ISSN 0973-3930

Volume 41 | Issue 1 | January–March 2021

International Journal of Diabetes in Developing Countries

Official Publication of
Research Society for the
Study of Diabetes in India

Springer

International Journal of Diabetes in Developing Countries
Incorporating Diabetes Bulletin

INTERNATIONAL ADVISORY BOARD

Aims and Scope

International Journal of Diabetes in Developing Countries targets a readership consisting of clinicians, research workers, paramedical personnel, nutritionists and health care personnel working in the field of diabetes. Original research work and reviews of interest to the above group of readers is considered for publication in the journal.

The journal has a goal of serving as an important resource material in diabetes for its readers, mainly in the developing world.

Copyright Information

For Authors

As soon as an article is accepted for publication, authors will be requested to assign copyright of the article (or to grant exclusive publication and dissemination rights) to the publisher (respective the owner if other than Springer Nature). This will ensure the widest possible protection and dissemination of information under copyright laws.

More information about copyright regulations for this journal is available at www.springer.com/13410

For Readers

While the advice and information in this journal is believed to be true and accurate at the date of its publication, neither the authors, the editors, nor the publisher can accept any legal responsibility for any errors or omissions that may have been made. The publisher makes no warranty, express or implied, with respect to the material contained herein.

Subscription Information

International Journal of Diabetes in Developing Countries is published 4 times a year. Volume 41 (4 issues) of will be published in 2021.

ISSN 0973-3930 print version
ISSN 1998-3832 electronic version

For information on subscription rates please contact Springer Nature Customer Service Center: customerservice@springernature.com

The Americas (North, South, Central America and the Caribbean)
Springer Journal Fulfillment, 233 Spring Street, New York, NY, 10013-1578, USA
Tel. 800-SPRINGER (777-4643); 212-460-1500 (outside North America)

Outside the Americas
Springer Nature Customer Service Center GmbH
Tiergartenstr. 15, 69121 Heidelberg, Germany
Tel.: +49-6221-345-4303

Advertisements

Disclaimer

Journal Website

www.springer.com/13410
Electronic edition: link.springer.com/journal/13410

For the actual version of record please always check the online version of the publication.

Office of Publication

Springer Science+Business Media B.V., Van Godewijckstraat 30, 3311 GX Dordrecht, The Netherlands

International Journal of Diabetes in Developing Countries

Volume 41 · Number 1 · January–March 2021

EDITORIAL

Sleep: an emerging therapeutic target in diabetes care
N. Raizada · S.V. Madhu **1**

CONSENSUS

RSSDI clinical practice recommendations for screening, diagnosis, and treatment in type 2 diabetes mellitus with obstructive sleep apnea
V. Viswanathan · N. Ramakrishnan · B. Saboo · S. Agarwal **4**

ORIGINAL ARTICLES

Interaction effect between long sleep duration in early pregnancy and prepregnancy overweight/obesity on gestational diabetes mellitus
W. Wang · J. Zou · Q. Fu · T. Huang · M. Li · Z. Yao · L. Zou **22**

The effect of Ellagic acid on sleep quality in patients with type 2 diabetes: a randomized double blind clinical trial
M. Ghadimi · S. Hashemipour · M.R. Nooshabadi · M. Kavianpour · H.K. Haghighian **29**

Association of CAPN10 (SNP-19) genetic polymorphism and obesity with T2DM: a study on Bengali Hindu caste population
P. Sarkar · D. Chatterjee · A.R. Bandyopadhyay **37**

Association analysis of *HHEX* gene variant with type 2 diabetes risk
P. Banihashemi · H.R. Aghaei Meybodi · M. Afshari · N. Sarhangi · M. Hasanzad **43**

The value of adenosine deaminase activity in latent autoimmune diabetes in adults and type 2 diabetes patients
Y. Zhou · W.-D. Jin · Z.-Z. Pang · J. Xia · S.-F. Chen **48**

Estimation and SVM classification of glucose-insulin model parameters from OGTT data: a comparison with the ADA criteria · Estimation and SM classification of glucose-insulin model parameters
P. Vargas · M.A. Moreles · J. Peña · A. Monroy · S. Alavez **54**

Human ENPP1 gene polymorphism in DKD patients: a hospital-based case control study
S. Chandra · A.K. Singh · M. Singh · P. Pandey · C.S. Azad · S. Singh · P. Das · R.G. Singh **63**

Associations between plant-based dietary indices and dietary acid load with cardiovascular risk factors among diabetic patients
E. Daneshzad · F. Jahangir · J. Heshmati · B. Larijani · P.J. Surkan · L. Azadbakht **71**

Left ventricle diastolic dysfunction in a sample of prediabetic adults from Baghdad, Iraq
M.H. Alogaily · A.J. Alsaffar · M.B. Hamid **84**

Evaluation of platelet volume indices as predictive biomarkers of microvascular complications in patients with type 2 diabetes
R. Chawla · J. Sahu · H. Punyani · S. Jaggi **89**

Foot care practices among Diabetologists in India: A descriptive study by the Diabetic Foot Research India
V. Viswanathan · S. Nachimuthu · B.A. Khan **94**

Prevalence of musculoskeletal complications of type-2 diabetes mellitus in population of southern Punjab, Pakistan
M.N. Kalam · S.N.H. Shah · M.F. Rasool · A. Majeed · N. Ahmed · M. Saqlain **101**

Determinants of gestational diabetes mellitus: a hospital-based case–control study in coastal South India
A. Ganapathy · R. Holla · B.B. Darshan · N. Kumar · V. Kulkarni · B. Unnikrishnan · R. Thapar · P. Mithra · A. Kumar **108**

Maternal age at pregnancy and risk for gestational diabetes mellitus among Chinese women with singleton pregnancies
Y. Han · M. Tong · L. Jin · J. Yu · W. Meng · A. Ren · L. Jin **114**

Mediating effects of body composition at the first trimester on the occurrence of GDM at the early stage of the third trimester during the advanced maternal age
L. Jing · Z. Dan · W. Huiqun · L. Yanping · L. Yang · Y. Xuan · Z. Mengmou · L. Didong **121**

Barriers to postpartum follow-up of mothers with gestational diabetes mellitus and its implications: a mixed method study
N.P. Mathew · A.P. Rao · P. Narayanan **127**

Ocular surface assessment in maturity-onset diabetes of the young
O. Celikay · M. Kosker · S. Çalışkan · E. Cakal · F.A. Pinarli · C. Gurdal **136**

Assessment of nutritional status and quality of life in individuals with and without diabetes over 65 years of age
G. Pamuk · G. Kaner · E.M. Koc · T. Toşur · S. Dasdelen · S. Aksun · B.Ö. Pamuk **141**

Geriatric syndromes and the cumulative impacts on quality of life in older people with type 2 diabetes mellitus
T. Liu · X. Man · X. Miao **148**

Assessment of a group-based comprehensive diabetes management program to improve glycemic control, quality of life and self-care behavior in patients with type 2 diabetes mellitus in a primary healthcare setting of a metropolitan city in India: CDMP MUM Trial
M.P. Baviskar · S. Rangari · S. Mishra · B.S. Mohanta **156**

The predictors of perceived stress in patients with type 2 diabetes in Turkey: styles of coping with stress and metabolic variables
G. Bakan · F.H. Inci **164**

CASE REPORT

Concomitant diabetic ketocacidosis and renal tubular acidosis in a type 1 diabetes mellitus patient
K. Agarwal · D. Thakur · A. Gupta **172**

LETTER TO THE EDITOR

Missing the wood for the trees: cardiocentricity in current diabetes guidelines
S. Kalra · B. Saboo · N.K. Singh · S. Bajaj **175**

Further articles can be found at www.springerlink.com

Abstracted/Indexed in *CAB Abstracts, CNKI, Chemical Abstracts Service (CAS), Dimensions, EBSCO Academic Search, EBSCO CINAHL, EBSCO Discovery Service, EBSCO STM Source, EMBASE, EMCare, Google Scholar, Institute of Scientific and Technical Information of China, Japanese Science and Technology Agency (JST), Journal Citation Reports/Science Edition, Naver, OCLC WorldCat Discovery Service, ProQuest Biological Science Database, ProQuest Central, ProQuest Health & Medical Collection, ProQuest Health Research Premium Collection, ProQuest Immunology Abstracts, ProQuest India Database, ProQuest Natural Science Collection, ProQuest Nursing & Allied Health Database, ProQuest Pharma Collection, ProQuest Research Library, ProQuest SciTech Premium Collection, ProQuest-ExLibris Primo, ProQuest-ExLibris Summon, SCImago, SCOPUS, Science Citation Index Expanded (SciSearch), Semantic Scholar, TD Net Discovery Service, UGC-CARE List (India)*

Instructions for Authors for *Int J Diabetes Dev Ctries* are available at www.springer.com/13410.

Compliance with Ethical Requirements

International journal of Diabetes in Developing Countries requests that all authors comply with Springer's ethical policies. To view our ethics statements please visit the following:

· Conflict of Interest and Ethical Standards: http://www.springer.com/authors?SGWID=0-111-6-791531-0

· Informed Consent: http://www.springer.com/authors?SGWID=0-111-6-608209-0

Statement of Human and Animal Rights: http://www.springer.com/authors?SGWID=0-111-6-608309-0

International Journal of Diabetes in Developing Countries (2021) 41:1–3
https://doi.org/10.1007/s13410-021-00932-w

EDITORIAL

Sleep: an emerging therapeutic target in diabetes care

Nishant Raizada[1] · S. V. Madhu[1]

Published online: 13 February 2021

The management of chronic disorders such as diabetes mellitus is a challenge for modern medicine. Although several pharmacological agents have been developed for the treatment of diabetes mellitus, most oral drugs have limited efficacy in terms of reduction in glycosylated hemoglobin (HbA1c) levels along with a gradual decline in response as beta cell failure worsens. Insulin, although effective at all ranges of hyperglycemia, is cumbersome to administer and titrate, thereby making it unsuitable for many patients with type 2 diabetes mellitus. Hence, the need for novel therapeutic modalities in diabetes cannot be overemphasized.

Type 2 diabetes mellitus has been described as a lifestyle disorder. On a background of genetic predisposition, poor lifestyle choices in terms of diet and physical activity have been implicated in the pathogenesis of type 2 diabetes mellitus. Accordingly, diabetic diet and physical activity have proven to be the cornerstone in the management of diabetes. The decline in HbA1c with diabetic diet and physical activity exceeds that seen with individual pharmacological agents with the exception of insulin. However, diet and physical activity are not the only lifestyle factors affecting diabetes. Among the other lifestyle factors, sleep has unfortunately received less attention until recently. While obstructive sleep apnea has been well recognized as a risk factor for several metabolic diseases, the role of sleep disorders other than obstructive sleep apnea seems to be equally important.

Sleep disorders are commonly seen in diabetes. The prevalence of sleep disturbances in clinic-based populations of type 2 diabetes mellitus patients ranges from 33 to 73% [1–4]. Data suggests that sleep and sleep disorders influence diabetes at multiple points in the natural history of the disease. Sleep duration of less than 8 h has been shown to increase the risk of developing type 2 diabetes mellitus in a Chinese cohort and a risk score incorporating sleep duration has also been developed [5]. Sleep efficiency (time spent as sleep as a percentage of time spent in bed) of less than 80% was associated with type 2 diabetes mellitus with an odds ratio close to two in the Sleep Heart Heath Study [6]. A survey in Taiwanese public servants also revealed an association between shorter sleep duration and type 2 diabetes [7].

Gestational diabetes mellitus (GDM) which is a common precursor of type 2 diabetes mellitus in women appears to have links with sleep disturbances. In this issue, Wang et al. report that sleep duration of 10 h or more along with prepregnancy obesity is a strong risk factor for GDM. Both too much and too little night sleep and too much day time napping is associated with GDM in Chinese women [8]. The risk of GDM increased by 32% in those with shorter sleep duration in another study from China [9]. A systematic review and meta-analysis also found that poor sleep quality increases risk of GDM [10].

Sleep disorders appear to be associated with poor glycemic control in patients already suffering from diabetes mellitus. Sleep fragmentation was associated with worse HbA1c and fasting plasma glucose in a black population in the USA [11]. Poor sleep quality was also associated with higher HbA1c, BP, and obesity [12]. Even in patients who did not report any subjective sleep disturbances, a negative correlation between objective parameters of sleep quality and HbA1c was found [6].

Furthermore, sleep disorders have not only been associated with complications of diabetes but also with mortality risk. In a large cohort of adults from the USA, risk of mortality in type 2 diabetes mellitus patients (compared to non-diabetics) increased from 1.84 with a sleep duration of 7 h to 2.78 and 3.67 for 5 and 10 h respectively [13]. Similarly, data from the Korean National health insurance program found that newly diagnosed type 2 diabetes mellitus patients with sleep disturbances have a higher incidence of cardiovascular disease and all-cause mortality [14]. Poor sleep quality has also been shown to be associated with microvascular complications such as diabetic retinopathy [15].

✉ S. V. Madhu
drsvmadhu@gmail.com

[1] Department of Endocrinology, Center for Diabetes, Endocrinology and Metabolism, University College of Medical Sciences & GTB Hospital, New Delhi, India

Although data in type 1 diabetes mellitus is limited, the role of sleep disorders cannot be discounted. Reduced sleep duration and quality were associated with higher glycemic variability along with greater stress and depression in type 1 diabetes mellitus [16]. In another study, type 1 diabetes mellitus patients with poor sleep quality had higher total and LDL cholesterol as compared to good sleepers [17].

While the high prevalence of sleep disorders and their impact on diabetes risk, glycemic control, diabetes complications, and mortality paint a gloomy picture, the proverbial cloud does have a silver lining. In this issue, Ghadimi et al. have reported that administration of ellagic acid, a polyphenol known to be present in fruits and nuts, leads to a significant improvement in sleep quality in patients with type 2 diabetes mellitus. Although the mechanism of this improvement is unclear and a subject for further research, the authors hypothesize that the antioxidant properties of ellagic acid may contribute to reduction in reactive oxygen species and chemicals such as malondialdehyde—these molecules may play a role in sleep disturbances. The results of this double blind randomized trial are encouraging despite being a short duration study carried out in a small number of patients and could mark the beginning of a new approach in the management of diabetic patients if these can be replicated in larger studies with longer follow-up.

Patients with sleep disturbances due to coexisting obstructive sleep apnea (OSA) benefit from continuous positive airway pressure (CPAP) ventilation. CPAP in patients with type 2 diabetes and OSA have shown to improve HbA1c and reduce insulin resistance [18]. Even in patients with well-controlled diabetes and undiagnosed OSA, CPAP improves daytime sleepiness. The Diabetes Sleep Treatment Trial, which is currently ongoing , will compare CPAP with sham-CPAP with respect to improvement in glycemic control in patients with type 2 diabetes [19]. Liraglutide has shown to reduce apnea hypopnea index in patients with diabetes and OSA, presumably by inducing weight loss [20].

In patients who do not have OSA but still have sleep disturbances, several non-pharmacological interventions have been documented to be beneficial. In a randomized control trial, improvement in sleep quality in type 2 diabetes mellitus patients using cognitive behavioral therapy led to improvement in HbA1c to the tune of 0.9% [21]. A structured sleep education program in patients with type 2 diabetes who were sleeping after midnight leads to significant improvements in their sleep quality and glycemic control [22]. Nutritional ketosis under a continuous care intervention has also shown to improve sleep quality in type 2 diabetes [23].

Aerobic exercise training in type 2 diabetes has shown to improve sleep quality as well as quality of life [24]. However ,both aerobic and resistance exercise can predispose to hypoglycemia and cause sleep loss in type 1 diabetes [25]. Management of hypoglycemias by utilizing flash glucose monitoring can improve sleep disturbances [26]. Closed loop continuous subcutaneous insulin infusion (CSII) systems have also been studied regarding impact on sleep quality in type 1 diabetes although data suggests that they may not offer any advantage over sensor augmented pumps in this regard [27].

Treatment of both painless and painful neuropathy along with restless legs syndrome can also improve sleep disturbances [28]. In patients with diabetic neuropathic pain, mirogabalin has shown reduction in average daily sleep interference scores [29]. Sustained release oxycodone has also led to improvement in sleep quality in patients with type 2 diabetes and severe painful neuropathy [30]. Other drugs like mexiletine and gabapentin may also improve sleep disturbances by ameliorating pain in diabetic neuropathy [31].

Vitamin D supplements [32] and reservetrol [33] have been tried in healthy adults with sleep disorders with variable success although these have not been specifically evaluated in diabetic subjects.

Sleep and interventions to control sleep disorders have the potential to become an important component of diabetes care in future. Considering the limitations of current diabetes management, these novel strategies may well be the much needed ray of hope.

References

1. Sridhar GR, Madhu K. Prevalence of sleep disturbances in diabetes mellitus. Diabetes Res Clin Pract. 1994;23(3):183–6.
2. Skomro RP, Ludwig S, Salamon E, Kryger MH. Sleep complaints and restless legs syndrome in adult type 2 diabetics. Sleep Med. 2001;2(5):417–22.
3. Khorasani ZM, Ravan VR, Hejazi S. Evaluation of the prevalence of sleep disorder among patients with type 2 diabetes mellitus referring to Ghaem hospital from 2016 to 2017. Curr Diabetes Rev. 2020.
4. Khalil M, Power N, Graham E, Deschênes SS, Schmitz N. The association between sleep and diabetes outcomes - a systematic review. Diabetes Res Clin Pract. 2020;161:108035.
5. Liu X, Li Z, Zhang J, Chen S, Tao L, Luo Y, et al. Novel Risk Score for Type 2 Diabetes Containing Sleep Duration: A 7-Year Prospective Cohort Study among Chinese Participants. J Diabetes Res. 2020. https://doi.org/10.1155/2020/2969105.
6. Yan B, Zhao B, Fan Y, Yang J, Zhu F, Chen Y, et al. The association between sleep efficiency and diabetes mellitus in community-dwelling individuals with or without sleep-disordered breathing. J Diabetes. 2020;12(3):215–23.
7. Shih DP, Lin PY, Liang WM, Tseng PC, Kuo HW, Wang JY. Sleep Duration and Effort-Reward Imbalance (ERI) Associated with Obesity and Type II Diabetes Mellitus (T2DM) among Taiwanese Middle-Aged Public Servants. Int J Environ Res Public Health. 2020. https://doi.org/10.3390/ijerph17186577.
8. Wang S, Xu L, Jonas JB, You QS, Wang YX, Yang H. Prevalence and associated factors of dyslipidemia in the adult Chinese population. PLOS ONE. 2011;6(3):e17326.
9. Du M, Liu J, Na H, Zhao Z, Luo S, Wang H. Association between sleep duration in early pregnancy and risk of gestational diabetes mellitus: A prospective cohort study. Diabetes Metab. 2020. https://doi.org/10.1016/j.diabet.2020.101217.

10. Zhu B, Shi C, Park CG, Reutrakul S. Sleep quality and gestational diabetes in pregnant women: a systematic review and meta-analysis. Sleep Med. 2020;67:47–55.
11. Yano Y, Gao Y, Johnson DA, Carnethon M, Correa A, Mittleman MA, et al. Sleep Characteristics and Measures of Glucose Metabolism in Blacks: The Jackson Heart Study. J Am Heart Assoc. 2020. https://doi.org/10.1161/JAHA.119.013209.
12. Bener A, Al-Hamaq AOAA, Agan AF, Öztürk M, Ömer A. Sleeping disturbances and predictor risk factors among type 2 diabetic mellitus patients. Ann Afr Med. 2020;19:230–6.
13. Wang Y, Huang W, O'Neil A, Lan Y, Aune D, Wang W, et al. Association between sleep duration and mortality risk among adults with type 2 diabetes: a prospective cohort study. Diabetologia. 2020;63:2292–304.
14. Choi Y, Choi JW. Association of sleep disturbance with risk of cardiovascular disease and all-cause mortality in patients with new-onset type 2 diabetes: data from the Korean NHIS-HEALS. Cardiovasc Diabetol. 2020;19(1):61.
15. Dutta S, Ghosh S, Ghosh S. Association of sleep disturbance with diabetic retinopathy. Eur J Ophthalmol. 2020;1120672120974296.
16. Rechenberg K, Griggs S, Jeon S, Redeker N, Yaggi HK, Grey M. Sleep and glycemia in youth with type 1 diabetes. J Pediatr Health Care Off Publ Natl Assoc Pediatr Nurse Assoc Pract. 2020;34(4):315–24.
17. de Mattos ACMT, Campos YS, Fiorini VO, Sab Y, Tavares BL, Velarde LGC, et al. Relationship between sleep disturbances, lipid profile and insulin sensitivity in type 1 diabetic patients: a cross-sectional study. Arch Endocrinol Metab. 2020;64(4):412–7.
18. Martínez-Cerón E, Barquiel B, Bezos AM, Casitas R, Galera R, García-Benito C, et al. Effect of Continuous Positive Airway Pressure on Glycemic Control in Patients with Obstructive Sleep Apnea and Type 2 Diabetes. A Randomized Clinical Trial. Am J Respir Crit Care Med. 2016;194:476–85.
19. Chasens ER, Atwood CW, Burke LE, Korytkowski M, Stansbury R, Strollo PJ, et al. Diabetes sleep treatment trial: Premise, design, and methodology. Contemp Clin Trials. 2019;76:104–11.
20. Blackman A, Foster GD, Zammit G, Rosenberg R, Aronne L, Wadden T, et al. Effect of liraglutide 3.0 mg in individuals with obesity and moderate or severe obstructive sleep apnea: the SCALE Sleep Apnea randomized clinical trial. Int J Obes 2005. 2016;40(8):1310–9.
21. Zuo X, Dong Z, Zhang P, Zhang P, Chang G, Xiang Q, et al. Effects of cognitive behavioral therapy on sleep disturbances and quality of life among adults with type 2 diabetes mellitus: A randomized controlled trial. Nutr Metab Cardiovasc Dis. 2020;30:1980–8.
22. Li M, Li D, Tang Y, Meng L, Mao C, Sun L, et al. Effect of diabetes sleep education for T2DM who sleep after midnight: a pilot study from China. Metab Syndr Relat Disord. 2018;16(1):13–9.
23. Siegmann MJ, Athinarayanan SJ, Hallberg SJ, McKenzie AL, Bhanpuri NH, Campbell WW, et al. Improvement in patient-reported sleep in type 2 diabetes and prediabetes participants receiving a continuous care intervention with nutritional ketosis. Sleep Med. 2019;55:92–9.
24. S Delevatti R, Schuch FB, Kanitz AC, Alberton CL, Marson EC, Lisboa SC, et al. Quality of life and sleep quality are similarly improved after aquatic or dry-land aerobic training in patients with type 2 diabetes: a randomized clinical trial. J Sci Med Sport. 2018;21(5):483–8.
25. Reddy R, El Youssef J, Winters-Stone K, Branigan D, Leitschuh J, Castle J, et al. The effect of exercise on sleep in adults with type 1 diabetes. Diabetes Obes Metab. 2018;20:443–7.
26. Al Hayek AA, Al Dawish MA. Assessing Diabetes Distress and Sleep Quality in Young Adults with Type 1 Diabetes Using FreeStyle Libre: A Prospective Cohort Study. Diabetes Ther. 2020;11:1551–62.
27. Sharifi A, De Bock MI, Jayawardene D, Loh MM, Horsburgh JC, Berthold CL, et al. Glycemia, Treatment Satisfaction, Cognition, and Sleep Quality in Adults and Adolescents with Type 1 Diabetes When Using a Closed-Loop System Overnight Versus Sensor-Augmented Pump with Low-Glucose Suspend Function: A Randomized Crossover Study. Diabetes Technol Ther. 2016;18:772–83.
28. Choi D, Kim BY, Jung CH, Kim CH, Mok JO. Association between sleep quality and painless diabetic peripheral neuropathy assessed by current perception threshold in type 2 diabetes mellitus. Diabetes Metab J. 2020.
29. Merante D, Rosenstock J, Sharma U, Feins K, Hsu C, Vinik A, et al. Efficacy of mirogabalin (DS-5565) on patient-reported pain and sleep interference in patients with diabetic neuropathic pain: secondary outcomes of a phase II proof-of-concept study. Pain Med Malden Mass. 2017;18(11):2198–207.
30. Yao P, Meng L-X, Ma J-M, Ding Y-Y, Wang Z-B, Zhao G-L, et al. Sustained-release oxycodone tablets for moderate to severe painful diabetic peripheral neuropathy: a multicenter, open-labeled, postmarketing clinical observation. Pain Med Malden Mass. 2012;13(1):107–14.
31. Oskarsson P, Ljunggren JG, Lins PE. Efficacy and safety of mexiletine in the treatment of painful diabetic neuropathy. The Mexiletine Study Group. Diabetes Care. 1997;20(10):1594–7.
32. Majid MS, Ahmad HS, Bizhan H, Hosein HZM, Mohammad A. The effect of vitamin D supplement on the score and quality of sleep in 20–50 year-old people with sleep disorders compared with control group. Nutr Neurosci. 2018;21(7):511–9.
33. Wightman EL, Haskell-Ramsay CF, Reay JL, Williamson G, Dew T, Zhang W, et al. The effects of chronic trans-resveratrol supplementation on aspects of cognitive function, mood, sleep, health and cerebral blood flow in healthy, young humans. Br J Nutr. 2015;114(9):1427–37.

International Journal of Diabetes in Developing Countries (2021) 41:4–21
https://doi.org/10.1007/s13410-020-00909-1

CONSENSUS

RSSDI clinical practice recommendations for screening, diagnosis, and treatment in type 2 diabetes mellitus with obstructive sleep apnea

Vijay Viswanathan[1] · Nagarajan Ramakrishnan[2] · Banshi Saboo[3] · Sanjay Agarwal[4]

Received: 6 December 2020 / Accepted: 22 December 2020 / Published online: 25 February 2021

Abstract

Purpose of the study Type 2 diabetes mellitus (T2DM) and obstructive sleep apnea (OSA) are closely associated diseases with a significant impact on public health. Both diseases are highly prevalent and the common overlapping risk factors include obesity, old age, and a generally high preponderance in men. A growing body of evidence suggests a bidirectional association between OSA and T2DM; both, in turn, constitute as strong risk factors for cardiovascular diseases, the leading cause of premature deaths and morbidity in India. Several studies have reported a higher prevalence of OSA in Indian patients with T2DM, despite lower levels of obesity. However, OSA remains an underdiagnosed condition in Indian patients with T2DM due to lack of awareness, uncertainty about treatment options, and non-availability of diagnostic facilities.

Methods This document embodies evidence-based clinical practice recommendations and outlines an optimized care pathway for patients with T2DM and OSA, based on consensus multidisciplinary observations and clinical experiences.

Results and conclusions Implementation of screening, diagnosis, and treatment of OSA in patients with T2DM at initial stages could potentially alleviate the risk of cardiovascular disease and substantially improve their quality of life. The recommendations emphasize the need for collaborative efforts from diabetologists, endocrinologists, and sleep medicine specialists towards systematic screening, diagnosis, and treatment of coexisting T2DM and OSA for enhanced patient care.

Keywords Consensus guideline · Continuous positive airway pressure · India · Obstructive sleep apnea · Type 2 diabetes mellitus

Extended working group: Dr Anuj Maheshwari, Dr. B.M. Makkar, Dr Bikask Bhattacharjee, Dr C R Anand Moses, Dr C H Vasanth Kumar, Dr J K Sharma, Dr L Srinivas Murthy, Dr Pratap Jethwani, Dr Rajeev Chawla, Dr Ravindra Mehta, Dr Sanjay Reddy, Dr Shalini Jaggi, Dr Sudhir Bhandari, Dr Sujoy Ghosh, Dr Sunil Gupta, Dr Suresh Ramasubban, Dr Vijay Panikar

✉ Vijay Viswanathan
drvijay@mvdiabetes.com

[1] M.V. Hospital for Diabetes and Prof. M. Viswanathan Diabetes Research Centre (WHO Collaborating Centre for Research, Education and Training in Diabetes), No. 4, West MadaChruch Road, Royapuram, Chennai, Tamil Nadu 600013, India

[2] Nithra Institute of Sleep Sciences and Department of Critical Care and Sleep Medicine, Apollo Hospitals, Chennai, Tamil Nadu, India

[3] Dia Care-Diabetes and Hormone Centre, Ahmedabad, Gujarat, India

[4] Aegle Clinic Diabetes Care and Ruby Hall Clinic, Pune, Maharashtra, India

Background

The global burden of diabetes mellitus (DM) is rapidly escalating and nearly tripled over the past two decades [1]. According to estimates of the International Diabetes Federation (IDF) 2017,around 463 million or 9.3% of the world's population are living with DM and among these 90% of the cases are type 2 DM (T2DM). India, with over 77 million adults with DM, is ranked second in the world and the estimated number of affected individuals is expected to rise up to 134.2 million by 2045 [1].

Obstructive sleep apnea (OSA) is a common form of sleep-disordered breathing characterized by frequent episodes of partial or complete upper airway blockage resulting in oxygen desaturation and sleep fragmentation and non-refreshing sleep with daytime fatigue and sleepiness [2]. Recent literature-based evidence estimated that approximately 1 billion adults aged 30–69 years worldwide suffer from mild to severe OSA

and of these 425 million are moderate to severe for which treatment is usually recommended [3]. OSA is also a significant public health problem in India with a prevalence ranging between 3.5% and 19.5%, although a majority of them remain clinically undiagnosed [4].

A growing body of evidence suggests a positive and bidirectional association between OSA and the development of T2DM; both, in turn, constitute as strong risk factors for cardiovascular diseases (CVD), that affect millions worldwide [3, 5–9]. Both diseases share common determinants such as obesity and increasing age and have a higher predisposition in men. Several facets of OSA including short sleep duration and disturbances in the circadian rhythm have recently emerged as potential risk factors for insulin resistance (IR) and development of impaired glucose tolerance and T2DM [10]. Inadequately treated OSA in T2DM is also associated with higher risk of micro- and macrovascular complications including neuropathy, retinopathy, nephropathy, and peripheral arterial diseases. Despite this, OSA remains an underdiagnosed and untreated condition in adults with T2DM due to a general lack of awareness among public and healthcare professionals [11, 12]. The IDF and the American Diabetes Association (ADA) guidelines recommend screening for OSA in all adults with T2DM to reduce diabetes-associated complications [11, 13].

Epidemiology

The prevalence of OSA in adults with T2DM is challenging to define due to the lack of multicenter population-based studies in India. However, several cross-sectional studies across various geographies in India suggest a high prevalence of OSA in adults with T2DM, which varies widely between 23.7% and 95% depending on the study population, methods, and criteria used for diagnosis [14–18]. A hospital-based study from South India on 203 adults with T2DM showed that OSA is prevalent among 23.7% of its study population [14]. A similar prevalence rate for OSA (24.3%) was reported by *Ekka* et al among 325 patients with T2DM in north India [17]. In a study from Central India, among 33patients with T2DM, the prevalence rate for OSA was 27% [15]. A clinic-based cross-sectional study from the Western part of India reported a prevalence rate of 54% among patients with T2DM, which is almost double than other geographies across India [18]. Interestingly, a long-term study by *Malik* et al reported a prevalence rate of over 95% for OSA in north Indian patients with T2DM, which is the highest reported prevalence so far in India [16]. Research studies consistently reported a higher prevalence of OSA in T2DM among men than in women [14, 17, 18]. Moreover, the prevalence of OSA in patients with T2DM among the urban population is higher compared with the rural population [18].

Need for recommendations

The evidence-based Indian initiative on obstructive sleep apnea (INOSA) consensus guidelines stated that the prevalence of OSA in patients with T2DM is higher than those without disease and the risk of developing T2DM increases with the severity of OSA [19]. Existing evidence also suggests that OSA worsens glycemic control in T2DM and may contribute to a higher percentage of diabetes-related complications such as CVD, diabetic neuropathy as well as retinopathy [20].

Given the enormous public health burden, better clinical practices are needed to ensure that patients presenting with either OSA or T2DM are assessed for the co-existence of the other. Both IDF and ADA have already recommended screening for OSA in patients with T2DM [11, 13]. In India, current guidelines for the management of patients with T2DM do not include evaluation for possible OSA. Leading endocrinologists and diabetologists collaborated with sleep specialists to develop this clinical recommendation that provides evidence-based guidance to researchers, patients, clinicians, public health policy-makers, and all other stakeholders' for the screening, diagnosis, and treatment of OSA in patients with T2DM. These recommendations are convened to enable clinical decision making and should be appropriately amended based on the individual patient requirements such as comorbidities, and other factors based on good clinical judgment and practices. These are consensus-based recommendations from the published literature evidence and does not contain any work on animals or human participants conducted by any of the authors.

Obstructive sleep apnea

Signs and symptoms

The pathophysiology of OSA is multifactorial, and the signs and symptoms may vary among the affected individuals. OSA can be mainly grouped under two conditions: present during or around sleep and while the individual is wide-awake [21]. Recurrent collapse of nasopharyngeal and oropharyngeal airways during sleep resulting in substantially reduced (hypopnea) or completely interrupted (apnea) airflow even with constant breathing efforts is a hallmark feature of OSA. Loud snoring is another characteristic manifestation of OSA and in most cases is associated with a brief revival from sleep [22]. An autonomic nervous system stimulation that results in sudden awakening with gasping of air can lead to palpitations, sweating, or even panic [23]. Once the individual is awake, this shortness of breath quickly settles. This sudden awakening can make it difficult to fall asleep again and this is later diagnosed as an OSA. The fragmented sleep, daytime fatigue,

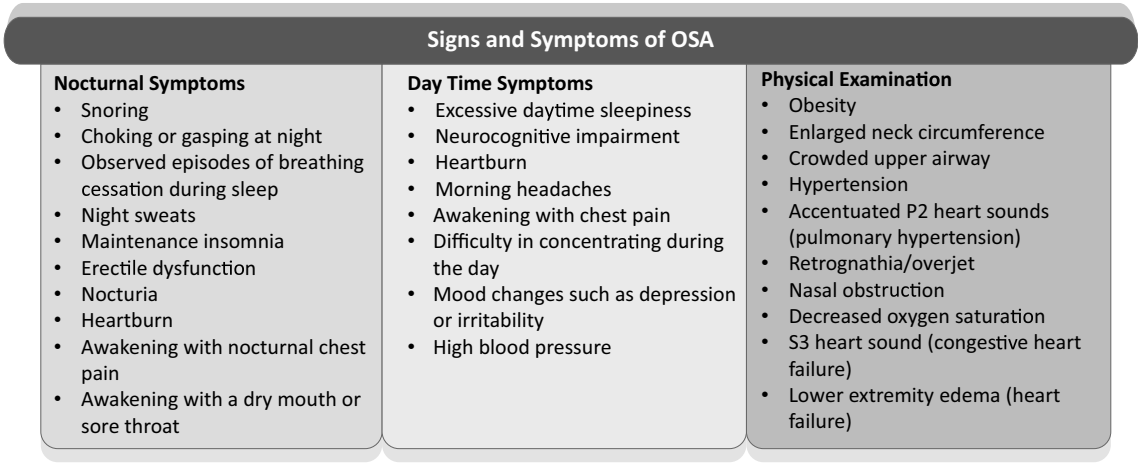


Fig. 1 Signs and symptoms of obstructive sleep apnea. OSA, obstructive sleep apnea; P2, pulmonary valve; S3, third heart sound

and sleepiness are broadly identified as symptoms of OSA [24].

The classical symptoms of OSA mainly involve snoring, excessive daytime sleepiness, choking or gasping at night, night sweats, neurocognitive impairment, heartburn, morning headaches, maintenance insomnia, erectile dysfunction, and nocturia (Fig. 1) [25]. The physical examination findings of OSA mainly comprise obesity, enlarged neck circumference, crowded upper airway, hypertension, accentuated P2 heart sounds (pulmonary hypertension), retrognathia/overjet, nasal obstruction, decreased oxygen saturation, S3 heart sound (congestive heart failure), and lower extremity edema (heart failure) [25].

Screening and diagnosis

Over 80% of individuals with moderate-to-severe OSA remain undiagnosed [26]. Considering the overwhelming impact of OSA on health and quality of life, it is important to effectively diagnose patients with OSA and ensure optimal management [27–30]. Thus, an appropriate screening tool is essential to categorize patients based on their clinical symptoms and physiological risk factors.

The prospective diagnostic strategy mainly includes an assessment of a formal clinical history using screening questionnaires, clinical questionnaire tools, and prediction tools [23]. Screening questionnaires are simple, low-cost tools that can be used to segregate patients eligible for diagnostic tests. The US Preventive Services Task Force recommends the Epworth Sleepiness Scale (ESS) [30], STOP Questionnaire (Snoring, Tiredness, Observed Apnea, High Blood Pressure) [31], STOP-Bang Questionnaire (STOP Questionnaire plus body mass index [BMI], Age, Neck Circumference, and Gender) [32], the Berlin questionnaire [33], and the Wisconsin Sleep Questionnaire [34] to screen patients with OSA in primary care setting [35] (Fig. 2).

In addition to the risk factors, physical examination comprising BMI, reduced distance and increased angles from the chin to the thyroid cartilage, and a narrow oropharyngeal opening can be predictive of OSA [24, 25]. Individuals screened positive would have to undergo a diagnostic test to confirm the presence of OSA. Polysomnography (PSG) is a noninvasive, sleep laboratory, or home-based testing using a portable monitor to quantify apnea-hypopnea index (AHI), calculated by adding all number apnea and hypopnea events and then dividing it by total sleep time. PSG involves simultaneous recording of OSA determinant physiologic sleep variables such as electroencephalogram, electrooculogram (eye movements), chin electromyogram (muscle tone), electrocardiogram, respiratory effort, airflow, and oxygenation [36].

OSA-excess weight-T2DM

Several longitudinal cohort studies and meta-analysis have reported an increased risk of poor metabolic control and incident T2DM in patients with severe to moderate sleep apnea [37–40]. Findings also suggest that self-reported history of sleep-disordered breathing and snoring are independently associated with glucose intolerance and insulin resistance in patients with T2DM [27, 41, 42]. Aggregated with aging and obesity, OSA has shown to increase the risk and severity of T2DM [43, 44]. Interestingly, Indian patients with T2DM with a low BMI are also at risk of developing OSA. In an Indian study with over 400 T2DM patients from a specialty diabetes clinic, nearly 28% of patients with BMI ≤ 25 kg/m^2 had a high risk of OSA as assessed on the Epworth Sleepiness Scale [45]. Thus, epidemiologically Indians and South Asians have a similar risk of OSA as compared with Americans or Europeans despite lower rates of obesity [46].

Obesity is a predominant risk factor and a 10% increase in body weight has been correlated with a six-fold increase in the risk of developing OSA [47]. Sleep fragmentation due to

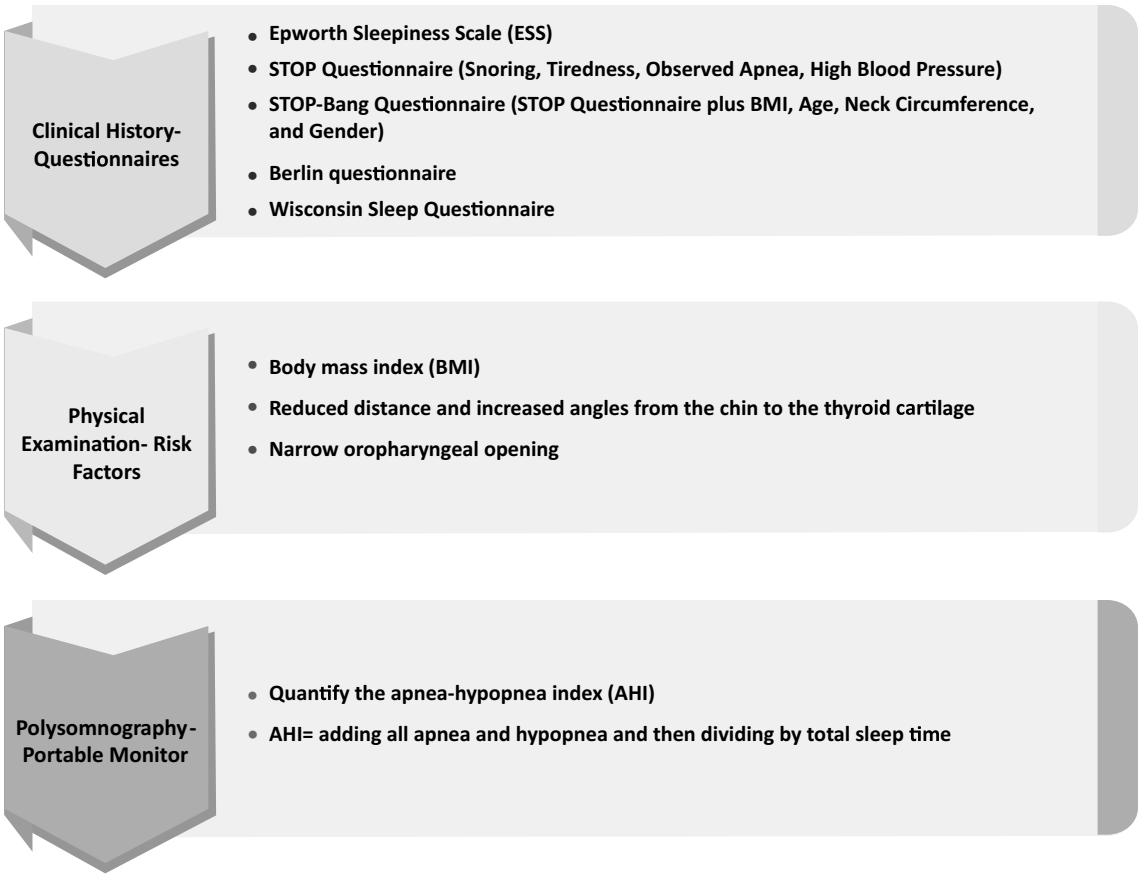


Fig. 2 Diagnosis of obstructive sleep apnea

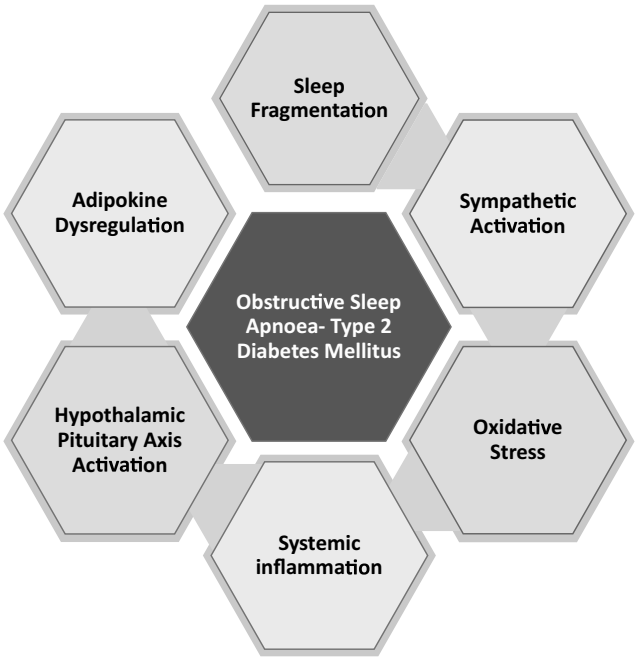


Fig. 3 Plausible mechanisms of development of glucose intolerance in OSA-T2DM patients

sudden awakening during sleep leads to activation of the autonomic nervous system. Augmented oxidative stress and chronic inflammation due to intermittent hypoxia and enhanced sympathetic activity dysregulate the glucose metabolism. It is further postulated that hypoxia may have damaging effect on pancreatic β-cell, liver, and adipose tissue function, which could further disrupt the glucose homeostasis [48] (Fig. 3).

Insufficient (≤ 5 h/day) or excessive (≥ 9 h/day) sleep interval is associated with a risk of developing T2DM, which is comparable with physical immobility or inactivity. The mechanism of glycemic dysregulation and obesity with sleep disturbances is mainly linked with overactivity of the sympathetic nervous system that triggers over-eating or binge eating and commonly reported in patients with T2DM; this is ultimately linked to excessive weight gain [43]. Dysregulation of neuroendocrine control of appetite associated with disturbed sleep leads to increased circulating levels of hunger promoting hormone (ghrelin) and decreased satiety factor (leptin) [49]. Prevalence of undiagnosed OSA (86.6%) is currently high in obese patients with T2DM [50].

Earlier studies comparing insulin resistance in total sleep-deprived individuals versus those with normal sleep conditions demonstrated that insulin resistance was induced by

acute sleep deprivation [51, 52]. Findings from a randomized controlled study suggested that an improvement of sleep pattern in obese patients with T2DM significantly reduced body weight, BMI, and glycated hemoglobin (HbA1c) as compared with the control group [53]. Confirming the two-way link, the Sleep AHEAD randomized controlled study reported clinically relevant improvements in OSA among obese patients with T2DM following lifestyle-based behavioral weight loss program and increased physical activity [54]. The benefit of weight loss was observed in men with severe OSA at baseline and participants with a weight loss of 10 kg or more who showed the greatest reductions in AHI [48]. Thus, effective weight loss strategies could produce meaningful improvements in outcomes among patients with OSA and T2DM.

Current treatment options for OSA

There are several options available for the management of OSA according to disease severity, which include:

- Lifestyle modifications such as weight loss through physical exercise for all obese people with OSA should be recommended for regardless of other interventions. Additionally, risk factors such as alcohol, smoking, and sedative medications should be avoided.
- Continuous positive airway pressure (CPAP) is regarded as the gold standard treatment for OSA. Patients with AHI ≥ 5 with symptoms or AHI ≥ 15 are primarily recommended for treatment with auto-CPAP therapy, as a first line of therapy [55]. During CPAP treatment, the pressurized air is delivered into the upper airways, to relieve obstruction during sleep [56, 57]. The mask is worn over the nose and/or mouth while sleeping, which is connected to the machine that delivers pressurized air continuously [58]. Despite all benefits, patient's intolerance, unacceptance, and non-adherence due to mask discomfort may limit the use of CPAP. Therefore, counseling, and open discussion with patients should be encouraged to mitigate apprehensions and negative perceptions about CPAP. Bi-level positive airway pressure (PAP) may be considered in patients who are intolerant to CPAP and is suitable for non-obstructive sleep-related hypoventilation and patients with overlap syndromes leading to hypoventilation. It works by administering pressure between inspiratory and expiratory cycles thereby combating the inspiratory flow limitation of the upper airway and increasing the tidal volume. This noninvasive method is considered more appropriate for obese patients with hypoventilation, patients with chronic obstructive pulmonary disorder, or alveolar hypoventilation associated with neuromuscular disorders [59].
- Oral appliances are widely prescribed as a treatment for OSA in patients with mild-to-moderate OSA, particularly in patients who are unable to use CPAP [60]. These are known to alleviate airway obstruction by enlarging the upper airway or reducing its collapsibility during sleep [61]. A dental device can be used to keep the airway open, which is noninvasive in nature and has emerged as an alternative treatment for OSA. Oral devices are categorized as tongue-retaining devices and mandibular advancement devices (MAD) or mandibular advancement splints (MAS) or mandibular repositioning appliances (MRA) [62, 63]. The MAD is more widely applied in clinical practice. MAD appliances enable mandibular protrusion with respect to the resting position by covering the upper and lower teeth, which in turn advances the tongue position and subsequently increases oropharyngeal volume. Accredited sleep specialists may predict an effective mandibular protrusion position by using polysomnographic evaluation with a remotely controlled mandibular positioner to help customize the device [64]. In patients with mild-to-moderate OSA, a > 50% reduction in AHI to < 5/h was reported in 42.8% of patients using an oral appliance and 73.2% of patients using CPAP. The odds of achieving AHI < 5/h was 49 times greater and < 10/h 89 times greater in patients treated with the oral appliance when compared with the control untreated group [65, 66]. The tongue-retaining devices hold the tongue forward by suction, thereby avoiding its collapse into airways. However, these devices have poor tolerance among patients and have shown inadequate efficacy [60, 66]. Objective tracking of device use and adherence is usually challenging. However, the difference between the mean subjective adherence rates for oral appliance users was 0.70 more hours per night than the objective adherence rates among CPAP users [65, 66].
- Surgery is warranted in few conditions that need anatomical restructuring to reduce the obstruction in nose, oropharynx, or hypopharynx. Such conditions include retro positioned maxilla or mandible, enlarged pharyngeal fat pads, soft palate or tongue, narrow posterior airway space, and upper airway hypotonia. Nasal surgeries, palatal surgeries (uvulopalatopharyngoplasty), and tongue-based surgeries (genioglossus advancement with hyoid suspension) are performed at level 1 to alleviate OSA maxillomandibular advancement (MMA) is a more complex procedure and generally reserved for patients with major OSA and for obstructions that could not be resolved in level 1 surgeries [63, 67].

Biological links/pathophysiology between OSA and T2DM

Obesity is a major confounder for the association between T2DM and OSA [68]. Nearly 60–90% of individuals with

OSA are overweight, and the relative risk of OSA in obesity (BMI > 29 kg/m^2) is ≥ 10 [11]. Several studies report a substantial impact of fat accumulation around the abdomen and neck regions on upper airway size and function. Increasing neck circumference, reduced pharyngeal lumen size, and compromised upper airway muscle force due to fat deposition in obese individuals leads to occlusion of the upper airway during sleep, resulting in OSA [69]. Additionally, reduced lung volume and upper airway size secondary to increasing mass effect of central obesity on the chest wall and reduced tracheal traction worsens hypoxia and contributes to increased risk of OSA [70]. The relationship between OSA and T2DM is bidirectional: OSA is a risk factor for T2DM and vice versa [9]. The following sections outline the postulated links.

Does OSA play any role in the development of T2DM?

OSA may directly or indirectly play a role in development of T2DM. The intermittent hypoxia and sleep fragmentation are postulated to induce metabolic dysfunction and alteration of glucose metabolism that eventually contributes to the development of T2DM [71] (Fig. 4). Episodes of hypoxia also reduce insulin sensitivity without a compensatory increase in insulin secretion, potentially suggesting β-cell dysfunction. Other mechanisms associated with disrupted glucose metabolism include hormonal changes such as activation of the hypothalamic–pituitary–adrenal axis; higher release of catecholamines, ghrelin, leptin; and decreased adiponectin in OSA. Further, recurrent hypoxia and sleep arousals contribute to increased sympathetic activity leading to dysregulation of glucose and fat metabolism and the development of T2DM. Oxidative stress in OSA and the elevated levels of inflammatory cytokines such as tumor necrosis factor alpha, interleukin-6 (IL-6), IL-8, and nuclear factor kappa light chain enhancer of activated B cells NF-κB also contribute to IR and β-cell dysfunction [72].

Does OSA have any effect on components of metabolic syndrome?

Metabolic syndrome is a group term for several metabolic abnormalities including central obesity, IR or glucose intolerance, hypertension, and dyslipidemia, and is associated with an increased risk for T2DM and CVDs [73]. Several studies identified an independent and bidirectional risk association between OSA and metabolic syndrome or its individual components [74, 75]. Moreover, the severity of OSA is associated with poor control of components of metabolic syndrome such as hyperglycemia, hypertension and dyslipidemia [76, 77]. Existing literature evidence suggests that OSA initiates several intermediary mechanisms such as inflammation, neurohumoral alterations including sympathetic activation, and oxidative stress that have been suggested to increase the risk of metabolic syndrome as well as its components [78].

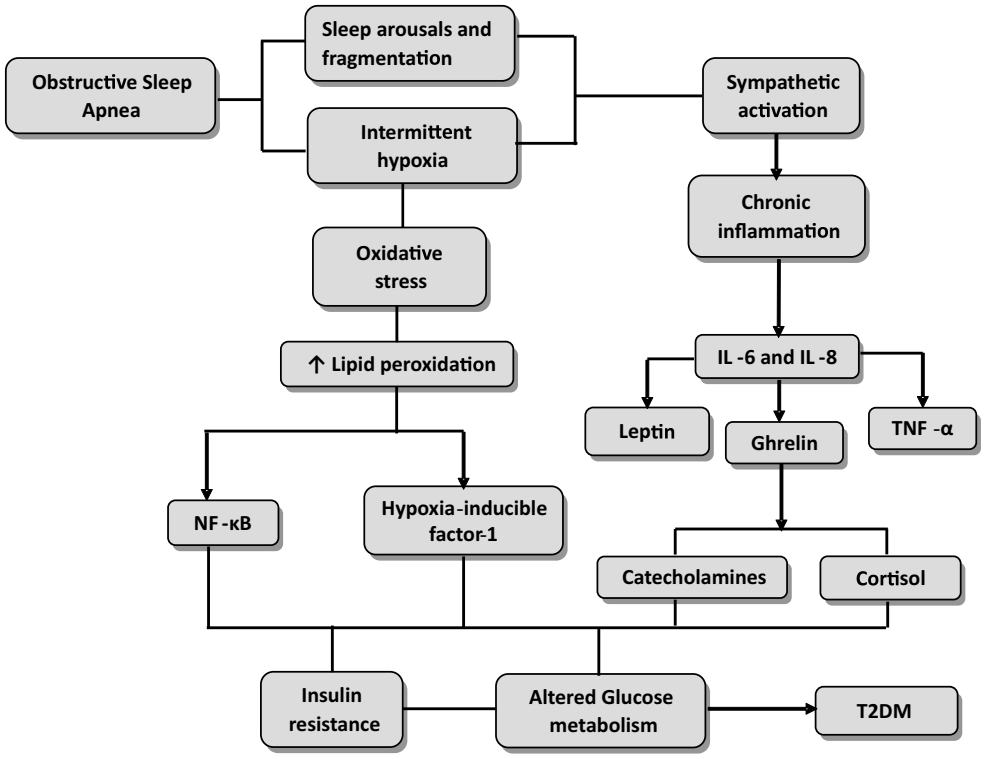


Fig. 4 Link between OSA and T2DM. OSA, obstructive sleep apnea; IL, interleukin; NF-κB, nuclear factor kappa light chain enhancer of activated B cells; T2DM, type 2 diabetes mellitus; TNF-α, tumor necrosis factor alpha

Does T2DM play a role in the development of OSA?

The role of T2DM as a risk factor for OSA is less explored. In a study evaluating risk factors (odds ratio [95% CI]) for OSA in a population of 3565 individuals, waist circumference (1.34 [1.19–1.52]), fasting insulin, homeostasis model assessment of insulin resistance (HOMA-IR) (1.31 [1.13–1.51] both) and triglycerides (1.24 [1.09–1.41]) were associated with a greater risk of observed apnea [79]. Notably, these risk factors are commonly associated with T2DM. It is postulated that T2DM can contribute to the development or exacerbated progression of pre-existing OSA. Increasing body weight in T2DM is the most common determinant of OSA that has a higher preponderance in men [80]. Among other possible mechanisms, loss of upper airway innervation and reduced neuromuscular response due to autonomic neuropathy in diabetes explains the abnormal control of breathing and frequent sleep apnea in these patients [80–82].

Does OSA have any effect on glycemic control of existing T2DM patients?

Several cross-sectional studies have suggested that OSA has a significant impact on the glucose metabolism and glycemic control in both patients with or without T2DM [83, 84]; however, data from long-term prospective longitudinal studies are lacking. Existing literature shows that both presence and severity of untreated OSA independently associate with poor glycemic control (increased HbA1c levels) in patients with T2DM; despite adjusting for relevant confounding factors such as age, sex, race, BMI, diabetes duration, lipids, exercise, blood pressure and insulin therapy [83, 85, 86]. Moreover, both duration and quality of sleep were also correlated with worse glycemic control; necessitating the need for treatment of OSA in patients with T2DM [87]. However, the effect of CPAP therapy on glycemic control or IR have had mixed results in patients with T2DM (Table 1).

Public health implications of OSA and T2DM

Both OSA and T2DM are closely associated diseases with high prevalence and having major impact on public health. There is increasing evidence that OSA is associated with risk of both T2DM and CVD complications; resulting in significant socioeconomic burden [1, 11, 13, 88]. It is likely that over half of the people with T2DM suffer from sleep disturbances and of these, up to one-third who are severely affected may require treatment [11].

Literature evidence shows that the presence and severity of OSA in patients with T2DM have been implicated in macro- and microvascular complications of diabetes mellitus [88]. OSA shares several common molecular mechanisms with hyperglycemia which results in microvascular complications in patients with T2DM [88]. In patients with T2DM, the presence of OSA was associated with the risk of diabetic retinopathy [89], diabetic macular edema [90], diabetic neuropathy [91], and diabetic nephropathy [92]. It was also well-established that OSA in patients with or without T2DM was strongly implicated for CVD in numerous cross-sectional and longitudinal studies [93, 94]. In both Sleep Heart Health Study and the Wisconsin Sleep Cohort Study, OSA was strongly associated with hypertension [7, 95, 96]. Moreover, OSA is the most common secondary cause of drug-resistant hypertension and linked to a non-dipping form of hypertension [7, 97]. Though studies in patients with T2DM are lacking, OSA has been associated with a higher risk of developing coronary artery disease [5], stroke [98], atrial fibrillation [99], atherosclerosis [100], dyslipidemia [101], arterial stiffness [102], cardiac arrhythmias [103], and heart failure [5]. Other comorbidities that have been associated with OSA include cognitive impairment, depression, decreased quality of life, motor vehicle accidents, psychological social function, and erectile dysfunction [88].

The high prevalence of OSA in T2DM raises the likelihood that some of the morbidity and mortalities linked to T2DM may be attributable to undiagnosed OSA [50]. This was clearly evident in the Sleep AHEAD trial where alarmingly 86% of obese patients with T2DM were found to have undiagnosed OSA with an AHI of 20.5 events/h [50]. In a 10-year long-term observation study, obese patients with OSA had higher healthcare resource utilization and associated costs; interestingly these were increased over time until diagnosis and decreased after treatment [104]. Therefore, even in the absence of clinical manifestations, physicians handling obese patients with T2DM should be screened and treated appropriately as a part of routine clinical practice for better health outcomes associated with adverse consequences of OSA [50, 105].

Recommendations for screening of patients for OSA for diabetologists [19, 106–108]

Figure 5 outlines the recommendations for screening, diagnosis, and treatment of OSA in patients with T2DM. As discussed, any of the validated, easy-to-use questionnaires (STOP, STOP-Bang, Berlin Questionnaire, Epworth Sleepiness Scale, or Wisconsin Sleep Questionnaire) can be used for screening. STOP and STOP-Bang are reliable tools with high sensitivity to detect mild, moderate, and severe OSA in clinical settings [109]. Patients presenting with the following signs and symptoms are considered as high risk and should be screened for OSA:

Table 1 Studies on the effects of CPAP on glucose metabolism in prediabetes and T2DM

Studies	Year	Number of study population	Study design	OSA definition	Baseline characteristics	Duration	Adherence (hours/night)	Glucose parameters measured	Findings
Prediabetes									
Weinstock et al. [110]	2012	50 adults (CPAP/sham: 25 and sham/CPAP: 25) with IGT (2-h OGTT ≥140 mg/dL)	Crossover	AHI ≥15		8-weeks	CPAP: 4.8 Sham: 3.4	Fasting and 2-h glucose, fasting, and 2-h insulin, insulin sensitivity (Gutt index), HOMA	No difference in glucose parameters. No reversal of IGT. Insulin sensitivity and 2-h insulin level improved only in severe OSA (AHI ≥30)
Pamidi et al. [111]	2015	39 (CPAP: 26 and placebo: 13) with prediabetes (FPG 100–125 or 2-h glucose 140–199 mg/dL, or both)	Parallel group	AHI ≥5		2 weeks	CPAP: 8	Fasting and 2-h glucose and insulin, insulin, AUC glucose, and insulin (OGTT); insulin sensitivity (IVGTT)	Improvement in insulin sensitivity and AUC glucose but no differences in other parameters
T2DM									
West et al. [112]	2007	42 (CPAP: 20 and control: 22)	Parallel group	ODI ≥10	CPAP: HbA1c 8.5%. Sham: HbA1c 8.4%	3 months	CPAP: 3.3 Sham: 3.5	HbA1c, insulin sensitivity by HOMA, and euglycemic hyperinsulinemia clamp	No difference in glucose parameters but improved sleepiness
Myhill et al. [113]	2012	44 early (i.e., 1 week) or late (i.e., 1 to 2 months) CPAP start	Parallel group	AHI ≥15	CPAP: HbA1c 6.9% (9.3% diet controlled, 62.8% OHA, 27.9% insulin and OHA)	3 months	CPAP: 5.4	HbA1c	No difference in glucose parameters. Significant reduction in SBP and DBP (9 and 7 mmHg, respectively)
Shaw et al. [114]	2016	256 (CPAP: 119 and usual care: 137)	Parallel group	ODI ≥15	CPAP: HbA1c 7.3% (47% diet controlled, 53% medications). Usual care: HbA1c 7.3% (54% diet controlled, 46% medications)	6 months	CPAP: 4.3 (at 3 months) and 4.9 (at 6 months)	HbA1c, fasting glucose	No difference; decreased DBP in adherent group, improved QOL, and decreased sleepiness
Martinez-Ceron et al. [115]	2016	50 (CPAP: 26 and untreated: 24)	Parallel group	AHI ≥5	CPAP: HbA1c, 7.6%. No treatment: HbA1c, 7.6% (58% OHA, 36% insulin, 6% OHA and insulin)	6 months	CPAP: 5.2	HbA1c, fasting glucose and insulin, insulin sensitivity (HOMA and QUICKI)	Decreased HbA1c levels, mean difference, 0.4% Decreased fasting insulin levels Improved insulin sensitivity decreased IL-1b, IL-6, and adiponectin
Mokhlesi et al. [116]	2016	19 (CPAP:13 and sham: 6)	Parallelgroup	AHI ≥5	CPAP: HbA1c, 7.3% (46% diet controlled, 54% OHA) Sham: HbA1c, 7.0% (33% diet controlled, 66% OHA)	1 week	CPAP: 7.9 Sham: 7.9	Plasma glucose measured by 24-h blood sampling	Decreased plasma glucose, predominantly at night and morning fasting, reduced serum insulin (nonsignificant trend)

Table 1 (continued)

Studies	Year	Number of study population	Study design	OSA definition	Baseline characteristics	Duration	Adherence (hours/night)	Glucose parameters measured	Findings
Morariu et al. [117]	2017	23 (CPAP:12 and sham: 11)	Parallel group	Previously untreated OSA	CPAP: HbA1c 6.6%Sham: HbA1c 6.9% (OHA only)	1 month	CPAP: 4.1 Sham: 4.5	Fructosamine, 24-h interstitial glucose profile by continuous glucose monitoring for 3 days	Significant reduction in fructosamine No difference in 24-h glucose profile
Lam et al. [118]	2017	64 (CPAP: 32 and untreated: 32)	Parallel group	AHI ≥ 15	CPAP: HbA1c 8.1% (78% OHA, 22% OHA and insulin). No treatment: HbA1c 8.4% (62% OHA, 38% OHA and insulin)	3 months	CPAP: 2.5	HbA1c, fasting glucose	No difference in glucose parameters but after excluding dropouts and those with medication changes, CPAP resulted in a reduction in HbA1c of 0.4%. Significant reduction in SBP and DBP (10 and 6 mmHg, respectively)

AHI, apnea-hypopnea index; *AUC*, area under the curve; *CPAP*, continuous positive airway pressure; *DBP*, diastolic blood pressure; *FPG*, fasting plasma glucose; *HbA1c*, hemoglobin A1c; *HOMA*, homeostatic model assessment; *IGT*, impaired glucose tolerance; *IL*, interleukin; *IVGTT*, IV glucose tolerance test; *ODI*, oxygen desaturation index; *OGTT*, oral glucose tolerance test; *OHA*, oral hypoglycemic agent; *OSA*, obstructive sleep apnea; *QOL*, quality of life; *QUICKI*, quantitative insulin sensitivity check index; *SBP*, systolic blood pressure; *T2DM*, type 2 diabetes mellitus

- Patients complaining of sleepiness, non-restorative sleep, fatigue or symptoms of insomnia, waking up with gasping or choking, habitual snoring, interruptions in breathing
- Upper airway evaluation showing retrognathia, high arched palate, macroglossia, tonsillar hypertrophy, enlarged uvula, and nasal abnormality
- Patients who demonstrate PSG or level 3 portable sleep test or home-based cardiorespiratory sleep tests with five or more obstructive respiratory events per hour of sleep OR with fifteen or more obstructive respiratory events per hour of sleep in the absence of symptoms
- Existing comorbidities such as hypertension, prediabetes or overt T2DM, congestive heart failure, atrial fibrillation, coronary artery disease, and cognitive dysfunction
- Patients with BMI > 22–25 kg/m^2 [119]
- Abdominal obesity (cm) in the range of > 90 for males and > 80 for females (Asian population) and waist circumference men: 78 cm, women: 72 cm
- Neck circumference—women: > 16 in.; men: > 17 in.
- Body fat cut—men: 25%, women: 30%
- Fasting plasma glucose (FPG) ≥ 100 mg/dL
- Hypertension (mmHg) ranging in ≥ 130/≥ 85
- High triglycerides levels of ≥ 150 and low levels of high-density lipoprotein (HDL)men: < 40; women: < 50

Additionally, patients with congestive heart failure, atrial fibrillation, treatment-refractory hypertension, nocturnal dysrhythmias, hypothyroidism, stroke, and pulmonary hypertension are also at increased risk of OSA and should be screened for the same [108]. Notably, candidates for bariatric surgery and individuals on high-risk jobs (machine operators, pilots, truck or bus drivers) experiencing excessive daytime somnolence should be screened for possible OSA.

Recommendations for diagnosis

Diagnosis of T2DM in patients with OSA

Diagnosis of prediabetes and T2DM is recommended in family members of patients with existing T2DM and overweight children and adolescents at the onset of puberty. Overweight individuals exhibiting signs and symptoms of OSA especially habitual snoring, witnessed apnea, and daytime sleepiness should be diagnosed for the co-existence of T2DM.

Prediabetes can be diagnosed based on the following criteria [120]:

- Impaired fasting glucose (IFG): FPG 100 mg/dL to 125 mg/dL or

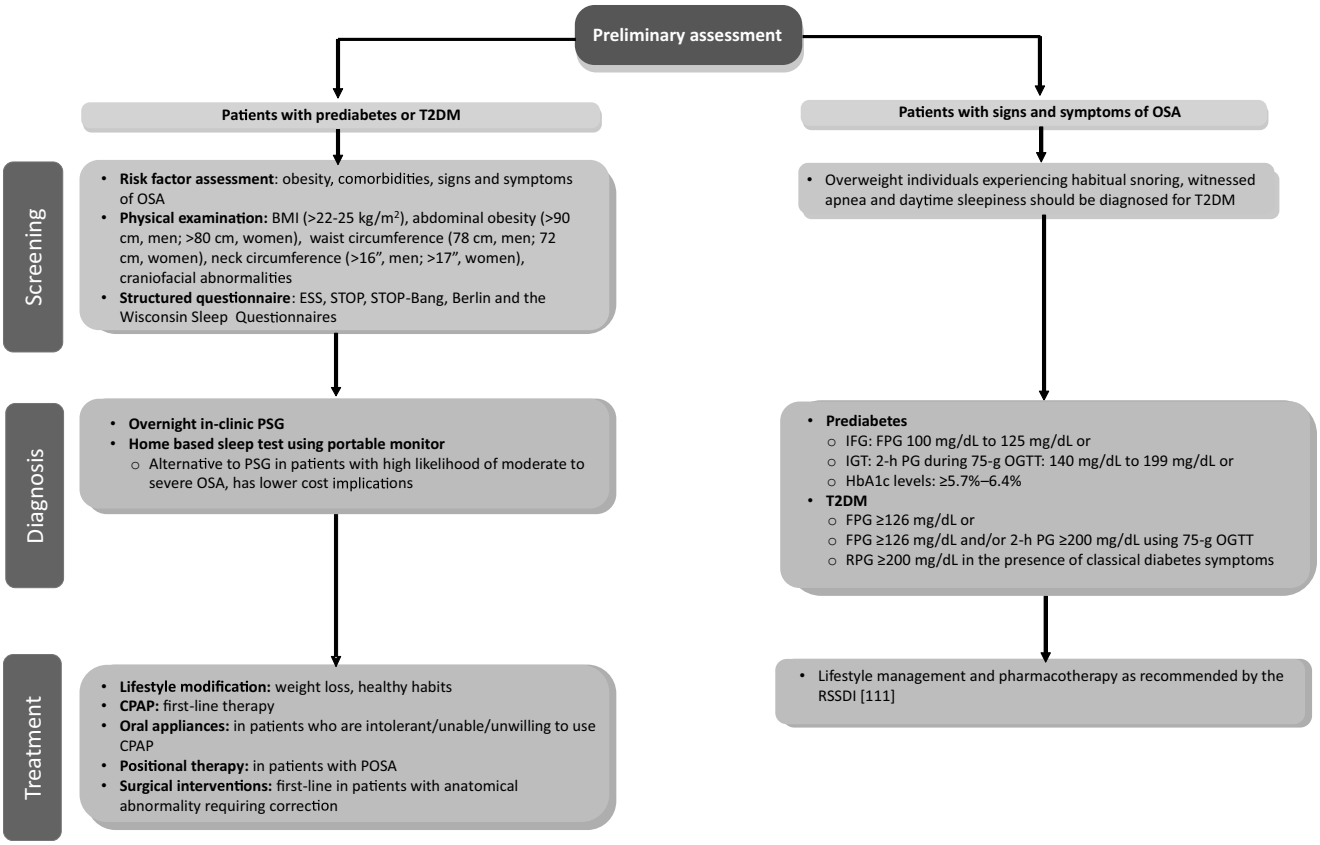


Fig. 5 Recommendations for screening, diagnosis, and treatment of OSA in patients with T2DM. AHI, apnea-hypopnea index; BMI, body mass index; CPAP, Continuous positive airway pressure; ESS, Epworth Sleepiness Scale; FPG, fasting plasma glucose; IFG, Impaired fasting glucose IGT, Impaired glucose tolerance OGTT, oral glucose tolerance test OSA, obstructive sleep apnea; POSA, Positional obstructive sleep apnea; PSG, polysomnography RPG, random plasma glucose; RSSDI, Research Society for Study of Diabetes in India; T2DM, type-2 diabetes mellitus

- Impaired glucose tolerance (IGT): 2-h plasma glucose (2-h PG) during 75-g, oral glucose tolerance test (OGTT) 140 mg/dL to 199 mg/dL or
- HbA1c levels ranging from ≥ 5.7%–6.4%

The diagnosis of T2DM should follow the following criteria [120]:

- FPG ≥ 126 mg/dL or
- FPG ≥ 126 mg/dL and/or 2-h PG ≥ 200 mg/dL using 75-g OGTT
- Random plasma glucose ≥ 200 mg/dL in the presence of classical diabetes symptoms

Diagnosis of OSA in patients with T2DM

Eligible patients (determined using questionnaire-based prediction algorithm) should be subjected to overnight monitoring using PSG by trained personnel [109, 121]. The diagnosis of OSA is to be confirmed if one of the two conditions exist [122]:

≥15 events of apnea, hypopnea, or increased respiratory effort leading to sleep arousals per hour of sleep in asymptomatic patient, with >75% of apnea/hypopnea events being obstructive	≥5 events of apnea, hypopnea, or increased respiratory effort leading to sleep arousals per hour of sleep in patients with signs and symptoms of disturbed sleep, with >75% of apnea/hypopnea events being obstructive

The AHI cut-offs for diagnosis of OSA measured on the PSG are as follows [26]:

- Mild OSA: 5 to 15 episodes/h
- Moderate OSA: 15–30 episodes/h
- Severe OSA: ≥ 30 episodes/h

Do not exclude the diagnosis of OSA based on a negative or indecisive polysomnogram. It is highly recommended to

repeat PSG especially in high-risk patients with predisposing variables [123].

As PSG is an expensive, time-consuming test requiring trained technicians, portable monitoring may be used as an alternative to diagnose OSA only in patients screened as highly probable to manifest moderate to severe OSA [124]. Testing of sleep-disordered breathing using level 3 portable devices may expedite diagnosis and considerably lower the costs associated with level 1 in-clinic PSG [19]. Level 3 portable devices have demonstrated adequate diagnostic performance compared with level 1 sleep tests in adult patients with moderate to severe OSA and having a high pretest probability with no unstable comorbidities [125]. Home-based cardiorespiratory sleep tests or cardiopulmonary may also be an acceptable approach for initial screening of patients with a suspicion of OSA, especially children or morbidly obese individuals [126, 127]. If the portable or home-based test is negative or inclusive, PSG should be performed for confirmatory diagnosis. Portable or home-based testing is discouraged in patients with comorbid neuromuscular diseases, moderate to severe pulmonary diseases, congestive heart failure, movement disorders, severe insomnia, history of stroke, sleep seizures, etc. [109, 121].

Patients diagnosed with OSA and recommended CPAP should be brought back to the sleep clinic for follow-up PSG to enable titration of CPAP pressure. If clinically suitable, a split-night protocol may be followed: part 1-diagnosis of OSA with at least 2 h of recorded sleep; part 2—titration of CPAP. This is recommended in high-risk patients with ≥ 20 events/h or ≥ 40 events/h in low-risk patients in part 1. The split-night protocol is a cost-effective approach that facilitates timely delivery of treatment [109].

Recommendations for treatment of OSA in prediabetes and T2DM

The treatment options for OSA in patients with T2DM who are at CVD risk include lifestyle modification, pharmacotherapy, and medical management, which include devices such as positioning therapy, CPAP therapy or dental appliances, and surgical interventions [128].

Lifestyle modification

- **Weight loss:** Weight loss achieved from either dietary or surgical procedures are shown to be associated with improvement in OSA severity [129, 130].

– Weight loss is considered as the primary treatment strategy for OSA in individuals who are overweight or obese and therefore, should be recommended in addition to other therapies

  Exercise and a healthy diet should be adopted for weight control.
  Bariatric surgery is another major option for patients in which weight loss through diet and exercise has failed.

- **Healthy habits:** [128]

– Alcohol and smoking cessation should be promoted to minimize further risk.
– Avoid medications that could aggravate sleep apnea, such as central nervous system depressants (i.e., opiates and benzodiazepines).

Pharmacotherapy

Role of pharmacotherapy in the management of OSA is limited [131].

- Currently, there are no widely effective pharmacotherapies to recommend for patients with OSA. However, treatment of chronic or seasonal nasal congestion could be improved with an appropriate regimen of antihistamines, decongestants, nasal steroids, and/or saline irrigation.

Medical management

Positional therapy

In this therapy, patients are maintained in a non-supine position during sleep using a positioning device, as it is associated with a higher reduction in upper airway dimension. It was found that over half of all patients with an AHI >5 events/h have a positional component to their OSA [132].

- Positional therapy can be used as a supplemental to primary therapy.

Continuous positive airway pressure

There are conflicting results on the effect of CPAP on the glucose metabolism in people with prediabetes and T2DM (Table 1**),** owing to small sample size and lack of control subjects. However, CPAP is considered as a gold standard and first line of therapy due to its favorable effects on sleep quality and quantity and subsequent prevention of T2DM and CVDs. Literature

evidence shows that patients with OSA who are highly adherent to CPAP therapy may have a greater likelihood of deriving metabolic benefit and lower risk of all-cause mortality due to CVDs [133, 134]. Moreover, the CPAP treatment has beneficial effects on quality of sleep that improves the fatigue and daytime sleepiness and consequently reduces vehicle accidents and work impairment [135].

- CPAP should be considered as a first-line of therapy.

Oral appliances

Oral appliances, such as tongue-retaining and mandibular advancement devices, works by mechanically enlarging the upper airway tract; however, these are less efficacious than CPAP therapy [66, 136].

- Oral appliances are recommended for the treatment of OSA in patients who are intolerant to or unable or unwilling to use CPAP therapy.

Surgical interventions

Surgery is performed to unblock the obstruction in the upper airway using different surgical procedures depending on the anatomic level at which obstruction occurs (nasal, upper pharyngeal, lower pharyngeal, or global upper airway) [128]. Inherent to any other surgical procedures these are limited by pre and post-operative complications [128].

- Surgery should be considered as a second-line of therapy for patients with OSA who are intolerant or experience a poor response to or CPAP therapy. However, it is considered as a first-line of therapy in pediatric patients and in those with significant anatomical abnormality requiring correction [137].

Recommendations of patient care

There is an evident need for healthcare professionals to be conscious, educated, and well-trained in the area of OSA and T2DM.

These clinical recommendations endorse IDF consensus guidelines on OSA in patients with T2DM and recommend the following:

- Healthcare professionals working on both T2DM and OSA should adopt adequate clinical practices to ensure that individual presenting with one clinical condition is considered for treatment of the other.
- Healthcare professionals should aim to develop routine interventions that are appropriate for both T2DM and OSA.

Sleep services

- Individuals with OSA must be routinely screened for markers of metabolic abnormalities and cardiovascular risk factors

  – At least waist circumference, blood pressure, fasting lipids, fasting glucose should be included as a part of the screening process.

Diabetes services

- The possibility of OSA should be considered in the assessment of all individuals with T2DM and metabolic syndrome.

  – All individuals with T2DM should be assessed for symptoms of OSA: snoring, apnea during sleep, and daytime sleepiness.

- Individuals with T2DM should be referred to a specialist at an early stage so as to diagnose and subsequent therapy owing to confirmed benefits on hypertension and quality of life.

  – Management of OSA should focus primarily on weight reduction in overweight and obese people.
  – CPAP is the current best treatment for moderate to severe OSA and should be considered where appropriate.
  – Recommend cessation of smoking and alcohol intake.
  – Recommend oral appliances or surgery where appropriate.

Recommendations for promotion of research

Further research is warranted, owing to the direct impact of OSA in T2DM on the individual's life and its economic burden on both individuals as well as society, in the following areas:

- **Epidemiological studies**

  – All data in patients with T2DM are limited with cross-sectional studies; hence, long-term nationwide

prevalence studies of OSA in adults with T2DM are warranted.

Studies in children with obesity, different ethnic groups, and adults with gestational diabetes and pre-eclampsia are of more interest.

- **Mechanistic studies**
  It is recommended that all healthcare professionals who are involved in T2DM or OSA must be educated about the links between the two conditions and trained appropriately for patient care.

– Further studies are required to better understand the biological links between OSA and T2DM, and to improve treatment and patient care, in the following areas.

  - Effect of OSA on insulin secretion, IR, mitochondrial function, and inflammatory markers
  - Newer and better biomarkers
  - Incidence and severity of micro- and macrovascular complications of T2DM in patients with OSA
  - Bidirectional link between T2DM and OSA

- **Intervention studies**

– Large randomized controlled trials of CPAP and other therapies in people with T2DM particularly prediabetes with special emphasis on CVD risk factors and outcomes and glycemic control are required.

  - Additional outcomes should also include oxidative stress, inflammation markers, and lipid metabolism.

– Studies on the weight loss interventions including the use of anti-obesity agents in people with OSA and T2DM should be investigated.
– Studies on combination therapies are also warranted.
– Therapies for OSA that are easier to use and cheaper than CPAP could be evaluated.

- **Economic analysis studies:** Studies evaluating the economic impact of OSA in patients with T2DM are warranted in terms of:

– Cost-effectiveness of OSA screening in T2DM-related outcomes.
– Cost-effectiveness of OSA treatment with CPAP on T2DM-related outcomes.

- **Resource development**
  Although PSG is the gold standard in the diagnosis of OSA, these studies are frequently limited by high costs and available resources such as hospital beds, waiting times, unwilling to stay overnight in the hospital for diagnosis, and labor requirements. In resource-limiting countries like India:

– A reliable but inexpensive diagnostic technique for OSA is needed in primary care settings.
– A precise and easy to handle clinical screening tools (i.e., home monitoring devices) are required to better diagnose and predict the severity of OSA, and for better risk stratification and to facilitate the efficiency of patient management.

Acknowledgments The preparation of this article was supported by ResMed India Pvt. Ltd. Priya Ganpathy, MPharm CMPP, provided medical writing assistance and Sangita Patil, PhD CMPP (SIRO Clinpharm Pvt. Ltd), provided additional editorial support. Manasi Date, PhD (SIRO Clinpharm Pvt. Ltd), provided support with literature search and data extraction.

Authorship contributions All named authors meet the International Committee of Medical Journal Editors (ICMJE) criteria for authorship for this article, take responsibility for the integrity of the work as a whole, and have given their approval for this version to be published.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflicts of interest.

References

1. International Diabetes Federation. IDF Diabetes Atlas teB, Belgium: International Diabetes Federation. 2017.
2. Sateia MJ. International classification of sleep disorders-third edition: highlights and modifications. Chest. 2014;146(5):1387–94. https://doi.org/10.1378/chest.14-0970.
3. Benjafield AV, Ayas NT, Eastwood PR, Heinzer R, Ip MSM, Morrell MJ, et al. Estimation of the global prevalence and burden of obstructive sleep apnoea: a literature-based analysis. Lancet Respir Med. 2019;7(8):687–98. https://doi.org/10.1016/S2213-2600(19)30198-5.
4. Raju YSSD, Yadati R, Alekhya A. Diabetes mellitus and obstructive sleep apnoea: implications for clinicians. J Clin Sci Res. 2016;5:225–33. https://doi.org/10.15380/2277-5706.JCSR.16.05.003.
5. Hla KM, Young T, Hagen EW, Stein JH, Finn LA, Nieto FJ, et al. Coronary heart disease incidence in sleep disordered breathing: the Wisconsin Sleep Cohort Study. Sleep. 2015;38(5):677–84. https://doi.org/10.5665/sleep.4654.
6. Gottlieb DJ, Yenokyan G, Newman AB, O'Connor GT, Punjabi NM, Quan SF, et al. Prospective study of obstructive sleep apnea and incident coronary heart disease and heart failure: the sleep heart health study. Circulation. 2010;122(4):352–60. https://doi.org/10.1161/CIRCULATIONAHA.109.901801.
7. Hou H, Zhao Y, Yu W, Dong H, Xue X, Ding J, et al. Association of obstructive sleep apnea with hypertension: a systematic review and meta-analysis. J Glob Health. 2018;8(1):010405. https://doi.org/10.7189/jogh.08.010405.

8. Resnick HE, Redline S, Shahar E, Gilpin A, Newman A, Walter R, et al. Diabetes and sleep disturbances: findings from the Sleep Heart Health Study. Diabetes Care. 2003;26(3):702–9. https://doi.org/10.2337/diacare.26.3.702.
9. Huang T, Lin BM, Stampfer MJ, Tworoger SS, Hu FB, Redline S. A population-based study of the bidirectional association between obstructive sleep apnea and type 2 diabetes in three prospective U.S. cohorts. Diabetes Care. 2018;41(10):2111–9. https://doi.org/10.2337/dc18-0675.
10. Bonsignore MR, Baiamonte P, Mazzuca E, Castrogiovanni A, Marrone O. Obstructive sleep apnea and comorbidities: a dangerous liaison. Multidiscip Respir Med. 2019;14:8. https://doi.org/10.1186/s40248-019-0172-9.
11. Pillar G, Shehadeh N. Abdominal fat and sleep apnea: the chicken or the egg? Diabetes Care. 2008;31(Suppl 2):S303–9. https://doi.org/10.2337/dc08-s272.
12. Ioja SCE, Ng J, et al. Obstructive sleep apnea in adults with type 1 and type 2 diabetes: perspectives from a quality improvement initiative in a university-based diabetes center. BMJ Open Diabetes Res Care. 2017;5(1):1–5. https://doi.org/10.1136/bmjdrc-2017-000433.
13. American Diabetes Association (ADA). Standards of medical care in diabetes. Diabetes Care. 2014;37:S14–80.
14. Viswanathan V, Ramalingam IP, Ramakrishnan N. High prevalence of obstructive sleep apnea among people with type 2 diabetes mellitus in a tertiary care center. J Assoc Physicians India. 2017;65(11):38–42.
15. Singh APS, Jain J, Singh R. Polysomnographic study in diabetes mellitus in central Indian subjects. J Mahatma Gandhi Inst Med Sci. 2012;17:17–21.
16. Malik JA, Masoodi SR, Shoib S. Obstructive sleep apnea in type 2 diabetes and impact of continuous positive airway pressure therapy on glycemic control. Indian J Endocrinol Metab. 2017;21(1):106–12. https://doi.org/10.4103/2230-8210.196005.
17. Ekka RSJ, Singh C, Sen MK, Gupta A. Prevalence of obstructive sleep apnea in type 2 diabetes mellitus. 2010;5:18–26.
18. Bhimwal RKMM, Jangid R, Bhati RL. To study the prevalence of obstructive sleep apnoea in type 2 diabetes patients in Western Rajasthan, India. Int J Adv Med. 2017;4(4):894–902. https://doi.org/10.18203/2349-3933.ijam20172569.
19. Sharma SK, Katoch VM, Mohan A, Kadhiravan T, Elavarasi A, Ragesh R, et al. Consensus and evidence-based Indian initiative on obstructive sleep apnea guidelines 2014 (first edition). Lung India. 2015;32(4):422–34. https://doi.org/10.4103/0970-2113.159677.
20. Siwasaranond N, Nimitphong H, Manodpitipong A, Saetung S, Chirakalwasan N, Thakkinstian A, et al. The relationship between diabetes-related complications and obstructive sleep apnea in type 2 diabetes. J Diabetes Res. 2018;2018:9269170. https://doi.org/10.1155/2018/9269170.
21. Eckert DJ, Malhotra A. Pathophysiology of adult obstructive sleep apnea. Proc Am Thorac Soc. 2008;5(2):144–53. https://doi.org/10.1513/pats.200707-114MG.
22. Levy P, Kohler M, McNicholas WT, Barbe F, McEvoy RD, Somers VK, et al. Obstructive sleep apnoea syndrome. Nat Rev Dis Primers. 2015;1:15015. https://doi.org/10.1038/nrdp.2015.15.
23. Laratta CR, Ayas NT, Povitz M, Pendharkar SR. Diagnosis and treatment of obstructive sleep apnea in adults. CMAJ. 2017;189(48):E1481–E8. https://doi.org/10.1503/cmaj.170296.
24. Semelka M, Wilson J, Floyd R. Diagnosis and treatment of obstructive sleep apnea in adults. Am Fam Physician. 2016;94(5):355–60.
25. Stansbury RC, Strollo PJ. Clinical manifestations of sleep apnea. J Thorac Dis. 2015;7(9):E298–310. https://doi.org/10.3978/j.issn.2072-1439.2015.09.13.
26. Amra B, Rahmati B, Soltaninejad F, Feizi A. Screening questionnaires for obstructive sleep apnea: an updated systematic review. Oman Med J. 2018;33(3):184–92. https://doi.org/10.5001/omj.2018.36.
27. Punjabi NM, Shahar E, Redline S, Gottlieb DJ, Givelber R, Resnick HE, et al. Sleep-disordered breathing, glucose intolerance, and insulin resistance: the Sleep Heart Health Study. Am J Epidemiol. 2004;160(6):521–30. https://doi.org/10.1093/aje/kwh261.
28. Bhat SUH, DeBari VA, Ahmad M, Polos PG, Chokroverty S. The utility of patient-completed and partner-completed Epworth Sleepiness Scale scores in the evaluation of obstructive sleep apnea. Sleep Breath. 2016;20(4):1347–54. https://doi.org/10.1007/s11325-016-1370-8.
29. Prasad KTSI, Agarwal R, Aggarwal AN, Behera D, Dhooria S. Assessing the likelihood of obstructive sleep apnea: a comparison of nine screening questionnaires. Sleep Breath. 2017;21(4):909–17. https://doi.org/10.1007/s11325-017-1495-4.
30. Johns MW. A new method for measuring daytime sleepiness: the Epworth Sleepiness Scale. Sleep. 1991;14(6):540–5. https://doi.org/10.1093/sleep/14.6.540.
31. Chung F, Yegneswaran B, Liao P, Chung SA, Vairavanathan S, Islam S, et al. STOP questionnaire: a tool to screen patients for obstructive sleep apnea. Anesthesiology. 2008;108(5):812–21. https://doi.org/10.1097/ALN.0b013e31816d83e4.
32. Chung F, Subramanyam R, Liao P, Sasaki E, Shapiro C, Sun Y. High STOP-Bang score indicates a high probability of obstructive sleep apnoea. Br J Anaesth. 2012;108(5):768–75. https://doi.org/10.1093/bja/aes022.
33. Netzer NC, Stoohs RA, Netzer CM, Clark K, Strohl KP. Using the Berlin questionnaire to identify patients at risk for the sleep apnea syndrome. Ann Intern Med. 1999;131(7):485–91. https://doi.org/10.7326/0003-4819-131-7-199910050-00002.
34. Young T, Palta M, Dempsey J, Skatrud J, Weber S, Badr S. The occurrence of sleep-disordered breathing among middle-aged adults. N Engl J Med. 1993;328(17):1230–5. https://doi.org/10.1056/NEJM199304293281704.
35. Force USPST, Bibbins-Domingo K, Grossman DC, Curry SJ, Davidson KW, Epling JW Jr, et al. Screening for obstructive sleep apnea in adults: US Preventive Services Task Force recommendation statement. JAMA. 2017;317(4):407–14. https://doi.org/10.1001/jama.2016.20325.
36. Comparative effectiveness of diagnosis and treatment of obstructive sleep apnea in adults. Comparative effectiveness review summary guides for clinicians. AHRQ comparative effectiveness reviews. Rockville (MD) 2007.
37. Reichmuth KJ, Austin D, Skatrud JB, Young T. Association of sleep apnea and type II diabetes: a population-based study. Am J Respir Crit Care Med. 2005;172(12):1590–5. https://doi.org/10.1164/rccm.200504-637OC.
38. Marshall NS, Wong KK, Phillips CL, Liu PY, Knuiman MW, Grunstein RR. Is sleep apnea an independent risk factor for prevalent and incident diabetes in the Busselton Health Study? J Clin Sleep Med. 2009;5(1):15–20.
39. Rajan P, Greenberg H. Obstructive sleep apnea as a risk factor for type 2 diabetes mellitus. Nat Sci Sleep. 2015;7:113–25. https://doi.org/10.2147/NSS.S90835.
40. Qie R, Zhang D, Liu L, Ren Y, Zhao Y, Liu D, et al. Obstructive sleep apnea and risk of type 2 diabetes mellitus: a systematic review and dose-response meta-analysis of cohort studies. J Diabetes. 2019;12:455–64. https://doi.org/10.1111/1753-0407.13017.
41. Elmasry A, Janson C, Lindberg E, Gislason T, Tageldin MA, Boman G. The role of habitual snoring and obesity in the development of diabetes: a 10-year follow-up study in a male

population. J Intern Med. 2000;248(1):13–20. https://doi.org/10.1046/j.1365-2796.2000.00683.x.
42. Al-Delaimy WK, Manson JE, Willett WC, Stampfer MJ, Hu FB. Snoring as a risk factor for type II diabetes mellitus: a prospective study. Am J Epidemiol. 2002;155(5):387–93. https://doi.org/10.1093/aje/155.5.387.
43. Koren D, O'Sullivan KL, Mokhlesi B. Metabolic and glycemic sequelae of sleep disturbances in children and adults. Curr Diabetes Rep. 2015;15(1):562. https://doi.org/10.1007/s11892-014-0562-5.
44. Pugliese G, Barrea L, Laudisio D, Salzano C, Aprano S, Colao A, et al. Sleep apnea, obesity, and disturbed glucose homeostasis: epidemiologic evidence, biologic insights, and therapeutic strategies. Curr Obes Rep. 2020;9(1):30–8. https://doi.org/10.1007/s13679-020-00369-y.
45. Viswanathan V, Ramakrishnan N, Sunaina S, Vigneswari A, Satyavani K. Subjects with type 2 diabetes may have obstructive sleep apnoea even at lower BMI values. Indian J Sleep Med. 2012;7(2):45–7.
46. Reddy EV, Kadhiravan T, Mishra HK, Sreenivas V, Handa KK, Sinha S, et al. Prevalence and risk factors of obstructive sleep apnea among middle-aged urban Indians: a community-based study. Sleep Med. 2009;10(8):913–8. https://doi.org/10.1016/j.sleep.2008.08.011.
47. Peppard PE, Young T, Palta M, Dempsey J, Skatrud J. Longitudinal study of moderate weight change and sleep-disordered breathing. JAMA. 2000;284(23):3015–21. https://doi.org/10.1001/jama.284.23.3015.
48. Mok Y, Tan CW, Wong HS, How CH, Tan KL, Hsu PP. Obstructive sleep apnoea and Type 2 diabetes mellitus: are they connected? Singap Med J. 2017;58(4):179–83. https://doi.org/10.11622/smedj.2017027.
49. Taheri S, Lin L, Austin D, Young T, Mignot E. Short sleep duration is associated with reduced leptin, elevated ghrelin, and increased body mass index. PLoS Med. 2004;1(3):e62. https://doi.org/10.1371/journal.pmed.0010062.
50. Foster GD, Sanders MH, Millman R, Zammit G, Borradaile KE, Newman AB, et al. Obstructive sleep apnea among obese patients with type 2 diabetes. Diabetes Care. 2009;32(6):1017–9. https://doi.org/10.2337/dc08-1776.
51. Wang X, Greer J, Porter RR, Kaur K, Youngstedt SD. Short-term moderate sleep restriction decreases insulin sensitivity in young healthy adults. Sleep Health. 2016;2(1):63–8. https://doi.org/10.1016/j.sleh.2015.11.004.
52. Khandelwal D, Dutta D, Chittawar S, Kalra S. Sleep disorders in type 2 diabetes. Indian J Endocrinol Metab. 2017;21(5):758–61. https://doi.org/10.4103/ijem.IJEM_156_17.
53. Mussa BM, Schauman M, Kumar V, Skaria S, Abusnana S. Personalized intervention to improve stress and sleep patterns for glycemic control and weight management in obese Emirati patients with type 2 diabetes: a randomized controlled clinical trial. Diabetes Metab Syndr Obes. 2019;12:991–9. https://doi.org/10.2147/DMSO.S201142.
54. Foster GD, Borradaile KE, Sanders MH, Millman R, Zammit G, Newman AB, et al. A randomized study on the effect of weight loss on obstructive sleep apnea among obese patients with type 2 diabetes: the Sleep AHEAD study. Arch Intern Med. 2009;169(17):1619–26. https://doi.org/10.1001/archinternmed.2009.266.
55. Mesman RLS, Calleja F, Hendriks G, Morolli B, Misovic B, Devilee P, et al. The functional impact of variants of uncertain significance in BRCA2. Genet Med. 2019;21(2):293–302. https://doi.org/10.1038/s41436-018-0052-2.
56. Calik MW. Treatments for obstructive sleep apnea. Journal of clinical outcomes management. J Clin Outcomes Manag. 2016;23(4):181–92.
57. Stasche N. Selective indication for positive airway pressure (PAP) in sleep-related breathing disorders with obstruction. GMS Curr Top Otorhinolaryngol Head Neck Surg. 2006;5:Doc06.
58. Sutherland K, Vanderveken OM, Tsuda H, Marklund M, Gagnadoux F, Kushida CA, et al. Oral appliance treatment for obstructive sleep apnea: an update. J Clin Sleep Med. 2014;10(2):215–27. https://doi.org/10.5664/jcsm.3460.
59. Morgenthaler TI, Aurora RN, Brown T, Zak R, Alessi C, Boehlecke B, et al. Practice parameters for the use of autotitrating continuous positive airway pressure devices for titrating pressures and treating adult patients with obstructive sleep apnea syndrome: an update for 2007. An American Academy of Sleep Medicine report. Sleep. 2008;31(1):141–7. https://doi.org/10.1093/sleep/31.1.141.
60. Hoffstein V. Review of oral appliances for treatment of sleep-disordered breathing. Sleep Breath 2007;11(1):1–22. https://doi.org/10.1007/s11325-006-0084-8, 1.
61. Gagnadoux F, Fleury B, Vielle B, Petelle B, Meslier N, N'Guyen XL, et al. Titrated mandibular advancement versus positive airway pressure for sleep apnoea. Eur Respir J. 2009;34(4):914–20. https://doi.org/10.1183/09031936.00148208.
62. Dieltjens M, Vanderveken O. Oral appliances in obstructive sleep apnea. Healthcare (Basel). 2019;7(4). https://doi.org/10.3390/healthcare7040141.
63. Zaghi S, Holty JE, Certal V, Abdullatif J, Guilleminault C, Powell NB, et al. Maxillomandibular advancement for treatment of obstructive sleep apnea: a meta-analysis. JAMA Otolaryngol Head Neck Surg. 2016;142(1):58–66. https://doi.org/10.1001/jamaoto.2015.2678.
64. Remmers J, Charkhandeh S, Grosse J, Topor Z, Brant R, Santosham P, et al. Remotely controlled mandibular protrusion during sleep predicts therapeutic success with oral appliances in patients with obstructive sleep apnea. Sleep. 2013;36(10):1517–25,25A. https://doi.org/10.5665/sleep.3048.
65. Deane SA, Cistulli PA, Ng AT, Zeng B, Petocz P, Darendeliler MA. Comparison of mandibular advancement splint and tongue stabilizing device in obstructive sleep apnea: a randomized controlled trial. Sleep. 2009;32(5):648–53. https://doi.org/10.1093/sleep/32.5.648.
66. Ramar K, Dort LC, Katz SG, Lettieri CJ, Harrod CG, Thomas SM, et al. Clinical practice guideline for the treatment of obstructive sleep apnea and snoring with oral appliance therapy: an update for 2015. J Clin Sleep Med. 2015;11(7):773–827. https://doi.org/10.5664/jcsm.4858.
67. Li HY, Lee LA, Hsin LJ, Fang TJ, Lin WN, Chen HC, et al. Intrapharyngeal surgery with integrated treatment for obstructive sleep apnea. Biom J. 2019;42(2):84–92. https://doi.org/10.1016/j.bj.2019.02.002.
68. Kahn SE, Hull RL, Utzschneider KM. Mechanisms linking obesity to insulin resistance and type 2 diabetes. Nature. 2006;444(7121):840–6. https://doi.org/10.1038/nature05482.
69. Cizza G, de Jonge L, Piaggi P, Mattingly M, Zhao X, Lucassen E, et al. Neck circumference is a predictor of metabolic syndrome and obstructive sleep apnea in short-sleeping obese men and women. Metab Syndr Relat Disord. 2014;12(4):231–41. https://doi.org/10.1089/met.2013.0093.
70. Jehan S, Myers AK, Zizi F, Pandi-Perumal SR, Jean-Louis G, McFarlane SI. Obesity, obstructive sleep apnea and type 2 diabetes mellitus: epidemiology and pathophysiologic insights. Sleep Med Disord. 2018;2(3):52–8.
71. Doumit J, Prasad B. Sleep apnea in type 2 diabetes. Diabetes Spectr. 2016;29(1):14–9. https://doi.org/10.2337/diaspect.29.1.14.
72. Vgontzas AN, Papanicolaou DA, Bixler EO, Kales A, Tyson K, Chrousos GP. Elevation of plasma cytokines in disorders of excessive daytime sleepiness: role of sleep disturbance and obesity. J

Clin Endocrinol Metab. 1997;82(5):1313–6. https://doi.org/10.1210/jcem.82.5.3950.
73. Eckel RH, Grundy SM, Zimmet PZ. The metabolic syndrome. Lancet. 2005;365(9468):1415–28. https://doi.org/10.1016/S0140-6736(05)66378-7.
74. Xu S, Wan Y, Xu M, Ming J, Xing Y, An F, et al. The association between obstructive sleep apnea and metabolic syndrome: a systematic review and meta-analysis. BMC Pulm Med. 2015;15:105. https://doi.org/10.1186/s12890-015-0102-3.
75. Basoglu OK, Sarac F, Sarac S, Uluer H, Yilmaz C. Metabolic syndrome, insulin resistance, fibrinogen, homocysteine, leptin, and C-reactive protein in obese patients with obstructive sleep apnea syndrome. Ann Thorac Med. 2011;6(3):120–5. https://doi.org/10.4103/1817-1737.82440.
76. Soin D, Kumar PA, Chahal J, Chawla SPS, Kaur S, Garg R, et al. Evaluation of obstructive sleep apnea in metabolic syndrome. J Family Med Prim Care. 2019;8(5):1580–6. https://doi.org/10.4103/jfmpc.jfmpc_175_19.
77. Jun J, Polotsky VY. Metabolic consequences of sleep-disordered breathing. ILAR J. 2009;50(3):289–306. https://doi.org/10.1093/ilar.50.3.289.
78. SM TEaM. Obstructive sleep apnea and metabolic syndrome: alterations in glucose metabolism and inflammation. Proc Am Thorac Soc. 2008;5:207–17. https://doi.org/10.1513/pats.200708-139MG.
79. Balkau B, Vol S, Loko S, Andriamboavonjy T, Lantieri O, Gusto G, et al. High baseline insulin levels associated with 6-year incident observed sleep apnea. Diabetes Care. 2010;33(5):1044–9. https://doi.org/10.2337/dc09-1901.
80. Newman AB, Foster G, Givelber R, Nieto FJ, Redline S, Young T. Progression and regression of sleep-disordered breathing with changes in weight: the sleep heart health study. Arch Intern Med. 2005;165(20):2408–13. https://doi.org/10.1001/archinte.165.20.2408.
81. Licinio J, Wong ML. Sequence and function in pharmacogenomics. Pharm J. 2003;3(3):123. https://doi.org/10.1038/sj.tpj.6500185.
82. Tantucci C, Scionti L, Bottini P, Dottorini ML, Puxeddu E, Casucci G, et al. Influence of autonomic neuropathy of different severities on the hypercapnic drive to breathing in diabetic patients. Chest. 1997;112(1):145–53. https://doi.org/10.1378/chest.112.1.145.
83. Kent BD, Grote L, Ryan S, Pepin JL, Bonsignore MR, Tkacova R, et al. Diabetes mellitus prevalence and control in sleep-disordered breathing: the European Sleep Apnea Cohort (ESADA) study. Chest. 2014;146(4):982–90. https://doi.org/10.1378/chest.13-2403.
84. Papanas N, Steiropoulos P, Nena E, Tzouvelekis A, Maltezos E, Trakada G, et al. HbA1c is associated with severity of obstructive sleep apnea hypopnea syndrome in nondiabetic men. Vasc Health Risk Manag. 2009;5:751–6. https://doi.org/10.2147/vhrm.s7057.
85. Pillai AWG, Gunathilake W, Idris I. Control in patients with type 2 diabetes prior to continuous positive airway pressure treatment. Diabetes Technol Ther. 2011;13:945–9. https://doi.org/10.1089/dia.2011.0005.
86. Aronsohn RS, Whitmore H, Van Cauter E, Tasali E. Impact of untreated obstructive sleep apnea on glucose control in type 2 diabetes. Am J Respir Crit Care Med. 2010;181(5):507–13. https://doi.org/10.1164/rccm.200909-1423OC.
87. Tsai YW, Kann NH, Tung TH, Chao YJ, Lin CJ, Chang KC, et al. Impact of subjective sleep quality on glycemic control in type 2 diabetes mellitus. Fam Pract. 2012;29(1):30–5. https://doi.org/10.1093/fampra/cmr041.
88. Tahrani AA. Obstructive sleep apnoea in diabetes: does it matter? Diab Vasc Dis Res. 2017;14(5):454–62. https://doi.org/10.1177/1479164117714397.
89. Altaf QA, Dodson P, Ali A, Raymond NT, Wharton H, Fellows H, et al. Obstructive sleep apnea and retinopathy in patients with type 2 diabetes. A longitudinal study. Am J Respir Crit Care Med. 2017;196(7):892–900. https://doi.org/10.1164/rccm.201701-0175OC.
90. Mason RH, West SD, Kiire CA, Groves DC, Lipinski HJ, Jaycock A, et al. High prevalence of sleep disordered breathing in patients with diabetic macular edema. Retina. 2012;32(9):1791–8. https://doi.org/10.1097/IAE.0b013e318259568b.
91. Fujihara K, Kodama S, Horikawa C, Yoshizawa S, Sugawara A, Hirasawa R, et al. The relationship between diabetic neuropathy and sleep apnea syndrome: a meta-analysis. Sleep Disord. 2013;2013:150371. https://doi.org/10.1155/2013/150371.
92. Tahrani AA, Ali A, Raymond NT, Begum S, Dubb K, Altaf QA, et al. Obstructive sleep apnea and diabetic nephropathy: a cohort study. Diabetes Care. 2013;36(11):3718–25. https://doi.org/10.2337/dc13-0450.
93. Seicean S, Strohl KP, Seicean A, Gibby C, Marwick TH. Sleep disordered breathing as a risk of cardiac events in subjects with diabetes mellitus and normal exercise echocardiographic findings. Am J Cardiol. 2013;111(8):1214–20. https://doi.org/10.1016/j.amjcard.2012.12.053.
94. Rivas M, Ratra A, Nugent K. Obstructive sleep apnea and its effects on cardiovascular diseases: a narrative review. Anatol J Cardiol. 2015;15(11):944–50. https://doi.org/10.5152/AnatolJCardiol.2015.6607.
95. Nieto FJ, Young TB, Lind BK, Shahar E, Samet JM, Redline S, et al. Association of sleep-disordered breathing, sleep apnea, and hypertension in a large community-based study. Sleep Heart Health Study. JAMA. 2000;283(14):1829–36. https://doi.org/10.1001/jama.283.14.1829.
96. Young T, Peppard P, Palta M, Hla KM, Finn L, Morgan B, et al. Population-based study of sleep-disordered breathing as a risk factor for hypertension. Arch Intern Med. 1997;157(15):1746–52.
97. Hla KM, Young T, Finn L, Peppard PE, Szklo-Coxe M, Stubbs M. Longitudinal association of sleep-disordered breathing and nondipping of nocturnal blood pressure in the Wisconsin Sleep Cohort Study. Sleep. 2008;31(6):795–800. https://doi.org/10.1093/sleep/31.6.795.
98. Yaggi HK, Concato J, Kernan WN, Lichtman JH, Brass LM, Mohsenin V. Obstructive sleep apnea as a risk factor for stroke and death. N Engl J Med. 2005;353(19):2034–41. https://doi.org/10.1056/NEJMoa043104.
99. Tung P, Levitzky YS, Wang R, Weng J, Quan SF, Gottlieb DJ, et al. Obstructive and central sleep apnea and the risk of incident atrial fibrillation in a community cohort of men and women. J Am Heart Assoc. 2017;6(7). https://doi.org/10.1161/JAHA.116.004500.
100. Kylintireas I, Craig S, Nethononda R, Kohler M, Francis J, Choudhury R, Stradling J, Neubauer S. Atherosclerosis and arterial stiffness in obstructive sleep apnea–a cardiovascular magnetic resonance study. Atherosclerosis. 2012;222(2):483–489. https://doi.org/10.1016/j.atherosclerosis.2012.03.036.
101. Kawano Y, Tamura A, Kadota J. Association between the severity of obstructive sleep apnea and the ratio of low-density lipoprotein cholesterol to high-density lipoprotein cholesterol. Metabolism. 2012;61(2):186–92. https://doi.org/10.1016/j.metabol.2011.06.004.
102. Doonan RJ, Scheffler P, Lalli M, Kimoff RJ, Petridou ET, Daskalopoulos ME, et al. Increased arterial stiffness in obstructive sleep apnea: a systematic review. Hypertens Res. 2011;34(1):23–32. https://doi.org/10.1038/hr.2010.200.
103. Selim BJ, Koo BB, Qin L, Jeon S, Won C, Redeker NS, et al. The association between nocturnal cardiac arrhythmias and sleep-disordered breathing: the DREAM study. J Clin Sleep Med. 2016;12(6):829–37. https://doi.org/10.5664/jcsm.5880.

104. Banno K, Ramsey C, Walld R, Kryger MH. Expenditure on health care in obese women with and without sleep apnea. Sleep. 2009;32(2):247–52. https://doi.org/10.1093/sleep/32.2.247.
105. Seetho IWWJ. Screening for obstructive sleep apnoea in obesity and diabetes – potential for future approaches. Eur J Clin Investig. 2013;43(6):640–55.
106. Misra A, Chowbey P, Makkar BM, Vikram NK, Wasir JS, Chadha D, et al. Consensus statement for diagnosis of obesity, abdominal obesity and the metabolic syndrome for Asian Indians and recommendations for physical activity, medical and surgical management. J Assoc Physicians India. 2009;57:163–70.
107. Edmonds PJ, Gunasekaran K, Edmonds LC. Neck grasp predicts obstructive sleep apnea in type 2 diabetes mellitus. Sleep Disord. 2019;2019:3184382. https://doi.org/10.1155/2019/3184382.
108. Epstein LJ, Kristo D, Strollo PJ Jr, Friedman N, Malhotra A, Patil SP, et al. Clinical guideline for the evaluation, management and long-term care of obstructive sleep apnea in adults. J Clin Sleep Med. 2009;5(3):263–76.
109. Foroughi M, Razavi H, Malekmohammad M, Adimi Naghan P, Jamaati H. Diagnosis of obstructive sleep apnea syndrome in adults: a brief review of existing data for practice in Iran. Tanaffos. 2016;15(2):70–4.
110. Weinstock TG, Wang X, Rueschman M, Ismail-Beigi F, Aylor J, Babineau DC, et al. A controlled trial of CPAP therapy on metabolic control in individuals with impaired glucose tolerance and sleep apnea. Sleep. 2012;35(5):617–25B. https://doi.org/10.5665/sleep.1816.
111. Pamidi S, Wroblewski K, Stepien M, Sharif-Sidi K, Kilkus J, Whitmore H, et al. Eight hours of nightly continuous positive airway pressure treatment of obstructive sleep apnea improves glucose metabolism in patients with Prediabetes. A randomized controlled trial. Am J Respir Crit Care Med. 2015;192(1):96–105. https://doi.org/10.1164/rccm.201408-1564OC.
112. West SD, Nicoll DJ, Wallace TM, Matthews DR, Stradling JR. Effect of CPAP on insulin resistance and HbA1c in men with obstructive sleep apnoea and type 2 diabetes. Thorax. 2007;62(11):969–74. https://doi.org/10.1136/thx.2006.074351.
113. Myhill PC, Davis WA, Peters KE, Chubb SA, Hillman D, Davis TM. Effect of continuous positive airway pressure therapy on cardiovascular risk factors in patients with type 2 diabetes and obstructive sleep apnea. J Clin Endocrinol Metab. 2012;97(11):4212–8. https://doi.org/10.1210/jc.2012-2107.
114. Shaw JE, Punjabi NM, Naughton MT, Willes L, Bergenstal RM, Cistulli PA, et al. The effect of treatment of obstructive sleep apnea on glycemic control in type 2 diabetes. Am J Respir Crit Care Med. 2016;194(4):486–92. https://doi.org/10.1164/rccm.201511-2260OC.
115. Martinez-Ceron E, Barquiel B, Bezos AM, Casitas R, Galera R, Garcia-Benito C, et al. Effect of continuous positive airway pressure on glycemic control in patients with obstructive sleep apnea and type 2 diabetes. A randomized clinical trial. Am J Respir Crit Care Med. 2016;194(4):476–85. https://doi.org/10.1164/rccm.201510-1942OC.
116. Mokhlesi B, Grimaldi D, Beccuti G, Abraham V, Whitmore H, Delebecque F, et al. Effect of one week of 8-hour nightly continuous positive airway pressure treatment of obstructive sleep apnea on glycemic control in type 2 diabetes: a proof-of-concept study. Am J Respir Crit Care Med. 2016;194(4):516–9. https://doi.org/10.1164/rccm.201602-0396LE.
117. Morariu EM, Chasens ER, Strollo PJ Jr, Korytkowski M. Effect of continuous positive airway pressure (CPAP) on glycemic control and variability in type 2 diabetes. Sleep Breath. 2017;21(1):145–7. https://doi.org/10.1007/s11325-016-1388-y.
118. Lam JCM, Lai AYK, Tam TCC, Yuen MMA, Lam KSL, Ip MSM. CPAP therapy for patients with sleep apnea and type 2 diabetes mellitus improves control of blood pressure. Sleep Breath. 2017;21(2):377–86. https://doi.org/10.1007/s11325-016-1428-7.
119. Consultation WHOE. Appropriate body-mass index for Asian populations and its implications for policy and intervention strategies. Lancet. 2004;363(9403):157–63. https://doi.org/10.1016/S0140-6736(03)15268-3.
120. Chawla R, Madhu SV, Makkar BM, Ghosh S, Saboo B, Kalra S, et al. RSSDI-ESI clinical practice recommendations for the management of type 2 diabetes mellitus 2020. Indian J Endocrinol Metab. 2020;24(1):1–122. https://doi.org/10.4103/ijem.IJEM_225_20.
121. Kapur VK, Auckley DH, Chowdhuri S, Kuhlmann DC, Mehra R, Ramar K, et al. Clinical practice guideline for diagnostic testing for adult obstructive sleep apnea: an Ac clinical practice guideline. J Clin Sleep Med. 2017;13(3):479–504. https://doi.org/10.5664/jcsm.6506.
122. Littner MR, Kushida C, Wise M, Davila DG, Morgenthaler T, Lee-Chiong T, et al. Practice parameters for clinical use of the multiple sleep latency test and the maintenance of wakefulness test. Sleep. 2005;28(1):113–21. https://doi.org/10.1093/sleep/28.1.113.
123. Levendowski DJ, Zack N, Rao S, Wong K, Gendreau M, Kranzler J, et al. Assessment of the test-retest reliability of laboratory polysomnography. Sleep Breath. 2009;13(2):163–7. https://doi.org/10.1007/s11325-008-0214-6.
124. Collop NA, Anderson WM, Boehlecke B, Claman D, Goldberg R, Gottlieb DJ, et al. Clinical guidelines for the use of unattended portable monitors in the diagnosis of obstructive sleep apnea in adult patients. Portable Monitoring Task Force of the American Academy of Sleep Medicine. J Clin Sleep Med. 2007;3(7):737–47.
125. El Shayeb M, Topfer LA, Stafinski T, Pawluk L, Menon D. Diagnostic accuracy of level 3 portable sleep tests versus level 1 polysomnography for sleep-disordered breathing: a systematic review and meta-analysis. CMAJ. 2014;186(1):E25–51. https://doi.org/10.1503/cmaj.130952.
126. Calleja JM, Esnaola S, Rubio R, Duran J. Comparison of a cardiorespiratory device versus polysomnography for diagnosis of sleep apnoea. Eur Respir J. 2002;20(6):1505–10. https://doi.org/10.1183/09031936.02.00297402.
127. Kingshott RN, Gahleitner F, Elphick HE, Gringras P, Farquhar M, Pickering RM, et al. Cardiorespiratory sleep studies at home: experience in research and clinical cohorts. Arch Dis Child. 2019;104(5):476–81. https://doi.org/10.1136/archdischild-2018-315676.
128. Tietjens JR, Claman D, Kezirian EJ, De Marco T, Mirzayan A, Sadroonri B, et al. Obstructive sleep apnea in cardiovascular disease: a review of the literature and proposed multidisciplinary clinical management strategy. J Am Heart Assoc. 2019;8(1):e010440. https://doi.org/10.1161/JAHA.118.010440.
129. Ashrafian H, Toma T, Rowland SP, Harling L, Tan A, Efthimiou E, et al. Bariatric surgery or non-surgical weight loss for obstructive sleep apnoea? A systematic review and comparison of meta-analyses. Obes Surg. 2015;25(7):1239–50. https://doi.org/10.1007/s11695-014-1533-2.
130. Kuna ST, Reboussin DM, Borradaile KE, Sanders MH, Millman RP, Zammit G, et al. Long-term effect of weight loss on obstructive sleep apnea severity in obese patients with type 2 diabetes. Sleep. 2013;36(5):641–9A. https://doi.org/10.5665/sleep.2618.
131. Gaisl T, Haile SR, Thiel S, Osswald M, Kohler M. Efficacy of pharmacotherapy for OSA in adults: a systematic review and network meta-analysis. Sleep Med Rev. 2019;46:74–86. https://doi.org/10.1016/j.smrv.2019.04.009.
132. Ravesloot MJL, White D, Heinzer R, Oksenberg A, Pepin JL. Efficacy of the new generation of devices for positional therapy for patients with positional obstructive sleep apnea: a systematic

review of the literature and meta-analysis. J Clin Sleep Med. 2017;13(6):813–24. https://doi.org/10.5664/jcsm.6622.
133. Javaheri S, Barbe F, Campos-Rodriguez F, Dempsey JA, Khayat R, Javaheri S, et al. Sleep apnea: types, mechanisms, and clinical cardiovascular consequences. J Am Coll Cardiol. 2017;69(7):841–58. https://doi.org/10.1016/j.jacc.2016.11.069.
134. Young T, Finn L, Peppard PE, Szklo-Coxe M, Austin D, Nieto FJ, et al. Sleep disordered breathing and mortality: eighteen-year follow-up of the Wisconsin sleep cohort. Sleep. 2008;31(8):1071–8.
135. Rabelo Guimaraes Mde L, Hermont AP. Sleep apnea and occupational accidents: are oral appliances the solution? Indian J Occup Environ Med. 2014;18(2):39–47. https://doi.org/10.4103/0019-5278.146887.
136. Sutherland K, Deane SA, Chan AS, Schwab RJ, Ng AT, Darendeliler MA, et al. Comparative effects of two oral appliances on upper airway structure in obstructive sleep apnea. Sleep. 2011;34(4):469–77. https://doi.org/10.1093/sleep/34.4.469.
137. Marcus CL, Brooks LJ, Draper KA, Gozal D, Halbower AC, Jones J, et al. Diagnosis and management of childhood obstructive sleep apnea syndrome. Pediatrics. 2012;130(3):576–84. https://doi.org/10.1542/peds.2012-1671.

International Journal of Diabetes in Developing Countries (2021) 41:22–28
https://doi.org/10.1007/s13410-020-00871-y

ORIGINAL ARTICLE

Interaction effect between long sleep duration in early pregnancy and prepregnancy overweight/obesity on gestational diabetes mellitus

Weiye Wang[1,2] · Jiuming Zou[3] · Qiang Fu[2] · Tao Huang[2] · Mengjun Li[2] · Zhao Yao[2] · Linbing Zou[4]

Received: 29 January 2020 / Accepted: 2 September 2020 / Published online: 19 September 2020

Abstract

Objective To explore the individual and interaction effects of sleep duration and prepregnancy body mass index (BMI) on gestational diabetes mellitus (GDM).

Methods Pregnant women who attended antenatal visits in 2 hospitals were selected as the study population. Sleep duration and prepregnancy BMI were collected through a semiquantitative questionnaire, and GDM was diagnosed according to an oral glucose tolerance test (OGTT).

Results In total, 196 women with GDM and 304 controls were included. Compared with the normal weight women, the prepregnancy overweight/obese group had an increased risk of GDM (OR: 7.04, $p < 0.001$); compared with women who slept 7–8.9 h/night, women who slept < 7, 9–9.9, or ≥ 10 h/night had an increased risk of GDM (all $p < 0.001$). The participants with a combination of long sleep duration (≥ 10 h/night) and prepregnancy overweight/obesity had the highest risk of GDM (OR = 39.57, $p < 0.001$), and this effect was greater than the product of the individual effects of a long sleep duration and overweight/obesity (p value of the interaction 0.007).

Conclusions A short or long sleep duration in early pregnancy and prepregnancy overweight/obesity is related to GDM, and there is a synergic effect between a long sleep duration and overweight/obesity on GDM.

Keywords Gestational diabetes mellitus · Body mass index · Obesity · Sleep duration · Interaction

Introduction

Gestational diabetes mellitus (GDM) is among the most common diseases occurring during pregnancy [1]. GDM continues to be a major cause of morbidity and is currently a major public health issue worldwide [2]. According to the statistics of the International Diabetes Association (IDF), approximately one-seventh of newborns worldwide in 2015 were affected by GDM [3]. As the proportions of overweight and obese people and older pregnant women increase, and due to the continuous changes in the diagnostic criteria for GDM, the global incidence of GDM has increased yearly in recent years [4, 5]. GDM can cause great harm to the health of mothers and their fetuses, affect the ability of pregnant women to deliver various nutrients to the fetus, and cause various adverse pregnancy outcomes, such as preterm birth and a low birth weight [6]. In addition, GDM is associated with an increased risk of future cardiovascular diseases in pregnant women [7].

✉ Jiuming Zou
zoujiuming@hbuas.edu.cn

[1] Department of Epidemiology and Health Statistics, School of Public Health, Qingdao University, 38 Dengzhou Road, Qingdao, Shandong 266021, People's Republic of China

[2] Department of Public Health and Preventive Medicine, School of Medicine, Jinggangshan University, 28 Xueyuan Road, Ji'an, Jiangxi 343009, People's Republic of China

[3] Department of Clinical Laboratory, Xiangyang Central Hospital, Affiliated Hospital of Hubei University of Arts and Science, 136 Jingzhou Road, Xiangyang, Hubei 441021, People's Republic of China

[4] Department of Obstetrics, Anhui Women and Children's Health Care Hospital, 15 Yimin Road, Hefei, Anhui 230001, People's Republic of China

Sleep duration and body mass index (BMI) have been recognized as factors that influence GDM. Some studies have shown that a shortened or prolonged sleep duration could increase the risk of GDM [8]. Other studies revealed that BMI could play a vital role in the development of hyperglycemia during pregnancy and that overweight or obese women could be at an increased risk of developing GDM [9, 10]. Sleep duration and overweight/obesity act through the same biological mechanism to increase the occurrence of GDM, and both may be associated with elevated inflammatory responses [11–13]. However, knowledge regarding the interaction effect between sleep and BMI on GDM is currently limited.

The goals of the present study are as follows: (1) to assess the impact of sleep duration in early pregnancy and prepregnancy BMI on GDM and (2) to examine whether any interaction effect exists.

Methods

Study population

In this retrospective study, the study population included pregnant women who visited Ji'an Women and Children's Health Care Hospital (Ji'an city, Jiangxi Province) and Anhui Women and Children's Health Care Hospital (Hefei city, Anhui Province) between April 2018 and May 2019. Pregnant women diagnosed with GDM were selected as cases, and women without GDM were selected as controls. The inclusion criteria were as follows: pregnant women who had (1) completed GDM screening and (2) a live singleton birth. The exclusion criteria were as follows: pregnant women who had (1) a previous diagnosis of diabetes or GDM; (2) a previous stillbirth, spontaneous abortion, delivery of a large baby, fetal malformation with unknown causes, or hydramnios; (3) polycystic ovary syndrome; or (4) communication disorders.

Sleep duration

The sleep duration information was assessed by a semi-quantitative questionnaire, which referenced the Pittsburg Sleep Quality Index (PSQI) scale [14]. Pregnant women were asked when they go to bed at night, how long it takes them to fall asleep, and when they wake in the morning in order to calculate the sleep duration during early pregnancy. In the present study, early pregnancy was defined as the period of 1~13 gestational weeks. The sleep duration responses were categorized into the following four groups: < 7 h/night, 7–8.9 h/night, 9–9.9 h/night, and ≥ 10 h/night.

Prepregnancy BMI

Pregnant women attended face-to-face interviews by trained investigators to assess the prepregnancy BMI information. The following question was asked: "How much did you weigh before pregnancy?" Since height values are generally constant, the prepregnancy height was replaced by the height value measured at enrollment. The maternal prepregnancy BMI was calculated by the following equation: $BMI = weight/height^2$. According to the criteria established by the Health Industry Standards of the People's Republic of China, the BMI was categorized into the following groups: thin (< 18.5 kg/m^2), normal weight (18.5~23.9 kg/m^2), overweight (24~27.9 kg/m^2), and obese (≥ 28 kg/m^2) group [15]. In the present study, the overweight and obese groups were combined into one group (overweight/obesity group) due to the limited sample size.

GDM diagnosis

In this study, GDM was diagnosed according to a routine oral glucose tolerance test (OGTT). According to the standard proposed by the National Health Commission of China and the International Association of Diabetes and Pregnancy Study Group (IADPSG), pregnant women are required to undergo screening for GDM by OGTT between 24 and 28 gestational weeks [16, 17]. The GDM diagnostic criteria were as follows: fasting blood glucose ≥ 5.1 mmol/L, blood glucose ≥ 10.0 mmol/L 1 h after consuming 75 g of glucose, and/or blood glucose ≥ 8.5 mmol/L 2 h after consuming 75 g of glucose [17].

Covariates

The following covariates were obtained from the structured questionnaire administered at enrollment: age, ethnicity (Han Chinese/other), education, smoking (smoking during early pregnancy, defined as ≥ 3 times a week, yes/no), drinking (drinking during early pregnancy, defined as ≥ 3 times a week, yes/no), gestational age, progesterone use (yes/no), midday napping duration (≤ 1/> 1 h/day), and parity (primiparous/multiparous). Education was recorded according to the number of completed years of school (≤ 9/10–12/13–16/≥ 17).

Statistical analysis

The distributions of continuous and categorical variables are described using the means ± SDs, frequencies, and percentages. The Pearson χ^2 test was adopted to compare the sleep duration and prepregnancy BMI category of pregnant women grouped by GDM. Binomial logistic regression analyses were

used to assess the relationship of sleep duration in early pregnancy and prepregnancy BMI category with GDM, and the results are presented as odds ratios (ORs) with 95% confidence intervals (CIs). The following covariates were chosen to control for the impact of potential confounders: age, education, drinking, smoking, progesterone use, ethnicity, midday napping duration, gestational age, and parity.

A stratified analysis was used to explore the interaction effect between sleep duration and prepregnancy BMI on GDM, and the Wald test was used by adding interaction terms between the prepregnancy BMI category and sleep duration (< 7 or ≥ 10 h/night) in early pregnancy. All statistical analyses were performed using R software (The R Project for Statistical Computing, version 3.6.1), and $p < 0.05$ was considered statistically significant.

Results

In total, 196 women with GDM and 304 women without GDM were selected as cases and controls, respectively. The average age of the participants was 28.03 ± 3.27 years. In total, 63 women (12.6%) had a family history of diabetes, and 88.6% of the women were primiparous. The average gestational age at the OGTT was 26.44 ± 2.65 weeks. Additional details of the demographic and clinical characteristics are provided in Table 1.

On an average, the prepregnancy BMI of women in the case group was 22.25 ± 3.09 kg/m^2, which was higher than that of the control group ($p < 0.001$). The Pearson χ^2 test showed that the distributions of the BMI category and sleep duration significantly differed between the two groups (both $p < 0.001$). Additional details are shown in Table 2. After adjusting for several confounding factors, compared with the normal weight women, prepregnancy overweight/obese women had a statistically significant increased risk of GDM (OR: 7.04, 95% CI: 3.65, 13.59, $p < 0.001$); compared with women who slept 7–8.9 h/night in early pregnancy, those who slept < 7 h/night, 9–9.9 h/night, or ≥ 10 h/night, all had a statistically significant increased risk of GDM. Additional details are provided in Table 3.

The combined effect of a long sleep duration in early pregnancy and prepregnancy overweight/obesity was also observed. As shown in Table 4, the combination of a long sleep duration (≥ 10 h/night) in early pregnancy and prepregnancy overweight/obesity resulted in the highest risk of GDM (OR = 39.57, $p < 0.001$), which was greater than the product of the individual effects of a long sleep duration and overweight/obesity. Furthermore, the interaction effect of long sleep duration in early pregnancy and prepregnancy overweight/obesity was examined using the Wald test. Compared with the GDM risk in the 7–8.9 h/night sleep duration group, the GDM risk in the ≥ 10 h/night sleep duration group was increased in the prepregnancy overweight/obese group, and the impact was much stronger than that in the normal weight group (p value of the interaction 0.007). Additional details are provided in Table 5.

Discussion

In this case-control study, we observed that short or long sleep durations in women during early pregnancy and those with prepregnancy overweight/obesity were associated with GDM. In addition, we examined the interaction effect of sleep duration on GDM in women during early pregnancy and those with prepregnancy overweight/obesity for the first time. There was a synergic effect between a long sleep duration and overweight/obesity on GDM, and the effect of a long sleep duration on GDM was stronger among the overweight/obese women.

Sleep disturbance is a common disorder in pregnant women with GDM [18]. Some studies have observed a relationship between sleep duration during pregnancy and GDM. Reutrakul et al. [19] found a significant negative impact of sleep duration < 7 h/night during the second trimester on OGTT blood glucose levels. Other studies observed that a short sleep duration could increase the occurrence of GDM among pregnant women [20, 21]. Several studies conducted in the USA and China showed that a U-type relationship exists between sleep duration and the risk of GDM and that short or long sleep durations in early pregnancy could lead to an increased risk of GDM [22, 23]. However, a study observed no significant correlation between sleep duration and GDM after adjusting for a series of covariates [24]. This study found that short and long sleep durations were both related to an increased occurrence of GDM, and our findings were consistent with the results of most of the previous studies.

The mechanisms underlying the effect of sleep duration on GDM remain unclear, but this effect could be affected by the following pathways. First, sleep disturbances can increase inflammatory responses that cause endothelial dysfunction and elevated oxidative stresses, and endothelial dysfunction is related to insulin resistance in type 2 diabetes [12, 13]. Second, the reduction in the basal glucagon concentrations caused by sleep disorders has been proposed as a mechanism to explain the impact of sleep duration [25]. In addition, high sympathetic nervous system activity, dysfunction of the hypothalamic pituitary axis (HPA), alteration of the synthesis and release of cytokines and adipokines, elevated serum cortisol levels, and peripheral vasoconstriction have all been reported to be potential biological mechanisms [26, 27].

As mentioned above, the relationship between prepregnancy BMI and GDM is relatively clear; prepregnancy overweight/obesity is reportedly associated with an increased

Table 1 Demographic and clinical characteristics of the study population†

Characteristics	Total ($n = 500$)	GDM	
		Yes ($n = 196$)	No ($n = 304$)
Age (years)	28.03 ± 3.27	28.99 ± 3.64	27.40 ± 2.82
Ethnicity			
Han Chinese	492 (98.4%)	193 (98.5%)	299 (98.4%)
Other	8 (1.6%)	3 (1.5%)	5 (1.6%)
Education (years of school)			
≤ 9	14 (2.8%)	3 (1.5%)	11 (3.6%)
10–12	74 (14.8%)	33 (16.8%)	41 (13.5%)
13–16	313 (62.6%)	126 (64.3%)	187 (61.5%)
≥ 17	99 (19.8%)	34 (17.3%)	65 (21.4%)
Height (cm)	160.61 ± 4.80	160.77 ± 4.94	160.50 ± 4.71
Prepregnancy weight (kg)	53.76 ± 7.57	57.50 ± 8.29	51.34 ± 5.95
Parity			
Primiparous	443 (88.6%)	167 (85.2%)	276 (90.8%)
Multiparous	57 (11.4%)	29 (14.8%)	28 (9.2%)
Family history of diabetes			
Yes	63 (12.6%)	34 (17.3%)	29 (9.5%)
No	437 (87.4%)	162 (82.7%)	275 (90.5%)
Drinking			
Yes	5 (1.0%)	4 (2.0%)	1 (0.3%)
No	495 (99.0%)	192 (98.0%)	303 (99.7%)
Smoking			
Yes	4 (0.8%)	3 (1.5%)	1 (0.3%)
No	496 (99.2%)	193 (98.5%)	303 (99.7%)
Progesterone use			
Yes	109 (21.8%)	47 (24.0%)	62 (20.4%)
No	391 (78.2%)	149 (76.0%)	242 (79.6%)
Gestational age at OGTT	26.44 ± 2.65	26.66 ± 4.17	26.31 ± 0.58
Midday napping duration			
≤ 1 h/day	339 (67.8%)	103 (52.6%)	236 (77.6%)
> 1 h/day	161 (32.2%)	93 (47.4%)	68 (22.4%)

GDM gestational diabetes mellitus, *OGTT* oral glucose tolerance test. †The data are presented as the mean ± SD or frequency (percentage)

Table 2 Univariate association between prepregnancy BMI or sleep duration in early pregnancy and GDM†

Characteristics	Total ($n = 500$)	GDM		
		Yes ($n = 196$)	No ($n = 304$)	p
BMI (kg/m^2)	20.83 ± 2.77	22.25 ± 3.09	19.92 ± 2.10	< 0.001
BMI category				< 0.001
Thin	107 (21.4%)	109 (55.6%)	210 (69.1%)	
Normal	319 (63.8%)	24 (12.2%)	83 (27.3%)	
Overweight/Obesity	74 (14.8)	63 (32.2%)	11 (3.6%)	
Sleep duration				< 0.001
< 7 h/night	20 (4.0%)	17 (8.7%)	3 (1.0%)	
7–8.9 h/night	250 (50.0%)	62 (31.6%)	188 (61.8%)	
9–9.9 h/night	119 (23.8%)	47 (24.0%)	72 (23.7%)	
≥ 10 h/night	111 (22.2%)	70 (35.7%)	41 (13.5%)	

BMI body mass index. †The data are presented as the frequency (percentage)

Table 3 Multivariate association between prepregnancy BMI category or sleep duration in early pregnancy and GDM

Characteristics	*n*	Crude		Adjusted[a]	
		OR (95% CI)	*p*	OR (95% CI)	*p*
BMI category					
Thin	107	0.56 (0.34, 0.93)	0.024	0.64 (0.39, 1.05)	0.096
Normal	319	1.00 (1.00, 1.00)	1.000	1.00 (1.00, 1.00)	1.000
Overweight/obesity	74	11.03 (5.59, 21.80)	< 0.001	7.04 (3.65, 13.59)	< 0.001
Sleep duration					
< 7 h/night	20	17.18 (4.87, 60.61)	< 0.001	10.27 (3.66, 28.82)	< 0.001
7–8.9 h/night	250	1.00 (1.00, 1.00)	1.000	1.00 (1.00, 1.00)	1.000
9–9.9 h/night	119	1.98 (1.24, 3.16)	0.004	2.00 (1.06, 3.77)	0.016
≥ 10 h/night	111	5.18 (3.20, 8.37)	< 0.001	4.25 (2.39, 7.56)	< 0.001

OR Odds ratio, *CI* confidence interval. [a] All models were adjusted for age, ethnicity, education, drinking, smoking, gestational age, parity, progesterone use, midday napping duration, and family history of diabetes

risk of GDM, and similar results were observed in the present study [9, 10]. The mechanisms underlying the effect may be related to elevated inflammatory responses [11].

To the best of our knowledge, this study is the first to observe the interaction effect of sleep duration and prepregnancy BMI on GDM. We found a synergic effect between a long sleep duration and overweight/obesity on GDM and that the effect of a long sleep duration on GDM was stronger among overweight/obese women. Our findings suggest that increased attention should be paid to overweight/obese pregnant women with long sleep durations to prevent and control the incidence of GDM. The mechanisms underlying the interaction effect remain unclear and may be related to the inflammatory response. Future studies are needed to confirm our findings.

The main strength of the present study is the use of abundant and accurate covariate data, which were collected and included in the multivariate analyses to avoid potential confounding bias. However, there are a few limitations that deserve acknowledgment. First, compared with a prospective cohort study design, the retrospective design of the present study weakens its ability to verify causality. In the present study, the prepregnancy weight and sleep duration in early pregnancy were self-reported through a questionnaire survey conducted at 24 to 28 gestational weeks, which could cause recall bias. Second, the sample size was not large. Due to the small size, the confidence interval of the effect was very large when observing the combined effect of a long sleep duration and prepregnancy overweight/obesity on GDM. Third, sleep duration was examined by a questionnaire referring to the PSQI scale, and self-reported sleep duration has been reported to be somewhat longer than objectively measured sleep duration in pregnant women according to a validation study of the PSQI [28].

In summary, this study revealed a significant relationship between sleep duration in early pregnancy or prepregnancy

Table 4 Combined effect of a long sleep duration in early pregnancy and prepregnancy overweight/obesity on GDM

	n	Crude OR (95% CI)	*p*	Adjusted[a] OR (95% CI)	*p*
Group A	178	1.00 (1.00, 1.00)	1.000	1.00 (1.00, 1.00)	1.000
Group B	86	1.75 (1.01, 3.04)	0.047	1.89 (1.06, 3.70)	0.033
Group C	44	3.24 (1.64, 6.40)	0.001	3.09 (1.04, 9.18)	0.036
Group D	13	3.45 (1.10, 10.80)	0.034	3.19 (1.06, 9.60)	0.031
Group E	13	16.26 (3.47, 76.14)	< 0.001	18.05 (5.83, 55.88)	< 0.001
Group F	42	38.42 (11.32, 130.40)	< 0.001	39.57 (16.01, 97.80)	< 0.001

Group A: normal weight combined with a sleep duration of 7–8.9 h/night; Group B: normal weight combined with a sleep duration of 9–9.9 h/night; Group C: normal weight combined with a sleep duration of ≥ 10 h/night; Group D: overweight/obese participants combined with a sleep duration of 7–8.9 h/night; Group E: overweight/obese participants combined with a sleep duration of 9–9.9 h/night; Group F: overweight/obese participants combined with a sleep duration of ≥ 10 h/night. [a] All models were adjusted for age, ethnicity, education, drinking, smoking, gestational age, parity, progesterone use, midday napping duration and family history of diabetes

Table 5 Interaction effect between prepregnancy BMI and a long sleep duration in early pregnancy on GDM

BMI category	n_1 [a]	n_2 [b]	OR (95% CI) [c]	p [d]	p_{int} [e]
Normal	178	44	3.09 (1.04, 9.18)	0.036	
Thin	59	25	3.45 (1.05, 11.33)	0.035	0.287
Overweight/obesity	13	42	10.51 (2.62, 42.16)	0.001	0.007

All models were adjusted for age, ethnicity, education, drinking, smoking, gestational age, parity, progesterone use, midday napping duration, and family history of diabetes. [a] Sample size of women who slept 7–8.9 h/night in each subgroup; [b] Sample size of women who slept ≥ 10 h/night in each subgroup; [c] Odds ratio (95% confidence interval) of ≥ 10 sleeping hours/night compared to 7–8.9 h/night in each subgroup; [d] Significance of the effect in each subgroup; [e] The p value of the interaction was based on the Wald test

BMI and GDM in Chinese women. More interestingly, the current study showed that the impact of a long sleep duration on GDM was stronger among overweight/obese women than that among normal weight women and that there was a synergic effect between a long sleep duration and overweight/obesity on GDM. Our findings suggest that special attention should be paid to prepregnancy overweight/obese pregnant women with long sleep duration to prevent GDM.

Acknowledgments We are sincerely indebted to the participants for their cooperation, and we gratefully acknowledge Mrs. Sisi Hu for her help in polishing the English language use in this manuscript.

Funding This study was funded by grants from the Natural Science Foundation of Jiangxi Province (No. 20202BABL216043), the Natural Science Project of Jinggangshan University (No. JZB1817), and the Science and Technology Research Project of the Department of Education of Jiangxi Province (No. GJJ170647).

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval The study was approved by Jinggangshan University Medical Ethics Review Committee (No. 201801), and all participants provided written informed consent.

References

1. McIntyre HD, Catalano P, Zhang C, Desoye G, Mathiesen ER, Damm P. Gestational diabetes mellitus. Nat Rev Dis Primers. 2019;5(1):47.
2. Lee KW, Ching SM, Hoo FK, Ramachandran V, Chong SC, Tusimin M, et al. Neonatal outcomes and its association among gestational diabetes mellitus with and without depression, anxiety and stress symptoms in Malaysia: a cross-sectional study. Midwifery. 2020;81:102586.
3. Mishra S, Rao CR, Shetty A. Trends in the diagnosis of gestational diabetes mellitus. Scientifica (Cairo). 2016;2016:5489015.
4. Szmuilowicz ED, Josefson JL, Metzger BE. Gestational diabetes mellitus. Endocrinol Metab Clin N Am. 2019;48(3):479–93.
5. Basha AS, Fram KM, Thekrallah FM, Irshaid ZA, Maswady AM, Obeidat ZN. Prevalence of gestational diabetes and contributing factors among pregnant Jordanian women attending Jordan University Hospital. Int J Diabetes Dev C. 2019;39(1):132–8.
6. Yang Y, Wang Z, Mo M, Muyiduli X, Wang S, Li M, et al. The association of gestational diabetes mellitus with fetal birth weight. J Diabetes Complicat. 2018;32(7):635–42.
7. Kramer CK, Campbell S, Retnakaran R. Gestational diabetes and the risk of cardiovascular disease in women: a systematic review and meta-analysis. Diabetologia. 2019;62(6):905–14.
8. Xu YH, Shi L, Bao YP, Chen SJ, Shi J, Zhang RL, et al. Association between sleep duration during pregnancy and gestational diabetes mellitus: a meta-analysis. Sleep Med. 2018;52:67–74.
9. Martin KE, Grivell RM, Yelland LN, Dodd JM. The influence of maternal BMI and gestational diabetes on pregnancy outcome. Diabetes Res Clin Pract. 2015;108(3):508–13.
10. Torloni MR, Betran AP, Horta BL, Nakamura MU, Atallah AN, Moron AF, et al. Prepregnancy BMI and the risk of gestational diabetes: a systematic review of the literature with meta-analysis. Obes Rev. 2009;10(2):194–203.
11. Pantham P, Aye IL, Powell TL. Inflammation in maternal obesity and gestational diabetes mellitus. Placenta. 2015;36(7):709–15.
12. Jelic S, Le Jemtel TH. Inflammation, oxidative stress, and the vascular endothelium in obstructive sleep apnea. Trends Cardiovasc Med. 2008;18(7):253–60.
13. Meigs JB, Hu FB, Rifai N, Manson JE. Biomarkers of endothelial dysfunction and risk of type 2 diabetes mellitus. JAMA. 2004;291(16):1978–86.
14. Buysse DJ, Reynolds CF 3rd, Monk TH, Berman SR, Kupfer DJ. The Pittsburgh Sleep Quality Index: a new instrument for psychiatric practice and research. Psychiatry Res. 1989;28(2):193–213.
15. Zhang B, Yang S, Yang R, Wang J, Liang S, Hu R, et al. Maternal prepregnancy body mass index and small for gestational age births in Chinese women. Paediatr Perinat Epidemiol. 2016;30(6):550–4.
16. Yang HX. Diagnostic criteria for gestational diabetes mellitus (WS 331-2011). Chin Med J. 2012;125(7):1212–3.
17. International Association of Diabetes and Pregnancy Study Groups Consensus Panel, Metzger BE, Gabbe SG, Persson B, Buchanan TA, Catalano PA, et al. International association of diabetes and pregnancy study groups recommendations on the diagnosis and classification of hyperglycemia in pregnancy. Diabetes Care. 2010;33(3):676–82.
18. Devaraj NK. Knowledge, attitude, and practice regarding obstructive sleep apnea among primary care physicians [J]. Sleep Breath. 2020;24:1–10.
19. Reutrakul S, Zaidi N, Wroblewski K, Kay HH, Ismail M, Ehrmann DA, et al. Sleep disturbances and their relationship to glucose tolerance in pregnancy. Diabetes Care. 2011;34(11):2454–7.
20. Cai S, Tan S, Gluckman PD, Godfrey KM, Saw SM, Teoh OH, et al. Sleep quality and nocturnal sleep duration in pregnancy and risk of gestational diabetes mellitus. Sleep. 2017;40(2):zsw058.
21. Facco FL, Grobman WA, Kramer J, Ho KH, Zee PC. Self-reported short sleep duration and frequent snoring in pregnancy: impact on glucose metabolism. Am J Obstet Gynecol. 2010;203(2):142 e1–5.
22. Rawal S, Hinkle SN, Zhu Y, Albert PS, Zhang C. A longitudinal study of sleep duration in pregnancy and subsequent risk of gestational diabetes: findings from a prospective, multiracial cohort. Am J Obstet Gynecol. 2017;216(4):399.e1–8.
23. Wang H, Leng J, Li W, Wang L, Zhang C, Li W, et al. Sleep duration and quality, and risk of gestational diabetes mellitus in pregnant Chinese women. Diabet Med. 2017;34(1):44–50.

24. Izci Balserak B, Jackson N, Ratcliffe SA, Pack AI, Pien GW. Sleep-disordered breathing and daytime napping are associated with maternal hyperglycemia. Sleep Breath. 2013;17(3):1093–102.
25. Schmid SM, Jauch-Chara K, Hallschmid M, Schultes B. Mild sleep restriction acutely reduces plasma glucagon levels in healthy men. J Clin Endocrinol Metab. 2009;94(12):5169–73.
26. Izci-Balserak B, Pien GW. The relationship and potential mechanistic pathways between sleep disturbances and maternal hyperglycemia. Curr Diab Rep. 2014;14(2):459.
27. Arnardottir ES, Mackiewicz M, Gislason T, Teff KL, Pack AI. Molecular signatures of obstructive sleep apnea in adults: a review and perspective. Sleep. 2009;32(4):447–70.
28. Herring SJ, Foster GD, Pien GW, Massa K, Nelson DB, Gehrman PR, et al. Do pregnant women accurately report sleep time? A comparison between self-reported and objective measures of sleep duration in pregnancy among a sample of urban mothers. Sleep Breath. 2013;17(4):1323–7.

International Journal of Diabetes in Developing Countries (2021) 41:29–36
https://doi.org/10.1007/s13410-020-00849-w

ORIGINAL ARTICLE

The effect of Ellagic acid on sleep quality in patients with type 2 diabetes: a randomized double blind clinical trial

Mahnaz Ghadimi[1] · Sima Hashemipour[2,3] · Mohamadreza Rashidi Nooshabadi[4] · Maria Kavianpour[5] · Hossein Khadem Haghighian[1,3]

Received: 24 April 2020 / Accepted: 2 July 2020 / Published online: 24 July 2020

Abstract

Background Oxidative stress can reduce the quality of sleep in patients with type 2 diabetes. Antioxidants such as polyphenols may increase sleep quality by improving oxidative stress conditions.

Objective Considering the antioxidant properties of Ellagic acid (EA), this study was designed to evaluate the effect of EA on sleep quality in diabetic patients.

Methods In this study, 44 diabetic patients were recruited. Patients who met the inclusion criteria that were randomly allocated consumed a capsule containing 180 mg of EA per day ($n = 22$) or placebo ($n = 22$) for 8 weeks. Anthropometric factors, physical activity, food intake, and Petersburg's Sleep Quality (PSQI) questionnaire were assessed at the beginning and end of the study. Kolmogorov-Smirnov test, paired sample t test and independent sample t test were used to analyze the data.

Results At the end of the study, the mean scores of PSQI and sleep subgroups in the intervention group were significantly lower than in the placebo group ($p < 0.05$). According to intragroup comparisons, these changes were significant in the intervention group at the end of the study compared to the beginning of the study ($p < 0.05$) and were not significant in the placebo group ($p > 0.05$).

Conclusion According to these findings, intake of EA may help to improve the sleep quality in patients with type 2 diabetes. These effects may be due to the antioxidant effects of this polyphenol.

Keywords Ellagic acid · Sleep quality · Stress oxidative · Type 2 diabetes

IRCT code IRCT20141025019669N13

Abbreviations

AGEs	Advanced glycation end products
BMI	Body mass index
DM	Diabetes mellitus
EA	Ellagic acid
FBS	Fasting blood sugar
GPx	Glutathione peroxidase
GSH	Glutathione
GSSG	Oxidized glutathione
HbA_1c	Hemoglobin A_1c

Maria Kavianpour and Hossein Khadem Haghighian contributed equally to this work.

✉ Maria Kavianpour
Kavianpour.maria@gmail.com

✉ Hossein Khadem Haghighian
khademnut@yahoo.com

[1] Department of Nutrition, School of Health, Qazvin University of Medical Sciences, Qazvin, Iran

[2] Department of Internal Medicine, Velayat Clinical Research Development Unit, Qazvin University of Medical Sciences, Qazvin, Iran

[3] Metabolic Diseases Research Center, Research Institute for Prevention of Non-Communicable Diseases, Qazvin University of Medical Sciences, Qazvin, Iran

[4] Department of Pharmacology and Toxicology, School of Pharmacy, Ahvaz Jundishapur University of Medical Sciences, Ahvaz, Iran

[5] Department of Tissue Engineering and Applied Cell Sciences, Faculty of Advanced Technologies in Medicine, Tehran University of Medical Sciences, Tehran, Iran

IL-1	Interleukin-1
IPAQ	International Physical Activity Questionnaire
MDA	Malondialdehyde
PSQI	Petersburg's Sleep Quality Index
ROS	Reactive oxygen species
SOD	Superoxide dismutase
TNFα	Tumor necrosis factor alpha

Background

One of the most common problems in people with type 2 diabetes is sleep disorders, which affect the control and treatment of this hormonal condition. Diabetes mellitus (DM) is a group of metabolic diseases characterized by hyperglycemia due to a defect in insulin secretion, insulin activity, or both [1]. Chronic hyperglycemia is associated with long-term damage, dysfunction, and failure of various organs such as the kidneys, nerves, heart, and blood vessels [2]. DM is divided into two main types: type 1 and type 2 diabetes. In general, the detrimental effects of hyperglycemia include macrovascular complications (coronary artery disease, peripheral arterial disease, and stroke), microvascular complications (diabetic nephropathy, retinopathy, neuropathy), and retinopathy [3]. Due to the prevalence of this disease and the heavy financial costs on the economic and health system of societies, proper control of blood sugar in diabetic patients and adjustment of factors affecting proper control of blood sugar are very important [4]. Lifestyle is one of the most important factors that can improve the outcome of care in these patients. Sleep in the face of physical activity is important component of a person's lifestyle and is two sides of the same coin. Disruption in each of them causes disruption in the other [5].

Epidemiological studies in recent years on the relationship between the quality of sleep at night and the prevalence of diabetes have shown that increasing or decreasing the duration of sleep at night has been associated with increased prevalence and incidence of diabetes and poor blood sugar control in diabetics [6]. Laboratory studies indicated the complete deprivation of 24 h of sleep, relative deprivation of several days of sleep, and sleeping for 12 h for several consecutive days, all of which increase the resistance of the surrounding tissue to insulin, impair glucose tolerance, and increase the feeling of hunger and consume foods with low nutritional value [7]. According to psychological studies, there is a two-way relationship between diabetes and sleep disorders. Decreased sleep quality in diabetic patients may be considered equivalent to insulin resistance caused by sleep disorders. The results of laboratory and clinical studies consider oxidative stress to be the missing link between the two conditions [8]. Oxidative stress can cause degenerative diseases by speeding up the cell cycle and increasing the cell death, which causes degenerative diseases. The results of studies indicated that during sleep, the antioxidant system increases the quality of sleep by eliminating free radicals and reducing the rate of formation of these active species, and it is clear that reducing the antioxidant power will worsen sleep [9]. However, the exact relationship between oxidative stress and sleep disorders has not been clearly established. Antioxidant consumption with dietary origin can help increase the strength of the antioxidant system power [10]. One of the known groups of antioxidants is polyphenols, and one of the polyphenols with high metabolic power is Ellagic acid (EA). EA (4,4,5,5,6,6-hexahydroxidifenic acid 2,6,2,6-dilactone) is a polyphenolic acid found in many foods such as fruits (especially pomegranates), nuts, and plant extracts. Antioxidant, anti-apoptotic, and anti-inflammatory properties of EA have been reported in several studies [11]. Considering the role of oxidative stress in sleep disorders and antioxidant properties of EA and the relations between diabetes, sleep quality, and oxidative stress, the purpose of this study was to evaluate the effect of EA supplementation on sleep quality in people with DM,

Methods

Study design

This randomized, double-blind, placebo-controlled clinical trial study was aimed to assess the 180 mg of Ellagic acid daily for 8 weeks to determine the effect of this dose of EA on sleep quality in diabetic patients. Demographic characteristics, medical history, and current medications of patients were recorded. Then, height, weight, and body mass index (BMI) were measured. Eligible patients were matched for age, sex, and weight. Patients were randomly assigned to two groups of intervention (EA) ($n = 22$) and placebo ($n = 22$). Sampling in this study was carried out using simple random sampling with using random numbers. Each person received an Ellagic acid capsule (180 mg) or placebo capsule containing wheat flour once daily with meals. The color, shape, and size of the supplement capsule were similar to that of the placebo capsule. In this study, the patient, researcher, and specialist physician were blind to supplement and placebo. Capsules prepared by someone else outside the study in A and B groups that were placed in the same package so that the investigator would be unaware of the contents of the capsules. Supplement was purchased from supplement spot and the placebo was made by School of Pharmacy, Tabriz University of Medical Sciences. Pharmacological remedy was similar in the two groups. All patients were advised not to alter their diet and physical activity habits along the study. The effective selective dose for Ellagic acid supplement was taken from Falsaperela et al. article [12]. At the beginning and at the end of the study, patients completed the PSQI questionnaire. The PSQI questionnaire is a tool for measuring sleep quality and determining

sleep disorder over the past month and consists of seven questions with a total score ranging from 0 to 21. Earning a score of 5 and above indicates sleep disorder in the individual. The questionnaire consists of seven parts. Also, to control for confounding effects of diet and physical activity, at the beginning and at the end of the study, patients were interviewed by a 3-day dietary recall questionnaire and subjects with moderate physical activity level were enrolled. Three-day food recalls were used to assess dietary intake, and Nutritionist IV program (San Bruno, CA) modified for Iranian food composition was used for estimating the dietary intake of participants. Also, to evaluate the physical activity, we used the International Physical Activity Questionnaire (IPAQ) [13]. Data from the IPAQ were converted to metabolic equivalent-minutes/week using existing guidelines.

Inclusion and exclusion criteria

In this study, patients were selected based on the Pittsburg Sleep Quality Index (PSQI) [14] which was referred to the Endocrinology and Metabolism Clinic of Qazvin University of Medical Sciences, Qazvin, Iran, with the opinion of the project's clinical consultant (endocrinologist). Patients who scored 5 or higher on this questionnaire were included in this study. This questionnaire has been validated in various scientific studies in Iran. Other inclusion criteria for this study include type 2 diabetes ($HbA_1c \geq 6\%$ or FBS ≥ 126 mg/dL (7 mmol/L)), 2-hp glucose ≥ 200 mg/dL (11.1 mmol/L), or random plasma glucose (≥ 200 mg/dL). Willingness to cooperate, moderate physical activity level, no change in treatment and medication in at least the last 2 months and having a 2-year history of diabetes. Exclusion criteria for this study included:taking of insulin, having diabetes for more than 10 years, pregnancy and lactation, BMI > 30, patients with severe renal and hepatic impairment, any acute illness that may affect the study (cardiovascular, pulmonary, renal, cancer, infection), change in dosage of hypoglycemic drugs, taking any dietary supplement from 2 months before the study, a history of any allergies, alcohol consumption, any side effects of the intervention, and unwillingness to participate or continue cooperation.

Patients

Participants in this study were patients with type 2 diabetes aged 24–55 years. In the beginning of the study, 54 diabetic patients were invited to the study with the approval of a specialist physician, of whom 10 were excluded because of unwillingness to participate in the study, and eventually, 44 patients entered the study (22 in each group).

Sample size

The level of malondialdehyde factor was used to calculate sample size before and after administration of pomegranate extract in the study of Hosseini et al. [15]. The sample size of 18 was obtained in each group. Considering 30% probable drop-out, the sample size was considered 22. In this research, 44 diabetic patients were studied.

Monitoring the intervention

Patients were followed up to control their consumption of Ellagic acid capsules and prevented from falling out once every 7 days by telephone. At the end of the study, each person should return the bottle containing their supplement for count of capsules. Patients who consumed less than 10% of capsules were removed from the study.

Statistical analyses

Statistical analysis was performed by SPSS version 20. The data were reported as mean ± SD. The normality of data distribution was assessed by Kolmogorov-Smirnov test. To compare the mean values before and after intervention in each group, paired sample *t* test was used, and to compare the mean values between two groups, independent sample *t* test was used. For all analysis, p value < 0.05 was considered statistically significant.

Results

In this study, in 44 patients who participated, 22 patients received Ellagic acid and 22 patients received placebo. In the present study, one person from each group did not complete the study for personal reasons. Also, no side effects were reported in the study. The characteristics of the participants are shown in Table 1. There was no statistically significant difference in the baseline characteristics of the participants between the two groups. The mean age of participants in intervention and placebo groups was 47 ± 5.06 and 44.04 ± 6.88 years old, respectively ($p > 0.05$). Also, there was no significant difference between the two groups in terms of anthropometric factors at the beginning of the study. The mean and standard deviation of weight (67.23 ± 10.24 vs 68.95 ± 9.02), height (162.43 ± 10.59 vs 163.71 ± 9.12), and BMI (25.32 ± 1.07 vs 25.61 ± 0.86) were in the intervention and placebo groups, respectively. Also, there was no significant difference in the amount of physical activity (36.15 ± 2.99 vs 37.27 ± 2.78) between groups at the beginning of the study ($p > 0.05$; Table 1). It is also noteworthy that at the end of the study, there was no difference in terms of weight, BMI, and physical activity between the two groups as well as within the group ($p > 0.05$; Table 1). The

Table 1 The comparison of baseline characteristics of the participants

Variable		Mean ± SD placebo ($n = 21$)	Mean ± SD Ellagic acid ($n = 21$)	P1
Age (years, mean ±SD)		44.04 ± 6.88	47 ± 5.06	0.701
Height(cm)		163.71 ± 9.12	162.43 ± 10.59	0.814
Weight (kg)	Before	68.95 ± 9.02	67.23 ± 10.24	0.621
	After	68.49 ± 9.21	66.98 ± 10.31	0.59
	P2	0.78	0.619	
Body mass index (kg/m^2, mean ±SD)	Before	25.61 ± 0.86	25.32 ± 1.07	0.562
	After	25.43 ± 0.9	25.22 ± 1.09	0.604
	P2	0.719	0.807	
Physical activity	Before	37.27 ± 2.78	36.15 ± 2.99	0.132
	After	37.5 ± 3.31	36.43 ± 2.89	0.108
	P2	0.429	0.49	
Metformin dose	1509.63 ± 284.27	1523.07 ± 301.71	0.83	Metformin dose
Diabetes duration	6.07 ± 1.27	5.87 ± 1.03	0.21	Diabetes duration

Data are expressed as means ± SD

P1: Comparison of the mean of baseline characteristics between the two groups of Ellagic acid and placebo (independent sample *t* test)

P2: Comparison of mean of baseline characteristics in each group at baseline and end of study (paired sample *t* test)

mean of energy and macronutrient intake at baseline and the end of the study are shown in Table 2. As shown, there was no statistically significant difference between the groups in terms of average daily intake of the energy, protein, fat, saturated fatty acids, unsaturated fatty acids, and some micronutrients at the beginning and the end of the study ($p > 0.05$). Also, there were no significant differences in metformin dose and duration of diabetes between two groups at the baseline ($p > 0.05$. Table 1). The data summarized in Table 3 shows the mean and standard deviation of sleep and sleep subgroups scores. There was no significant difference between the groups at the beginning of the study in terms of overall sleep quality and sleep subgroup scores (PSQI (score): 8.92 ± 1.39 vs 8.86 ± 2.07 in placebo and intervention groups, respectively, $p > 0.05$). EA intake caused significant changes in mean sleep score (PSQI), sleep duration, sleep latency, sleep efficiency, sleep disturbances, use of sleep medications, day time dysfunction, and the subjective sleep quality at the end of the study compared to the beginning of the study ($p < 0.05$). Also, comparing the mean changes of these factors at the end of the study between the two groups was significant ($p < 0.05$). But, according to intergroup comparisons, the results of the study showed no change in the placebo group in terms of sleep score and sleep subgroup score at the end of the study compared to the beginning of the study ($p > 0.05$).

Discussion

In the present study, the role of Ellagic acid on sleep quality in diabetic patients was assessed. In this research, the intervention group received 180 mg of Ellagic acid for 8 weeks. Sleep quality in diabetic patients significantly improved after intervention. The PSQI score significantly decreased in intervention group. Sleep disorders are associated with psychological disorders and are one of the causes of diseases such as anxiety and suicide [16]. Meanwhile, the results of descriptive-analytical studies suggest that blood sugar control worsens as a result of sleep disorders. Of course, considering animal and human studies, the relationship between these two scientific categories is two-way. Considering the results of experimental and clinical studies, it can be concluded that one of the conditions that can affect these two diseases and make both conditions worse is the high intensity of oxidative stress in the body system [17]. Eating antioxidants of nutritional origin can help maintain balance and control oxidative stress. One of the strongest antioxidants in the polyphenols group is Ellagic acid. Several animal and clinical scientific studies have assessed the antioxidant functions on sleep quality in several diseases [18]. Alzoubi et al. assessed the effect of vitamin E as an antioxidant supplement in sleep-deprived rats. The animals were given 100 mg/kg vitamin E, and antioxidant biomarkers such as glutathione (GSH), oxidized glutathione (GSSG) and GSH/GSSG ratio, glutathione peroxidase (GPx), and superoxide dismutase (SOD) were measured. The results of study showed that vitamin E improved chronic insomnia by reducing hippocampal GSH/GSSG ratio, catalase, SOD, and GPx activity [19]. Also, the results of Mohammad Shahi et al. study indicated that the use of vitamin D supplement improves sleep quality, reduces sleep latency, raises sleep duration, and improves subjective sleep quality in people of 20–50 years old with sleep disorder.

Table 2 The comparison of the dietary intake at the baseline and the end of the study in patients with IBS

Variables		Mean ± SD placebo ($n = 21$)	Mean ± SD Ellagic acid ($n = 21$)	P1
Energy (kcal)	Baseline	2057.71 ± 230.546	2034.41 ± 286.84	0.405
	End	2043.49 ± 241.269	2026.38 ± 292.2	0.3
	P2	0.58	0.48	
Protein (g)	Baseline	82.19 ± 16.3	80.81 ± 23.11	0.55
	End	81.07 ± 21.03	78.27 ± 18.01	0.41
	P2	0.68	0.6	
Carbohydrate (g)	Baseline	263.61 ± 62.19	260.71 ± 49.13	0.59
	End	261.39 ± 51.09	259.14 ± 52.2	0.52
	P2	0.63	0.69	
Fat (g)	Baseline	77.5 ± 16.22	76.3 ± 15.07	0.801
	End	75.18 ± 24.18	74.17 ± 15.07	0.705
	P2	0.309	0.41	
Saturated fatty acids (g)	Baseline	24.08 ± 6.31	23.27 ± 6.17	0.77
	End	23.1 ± 6.07	22.06 ± 5.17	0.662
	P2	0.47	0.6	
Monounsaturated fatty acid (g)	Baseline	28.09 ± 8.14	26.45 ± 7.3	0.807
	End	28.22 ± 7.12	26.65 ± 5.3	0.59
	P2	0.63	0.74	
Polyunsaturated fatty acid (g)	Baseline	25.28 ± 7.64	25.56 ± 7.03	0.82
	End	24.15 ± 5.18	24.27 ± 5.17	0.86
	P2	0.354	0.29	
Fiber (g)	Baseline	9.27 ± 1.03	8.95 ± 1	0.48
	End	8.65 ± 1.44	8.07 ± 1.2	0.401
	P2	0.28	0.25	
Vitamin C (mg)	Baseline	69.14 ± 20.19	68.47 ± 13.27	0.74
	End	67.47 ± 23.01	68.03 ± 14.37	0.52
	P2	0.583	0.641	
Vitamin E (IU)	Baseline	8.24 ± 1.01	7.66 ± 1.24	0.456
	End	7.53 ± 1.21	7.87 ± 1.01	0.11
	P2	0.353	0.84	
Selenium	Baseline	121.77 ± 41.02	120 ± 25.07	0.481
	End	120.16 ± 44.53	119.44 ± 19.61	0.3
	P2	0.347	0.491	

Data are expressed as means ± SD

P1: Comparison of the mean of dietary intake between the two groups of Ellagic acid and placebo (independent sample *t* test)

P2: Comparison of mean of baseline characteristics in each group at baseline and end of study (paired sample *t* test)

In their clinical trial that required 89 people with sleep disorders, intervention group received a 50,000-unit vitamin D supplement, one in a fortnight for 8 weeks. At the end of the study sleep score reduced significantly in vitamin recipients as compared with placebo recipients Majid et al. [20]. However, in a study conducted on healthy adults, the impact of resveratrol supplementation on cognitive function, sleep quality, and blood flow was assessed. No significant differences were observed in the use of resveratrol and improvement of sleep quality [21]. One of the reasons that mentioned study did not observe the beneficial effect of resveratrol on sleep quality was probably that healthy people with no sleep disorders were included in the study. Due to the fact that a detailed study on the effect of antioxidant supplementation in diabetics on sleep quality has not been performed and its exact mechanism is not fully known, but by summarizing scientific results from various studies, this conclusion can be reached that receiving antioxidants can improve sleep quality by reducing oxidative stress and inflammation [22]. In patients with sleep disorders, reoxygenation/hypoxia cycles occur intermittently during

Table 3 The comparison of the sleep score and sleep subgroup score changes in the study groups at the beginning and the end of the study

Variables		Mean ± SD placebo($n = 21$)	Mean ± SD Ellagic acid ($n = 21$)	P1
PSQI (score)	Baseline	8.92 ± 1.39	8.86 ± 2.07	0.103
	End	8.77 ± 2.04	5.39 ± 1.11	0.033
	P2	0.1	0.001	
Sleep duration (h)	Baseline	5.07 ± 1.18	5.69 ± 1.2	0.41
	End	5.63 ± 1.91	7.47 ± 2.1	0.002
	P2	0.304	0.011	
Sleep latency (min)	Baseline	52.07 ± 18.14	52.1 ± 14.21	0.504
	End	47.18 ± 13.09	29.22 ± 9.13	0.02
	P2	0.306	0.001	
Sleep efficiency (%)	Baseline	79.64 ± 17.3	80.81 ± 16.12	0.401
	End	81.11 ± 17.8	88.01 ± 19.02	0.04
	P2	0.108	0.03	
Sleep disturbances (score)	Baseline	2.07 ± 0.25	2.1 ± 0.36	0.37
	End	1.84 ± 0.2	0.95 ± 0.1	0.03
	P2	0.11	0.003	
Use of sleep medications (time per week)	Baseline	2.4 ± 0.49	2.32 ± 0.33	0.23
	End	2.1 ± 0.23	0.94 ± 0.06	0.019
	P2	0.27	0.01	
Day time dysfunction (score)	Baseline	2.05 ± 0.5	1.96 ± 0.31	0.51
	End	1.88 ± 0.46	0.7 ± 0.13	0.031
	P2	0.23	0.001	
Subjective sleep quality (score)	Baseline	1.71 ± 0.32	1.8 ± 0.52	0.32
	End	1.68 ± 0.17	1.04 ± 0.23	0.041
	P2	0.35	0.03	

Data are expressed as means ± SD

P1: Comparison of the mean of sleep score and sleep subgroup score between the two groups of Ellagic acid and placebo (independent sample *t* test)

P2: Comparison of mean of baseline sleep score and sleep subgroup score changes in each group at baseline and end of study (paired sample *t* test)

each hour of sleep [23]. These cycles result in the production of ROS. Antioxidant enzymes in the body and antioxidants such as melatonin neutralize their harmful effects [2]. Also, these bioactive substances such as polyphenols can inhibit the oxidation of the fatty acids in the nervous system's membrane cell and reduce the production of malondialdehyde (MDA), leading to the treatment of this disorder. According to the results of scientific studies, the amount of MDA in people with poor sleep quality increases [8]. It is known that by reducing the oxidation of fats in the cell membrane, insulin resistance is reduced and the glycemic status of diabetic patients is improved [2]. Decreased insulin resistance due to EA intake can improve sleep quality. On the other hand, the inverse relationship between advanced glycation end products (AGEs) and sleep quality has been observed in human studies [24]. Getting antioxidants by preventing the formation of AGEs such as hemoglobin A_1c (HbA_1c) can improve glycemic status and oxidative stress in these patients [25]. In the cross-sectional study conducted by Hedayati et al., significant relation was observed between hemoglobin A1C levels and sleep quality [24]. Meanwhile, the role of inflammation in people with sleep disorders has been of interest to researchers and extensively studied. Sleep disorders, by disrupting the regular secretion of hormones that regulate metabolic pathways, can induce inflammatory cytokines such as interleukin-1 (IL-1) and tumor necrosis factor alpha (TNFα) that are active in the central nervous system [26]. According to scientific research reports, increased inflammation, fat oxidation, and production of MDA are associated. The intake of EA antioxidant, by reducing inflammation and IR, leads to a decrease in oxidative stress and thereby increases the quality of sleep.

One of the strengths of this study is that for the first time, the effect of pure supplement of EA was investigated in diabetic patients on quality of sleep. Also, the design of this study is a double-blind randomized clinical trial that had parallel groups, making the results of this study remarkable. However, a small number of the participant is an important limitation considering

the large number of potential variables that can affect both glycemic control and other DM-related variables and the variables affecting the sleep quality itself. Comorbid illnesses and having other medications are examples of these variables. It should be noted that in order to draw clinical conclusions and examine the clinical effects, it is necessary to conduct a study with a larger number of participants and intervention period.

Conclusion

In conclusion, the results of our study indicated that supplementation with EA, 180 mg/day for 8 weeks, in diabetic patients, improved the quality of sleep. These results provide evidence to support the view that polyphenol antioxidant group with reducing the complications of diabetes can play an important role in enhancing quality of sleep in people with metabolic diseases.

Acknowledgments The authors would like to thank all of the participants who completed the study protocol. This work was financially supported by a Grant (Number: IR.QUMS.REC.1398.079) from Vice-Chancellor for Research Affairs of Qazvin University of Medical Sciences, Qazvin, Iran.

Authors' details MG: MSc, Department of Nutrition, School of Health, Qazvin University of Medical Sciences.

SH: Department of Internal Medicine, Velayat Clinical Research Development Unit, Qazvin University of Medical Sciences.

MRN: Department of Pharmacology and Toxicology, School of Pharmacy, Ahvaz Jundishapur University of Medical Sciences.

MK: Department of Tissue Engineering and Applied Cell Sciences, Faculty of Advanced Technologies in Medicine, Tehran University of Medical Sciences.

HKH: Metabolic Diseases Research Center, Research Institute for Prevention of Non-Communicable Diseases, Qazvin University of Medical Sciences.

Authors' contributions All persons who met authorship criteria are listed as authors. M Ghadimi, M Kavianpour, and H Khadem Haghighian contributed significantly to the work's conception, participated in the writing and critical revision of the manuscript in a manner sufficient to establish the ownership of the intellectual content. S Hashemipour and M Rashidi Nooshabadi contributed significantly to the work's conception. H Khadem Haghighian analyzed and interpreted data. M Ghadimi and M Kavianpour involved in the design of work. All authors approved the final version of the manuscript to be published.

Funding information This work was financially supported by a Grant (Number: IR.QUMS.REC.1398.079) from Vice-Chancellor for Research Affairs of Qazvin University of Medical Sciences, Qazvin, Iran. This investigation was extracted from the Master of Science thesis of Zahra Mirzaie, student of Qazvin University of Medical Sciences.

Data availability Data from this project will not be shared because additional results from the study are yet to be published.

Compliance with ethical standards

Ethics The study was approved by the Ethical Committee of Qazvin University of Medical Sciences with grant number of IR.QUMS.REC.1398.079 and also registered by the identification code IRCT20141025019669N13 in clinical trial registry of Iran. Informed consent was obtained from these individuals to participate in the study.

Conflict of interest The authors declare that they have no conflict of interest.

Ethics approval and consent to participate The protocol of the study after approving with the ethic committee of Qazvin University of Medical Sciences, Qazvin, Iran, with grant number of Number: IR.QUMS.REC.1398.079, was registered in the Iranian Registry of Clinical Trials website by the IRCT20141025019669N13 code. Also, written informed consent was given by all the participants before their inclusion in the study.

References

1. Öztürk E, Arslan AKK, Yerer MB, Bishayee A. Resveratrol and diabetes: a critical review of clinical studies. Biomed Pharmacother. 2017;95:230–4.
2. Dos Santos JM, Tewari S, Mendes RH. The role of oxidative stress in the development of diabetes mellitus and its complications. J Diabetes Res. 2019;2019.
3. Buzdar MJ, Ahmed F, Mengal A, Hameed T, Abbas F, Tahir HM, et al. Study on the complications by chronic diabetes mellitus type II in Quetta Balochistan. Pak-Euro J Med Life Sci. 2019;2(3):65–8.
4. Fasil A, Biadgo B, Abebe M. Glycemic control and diabetes complications among diabetes mellitus patients attending at University of Gondar Hospital, Northwest Ethiopia. Diabetes Metab Syndr Obes: Targets Ther. 2019;12:75.
5. Goldberg RB, Bray GA, Marcovina SM, Mather KJ, Orchard TJ, Perreault L, et al. Non-traditional biomarkers and incident diabetes in the diabetes prevention program: comparative effects of lifestyle and metformin interventions. Diabetologia. 2019;62(1):58–69.
6. Tracy EL, Berg CA, Baucom KJ, Turner SL, Kelly CS, Van Vleet M, et al. Daily sleep quality and daily stressors in couples coping with type 1 diabetes. Health Psychol. 2019;38(1):75–83.
7. Telford O, Diamantidis CJ, Bosworth HB, Patel UD, Davenport CA, Oakes MM, et al. The relationship between Pittsburgh sleep quality index subscales and diabetes control. Chronic Illn. 2019;15(3):210–9.
8. Barakat S, Abujbara M, Banimustafa R, Batieha A, Ajlouni K. Sleep quality in patients with type 2 diabetes mellitus. J Clin Med Res. 2019;11(4):261–6.
9. Borges YG, Cipriano LHC, Aires R, Zovico PVC, Campos FV, de Araújo MTM, et al. Oxidative stress and inflammatory profiles in obstructive sleep apnea: are short-term CPAP or aerobic exercise therapies effective? Sleep Breath. 2019:1–9.
10. Hahad O, Prochaska JH, Daiber A, Muenzel T. Environmental noise-induced effects on stress hormones, oxidative stress, and vascular dysfunction: key factors in the relationship between cerebrocardiovascular and psychological disorders. Oxidative Med Cell Longev. 2019;2019:1–13.
11. BenSaad LA, Kim KH, Quah CC, Kim WR, Shahimi M. Anti-inflammatory potential of ellagic acid, gallic acid and punicalagin

A&B isolated from Punica granatum. BMC Complement Altern Med. 2017;17(1):47.
12. Falsaperla M, Morgia G, Tartarone A, Ardito R, Romano G. Support ellagic acid therapy in patients with hormone refractory prostate cancer (HRPC) on standard chemotherapy using vinorelbine and estramustine phosphate. Eur Urol. 2005;47(4):449–55.
13. Craig C, Marshall A, Sjostrom M, Bauman A, Lee P, Macfarlane D et al (2017) International Physical Activity Questionnaire-Short Form
14. Mollayeva T, Thurairajah P, Burton K, Mollayeva S, Shapiro CM, Colantonio A. The Pittsburgh sleep quality index as a screening tool for sleep dysfunction in clinical and non-clinical samples: a systematic review and meta-analysis. Sleep Med Rev. 2016;25:52–73.
15. Hosseini B, Saedisomeolia A, Wood LG, Yaseri M, Tavasoli S. Effects of pomegranate extract supplementation on inflammation in overweight and obese individuals: a randomized controlled clinical trial. Complement Ther Clin Pract. 2016;22:44–50.
16. Thorpy M. International classification of sleep disorders. Sleep disorders medicine. Springer;2017. p 475–84.
17. Tauman R, Shalitin S, Lavie L. Oxidative stress in obese children and adolescents with and without type 2 diabetes mellitus is not associated with obstructive sleep apnea. Sleep Breath. 2019;23(1):117–23.
18. Dalvi LT, Moreira DC, Andrade R Jr, Ginani J, Alonso A, Hermes-Lima M. Ellagic acid inhibits iron-mediated free radical formation. Spectrochim Acta A Mol Biomol Spectrosc. 2017;173:910–7.
19. Alzoubi KH, Khabour OF, Rashid BA, Damaj IM, Salah HA. The neuroprotective effect of vitamin E on chronic sleep deprivation-induced memory impairment: the role of oxidative stress. Behav Brain Res. 2012;226(1):205–10.
20. Majid MS, Ahmad HS, Bizhan H, Hosein HZM, Mohammad A. The effect of vitamin D supplement on the score and quality of sleep in 20–50 year-old people with sleep disorders compared with control group. Nutr Neurosci. 2018;21(7):511–9.
21. Wightman EL, Haskell-Ramsay CF, Reay JL, Williamson G, Dew T, Zhang W, et al. The effects of chronic trans-resveratrol supplementation on aspects of cognitive function, mood, sleep, health and cerebral blood flow in healthy, young humans. Br J Nutr. 2015;114(9):1427–37.
22. Kanagasabai T, Ardern CI. Inflammation, oxidative stress, and antioxidants contribute to selected sleep quality and cardiometabolic health relationships: a cross-sectional study. Mediat Inflamm. 2015;2015:1–11.
23. Morris G, Stubbs B, Köhler CA, Walder K, Slyepchenko A, Berk M, et al. The putative role of oxidative stress and inflammation in the pathophysiology of sleep dysfunction across neuropsychiatric disorders: focus on chronic fatigue syndrome, bipolar disorder and multiple sclerosis. Sleep Med Rev. 2018;41:255–65.
24. Hedayati A, Gholampour Y, Dehghan A. The relation between sleep disorders and hemoglobin A1c levels in patients with type II diabetes mellitus. Med J Mashhad Univ Med Sci. 2016;59(3):179–87.
25. Talaei B, Amouzegar A, Sahranavard S, Hedayati M, Mirmiran P, Azizi F. Effects of cinnamon consumption on glycemic indicators, advanced glycation end products, and antioxidant status in type 2 diabetic patients. Nutrients. 2017;9(9):991.
26. Ryan S. Adipose tissue inflammation by intermittent hypoxia: mechanistic link between obstructive sleep apnoea and metabolic dysfunction. J Physiol. 2017;595(8):2423–30.

International Journal of Diabetes in Developing Countries (2021) 41:37–42
https://doi.org/10.1007/s13410-020-00861-0

ORIGINAL ARTICLE

Association of CAPN10 (SNP-19) genetic polymorphism and obesity with T2DM: a study on Bengali Hindu caste population

Pranabesh Sarkar[1] · Diptendu Chatterjee[1] · Arup Ratan Bandyopadhyay[1]

Received: 12 December 2019 / Accepted: 10 August 2020 / Published online: 1 September 2020

Abstract

Background Type 2 diabetes mellitus (T2DM) is a multifactorial disease caused due to induced obesity and the influence of genetic polymorphism. Calpain-10 (CAPN10) gene and its genetic polymorphism (SNP-19) significantly alter insulin sensitivity assayed by the synthesis of a mutant protein and/or altered transcriptional regulation, which could contribute to the diabetes risk. The role of CAPN10 genetic polymorphism in T2DM has widely been studied in different populations and obtained variable results. Considering the variable results in different ethnic groups, the present study is an attempt to understand the association of CAPN10 (SNP-19) genetic polymorphism and obesity with T2DM in Bengali Hindu caste population.

Methods The present study consisted of 104 clinically diagnosed type 2 diabetes mellitus male patients (Age range = 23–80 years) and 176 apparently healthy males (Age range = 18–79 years) without T2DM and family history of T2DM from Bengali Hindu caste population. Genomic DNA was isolated from mouthwash using phenol-chloroform method with slight modifications. PCR method has been used to perform genotyping of (CAPN10 snp-19). Anthropometric and physiological variables have been collected by using standard method.

Results Overall, the distribution CAPN10 (SNP-19) genotypes revealed no significant difference between T2DM patients and control group. However, T2DM patients with II genotype have significantly ($p < 0.05$) higher WHR compared to ID and DD genotypes. Individuals with DD genotype demonstrated significant ($p < 0.05$) higher risk associated with T2DM than (II+ID) genotype with increase in BMI, WC, and HC.

Conclusions The findings of the present study might incorporate as criterion for early prognosis of T2DM in Bengali Hindu caste.

Keywords CAPN10 · Genetic polymorphism · Obesity · Hazard ratio · Bengali Hindu caste

Introduction

Type 2 diabetes mellitus (T2DM) is a metabolic disorder characterized by high blood glucose levels due to defects in insulin secretion from pancreatic β cells and peripheral insulin resistance [1, 2]. The global prevalence of T2DM is increasing rapidly and will reach around 366 million in 2030 [3]. The International Diabetes Federation (2019) reported that the prevalence of diabetes in India was 77 million, that is, 10.4% of the total adult population [4]. Genetic polymorphism and induced obesity mainly trigger the progression of T2DM [5, 6].

✉ Pranabesh Sarkar
p.sarkar1991@gmail.com; psanthro_rs@caluniv.ac.in

[1] Department of Anthropology, University of Calcutta, 35 Ballygunge Circular Road, Kolkata, West Bengal 700019, India

Calpain-10 (CAPN10) is a T2DM candidate gene [7] identified through positional cloning and genome-wide association studies [8]. It is located on chromosome 2q 37.3, comprised of 15 exons spanning 31 kb of genomic sequence and encodes a 672- amino-acid intracellular protease [9]. CAPN10 gene consists of an isoform-specific large subunit (80 kDa) and a common small subunit (30 kDa) that functions as intracellular calcium–dependent cysteine protease in calcium-regulated signalling pathways [1]. The gene is expressed at the mRNA and protein levels by several tissue types, with different mRNA isoforms, those involved in the regulation of glucose homeostasis, such as pancreatic β islet cells, liver, skeletal muscle and adipocytes [10]. Genetic polymorphisms (SNP-43, SNP-63, SNP-19) of CAPN10 have been involved in the development of T2DM and obesity [11]. CAPN10 (SNP 19) might alter the risk of T2DM by affecting its transcriptional regulation. In addition to that, SNP-19 significantly alters insulin sensitivity assayed by the synthesis of a mutant

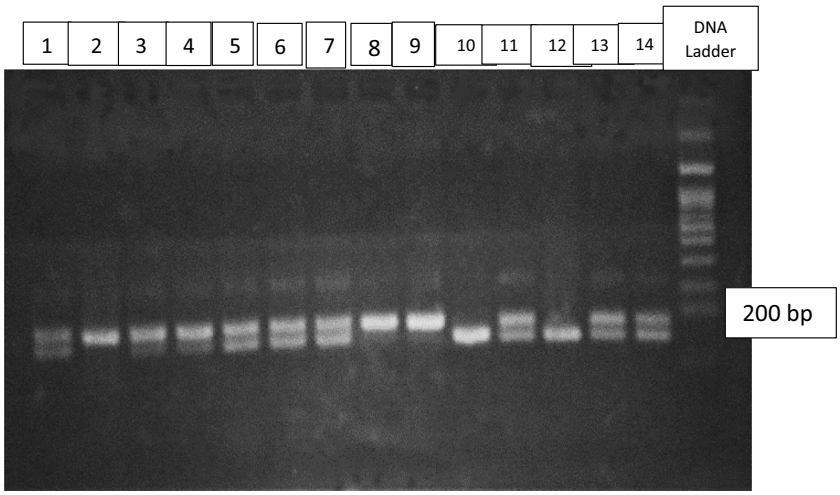


Fig. 1 Gel documentation of CAPN10 Snp-19 (I/D) polymorphism

Lane 1, 3, 4, 5, 6, 7, 11, 13 and 14 shows ID genotype

Lane 2, 8 and 9 shows II genotype

Lane 10 and 12 shows DD genotype

protein and/or altered transcriptional regulation, which could contribute to the diabetes risk [5].

Initially, the association between SNP-43 and T2DM was reported. GG genotypes (SNP-43) were found as factors for reducing CAPN10 mRNA expression level in skeletal muscle. That leads to decreased rate of insulin-mediated glucose turnover or insulin resistance [12]. Several studies on linkage and a high-risk haplotype combination of SNP-43, SNP-19, and SNP-63 were found to have 2–3-fold increased risk of T2DM due to elevated fatty acid and insulin resistance in Mexican Americans, Finns and Germans [7, 9, 10, 13, 14], British population, Pima Indians, African Americans and South Indians [13].However, the variable results have been observed in Japanese [9] and Samoans [15].

Considering the variable results in different ethnic groups, the effect of genetic polymorphism with induced obesity might yield imperative result in Bengali Hindu caste population for pathogenesis of T2DM. On this background, the present study is an attempt to understand the association of CAPN10 (SNP-19) genetic polymorphism and obesity with T2DM in Bengali Hindu caste population.

Materials and methods

The present study consisted of 104 clinically diagnosed Type 2 diabetes mellitus male patients (age range = 23–80 years) from Bengali Hindu caste population. The T2DM cases were identified as individuals having a fasting plasma glucose level of more than 126 mg/dL. The T2DM patients have also been selected by physician recommendation.

One hundred seventy-six apparently healthy males (age range = 18–79 years) participated from the same group as controls. The healthy controls have been selected as those with a fasting blood glucose level below110 mg/dL, and none of them have any family history of T2DM.

Bengali Hindu caste is an endogamous ethnolinguistic group of northern-south Asia. Purposive sampling method was used to select participants from Kolkata and its suburban area of West Bengal, India. Data has been collected from November 2017 to January 2019 in several phases.

Genomic DNA was isolated from mouthwash using the phenol-chloroform method [16] with slight modifications.

Table 1 Distribution of CAPN10 genotypes and alleles among control group and T2DM patients

	CAPN10 (SNP-19) genotypes			Total	*p* value	Allele frequency	
	II	ID	DD			I	D
Control	45 (25.6)	98 (55.7)	33 (18.8)	176	$p = 0.454$	0.53	0.47
T2DM patients	30 (28.8)	50 (48.1)	24 (23.1)	104		0.52	0.48
Total	75	148	57	280			

*Value in the (parentheses) denotes percentage

Table 2 Distribution of anthropometric and physiological variables among control group and T2DM patients classified according to CAPN10 (SNP-19) genotypes

Variables	Control group (mean ± SD) $n = 176$				T2DM patients (mean ± SD) $n = 104$			
	II ($n = 45$)	ID ($n = 98$)	DD ($n = 33$)		II ($n = 30$)	ID ($n = 50$)	DD ($n = 24$)	
PBF	29.04 ± 7.23	30.42 ± 7.00	27.54 ± 7.50	$p = 0.121$	32.69 ± 7.27	33.31 ± 6.39	31.45 ± 6.89	$p = 0.545$
BMI	23.66 ± 4.37	24.92 ± 4.46	22.43 ± 3.70	$p = 0.013$*	25.62 ± 3.43	26.05 ± 3.68	23.90 ± 4.01	$p = 0.065$
WC (cm)	87.39 ± 10.7	88.86 ± 10.48	86.68 ± 9.37	$p = 0.507$	96.78 ± 11.0	93.75 ± 10.0	89.90 ± 10.7	$p = 0.062$
HC (cm)	93.14 ± 7.69	94.19 ± 9.58	91.68 ± 7.64	$p = 0.355$	96.06 ± 8.14	97.02 ± 9.40	94.40 ± 8.56	$p = 0.497$
WHR	.9374 ± .07	.9451 ± .079	.9457 ± .07	$p = 0.841$	1.00 ± .06	.9673 ± .063	.9512 ± 058	p = 0.004*
SBP (mmHg)	133.58 ± 20.5	139.30 ± 22.7	131.24 ± 21.7	p = 0.121	144.40 ± 24.6	141.34 ± 21.7	145.42 ± 24.3	$p = 0.735$
DBP (mmHg)	79.60 ± 11.84	80.36 ± 10.62	78.03 ± 13.0	$p = 0.598$	78.17 ± 10.4	79.58 ± 14.3	82.42 ± 10.47	$p = 0.455$
MAP (mmHg)	97.59 ± 13.10	100.00 ± 13.14	95.76 ± 15.0	$p = 0.255$	100.24 ± 13.4	100.17 ± 15.0	103.41 ± 13.25	$p = 0.625$

*$p < 0.05$

The quantity and quality of DNA was checked using OD (optical density) by spectrophotometry and identified by using 1% agarose gel electrophoresis and visualized in UV transilluminator with ethidium bromide staining.

The PCR method has been used to perform genotyping of (CAPN10 snp-19) genetic polymorphism using (Forward Primer-5_- GTTTGGTTCTCTTCAGCGTGGAG-3_, Reverse Primer-5_-CATGAACCCTGGCAGGGTCTAAG-3). PCR was performed in thermocycler with 10 μl volume containing 10× PCR buffer, 10 mM dNTPs, 25 mM MgCl2, 250 nM of each primer, 1 U of Hot start Taq and 50 ng DNA. The PCR cycle was as follows: initialization was carried out at 95 °C for 15 min followed by 35 cycles for denaturation (94 °C for 1 min), annealing (60 °C for 1 min) and extension (72 °C for 1 min). The final extension or elongation was carried out at 72 °C for 10 min. The amplified PCR products were verified in 3% agarose gel electrophoresis with ethidium bromide staining. Allele D (two repeats of 32-bp sequence) produced band in 155 bp region of DNA ladder and allele I (three repeats) produced band in 187 bp region of DNA ladder (Fig. 1).

Anthropometric data (WC, HC) has been collected by using standard method [17].Waist-hip ratio was calculated using standard formula (waist circumference/hip circumference). Percent body fat (PBF) and body mass index (BMI) were obtained from OMRON bio electrical impendence (Hbf-375) strictly following the manufacturer manual. Blood pressure (SBP, DBP) was measured at resting condition of an individual using Omron Automatic blood pressure monitor. Mean arterial pressure (MAP) has been calculated by using standard formula [MAP = DBP + 1/3(SBP-DBP)].

Allele frequencies were computed by using maximum likelihood estimation [18]. Statistical analysis was performed using SPSS-25 software. The cut-off was set as $p = 0.05$. Inferential statistics (chi-square) was used to find differences in the genotypic distribution of CAPN10 (SNP-19). However, one-way

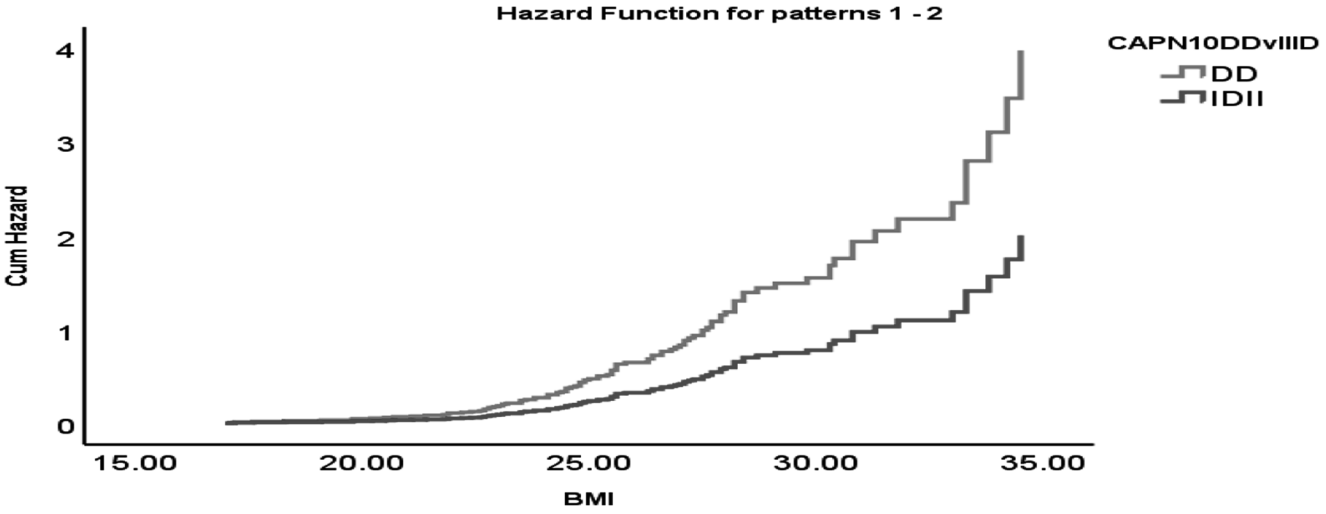


Fig. 2 Hazard ratio (HR) of T2DM by BMI in Bengali Hindu caste classified according to CAPN10 DD vs (II + ID) genotypes

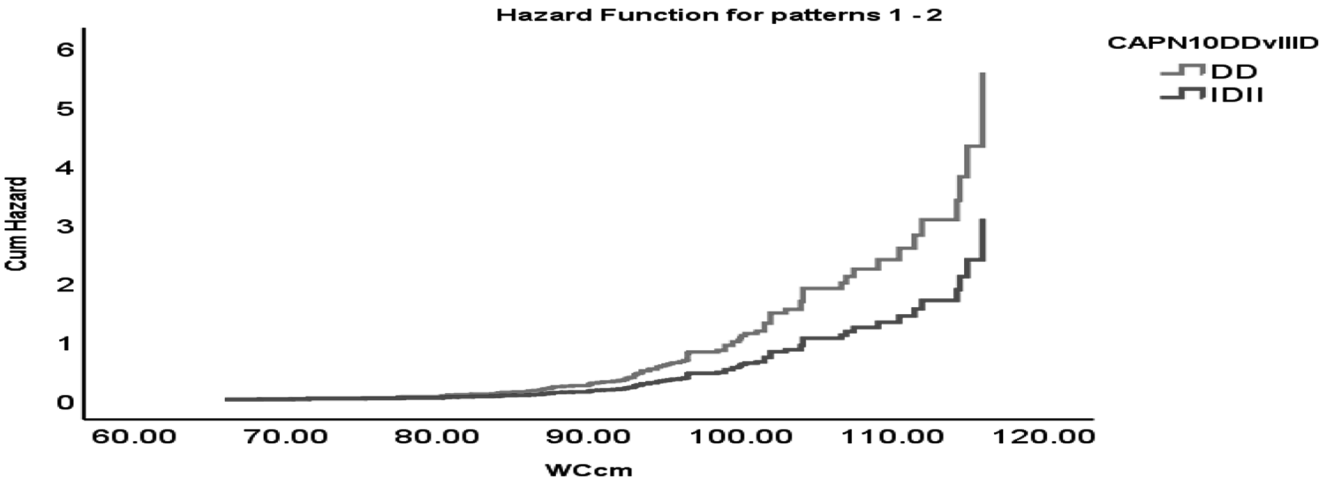


Fig. 3 Hazard ratio (HZ) of T2DM by WC in Bengali Hindu caste classified according to CAPN10 DD vs (II + ID) genotypes

ANOVA was performed to compare the distribution of anthropometric and physiological variables among control group and T2DM patients according to CAPN10 (SNP-19) genotypes. To find out independent risk factor of T2DM, Cox regression analysis was performed to access the magnitude of risk.

Results

Distribution of CAPN10 genotypes and alleles among control group and T2DM patients in Table 1 showed no difference in the genotypic distribution between the controls and T2DM patients and found to be in Hardy-Weinberg equilibrium among the controls and T2DM patients. Homozygous genotypes (II and DD) were comparatively higher in T2DM patients (28.8% and 23.1%) than controls (25.6% and 18.8%).

Examination on CAPN10 (SNP-19) genotypes revealed (Table 2), among the T2DM patients, II genotype was found to have significantly ($p < 0.05$) higher WHR compared with ID and DD genotypes. However, no significant association was observed in PBF, BMI, WC, HC, SBP, DBP and MAP with CAPN10 genotypes among T2DM patients. In case of control group, ID genotype was found to have significantly ($p < 0.05$) higher BMI compared with II and DD genotypes. However, no significant association was observed in PBF, WC, HC, WHR, SBP, DBP and MAP with CAPN10 genotypes. To assess the hazard ratio, the Cox regression analysis was performed where BMI, WC and HC were used as independent risk factors for T2DM and discerned that the DD genotype has 1.98 times [HR = 1.98(1.244–3.153, p = 0.004)] more risk associated with T2DM than (II + ID) genotype with increase in BMI (Fig. 2). A similar trend was observed with increased risk in WC as 1.8 times [HR = 1.8 (1.13–2.88, $p = 0.01$)] (Fig. 3) followed by HC as 1.6 times [HR = 1.6 (1.037–2.608, $p = 0.03$)] (Fig. 4).

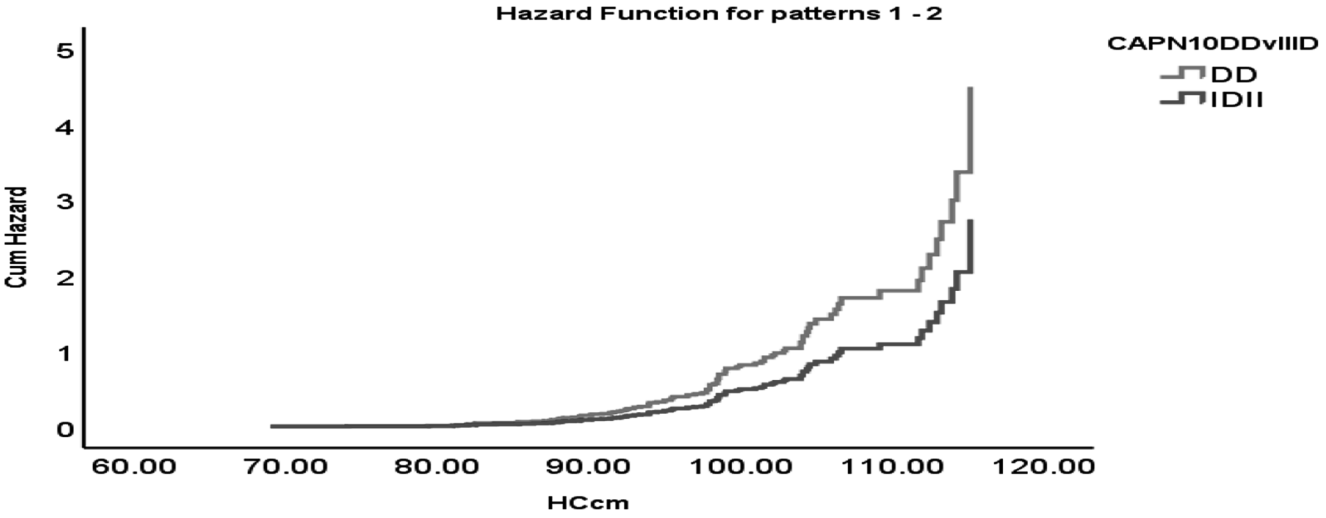


Fig. 4 Hazard ratio (HZ) of T2DM by HC in Bengali Hindu caste classified according to CAPN10 DD vs (II + ID) genotypes

Discussion

CAPN10 has been identified as a candidate gene for T2DM susceptibility [7]. The role of CAPN10 genetic polymorphism has also been observed in human brain development [19], cognitive impairment in Cerebral Small Vessel Disease (SVD) patients [20], cardiomyopathy [21] and Polycystic Ovary Syndrome (PCOS) [6, 22]. Genetic polymorphisms of CAPN10 (SNP-44, SNP-43, SNP-19, SNP-63) have widely been studied to assess the risk of T2DM in different populations [5]. To fulfil the objective of the present study, CAPN10 (SNP-19) genetic polymorphism has been selected to find its association with T2DM. The present study found slightly higher T2DM risk with presence of homozygous (II and DD) allele compared with heterozygous ID allele. However, no significant genotypic differences of CAPN10 (SNP-19) genetic polymorphism were observed between T2DM patients and control among the Bengali Hindu caste population. In spite of the heterogenous nature of the Indian population, the findings of the present study corroborate with earlier works conducted in the Eastern Indian population [23], Southern Indian population [7] and Banias and Jat Sikh of North-west Indian population [5]. A similar trend has been found in terms of no association between CAPN10 (SNP-19) genetic polymorphism and T2DM among Mexican mestizos [24], Koreans [9], population of UK [25], Finnish [26] and Javanese [27].

In contrast, the homozygous II allele of CAPN10 (SNP-19) genetic polymorphism had increased the risk of T2DM in Gujrati people [21], Tunisian Arabs [1], Spanish [28] and the Caucasian population [29]. The higher Insulin resistance index (HOMA index) was found to be associated with the homozygous II genotype for development of T2DM [28]. On the other hand, the DD genotype of CAPN10(SNP-19) genetic polymorphism had significantly enhanced T2DM risk among patients of the Tunisian Arab population [1] and Turkish population[30].

Additionally, haplotype combination of CAPN10 (SNP-44/43/19/63 1121/1121) with presence of D allele of SNP-19 was associated with increased risk of T2DM in Finnish [10]. Another haplotype combination of SNP-43/19/63 112/121 with presence of I allele of SNP-19 was found to be associated with higher risk in T2DM patients of South Indians [31] and Pima Indians [32].

Obesity is one of the main risk factors for non-communicable diseases like hypertension, coronary heart disease and Type 2 diabetes [33]. Genetic polymorphism as well as genetic predisposition to obesity or excess weight is a strong risk factor for T2DM [34], although no significant differences were observed in anthropometric (except WHR) variables pertaining to obesity and physiological variables according to CAPN10 (SNP-19) genotypes among male T2DM patients in Bengali Hindu caste . Waist-hip ratio (WHR) was significantly increased in homozygous II genotype compared with heterozygous ID and homozygous DD genotypes in male T2DM patients of the Bengali Hindu caste group. Similarly, studies in Turkish individuals [35], Colombian population [36], Tunisian Arabs [1] and north-west Indians [5] revealed significant association between BMI and homozygous II genotypes of CAPN10 (SNP-19). Contrary to previous studies, no association has been found between CAPN10 (SNP-19) genetic polymorphism and BMI in Mexican [27], Javanese [37] and Finnish [26] T2DM patients.

Moreover, hazard risk assessment revealed that DD genotype has significant risk associated with T2DM than II + ID genotypes with increase in BMI, WC and HC. This finding contradicts with work in Brahmins of north-west India, where the II genotype has 1.5-fold increased risk associated with T2DM than (ID/DD) genotypes with the increase in BMI [5].

The inconsistency of the results has been obtained due to genetic heterogeneity and ethnic variability. Ethnolinguistic group like Bengali Hindu caste population has no effect of CAPN10 (SNP-19) genetic polymorphism in T2DM but, the effect of homozygous II allele of CAPN10 (SNP-19) genetic polymorphism was observed in central obesity (WHR) among T2DM patients of the studied population. However, the combination of CAPN10 (SNP-19) genetic polymorphism and obesity markers revealed CAPN10 (SNP-19) DD genotypes with increased obesity have significant impact on progression of T2DM. Further studies in large sample size will validate the result and include these facts as a criterion for early prognosis of T2DM.

Acknowledgments The authors are grateful to the participants of the present study.

Funding information The University Grant Commission (UGC-NET JRF Fellowship) and UPE-II grant provided the financial support.

Compliance with ethical standards

The present study has been approved by the Institutional Ethical Committee (Ref No. 009/17-18/1682 dated-30/11/2017).

Conflict of interest The authors declare that they have no conflict of interest.

Informed consent The data was obtained from each participant prior to data collection.

References

1. Ezzidi I, Turki A, Messaoudi S, Chaieb M, Kacem M, Al-Khateeb GM, et al. Common polymorphisms of calpain-10and the risk of type 2 diabetes in a Tunisian Arab population: a case-control study. BMC Med Genet. 2010;11:75.
2. Ridderstråle M, Nilsson E. Type 2 diabetes candidate gene CAPN10: first, but not last. Curr Hypertens Rep. 2008;10(1):19–24. https://doi.org/10.1007/s11906-008-0006-1.

3. Raza ST, Abbas S, Ahmed F, Fatima J, Zaidi ZH, Mahdi F. Association of MTHFR and PPARγ2 gene polymorphisms in relation to type 2 diabetes mellitus cases among north Indian population. Gene. 2012;511:375–9.
4. IDF. IDF Diabetic Atlas (9th edition). International Diabetes Federation. 2019.
5. Sharma R, Matharoo K, Kapoor R, Chopra H, Bhanwer A. Ethnic differences in CAPN10 SNP-19 in type 2 diabetes: a North-West Indian case-control study and evidence from meta-analysis. Genet Res (Camb). 2013;95:146–55.
6. Márquez JL, Pacheco A, Valdés P, Salazar LA. Association between CAPN10 UCSNP-43 gene polymorphism and polycystic ovary syndrome in Chilean women. Clin Chim Acta. 2008;398(1–2):5–9. https://doi.org/10.1016/j.cca.2008.07.028.
7. Bodhini D, Radha V, Ghosh S, Sanapala KR, Majumder PP, Rao MR, et al. Association of calpain 10 gene polymorphisms with type 2 diabetes mellitus in Southern Indians. Metabolism. 2011;60:681–8.
8. Cheekurthy AJP, Rambabu C, Kumar A. Gene as a risk factor for type 2 diabetes mellitus and its related complications. Biomed Res Clin Prac. 2016;1(1):14–7.
9. Kang ES, Kim HJ, Nam M, Nam CM, Ahn CW, Cha BS, et al. A novel 111/121 diplotype in the Calpain-10 gene is associated with type2 diabetes. J Hum Genet. 2006;51:629–33.
10. Orho-Melander M, Klannemark M, Svensson MK, Ridderstråle M, Lindgren MC, Groop L. Variants in the calpain-10 gene predispose to insulin resistance and elevated free fatty acid levels. Diabetes. 2002;51:2658–64.
11. Nam JS, Han JW, Lee SB, et al. Calpain-10 and adiponectin gene polymorphisms in Korean type 2 diabetes patients. Endocrinol Metab (Seoul). 2018;33(3):364–71. https://doi.org/10.3803/EnM.2018.33.3.364.
12. Baier LJ, Permana PA, Yang X, et al. A calpain-10 gene polymorphism is associated with reduced muscle mRNA levels and insulin resistance. J Clin Invest. 2000;106(7):R69–73. https://doi.org/10.1172/JCI10665.
13. Cox NJ, Hayes G, Roe CA, Tsuchiya T, Bell GI. Linkage of Calpain 10 to type 2 diabetes: the biological rationale. Diabetes. 2004;53(1):S19–S-25. https://doi.org/10.1007/s00438-015-1097-4.
14. Malecki MT, Moczulski DK, Klupa T, Wanic K, Cyganek K, Frey J, et al. Homozygous combination of calpain 10 gene haplotypes is associated with type 2 diabetes mellitus in a Polish population. Eur J Endocrinol. 2002;146:695–9.
15. Tsai HJ, Sun G, Weeks DE, et al. Type 2 diabetes and three calpain-10 gene polymorphisms in Samoans: no evidence of association. Am J Hum Genet. 2001;69(6):1236–44. https://doi.org/10.1086/324646.
16. Zayats T, Young TL, Mackey DA, Malecaze F, Calvas P, Guggenheim J. Quality of DNA extracted from mouthwashes. PLoS One. 2009;4:1–5.
17. Weiner JS, Lourie JA. Practical human biology. London: Academic Press; 1981.
18. Cavalli-Sforza LL, Bodmer WF. The genetics of human populations. New York: W.H Freeman and Company; 1971.
19. Oladnabi M, Musante L, Larti F, et al. New evidence for the role of calpain 10 in autosomal recessive intellectual disability: identification of two novel nonsense variants by exome sequencing in Iranian families. Arch Iran Med. 2015;18(3):179–84.
20. Wu K, Cai Y. The SNP43 (G/A) polymorphism in CAPN10 gene confers an increased risk of cognitive impairment in cerebral small vessel disease. J Clin Lab Anal. 2018;32(9):e22615. https://doi.org/10.1002/jcla.22615.
21. Mistry KN, Sanjay L, Solanki A, Thakkar A. Calpain 10 gene polymorphism and its association in cardiomyopathy and type 2 diabetes. Int J Adv Diabetes Res. 2013;1(1):16–22.
22. Dasgupta S, Sirisha PV, Neelaveni K, Anuradha K, Reddy BM. Association of CAPN10 SNPs and haplotypes with polycystic ovary syndrome among South Indian Women. PLoS One. 2012;7(2):e32192. https://doi.org/10.1371/journal.pone.0032192.
23. Adak S, Sengupta S, Chowdhury S, Bhattacharyya M. Co-existence of risk and protective haplotypes of Calpain 10 gene to type 2 diabetes in the eastern Indian population. Diab Vasc Dis Res. 2010;7:63–8.
24. Picos-Cardenas VJ, Sainz-Gonzalez E, Miliar-García A, Romero-Zazueta A, Quintero-Osuna R, Leal-Ugarte E, et al. Calpain-10 gene polymorphisms and risk of type 2 diabetes mellitus in Mexican mestizos. Genet Mol Res. 2015;14(1):2205–15.
25. Evans JC, Frayling TM, Cassell PG, Saker PJ, Hitman GA, Walker M, et al. Studies of association between the gene for calpain-10 and type 2 diabetes mellitus in the United Kingdom. Am J Hum Genet. 2001;69:544–52.
26. Pihlajamäki J, Salmenniemi U, Vänttinen M, et al. Common polymorphisms of calpain-10 are associated with abdominal obesity in subjects at high risk of type 2 diabetes. Diabetologia. 2006;49(7):1560–6. https://doi.org/10.1007/s00125-006-0270-z.
27. Tursinawati Y, Kartikadewi A, Hakim RF. Association of CAPN10 SNP-19 (rs3842570) Polymorphism on fasting plasma glucose, blood pressure and body mass index of Javanese type-2 diabetes patients. Int Conf Food Sci Technol. 2019;292:012031.
28. Saez ME, Gonzalez-Sanchez JL, Ramírez-Lorca R, Martinez-Larrad MT, Zabena C, Gonzalez A, et al. The CAPN10 gene is associated with insulin resistance phenotypes in the Spanish population. PLoS One. 2008;3(8):e2953. https://doi.org/10.1371/journal.pone.0002953.
29. Elbein SC, Chu W, Ren Q, Hemphill C, Schay J, Cox NJ, et al. Role of calpain-10 gene variants in familial type 2 diabetes in Caucasians. J Clin Endocrinol Metab. 2002;87(2):650–4.
30. Bayramci NS, Acik L, Kalkan C, Yetkin I. Investigation of glucocorticoid receptor and calpain-10 gene polymorphisms in Turkish patients with type 2 diabetes mellitus. Turk J Med Sci. 2017;47:1568–75.
31. Cassell PG, Jackson AE, North BV, et al. Haplotype combinations of calpain 10 gene polymorphisms associate with increased risk of impaired glucose tolerance and type 2 diabetes in South Indians. Diabetes. 2002;51(5):1622–8. https://doi.org/10.2337/diabetes.51.5.1622.
32. Lynn S, Evans JC, White C, et al. Variation in the calpain-10 gene affects blood glucose levels in the British population. Diabetes. 2002;51(1):247–50. https://doi.org/10.2337/diabetes.51.1.247.
33. Leitner DR, Frühbeck G, Yumuk V, et al. Obesity and Type 2 Diabetes: two diseases with a need for combined treatment strategies - EASO Can Lead the Way. Obes Facts. 2017;10(5):483–92. https://doi.org/10.1159/000480525.
34. Khan IA, Movva S, Shaik NA, et al. Investigation of Calpain 10 (rs2975760) gene polymorphism in Asian Indians with gestational diabetes mellitus. Meta Gene. 2014;2:299–306. Published 2014 Apr 17. https://doi.org/10.1016/j.mgene.2014.03.001.
35. Arslan E, Acik L, Gunaltili G, Ayvaz G, Altinova AE, Arslan M. The effect of calpain-10 gene polymorphism on the development of type 2 diabetes mellitus in a Turkish population. Endokrynologia Polska. 2014;65:90–5.
36. Orozco AC, Muñoz AM, Velásquez CM, Uscátegui RM, Parra MV, Patiño FA, et al. Variant in CAPN10 gene and environmental factors show evidence of association with excess weight among young people in a Colombian population. Biomédica. 2014;34(4):546–55. https://doi.org/10.7705/biomedica.v34i4.2246.
37. Meza-Espinoza JP, Leal-Ugarte E, Peralta-Leal V, Flores-Villarreal LE, Picos-Cardenas VJ, Sainz-Gonzalez E, et al. Association of CAPN10 SNP-19 with metabolic traits in Mexican patients with type 2 diabetes. Int J Hum Genet. 2019;19(1):48–53.

International Journal of Diabetes in Developing Countries (2021) 41:43–47
https://doi.org/10.1007/s13410-020-00870-z

ORIGINAL ARTICLE

Association analysis of *HHEX* gene variant with type 2 diabetes risk

Pantea Banihashemi[1] · Hamid Reza Aghaei Meybodi[2] · Mahdi Afshari[3] · Negar Sarhangi[2] · Mandana Hasanzad[1]

Received: 27 March 2020 / Accepted: 1 September 2020 / Published online: 18 September 2020

Abstract

Background Type 2 diabetes mellitus (T2DM) is a common, chronic, and complex disorder that is influenced by interactions between genetic and environmental factors. The *hematopoietically expressed homeobox* (*HHEX*) gene, which affects insulin sensitivity and secretion, is a candidate gene for the pathogenesis of T2DM.

Objective The purpose of this study was to investigate the effect of the *HHEX* genetic variant, rs1111875, on the T2DM risk in a group of Iranian patients for the first time.

Methods A total of 108 T2DM patients and 100 normal subjects were genotyped using polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP).

Results There was significant difference in genotypes and alleles frequency of rs1111875 between both case and control groups ($p < 0.001$). The frequency of the G allele was significantly higher in cases (87.5%) compared with controls (27%) (OR = 18.92, CI = 11.06–32.69, $p < 0.001$).

Conclusion Our findings suggest that the rs1111875 variant of the *HHEX* gene could be considered a strong risk factor for T2DM development.

Keywords *HHEX* · rs1111875 · T2DM · Genetic variant

Introduction

Type 2 diabetes mellitus (T2DM) is a complex, metabolic, multifactorial disorder resulting from interactions between genetic and environmental factors [1–3]. T2DM is caused by insulin resistance in muscles and fat cells, decreased insulin secretion, and increased hepatic glucose production [4, 5]. Patients with T2DM may suffer from macrovascular and microvascular complications such as cardiovascular disease, neuropathy, nephropathy, and retinopathy, which are associated with a marked morbidity and mortality, decrease the quality of life, and have negative economic effects on countries [6, 7].

Genome-wide association studies (GWAS) have shown a large number of genes associated with T2DM, including *TCF7L2*, *SLC30A8*, *IGF2BP2*, *CDKN2A*, *CDKN2B*, *CDKAL1*, *HHEX*, *EXT2*, *LOC387761*, *FTO*, *PPARG*, *KCNQ1*, and *JAZF1* [2, 8]. Identifying more genetic variants can help to predict and prevent T2DM [9]. In 2007, *hematopoietically expressed homeobox* (*HHEX*) was introduced as a new locus for T2DM [8]. It encodes a transcription factor involved in Wnt signaling and is required for pancreatic development and differentiation.

HHEX gene locus is on chromosome 10q23. It contains four exons with the size of 5.7 kb and encodes a protein which contains 270 amino acids [10]. Rs1111875 variant is located in a region nearby the end of a 270-kb linkage disequilibrium block of telomeric part on chromosome 10 [11]. Rs1111875 is near an exon of *HHEX* gene; therefore, no changes happen in the amino acid sequence [12]. Rs1111875 variant is crucial to ventral pancreatic development. The *HHEX* gene is a second most powerful biological candidate given the postulated

✉ Hamid Reza Aghaei Meybodi
dr_aghai@yahoo.com

✉ Mandana Hasanzad
mandanahasanzad@yahoo.com

[1] Medical Genomics Research Center, Tehran Medical Sciences, Islamic Azad University, Khaghani Avenue, Shariati St, Tehran 193951459, Iran

[2] Personalized Medicine Research Center, Endocrinology and Metabolism Clinical Sciences Institute, Tehran University of Medical Sciences, No.10- Jalal -e-Ale-Ahmad Street, Chamran Highway, Tehran 1411713119, Iran

[3] Department of Community Medicine, Zabol University of Medical Sciences, Zabol, Iran

effects on both insulin signaling and islet function [10, 13]. Various studies reported *HHEX*/insulin-degrading enzyme variants (rs1111875) and the response of insulin secretion following a glucose load, which suggests that the *HHEX* gene can influence the T2D risk primarily by affecting β-cell function [13, 14].

Accordingly, it is a strong candidate gene for the pathogenesis of T2DM and is known to affect the insulin secretion and pancreatic function.

The aim of the present study was to investigate the association of the rs1111875 variant in the *HHEX* gene with T2DM development among a group of Iranian patients.

Methods

Two hundred and eight subjects, including 108 T2DM patients and 100 healthy controls, were enrolled in this study. Diabetes patients were selected according to the American Diabetic Association (ADA) criteria [15].

A 5-ml blood sample was collected from each subject in EDTA-coated tubes and DNA was extracted using the standard salting out method [16]. Genotyping of the *HHEX* rs1111875 variant was performed using the polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP) method. The 161-bp fragment of the *HHEX* gene was amplified using 5′-CATCATAACTTCTCACTCCC TTCC-3′ as the forward primer and 5′- GCTGCTTA TGGAAACTGCATTACT-3′ as the reverse primer [10] in a 25-μl PCR reaction mixture containing 1-μl template DNA, 10-μl Red Master Mix (Amplicon), 1 μl of each primer, and 12-μl dH2O. Thermocycling conditions were as follows: 95 °C for 1 min followed by 35 cycles of 1 min at 95 °C, 40 s at 60 °C, and 40 s at 74 °C, with a final incubation at 74 °C for 5 min.

The XbaI restriction enzyme was used to digest the PCR product at 37 °C for about 20 h. In the presence of the A allele, the 161-bp PCR product was cleaved into two fragments, 111 bp and 50 bp (AA homozygote genotype). The fragment of the PCR product was not digested when the restriction enzyme-cutting site was at base G, with the initial segment size being 161 bp (GG genotype). Three fragments, 161 bp, 111 bp, and 50-bp, represented the AG heterozygote genotype [17]. The digested fragments were separated by electrophoresis on a 2% agarose gel. Some PCR products were randomly sequenced to confirm the results (Fig. 1).

Table 1 Comparing cases and controls regarding different factors

Factors		Normal	T2DM	*p* value
Sex	Male	34 (34)	37 (34.26)	0.969
	Female	66 (66)	71 (65.74)	
Ethnicity	Fars	50 (50)	34 (31.48)	0.007
	Others	50 (50)	74 (68.52)	
Age		38.78 (14.89)	54.36 (10.93)	0.0001

T2DM, type 2 diabetes mellitus

Results

Totally, 108 patients with T2DM were compared with 100 normal subjects. As illustrated in Table 1, gender frequency was similar in cases and controls ($p = 0.969$). The results showed that Fars ethnicity was significantly less frequent in the diabetic patients vs. normal subjects (31.48 vs. 50% respectively, $p = 0.007$). Moreover, the cases were significantly older than controls (54.36 vs. 38.78, respectively, $p = 0.0001$). The frequency of the GG genotype was 82.41% and 27% and the frequency of the AG genotype was 10.19% and 0% in the cases and controls, respectively ($p < 0.001$). After adjusting for the effect of ethnicity, the adjusted odds ratio for AG and GG genotypes were 120.82 ($p < 0.001$) and 32.32 ($p < 0.001$), respectively (Table 2).

The frequency of the G allele was significantly higher in cases vs. controls (87.5 vs. 27%, respectively, $p < 0.001$) (Table 3). The mean (SD) hemoglobin A1c (HbA1c) was 8.63 (1.50), 7.94 (1.73), and 7.89 (1.73) in diabetic patients with AA, AG, and GG genotypes, respectively (Table 4) ($p = 0.313$).

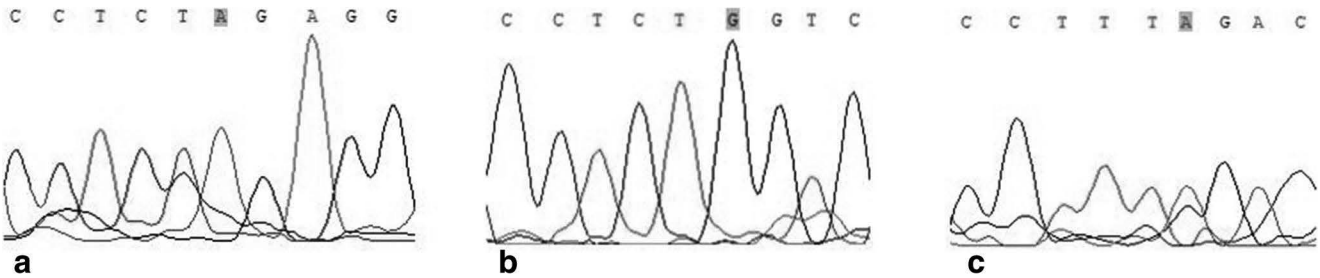


Fig. 1 Results of DNA sequencing for rs1111875 of *HHEX* gene. **a** AA genotype. **b** GG genotype. **c** AG genotype

Table 2 Genotypes distribution of rs1111875 *HHEX* gene variant in T2DM ($n = 108$) and controls ($n = 100$) and their Crude and adjusted association with T2DM risk

Genotype	Normal	T2DM	*p* value	Crude OR	*p* value	95% CI	Adjusted OR	*p* value	95% CI
AA	73 (73)	8 (7.41)	< 0.001	1			1		
AG	0 (0)	11 (10.19)		120.58	< 0.001	17.55-inf	120.82	< 0.001	17- inf*
GG	27 (27)	89 (82.41)		29.40	< 0.001	12–79.73	32.32	< 0.001	13–91
AA	73 (73)	8 (7.41)	< 0.001	1			1		
AG+GG	27 (27)	100 (92.59)		33.80	< 0.001	14–78.65	38.13	< 0.001	15.75–92

T2DM, type 2 diabetes mellitus; *OR*, odds ratio; *CI*, confidence interval

Discussion

Many GWAS have described common variants in different populations and have identified many single nucleotide polymorphisms (SNPs) in association with type 2 diabetes [4]. This study found a significantly higher genotype frequency of the *HHEX* rs1111875 variant in patients with T2DM compared with the normal population. The homozygous GG genotype of this variant significantly increased the odds of diabetes mellitus by more than 30%. In addition, the heterozygous genotype caused an approximately 120-fold increase in the risk of diabetes mellitus. Moreover, allelic investigation showed that G allele carriers had an approximately 19-fold higher risk of developing diabetes mellitus compared with A allele carriers. Therefore, this allele can be a strong risk factor for diabetes mellitus. Sladek et al. were the first researchers who found that rs1111875 was located near the telomeric end of the chromosome 10. They also showed the *HHEX* was essential for hepatic and pancreatic development, a target of Wnt signaling pathway in the French population. They suggested a significant association between the G allele, as a risk allele, and type 2 diabetes [11]. In 2008, Furukawa et al. studied 405 cases and 340 controls in Japan and found that the G allele of the rs1111875 variant of the *HHEX* gene was associated with T2D, which was in agreement with Sladek et al. [18]. According to a study of 1529 cases and 1439 controls by Lin et al., the G allele of the rs1111875 variant of the *HHEX* gene is significantly more frequent in the diabetic patients than controls with a risk allele frequency of 0.32. Moreover, it is associated with T2D in the Chinese population [19]. This association in the Chinese population was further proven by Hu et al. in a study of 1849 T2D patients and 1785 controls with an almost similar risk allele frequency [20]. In another study, Chauhan et al. found an association between the G allele (risk allele) of the rs1111875 variant and T2D in a study of 2486 patients and 2678 controls in an Indian population with a risk allele frequency of 0.47 [21]. Kifagi et al. found an association between increased risk of T2D and the presence of the GG genotype of the rs1111875 variant in a Tunisian population comprising 403 healthy controls and 331 patients [1]. Similarly, van Vliet-Ostaptchouk et al [22]. reported an association between the G allele of the rs1111875 variant as the risk allele with a risk allele frequency of 0.60 and T2D in a Dutch population including 501 T2D patients and 920 healthy controls [22].

There are two other studies in Iranian ethnicity; Galavi et al. [23] have indicated dramatically increase in the risk of T2D in southeast Iranian population, which is similar to the result of the current study; in the second report by Mansoori et al. [24], a significant association was found between GG genotype and the G allele of the rs1111875 (A/G) SNP and susceptibility to T2DM that is consistent with our findings. However, in genetic association analysis, the ethnicity is the most important issue which should be considered, as Iran consists of Iranian Persians (Fars), who constitute 51% of Iran's population and the rest of the population are Iranian Azeris (24%) and Iranian Gilakis and Mazandaranis, Iranian Kurds, Iranian Arabs, Iranian Lurs, Iranian Balochs Iranian Turkmen, and others [25]. Consequently, in the genetic studies, we should include all of the mentioned ethnicities according to the reported percentages. One of the important factor that is seen in the present study was inclusion of the ethnicities which has not been observed in the previous studies in Iranian population. The second strength point of the present study in comparison with the abovementioned studies in Iranian

Table 3 Alleles distribution of rs1111875 *HHEX* gene variant in T2DM ($n = 108$) and controls ($n = 100$) and their association with T2DM risk

Alleles	Normal (%)	T2DM (%)	*p* value	OR	*p* value	95% CI
A	73	12.5	< 0.001	1		
G	27	87.5		18.92	< 0.001	11.06–32.69

T2DM, type 2 diabetes mellitus; *OR*, odds ratio; *CI*, confidence interval

Table 4 Mean HbA1C among T2DM patients with different genotypes of the rs1111875 variant

Genotype	Frequency	Mean	SD	*p* value
AA	8	8.63	1.50	0.313
AG	11	7.94	1.73	
GG	89	7.89	1.73	

SD, std. deviation

population was the association analysis between genotype and HbA1c as ADA has recommended HbA1c as a possible substitute to fasting blood glucose for diagnosis of diabetes [26].

In line with the results of the present study, all the above studies showed a link between the GG genotype and type 2 diabetes in different populations. However, this study showed a higher odds ratio (OR) for the risk allele.

However, some studies found no significant association between the rs1111875 variant of the *HHEX* gene and T2D. Some examples include a study by Tabara et al. in a Japanese population of 506 diabetic patients and 402 controls (p value = 0.602) [27], a study conducted by Wen et al. in 1165 T2D patients and 1136 healthy controls in China [28], and a study by Phani et al. in an Indian population [8].

Conclusion

The results of the present study provided an evidence that the rs1111875 variant of the *HHEX* gene could be considered a strong risk factor for diabetes mellitus. Further studies with larger sample sizes are required to confirm the role of this variant in developing diabetes mellitus.

Acknowledgments The researchers would like to express their gratitude to all participants enrolled in this project.

Author contributions MH managed the project and provided guidance to the research; PB and NS drafted the manuscript and performed lab genotyping; MA performed the statistical analysis; HRAM provided clinical guidance to the research.

All authors contributed to and approved the final version of the manuscript.

Data availability Data from this project will be available to share.

Compliance with ethical standards

Ethical approval and consent to participate This study was approved by Ethical committee (ID research ethics: IR.IAU.PS.REC.1398.031) and informed consent was obtained from all individual participants included in the study.

Competing interests The authors declare that they have no conflict of interest.

References

1. Kifagi C, Makni K, Boudawara M, Mnif F, Hamza N, Abid M, et al. Association of genetic variations in TCF7L2, SLC30A8, HHEX, LOC387761, and EXT2 with type 2 diabetes mellitus in Tunisia. Genet Test Mol Biomarkers. 2011;15(6):399–405.
2. Shahbazian H, Aleali AM, Amani R, Namjooyan F, Cheraghian B, Latifi SM, et al. Effects of saffron on homocysteine, and antioxidant and inflammatory biomarkers levels in patients with type 2 diabetes mellitus: a randomized double-blind clinical trial. Avicenna J Phytomed. 2019;9(5):436–45.
3. Hindy G, Mollet IG, Rukh G, Ericson U, Orho-Melander M. Several type 2 diabetes-associated variants in genes annotated to WNT signaling interact with dietary fiber in relation to incidence of type 2 diabetes. Genes Nutr. 2016;11(1):6.
4. Votsi C, Toufexis C, Michailidou K, Antoniades A, Skordis N, Karaolis M, et al. Type 2 diabetes susceptibility in the Greek-Cypriot population: replication of associations with TCF7L2, FTO, HHEX, SLC30A8 and IGF2BP2 polymorphisms. Genes. 2017;8(1):16.
5. García-Chapa EG, Leal-Ugarte E, Peralta-Leal V, Durán-González J, Meza-Espinoza JP. Genetic epidemiology of type 2 diabetes in Mexican mestizos. Biomed Res Int. 2017;2017:1–10.
6. Zeng C, Zhou Z, Han Y, Wen Z, Guo C, Huang S, et al. Interactions of TRAF6 and NLRX1 gene polymorphisms with environmental factors on the susceptibility of type 2 diabetes mellitus vascular complications in a southern Han Chinese population. J Diabetes Complicat. 2017;31(12):1652–7.
7. Wu Y, Ding Y, Tanaka Y, Zhang W. Risk factors contributing to type 2 diabetes and recent advances in the treatment and prevention. Int J Med Sci. 2014;11(11):1185–200.
8. Phani NM, Adhikari P, Nagri SK, D'Souza SC, Satyamoorthy K, Rai PS. Replication and relevance of multiple susceptibility loci discovered from genome wide association studies for type 2 diabetes in an Indian population. PLoS One. 2016;11(6):e0157364.
9. Meybodi HRA, Hasanzad M, Larijani B. Path to personalized medicine for type 2 diabetes mellitus: reality and hope. Acta Med Iran. 2017:166–74.
10. Xu P, Che Y, Cao Y, Wu X, Sun H, Liang F, et al. Polymorphisms of TCF7L2 and HHEX genes in Chinese women with polycystic ovary syndrome. J Assist Reprod Genet. 2010;27(1):23–8.
11. Sladek R, Rocheleau G, Rung J, Dina C, Shen L, Serre D, et al. A genome-wide association study identifies novel risk loci for type 2 diabetes. Nature. 2007;445(7130):881–5.
12. Lee Y-H, Kang ES, Kim SH, Han SJ, Kim CH, Kim HJ, et al. Association between polymorphisms in SLC30A8, HHEX, CDKN2A/B, IGF2BP2, FTO, WFS1, CDKAL1, KCNQ1 and type 2 diabetes in the Korean population. J Hum Genet. 2008;53(11–12):991–8.
13. Lu CC, Chen YT, Chen SY, Hsu YM, Lin CC, Tsao JW, et al. Hematopoietically expressed homeobox gene is associated with type 2 diabetes in KK Cg-A(y)/J mice and a Taiwanese Han Chinese population. Exp Ther Med. 2018;16(1):185–91.
14. Wang Y, Qiao W, Zhao X, Tao M. Quantitative assessment of the influence of hematopoietically expressed homeobox variant (rs1111875) on type 2 diabetes risk. Mol Genet Metab. 2011;102(2):194–9.
15. Association AD. Standards of medical care in diabetes—2017 abridged for primary care providers. Clinical diabetes: a publication of the American Diabetes Association. 2017;35(1):5.
16. Miller S, Dykes D, Polesky H. A simple salting out procedure for extracting DNA from human nucleated cells. Nucleic Acids Res. 1988;16(3):1215.
17. Abdul-Hassan IA, Al-Tamimi MT, Al-dujaily SS. The association of HHEX gene polymorphism with the incidence of polycystic

ovary syndrome (PCOS) in Iraq. World J Pharm Res. 2014;3(10): 287–98.
18. Furukawa Y, Shimada T, Furuta H, Matsuno S, Kusuyama A, Doi A, et al. Polymorphisms in the IDE-KIF11-HHEX gene locus are reproducibly associated with type 2 diabetes in a Japanese population. J Clin Endocrinol Metab. 2008;93(1):310–4.
19. Lin Y, Li P, Cai L, Zhang B, Tang X, Zhang X, et al. Association study of genetic variants in eight genes/loci with type 2 diabetes in a Han Chinese population. BMC Med Genet. 2010;11:97.
20. Hu C, Zhang R, Wang C, Wang J, Ma X, Lu J, et al. PPARG, KCNJ11, CDKAL1, CDKN2A-CDKN2B, IDE-KIF11-HHEX, IGF2BP2 and SLC30A8 are associated with type 2 diabetes in a Chinese population. PLoS One. 2009;4(10):e7643.
21. Chauhan G, Spurgeon CJ, Tabassum R, Bhaskar S, Kulkarni SR, Mahajan A, et al. Impact of common variants of PPARG, KCNJ11, TCF7L2, SLC30A8, HHEX, CDKN2A, IGF2BP2, and CDKAL1 on the risk of type 2 diabetes in 5,164 Indians. Diabetes. 2010;59(8):2068–74.
22. van Vliet-Ostaptchouk JV, Onland-Moret NC, van Haeften TW, Franke L, Elbers CC, Shiri-Sverdlov R, et al. HHEX gene polymorphisms are associated with type 2 diabetes in the Dutch Breda cohort. Eur J Hum Genet. 2008;16(5):652–6.
23. Galavi H, Mollashahee-Kohkan F, Saravani R, Sargazi S, Noorzehi N, Shahraki H. HHEX gene polymorphisms and type 2 diabetes mellitus: a case-control report from Iran. J Cell Biochem. 2019;120(10):16445–51.
24. Mansoori Y, Daraei A, Naghizadeh M, Salehi R. The HHEX rs1111875A/G gene polymorphism is associated with susceptibility to type 2 diabetes in the Iranian population. Mol Biol. 2015;49(4): 535–42.
25. Hassan HD. Iran: ethnic and religious minorities. LIBRARY OF CONGRESS WASHINGTON DC CONGRESSIONAL RESEARCH SERVICE.
26. Sherwani SI, Khan HA, Ekhzaimy A, Masood A, Sakharkar MK. Significance of HbA1c test in diagnosis and prognosis of diabetic patients. Biomark Insights. 2016;11:BMI. S38440.
27. Tabara Y, Osawa H, Kawamoto R, Onuma H, Shimizu I, Miki T, et al. Replication study of candidate genes associated with type 2 diabetes based on genome-wide screening. Diabetes. 2009;58(2): 493–8.
28. Wen J, Rönn T, Olsson A, Yang Z, Lu B, Du Y, et al. Investigation of type 2 diabetes risk alleles support CDKN2A/B, CDKAL1, and TCF7L2 as susceptibility genes in a Han Chinese cohort. PLoS One. 2010;5(2):e9153.

International Journal of Diabetes in Developing Countries (2021) 41:48–53
https://doi.org/10.1007/s13410-020-00864-x

ORIGINAL ARTICLE

The value of adenosine deaminase activity in latent autoimmune diabetes in adults and type 2 diabetes patients

Yu Zhou[1] · Wei-Dong Jin[1] · Zhen-Zhen Pang[1] · Jun Xia[1] · Su-Feng Chen[1]

Received: 8 February 2020 / Accepted: 19 August 2020 / Published online: 4 September 2020

Abstract

Objective Adenosine deaminase (ADA) associated with cell-mediated immune responses is involved in many diseases. But little is known about the value of ADA activity in the progression of different diabetes types. The purpose of this study was to compare the ADA level between latent autoimmune diabetes in adults (LADA), type 2 diabetes (T2D) patients, and healthy controls (HC) and analyze its correlation with glycemic parameters and systemic cytokines.

Methods This hospital-based study included 28 LADA patients, 52 T2D patients, and 50 HC. Serum ADA activity and concentrations of inflammatory cytokines were measured. Correlations of ADA level with different indicators were assessed by using spearman's correlation method.

Results Serum ADA activity was significantly higher in T2D patients compared with LADA ($p = 0.008$) and HC ($p < 0.001$). Correlation analysis of ADA with HbA1c% ($r = 0.34$, $p = 0.003$) and inflammatory cytokines (IL-6, $r = 0.31$, $p = 0.007$; IL-10, $r = 0.22$, $p = 0.049$) showed significant positive correlations.

Conclusions Serum ADA activity may reflect the different immunopathogenesis between LADA and T2D patients.

Keywords Adenosine deaminase · Immunopathogenesis · Latent autoimmune diabetes in adults · Type 2 diabetes

Introduction

Diabetes is a heterogeneous disease, which can be classified as many subgroups based on the pathophysiology [1]. The term "latent autoimmune diabetes in adults" (LADA) was first introduced by Tuomi et al. in 1993 to define the type 1.5 diabetes that is positive for antibodies against the insulin producing beta cells but has phenotypical features with type 2 diabetes (T2D) [2]. As we all know, T2D is the most prevalent form of diabetes, accounting for 90–95% of total diabetic cases worldwide [3]. However, owing to the overlap of clinical characteristics, LADA patients are easily misdiagnosed as T2D. Previous articles have revealed that approximately 5–12% T2D cases in European populations are actually misdiagnosed LADA [4, 5]. It is widely accepted that insulin resistance and impaired insulin secretion are the main pathophysiological features of T2D. Comparatively, LADA patients have less insulin secretion and faster insulin dependence progress [6]. Moreover, chronic inflammation plays an important part in the development of insulin resistance and insulin secretion deficiency [7]. Oxidative stress mediators, such as pro-inflammatory cytokines IL-1β, IL-6, and TNF-α, are found to be important pathogenic factors in diabetes [8]. Nonetheless, the role of immune system, such as cellular immunology in the progression of different diabetes types, is not fully understood [9].

Adenosine deaminase (ADA) is an enzyme of purine metabolism that catalyzes the deamination of adenosine and deoxyadenosine into inosine nucleosides. This conversion is associated with the lymphocyte proliferation and differentiation; therefore, ADA is considered as a marker for the assessment of cellular immunity [10]. Moreover, since adenosine can increase the insulin sensitivity by stimulating glucose transport into cells and ADA consumes adenosine in the local microenvironment, ADA activity has been reported to be an indicator of insulin function as well [11]. Although ADA plays an important role in regulating insulin action, scarce reports are available about serum ADA activity in diabetic

✉ Yu Zhou
zhouyu201378@163.com

[1] Department of Clinical Laboratory, Zhejiang Provincial People's Hospital, People's Hospital of Hangzhou Medical College, Zhejiang 310014, Hangzhou, China

subjects, and its significance in the immunopathogenesis of different diabetes types has not been fully elucidated. Thus, the objective of this study was to investigate the ADA level in different types of diabetes and correlate the serum ADA activity with glycemic parameters and systemic cytokines to assess the role of ADA in the immunopathogenesis of LADA and T2D. To our knowledge, there are no published reports comparing the correlation of ADA and systemic cytokines among LADA and T2D patients so far.

Materials and methods

Participants

The study population for the current study consisted of 130 participants recruited in Zhejiang Provincial People's Hospital. Diabetes diagnosis was conformed to the World Health Organization (WHO) 2019 criteria. Patients with any microvascular and macrovascular complications were not excluded. Diagnosis of LADA were (1) no ketosis or ketoacidosis, (2) insulin independence for the first 6 months after diabetes diagnosis, (3) at least one positive islet autoantibody (glutamic acid decarboxylase 65 antibody (GADA) or insulinoma-associated protein-2 antibody (IA-2A)), and (4) age ≥ 30 years at onset of diabetes. Criteria for T2D were (1) negative for islet autoantibodies and (2) no immediate insulin treatment requirement [12]. In addition, patients having secondary diabetes, any malignancy, autoimmune disease, chronic or acute infection, or receiving immunosuppressive treatment were excluded.

General clinical conditions

Clinical data including sex, age, body mass index (BMI), systolic blood pressure (SBP), diastolic blood pressure (DBP), and diabetes duration; laboratory indicators including diabetes-related autoantibodies (GADA, IA-2A, islet cell antibody (ICA), and insulin autoantibody (IAA)), diabetes-unrelated autoantibodies (thyroid peroxidase antibody (TPO) and thyroglobulin antibody (Tg)), glycosylated hemoglobin (HbA1c), fasting plasma glucose (FPG), fasting C-peptide (FCP), and lipid profiles (triglycerides (TG), total cholesterol (TC), low-density lipoproteins cholesterol (LDL-C), and high-density lipoproteins cholesterol (HDL-C)) were recorded. GADA, IA-2A, ICA, and IAA were measured on iFlash 3000 using chemiluminescent immunoassay; TPO and TG were detected by UniCel DxI 800 (Beckman Coulter); high performance liquid chromatography on Bio-Rad was employed for HbA1c; FCP was conducted using electrochemiluminescence immunoassay on cobas e 602; other parameters (FPG, TG, TC, LDL-C, and HDL-C) were assessed by automatic biochemical analyzer (Beckman Coulter AU5821).

ADA and cytokine assays

Peripheral blood samples of all subjects were collected after more than 8 h overnight fasting. After centrifuging, the serums were stored at − 80 °C and thawed only when detected. Activity of serum ADA, which was expressed in enzyme units/liter (U/L), was measured on automatic biochemical analyzer (Beckman Coulter AU5821) using rate assay. ADA coupled with purine nucleoside phosphorylase (PNP), xanthine oxidase (XOD), and peroxidase (POD) were used to determine the catalytic concentration of ADA. Serum concentrations of cytokines IL-2, IL-4, IL-6, IL-10, IL-17A, IFN-γ, and TNF-α were measured by flow cytometry using the BD Cytometric Bead Array (CBA) Human Soluble Protein Flex Set System. CBA made use of microspheres with different fluorescence intensity, which contained capture antibodies that could identify specific proteins. After samples interacted with microspheres and PE-labeled detection antibodies, they were detected by flow cytometry. Data analyses were performed through FCAP Array v3.0. For cytokine concentrations below standard range and out of invertible range, 0.00 pg/mL was assigned. Inter-assay and intra-assay variations were < 20% and < 10%, respectively.

Statistical analysis

Due to lack of normality, the results were expressed as medians and quartiles for continuous variables. For categorical variables, frequencies were used. To examine differences of clinical and laboratory data between different groups, Student's t test, ANOVA, or Wilcoxon rank-sum test was employed for continuous data where appropriate. The Fisher's exact test or chi-square ($\chi 2$) test was used in categorical data with two or more classes. Data were not adjusted for multiple comparisons and were therefore descriptive. Spearman's correlation was used to correlate serum ADA level with different parameters in LADA and T2D patients. $p < 0.05$ was considered statistically significant. SPSS version 18.0 was applied for statistical analyses.

Results

Comparison of general characteristics among different groups

In total, 130 patients (28 LADA patients, 52 T2D patients, and 50 HC) are included in our study; detailed features are shown in Table 1. The median age of diabetes patients was 54.50 years (44.25; 62.75). There were no significant differences

Table 1 Comparison of clinical information and laboratory indicators in LADA, T2D patients, and healthy controls

Characteristics	LADA (n = 28)	T2D (n = 52)	HC (n = 50)	p value			
Demographics				All	LADA VS T2D	LADA vs HC	T2D vs HC
Male (n, %)	21 (75.00%)	39 (75.00%)	33 (66.00%)	0.542	1.000	0.409	0.319
Age (years)	55.00 (48.25; 61.75)	54.00 (43.00; 64.75)	52.00 (49.00; 57.00)	0.783	0.801	0.359	0.904
BMI (kg/m^2)	22.32 (21.32; 25.06)	23.20 (21.67; 25.64)	23.74 (21.09; 25.89)	0.740	0.476	0.501	0.871
SBP (mmHg)	124.00 (107.00; 142.00)	127.00 (116.30; 142.80)	124.00(113.00; 134.30)	0.152	0.321	0.791	0.056
DBP (mmHg)	80.00 (70.00; 88.00)	82.00 (71.00; 89.25)	75.00 (70.00; 83.00)	0.114	0.787	0.210	0.049*
Diabetes duration (years)	2.00 (0.46; 8.50)	2.00 (0.16; 10.00)	0	NA	0.910	NA	NA
Diabetes-related autoantibodies							
GADA (n, %)	19 (67.86%)	0	0	NA	NA	NA	NA
DIA-2A (n, %)	10 (35.71%)	0	0	NA	NA	NA	NA
ICA (n, %)	6 (21.43%)	0	0	NA	NA	NA	NA
IAA (n, %)	6 (21.43%)	0	0	NA	NA	NA	NA
Diabetes-unrelated autoantibodies							
TPO (n, %)	8 (28.57%)	5 (9.62%)	2 (4.00%)	0.008**	0.053[a]	0.003[a] **	0.437[a]
Tg (n, %)	8 (28.57%)	1 (1.92%)	0	NA	0.001[a] **	NA	NA
Other laboratory indicators							
HbA1c (%)	7.50 (6.50; 10.20)	9.95 (8.55; 11.38)	5.30 (5.00; 5.53)	< 0.001***	0.003**	< 0.001***	< 0.001***
FPG (mmol/L)	7.22 (5.54; 8.92)	8.25 (6.77; 10.48)	5.11 (4.85; 5.40)	< 0.001***	0.061	< 0.001***	< 0.001***
FCP (nmol/L)	1.51 (1.12; 2.19)	1.87 (1.29; 2.47)	2.01 (1.59; 2.43)	0.163	0.249	0.045*	0.500
TG (mmol/L)	1.52 (1.32; 2.36)	1.47 (0.98; 2.46)	1.26 (1.04; 1.87)	0.275	0.683	0.091	0.319
TC (mmol/L)	5.23(4.02; 6.12)	4.54 (3.77; 5.55)	4.96 (4.42; 5.74)	0.054	0.151	0.807	0.017*
LDL-C (mmol/L)	2.96 (2.06; 3.87)	2.73 (1.65; 3.27)	2.95 (2.41; 3.51)	0.052	0.071	0.949	0.026*
HDL-C (mmol/L)	1.04 (0.88; 1.26)	0.94 (0.79; 1.15)	1.29 (1.13; 1.44)	< 0.001***	0.095	0.001**	< 0.001***

Data are shown as medians and quartiles. *BMI* body mass index, *SBP* systolic blood pressure, *DBP* diastolic blood pressure, *GADA* glutamic acid decarboxylase 65 antibody, *IA-2A* insulinoma-associated protein-2 antibody, *ICA* islet cell antibody, *IAA* insulin autoantibody, *TPO* thyroid peroxidase antibody, *Tg* thyroglobulin antibody, *HbA1c* glycosylated hemoglobin, *FPG* fasting plasma glucose, *FCP* fasting C-peptide, *TG* triglycerides, *TC* total cholesterol, *LDL-C* low-density lipoproteins cholesterol, *HDL-C* high-density lipoproteins cholesterol, *NA* no analysis. [a] Fisher's exact test. *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$

among three different groups with regard to sex ratio, age, BMI, SBP, and DBP. Diabetes duration between LADA and T2D showed no significant difference, as well. As for laboratory indexes, comparison of diabetes-unrelated autoantibodies among LADA, T2D, and HC yielded significant difference in TPO ($p = 0.008$); the positive rate of Tg was higher in LADA than T2D patients ($p = 0.001$). HbA1c% tended to be higher in T2D patients than LADA ($p = 0.003$) and HC ($p < 0.001$). Although no statistical difference of FPG and HDL-C are found between LADA and T2D patients, diabetic patients (including LADA and T2D) showed higher FPG but lower HDL-C comparing with HC. Other indicators, including FCP, TG, TC, and LDL-C, had no significant differences among the three groups.

ADA and circulating cytokine concentrations in LADA and T2D patients

As shown in Fig. 1, serum ADA activity was significantly higher in T2D patients compared with LADA ($p = 0.008$) and HC ($p < 0.001$). Meanwhile, circulating concentrations of IL-6 ($p = 0.01$) and IL-10 ($p < 0.001$) had significant differences among three different groups; additionally, T2D patients showed higher IL-6 ($p = 0.038$) and IL-10 ($p < 0.001$) level than LADA. Although we also measured serum concentrations of IL-2, IL-4, IFN-γ, and TNF-α, most of them were below the detection limit. Therefore, no further analyses were performed in these cytokines.

Associations of ADA in different diabetes types

When considering both diabetes types together, Spearman's correlation of HbA1c% with serum ADA showed a significant positive correlation ($r = 0.34$, $p = 0.003$). Furthermore, ADA had significant positive associations with IL-6 cytokines ($r = 0.31$, $p = 0.007$) and IL-10 ($r = 0.22$, $p = 0.049$) (Table 2).

Table 2 Correlational study of ADA and the following variables in LADA and T2D patients

Variables	Correlation coefficient	*p* value
Glycemic parameters		
HbA1c (%)	0.34	0.003**
FPG (mmol/L)	0.12	0.285
Cytokines		
IL-6 (pg/mL)	0.31	0.007**
IL-10 (pg/mL)	0.22	0.049*

HbA1c glycosylated hemoglobin, *FPG* fasting plasma glucose. *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$

Discussion

Due to sharing genetic, metabolic, clinical, and immunologic characteristics with both type 1 and type 2 diabetes, LADA is often overlooked in clinical practice with a high rate of misdiagnosis [13, 14]. Biomarkers which can provide insights into the pathogenesis of different diabetes types are urgently needed. Purinergic network is noted a central player in several immune regulation-related diseases, which include diabetes [11]. ADA is the final key factor of adenosine pathway, involved in immunomodulatory by degrading adenosine in an irreversible deamination reaction [15, 16], but its clinical significance in diabetes has not received sufficient attention. There are several reports about ADA levels in diabetes, but uncertainty and controversy still remain. Our study was as a preliminary research to assess the value of serum ADA activity in the immunopathogenesis of LADA and T2D patients.

Generally, patients with LADA are leaner, with lower fasting C-peptide and less metabolic syndrome than T2D [13]. Different from the results of a multicenter study conducted in China [17], LADA patients shared most clinical features with T2D in our cohort. This controversy may be attributed to the differences in diagnostic criteria for LADA patients and

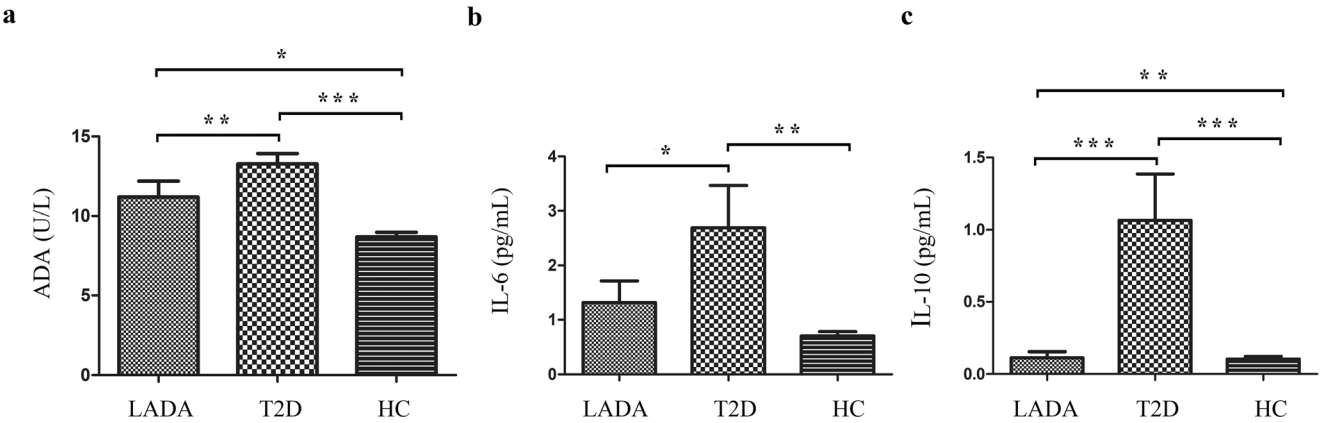


Fig. 1 Serum ADA activity and cytokine concentrations in LADA, T2D patients, and healthy controls. Results were expressed as the mean ± SD. **a** ADA, **b** IL-6, and **c** IL-10. *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$

regional divergence in the populations studied. It has been reported that the variability in the strength of GADA reactivity may partly be associated with substantial heterogeneity of LADA patients [18]. Although the small sample size limited statistical analysis to some extent, the frequency of TPO and Tg tend to be increased in patients with LADA, which was consistent with Schloot et al.'s study [8]. Researches had shown that comparing with normal people, ADA activity was elevated in patients with T2D [19] and gestational diabetes mellitus [20]. We got similar result when comparing serum ADA level between T2D and HC ($p < 0.001$); furthermore, higher ADA was found in T2D patients than LADA ($p = 0.008$). ADA is an important enzyme for regulating the bioactivity of insulin [11], and its activity is increased in the event of oxidative stress and cell membrane damage [21]. The phenomenon that T2D patients tend to have higher levels of ADA activity than LADA may be able to reveal the differences in the progression and severity of cellular immune responses between them.

Diabetes is a strong inflammatory nature disorder; some serum immune mediators have been found to increase in different diabetes types (e.g., systemic cytokines, chemokines, and adhesion molecules) [22]. Compared with LADA patients, IL-6 involved in the genesis of insulin resistance was higher in patients with T2D ($p = 0.038$), which was similar to the results of Pham et al. [22] and Xiang et al. [23]. Contrary to our finding, in the study of Pham et al., systemic concentration of anti-inflammatory cytokine IL-10 showed no differences between patients with LADA and T2D, which possibly because IL-10 was only detected in 44% of individuals in this study. The upregulation of anti-inflammatory mediators may reflect as a feedback to the rise of pro-inflammatory cytokines, associated with an attempt to ameliorate inflammation in patients with diabetes.

Hyperglycemia occurs due to peripheral insulin resistance. In line with other studies [19, 24, 25], the results of correlation analysis indicated that ADA was significantly correlated with HbA1c% ($r = 0.34$, $p = 0.003$), which further elucidated the role of ADA in modulation of insulin activity. Meanwhile, there was a significant positive correlation between ADA and cytokines (IL-6, $r = 0.31$, $p = 0.007$; IL-10, $r = 0.22$, $p = 0.049$), which has been barely studied in diabetes. Several immunological disturbances, including abnormal T-lymphocyte function and low-grade systemic inflammation, are involved in the development of different diabetes types, especially in T2D [26, 27]. ADA is thought to be an indicator of T-cell activation and has a direct correlation with the extent of inflammation [28]. The positive correlation between serum ADA levels and inflammatory cytokines supports that altered blood ADA activity may help predict the extent of immunological response associated with diabetes.

This study also had some limitations. First of all, it was a single-center study with a limited sample size. Secondly, lack of international consensus on the definition of LADA patients might confuse the results of different researches. Furthermore, two major isoenzymes of ADA (ADA1and ADA2) were not detected respectively in our study.

In summary, the assessment of ADA can suggest the heterogeneity of immune response participated in LADA and T2D progression. Its positive correlations with HbA1c% and inflammatory cytokines reveal the potential value of ADA in predicting the severity of diabetes progression in patients. There is a scarcity of information on the differences of ADA activity between LADA and T2D patients. Despite some emerging literature dealing with ADA activity in immune-related disease including diabetes, larger sample studies are needed to confirm these findings.

Acknowledgments The authors would like to thank the clinical and laboratory staff of the Zhejiang Provincial People's Hospital, Hangzhou, China, for facilitating the collection and measurement of the blood samples.

Authors' contributions All authors contributed to the study conception and design. Yu Zhou, literature search and original draft preparation; Wei-Dong Jin, patients recruitment and samples collection; Zhen-Zhen Pang, data analysis and interpretation; Jun Xia, statistical analysis, manuscript preparation, and review; Su-Feng Chen, intellectual content definition, manuscript review, and final approval. All authors read and approved the final manuscript.

Funding The study was supported by the Zhejiang Provincial Medical and Health Research Foundation of China (2017KY006) and the National Natural Science Foundation of China (81902132).

Data availability All the data on which the conclusions of the manuscript rely is presented in the main paper.

Compliance with ethical standards

This study was approved by the ethics committee of Zhejiang Provincial People's Hospital (2020QT223).

Conflict of interest The authors declare no competing interests.

Ethical approval All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Informed consent Informed consent was obtained from all individual participants included in the study.

Consent for publication Not applicable.

Code availability Not applicable.

References

1. Leslie RD, Palmer J, Schloot NC, Lernmark A. Diabetes at the crossroads: relevance of disease classification to pathophysiology

and treatment. Diabetologia. 2016;59(1):13–20. https://doi.org/10.1007/s00125-015-3789-z.
2. Tuomi T, Groop LC, Zimmet PZ, Rowley MJ, Knowles W, Mackay IR. Antibodies to glutamic acid decarboxylase reveal latent autoimmune diabetes mellitus in adults with a non-insulin-dependent onset of disease. Diabetes. 1993;42(2):359–62. https://doi.org/10.2337/diab.42.2.359.
3. Ogurtsova K, da Rocha Fernandes JD, Huang Y, Linnenkamp U, Guariguata L, Cho NH, et al. IDF Diabetes Atlas: global estimates for the prevalence of diabetes for 2015 and 2040. Diabetes Res Clin Pract. 2017;128:40–50. https://doi.org/10.1016/j.diabres.2017.03.024.
4. Liu L, Li X, Xiang Y, Huang G, Lin J, Yang L, et al. Latent autoimmune diabetes in adults with low-titer GAD antibodies: similar disease progression with type 2 diabetes: a nationwide, multicenter prospective study (LADA China Study 3). Diabetes Care. 2015;38(1):16–21. https://doi.org/10.2337/dc14-1770.
5. Zaharieva ET, Velikova TV, Tsakova AD, Kamenov ZA. Prevalence of positive diabetes-associated autoantibodies among type 2 diabetes and related metabolic and inflammatory differences in a sample of the Bulgarian population. J Diabetes Res. 2017;2017:9016148–6. https://doi.org/10.1155/2017/9016148.
6. Zampetti S, Campagna G, Tiberti C, Songini M, Arpi ML, De Simone G, et al. High GADA titer increases the risk of insulin requirement in LADA patients: a 7-year follow-up (NIRAD study 7). Eur J Endocrinol. 2014;171(6):697–704. https://doi.org/10.1530/eje-14-0342.
7. Donath MY, Shoelson SE. Type 2 diabetes as an inflammatory disease. Nat Rev Immunol. 2011;11(2):98–107. https://doi.org/10.1038/nri2925.
8. Schloot NC, Pham MN, Hawa MI, Pozzilli P, Scherbaum WA, Schott M, et al. Inverse relationship between organ-specific autoantibodies and systemic immune mediators in type 1 diabetes and type 2 diabetes: action LADA 11. Diabetes Care. 2016;39(11):1932–9. https://doi.org/10.2337/dc16-0293.
9. Radenkovic M, Silver C, Arvastsson J, Lynch K, Lernmark A, Harris RA, et al. Altered regulatory T cell phenotype in latent autoimmune diabetes of the adults (LADA). Clin Exp Immunol. 2016;186(1):46–56. https://doi.org/10.1111/cei.12834.
10. Passos DF, Bernardes VM, da Silva JLG, Schetinger MRC, Leal DBR. Adenosine signaling and adenosine deaminase regulation of immune responses: impact on the immunopathogenesis of HIV infection. Purinergic Signal. 2018;14(4):309–20. https://doi.org/10.1007/s11302-018-9619-2.
11. Fotino C, Dal Ben D, Adinolfi E. Emerging roles of purinergic signaling in diabetes. Med Chem. 2018;14(5):428–38. https://doi.org/10.2174/1573406414666180226165204.
12. Castelblanco E, Hernandez M, Castelblanco A, Gratacos M, Esquerda A, Mollo A et al. Low-grade inflammatory marker profile may help to differentiate patients with LADA, Classic Adult-Onset Type 1 Diabetes, and Type 2 Diabetes. 2018;41(4):862-8. doi: https://doi.org/10.2337/dc17-1662.
13. Ostergaard JA, Laugesen E, Leslie RD. Should there be concern about autoimmune diabetes in adults? Current Evidence and Controversies. Curr Diab Rep. 2016;16(9):82. https://doi.org/10.1007/s11892-016-0780-0.
14. Mishra R, Hodge KM, Cousminer DL, Leslie RD, Grant SFA. A global perspective of latent autoimmune diabetes in adults. Trends Endocrinol Metab. 2018;29(9):638–50. https://doi.org/10.1016/j.tem.2018.07.001.
15. Ohta A, Sitkovsky M. Extracellular adenosine-mediated modulation of regulatory T cells. Front Immunol. 2014;5:304. https://doi.org/10.3389/fimmu.2014.00304.
16. Ciruela F, Saura C, Canela EI, Mallol J, Lluis C, Franco R. Adenosine deaminase affects ligand-induced signalling by interacting with cell surface adenosine receptors. FEBS Lett. 1996;380(3):219–23. https://doi.org/10.1016/0014-5793(96)00023-3.
17. Zhou Z, Xiang Y, Ji L, Jia W, Ning G, Huang G, et al. Frequency, immunogenetics, and clinical characteristics of latent autoimmune diabetes in China (LADA China study): a nationwide, multicenter, clinic-based cross-sectional study. Diabetes. 2013;62(2):543–50. https://doi.org/10.2337/db12-0207.
18. Tuomi T, Santoro N, Caprio S, Cai M, Weng J, Groop L. The many faces of diabetes: a disease with increasing heterogeneity. Lancet. 2014;383(9922):1084–94. https://doi.org/10.1016/s0140-6736(13)62219-9.
19. Niraula A, Thapa S, Kunwar S, Lamsal M, Baral N, Maskey R. Adenosine deaminase activity in type 2 diabetes mellitus: does it have any role? BMC Endocr Disord. 2018;18(1):58. https://doi.org/10.1186/s12902-018-0284-9.
20. Khosrowbeygi A, Shiamizadeh N, Taghizadeh N. Maternal circulating levels of some metabolic syndrome biomarkers in gestational diabetes mellitus. Endocrine. 2016;51(2):245–55. https://doi.org/10.1007/s12020-015-0697-4.
21. Sargisova YG, Andreasyan NA, Hayrapetyan HL, Harutyunyan HA. Nitric oxide - an activating factor of adenosine deaminase 2 in vitro. Biochemistry. 2012;77(1):92–7. https://doi.org/10.1134/s0006297912010117.
22. Pham MN, Hawa MI, Pfleger C, Roden M, Schernthaner G, Pozzilli P, et al. Pro- and anti-inflammatory cytokines in latent autoimmune diabetes in adults, type 1 and type 2 diabetes patients: Action LADA 4. Diabetologia. 2011;54(7):1630–8. https://doi.org/10.1007/s00125-011-2088-6.
23. Xiang Y, Zhou P, Li X, Huang G, Liu Z, Xu A, et al. Heterogeneity of altered cytokine levels across the clinical spectrum of diabetes in China. Diabetes Care. 2011;34(7):1639–41. https://doi.org/10.2337/dc11-0039.
24. Lee JG, Kang DG, Yu JR, Kim Y, Kim J, Koh G, et al. Changes in adenosine deaminase activity in patients with type 2 diabetes mellitus and effect of DPP-4 inhibitor treatment on ADA activity. Diabetes Metab J. 2011;35(2):149–58. https://doi.org/10.4093/dmj.2011.35.2.149.
25. Kurtul N, Pence S, Akarsu E, Kocoglu H, Aksoy Y, Aksoy H. Adenosine deaminase activity in the serum of type 2 diabetic patients. Acta Med (Hradec Kralove). 2004;47(1):33–5.
26. Zhou T, Hu Z, Yang S, Sun L, Yu Z. Role of adaptive and innate immunity in type 2 diabetes mellitus. 2018;2018:7457269. doi: https://doi.org/10.1155/2018/7457269.
27. Stentz FB, Kitabchi AE. Activated T lymphocytes in Type 2 diabetes: implications from in vitro studies. Curr Drug Targets. 2003;4(6):493–503. https://doi.org/10.2174/1389450033490966.
28. Desrosiers MD, Cembrola KM, Fakir MJ, Stephens LA, Jama FM, Shameli A, et al. Adenosine deamination sustains dendritic cell activation in inflammation. J Immunol. 2007;179(3):1884–92. https://doi.org/10.4049/jimmunol.179.3.1884.

International Journal of Diabetes in Developing Countries (2021) 41:54–62
https://doi.org/10.1007/s13410-020-00851-2

ORIGINAL ARTICLE

Estimation and SVM classification of glucose-insulin model parameters from OGTT data: a comparison with the ADA criteria

Estimation and SM classification of glucose-insulin model parameters

Paola Vargas[1] · Miguel Angel Moreles[1] · Joaquin Peña[1] · Adriana Monroy[2] · Silvestre Alavez[3]

Received: 24 February 2020 / Accepted: 27 July 2020 / Published online: 12 September 2020

Abstract

Background: The oral glucose tolerance test (OGTT) is used for the diagnosis of diabetic conditions. The American Diabetes Association (ADA) provides criteria for definition, namely, impaired fasting glucose (IFG), impaired glucose tolerance (IGT), type 2 diabetes mellitus (T2DM), and normal glucose tolerance (NGT).

Purpose of the study: To examine the application of parameters estimated in models of the glucose-insulin regulatory system during the OGTT, as a classification tool of diabetic conditions.

Methods: Given a set of OGTT data, parameters for each subject are estimated from and ODE model using a Bayesian approach. Point clouds are constructed with parameter pairs and inspected for classification, using a support vector machine (SVM) learning technique. The classical train-test split is used for validation. The training set is comprised of 80 non-related, female volunteers recruited at the Mexico General Hospital.

Results: The parameters peak glucose concentration and average of glucose removal rates are suitable for classification. For the training set, the classification was successful for at least 85% of subjects. Noteworthy, a linear separation suffices. The classification is tested on an independent population of OGTT data for 24 males and 33 females. Classification is successful for 91% of males and 87% of females. Ill patients are correctly classified.

Conclusion: Peak glucose concentration and average of glucose removal rates are proposed as potential patient's indices for diabetic condition. As a graphical tool, clinicians may interpret the SVM classification diagrams. These show a transition from healthy to diabetic. The gray area might suggest pre-diabetic subjects.

Keywords Oral glucose tolerance test · ODE model · Parameter estimation · SVM classification

Introduction

The oral glucose tolerance test (OGTT) is used for the diagnosis of diabetic conditions, namely, impaired fasting glucose (IFG), impaired glucose tolerance (IGT), type 2 diabetes mellitus (T2DM), and normal glucose tolerance (NGT). The American Diabetes Association (ADA) [4] provides criteria for definition.

Glucose curves (patterns) are also fitted to OGTT data by means of statistical methods, in order to extract more information for patient care. Recent studies of this sort are available [5, 10, 11, 14, 15, 19]. In particular, a common aim is to relate OGTT patterns with risk of diabetes.

In this work we are concerned with an alternative approach, to extract information from mathematical models of the underlying dynamics of the glucose-insulin regulatory system during the OGTT. This approach is reviewed [16]. Modeling is a very active research area, current models are complemented with more sophisticated processes [7]. Therein, an integrated model describing both hepatic glucose and pancreatic insulin regulation is introduced.

These models are in terms of ordinary differential equations (ODE), and are of short term in the sense that

✉ Miguel Angel Moreles
moreles@cimat.mx

[1] Computer Science and Mathematics Departments, Centro de Investigación en Matemáticas, Jalisco s/n, Valenciana, 36240 Guanajuato, GTO, Mexico

Extended author information available on the last page of the article.

they describe the dynamics after an external source excites the system. The external source is a load of glucose administered to the patient after an overnight fast. An important application is the computation of a patient's insulin sensitivity from mathematical models linked to clinical protocols [6]. This computation rests on the robust estimation of parameters in the models. This is a classic subject, and most methodologies have been attempted. See the literature review of Parameter Estimation Techniques [18].

Models for the glucose-insulin system can be quite complex, involving several parameters in a system of ordinary differential equations. Consequently, second order minimal models are preferred. As a first test of the methodology to be presented, we use the minimal model of Ackerman et al [1]. This model is probably the simplest, but we shall argue that the obtained results are highly satisfactory to merit the report of our findings.

The basic model is reduced to a single differential equation corresponding to a harmonic oscillator. The state variable, is the deviation from the stable fasting level of the glucose concentration. A simple expression is obtained with four parameters to be estimated for a patient's OGTT data.

This parameter estimation problem is solved following the Bayesian approach. The MAP estimator is chosen from the posterior. The handling of noisy data and assessment of the quality of point estimators, is straightforward in Bayesian estimation. These features have made Bayesian estimation a natural choice. A strong case is made [17]. Therein, a comparison is carried out of Fisherian, such as maximum likelihood, and Bayesian parameter estimation techniques. It is found that Bayesian parameter estimation techniques are less sensitive, in terms of both accuracy and precision, for the estimation of insulin sensitivity in the so-called minimal model of glucose kinetics.

An alternative for aiding on early diagnosis of diabetes is based on learning techniques. As pointed out [9], *the use of intelligent techniques has increased in disease classifications and a lot of research studies have been carried out in making vital medical predictions.* (See a comparison of various classification algorithms therein).

The novelty of this work is on this line, we develop a classification tool to determine a patient's diabetic condition, as well as diabetic risk. In contrast to previous works, instead of the raw data set, we propose two of the estimated parameters in an ODE model for classification.

The proposed parameters are the *peak glucose concentration* and the *average of glucose removal rates*. Classification is accomplished by means of a SVM technique applied to the full population. Noteworthy, a linear kernel suffices.

The analysis was based on actual data gathered at the Mexico General Hospital. For validation, a statistical learning technique is applied. Patients are divided into two populations: a training set to construct the classification and a test set for validation. The former is a population of 80 non-related females. The latter is an independent population of 24 males and 33 females.

The outline is as follows.

In the "Materials and methods" section, we describe the OGTT and introduce the glucose patterns associated with the Ackerman's model. Also, the parameter estimation problem is posed, and SVM classification is described. In the "Technical information" section, the basic glucose-insulin regulatory system model is introduced as well as some technical details on Bayesian estimation. The "Results" and "Discussion" sections present the quantitative results of the case under study and their robustness, and make a case for the potential application on diabetes diagnostics. We close this exposition with conclusions and future lines of research.

Materials and methods

For several years, OGTT data has been collected at the Mexico General Hospital as a monitoring project. In this study, we had access to data from the years 2016 and 2017.

The age range of the subjects was between 18 and 45 years. Anthropometric and clinical information was available, but we stress that our methodology, modeling-estimation-classification, is solely based on raw OGTT data.

An OGTT study and cutoffs for classification

We included 80 non-related, female volunteers, recruited at the Mexico General Hospital. They did not take any medication known to affect glucose tolerance, insulin sensitivity, or insulin secretion.

Volunteers underwent a 2-h OGTT after 8 h overnight fast. They ingested a 75 g dextrose solution (Dextrosol, Hycel, Mexico), and venous blood samples were obtained at 0, 30, 60, 90, and 120 min for determination of plasma glucose and plasma insulin concentrations. Plasma glucose was determined by the glucose oxidase method.

IFG, IGT, and T2DM were defined according to the American Diabetes Association (ADA) 2018 criteria [4]. Namely,

– Diabetes mellitus: fasting plasma glucose $\geq$ 7 mmol/l (126 mg/dl) or 2 h post-load plasma glucose $\geq$ 11.1 mmol/l (200 mg/dl).

– IFG: fasting plasma glucose $\geq$ 5.6 to 6.9 mmol/l (100–125 mg/dl),
– IGT: as 2 h plasma glucose $\geq$ 7.8 to 11 mmol/l (140–199 mg/dl).

The 80 subjects were clustered as follows: 5 IFG, 14 IGT, 7 IFG+IGT, 3 T2DM, and 51 NGT.

OGTT curves from the Ackerman model

In time t, let $G(t)$ be the plasma glucose concentration in the blood and $H(t)$ be the net concentration of hormones that influence the blood glucose levels. For OGTT conditions, insulin is considered predominant and $H(t)$ is essentially its concentration.

It is assumed that after fasting, the patient's concentrations have stabilized to G_0 and H_0.

We study the small deviations

$$g(t) = G(t) - G_0, \quad h(t) = H(t) - H_0.$$

Below, we show that

$$g(t) = Ae^{-\alpha t}\cos(\omega t - \delta). \tag{1}$$

This is the solution of the Ackerman model [1], after the glucose load has been absorbed.

The graph of the function $g(t)$ is the so-called OGTT curve (pattern).

For classification, we consider the parameters:

– A, *peak glucose concentration*,
– α, *average of glucose removal rates*.

The parameters ω and δ need to be estimated, but are of no interest in this work.

The parameter estimation problem given OGTT data

Patients are numbered from $j = 1$ to $j = 80$. Let us define $g_t^j = G^j(t) - G^j(0)$ for $t = 30, 60, 90, 120$.

The problem of concern is as follows: Given OGTT data for patient j: g_{30}^j, g_{60}^j, g_{90}^j, g_{120}^j, estimate the parameters

$$\mathbf{u}_j = (A_j, \alpha_j, \omega_j, \delta_j)^t, \tag{2}$$

given that

$$g^j(t) = A_j e^{-\alpha_j t}\cos(\omega_j t - \delta_j). \tag{3}$$

To solve this optimization problem, we choose the Bayesian approach. The maximum a posteriori (MAP) and conditional mean (CM) point estimators are computed. By design, the Bayesian approach gauges robustness and reliability of estimators [13, 20]. Details are below.

SVM classification

The 80 subjects above are regarded as a training set. Subjects are divided into two classes: healthy (+ 1) and ill (− 1). For patient j, the parameters A_j and α_j are considered for binary classification in the A-α plane. Classification is carried out by finding a *soft margin support vector machine*. Slack variables are introduced to find a separating line with a high success rate. This methodology is classic [12].

To evaluate the performance of the linear separator, an independent test set is considered:

– 24 males: 15 NGT, 3 IGT, 2 IFG-IGT, 4 T2DM,
– 33 females: 17 NGT, 1 IFG, 11 IGT, 2 IFG-IGT, 2 T2DM.

We remark that this study is a first step to test the proposed methodology, modeling-estimation-classification. The sample size is adequate for the application and conclusions of the support vector machine method.

Chronologically, the training set was selected because it contained all diabetic conditions and appropriate size for classification. To avoid gender as a factor, an all female population was chosen for the training set. After the separating line was constructed, the population above, of comparable size, was chosen for testing. An added difficulty was to choose a mixed population.

A fortiori, gender plays no role. Raw OGTT data led to our results.

Technical information

The Ackerman model for the glucose-insulin regulatory system

The simple model derived in Ackerman et al. [1] is

$$\dot{g} = -m_1 g - m_2 h + J$$
$$\dot{h} = -m_3 h + m_4 g$$

where m_1, m_2, m_3, and m_4 are nonnegative constants.

Therein, the meaning of the parameters is as follows:

– m_1: rate of glucose removal independent of insulin,
– m_2: rate of glucose removal dependent of insulin,
– m_3: rate of insulin removal independent of glucose,

– m_4: rate of release of insulin due to glucose.

After some time, the glucose load $J(t)$ is absorbed in the system and $J(t) \equiv 0$. It is straightforward to eliminate h of the system to obtain the following second-order differential equation,

$$\ddot{g} + 2\alpha\dot{g} + \omega_0^2 g = 0 \tag{4}$$

Here

$$\alpha = \frac{1}{2}(m_1 + m_3),$$

and

$$\omega_0 = \sqrt{m_1 m_3 + m_2 m_4},$$

is the natural or resonant frequency of the system.

A viable interpretation of the glucose-insulin system is that of a damped harmonic oscillator. Hence, it is assumed that

$$\alpha^2 - \omega_0^2 < 0.$$

It is readily seen that the general solution of (4) is

$$g(t) = Ae^{-\alpha t}\cos(\omega t - \delta), \tag{5}$$

where

$$\omega = \sqrt{\omega_0^2 - \alpha^2}.$$

Bayesian estimation

In Bayesian estimation, all variables are random; thus, we consider the model

$$\mathbf{y} = \mathcal{G}(\mathbf{u}) + \eta$$

where $\mathbf{u}$ is the parameter to estimate, $\mathbf{y}$ the data, η the noise, and $\mathcal{G}$ the observation operator.

An important feature of Bayesian estimation is to propose a *prior* probability density function, π_0, for the parameter $\mathbf{u}$. This prior encompasses all we know about $\mathbf{u}$. In essence, it is a modeling problem.

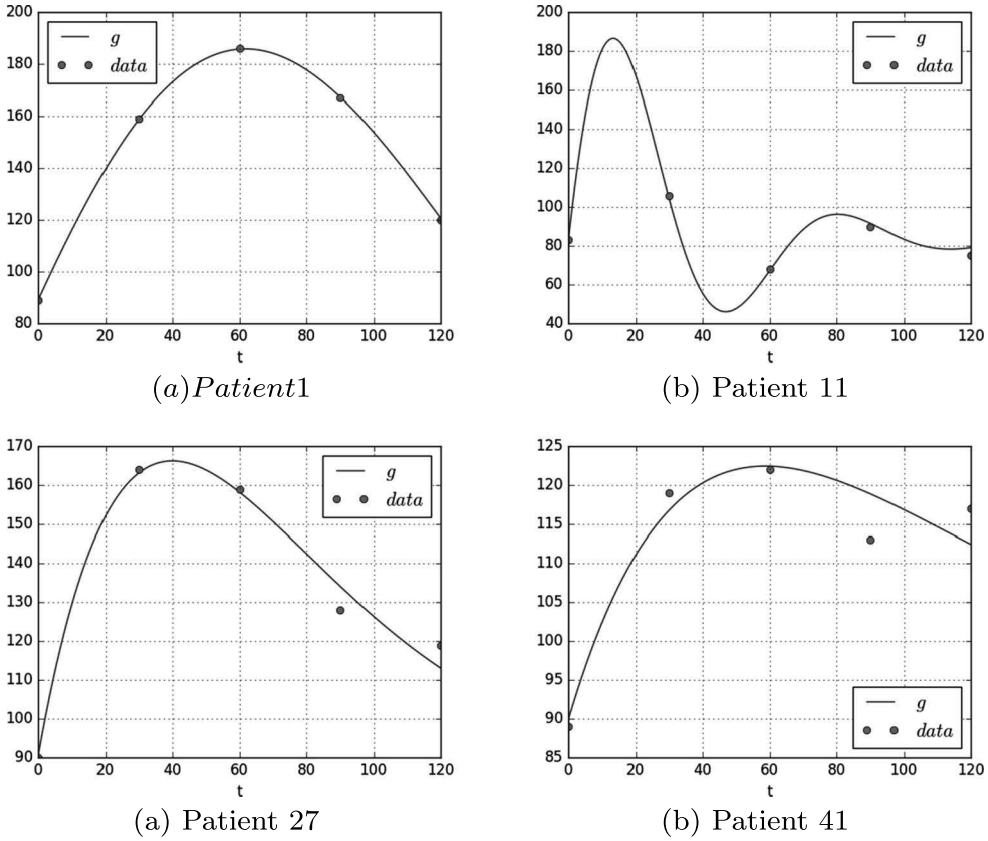
Noise is supposed to be known with density ρ and independent of $\mathbf{u}$. Consequently, the conditional density $\pi^{\mathbf{y}}(\mathbf{u}) \equiv \pi(\mathbf{u}|\mathbf{y})$, the *posterior*, is given from Bayes' formula

$$\pi^{\mathbf{y}}(\mathbf{u}) \propto \rho(\mathbf{y} - \mathcal{G}(\mathbf{u}))\pi_0(\mathbf{u}).$$

The point of Bayesian estimation is to determine the posterior. From the latter, point estimators can be obtained, namely, the conditional mean (CM) and the MAP estimator.

The CM estimate is given by the integral

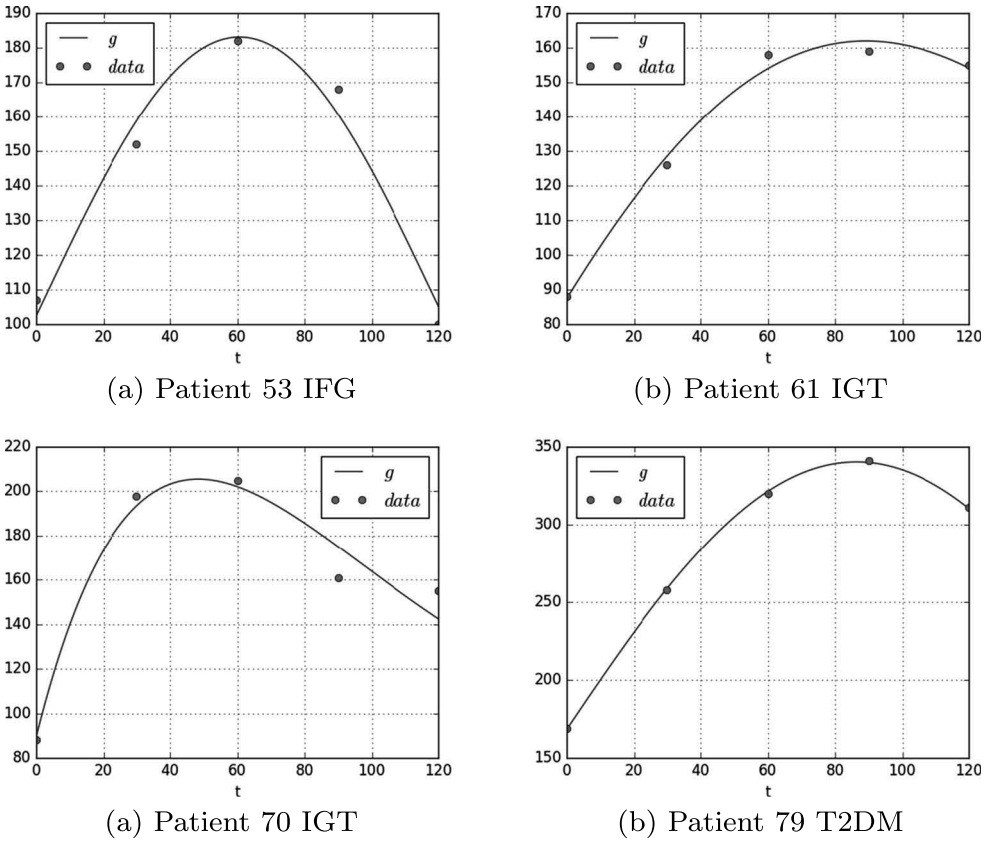
$$\mathbf{u}_{CM} = \int \mathbf{u}\pi^{\mathbf{y}}(\mathbf{u})d\mathbf{u}.$$



(a) $Patient1$ (b) Patient 11

(a) Patient 27 (b) Patient 41

Fig. 1 OGTT curves for some NGT patients. **a** Patient 1. **b** Patient 11. **c** Patient 27. **d** Patient 41



(a) Patient 53 IFG (b) Patient 61 IGT

(a) Patient 70 IGT (b) Patient 79 T2DM

Fig. 2 OGTT curves for some ill patients. **a** Patient 53 IFG. **b** Patient 61 IGT. **c** Patient 70 IGT. **d** Patient 79 T2DM

whereas for the MAP, an optimization problem is to be solved,

$$\mathbf{u}_{MAP} = \arg\max\ \rho(\mathbf{y} - \mathcal{G}(\mathbf{u}))\pi_0(\mathbf{u}).$$

In our problem, Gaussian noise is assumed with zero mean and standard deviation γ. From Bayes' formula, we have,

$$\pi^y(\mathbf{u}) \propto \exp\left(-\frac{1}{\gamma^2}\,|y - \mathcal{G}(\mathbf{u})|^2\right)\pi_0(\mathbf{u})$$

The prior may influence artificially the determination of the posterior, this is unwanted. Uninformative (uniform density) priors are preferred. Hence, we use uniform densities:

- $A \sim U[0.5\, g_m, 2.5\, g_M + 150]$.
- $\alpha \sim U[0, 0.1]$.
- $\omega_0 \sim U[0, 0.15]$.
- $\delta \sim U[-2\pi, 2\pi]$.

where g_m and g_M are respectively the minimum and maximum of absolute values of G_0 shifted glucose concentration data. Only an estimate of the magnitude is required.

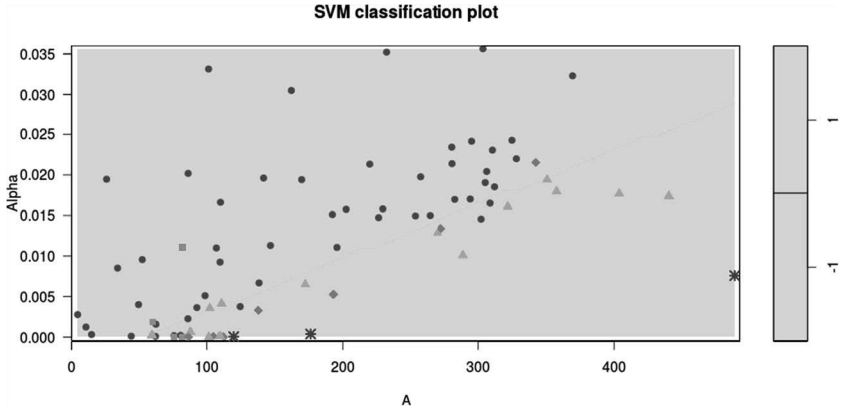


Fig. 3 NGT (•), IFG (□), IGT (△), IFG-IGT (◇), T2DM (∗)

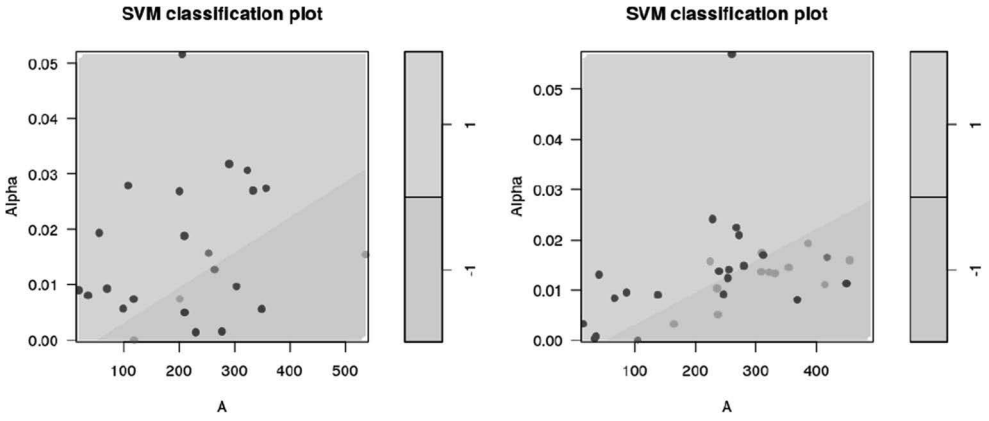


Fig. 4 Test set: males (left), females (right)

Also, we allow large observation errors with a Gaussian model with standard deviation $\gamma = 5$, a typical value in the literature.

In practice, a sample of the posterior is constructed. We use *emcee*, an affine invariant Markov chain Monte Carlo (MCMC) ensemble sampler [8]. Consequently, for each patient's parameters, a posterior distribution is determined and its MAP estimator is obtained.

An advantage of the Bayesian approach is that uncertainty of point estimates is readily quantified by means of the marginal densities of the posterior.

For instance, given the posterior density $\pi^y(\mathbf{u})$, the marginal for α is found by integrating on the other variables.

$$\pi^y(\alpha) = \int\int\int \pi^y(A, \alpha, \omega, \delta) dA d\omega d\delta.$$

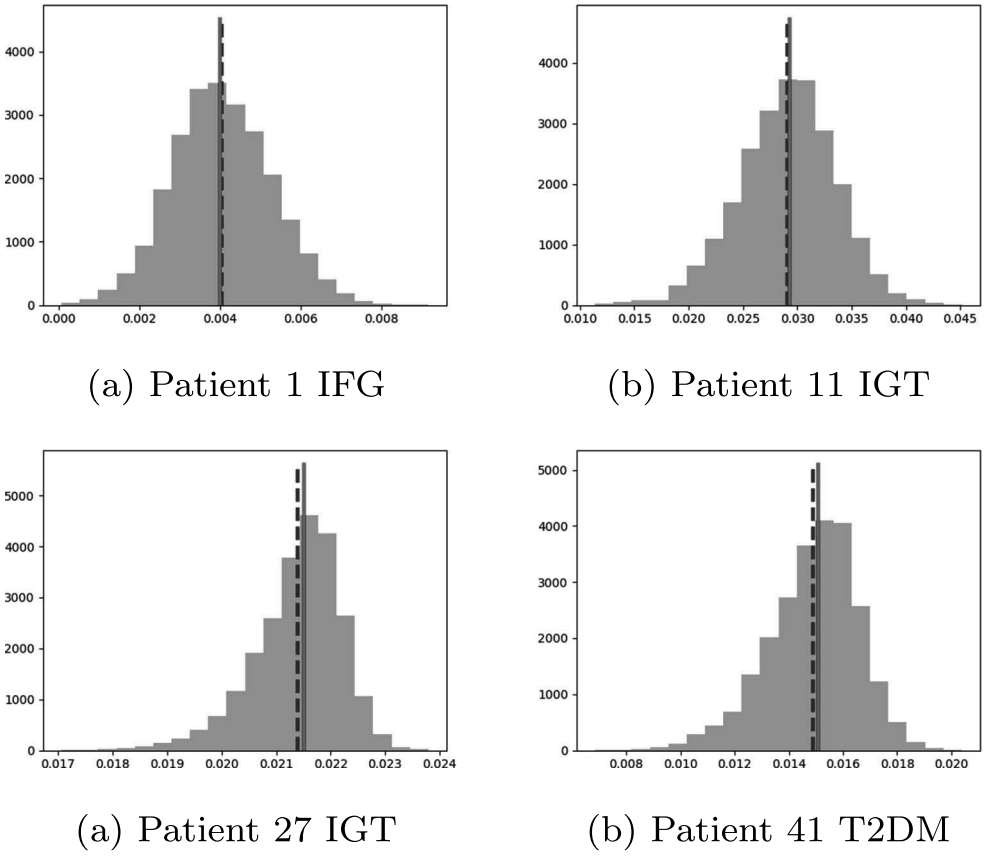


Fig. 5 Marginal posterior densities for NGT patients. MAP solid line. CM dashed line. **a** Patient 1 IFG. **b** Patient 11 IGT. **c** Patient 27 IGT. **d** Patient 41 T2DM

Results

Curve fitting For each patient, the parameters (2) are estimated to construct the functions (3).

We plot OGTT curves for some patients in different diabetic conditions, healthy patients in Fig. 1, whereas ill patients in Fig. 2.

Modeling and parameter estimation is a well-known methodology. The first part of our findings follows this line of thought. The OGTT curves in figures show that our parameter estimation methodology fits the data accurately.

SVM classification In Fig. 3, the binary classification in the A-α plane, obtained by the soft margin support vector machine, is shown. The classification is successful for 85% of patients.

It is remarkable that a linear separator suffices.

Training-test split The classification is tested on an independent population of OGTT data for 24 males and 33 females (Fig. 4). Classification is successful for 91% of males and 87% of females.

Ill patients There are two potential classification errors: to classify a healthy subject as ill, and an ill subject as healthy. The latter is the worst-case scenario. As shown in Figs. 3 and 4, patients with IGT, IFG-IGT, and T2DM conditions are below the separating line. Consequently, ill patients were correctly classified.

Discussion

Based on experiments, it was observed [2] that the parameter α is very sensitive to errors on glucose concentration. Its use was not recommended for a diagnosis criterion. Our Bayesian approach suggests otherwise. For a worst-case scenario, we have considered large observation errors with a Gaussian model. Nevertheless, the obtained marginal densities of the posterior for the parameter α are unimodal and the point estimators are unambiguous. These features simplify the estimation problem. In Figs. 5 and 6, we show a sample of healthy and ill subjects.

In light of the results of SVM classification (Fig. 3), a clinical implication of the current study is that the

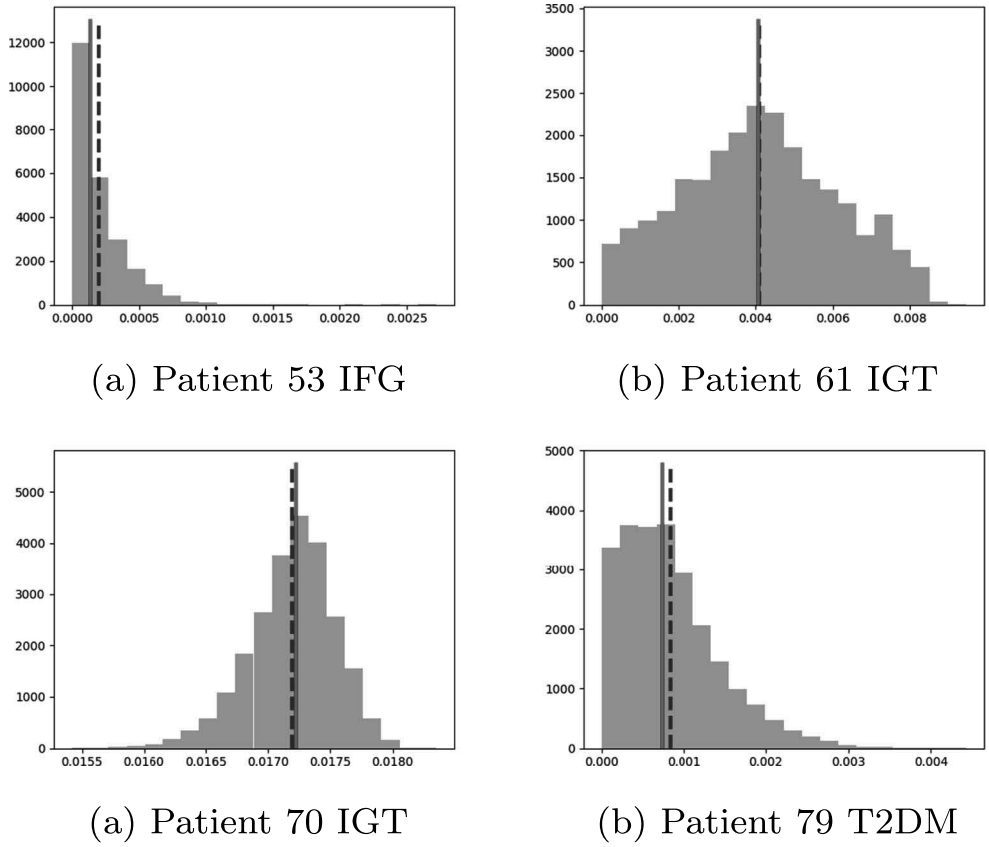


Fig. 6 Marginal posterior densities for ill patients. MAP solid line. CM dashed line. **a** Patient 53 IFG. **b** Patient 61 IGT. **c** Patient 70 IGT. **d** Patient 79 T2DM

peak glucose concentration A, and the average of glucose removal rates α, can be regarded as patient's indices with the potential to be used for classification of diabetic conditions.

At first sight, one may question the quality of classification in the neighborhood of the separating line. Nevertheless, there is an apparent *clockwise* transition from healthy to type 2 diabetes mellitus. The latter might have another clinical implication, namely, early detection of pre-diabetic patients.

On the downside, one may argue that this population set is somewhat limited and that more data and planned sampling are required to be conclusive. However, based on the research process, we are optimistic on the potential for clinical application.

More precisely, the OGTT data in this study was gathered independently according to protocol, by the medical researchers in our group. The computational modeling specialists came later. There was no special criterion for the train-test split. In particular, we just used a size criterion for choosing the training set.

We stress that, our initial plan was to tune our methodology on the simple minimal model (4). It is apparent that in spite of its simplicity, the results are remarkable. It is also surprising that in the application of the SVM method, a linear kernel sufficed.

Finally, let us discuss some limitations of the present study. As pointed out before, to be conclusive, a larger sample and a thorough statistical study is required. Also, the first part of the process is the parameter estimation problem for a given subject. An optimization specialist will have a preferred method to implement an in-house solution. But for wider use, this part of the process requires automatization. The same applies to SVM classification.

The glucose-insulin model used in this study is mostly limited to raw OGTT data. To introduce anthropometric and clinical information, which is essential on a risk assessment application, would require to consider alternative models.

Conclusions

In this work, we have delved on the classification properties of parameter in glucose-insulin models. SVM techniques are proposed for classification. As a proof of concept, the methodology is tested on the simple Ackerman model (4). The obtained results are promising, which have led us to introduce peak glucose concentration A, and the average of glucose removal rates α as indices associated with patient's diabetic condition.

It is of current and future research, to consider more sophisticated glucose-insulin models [16, 18]. It is expected that clustering may vary, and kernel selection in SVM is an issue to be resolved [3].

Also, a larger sample is under study to test further the proposed methodology and classification indices.

Funding M. A. Moreles received financial support from CONACYT through the project CIB 180723.

Availability of data and material Raw OGTT data is available upon request.

Code availability Available upon request

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethics approval The study protocol was approved by the Ethical Committee of the General Hospital of Mexico, and informed written consent was obtained.

Consent to participate Informed written consent was obtained from all volunteers.

Consent for publication Not applicable

References

1. Ackerman E, Rosevear JW, McGuckin W. A mathematical model of the glucose-tolerance test. Phys Med Biol. 1964;2(9):203–213.
2. Ackerman E, Gatewook L, Rosevear J, Molnar GI. Blood glucose regulation and diabetes. Concepts and models of biomathematics. In: Heinmets F, editors; 1969. p. 131–156.
3. Hernandez-Aguirre A, Mendez-Davila HD, Moreles-Vazquez MA. What kernel size separates my data? In: Proceedings of the Fifth Mexican International Conference in Computer Science, 2004. ENC 2004. IEEE; 2004.
4. American Diabetes Association. Classification and diagnosis of diabetes: standards of medical care–2018. Diabetes Care, Suppl. 2018;1(41):S13–S17. https://doi.org/10.2337/dc18-S002.
5. Alyass A, Almgren P, Akerlund M, Dushoff J, Isomaa B, Nilsson P, Tuomi T, Lyssenko V, Groop L, Meyre D. Modelling of OGTT curve identifies 1 h plasma glucose level as a strong predictor of incident type 2 diabetes: results from two prospective cohorts. Diabetologia. 2015;1(58):87–97.
6. Caumo A, Bergman RN, Cobelli C. Insulin sensitivity from meal tolerance tests in normal subjects: a minimal model index. J Clin Endocrinol Metab. 2000;85:4396.
7. Erlandsen M, Martinussen C, Gravholt CH. Integrated model of insulin and glucose kinetics describing both hepatic glucose and pancreatic insulin regulation. Computer Methods and Programs in Biomedicine. 2018;156:121–131.
8. Foreman-Mackey D, Hogg D, Lang D, Goodman J. Emcee: the MCMC hammer. Publ Astron Soc Pac. 2013;125(925):306–12. https://doi.org/10.1086/670067.
9. Heydari M, Teimouri M, Heshmati Z, et al. Comparison of various classification algorithms in the diagnosis of type 2 diabetes in Iran. Int J Diabetes Dev Ctries. 2016;36:167–173. https://doi.org/10.1007/s13410-015-0374-4.
10. Hulman A, Vistisen D, Glümer C, Bergman M, Witte D, Færch K. Glucose patterns during an oral glucose tolerance test and associations with future diabetes, cardiovascular disease and all-cause mortality rate. Diabetologia. 2018;61(1):101–107.

11. Ismail HM, Xu P, Libman IM, Becker DJ, Marks JB, Skyler JS, Palmer JP, Sosenko J. Type 1 diabetes TrialNet study group the shape of the glucose concentration curve during an oral glucose tolerance test predicts risk for type 1 diabetes. Diabetologia. 2018;1(61):84–92.
12. James G, Witten D, Hastie T, Vol. 112. Tibshirani r. An introduction to statistical learning. New York: Springer; 2013.
13. Kaipio J, Somersalo E. Statistical and computational inverse problems. Berlin: Springer; 2004.
14. Khan ZAW, Vidyasagar S, Varma DM, Nandakrishna B, Holla A, Binu VS. The clinical and biochemical profiles of patients with IFG. International Journal of Diabetes in Developing Countries. 2019;39(1):94–99.
15. Morbiducci U, et al. Predicting the metabolic condition after gestational diabetes mellitus from oral glucose tolerance test curves shape. Annals of Biomedical Engineering. 2014;42.5:1112–1120.
16. Palumbo P, Ditlevsen S, Bertuzzi A, De Gaetano A. Mathematical modeling of the glucose-insulin system: a review. Mathematical Biosciences. 2013;244(2):69–81.
17. Pillonetto G, Sparacino G, Cobelli C. Numerical non-identifiability regions of the minimal model of glucose kinetics: superiority of Bayesian estimation. Math Biosci. 2003;184:53–67.
18. Rathee SODE. Models for the management of diabetes: a review. Int J Diabetes Dev Ctries. 2017;37:4. https://doi.org/10.1007/s13410-016-0475-8.
19. Rauf M, Sevil E, Ebru C, Cemil C. Early diagnosis of gestational diabetes mellitus during the first trimester of pregnancy based on the one-step approach of the International Association of Diabetes and Pregnancy Study Groups. International Journal of Diabetes in Developing Countries. 2018;38(1):20–25.
20. Stuart AM. Inverse problems: a Bayesian perspective. Acta Numerica. 2010;19:451–559.

Affiliations

Paola Vargas[1] · Miguel Angel Moreles[1] · Joaquin Peña[1] · Adriana Monroy[2] · Silvestre Alavez[3]

[2] Mexico General Hospital, Mexico City, Mexico

[3] Health Sciences Department, Metropolitan Autonomous University, Lerma, Mexico

International Journal of Diabetes in Developing Countries (2021) 41:63–70
https://doi.org/10.1007/s13410-020-00841-4

ORIGINAL ARTICLE

Human ENPP1 gene polymorphism in DKD patients: a hospital-based case control study

S. Chandra[1] · A. K. Singh[2] · M. Singh[2] · P. Pandey[2] · C. S. Azad[2] · S. Singh[1] · P. Das[3] · R. G. Singh[1]

Received: 31 May 2019 / Accepted: 13 June 2020 / Published online: 20 August 2020

Abstract

Aim The aim of present study was to investigate the association of the ENPP1 (K121Q) gene polymorphism with DKD in eastern Uttar Pradesh population.

Methods A total of 162 DKD and 155 apparently healthy controls were enrolled and K121Q polymorphism was determined by PCR-RFLP followed by sequencing.

Results It was observed that genotype KQ was more frequent in patients than controls and allelic frequencies of Q allele were higher in DKD (Q 21.3%) than control (Q 15.81%). It depicted that Q allele may be associated with DKD patient and has more risk in eastern UP population.

Conclusion The study indicated a significant association of KQ polymorphism of the ENPP1 gene with DKD in the eastern Uttar Pradesh population.

Keywords Nephropathy · Chronic kidney disease · Diabetic kidney disease · ENPP1 · Polymorphism · DKD

Introduction

Diabetic kidney disease (DKD) is the common cause of end-stage renal disease. It is associated with high morbidity and mortality rates among diabetic patients [1–2]. DKD is a common diabetic chronic complication, characterized by reduced GFR (glomerular filtration rate) and albuminuria. It is also called DKD or diabetic nephropathy (DN). It has a multifactorial origin caused by a delicate interaction between susceptible genes and environmental factors. Due to the increasing prevalence of obesity, hypertension, and physical inactivity, millions of peoples (about 6–7%) of the world's population are affected by DKD. Twenty-four million people in the USA have diabetes and approximately 180,000 people are living with renal failure as a result of diabetes [3–4]. The occurrence of DKD in India was less (8.9% in Vellore, 5.5% in Chennai) when compared with the occurrence in Asian Indians (22.3%) in the UK [5]. Risk factors such as dyslipidemia, arterial hypertension, and hyperglycemia are recognized to play a key role in the development of DKD. In addition to this risk factor, there is the involvement of some molecular markers, which may serve as diagnostic/ prognostic tools. Ectoenzyme

✉ R. G. Singh
rgsinghbhu@gmail.com; rgsingh@bhu.ac.in

S. Chandra
ayunephro@gmail.com

A. K. Singh
alokbiochem@gmail.com

M. Singh
mritunjai.singh@gmail.com

P. Pandey
preetypreet23@gmail.com

C. S. Azad
csazad.ims.bhu14@gmail.com

S. Singh
shivendrasbhu@hotmail.com

P. Das
parimal@bhu.ac.in

[1] Department of Nephrology, Institute of Medical Sciences, Banaras Hindu University, Varanasi 221005, India

[2] Department of Medicine, IMS, Banaras Hindu University, Varanasi, India

[3] Centre for Genetic Disorders, Institute of Science, Banaras Hindu University, Varanasi, India

nucleotide pyrophosphate phosphodiesterase 1 (ENPP1) is a good candidate gene for DKD. The cytogenetic location of ENPP1 is $6q^{23.2}$ which transcribes 7442 bp mRNA (Mm-006208.2). This gene spanning 87 kilobases contains 25 exons and 24 introns, which codes for 925 amino acids long protein. ENPP1 gene blocks the tyrosine kinase activity of the insulin receptor in several cells, causing insulin resistance by missense polymorphism at 121 positions in the protein. The corresponding single nucleotide substitution leads to a change of class of amino acid from charge to uncharged [6]. This alters the behavior by influencing the protein-protein interaction. Change in nucleotide at rs537 (A > C) in exon 4 of the ENPP1 gene, alters the amino acid lysine to glutamine (K121Q). In different studies, it has been found that the Q allele is associated with the inhibition of insulin signaling more effectively than K allele, the identification of such genes will help to detect an individual's risk of developing DKD and may provide a better understanding of its pathophysiology [7]. However, the results of the association of ENPP1 with DKD have been controversial [8]. Several studies on the association between the ENPP1 (K121Q) polymorphism and DKD in various populations observed both positive [9, 10] and negative association [11]. Most genetic studies of DKD used a candidate gene approach. Several studies from different parts of the world have examined ENPP1 gene polymorphism as a candidate for DKD [12].

The genetic study of DKD in the eastern UP population has public health significance because the disease has reached outbreak proportions in this particular region. The prevalence of diabetes with kidney disease rises alarmingly in India and projected to reach up to 124 million by 2040 [13]. So, this study was planned to investigate the association of the ENPP1/PC-1 (K121Q) in polymorphism in DKD patients in the eastern UP population and their association with other risk factors.

Materials and methods

Study of population and clinical evaluation of patients

All patients included in this study were taken from Nephrology OPD, Sir Sundar Lal Hospital, Banaras Hindu University, Varanasi, UP, India, during the period from 2011 to 2014. Signed informed consent was taken from every subject.

Exclusion criteria included patients < 18 years of traumatic cases and patients who were mentally incapable of giving their own consent. If the patient met the appropriate criteria, we visited the patient before treatment to explain the study and asked for the patient's participation along with informed consent form. We conducted a 30-min interview with the patient. The interview was based on a predesigned questionnaire that included questions about height and weight, age, health care, and utilization, health habits, and physical activity level. Data involving presentation, diagnosis, and staging were collected from office charts, hospital charts, and a face-to-face interview with the study subjects. A total of 162 DKD patients and 155 normal controls were prospectively enrolled for the study.

Estimation of body mass index

According to WHO-1998, the BMI cutoff in Europids for overweight (≥ 25.0 kg/m^2) and obesity (≥ 30.0 kg/m^2) is higher than the Asian-pacific region. Steering committee (WHO Western Pacific Region 2000, the international association for the study of obesity and international obesity task force) recommended in Asians the cutoff for overweight ($\geq$ 23.0 kg/m^2) and obesity (≥ 25.0 kg/m^2) and the exact classification criteria as discussed in our previous study [14]. BMI was measured at the time of hospital admission for treatment and the above criteria were used to categorize patients.

Glomerular filtration rate estimations

Serum creatinine measurements were performed the day before the measurement of isotopic GFR (i-GFR) through Cockroft and Gault formula (CG) [15]. The CG equation predicts a better outcome than either the CKD-EPI or the MDRD equation [16–17]

$$\mathrm{CG} = \frac{[140 - \mathrm{age(years)}] * \mathrm{Body\ weight\ (Kilograms)} * [0.85\ (\mathrm{if\ female})]}{72 * \mathrm{Serum\ creatinine\ (Milligrams\ per\ deciliter)}}$$

The equation was designed to estimate creatinine clearance expressed in milliliters per minute.

Kidney disease staging

The stages of DKD are mainly based on measured or estimated GFR. There are five stages, but kidney function is normal in stage 1 and the end-stage in stage 5. In our study, we classify our patients in three groups: mild CKD (60–89), moderate CKD (30–59), and severe CKD (15–29) on the basis of GFR.

Blood samples

In all patients, routine biochemical tests were carried out for blood glucose, renal function test, liver function test, and complete blood count. Blood (~ 3 ml) was drawn through sterile syringe from the peripheral vein, collected into EDTA coated vials and refrigerated at − 20 °C till used for DNA extraction.

Isolation of genomic DNA

Genomic DNA was extracted from collected blood samples. The entire experimental steps were carried out at 4 °C. The

blood was homogenized in ice-cold sucrose, EDTA, and Tris-HCl buffer (sucrose 10.8 g, 0.5 mol/l of EDTA 1 ml, 1 mol/l of Tris-Cl 2.5 ml, pH 8 to 100 ml water, autoclaved, and stored at 4 °C) and centrifuged at 4000 rpm for 10 min at 4 °C to pellet, was washed once again with sucrose, EDTA and Tris-HCl buffer. The pure nuclear pellet was suspended in Tris HCl, EDTA, and NaCl buffer (1 mol/l Tris-Cl 2.0 ml, 0.5 mol/l EDTA 1 ml, 5 mol/l NaCl 8 to 100 ml water, autoclaved and stored at 4 °C), SDS was added to a final concentration of 1%, which was mixed gently to lyse the nuclei. Proteinase-K was added to the lysate (to a final concentration of 100 μg/ml and was incubated at 37 °C overnight). The proteinase-K treated lysate was mixed with an equal volume of tris saturated phenol, pH 8, twice with phenol: chloroform (1:1) mixture and once with chloroform: isoamyl alcohol (24:1) mixture following centrifugation at 10000 rpm after each extraction. The final aqueous phase was transferred to a fresh tube. To this aqueous phase, 1/10th volume of 3 mol/l of sodium acetate (pH 5.2) and two volumes of ice-cold absolute alcohol were added. The DNA was then precipitated, washed with 70% ethanol, air dried, and dissolved in an appropriate volume of TE buffer (pH 8). The isolated DNA was kept at 4 °C for further experiments.

ENPP1 PCR-RFLP (restriction fragment length polymorphism) genotyping

Polymerase chain reaction (PCR) amplification using a primer pair as follows:

Forward primer 5'-CTGTGTTCACTTTGGACATG TTG-3′;

Reverse primer 5'-GACGTTGGAAGATACCAGGT TG-3′.

The primers amplified a 238 bp product. PCR was carried out in Veriti 96 well Thermal cycler (Applied Biosystems) at respective annealing temperature using the standard protocol. Primers used have been chosen from the previous work done for the ENPP1 gene [9]. PCR reaction was carried out in a volume of 25 μl with, 1× PCR master mix (fermentas), 10 μM of both forward and reverse primer with the following conditions: 35 cycles of 95 °C for 30 s, respective annealing at 55.5 °C for 40 s, and 72 °C for 40 s, with initial denaturation at 94 °C for 3 min and final extension at 72 °C for 5 min. PCR products were analyzed on 2% agarose gels (BIORON, Ludwigshafen, Germany) in 1X TBE (89 mmol/l of Tris borate and 2 mmol/l of EDTA, pH 8.3). PCR products were digested by AvaII restriction enzyme (20 units) for 2 h at 37 °C and separated on 3% agarose gel by electrophoresis. The digested product produced three distinct bands having different molecular sizes (Fig. 1). KK and KQ amplicon were also sequenced using the automated DNA sequence, genetic analyzer 3130. To minimize the sequencing artifacts induced by PCR, products from at least two different PCRs were sequenced using forward and reverse primers. Sequences were compared against the human (rs1044498 A > C) sequence. Sequences variants in the corresponding sequence that were found in both the DKD and control at a particular location were classified as polymorphisms.

Statistical analysis

All statistical analysis was performed using SPSS 16.0 for windows software. The result was presented as a mean ± standard deviation for the continuous variables. Analysis of variance (ANOVA) was used to assess the association between genotypes and baseline characteristics. Chi-square test was carried out ascertaining the homogeneity of proportions and to determine the significance of polymorphism and risk factors. Hardy-Weinberg equilibrium was considered to determine the allelic frequency. Bivariate analysis was performed to determine associations of the presence of genotypes with DKD. Statistical significance was considered when $p = 0.05$.

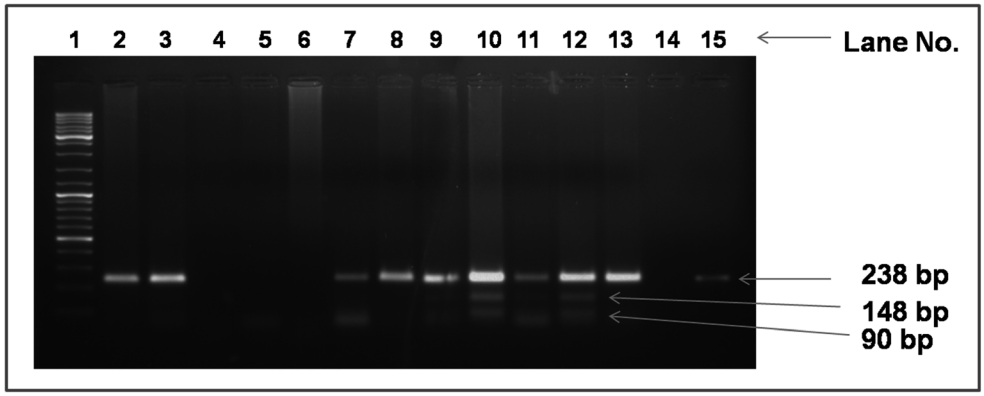


Fig. 1 Human ENPP1 gene polymorphism (restriction digestion method): lane no. 1; 100 bp DNA Ladder, lane no. 2, 3, 8, 9, 11, 13, 15; undigested PCR product (KK), lane no. 4, 5,6,14; no PCR bands suggest no results, lane no. 10 and 12; 2 bands of 148 bp and 90 bp long digested PCR products (KQ)

Table 1 Baseline Characteristics of study groups

Characteristics	Cases ($N = 162$)	Control ($N = 155$)	*P Value*
BMI Mean ± SD	25.3 ± 3.4	23.98 ± 4.3	0.003
SBP Mean ± SD	133.2 ± 18.8	127.4 ± 18.4	0.006
DBP Mean ± SD	82.4 ± 12.1	80.9 ± 11.2	0.238
Hb Mean ± SD	10.2 ± 4.7	11.7 ± 2.14	0.000
FBS Mean ± SD	131.1 ± 67.5	126.2 ± 67.5	0.518
PP Mean ± SD	181.7 ± 76.6	135.8 ± 68.9	0.000
Urea Mean ± SD	69.7 ± 34.34	31.9 ± 22.16	0.000
Creatinine Mean ± SD	2.15 ± .96	1.58 ± 1.31	0.000
Albumin Mean ± SD	3.84 ± .65	4.33 ± .61	0.000
Cholesterol Mean ± SD	147.7 ± 42.29	166.8 ± 44.42	0.000
HDL Mean ± SD	38.2 ± 12.03	39.3 ± 10.97	0.409
LDL Mean ± SD	71.73 ± 27.37	103.3 ± 35.52	0.000
VLDL Mean ± SD	28.9 ± 12.1	32.1 ± 16.4	0.050
TG Mean ± SD	137.3 ± 53.3	132.0 ± 58.56	0.399
Na Mean ± SD	136.5 ± 6.8	137.2 ± 13.06	0.537
K Mean ± SD	4.9 ± 2.79	4.6 ± .86	0.121

Results

The baseline characteristics of patients

The mean age of DKD patient and control was 57.70 ± 9.8 years and 55.21 ± 10.5 years respectively. The preponderance of study showed a significant positive association between DKD risk with body mass index (BMI), hemoglobin (Hb), post prandial sugar (PP), urea, creatinine (CREAT), albumin (ALB), cholesterol (CHOL), low-density lipoprotein (LDL), systolic blood pressure (SBP), and very low–density lipoprotein (VLDL) while factors like diastolic blood pressure (DBP), fasting blood Sugar (FBS), high-density lipoprotein (HDL), triglyceride (TG), sodium (NA), and potassium (K) did not show any significant association between study groups (Table 1).

Genotype distribution of ENPP1 gene

The frequencies of KK, KQ and QQ genotype (Fig. 2) among the DKD group were 60.5% ($n = 98$), 36.4% ($n = 59$), and 3.1% ($n = 5$) respectively, whereas, in healthy control group

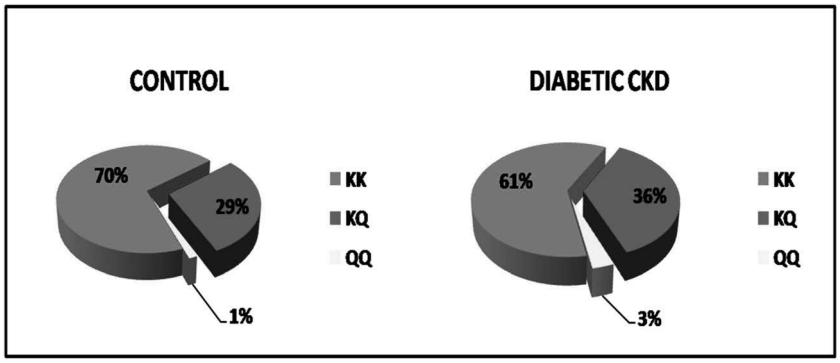


Fig. 2 Distribution of genotype with study subject ($N = 317$).

Table 2 Genotype distribution of ENPP1 gene in study groups

Variable		Cases (162)	Control (155)	*P* Value
ENPP1 genotypes	KK	98 (60.5%)	108 (69.7%)	0.174
	KQ	59 (36.4%)	45 (29.0%)	
	QQ	5 (3.1%)	2 (1.3%)	

the same were found to be 69.7% ($n = 108$), 29.0% ($n = 45$), and 1.3% ($n = 2$) respectively (Table 2).

Distribution of ENPP1 genotype showed no significant difference among the study.

Allelic frequencies of K and Q allele in the ENPP1 gene were determined by using Hardy-Weinberg equilibrium. Prevalence of K allele with control and case was found 84.19% and 78.7% respectively. The allelic frequencies show significance between the two groups (Table 3).

Association of ENPP1 genotype with risk factors

TG, BMI, LDL, HDL, SBP, DBP, Diabetes, and urea creatinine were compared with ENPP1 genotypes KK, KQ, and QQ in study groups (Table 4). They were not found to be associated with genotype distribution in the study group.

Bivariate analysis of the ENPP1 genotype and DKD patients

Bivariate analysis (Table 5) suggests that there were 26% more risk with the KQ genotype in comparison to QQ for moderate DKD. KQ genotype was found to have 16% lower risk in severe DKD, and KK genotype was found with equal possibility as reference category (QQ), but data is not statistically significant.

Discussion

DKD/DN is a disease with complex pathogenesis that depends on genetic predisposition as well as factors associated with lifestyle and environment. The gap between onset and clinical diagnosis of diabetes leads to the development of numerous chronic complications, which are the leading causes of premature mortality among diabetic patients. DKD is one of the most microvascular complications of non-insulin dependent diabetes mellitus (NIDDM) and has been reported to occur infrequently in 10 years after the occurrence of diabetes. However, if the risk factors are uncontrolled, DKD may occur too early. It is a widely accepted fact that one can do quite well to delay the onset of DKD, but once occurred, it cannot be reversed rather with early diagnosis.

The molecular mechanisms accounting for the involvement between Q variant of ENPP1gene and progress of advanced stages of DKD have been hypothesized earlier and indicated that it is expressed in numerous tissues including the muscle, liver, fat, and kidney [18]. This study is a comprehensive investigative report with reference to the association of ENPP1, K121Q gene polymorphism with DKD in adult Asian Indians living in the eastern part of Uttar Pradesh. The study was carried out between two groups (control and DKD) which were matched for age and sex. However, they were significantly different with respect to BMI, TG, LDL, FBS, PP, SBP, and DBP. The results of different studies between ENPP1/ K121Q variants and DKD in numerous races are conflicting [9–11]. In the present study, the genotype distribution of K121Q was not associated with DKD. Further, we investigated the role of ENPP1/ K121Q polymorphism with DKD related various phenotypes like SBP, DBP, fasting glucose, and lipid profile. There were significant associations found between DKD and BMI, HB, PP, UREA, CREAT, ALB, CHOL, and LDL, but no significant associations were detected with SBP, DBP, FBS, HDL, TG, NA, and K in the present study.

In different studies, it has been found that the Q allele is associated with the inhibition of insulin signaling more effectively than K allele [10]. This study included 162 cases (DKD) and 155 controls (healthy normal) in which Q allele is more frequently found in cases (21.3%) than controls (15.18%) (Table 3) which supports that Q allele is commonly associated in DKD patient. Homozygous genotype (QQ) was more frequent in patients than controls and more affected than heterozygous genotype when compared with other parameters.

Table 3 Representation of ENPP1 Genotype and allele frequencies with study groups (Hardy-Weinberg)

	Genotype Frequency (Expected)			Allele Frequency		*P Value*
	KK	KQ	QQ	K allele	Q allele	
Cases	100.35 (61.94%)	54.31 (33.52%)	7.35 (4.54%)	78.7%	21.3%	0.271
Control	109.87 (70.89%)	41.25 (26.62%)	3.87 (2.50%)	84.19%	15.81%	0.258

Table 4 Association of ENPP1 Gene Polymorphism with risk factors

Variables	Diabetic CKD Patient				Normal Control			
	KK ($N = 98$)	KQ ($N = 59$)	QQ ($N = 5$)	*P Value*	KK ($N = 108$)	KQ ($N = 45$)	QQ ($N = 2$)	*P* Value
Age	58.36 ± 10.12	56.46 ± 9.53	59.40 ± 9.96	0.473	55.93 ± 10.78	53.87 ± 10.17	47.00 ± 1.41	0.300
BMI	25.08 ± 3.35	25.56 ± 3.82	26.32 ± 2.26	0.569	24.10 ± 4.84	23.53 ± 2.84	27.75 ± 0.35	0.358
SBP	135.69 ± 17.26	129.73 ± 20.01	128.00 ± 31.14	0.130	126.54 ± 18.70	129.18 ± 18.09	138.0 ± 2.82	0.521
DBP	83.20 ± 12.37	81.20 ± 12.04	83.60 ± 11.61	0.599	80.48 ± 11.85	81.33 ± 9.40	96.00 ± .00	0.147
Hb	10.43 ± 5.83	9.81 ± 2.20	10.28 ± .58	0.734	11.62 ± 2.18	12.11 ± 2.04	13.05 ± .77	0.301
FBS	124.63 ± 72.10	138.2 ± 53.94	173.8 ± 106.03	0.168	131.97 ± 73.17	114.54 ± 50.82	77.05 ± 23.54	0.204
PP	179.91 ± 80.50	191.55 ± 69.87	160.4 ± 78.14	0.421	139.38 ± 67.37	129.58 ± 73.69	90.00 ± 28.28	0.464
Urea	70.09 ± 33.99	70.31 ± 34.79	56.28 ± 40.67	0.675	34.43 ± 25.07	26.70 ± 11.82	19.54 ± 4.83	0.105
Creatinine	2.21 ± .99	2.05 ± .92	2.15 ± .74	0.604	1.66 ± 1.31	1.44 ± 1.33	.62 ± .007	0.369
Cholesterol	150.58 ± 37.81	143.85 ± 49.22	138.42 ± 40.72	0.556	167.86 ± 46.51	163.79 ± 40.27	177.65 ± 17.74	0.826
HDL	39.13 ± 13.58	36.88 ± 9.41	37.24 ± 4.84	0.520	39.20 ± 10.43	39.57 ± 12.28	40.40 ± 15.27	0.973
LDL	72.30 ± 30.24	69.23 ± 22.16	90.04 ± 18.39	0.251	106.25 ± 36.93	96.15 ± 32.01	105.55 ± 3.74	0.278
VLDL	29.14 ± 13.11	28.97 ± 10.81	24.42 ± 6.14	0.700	30.42 ± 15.31	31.56 ± 19.46	28.01 ± 5.24	0.901
TG	137.19 ± 56.13	139.13 ± 50.64	120.80 ± 26.49	0.673	129.54 ± 51.92	137.51 ± 73.39	146.80 ± 16.68	0.702
Na	136.48 ± 6.89	136.27 ± 6.79	141.48 ± 5.31	0.260	136.34 ± 14.97	139.26 ± 6.62	143.50 ± 2.12	0.362
K	4.68 ± .91	5.55 ± 4.39	4.02 ± 2.34	0.127	4.70 ± .86	4.46 ± .85	3.55 ± .07	0.063

However, K allele was found to be the vaguely similar frequency in both disease and healthy control group. Earlier studies have reported that the frequency of 121Q allele carriers in Chinese population is 18.8%; in Caucasians, 23.2–36.4%; in South Indians (living in Chennai and Dallas, USA), 27.5–34.2%, and in Dominicans, 78.4%; and in African-Americans, 67.0%. Further, a significant alliance between the 121Q allele and T2D (type 2 diabetes) has been shown in Caucasian populations in the USA and Finland [19–21]. In addition to diabetes, the Q121 (PC-1) allele has also recently been reported to stimulate the risk of obesity [22]. Many case-control studies have accomplished the possible contribution of ENPP1gene in genetic accountability to obesity and T2D, while several studies revealed that ENPP1 is associated with neither diabetes nor obesity because of the multifactorial nature of the disease [23–27]. Our findings show that the allele frequency and genotype pattern of ENPP1 (K121Q) were not significantly different between cases and healthy controls. Kubaszek et al. showed similar findings of K121Q polymorphism, in which no significant difference was found in south Asian inhabitants [21]. The present study was also deficient in the association of K121Q with any risk factors (Table 4). Though various negative association was observed in Chinese population [25]. Lee et al. [27] had demonstrated that T2D patients with the ENPP1 (121Q) variant have a higher rate of aortic calcification which is well-known predictor of cardiovascular outcomes.

The K121Q (PC-1) polymorphism is associated with the presence of vascular calcification, which can raise the risk of coronary heart disease (CHD) in T2D patients. DKD stages (Table 5) were also not found to be significant with K121Q polymorphism in the present study but De Cosmo et al. [28] demonstrated that the 121Q variant of ENPP1 gene is coupled with decreased renal function (reduced GFR) among patients with T2D. Consequently, a possible mechanism accounting for the 121Q variant is the predisposing effect of atherosclerosis and insulin resistance, which are independent risk factors for renal dysfunction in patients with T2D. The study has some limitations; Creatinine cutoff is high; hence, the reported figure is slightly lower than reported by the other Indian studies. The larger sample size is needed in future studies to exclude the interference factors of genetic risk contributed by Q variants. Other gene polymorphism should have examined comprehensively to explore the virtual role of ENPP1 K121Q gene polymorphism in DKD patients.

Conclusion

In the present study, we concluded that the KQ genotype of the ENPP1 gene is strongly associated with disease stages and has a greater risk for disease severity. It is also associated with

Table 5 Association of ENPP1 Genotypes with DKD Stages through regression analysis

Variables			Odds Ratio (OR)	95% Confidence Interval (CI)		*P* Value
				Lower	Higher	
ENPP1 Genotype	Severe DKD (15–30 ml/Min)	KK	1.082	0.090	13.008	*0.950*
		KQ	0.841	0.065	10.866	*0.894*
		QQ	–	–	–	–
	Moderate DKD (30–50 ml/Min)	KK	0.556	0.047	6.898	*0.644*
		KQ	1.264	0.099	15.980	*0.856*
		QQ	–	–	–	–

the decrement of GFR in the study groups. On the other hand, KK genotype was not found to be associated with any risk factors.

Since the present study has limited sample size of ethnic group, further confirmatory studies needed to confirm the significant association of ENPP1 gene with the pathogenesis of DKD and other correlated factors.

Acknowledgments We thank all medical and nursing staffs of Nephrology and Diabetes Care for the design and conduct of the study and team members of Department of Nephrology and Centre for Genetic Disorder, Banaras Hindu University of India for the guidance and comments.

Compliance with ethical standards

The study was ethically approved by institutional ethical committee (Dean/2012-13/485).

Conflict of interest The authors declare that they have no conflict of interest.

References

1. Carpena MP, Rados DV, Sortica DA, Souza BM, Reis AF, Canani LH, et al. Genetics of diabetic nephropathy. Arq Bras Endocrinol Metabol. 2010;54:253–61.
2. Sortica DA, Crispim D, Zaffari GP, Friedman R, Canani LH. The role of ecto-nucleotide pyrophosphatase/phosphodiesterase 1 in diabetic nephropathy. Arq Bras Endocrinol Metabol. 2011;55:677–85.
3. Singh M, Singh AK, Singh S, Pandey P, Chandra S, Gambhir IS. Angiotensin-converting enzyme gene I/D polymorphism increases the susceptibility to hypertension and additive diseases: a study on North Indian patients. Clin Exp Hypertens. 2016;38:305–11.
4. Bethesda MD. National Institute of Diabetes and Digestive and Kidney Diseases. National Diabetes Statistics, 2007, National Institutes of Health, U.S. Department of Health and Human Services. 2008
5. Ramachandran A. Epidemiology of diabetes in India-three decades of research. J Assoc Physicians India. 2005;53:34–8.
6. Maddux BA, Goldfine ID. Membrane glycoprotein PC-1 inhibition of insulin receptor function occurs via direct interaction with the receptor alpha-subunit. Diabetes. 2000;49:13–9.
7. Palmer ND, Freedman BI. Insights into the genetic architecture of diabetic nephropathy. Curr Diab Rep. 2012;12:423–31.
8. Keshavarz P, Inoue H, Sakamoto Y, Kunika K, Tanahashi T, Nakamura N, et al. No evidence for association of the ENPP1 (PC-1) K121Q variant with risk of type 2 diabetes in a Japanese population. J Hum Genet. 2006;51:559–66.
9. Pizzuti A, Frittitta L, Argiolas A, Baratta R, Goldfine ID, Bozzali M, et al. A polymorphism (K121Q) of the human glycoprotein PC-1 gene coding region is strongly associated with insulin resistance. Diabetes. 1999;48:1881–4.
10. Prakash J, Mittal B, Awasthi S, Agarwal CG, Srivastava N. K121Q ENPP1/PC-1 gene polymorphism is associated with insulin resistance in a North Indian population. J Genet. 2013;92(3):571–6.
11. Son M, Ye BJ, Kim JI, Kang S, Jung KY. Association between shift work and obesity according to body fat percentage in Korean wage workers: data from the fourth and the fifth Korea National Health and Nutrition Examination Survey (KNHANES 2008–2011). Ann Occup Environ Med. 2015;23:27–32.
12. Sortica DA, Buffon MP, Souza BM, Nicoletto BB, Santer A, Assmann TS, et al. Association between the ENPP1 K121Q polymorphism and risk of diabetic kidney disease: a systematic review and meta-analysis. PLoS One. 2015;10(3):e0118416.
13. Rajput R, KM Kumar P, Seshadri K, Agarwal P, Talwalkar P, et al. Prevalence of chronic kidney disease in type 2 diabetes mellitus patients: START- India study. J Diabetes Metab. 2017;8:722.
14. Chandra S, Singh AK, Singh M, Das P, Singh S, Pandey HP, et al. Prolonged elevated postprandial sugar augments severity in kidney disease: a hospital based study of Indian patients. Ren Fail. 2014;36:50–4.
15. Cockcroft DW, Gault HM. Prediction of creatinine clearance from serum creatinine. Nephron. 1976;16:31–41.
16. Szummer K, Evans M, Carrero JJ, Alehagen U, Dahlström U, Benson L, et al. Comparison of the chronic kidney disease epidemiology collaboration, the modification of diet in renal disease study and the Cockcroft-Gault equation in patients with heart failure. Open Heart. 2017;4:e000568. https://doi.org/10.1136/openhrt-2016-000568.
17. Melloni C, Peterson ED, Chen AY, et al. Cockcroft-Gault versus modification of diet in renal disease: importance of glomerular filtration rate formula for classification of chronic kidney disease in patients with non-ST segment elevation acute coronary syndromes. J Am Coll Cardiol. 2008;51:991–6.
18. Goding JW, Howard MCE. Ecto-enzymes of lymphoid cells. Immunol Rev. 1998;161:5–10.
19. Abate N, Carulli L, Cabo-Chan A Jr, Chandalia M, Snell PG, Grundy SM. Genetic polymorphism PC-1 K121Q and ethnic susceptibility to insulin resistance. J Clin Endocrinol Metab. 2003;88:5927–34.

20. Abate N, Chandalia M, Satija P, Adams-Huet B, Grundy SM, Sandeep S, et al. ENPP1/PC-1 K121Q polymorphism and genetic susceptibility to type 2 diabetes. Diabetes. 2005;54:1207–13.
21. Kubaszek A, Markkanen A, Eriksson JG, Forsen T, Osmond C, Barker DJP, et al. The association of the K121Q polymorphism of the plasma cell glycoprotein-1 gene with type2 diabetes and hypertension depends on size at birth. J Clin Endocrinol Metab. 2004;89:2044–7.
22. Hsiao TJ, Lin E. The ENPP1 K121Q polymorphism is associated with type 2 diabetes and related metabolic phenotypes in a Taiwanese population. Mol Cell Endocrinol. 2016;433:20–5.
23. Yako YY, Madubedube JH, Kengne AP, Erasmus RT, Pillay TS, Matsha TE. Contribution of ENPP1, TCF7L2, and FTO polymorphisms to type 2diabetes in mixed ancestry ethnic population of South Africa. Afr Health Sci. 2015;15:1149–60.
24. Zhao T, Liu Z, Zhang D, Liu Y, Yang Y, Zhou D, et al. The ENPP1 K121Q polymorphism is not associated with type 2 diabetes or obesity in the Chinese Han population. J Hum Genet. 2011;56:12–6.
25. Bhatti JS, Bhatti GK, Mastana SS, Ralhan S, Joshi A, Tewari R. ENPP1/PC-1 K121Q polymorphism and genetic susceptibility to type 2 diabetes in North Indians. Mol Cell Biochem. 2010;345:249–57.
26. Shi X, Wang L, Jin F, Sun JF, Sun L. The ENPP1 K121Q polymorphism is not associated with type 2 diabetes in northern Chinese. Acta Diabetol. 2011;48:303–10.
27. Lee JE, Choi YK, Seo HA, Jeon JH, Jeong JY, Moon SS, et al. Impact of ENPP1 and MMP3 gene polymorphisms on aortic calcification in patients with type 2 diabetes in a Korean population. Diabetes Res Clin Pract. 2010;88:87–96.
28. De-Cosmo S, Trevisan R, Minenna A, Vedovato M, Viti R, Santini SA, et al. Insulin resistance and the cluster of abnormalities related to the metabolic syndrome are associated with reduced glomerular filtration rate in patients with type 2. Diabetes Care. 2006;29:432–4.

International Journal of Diabetes in Developing Countries (2021) 41:71–83
https://doi.org/10.1007/s13410-020-00862-z

ORIGINAL ARTICLE

Associations between plant-based dietary indices and dietary acid load with cardiovascular risk factors among diabetic patients

Elnaz Daneshzad[1] · Farzaneh Jahangir[1] · Javad Heshmati[2] · Bagher Larijani[3] · Pamela J. Surkan[4] · Leila Azadbakht[1,5]

Received: 22 January 2020 / Accepted: 10 August 2020 / Published online: 15 September 2020

Abstract

Aim To study how a plant-based diet and dietary acid load (DAL) are related to cardiovascular disease (CVD) risk factors among type 2 diabetic women.

Methods In this cross-sectional study, a validated food frequency questionnaire was used to assess the dietary intake of 230 diabetic women. We created a plant-based dietary index (PDI), healthy PDI (hPDI), and unhealthy PDI (uPDI). DAL was calculated based on potential renal acid load (PRAL) and net endogenous acid production (NEAP).

Results Patients in the highest tertile of PDI had lower fasting blood sugar (146.74 ± 6.16 vs. 152.87 ± 6.09, $p = 0.009$) and lower 2-h postprandial glucose (181.76 ± 7.03 vs. 203.01 ± 6.94, $p = 0.002$). Patients in the highest tertile of PDI were at lower risk of high waist circumference in a crude model (odd ratios: 0.43; 95% CI: 0.19; 0.96, $p = 0.039$); however, this association did not reach statistical significant after adjustment for confounders.

Conclusion Our study suggests that plant-based dietary indices and DAL are not associated with CVD risk factors among diabetic patients.

Keywords Dietary acid load · The plant-based dietary index · Cardiovascular risk factors · Diabetic patients · NEAP · PRAL

Introduction

Diabetes is a chronic metabolic disorder and an independent risk factor for cardiovascular diseases (CVD) [1]. Poor glycemic control can lead to micro- and macrovascular complications as well as cardiovascular events [2]. CVD is a major cause of death throughout the world [3]. Ischemic heart disease, hypertensive heart disease, and other cardiovascular diseases rank as the first, fourth, and ninth most common causes of mortality in Iran [4], respectively. Worldwide, it is projected that diabetes prevalence will rise to 10.1% by 2035 [5]. Several factors—including genes, family history, environmental factors, physical inactivity, stress, and diet—play an important role in development of diabetes and CVD [3, 4].

Medical nutrition therapy is important in the management of diabetes and the prevention of the related complications. As Western diets contain more protein and fewer fruits and vegetables, they can induce higher acid load and raise the risk of metabolic disorders [6]. Also, refined grains such as bread, potato, and pasta (which are food sources with a high glycemic index) can result in harmful cardio-metabolic effects [7]. A low-carbohydrate diet with more vegetable sources has been associated with lower CVD and all-cause mortality [8].

Plant-based diets (PBDs) and vegetarian diets may be beneficial in preventing diabetes [9], high blood pressure [10], obesity [11], and cardiovascular disease [12]. PBDs are based on whole plant-based foods such as fruits and vegetables and typically exclude or reduce the intake of dairy products, eggs,

Electronic supplementary material The online version of this article (https://doi.org/10.1007/s13410-020-00862-z) contains supplementary material, which is available to authorized users.

✉ Leila Azadbakht
azadbakhtleila@gmail.com

[1] Department of Community Nutrition, School of Nutritional Sciences and Dietetics, Tehran University of Medical Sciences, Tehran, Iran

[2] Department of Nutritional Science, School of Nutritional Science and Food Technology, Kermanshah University of Medical Sciences, Kermanshah, Iran

[3] Endocrinology and Metabolism Research Center, Endocrinology and Metabolism Clinical Sciences Institute, Tehran University of Medical Sciences, Tehran, Iran

[4] Department of International Health, Johns Hopkins Bloomberg School of Public Health, Baltimore, MD, USA

[5] Diabetes Research Center, Endocrinology and Metabolism Clinical Sciences Institute, Tehran University of Medical Sciences, Tehran, Iran

and meats [13]. The beneficial role of PBDs in health promotion may be due to consumption of low calorie nutritious foods [14]. On the other hand, some have emphasized the role of acid-base homeostasis in the development of cardiovascular and metabolic disorders [15]. Fruits and vegetables are net alkalinizing in nature and contribute to an increased base load, but animal proteins, meats, cheese, and eggs can increase acid load [16]. Metabolic acidosis can increase insulin resistance [15]. Several studies indicate that dietary acid load (DAL) may increase the risk of diabetes and cardiometabolic risk factors in healthy people [17, 18].

To the best of our knowledge, there is just one study on the association between DAL and CVD risk factors among diabetic nephropathy patients [19], and no studies on plant-based dietary indices and DAL in relation to CVD risk factors among diabetic patients. While the association between fruit and vegetable intake and CVD risk factors (e.g., high lipid profile, high blood pressure) has been studied, in the present study, we examine in detail how healthy and less healthy food groups as well as animal-based foods are related to CVD risk factors. Given the beneficial effects of vegan diets in preventing chronic diseases [9–12], the study of adherence to plant-based diets and their association with complications of diseases is of interest. Therefore, we aimed to explore associations between plant-based dietary indices and DAL with CVD risk factors in a sample population of type 2 diabetic women in Tehran.

Methods

In the present cross-sectional study, we enrolled 230 type 2 diabetic women (≥ 30 years old) who were referred to a diabetes research center in Tehran province, Iran (between October 2017 and October 2018). The sample size was calculated based on the mean and standard deviation (SD) of fasting blood sugar (FBS) ($[1.96^2 \times 10.9^2] / [0.02 \times 100.2]^2$) [20]. Simple random sampling was performed, and all participants provided written informed consent. As this clinic serves residents throughout Tehran, participants represented the full range of socioeconomic status. Type 2 diabetic non-pregnant women from Iran (not immigrants) who had no major medical problems were eligible for the study. Specifically, women with important illness or chronic diseases such as thyroid dysfunction, cancers, complicated cardiovascular disease, and kidney disease as well as patients who were following specific diets were excluded. We also excluded individuals who reported a total energy intake of < 800 or > 4200 kcal.

Assessment of anthropometric measures

Body weight was measured using a calibrated digital SECA scale (803, Germany) to the nearest 100 g when participants were minimally clothed and not wearing shoes. Participant height was measured to the nearest 0.1 mm while in the standing position using an unstretched tape measure. Body mass index (BMI) was calculated as weight divided by height squared (kg/m^2). Waist circumference was measured using an unstretched tape measure to the nearest 0.1 cm above light clothing at the narrowest point of the waist between the iliac crest and lowest rib.

Assessment of biochemical tests and other variables

Socio-demographic information including age, women's and their husband's education level, job, income, smoking habits, and medical history, current medications, and vitamin/mineral supplement consumption were collected on a questionnaire. Physical activity levels were recorded for a week and then converted to metabolic equivalent hours per week (MET h/wk) [21]. Blood pressure was measured twice using a sphygmomanometer with the average considered as the participant's blood pressure. Biochemical markers including FBS, 2-h postprandial blood sugar (2hpp), hemoglobin A1C (Hb A1C), total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), and triglycerides (TG) were recorded from the medical records. CVD risk factors were defined based on the Adult Treatment Panel III (ATPIII) guidelines [22]. Based on this panel, high risk on metabolic indicators was defined as follows: abdominal obesity (WC ≥ 88 in women), high serum triglycerides levels (≥ 150 mg/dL), elevated blood pressure (≥ 130/85 mmHg), low serum HDL-C (< 50), high serum LDL-C levels (≥ 130 mg/dL), abnormal serum glucose levels (FBS ≥ 110 mg/dL), high serum total cholesterol levels (≥ 200 mg/dL), and obesity (BMI ≥ 30).

Assessment of dietary intake

Dietary intake during the past year was assessed using a 168-item semi-quantitative food frequency questionnaire (FFQ), which has been validated and found reliable in the Iranian population [23]. All patients recorded the amount and frequency of each food consumed on a daily, weekly, or monthly basis during the past year. The reported portion sizes of consumed foods were converted to grams per day [24]. Nutritionist IV software (Version 7.0; N-Squared Computing, Salem, OR, USA) adapted for Iranian foods was used for nutrient analysis and estimation of energy intake.

Assessment of the plant-based dietary index

We created a plant-based dietary index (PDI), healthy plant-based dietary index (hPDI), and unhealthy plant-based dietary index (uPDI), as reported previously [25, 26]. First, 18 foods groups were created based on nutrient similarity. Then, the 18

Table 1 Participant characteristics by tertiles of plant-based indices and dietary acid load

Variables	Total	PDI tertiles			*p* value[a]	hPDI tertiles			*p* value	uPDI tertiles
		1	2	3		1	2	3		1
Number	230	77	79	74		76	76	78		76
Age (*n* (%))										
30–50 years	28 (12.2)	7 (25.0)	14 (50.0)	7 (25.0)	0.467	10 (35.7)	13 (46.4)	5 (17.9)	0.311	8 (28.6)
51–70 years	176 (76.5)	61 (34.7)	56 (31.8)	59 (33.5)		56 (31.8)	55 (31.2)	65 (36.9)		61 (34.7)
> 71 years	26 (11.3)	9 (34.6)	9 (34.6)	8 (30.8)		10 (38.5)	8 (30.8)	8 (30.8)		7 (26.9)
Weight (kg)	73.42 (11.83)	74.70 (11.40)	73.56 (13.16)	71.94 (10.72)	0.356	73.64 (12.80)	73.85 (11.80)	72.78 (11.83)	0.836	76.41 (13.47)
BMI (*n* (%)										
< 25 kg/m^2	30 (13)	8 (26.7)	7 (23.3)	15 (50.0)	0.071	9 (30.0)	9 (30.0)	12 (40.0)	0.730	8 (26.7)
≥ 25 kg/m^2	200 (87)	69 (34.7)	72 (36.2)	58 (29.1)		67 (33.7)	67 (33.7)	65 (32.7)		67 (33.7)
WC (*n* (%))										
< 88 cm	52 (22.6)	12 (23.1)	18 (34.6)	22 (42.3)	0.115	15 (28.8)	15 (28.8)	22 (42.3)	0.348	19 (36.5)
≥ 88 cm	178 (177.4)	65 (36.5)	61 (34.3)	62 (29.2)		61 (34.3)	61 (34.3)	56 (31.5)		57 (32.0)
Years having diabetes	6.58 (0.20)	6.73 (3.15)	5.95 (3.27)	7.11 (3.01)	0.069	6.09 (3.17)	6.79 (3.23)	6.86 (3.10)	0.257	6.57 (3.32)
Years post-menopause	10.81 (0.58)	11.96 (8.46)	9.29 (8.72)	11.23 (9.31)	0.151	10.05 (7.66)	11.25 (8.62)	11.12 (10.17)	0.661	10.76 (9.56)
SES score	20.59 (4.72)	19.84 (4.83)	20.78 (4.56)	21.16 (4.73)	0.209	19.32 (5.04)	20.32 (4.61)	22.07 (4.11)	*0.001*	22.27 (4.07)
PA (met.h/wk)					0.477				0.171	
Light (*n* (%)	76 (33.0)	28 (36.8)	29 (38.2)	19 (25.0)		33 (43.4)	22 (28.9)	21 (27.6)		25 (32.9)
Moderate	77 (33.5)	24 (31.2)	23 (29.9)	30 (39.0)		22 (28.6)	29 (37.7)	26 (33.8)		21 (27.3)
Vigorous	77 (33.5)	25 (32.5)	27 (35.1)	25 (32.5)		21 (27.3)	25 (32.5)	31 (40.3)		30 (39.0)
Supplement consumption (*n* (%))										
Yes	59 (25.70)	17 (28.80)	18 (30.50)	24 (40.70)	0.267	17 (28.80)	18 (30.50)	24 (40.70)	0.437	20 (33.90)
No	171 (74.30)	60 (35.10)	61 (35.70)	50 (29.20)		59 (34.50)	58 (33.90)	54 (31.60)		56 (32.70)

Variables	uPDI tertiles		*p* value	NEAP tertiles			*p* value	PRAL tertiles			*p* value
	2	3		1	2	3		1	2	3	
Number	73	81		76	77	77		76	77	77	
Age (*n* (%))											
30–50 years	13 (46.4)	7 (25.0)	0.123	10 (35.7)	12 (42.9)	6 (21.4)	0.547	9 (32.1)	13 (46.4)	6 (21.4)	0.555
51–70 years	55 (31.2)	60 (34.1)		59 (33.5)	57 (32.4)	60 (34.1)		58 (33.0)	56 (31.8)	62 (35.2)	
> 71 years	5 (19.2)	14 (53.8)		7 (26.9)	8 (30.8)	11 (42.3)		9 (34.6)	8 (30.8)	9 (34.6)	
Weight (kg)	71.67 (10.77)	72.20 (10.65)	*0.025*	74.12 (12.77)	73.17 (11.09)	72.98 (11.70)	0.815	74.41 (12.60)	72.96 (10.62)	72.90 (12.28)	0.673
BMI (*n* (%)											
< 25 kg/m^2	10 (33.3)	12 (40.0)	0.733	8 (26.7)	11 (36.7)	11 (36.7)	0.718	5 (16.7)	13 (43.3)	12 (40.0)	0.128

Table 1 (continued)

≥ 25 kg/m^2	63 (31.7)	69 (34.7)		68 (34.2)	65 (32.7)	66 (33.2)		70 (35.2)	64 (32.2)	65 (32.7)	
WC (*n* (%))											
< 88 cm	12 (23.1)	21 (40.4)	0.309	20 (38.5)	17 (32.7)	15 (28.8)	0.595	17 (32.7)	22 (42.3)	13 (25.0)	0.222
≥ 88 cm	61 (34.3)	60 (33.7)		56 (31.5)	60 (33.7)	62 (34.8)		59 (33.1)	55 (30.9)	64 (36.0)	
Years having diabetes	6.85 (3.31)	6.36 (2.91)	0.633	6.68 (2.92)	6.49 (3.35)	6.57 (3.27)	0.933	6.74 (3.10)	6.32 (3.26)	6.69 (3.18)	0.681
Years post-menopause	9.62 (8.59)	11.93 (8.39)	0.273	10.70 (10.14)	9.45 (7.39)	12.27 (8.76)	0.142	10.79 (10.09)	9.66 (8.36)	11.97 (8.87)	0.271
SES score	20.75 (4.89)	18.86 (4.57)	*< 0.0001*	21.28 (4.01)	20.77 (4.96)	19.71 (5.04)	0.108	22.11 (4.09)	20.16 (5.01)	19.50 (4.68)	*0.002*
PA (met.h/wk)			*0.001*				0.066				0.202
Light (*n* (%)	14 (18.4)	37 (48.7)		21 (27.6)	26 (34.2)	29 (38.2)		19 (25.0)	24 (31.6)	33 (43.4)	
Moderate	27 (35.1)	29 (37.7)		33 (42.9)	18 (23.4)	26 (33.8)		29 (37.7)	25 (32.5)	23 (29.9)	
Vigorous	32 (41.6)	15 (19.5)		22 (28.6)	33 (42.9)	22 (28.6)		28 (36.4)	28 (36.4)	21 (27.3)	
Supplement consumption (*n* (%))											
Yes	16 (27.10)	23 (39.00)	0.647	26 (44.10)	16 (27.10)	17 (28.80)	0.111	22 (37.30)	20 (33.90)	17 (28.80)	0.621
No	57 (33.30)	58 (33.90)		50 (29.20)	61 (35.70)	60 (35.10)		54 (31.60)	57 (33.30)	60 (35.10)	

Significant *p*-values present as italic format

PDI plant-based diet index, *hPDI* healthy plant-based diet index, *uPDI* unhealthy plant-based diet index, *NEAP* net endogenous acid production, *PRAL* potential renal acid load, *BMI* body mass index, *WC* waist circumference, *PA* physical activity, *SES* socio-economic status

[a]Calculated using Chi-square tests and *t* tests and presented as mean (SD) or number (%)

food groups were classified into three groupings: animal foods, healthy plant foods, and unhealthy plant foods. The healthy plant food grouping included whole grains, legumes, nuts, fruits, vegetables, vegetable oils, tea, and coffee, whereas the less healthy plant food grouping included potatoes, grains, sweets, sugar-sweetened beverages, refined fruit juices, and desserts. The animal food grouping included animal fats, dairy, fish and seafood, eggs, poultry, and red meat and miscellaneous animal-based foods. Details of the food groupings are shown in Supplemental Table 1. We divided the 18 food groups into deciles of consumption designated with scores from 1 to 10. For the PDI, a high score of 10 corresponded to the highest decile and 1 corresponded to the lowest decile of all plant-based foods. Participants scoring in the highest decile of the animal diet were given a score of 1 and those in the lowest decile a score of 10. For the hPDI, healthy plant food groups were assigned higher scores, and lower scores corresponded to higher consumption in the unhealthy plant food groups and animal food groups. For uPDI, a score of 1 indicated the lowest decile of the unhealthy plant food groups and a score of 10 indicated the highest decile (Supplemental Table 1) [26]. Finally, all 18 food group scores were summed (range 18–180), such that a higher overall score (combining all three indices) reflected lower animal food intake. Finally, these three PDI indices were categorized into tertiles to assess their relationships with CVD risk factors.

Assessment of dietary acid load

DAL was calculated based on potential renal acid load (PRAL) [27, 28] and net endogenous acid production (NEAP) methods [6]. PRAL was calculated based on protein, potassium (K), calcium (Ca), and magnesium (mg) intake.

$$\begin{aligned}\mathrm{PRAL(mEq/d)} = {} & (\mathrm{protein[g/d]} \times 0.49) \\ & + (\mathrm{P[mg/d]} \times 0.037) - (\mathrm{K[mg/d]} \times 0.021) \\ & - (\mathrm{Ca[mg/d]} \times 0.013) - (\mathrm{mg[mg/d]} \times 0.026)\end{aligned}$$

NEAP was estimated: mEq/d = (54.5 × protein intake[g/d]/K intake[mEq/d]) − 10.2.

Statistical analysis

Participant characteristics were assessed in relation to dietary scores using ANOVA or Chi-square tests for continuous and categorical variables, respectively, and reported as the mean ± standard deviation (SD) or percentage. Dietary intakes were calculated across tertiles of adherence to plant-based dietary indices and DAL after adjustment for energy intake using analysis of covariance (ANCOVA). Risk of metabolic factors in diabetic patients is presented in crude and adjusted models

Table 2 Dietary intakes by tertiles of plant-based diet indices and dietary acid load

Variables	Total ($n = 230$)	PDI			p value[a]	hPDI			p value[a]	uPDI	
		1	2	3		1	2	3		1	2
Energy (kcal/day)	2294.8 (496.34)	2360.2 (486.91)	2262.2 (475.74)	2261.6 (526.66)	0.368	2295 (445.76)	2117.4 (405.27)	2467.5 (563.81)	*< 0.0001*	2334.9 (574.46)	2280 (473.56)
CHO (%)	58.37 (0.56)	56.34 (0.88)	58.87 (0.87)	59.94 (0.90)	*0.015*	59.84 (0.89)	58.55 (0.91)	56.77 (0.90)	0.055	55.69 (0.87)	58.32 (0.88)
Protein (%)	12.38 (0.10)	12.07 (0.18)	12.54 (0.18)	12.54 (0.19)	0.119	11.56 (0.17)	12.27 (0.17)	13.30 (0.17)	*< 0.0001*	13.72 (0.14)	12.43 (0.14)
Fat (%)	32.41 (0.58)	34.42 (0.88)	31.69 (0.86)	31.09 (0.89)	*0.019*	31.02 (0.88)	32.27 (0.90)	33.90 (0.89)	0.077	34.48 (0.88)	32.29 (0.89)
Cholesterol (mg/day)	171.86 (4.27)	201.4 (6.77)	169.9 (6.67)	143.2 (6.89)	*< 0.0001*	201.7 (6.79)	171.2 (6.95)	143.4 (6.86)	*< 0.0001*	165.6 (7.06)	196.3 (7.19)
SFA (mg/day)	20.45 (0.44)	22.95 (0.47)	20.21 (0.46)	18.10 (0.48)	*< 0.0001*	22.43 (0.49)	20.64 (0.50)	18.32 (0.49)	*< 0.0001*	19.76 (0.52)	21.59 (0.53)
EPA	0.01 (0.001)	0.01 (0.002)	0.02 (0.002)	0.01 (0.003)	0.099	0.01 (0.002)	0.01 (0.003)	0.02 (0.003)	0.192	0.02 (0.002)	0.01 (0.002)
DHA	0.04 (0.003)	0.03 (0.006)	0.05 (0.006)	0.03 (0.006)	0.099	0.03 (0.006)	0.03 (0.006)	0.05 (0.006)	0.192	0.06 (0.006)	0.04 (0.006)
Fiber (g/day)	20.84 (0.43)	18.51 (0.50)	20.97 (0.49)	23.13 (0.51)	*< 0.0001*	16.63 (0.39)	21.07 (0.40)	24.72 (0.39)	*< 0.0001*	24.67 (0.43)	20.44 (0.44)
Sodium	3546.1 (93.74)	3905 (148.55)	3395 (146.34)	3334 (151.20)	*0.013*	3989 (146.86)	3498 (150.22)	3162 (148.19)	*< 0.0001*	3083 (146.37)	3579 (149.21)
Potassium	3595.8 (69.03)	3246 (81.82)	3656 (80.60)	3895 (83.28)	*< 0.0001*	3173 (77.58)	3557 (79.36)	4045 (78.29)	*< 0.0001*	4153 (73.33)	3505 (74.76)
Iron	17.44 (0.24)	16.18 (0.28)	17.77 (0.28)	18.39 (0.29)	*< 0.0001*	16.22 (0.27)	17.16 (0.28)	18.90 (0.28)	*< 0.0001*	19.09 (0.27)	17.16 (0.27)
Calcium	837.50 (16.87)	799.1 (25.80)	869.3 (25.42)	843.5 (26.26)	0.150	759.7 (25.01)	830.6 (25.58)	920 (25.24)	*< 0.0001*	968.3 (22.82)	851.3 (23.26)
Phosphorus	1110.55 (21.26)	1058 (22.24)	1135 (21.91)	1139 (22.64)	*0.016*	1010 (20.69)	1108 (21.16)	1211 (20.88)	*< 0.0001*	1253 (18.67)	1107 (19.04)
Vitamin A (RAE/day)	0.005 (0.0002)	954.1 (68.88)	1164 (67.85)	1328 (70.11)	*0.001*	996.4 (67.31)	995.4 (68.85)	1440 (67.92)	*< 0.0001*	1454 (65.68)	1134 (66.95)
Vitamin E (mg/day)	187.12 (6.83)	3.40 (0.14)	3.87 (0.14)	4.03 (0.14)	*0.007*	3.54 (0.14)	3.75 (0.14)	4.01 (0.14)	0.083	4.11 (0.14)	3.65 (0.14)
Vitamin C (mg/day)	3.76 (0.08)	114.9 (4.93)	129 (4.86)	145.6 (5.02)	*< 0.0001*	108.2 (4.79)	131.4 (4.90)	148.7 (4.83)	*< 0.0001*	152.7 (4.63)	132.3 (4.72)
Vitamin B1	129.61 (3.05)	1.79 (0.02)	1.92 (0.02)	1.96 (0.02)	*< 0.0001*	1.84 (0.02)	1.90 (0.03)	1.93 (0.03)	0.114	1.86 (0.03)	1.93 (0.03)
Vitamin B2	1.89 (0.02)	1.31 (0.04)	1.39 (0.03)	1.34 (0.04)	0.425	1.24 (0.03)	1.31 (0.03)	1.48 (0.03)	*< 0.0001*	1.58 (0.03)	1.36 (0.03)
Vitamin B3	1.35 (0.02)	20.87 (0.35)	22.54 (0.35)	23.52 (0.36)	*< 0.0001*	20.68 (0.34)	21.83 (0.34)	24.32 (0.34)	*< 0.0001*	23.58 (0.36)	21.88 (0.37)
Vitamin B6	22.29 (0.42)	1.28 (0.03)	1.43 (0.03)	1.50 (0.03)	*< 0.0001*	1.25 (0.03)	1.40 (0.03)	1.56 (0.03)	*< 0.0001*	1.58 (0.03)	1.41 (0.03)
Vitamin B12	1.40 (0.02)	2.68 (0.11)	2.76 (0.11)	2.29 (0.11)	*0.012*	2.76 (0.11)	2.50 (0.12)	2.49 (0.12)	0.199	2.90 (0.11)	2.70 (0.11)
Folate	2.58 (0.06)	277.2 (11.40)	327.7 (11.23)	374.9 (11.60)	*< 0.0001*	254.8 (10.24)	320.1 (10.47)	401.1 (10.33)	*< 0.0001*	403.8 (10.18)	320.2 (10.38)
Red meat (g/day)	325.99 (8.24)	22.97 (1.22)	20.46 (1.20)	17.72 (1.24)	*0.012*	22.44 (1.23)	20.71 (1.26)	18.16 (1.24)	0.053	22.41 (1.24)	19.56 (1.26)
Organ meat (g/day)	20.42 (0.72)	1.93 (0.29)	1.31 (0.28)	0.60 (0.29)	*0.007*	2.58 (0.27)	0.95 (0.28)	0.35 (0.28)	*< 0.0001*	0.56 (0.29)	1.57 (0.29)
Processed meat (g/day)	1.29 (0.57)	5.93 (0.98)	3.14 (0.97)	1.57 (1.01)	*0.008*	7.88 (0.94)	2.06 (0.96)	0.84 (0.95)	*< 0.0001*	1.82 (0.99)	2.83 (1.01)
Fish (g/day)	3.57 (0.57)	6.28 (0.88)	8.14 (0.87)	6.19 (0.90)	0.215	6.17 (0.89)	6.75 (0.91)	7.72 (0.90)	0.470	9.68 (0.85)	7.37 (0.87)
Poultry (g/day)	6.89 (0.51)	14.89 (1.01)	15.32 (0.98)	13.23 (1.02)	0.305	14.86 (1.01)	14.37 (1.03)	14.28 (1.02)	0.911	17.15 (0.98)	14.66 (0.99)
Egg (g/day)	14.50 (0.58)	21.86 (1.24)	17.29 (1.22)	15.90 (1.26)	*0.002*	20.58 (1.26)	18.89 (1.29)	15.72 (1.27)	*0.025*	17.98 (1.24)	21.81 (1.27)
Whole grains (g/day)	18.37 (0.73)	36.90 (6.12)	51.46 (6.03)	68.99 (6.23)	*0.001*	28.65 (5.94)	52.27 (6.07)	75.17 (5.99)	*< 0.0001*	70.85 (6.08)	52.94 (6.20)
Refined grains (g/day)	52.23 (3.63)	367.61 (17.14)	378.14 (16.89)	339.17 (17.45)	0.257	424.56 (16.19)	369.75 (16.56)	293.72 (16.33)	*< 0.0001*	281.01 (15.64)	362.86 (15.94)
Legumes (g/day)	24.55 (0.79)	20.59 (1.28)	23.74 (1.26)	29.55 (1.30)	*< 0.0001*	19.58 (1.26)	24.18 (1.29)	29.77 (1.28)	*< 0.0001*	31.67 (1.20)	23.52 (1.23)
Total dairy (g/day)	274.48 (9.67)	300.57 (15.98)	285.34 (15.74)	235.73 (16.27)	*0.013*	286.69 (16.31)	273.16 (16.69)	263.86 (16.46)	0.611	315.73 (15.94)	272.45 (16.24)
Low-fat dairy (g/day)	362.08 (9.96)	145.66 (16.68)	184.35 (16.43)	178.34 (16.98)	0.213	111.45 (16.06)	175.71 (16.42)	219.90 (16.20)	*< 0.0001*	265.34 (14.80)	147.25 (15.08)

Table 2 (continued)

High-fat dairy (g/day)	169.46 (9.69)	170.04 (14.31)	113.01 (14.10)	65.59 (14.57)	*< 0.0001*	191.35 (13.65)	110.42 (13.96)	50.50 (13.77)	*< 0.0001*	60.16 (14.45)	135.95 (14.73)
Vegetables (g/day)	116.84 (8.79)	350.99 (20.78)	441.80 (20.47)	499.84 (21.15)	*< 0.0001*	332.40 (20.29)	443.93 (20.75)	511.72 (20.47)	*< 0.0001*	536.69 (19.54)	436.74 (19.92)
Starchy vegetables (g/day)	430.07 (12.60)	77.86 (6.34)	89.63 (6.24)	79.83 (6.45)	0.368	106.51 (5.92)	86.53 (6.05)	55.28 (5.97)	*< 0.0001*	57.73 (5.84)	76.52 (5.96)
Fruit (g/day)	82.53 (3.77)	236.20 (11.89)	264.58 (11.71)	313.36 (12.10)	*< 0.0001*	214.27 (11.37)	272.83 (11.63)	323.82 (11.47)	*< 0.0001*	325.50 (11.40)	271.70 (11.62)
Fruit juice (g/day)	270.78 (7.50)	0.97 (0.56)	3.35 (0.55)	3.45 (0.57)	*0.003*	3.08 (0.57)	1.37 (0.59)	3.29 (0.58)	*0.045*	3.40 (0.58)	2.54 (0.59)
Tea and coffee (g/day)	2.58 (0.33)	507.84 (35.92)	583.72 (35.39)	741.15 (36.56)	*< 0.0001*	575.54 (37.44)	583.69 (38.30)	666.17 (37.78)	0.181	660.96 (37.48)	603.40 (38.21)

Variables	uPDI	*p* value[a]	NEAP			*p* value[a]	PRAL			*p* value[a]
	3		1	2	3		1	2	3	
Energy (kcal/day)	2270.6 (437.77)	0.688	2300.7 (488.97	2222.6 (495.49)	2361.2 (501.05)	0.222	2441.3 (518.54)	2171.9 (516.66)	2273.1 (415.64)	*0.003*
CHO (%)	60.93 (0.84)	*< 0.0001*	61.41 (0.84)	59.12 (0.84)	54.62 (0.84)	*< 0.0001*	61.05 (0.86)	59.38 (0.85)	54.71 (0.84)	*< 0.0001*
Protein (%)	11.08 (0.13)	*< 0.0001*	11.83 (0.18)	12.58 (0.18)	12.73 (0.18)	*0.001*	12.43 (0.19)	12.36 (0.18)	12.36 (0.18)	0.960
Fat (%)	30.58 (0.85)	*0.007*	30.39 (0.86)	31.32 (0.85)	35.49 (0.85)	*< 0.0001*	30.30 (0.87)	31.26 (0.86)	35.64 (0.85)	*< 0.0001*
Cholesterol (mg/day)	155.7 (6.83)	*< 0.0001*	166.9 (7.26)	165.3 (7.24)	183.3 (7.24)	0.157	168.1 (7.40)	166.9 (7.32)	180.5 (7.24)	0.341
SFA (mg/day)	20.06 (0.50)	*0.032*	20.070 (0.51)	19.38 (0.51)	21.88 (0.51)	*0.002*	19.75 (0.51)	19.51 (0.51)	22.07 (0.50)	*0.001*
EPA	0.009 (0.002)	*< 0.0001*	0.016 (0.002)	0.02 (0.002)	0.014 (0.002)	*0.011*	0.021 (0.003)	0.018 (0.002)	0.015 (0.002)	0.174
DHA	0.02 (0.005)	*< 0.0001*	0.037 (0.006)	0.056 (0.006)	0.033 (0.006)	*0.011*	0.05 (0.006)	0.04 (0.006)	0.03 (0.006)	0.174
Fiber (g/day)	17.62 (0.41)	*< 0.0001*	22.96 (0.50)	20.98 (0.50)	18.62 (0.50)	*< 0.0001*	23.92 (0.47)	20.80 (0.47)	17.85 (0.46)	*< 0.0001*
Sodium	3951 (141.69)	*< 0.0001*	3377 (150.37)	3447 (149.93)	3812 (149.84)	0.091	3262 (149.81)	3390 (148.20)	3983 (146.67)	*0.001*
Potassium	3154 (70.99)	*< 0.0001*	4195 (67.03)	3603 (66.83)	2996 (66.80)	*< 0.0001*	4347 (59.57)	3491 (58.93)	2960 (58.32)	*< 0.0001*
Iron	16.14 (0.26)	*< 0.0001*	18.96 (0.27)	17.46 (0.26)	15.91 (0.26)	*< 0.0001*	19.40 (0.26)	17.03 (0.25)	15.91 (0.25)	*< 0.0001*
Calcium	702.3 (22.09)	*< 0.0001*	826.2 (26.01)	866.3 (25.93)	819.8 (25.91)	0.392	890.1 (26.16)	817.2 (25.88)	805.8 (25.61)	0.050
Phosphorus	980.1 (18.08)	*< 0.0001*	1121 (22.49)	1139 (22.43)	1071 (22.42)	0.090	1163 (22.69)	1087 (22.44)	1082 (22.21)	*0.021*
Vitamin A (RAE/day)	868.7 (63.58)	*< 0.0001*	1473 (65.72)	1070 (65.53)	901 (65.49)	*< 0.0001*	1594 (62.10)	1011 (61.43)	839.9 (60.80)	*< 0.0001*
Vitamin E (mg/day)	3.54 (0.13)	*0.011*	4.27 (0.13)	3.86 (0.13)	3.16 (0.13)	*< 0.0001*	4.42 (0.13)	3.68 (0.13)	3.20 (0.13)	*< 0.0001*
Vitamin C (mg/day)	105.5 (4.49)	*< 0.0001*	153.3 (4.48)	135.6 (4.47)	100.3 (4.47)	*< 0.0001*	162.1 (4.20)	130.7 (4.15)	96.39 (4.11)	*< 0.0001*
Vitamin B1	1.88 (0.02)	0.235	0.87 (0.02)	1.94 (0.02)	1.85 (0.02)	0.087	1.87 (0.03)	1.95 (0.02)	1.85 (0.02)	*0.036*
Vitamin B2	1.12 (0.03)	*< 0.0001*	1.33 (0.04)	1.37 (0.04)	1.34 (0.04)	0.827	1.43 (0.04)	1.29 (0.04)	1.32 (0.03)	*0.036*
Vitamin B3	21.46 (0.35)	*< 0.0001*	22.01 (0.38)	22.25 (0.38)	22.63 (0.37)	0.506	22.13 (0.38)	22.61 (0.38)	22.14 (0.37)	0.605
Vitamin B6	1.23 (0.03)	*< 0.0001*	1.57 (0.03)	1.39 (0.03)	1.25 (0.03)	*< 0.0001*	1.62 (0.03)	1.36 (0.03)	1.23 (0.03)	*< 0.0001*
Vitamin B12	2.17 (0.11)	*< 0.0001*	2.39 (0.11)	2.78 (0.11)	2.56 (0.11)	0.065	2.67 (0.12)	2.45 (0.12)	2.62 (0.11)	0.387

Table 2 (continued)

Folate	258.2 (9.85)	*< 0.0001*	357.2 (11.97)	325.6 (11.94)	295.6 (11.93)	*0.002*	380.4 (11.54)	318.9 (11.42)	279.4 (11.30)	*< 0.0001*
Red meat (g/day)	19.32 (1.20)	0.148	19.61 (1.25)	21.30 (1.24)	20.33 (1.24)	0.630	21.12 (1.27)	20.09 (1.25)	20.06 (1.24)	0.797
Organ meat (g/day)	1.71 (0.28)	*0.010*	1.21 (0.29)	1.09 (0.29)	1.56 (0.29)	0.507	0.95 (0.30)	1.36 (0.29)	1.54 (0.29)	0.374
Processed meat (g/day)	5.88 (0.96)	*0.010*	1.86 (1.01)	4.58 (0.99)	4.24 (0.99)	0.113	1.69 (0.99)	2.56 (0.98)	6.43 (0.97)	*0.002*
Fish (g/day)	3.84 (0.82)	*< 0.0001*	6.25 (0.88)	8.95 (0.87)	5.46 (0.87)	*0.015*	8.01 (0.90)	7.06 (0.89)	5.62 (0.88)	0.164
Poultry (g/day)	11.88 (0.94)	*0.001*	12.53 (0.99)	14.48 (0.99)	16.47 (0.99)	*0.021*	13.76 (1.02)	14.64 (1.01)	15.09 (1.01)	0.645
Egg (g/day)	15.65 (1.20)	*0.002*	18.47 (1.27)	17.26 (1.27)	19.39 (1.27)	0.499	18.50 (1.30)	18.01 (1.28)	18.61 (1.27)	0.939
Whole grains (g/day)	34.11 (5.89)	*< 0.0001*	49.31 (6.32)	55.15 (6.30)	52.18 (6.30)	0.807	62.94 (6.34)	40.82 (6.27)	53.05 (6.20)	0.051
Refined grains (g/day)	437.44 (15.14)	*< 0.0001*	360.4 (17.31)	365.1 (17.26)	360.7 (17.25)	0.978	324.1 (17.29)	389.2 (17.10)	372.5 (16.92)	*0.025*
Legumes (g/day)	18.81 (1.16)	*< 0.0001*	25.57 (1.34)	25.84 (1.33)	22.27 (1.33)	0.114	27.27 (1.33)	25.87 (1.32)	20.56 (1.30)	*0.001*
Total dairy (g/day)	237.60 (15.43)	*0.002*	247.43 (16.20)	292.68 (16.15)	282.97 (16.14)	0.117	272.05 (16.59)	270.95 (16.41)	280.41 (16.24)	0.904
Low-fat dairy (g/day)	99.52 (14.32)	*< 0.0001*	173.5 (16.80)	181.7 (16.75)	153.2 (16.74)	0.468	204.6 (16.84)	163.3 (16.65)	140.9 (16.48)	*0.026*
High-fat dairy (g/day)	152.79 (13.99)	*< 0.0001*	87.57 (14.96)	121.3 (14.91)	141.3 (14.90)	*0.039*	80.48 (15.04)	118.6 (14.87)	151 (14.72)	*0.004*
Vegetables (g/day)	324.03 (18.91)	*< 0.0001*	500.6 (20.80)	441 (20.74)	349.5 (20.73)	*< 0.0001*	535.9 (19.81)	436.6 (19.60)	319.1 (19.40)	*< 0.0001*
Starchy vegetables (g/day)	111.23 (5.66)	*< 0.0001*	108.3 (5.91)	82.96 (5.89)	56.70 (5.89)	*< 0.0001*	100.2 (6.30)	79.74 (6.23)	67.94 (6.17)	*0.001*
Fruit (g/day)	218.59 (11.03)	*< 0.0001*	315.5 (11.35)	288 (11.31)	209.4 (11.31)	*< 0.0001*	334.8 (10.94)	277.2 (10.82)	201.2 (10.71)	*< 0.0001*
Fruit juice (g/day)	1.85 (0.56)	0.161	3.04 (0.58)	2.53 (0.58)	2.19 (0.58)	0.586	3.97 (0.58)	1.91 (0.57)	1.89 (0.57)	*0.017*
Tea and coffee (g/day)	565.19 (36.28)	0.185	688.8 (37.16)	576.6 (37.05)	562.6 (37.03)	*0.033*	663.4 (37.95)	609.5 (37.54)	554.7 (37.15)	0.127

Significant *p*-values present as italic format

[a] Calculated using *t* tests for energy intake and multivariate analysis of covariance (ANCOVA) for other variables. All variables, except energy, were adjusted for energy intake

CHO carbohydrate, *SFA* saturated fatty acid, *MUFA* mono-unsaturated fatty acid, *PUFA* poly-unsaturated fatty acid

Table 3 Biochemical measures by tertiles of plant-based diet indices and dietary acid load

Variables	Total ($n = 230$)	PDI			p value[a]	hPDI			p value[a]	uPDI	
		1	2	3		1	2	3		1	2
FBS	157.40 (3.47)										
Crude		152.53 (5.01)	171.96 (6.66)	146.93 (5.88)	*0.008*	167.84 (6.20)	156.81 (6.56)	147.80 (5.06)	0.061	149.77 (6.02)	161.10 (5.88)
Adjusted model		152.87 (6.09)	171.80 (5.89)	146.74 (6.16)	*0.009*	167.89 (6.08)	159.09 (6.12)	145.53 (6.16)	*0.041*	150.31 (6.28)	162.50 (6.31)
2hPPG	200.93 (4.07)										
Crude		203.35 (6.70)	216.44 (7.49)	181.87 (6.34)	*0.002*	210.39 (6.60)	201.43 (7.72)	191.24 (6.70)	0.157	201.31 (8.24)	201.61 (6.76)
Adjusted model		203.01 (6.94)	216.88 (6.72)	181.76 (7.03)	*0.002*	209.41 (7.02)	203.77 (7.07)	189.91 (7.12)	0.150	204.84 (7.23)	203.99 (7.26)
HbA1c (%)	7.54 (0.07)										
Crude		7.38 (0.11)	7.81 (0.12)	7.44 (0.14	0.036	7.54 (0.11)	7.58 (0.13)	7.51 (0.13)	0.935	7.54 (0.14)	7.56 (0.13)
Adjusted model		7.31 (0.13)	7.83 (0.12)	7.48 (0.13)	*0.018*	7.52 (0.13)	7.60 (0.13)	7.51 (0.13)	0.868	7.55 (0.13)	7.56 (0.13)
TC	160.25 (2.68)										
Crude		157.70 (4.55)	159.11 (4.29)	164.13 (5.13)	0.597	161.38 (4.57)	158.73 (4.40)	160.64 (4.97)	0.919	162.03 (4.89)	155.12 (5.17)
Adjusted model		156.36 (4.76)	159.28 (4.60)	165.33 (4.82)	0.417	159.52 (4.73)	157.30 (4.76)	163.84 (4.79)	0.632	164.46 (4.82)	154.75 (4.84)
TG	160.59 (5.05)										
Crude		160.49 (7.46)	151.34 (7.43)	170.58 (11.07)	0.301	164.11 (8.09)	155.31 (6.98)	162.30 (10.71)	0.757	156.19 (9.28)	160.84 (8.12)
Adjusted model		159.95 (8.94)	149.03 (8.65)	173.60 (9.05)	0.150	161.38 (8.91)	151.65 (8.97)	168.53 (9.03)	0.429	157.26 (9.15)	161.50 (9.19)
LDL	97.70 (2.16)										
Crude		98.61 (3.99)	96.48 (3.08)	98.05 (4.18)	0.916	103.19 (3.73)	92.19 (3.42)	97.70 (3.99)	0.118	95.82 (3.64)	96.49 (4.05)
Adjusted model		97.51 (3.91)	96.77 (3.78)	98.87 (3.96)	0.928	102.41 (3.84)	91.34 (3.86)	99.34 (3.89)	0.116	96.39 (3.96)	95.80 (3.98)
HDL	45.87 (0.67)										
Crude		45.49 (0.90)	45.41 (1.33)	46.77 (1.21)	0.660	44.92 (1.31)	46.38 (0.95)	46.32 (1.19)	0.608	46.81 (1.10)	44.60 (0.90)
Adjusted model		45.17 (1.18)	45.66 (1.15)	46.84 (1.20)	0.609	45.01 (1.18)	46.45 (1.18)	46.17 (1.19)	0.658	46.91 (1.20)	44.70 (1.21)

Variables	uPDI	p value[a]	NEAP			p value[a]	PRAL			p value[a]
	3		1	2	3		1	2	3	
FBS										
Crude	161.22 (6.07)	0.305	155.64 (5.92)	161.36 (6.51)	155.18 (5.60)	0.721	153.72 (5.79)	151.16 (5.49)	167.27 (6.06)	0.125
Adjusted model	159.45 (6.16)	0.371	155.43 (6.13)	162.88 (6.09)	153.87 (6.11)	0.542	153.30 (6.17)	151.75 (6.10)	167.09 (6.07)	0.151
2hPPG										
Crude	199.97 (6.14)	0.985	193.51 (7.17)	201.80 (7.10)	207.40 (6.88)	0.377	192.32 (7.27)	201.55 (7.19)	208.81 (6.64)	0.255
Adjusted model	194.51 (7.09)	0.549	194.72 (7.04)	204.10 (7.01)	203.90 (7.02)	0.565	192.44 (7.11)	204.04 (7.03)	206.21 (7.00)	0.347
HbA1c (%)										
Crude	7.53 (0.11)	0.988	7.46 (0.12)	7.61 (0.13)	7.56 (0.12)	0.715	7.50 (0.13)	7.54 (0.11)	7.59 (0.13)	0.879
Adjusted model	7.53 (0.13)	0.981	7.50 (0.13)	7.62 (0.13)	7.51 (0.13)	0.797	7.49 (0.13)	7.58 (0.13)	7.56 (0.13)0	0.869
TC										
Crude	163.20 (3.89)	0.421	157.65 (5.03)	168.89 (4.65)	154.18 (4.09)	0.063	158.60 (5.08)	164.12 (4.72)	158.01 (4.10)	0.591

Table 3 (continued)

Adjusted model	161.26 (4.73)	0.354	159.01 (4.66)	168.91 (4.63)	152.82 (4.65)	0.052	160.85 (4.77)	163.25 (4.71)	156.67 (4.69)	0.609
TG										
Crude	164.49 (8.81)	0.796	154.82 (8.34)	163.45 (9.63)	163.42 (8.26)	0.727	159.03 (9.37)	156.32 (8.10)	166.40 (8.80)	0.702
Adjusted model	162.90 (8.97)	0.906	156.90 (8.91)	161.48 (8.86)	163.35 (8.89)	0.872	163.22 (9.01)	153.87 (8.90)	164.71 (8.86)	0.652
LDL										
Crude	100.54 (3.60)	0.623	94.60 (3.90)	101.57 (3.43)	96.88 (3.89)	0.410	95.82 (3.84)	101.92 (3.62)	95.32 (3.77)	0.384
Adjusted model	100.61 (3.88)	0.653	95.42 (3.85)	101.24 (3.83)	96.40 (3.84)	0.523	96.63 (3.90)	101.74 (3.85)	94.70 (3.83)	0.416
HDL										
Crude	46.14 (1.37)	0.400	45.96 (1.04)	45.51 (0.98)	46.15 (1.42)	0.925	45.02 (0.99)	46.22 (0.99)	46.37 (1.44)	0.672
Adjusted model	45.96 (1.18)	0.431	46.14 (1.17)	45.77 (1.17)	45.72 (1.17)	0.963	44.84 (1.18)	46.79 (1.17)	45.98 (1.16)	0.517

Significant *p*-values present as italic format

[a] Calculated using *t* tests, a crude model, and multivariate analysis of covariance (ANCOVA) for the adjusted models. Data are presented as mean and SE

Adjusted model: All the variables were adjusted for age, BMI, socio-economic status, physical activity, vitamin D, energy, and supplement intake

PDI plant-based diet index, *hPDI* healthy plant-based diet index, *uPDI* unhealthy plant-based diet index, *NEAP* net endogenous acid production, *PRAL* potential renal acid load, *FBS* fasting blood glucose, *2hppG* 2-hour post-prandial glucose, *HbA1c* hemoglobin A1C, *TC* total cholesterol, *TG* triglyceride, *LDL* low-density lipoprotein, *HDL* high-density lipoprotein

using binary logistic regression. Age, BMI, physical activity, socioeconomic status, vitamin/mineral supplement consumption, vitamin D, and energy intake were adjusted for in multivariable models.

Results

Participant characteristics are displayed in Table 1. Mean age was 59.90 (9.20) years. There was a significant association between weight and uPDI ($p = 0.025$). A significant difference was observed in socioeconomic status across tertiles of PRAL, hPDI, and uPDI ($p < 0.05$). Physical activity differed significantly across tertiles of uPDI ($p = 0.001$).

Table 2 presents energy-adjusted dietary intakes among plant-based dietary indices, NEAP and PRAL. Consumption of total carbohydrates, sodium, organ meats, processed meats, high-fat dairy, starchy vegetables, and refined grains was greater in the highest tertile of uPDI ($p < 0.05$). Consumption of carbohydrates, eicosapentaenoic acid, docosapentaenoic acid, fiber, potassium, iron, and vitamins A, E, C, and B6 was lower in the highest tertile of NEAP ($p < 0.05$). Consumption of total carbohydrates, fiber, potassium, iron, calcium, phosphorus, vitamins A, E, C, B6, and folate was lower in the highest tertile of PRAL ($p < 0.05$).

Table 3 displays biochemical tests and blood pressure for tertiles of plant-based dietary indices and DAL in crude and adjusted models. Participants in the highest tertile of PDI had lower levels of fasting blood sugar (146.74 ± 6.16 vs. 152.87 ± 6.09, $p = 0.009$), and 2-h post-prandial glucose (181.76 ± 7.03 vs. 203.01 ± 6.94, $p = 0.002$). Also, patients in the highest tertile of hPDI had lower levels of fasting blood sugar (145.53 ± 6.16 vs. 167.89 ± 6.08, $p = 0.041$).

Table 4 shows the odds ratios and 95% confidence intervals for risk of metabolic disorders and CVD risk factors in crude and adjusted models for tertiles of DAL scores and PDI indices. Among participants in the top tertile of PDI, 85% decreased the risk of abdominal obesity; however, associations lost significance after adjusting for cofounding variables. No statistical association was observed between plant-based dietary indices or DAL and CVD risk factors.

Discussion

Plant-based diets contain adequate quantities of plant proteins, vitamins, minerals, plant sterols, polyphenols, flavonoids, and also contain low amounts of salt, fat, animal products, oils, processed foods, cholesterol, and sugar which help to prevent CVD and control CVD risk factors. In the present cross-sectional study, we examined how plant-based dietary indices and DAL were associated with cardiovascular risk factors. We found that FBS, 2hPPG, and HbA1C decreased across tertiles

Table 4 Odd ratios and confidence intervals for cardiovascular risk factors among tertiles of dietary acid load and plant-based dietary indices

Variables	PDI tertiles			P trend[a]	hPDI tertiles			P trend	uPDI tertiles			P trend	NEAP tertiles			P trend	PRAL tertiles			P trend
	1	2	3		1	2	3		1	2	3		1	2	3		1	2	3	
FBS ≥ 110																				
Crude model	1	0.88 (0.30, 2.57)	0.42 (0.16, 1.13)	0.074	1	0.41 (0.14, 1.15)	0.58 (0.20, 1.69)	0.364	1	1.76 (0.65, 4.77)	1.33 (0.53, 3.29)	0.528	1	0.90 (0.36, 2.28)	1.30 (0.48, 3.51)	0.605	1	1.55 (0.62, 3.89)	2.06 (0.77, 5.49)	0.139
Adjusted model	1	0.89 (0.29, 2.73)	0.32 (0.11, 0.92)	*0.030*	1	0.43 (0.14, 1.24)	0.55 (0.18, 1.71)	0.310	1	1.39 (0.48, 3.97)	0.82 (0.30, 2.23)	0.742	1	1.01 (0.38, 2.64)	1.20 (0.42, 3.39)	0.731	1	1.55 (0.57, 4.21)	1.98 (0.70, 5.58)	0.190
SBP ≥ 130																				
Crude model	1	0.55 (0.28, 1.06)	0.68 (0.35, 1.32)	0.244	1	0.94 (0.49, 1.81)	0.72 (0.37, 1.40)	0.338	1	1.01 (0.51, 1.95)	0.95 (0.49, 1.83)	0.891	1	1.30 (0.67, 2.52)	1.23 (0.63, 2.39)	0.542	1	0.87 (0.45, 1.69)	1.09 (0.56, 2.10)	0.784
Adjusted model	1	0.79 (0.38, 1.63)	1.01 (0.48, 2.08)	0.997	1	1.09 (0.52, 2.26)	0.84 (0.40, 1.78)	0.677	1	1.03 (0.48, 2.17)	0.80 (0.37, 1.73)	0.589	1	1.28 (0.61, 2.67)	0.92 (0.44, 1.91)	0.817	1	1.05 (0.49, 2.24)	0.97 (0.47, 2.02)	0.938
DBP ≥ 85																				
Crude model	1	0.85 (0.31, 2.33)	0.66 (0.22, 1.97)	0.465	1	1.14 (0.41, 3.13)	0.70 (0.23, 2.14)	0.555	1	2.25 (0.73, 6.94)	1.55 (048, 4.98)	0.507	1	0.98 (0.30, 3.20)	1.94 (0.68, 5.55)	0.191	1	0.54 (0.15, 1.92)	1.82 (0.67, 4.90)	0.191
Adjusted model	1	1.02 (0.33, 3.08)	0.75 (0.22, 2.49)	0.875	1	1.26 (0.43, 3.71)	0.85 (0.24, 2.94)	0.851	1	2.55 (0.73, 8.94)	0.92 (0.23, 3.62)	0.812	1	1.16 (0.32, 4.16)	1.67 (0.53; 5.24)	0.362	1	0.62 (0.15, 2.52)	1.61 (0.53, 4.84)	0.303
HDL < 50																				
Crude model	1	1.27 (0.61, 2.62)	0.78 (0.38, 1.57)	0.486	1	0.62 (0.30, 1.28)	0.73 (0.35, 1.52)	0.422	1	1.01 (0.49, 2.07)	1.02 (0.50, 2.06)	0.947	1	0.88 (0.43, 1.83)	0.78 (0.38, 1.59)	0.500	1	0.58 (0.28, 1.22)	0.62 (0.30, 1.30)	0.224
Adjusted model	1	1.20 (0.56, 2.55)	0.69 (0.32, 1.46)	0.829	1	0.58 (0.27, 1.23)	0.72 (0.33, 1.57)	0.406	1	0.97 (0.45, 2.07)	1.01 (0.47, 2.19)	0.968	1	0.92 (0.44, 1.95)	0.88 (0.24, 1.84)	0.741	1	0.49 (0.22, 1.08)	0.61 (0.28, 1.32)	0.239
LDL ≥ 130																				
Crude model	1	0.56 (0.18, 1.17)	1.32 (0.61, 2.88)	0.453	1	0.40 (0.16, 1.01)	0.89 (0.41, 1.93)	0.775	1	0.96 (0.41, 2.21)	0.92 (0.40, 2.09)	0.853	1	1.29 (0.56, 2.97)	1.18 (0.50, 2.76)	0.700	1	1.17 (0.51, 2.66)	0.98 (0.42, 2.28)	0.970
Adjusted model	1	0.48 (0.18, 1.27)	1.41 (0.61, 3.27)	0.389	1	0.37 (0.14, 0.96)	1.07 (0.46, 2.46)	0.974	1	0.94 (0.38, 2.27)	0.82 (0.33, 2.05)	0.683	1	1.27 (0.53, 3.03)	1.16 (0.48, 2.79)	0.741	1	1.03 (042, 2.49)	0.89 (0.36, 2.17)	0.800
TC ≥ 200																				
Crude model	1	0.74 (0.32, 1.70)	1.05 (0.47, 2.33)	0.908	1	0.91 (0.39, 2.10)	1.05 (0.47, 2.36)	0.895	1	0.97 (0.43, 2.14)	0.58 (0.25, 1.36)	0.224	1	1.06 (0.49, 2.29)	0.49 (0.20, 1.20)	0.135	1	0.90 (0.41, 1.99)	0.62 (0.26, 1.45)	0.280
Adjusted model	1	0.70 (0.29, 1.68)	1.07 (0.45, 2.52)	0.869	1	0.90 (0.38, 2.14)	1.40 (0.59, 3.35)	0.456	1	0.87 (0.37, 2.03)	0.40 (0.15, 1.04)	0.064	1	0.97 (0.43, 2.17)	0.43 (0.17, 1.08)	0.084	1	0.67 (0.29, 1.57)	0.46 (0.18, 1.15)	0.098
TG ≥ 150																				
Crude model	1	0.81 (0.43, 1.53)	0.92 (0.48, 1.74)	0.800	1	1.11 (0.58, 2.09)	0.85 (0.45, 1.61)	0.633	1	1.08 (0.56, 2.05)	1.03 (0.55, 1.93)	0.925	1	0.83 (0.44, 1.57)	0.97 (0.51, 1.83)	0.938	1	0.83 (0.44, 1.57)	0.97 (0.51, 1.83)	0.938
Adjusted model	1	0.69 (0.34, 1.39)	0.94 (0.46, 1.93)	0.876	1	1.25 (0.62, 2.48)	1.12 (0.55, 2.28)	0.733	1	1.01 (0.49, 2.06)	0.71 (0.34, 1.46)	0.360	1	0.73 (0.37, 1.47)	0.84 (0.42, 1.69)	0.636	1	0.62 (0.30, 1.26)	0.73 (0.36, 1.48)	0.401
WC ≥ 88																				
Crude model	1	0.62 (0.27, 1.40)	0.43 (0.19, 0.96)	*0.039*	1	1.00 (0.45, 2.22)	0.62 (0.29, 1.32)	0.209	1	1.69 (0.75, 3.80)	0.95 (0.46, 1.95)	0.868	1	1.26 (0.60, 2.64)	1.47 (0.69, 3.15)	0.313	1	0.72 (0.34, 1.49)	1.41 (0.63, 3.17)	0.415
Adjusted model	1	0.68 (0.22, 2.05)	0.63 (0.20, 1.92)	0.432	1	1.04 (0.34, 3.14)	0.49 (0.16, 1.48)	0.192	1	3.25 (1.05, 10.08)	1.98 (0.66, 5.90)	0.206	1	0.80 (0.28, 2.30)	1.53 (0.53, 4.38)	0.456	1	0.72 (0.26, 1.95)	2.60 (0.83, 8.17)	0.129
BMI ≥ 30																				

Table 4 (continued)

Crude model	1	0.58 (0.30, 1.12)	0.52 (0.26, 1.04)	0.060	1	1.12 (0.57, 2.18)	0.85 (043, 1.67)	0.649	1	0.51 (0.26, 1.03)	0.65 (0.33, 1.24)	0.195	1	1.81 (0.90, 3.62)	1.91 (0.95, 3.82)	0.070	1	1.32 (0.67, 2.62)	1.48 (0.75, 2.91)	0.256
Adjusted model	1	0.56 (0.28, 1.11)	0.54 (0.26, 1.10)	0.085	1	1.15 (0.58, 2.28)	0.96 (0.47, 1.95)	0.931	1	0.48 (0.24, 0.99)	0.56 (0.27, 1.14)	0.104	1	1.73 (0.85, 3.52)	1.83 (0.90, 3.74)	0.097	1	1.26 (0.62, 2.57)	1.37 (0.68, 2.76)	0.381

Significant *p*-values present as italic format

PDI plant-based diet index, *hPDI* healthy plant-based diet index, *uPDI* unhealthy plant-based diet index, *NEAP* net endogenous acid production, *PRAL* potential renal acid load, *FBS* fasting blood sugar, *SBP* systolic blood pressure, *DBP* diastolic blood pressure, *TG* triglyceride, *TC* total cholesterol, *HDL* high-density lipoprotein, *LDL* low-density lipoprotein, *WC* waist circumference, *BMI* body mass index

[a] Calculated using logistic regression

[b] Odd ratio (95% confidence interval)

Adjusted model: All variables were adjusted for age, BMI, socio-economic status, physical activity, energy, vitamin D, and supplement intake

of PDI in crude and adjusted models. Also, FBS decreased across tertiles of hPDI after adjustment for age, BMI, socio-economic status, physical activity, energy, vitamin D intake, and vitamin and mineral supplement consumption. There was a positive association between PDI and WC in the crude model; however, statistical significance disappeared after taking confounders into account. To our knowledge, no prior study has examined the association between PBDs and DAL in patients with type 2 diabetes. However, we previously reported an association between DAL in patients with diabetic nephropathy and CVD risk factors (suggesting that HbA1C was associated with DAL) [19]. Moreover, other research has shown lower socio-economic status to be associated with higher PBD indices and lower DAL [29, 30]. Individuals with lower income and education have limited ability to purchase fresh vegetables and fruits which can lead to lower consumption of food groups with low dietary acid loads [29, 30].

Haghighatdoost et al. showed a positive association between higher DAL and HbA1C [19]. Based on a previous case-control study of women with and without gestational diabetes mellitus (GDM), women with higher DAL scores had higher risk of GDM during pregnancy [31]. Although we did not find a statistically significant association between HbA1C and DAL in the present study, we found that HbA1C was lower in participants in the upper tertiles of PDI, which may be related to the benefits of higher intake of PDI foods such as vegetables, fruits, and whole grains. Inconsistent findings between our study and others may be due to the inability of a cross-sectional study to fully account for confounding. Future randomized trials are needed that can also take into account unknown confounders.

Among the plant-based dietary indices, PDI included whole grains, fruits, vegetables, vegetable oils, nut, legumes, tea, and coffee. The beneficial effects of these healthy components may play an important role in decreasing glycemic factors, immunologic markers, inflammatory factors, and in increasing antioxidant capacity [32]. Also, lower intake of animal food groups leads to lower DAL and usually occurs along with more PBDs, which can increase urinary PH and improve the metabolic profile [33]. Fatahi et al. showed that a 10-week-long dietary intervention that was rich in whole grains had beneficial effects on CVD risk factors [34]. Also, a randomized clinical trial in 40 women who were overweight or obese found that a whole grain diet improved blood pressure [35]. Also, a randomized trial of patients with type 2 diabetes showed that legumes that are rich in fiber and vegetable protein (as a part of low-glycemic-index diet) can improve glycemic control and HbA1c levels [36]. Vegetables, fruits, and whole grains are high in fibers that are associated with lower HbA1c [37]. A cross-sectional study by Pham et al., conducted with 1440 Japanese aged 18–69, showed that coffee consumption can be associated with decreasing insulin resistance [38]. However, the study did not observe a significant association between coffee consumption and HbA1c [38].

Coffee contains antioxidants and polyphenols that can decrease oxidative stress and subsequent insulin resistance [39, 40].

Haghighatdoost et al. reported an inverse association between PRAL and FBS [19]. Also, a higher PRAL was associated with higher TG and SBP [19]. A study in a large representative sample of 6788 Germans (aged 18–79 years), which used both food frequency questionnaires and biomarker-based estimates, revealed a significant relationship between PRAL and hypertension [41]. A cross-sectional study of 1136 young women revealed that TC and HDL increased following PRAL [18]. They also showed that BMI and WC were significantly correlated with protein potassium ratio [18]. Another study found that HDL, TC, BMI, and SBP decreased following adherence to a low-fat, whole food, plant-based diet [42]. However, we did not observe any significant associations between DAL and glycemic factors, lipid profile, or blood pressure. In line with our findings, the Rotterdam Study did not find a positive association between DAL and hypertension [43]. Also, a study of Japanese women found that DAL was not significantly associated with FBS or HbA1c [44]. Possible reasons for differing results between studies include different sample sizes, populations of different ages, a variety of methods to assess dietary intake, different scoring to assess DAL, different categorizations of DAL score, specific habits of various populations, and varied adjustments for confounders. Moreover, given that our study focused on diabetic patients, it is possible that results may be different among healthy people.

To the best of our knowledge, this was the first study to assess the association between DAL and PBDs with CVD risk factors among patients with type 2 diabetes; we considered two different dietary indices including DAL and plant-based diet indices. This is the first study on this topic conducted in Middle East that has assessed healthy and unhealthy food groups and animal-based foods in detail. However, it also has some limitations. One important limitation is that the cross-sectional study design does not support causal inference. Further, various unknown unmeasured confounders may also affect the results. Urine acid status was not measured to validate metabolic acidosis and DAL in the participants. Glycemic factors decreased across tertiles of PDI; however, we did not find CVD risk factors to be significantly associated with PBDs or DAL, which may have been due to the cross-sectional nature of the study. Future cohort studies in both genders are needed to more fully understand clinical outcomes related to DAL and PBDs in diabetic patients.

Acknowledgments We would like to express are gratitude to Dr. Nasli, secretary of the Diabetes Research Center of Tehran University of Medical Sciences, and all staffs in this center in Tehran, Iran.

Authors' contributions ED, BL, and LA designed and LA supervised the study. ED and FJ conducted the study. ED and LA performed the statistical analyses. ED prepared a first draft of the manuscript, and LA, JH, and PS finalized it.

Funding This study is supported by Tehran University of Medical Sciences (grant number: 9601-161-34260).

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval This study was approved by the ethical committee of the Tehran University of Medical Sciences. All participants provided informed written consent to participate in the study.

References

1. Borch-Johnsen K, Kreiner S. Proteinuria: value as predictor of cardiovascular mortality in insulin dependent diabetes mellitus. Br Med J (Clin Res Ed). 1987;294(6588):1651–4.
2. Arredondo A. Diabetes: a global challenge with high economic burden for public health systems and society. Am J Public Health. 2013;103(2):e1–2.
3. Global, regional, and national age-sex specific all-cause and cause-specific mortality for 240 causes of death, 1990–2013: a systematic analysis for the Global Burden of Disease Study 2013. Lancet (London, England). 2015;385(9963):117–71.
4. What causes the most deaths? http://www.healthdata.org/iran [updated 2017; cited 2018 30 November].
5. International Diabetes Federation (IDF). Country estimates table 2011. diabetes atlas2012 [cited 2018 30 November]. files/EN_6E_Atlas_Full_0.pdf; 6th ed:[Available from: Available from http://www.idf.org/sites/default/.
6. Frassetto LA, Todd KM, Morris RC Jr, Sebastian A. Estimation of net endogenous noncarbonic acid production in humans from diet potassium and protein contents. Am J Clin Nutr. 1998;68(3):576–83.
7. Huang M, Li J, Ha MA, Riccardi G, Liu S. A systematic review on the relations between pasta consumption and cardio-metabolic risk factors. Nutr Metab Cardiovasc Dis. 2017;27(11):939–48.
8. Fung TT, van Dam RM, Hankinson SE, Stampfer M, Willett WC, Hu FB. Low-carbohydrate diets and all-cause and cause-specific mortality: two cohort studies. Ann Intern Med. 2010;153(5):289–98.
9. Rinaldi S, Campbell EE, Fournier J, O'Connor C, Madill J. A comprehensive review of the literature supporting recommendations from the Canadian Diabetes Association for the use of a plant-based diet for management of type 2 diabetes. Can J Diabetes. 2016;40(5):471–7.
10. Pettersen BJ, Anousheh R, Fan J, Jaceldo-Siegl K, Fraser GE. Vegetarian diets and blood pressure among white subjects: results from the Adventist Health Study-2 (AHS-2). Public Health Nutr. 2012;15(10):1909–16.
11. Tonstad S, Butler T, Yan R, Fraser GE. Type of vegetarian diet, body weight, and prevalence of type 2 diabetes. Diabetes Care. 2009;32(5):791–6.
12. Tuso PJ, Ismail MH, Ha BP, Bartolotto C. Nutritional update for physicians: plant-based diets. Perm J. 2013;17(2):61–6.
13. Lee V, McKay T, Ardern CI. Awareness and perception of plant-based diets for the treatment and management of type 2 diabetes in a

community education clinic: a pilot study. J Nutr Metab. 2015;2015:236234.
14. Appleby PN, Thorogood M, Mann JI, Key TJ. Low body mass index in non-meat eaters: the possible roles of animal fat, dietary fibre and alcohol. Int J Obes Relat Metab Dis. 1998;22(5):454–60.
15. Souto G, Donapetry C, Calvino J, Adeva MM. Metabolic acidosis-induced insulin resistance and cardiovascular risk. Metab Syndr Relat Disord. 2011;9(4):247–53.
16. Remer T. Influence of nutrition on acid-base balance–metabolic aspects. Eur J Nutr. 2001;40(5):214–20.
17. Fagherazzi G, Vilier A, Bonnet F, Lajous M, Balkau B, Boutron-Rualt MC, et al. Dietary acid load and risk of type 2 diabetes: the E3N-EPIC cohort study. Diabetologia. 2014;57(2):313–20.
18. Zhang L, Curhan GC, Forman JP. Diet-dependent net acid load and risk of incident hypertension in United States women. Hypertension (Dallas, Tex : 1979). 2009;54(4):751–5.
19. Haghighatdoost F, Najafabadi MM, Bellissimo N, Azadbakht L. Association of dietary acid load with cardiovascular disease risk factors in patients with diabetic nephropathy. Nutrition (Burbank, Los Angeles County, Calif). 2015;31(5):697–702.
20. Monfort-Pires M, Folchetti LD, Previdelli AN, Siqueira-Catania A, de Barros CR, Ferreira SR. Healthy Eating Index is associated with certain markers of inflammation and insulin resistance but not with lipid profile in individuals at cardiometabolic risk. Appl Physiol Nutr Metab. 2014;39(4):497–502.
21. Ainsworth BE, Haskell WL, Whitt MC, Irwin ML, Swartz AM, Strath SJ, et al. Compendium of physical activities: an update of activity codes and MET intensities. Med Sci Sports Exerc. 2000;32(9 Suppl):S498–504.
22. Third Report of the National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, and Treatment of High Blood Cholesterol in Adults (Adult Treatment Panel III) final report. Circulation. 2002;106(25):3143–421.
23. Azadbakht L, Esmaillzadeh A. Red meat intake is associated with metabolic syndrome and the plasma C-reactive protein concentration in women. J Nutr. 2009;139(2):335–9.
24. Ghaffarpour M, Houshiar-Rad A, Kianfar H. The manual for household measures, cooking yields factors and edible portion of foods. Tehran: Nashre Olume Keshavarzy; 1999. p. 1–40.
25. Martinez-Gonzalez MA, Sanchez-Tainta A, Corella D, Salas-Salvado J, Ros E, Aros F, et al. A provegetarian food pattern and reduction in total mortality in the Prevencion con Dieta Mediterranea (PREDIMED) study. Am J Clin Nutr. 2014;100(Suppl 1):320S–8S.
26. Satija A, Bhupathiraju SN, Rimm EB, Spiegelman D, Chiuve SE, Borgi L, et al. Plant-based dietary patterns and incidence of type 2 diabetes in US men and women: results from three prospective cohort studies. PLoS Med. 2016;13(6):e1002039.
27. Remer T, Manz F. Estimation of the renal net acid excretion by adults consuming diets containing variable amounts of protein. Am J Clin Nutr. 1994;59(6):1356–61.
28. Remer T, Dimitriou T, Manz F. Dietary potential renal acid load and renal net acid excretion in healthy, free-living children and adolescents. Am J Clin Nutr. 2003;77(5):1255–60.
29. Kant AK, Graubard BI. Secular trends in the association of socioeconomic position with self-reported dietary attributes and biomarkers in the US population: National Health and Nutrition Examination Survey (NHANES) 1971-1975 to NHANES 1999-2002. Public Health Nutr. 2007;10(2):158–67.
30. Miller RR, Sales AE, Kopjar B, Fihn SD, Bryson CL. Adherence to heart-healthy behaviors in a sample of the U.S. population. Prev Chronic Dis. 2005;2(2):A18.
31. Saraf-Bank S, Tehrani H, Haghighatdoost F, Moosavian SP, Azadbakht L. The acidity of early pregnancy diet and risk of gestational diabetes mellitus. Clin Nutr (Edinburgh, Scotland). 2018;37(6 Pt A):2054–9.
32. Griffiths K, Aggarwal BB, Singh RB, Buttar HS, Wilson D, De Meester F. Food antioxidants and their anti-inflammatory properties: a potential role in cardiovascular diseases and cancer prevention. Diseases (Basel, Switzerland). 2016;4(3).
33. Berardi JM, Logan AC, Rao AV. Plant based dietary supplement increases urinary pH. J Int Soc Sports Nutr. 2008;5:20.
34. Fatahi S, Daneshzad E, Kord-Varkaneh H, Bellissimo N, Brett NR, Azadbakht L. Impact of diets rich in whole grains and fruits and vegetables on cardiovascular risk factors in overweight and obese women: a randomized clinical feeding trial. J Am Coll Nutr. 2018;37(7):568–77.
35. Kirwan JP, Malin SK, Scelsi AR, Kullman EL, Navaneethan SD, Pagadala MR, et al. A whole-grain diet reduces cardiovascular risk factors in overweight and obese adults: a randomized controlled trial. J Nutr. 2016;146(11):2244–51.
36. Jenkins DJ, Kendall CW, Augustin LS, Mitchell S, Sahye-Pudaruth S, Blanco Mejia S, et al. Effect of legumes as part of a low glycemic index diet on glycemic control and cardiovascular risk factors in type 2 diabetes mellitus: a randomized controlled trial. Arch Intern Med. 2012;172(21):1653–60.
37. AlEssa HB, Ley SH, Rosner B, Malik VS, Willett WC, Campos H, et al. High fiber and low starch intakes are associated with circulating intermediate biomarkers of type 2 diabetes among women. J Nutr. 2016;146(2):306–17.
38. Pham NM, Nanri A, Kochi T, Kuwahara K, Tsuruoka H, Kurotani K, et al. Coffee and green tea consumption is associated with insulin resistance in Japanese adults. Metab Clin Exp. 2014;63(3):400–8.
39. Lee AH, Tan L, Hiramatsu N, Ishisaka A, Alfonso H, Tanaka A, et al. Plasma concentrations of coffee polyphenols and plasma biomarkers of diabetes risk in healthy Japanese women. Nutr Diabetes. 2016;6:e212.
40. Henriksen EJ, Diamond-Stanic MK, Marchionne EM. Oxidative stress and the etiology of insulin resistance and type 2 diabetes. Free Radic Biol Med. 2011;51(5):993–9.
41. Krupp D, Esche J, Mensink GBM, Klenow S, Thamm M, Remer T. Dietary acid load and potassium intake associate with blood pressure and hypertension prevalence in a representative sample of the German adult population. Nutrients. 2018;19:10(1).
42. Kent LM, Grant RS, Watts G, Morton DP, Rankin PM, Ward EJ. HDL subfraction changes with a low-fat, plant-based complete health improvement program (CHIP). Asia Pac J Clin Nutr. 2018;27(5):1002–9.
43. Engberink MF, Bakker SJ, Brink EJ, van Baak MA, van Rooij FJ, Hofman A, et al. Dietary acid load and risk of hypertension: the Rotterdam study. Am J Clin Nutr. 2012;95(6):1438–44.
44. Murakami K, Sasaki S, Takahashi Y, Uenishi K. Association between dietary acid-base load and cardiometabolic risk factors in young Japanese women. Br J Nutr. 2008;100(3):642–51.

International Journal of Diabetes in Developing Countries (2021) 41:84–88
https://doi.org/10.1007/s13410-020-00858-9

ORIGINAL ARTICLE

Left ventricle diastolic dysfunction in a sample of prediabetic adults from Baghdad, Iraq

Methaq H. Alogaily[1] · Atheer J. Alsaffar[1] · Moayed B. Hamid[2]

Received: 25 November 2019 / Accepted: 4 August 2020 / Published online: 26 August 2020

Abstract

Background Prediabetes is a metabolic state in which people are not diabetics but also cannot be considered normal in terms of cutoff points of glycated hemoglobin or fasting plasma glucose. Those people are at higher risk of progression to type 2 diabetes mellitus and to develop many of the diabetes micro- and macro-vascular complications even before a diagnosis of diabetes has been established. One of the earliest cardiac manifestations of diabetic heart failure is left ventricle diastolic dysfunction.

Objectives To identify diastolic dysfunction as early complication of prediabetes among adults in Baghdad, Iraq, and to determine associated risk factors among those individuals.

Subjects and methods This cross-sectional study enrolled 38 adults (20–79 years) with prediabetes in the setting of primary healthcare centers in Baghdad, Iraq, compared with 38 adults (age and sex matched) without prediabetes. People with any condition that interfere with glycated hemoglobin level were excluded. Data were collected through direct interview. Anthropometric measurements and laboratory analysis were done to measure fasting plasma glucose, glycated hemoglobin, and lipid profile. Echocardiography was done to assess left ventricle diastolic function.

Results Prediabetes was significantly associated with left ventricle diastolic dysfunction (95% CI 0.23, 0.53) than those without prediabetes (95% CI 0.03, 0.21) ($p = 0.013$).

Conclusions Left ventricle diastolic dysfunction is an early manifestation of cardiac abnormalities associated with prediabetes. Those people share same factors with those having type 2 diabetes mellitus. More attention should be paid to identify those individuals correctly in order to intervene instantly.

Keywords Prediabetes · Left ventricle diastolic dysfunction · Iraq

Introduction

Type 2 diabetes mellitus (T2DM) is an independent well-known risk factor for heart failure development, and diastolic dysfunction represents one of the early manifestations of diabetic cardiomyopathy [1]. It is well established that metabolic abnormalities precede the onset of overt diabetes by years, and at the time of diagnosis, many patients with T2DM have already had organ damage or advanced subclinical atherosclerosis [2–4]. Prediabetes (preDM), linked to decrease insulin sensitivity or increased insulin resistance [5], is associated with higher risk of progression to T2DM and to micro- and macro-vascular complications even before a diagnosis of diabetes has been established [6]. Not all people with preDM will develop future T2DM; however, the majority do. It was estimated that up to 70% of individuals with preDM will eventually progress to T2DM with annual conversion rate about 5–10% [7]. Left ventricular diastolic dysfunction (LVDD) is usually the result of impaired LV relaxation and increased LV chamber stiffness which leads to increase in cardiac filling pressures [8], and at time symptoms appear, it can lead to overt heart failure syndrome despite normal or mildly impaired LV systolic function [9]. Transthoracic echocardiography (TTE) is the most single and effective method in the diagnosis of LVDD. It gives information (mechanical and

✉ Methaq H. Alogaily
Methaq.h@colmed-alnahrain.edu.iq

Atheer J. Alsaffar
ahrsaffar@yahoo.com

Moayed B. Hamid
Moayed.basheer@yahoo.com

[1] Department of Community and Family Medicine, College of Medicine, Al-Nahrain University, Baghdad, Iraq

[2] Department of Medicine, College of Medicine, Al-Nahrain University, Baghdad, Iraq

functional) on cardiac abnormalities contributing to LVDD [10]. Previous studies suggested that LVDD in diabetic patients is associated with the subsequent development of heart failure and increased mortality independent of hypertension, coronary disease, or other echocardiographic parameters [11]. Data indicated that early detection with timely intervention may prevent or delay the development of heart failure, which is a major cause of morbidity and mortality in diabetic patients [12, 13].

Perreault et al. studied the regression from preDM to normal glucose regulation whether by life style changes or with metformin, and they found that regression to normoglycemia, independent of the approach and even if transient, is significantly associated with risk reduction of future diabetes [14].

Iraq is categorized by the International Diabetes Federation (IDF) 2017 in the Middle East and North Africa Region (MENA), and due to a lack of data, sources, and information about the real situation, the prevalence of preDM was estimated to be 6–8% using extrapolated data from similar ethnicity countries, geography, language, and income level [15]. Few recent Iraqi studies found preDM prevalence double or tripple this number [16, 17], and this was consistent with other studies in neighborhood countries in the region [18, 19]. Up to our knowledge, no study to assess LVDD in Iraqi prediabetic people has been done yet.

The identification of those with preDM and its early sequelae is crucial to halt or reverse the rising epidemic of T2DM and will guide more specific and cost-effective control measures.

Subjects and methods

Baghdad is the capital of Iraq (5169 km^2) with about 8 million population. Tigris River divides the city into nearly equal halves: Karkh and Rusafa. Study samples were people attending Baghdad's primary healthcare (PHC) centers (for 1 year from June 2018 to June 2019) which are distributed evenly throughout the city. Multistage random sampling technique was used to select health sectors of both Karkh and Rusafa directorates than PHC centers from selected sectors yielding a final sample size of 76 adults aged 20–79 years with certain inclusion criteria (no history of diabetes, hypertension, previous cardiovascular event, anemia or hemoglobinopathies, malignant disease, drugs, or alcohol abuse). Fasting plasma glucose (FPG) and glycated hemoglobin (A1C) were used to divide the sample into two groups, 38 individuals with preDM who were matched (age and sex) to another 38 individuals without preDM. Those appeared to be in diabetic range or those with cardiac abnormalities (other than LVDD) detected by echocardiography (e.g., low ejection fraction, valvular abnormalities, wall motion abnormalities, etc.) were excluded from the study. Direct interview with each participant had been done, and requested information regarding demographic data (age, sex, residence, occupation, etc.), history of smoking, hypertension, cardiac disease, and other medical conditions were taken.

The weight was measured (to nearest 0.5 kg), in erect position without shoes and light clothing using an electronic scale. Height was measured by using a height tape measure with an approximation of ± 0.1 cm. BMI was calculated as body weight/height^2 (kg/m^2). WHO criteria were used to classify people into normal, overweight, and obese [20]. Body surface area was calculated using Mosteller formula [21].

Blood pressure was measured in a participant's arm using a mercury sphygmomanometer in a sitting position. Two blood pressure readings were taken at 5 min interval, and the mean value was calculated in millimeters of mercury (mmHg).

A venous blood sample was obtained from each participant after confirmation of 8 h overnight fasting, 1 ml collected in a vacuum collection K3 EDTA tube (mixed thoroughly) and 1 ml in a gel and clot activator glass tube, both stored in ice-cool box (2–8 °C) and analyzed by laboratory technician (within 4–5 h).

Complete blood count was measured using Celltac Es MEK-7300K. Siemens Dimension EXL 200 was used to measure serum FPG concentrations and the lipid profile. Venous blood sample used for A1C measurement was analyzed using the enzymatic method with Arkray ADAMS A1C HA-8180 V (Menarini).

PreDM was defined as not having, but having A1C between 5.7 and 6.4% or FPG between 100 and 125 mg/dl or both according to the ADA classification. Diabetes is considered when the FPG was 126 mg/dl or more and when the A1C was 6.5 or more or on antidiabetic drugs [5].

Lipid profile was measured and the LDL-c was calculated using the Friedewald formula [22].

Both groups had TTE examination done by an echocardiography specialist who was blind to the purpose of study using Philips IE33 ultrasound system. Assessment of diastolic and systolic function and all cardiac chambers measurements was done according to the criteria defined by the American Society of Echocardiography [23]. LVM was calculated according to cube formula using linear method and indexed to BSA.

Data analysis

Data were coded, entered, and analyzed using SPSS (Statistical Packages for Social Sciences) program, version 24. Descriptive data were expressed as means and standard deviations for continuous measurements and as frequencies and percentages for categorical measurements.

Paired t test was used to compare continuous data, and McNemar test was used for the association of paired categorical data.

Conditional logistic regression (Cox survival analysis) was used to predict LVDD in preDM and comparison groups with adjustment for other co-variants. Statistical significance was accepted for a 2-sided $p < 0.05$.

Results

The age of the participants was between 23 and 70 years with a mean of 51.8 ± 11.3 years. Forty-two (55.3%) were females. The mean FPG and A1C for preDM group were 113.6 ± 8.5 and 6.0 ± 0.23 versus 88.1 ± 7.6 and 5.1 ± 0.43 for the comparison group ($p < .0001$).

There were no statistically significant differences between two groups regarding blood pressure readings, hemoglobin, and hematocrit levels.

There were statistically significant difference in means of both groups regarding BMI ($P = 0.001$), total cholesterol ($p = 0.001$), TGs ($p = 0.017$), LDL-c ($p = 0.003$), and HDL-c ($p = 0.001$) (Table 1).

Individuals with preDM had no statistically significant difference in LVM (except for females $p = 0.03$) and LVMI; however, there was significant difference regarding relative wall thickness RWT ($p = 0.009$). No significant difference was found in EF% of both groups (Table 2).

PreDM was significantly associated with LVDD (95% CI 0.23, 0.53) than those without preDM (95% CI 0.03, 0.21) ($p = 0.013$) (Table 3) (Fig. 1).

Conditional logistic analysis using Cox analysis showed that preDM is an independent predictor of LVDD with an odd ratio of 65.3 (95% CI 2.6–1627.2) ($p = 0.01$). Other co-variants, e.g., BMI ($p = 0.75$), cholesterol ($p = 0.64$), TGs ($p = 0.80$), LDL-c ($p = 0.67$), and HDL-c ($p = 0.63$), did not improve over all model fit.

Table 1 Clinical and laboratory measurements in prediabetes and comparison group*

	Prediabetes	Non-prediabetes	Significance
BPS	121.1 ± 11.7	121.5 ± 11.2	$p = 0.87$
BPD	76.3 ± 7.0	75.9 ± 8.01	$p = 0.76$
BMI	29.8 ± 3.6	26.6 ± 4.3	$p = 0.001$
Hb%	13.6 ± 1.0	13.8 ± 1.3	$p = 0.56$
Hct	41.4 ± 4.0	41.4 ± 4.3	$p = 0.95$
FBG	113.6 ± 8.3	88.1 ± 7.6	$p < 0.0001$
A1C	6.0 ± 0.23	5.1 ± 0.43	$p < 0.0001$
TC	224.5 ± 42.1	193.5 ± 40.3	$p = 0.001$
TGS	175.4 ± 73.3	138.5 ± 38.8	$p = 0.017$
LDL	138.5 ± 47.4	109.9 ± 42.5	$p = 0.003$
HDL	51.0 ± 6.7	55.8 ± 6.5	$p = 0.001$

*Paired t test. Values are expressed as mean ± SD

Table 2 Echocardiographic measurements in prediabetes and comparison group*

	Prediabetes	Non-prediabetes	p value
EF%	68.9 ± 5.5	69.4 ± 4.8	0.59
LV mass			
Males	184.6 ± 66.8	166.7 ± 45.3	0.34
Females	162.9 ± 47.2	131.3 ± 44.1	0.03
LV mass index			
Males	91.6 ± 32.6	88.5 ± 22.5	0.71
Females	86.5 ± 22.4	75.1 ± 21.9	0.13
RWT	0.38 ± 0.09	0.34 ± 0.06	0.009

*Paired t test. Values are expressed as mean ± SD

Discussion

The core of this study was to exclude any condition associated with LVDD as much as possible, starting from participant's selection criteria, age and sex matching of cases to the comparison group, and finally through analysis. In addition, five participants (two preDM and three none) were found to have cardiac abnormalities (as detected by echocardiography) other than LVDD and thus were excluded from the study. All these measures were to improve the strength of association between preDM and LVDD. Studies have shown that LVDD is one of the earliest signs of myocardial involvement in T2DM [24]; however, the exact prevalence is not known. Uday et al. studied the prevalence of LVDD in patients with T2DM in the absence of coronary artery disease or hypertension and found the prevalence of LVDD of 43% [25]. Recently, it was found that changes in LVDD precede the onset of diabetes [26]. In this study, preDM was independently associated with LVDD, and this could be associated with the persistent exposure to hyperglycemia. However, it has been suggested that subtle

Table 3 Diastolic dysfunction in prediabetes and comparison group*

		Non-preDM	
		LVDD	No
PreDM	LVDD	11 (28.9%)	14 (36.8%)
	No	3 (7.9%)	10 (26.3%)

*McNemar test ($p = 0.013$)

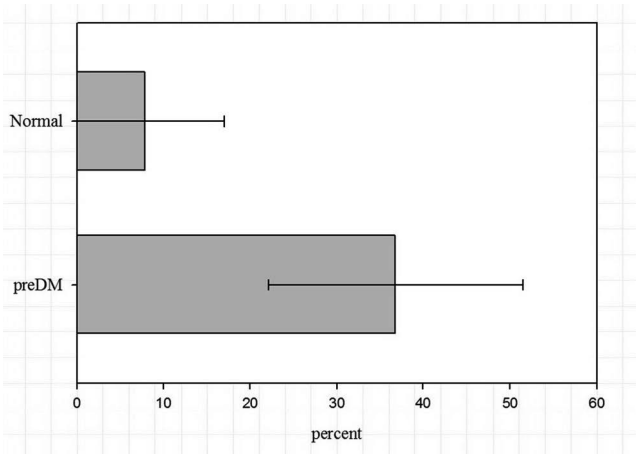


Fig. 1 Percent of individuals with and without preDM having LVDD

cardiac involvement may not be merely due to hyperglycemia, but also it could be associated with the state of insulin resistance [27], which is the main component of preDM. Participants with preDM were more dyslipidemic in all lipid components having higher weight compared with comparison group. This is in agreement with the hypothesized independent association between LVDD, obesity, and dyslipidemia [28]. Despite this, adjusting for these co-variants resulted in statistically insignificant difference regarding the association between preDM and LVDD.

Finding the mean age of 51.8 ± 11.3 years is consistent with the fact that preDM and diabetes in the Middle East and Arab countries dominated in those younger than 60 years old [29]. This age group is the most productive group in the community of these countries, and hence, targeting by this state will result in catastrophic disability both in man power as well as health expenditure.

The finding of insignificant association in LVMI between the two groups is consistent with the finding of Fuentes et al. [30]. Since the participants were normotensives and sex and age matched, this finding further supports the independence of preDM and LVDD association.

The study had points of strength and limitations; it throws light on the rising global interest in preDM state especially in the contest of extreme scarcity of studies in this part of the world. Besides, it focused on the most important, preventable, and earliest risk factor for developing diabetic cardiomyopathy. Most of the included participants were apparently healthy (e.g., mothers accompany children for immunization, relatives of patients, some adults working there, people coming to complete paperwork, etc.), and thus, the sample was almost general population based. However, of the limitations, this cross-sectional study precludes the link to cause effect type of association and the time course between preDM and LVDD (if causative). Furthermore, a larger sample size (based on assumption that prevalence of preDM in Iraq is much higher that estimated by IDF) is needed to confirm these findings. The collection of past medical conditions and conditions that interfere with A1C measurement or echocardiographic parameters were relied on history taken from the participants and not confirmed by laboratory tests. In addition, FPG or A1C was used to identify people with preDM, rather than oral glucose tolerance test to be in line with WHO recommendations.

Conclusions

We demonstrated that preDM is an independent predictor of LVDD which is the earliest preclinical cardiac abnormality leading to diabetic cardiomyopathy. Early identification of individuals with preDM is crucial. This group of people requires extensive evaluation including echocardiographic assessment once identified.

Acknowledgments The authors would like to thank in advance Iraqi Ministry of Health and Al-karkh and Al-Rusafa health directorates with all PHC centers staff for their cooperation.

Author contributions All authors contributed to the study conception and design. Material preparation, data collection, and analysis were performed by Methaq H. Alogaily, Atheer J. Alsaffar, and Moayed B. Hamid. The first draft of the manuscript was written by Methaq H. Alogaily, and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical considerations The study was approved by the institutional review board of the College of Medicine, Al-Nahrain University, and a request to facilitate the task of researcher was given. Participants directly interviewed were told about the research purpose and consent.

References

1. Di Pino A, Mangiafico S, Urbano F, et al. HbA1c identifies subjects with prediabetes and subclinical left ventricular diastolic dysfunction. J Clin Endocrinol Metab. 2017;102(10):3756–64. https://doi.org/10.1210/jc.2017-00954.
2. Kohner EM, Aldington SJ, Stratton IM, et al. United Kingdom prospective diabetes study, 30: diabetic retinopathy at diagnosis of non–insulin-dependent diabetes mellitus and associated risk factors. JAMA Ophthalmol. 1998;116(3):297–303. https://doi.org/10.1001/archopht.116.3.297.
3. Koopman RJ, Mainous AG 3rd, Liszka HA, et al. Evidence of nephropathy and peripheral neuropathy in US adults with undiagnosed diabetes. Ann Fam Med. 2006;4(5):427–32. https://doi.org/10.1370/afm.577.
4. Son J-W, Jang E-H, Kim M-K, et al. Diabetic retinopathy is associated with subclinical atherosclerosis in newly diagnosed type 2 diabetes mellitus. Diabetes Res Clin Pract. 2011;91(2):253–9. https://doi.org/10.1016/j.diabres.2010.11.005.

5. American Diabetes Association. 2. Classification and diagnosis of diabetes: standards of medical care in diabetes—2018. Diabetes Care. 2018;41(Suppl. 1):S13–27. https://doi.org/10.2337/dc18-S002.
6. Tabak AG, Herder C, Rathmann W, et al. Prediabetes: a high-risk state for diabetes development. Lancet. 2012;379:2279–90. https://doi.org/10.1016/S0140-6736(12)60283-9.
7. Gerstein HC, Santaguida P, Raina P, et al. Annual incidence and relative risk of diabetes in people with various categories of dysglycemia: a systematic overview and meta-analysis of prospective studies. Diab Res Clin Pract. 2007;78(3):305–12. https://doi.org/10.1016/j.diabres.2007.05.004.
8. Gardin JM, Arnold AM, Bild DE, et al. Left ventricular diastolic filling in the elderly: the cardiovascular health study. Am J Cardiol. 1998;82:345–51. https://doi.org/10.1016/S0002-9149(98)00339-7.
9. Aurigemma GP, Gottdiener JS, Shemanski L, et al. Predictive value of systolic and diastolic function for incident congestive heart failure in the elderly: the cardiovascular health study. J Am Coll Cardiol. 2001;37:1042–8. https://doi.org/10.1016/S0735-1097(01)01110-X.
10. Park K-T, Kim H-L, Oh S, et al. Association between reduced arterial stiffness and preserved diastolic function of the left ventricle in Korean middle-aged and elderly patients. J Clin Hypertens. 2017;19:620–6. https://doi.org/10.1111/jch.12968.
11. Nagueh SF, Smiseth OA, Appleton CP, et al. Recommendations for the evaluation of left ventricular diastolic function by echocardiography: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. J Am Soc Echocardiogr. 2016;29(4):277–314. https://doi.org/10.1016/j.echo.2016.01.011.
12. From AM, Scott CG, Chen HH. The development of heart failure in patients with diabetes mellitus and pre-clinical diastolic dysfunction a population-based study [published correction appears in J Am Coll Cardiol. 2010 Nov 2; 56(19):1612]. J Am Coll Cardiol. 2010;55(4):300–5. https://doi.org/10.1016/j.jacc.2009.12.003.
13. Fang ZY, Schull-Meade R, Downey M, et al. Diabetologia. 2005;48:394. https://doi.org/10.1007/s00125-004-1632-z.
14. Perreault L, Pan Q, Mather KJ, et al. Effect of regression from prediabetes to normal glucose regulation on long-term reduction in diabetes risk: results from the diabetes prevention program outcomes study. Lancet. 2012;379(9833):2243–51. https://doi.org/10.1016/S0140-6736(12)60525-X.
15. International Diabetes Federation. IDF Diabetes Atlas. 8th ed. Brussels: International Diabetes Federation; 2017. http://www.diabetesatlas.org
16. Al Azzawi O. Prevalence of prediabetes and metabolic syndrome and their association in an Iraqi sample. IOSR J Dent Med Sci. 2015;14:10–6. https://doi.org/10.9790/0853-14951016.
17. Mansour A, Al-Maliky A, Kasem B, Jabar A, Mosbeh K. Prevalence of diagnosed and undiagnosed diabetes mellitus in adults aged 19 years and older in Basrah, Iraq. Diab Met Syndr Obes: Targets Ther. 2014;7:139–44. https://doi.org/10.2147/DMSO.S59652.
18. Satman I, Omer B, Tutuncu Y, et al. Twelve-year trends in the prevalence and risk factors of diabetes and prediabetes in Turkish adults. Eur J Epidemiol. 2013;28(2):169–80. https://doi.org/10.1007/s10654-013-9771-5.
19. Zhang FF, Al Hooti S, Al Zenki S, et al. Vitamin D deficiency is associated with high prevalence of diabetes in Kuwaiti adults: results from a national survey. BMC Public Health. 2016;16:100. https://doi.org/10.1186/s12889-016-2758-x.
20. World Health Organization (WHO). Obesity and overweight factsheet from the WHO. 2018 Available from: https://www.who.int/news-room/fact-sheets/detail/obesity-and-overweight.
21. Mosteller RD. Simplified calculation of body-surface area. N Engl J Med. 1987;317:1098. https://doi.org/10.1056/NEJM198710223171717.
22. Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin Chem. 1972;18(6):499.
23. Lang RM, Badano LP, Mor-Avi V, et al. Recommendations for cardiac chamber quantification by echocardiography in adults: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. J Am Soc Echocardiogr. 2015;28(1):1–39.e14. https://doi.org/10.1016/j.echo.2014.10.003.
24. Zabalgoitia M, Ismaeil MF, Anderson L, Maklady FA. Prevalence of diastolic dysfunction in normotensive, asymptomatic patients with well-controlled type 2 diabetes mellitus. Am J Cardiol. 2001;87(3):320–3. https://doi.org/10.1016/S0002-9149(00)01366-7.
25. Uday G, Jayakumar S, Jnaneshwari M, et al. Evaluation of left ventricular diastolic dysfunction in type II diabetes mellitus - the role of valsalva maneuver. J Evol Med Dent Sci. 2014;3(11):2898–906. https://doi.org/10.14260/jemds/2014/2222.
26. Stahrenberg R, Edelmann F, Mende M, et al. Association of glucose metabolism with diastolic function along the diabetic continuum [published correction appears in Diabetologia. 2011 Apr;54(4):990. Schönbrunn, L [added]]. Diabetologia. 2010;53(7):1331–40. https://doi.org/10.1007/s00125-010-1718-8.
27. Fontes-Carvalho R, Ladeiras-Lopes R, Bettencourt P, Leite-Moreira A, Azevedo A. Diastolic dysfunction in the diabetic continuum: association with insulin resistance, metabolic syndrome and type 2 diabetes. Cardiovasc Diabetol. 2015;14(1):4. https://doi.org/10.1186/s12933-014-0168-x.
28. Sliem H, Nasr G. Left ventricular structure and function in prediabetic adults: relationship with insulin resistance. J Cardiovasc Dis Res. 2011;2(1):23–8. https://doi.org/10.4103/0975-3583.78583.
29. Mansour A, Douri F. Diabetes in Iraq: facing the epidemic. A systematic review. Wulfenia. 2015;22:258.
30. Fuentes L, de las Brown AL, Mathews SJ, et al. Metabolic syndrome is associated with abnormal left ventricular diastolic function independent of left ventricular mass. Eur Heart J. 2007;28(5):553–9. https://doi.org/10.1093/eurheartj/ehl526.

International Journal of Diabetes in Developing Countries (2021) 41:89–93
https://doi.org/10.1007/s13410-020-00852-1

ORIGINAL ARTICLE

Evaluation of platelet volume indices as predictive biomarkers of microvascular complications in patients with type 2 diabetes

Rajeev Chawla[1] · Jyotshnarani Sahu[1] · Hitesh Punyani[1] · Shalini Jaggi[2]

Received: 20 March 2020 / Accepted: 27 July 2020 / Published online: 14 August 2020

Abstract

Introduction Patients with type 2 diabetes are known to have a higher risk of developing micro- and macrovascular complications which enhance the risk of morbidity and mortality. Increased platelet activity is postulated to be a vital mechanism in the development of vascular complications. Platelet volume indices (PVI), including mean platelet volume (MPV), platelet distribution width (PDW), and plateletcrit (PCT) are indicators of enhanced platelet activity and these may be useful markers of complications.

Materials and methods We conducted a case-control study across 130 T2DM patients that were matched for age and gender with 130 people who did not have T2DM. Detailed demographic profile and clinical history including duration of diabetes along with complete physical examination was conducted. Fundus examination was conducted for direct ophthalmoscopy for retinopathy and Neuropathy Disability Score (NDS) was calculated to assess the severity of neuropathy. The biochemical investigations included were the platelet indices, fasting blood glucose (FPG), postprandial blood glucose, HbA1c, serum creatinine, and urine albumin.

Results We observed that platelet indices, such as MPV, PDW, and PCT were significantly higher in diabetic individuals than in matched controls. Higher MPV, PDW, and PCT were observed in diabetic patients with microvascular complications compared with those without microvascular complications. We observed that patients with neuropathy with NDS > 6 had higher value of MPV, PDW, and PCT than compared with those with NDS < 6. There was significant difference for MPV between diabetics with and without complications and non-diabetics ($p < 0.05$). PDW and MPV were positively correlated with duration of diabetes.

Conclusions The association between MPV, PDW, and PCT and diabetic retinopathy revealed that platelet indices were significantly higher than control group in both NPDR and PDR. However, we did not observe any difference in platelet indices in NPDR and patients without diabetic retinopathy. MPV, PDW, and PCT were higher in individuals with higher HbA1clevel and the correlation was found to be statistically significant.

Keywords Diabetic nephropathy · Diabetic neuropathy · Diabetic retinopathy · Type 2 DM · Microvascular complications · Platelet dysfunction · Platelet indices

Introduction

Diabetes mellitus is one of the most challenging clinical problems [1]. The prevalence of diabetes in Indian adult population is estimated to be 8.8% [2]. T2DM is a heterogeneous metabolic disorder characterized by chronic hyperglycemia, resulting from defects in insulin secretion, insulin action, or both. There are associated metabolic abnormalities that involve long-term micro- and macrovascular complications involving blood vessel, eyes, kidney, and nerves [3].

Long-term microvascular complications of diabetes include retinopathy with potential loss of vision; nephropathy leading to renal failure; peripheral neuropathy with risk of foot ulcers, amputations, and Charcot joints; and autonomic neuropathy causing gastrointestinal, genitourinary, and cardiovascular symptoms and sexual dysfunction [4].

T2DM is a now well documented to be prothrombotic state owing to sustained hyperglycemia, dyslipidemia, and insulin

✉ Rajeev Chawla
rajeevaastikchawla@yahoo.com

[1] Maharaja Agrasen Hospital, Punjabi Bagh, New Delhi, India

[2] Dr.Mohans' Diabetes Specialities Centre, Kirti Nagar, New Delhi, India

resistance causing endothelial and pericyte injury [5]. Hyperglycemia may represent a causal factor for in vivo platelet activation and may be responsible for non-enzymatic glycation of platelet glycoprotein causing changes in their structure and conformation as well as alteration in membrane lipid dynamics [6]. The increased platelet activity is believed to play a vital role in the development of vascular complications of this metabolic disease [7].

Platelets in diabetics are active and have increased aggregation because of dysregulated signaling pathway. This contributes to thrombus formation and microcapillary embolization. The release of constrictive, oxidative, and mitogenic substances such as platelet-derived growth factor accelerate the progression of local neovascularization of lens in diabetic Retinopathy. Microvascular complications have been described as factors that are predictive of cardiovascular and cerebrovascular morbidity and mortality among diabetic subjects [8] Thus, microvascular complications were pre-identified to be evaluated in our study.

Platelet volume indices such as mean platelet volume (MPV), platelet distribution width (PDW), plateletcrit (PCT), and platelet counts are indicators of increased platelet activity and can be considered as potential biomarkers for diabetic complications.

The newer hematological analyzers provide variety of platelet parameters which help in easy detection of change in platelet structure, and in early detection of prothrombotic state of platelets. These can act as an alarm for diagnosing and monitoring progression of diabetic complications.

Objectives

We evaluated the platelet indices in T2DM and compared them with non-diabetic individuals and then correlated platelet indices with microvascular complications. We also explored if platelet indices can predict the occurrence of microvascular complications in T2DM.

Materials and methods

The patient population included 130 T2DM patients who were newly diagnosed as per WHO criteria or individuals already on standard care of approach. Age- and sex-matched 130 non-diabetics individuals were taken as controls. The patients were excluded if they were female with hemoglobin < 10 g% and male patients with Hb < 12 g%, had history of blood transfusion in last 14 days, malignancy, and thrombocytopenia, were pregnant women, and if the patients were on antiplatelet drug such as aspirin and clopidogrel.

We conducted the investigations that included glycated hemoglobin, which was estimated by high-performance liquid chromatography. Platelet volume indices included MPV, PDW, and PCT. The FPG and PPG were investigated, twice, first after 8 h of fasting, and second 2 h after meals using semi-automated analyzer of the calorimetric method. Serum creatinine and urine albumin were evaluated. Tests for microvascular complications were for the evaluation of neuropathy which was evaluated by testing light touch using a 10 g monofilament, pain sensation using pinprick and vibration sense using a tuning fork of 128 Hz. The neuropathy disability score (NDS) was calculated to assess the severity of neuropathy. NDS score > 6 was diagnostic of peripheral neuropathy.

A detail history for duration of diabetes and other comorbid conditions was recorded. Neuropathy Disability Score (NDS) included the vibration perception threshold, temperature perception on dorsum of the foot, and pin prick evaluation. Score of 1 was considered abnormal and achilles reflex was also done.

Statistical analysis

Statistical analysis was performed using the IBMSPSS version 21. Data were expressed as mean ± standard deviation to characterize the study sample. For qualitative data, chi-square or Fischer's exact test was used to observe difference between proportions for independent groups. Pearson correlation coefficient was calculated between the outcome and quantitative independent demographic and clinical factors. p value < 0.05 was considered statistically significant (Fig. 1).

Results

Normal range of platelet volume indices were MPV (8.6–15.5 fl) PDW (9.0–14 fl), PCT (0.22–0.24%). The baseline characteristics of studied population are presented in Table 1.

Platelet volume indices in diabetes were evaluated. The mean of MPV, PDW, and PCT of cases was (mean ± SD) 11.34 ± 1.21 fl, 14.86 ± 0.94 fl, 0.24 ± 0% and in control these values were 8.35 ± 0.36 fl, 11.49 ± 0.59 fl, and 0.22 ± 0.01% respectively. MPV, PDW, and PCT were significantly higher in individuals with T2DM as compared with non-diabetics (Table 2).

Platelet volume indices and microvascular complications

Out of total 130 individuals with diabetes, 78 had microvascular complications including diabetic neuropathy, diabetic nephropathy, and diabetic retinopathy. The distribution of

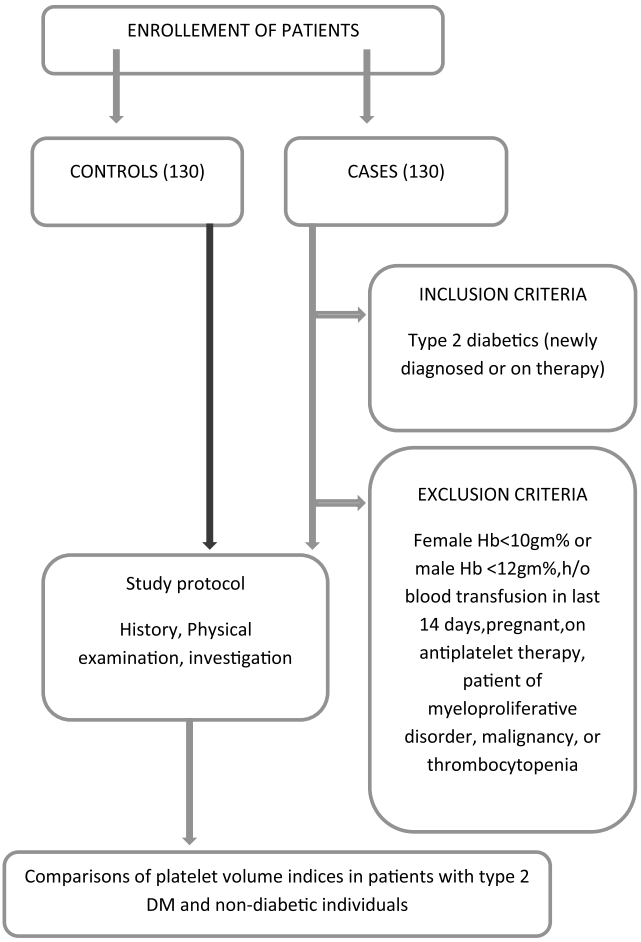


Fig. 1 Flow chart of the study protocol

Table 1 Baseline characteristics of study population

	Case	Control	*p* value
Sample size	130	130	
Age			0.947
Mean ± SD	56.82 ± 7.21	56.87 ± 7.19	
Median	58	58	
Min–max	36–74	36–74	
FBS			< 0.0001
Mean ± SD	148 ± 43.39	91.28 ± 8.51	
Median	142	90	
Min–max	84–334	77–143	
PPBS			< 0.0001
Mean ± SD	206.06 ± 59.43	141.91 ± 9.12	
Median	185.5	141	
Min–max	136–402	112–162	
HbA1c			< 0.0001
Mean ± SD	8.83 ± 2.22	4.89 ± 0.37	
Median	8.15	4.9	
Min–max	5.7–15.9	3.8–5.7	
Serum creatinine			< 0.0001
Mean ± stdev	1.25 ± 0.71	0.58 ± 0.14	
Median	1.02	0.6	
Min–max	0.35–3.8	0.3–0.9	
TPC			< 0.0001
Mean ± SD	2.58 ± 0.59	2.58 ± 0.59	
Median	2.62	2.62	
Min–max	1.55–4.28	1.55–4.28	
MPV			< 0.0001
Mean ± SD	11.34 ± 1.21	8.35 ± 0.36	
Median	11	8.4	
Min–max	9.1–13.7	7.5–8.8	
PDW			< 0.0001
Mean ± SD	14.86 ± 0.94	11.49 ± 0.59	
Median	14.8	11.4	
Min–max	12.65–16.8	10.5–12.8	
PCT			< 0.0001
Mean ± SD	0.24 ± 0.01	0.22 ± 0.01	
Median	0.24	0.22	
Min–max	0.21–0.26	0.18–0.3	

the patients with complications was as follows: 30 patients had diabetic neuropathy, 28 patients had retinopathy, and 28 had nephropathy. Neuropathy patients were divided into 2 subgroups depending on NDS Score (> 6 or < 6). Retinopathy patients were divided as NPDR and PDR.

We observed high value of mean of MPV, PDW, and PCT in diabetic patients with microvascular complications compared with those without microvascular complications (Table 3).

In a study conducted in patients in India [9], it was observed that platelet indices, namely, MPV, PDW, and PCT were significantly higher in diabetic individuals than those with age- and gender-matched controls. Moreover, the increase in MPV, PDW, and PCT were significant in diabetic subjects with microvascular complications when compared with those without microvascular complications. Platelet dysfunction also showed a positive association with HbA1C, retinopathy, nephropathy, and neuropathy individually.

We found that the mean of platelet volume indices was significantly higher in diabetic neuropathy patients with NDS score more than 6. MPV, PDW, and PCT were 12.92 ± 0.52 fl, 16.27 ± 0.39 fl, and 0.25 ± 0% (mean SD) in NDS > 6 and 11.3 ± 0.8 fl, 15.29 ± 0.56 fl, 0.24 ± 0% respectively in NDS < 6. The correlation was statistically significant ($p = <$ 0.001, 0.001, 0.003) correlating with the severity of neuropathy (Table 4).

We observed statistically significant correlation of MPV, PDW, and PCT with diabetic retinopathy and nephropathy. This study showed that mean of MPV, PDW, and PCT was higher in diabetic retinopathy patients with proliferative retinopathy (PDR) than non-proliferative retinopathy (NPDR)

Table 2 Comparison of platelet indices between two study groups

	Case	Control	*p* value
MPV			< 0.0001
Mean ± SD	11.34 ± 1.21	8.35 ± 0.36	
Median	11	8.4	
Min–max	9.1–13.7	7.5–8.8	
PDW			< 0.0001
Sample size	130	130	
Mean ± SD	14.86 ± 0.94	11.49 ± 0.59	
Median	14.8	11.4	
Min–max	12.65–16.8	10.5–12.8	
PCT			< 0.0001
Mean ± SD	0.24 ± 0.01	0.22 ± 0.01	
Median	0.24	0.22	
Min–max	0.21–0.26	0.18–0.3	

and this positive correlation was statistically significant. There was insignificant difference between the mean of platelet indices of NPDR and diabetic patient without retinopathy (Table 5). Similarly, higher values were noted in research from Turkey and India by Dinder et al. [10]; Kodiatte et al. [11] with MPV associated with retinal neovascularization of diabetic retinopathy (Table 6).

We noted that slightly higher MPV, PDW, and PCT were found in patient with albuminuria as compared with patient without albuminuria. But the relationship between platelet volume indices and nephropathy could not be established as there we could not do the repeated measurement of urine albumin to confirm a patient with nephropathy.

Table 3 Comparisons of platelet indices of cases with and without microvascular complications

	Microvascular complications		
Platelet indices	No	Yes	*p* value
MPV			< 0.0001
Sample size	52	78	
Mean ± SD	10.63 ± 0.72	11.82 ± 1.24	
Median	10.55	11.9	
Min–max	9.5–12.8	9.1–13.7	
PDW			< 0.0001
Sample size	52	78	
Mean ± SD	14.18 ± 0.63	15.31 ± 0.83	
Median	14.05	15.2	
Min–max	12.8–15.8	12.65–16.8	
PCT			< 0.0001
Sample size	52	78	
Mean ± SD	0.23 ± 0	0.24 ± 0.01	
Median	0.24	0.24	
Min–max	0.21–0.24	0.23–0.26	

Table 4 Comparisons of platelet indices with Neuropathy Disability Score (NDS)

	NDS		
Platelet indices	< 6	> 6	*p* value
Sample size	12	18	< 0.0001
MPV			
Mean ± SD	11.3 ± 0.8	12.92 ± 0.52	
Median	11	12.9	
Min–max	10–12.6	11.4–13.7	
PDW			< 0.0001
Mean ± SD	15.29 ± 0.56	16.27 ± 0.39	
Median	15.3	16.2	
Min–max	14.6–16.8	15.8–16.8	
PCT			*p* = 0.0003
Mean ± SD	0.24 ± 0	0.25 ± 0	
Median	0.24	0.25	
Min–max	0.23–0.25	0.24–0.26	

Platelet volume indices and HbA1c

Platelet parameters MPV, PDW, and PCT were found to be higher among DM with HbA1C > 7% as compared with DM with HbA1C < 7% and this was found to be statistically significant.

These results were quite similar to a study by Pujani et al. [12] in which there were higher values of platelet parameters including PC, MPV, PDW, PLCR, and PCT among DM with

Table 5 Comparison of platelet indices in diabetic retinopathy

	Fundus		*p* value
	NPDR	PDR	
Sample size	12	16	
MPV			*p* = 0.0003
Mean ± SD	10.76 ± 1.03	12.61 ± 0.91	
Median	10.8	12.65	
Min–max	9.2–12.9	10.4–13.6	
PDW			< 0.0001
Mean ± SD	14.58 ± 0.4	15.77 ± 0.49	
Median	14.7	15.8	
Min–max	13.6–15	14.86–16.8	
PCT			*p* = 0.002
Mean ± SD	0.24 ± 0	0.24 ± 0.01	
Median	0.24	0.25	
Min–max	0.23–0.24	0.23–0.25	

Table 6 Comparisons of platelet indices with HBA1c

	HbA1c		*p* value
	(1) < 7	(2) ≥ 7	
Sample size	58	72	< 0.0001
MPV			
Mean ± SD	10.54 ± 0.62	11.99 ± 1.17	
Median	10.5	12.45	
Min–max	9.1–12.1	9.2–13.7	
PDW			< 0.0001
Sample size	58	72	
Mean ± SD	14.32 ± 0.66	15.28 ± 0.91	
Median	14.2	15.4	
Min–max	13.2–16.6	12.65–16.8	
PCT			< 0.0001
Sample size	58	72	
Mean ± SD	0.24 ± 0.01	0.24 ± 0.01	
Median	0.24	0.24	
Min–max	0.21–0.25	0.23–0.26	

complication as compared with DM without complication, which was statistically significant. Among the platelet parameters, MPV, PCT, and PDW were found to be higher among DM with HbA1C > 7% as compared with DM with HbA1C < 7%.

Conclusions

There was a statistically significant difference for MPV between diabetics with and without complications and non-diabetics ($p < 0.05$). PDW and MPV were positively correlated with duration of diabetes. Duration of diabetes was significantly higher in diabetics with retinopathy ($p < 0.05$) and neuropathy ($p < 0.05$). MPV and PDW can be the predictive biomarkers of diabetic microvascular complications. A study conducted by Kshirsagar et al. [13] has demonstrated that MPV was significantly higher in diabetics than non-diabetic controls ($p < 0.05$). HbA1c ($p < 0.05$) and duration of diabetes ($p < 0.05$) were statistically significantly higher in diabetics with microvascular complications as compared with diabetics without microvascular complications. The study is limited for the small sample size, thereby limiting the wide confidence interval.

In our cross-sectional comparative study, we consistently demonstrated significant positive correlation between platelet volume indices (MPV, PDW, PCT) in diabetic patients with microvascular complications. Platelet indices had significant correlation with HbA1c. There was significant difference in platelet indices in patients with T2DM, especially with microvascular complications, when compared with non-diabetic individuals. Since the platelet indices analysis is simple, convenient, and cost-effective diagnostic tool to determine platelet dysfunction, these can be a prognostic marker for microvascular complications of diabetes.

References

1. Adeghate E, Schattner P, Dunn E. An update on the etiology and epidemiology of diabetes mellitus. Ann N Y Acad Sci. 2006;1084:1–29.
2. Cho NH, Shaw JE, Karuranga S, Huang Y, da Rocha Fernandes JD, Ohlrogge AW, et al. IDF diabetes atlas: global estimates of diabetes prevalence for 2017 and projections for 2045. Diabetes Res Clin Pract. 2018;138:271–81.
3. Powers AC. Diabetes mellitus. In: Longo DL, Fauci AS, Kasper DL, Hauser SL, Loscalzo J, Jameson JL, editors. Harrison's principles of internal medicine. 20th ed. New York: McGraw-Hill; 2018. p. 2968–3002.
4. Marshall SM, Barth JH. Standardization of HbA1c measurements–a consensus statement. Diabet Med. 2000;17(1):5–6.
5. Ferroni P, Basili S, Falco A, Davì G. Platelet activation in type 2 diabetes mellitus. J Thromb Haemost. 2004;2(8):1282–91.
6. Coughlin SR. Protease-activated receptors in hemostasis, thrombosis and vascular biology. J Thromb Haemost. 2005;3(8):1800–14.
7. Buch A, Kaur S, Nair R, Jain A. Platelet volume indices as predictive biomarkers for diabetic complications in type 2 diabetic patients. J Lab Physicians. 2017;9(2):84–8.
8. Miettinen H, Haffner SM, Lehto S, Rönnemaa T, Pyörälä K, Laakso M. Proteinuria predicts stroke and other atherosclerotic vascular disease events in nondiabetic and non-insulin-dependent diabetic subjects. Stroke. 1996;27(11):2033–9.
9. Walinjkar RS, Khadse S, Kumar S, Bawankule S, Acharya S. Platelet indices as a predictor of microvascular complications in type 2 diabetes. Indian J Endocrinol Metab. 2019;23(2):206–10.
10. Dindar S, Cinemre H, Sengul E, Annakkaya AN. Mean platelet volume is associated with glycaemic control and retinopathy in patients with type 2 diabetes mellitus. West Indian Med J. 2013;62(6):519–23.
11. Kodiatte TA, Manikyam UK, Rao SB, Jagadish TM, Reddy M, Lingaiah HK, et al. Mean platelet volume in type 2 diabetes mellitus. J Lab Physicians. 2012;4(1):5–9.
12. Pujani M, Gahlawat H, Agarwal C, Chauhan V, Singh K, Lukhmana S. Platelet parameters: can they serve as biomarkers of glycemic control or development of complications in evaluation of type 2 diabetes mellitus? Iraqi J Hematol. 2018;7:72–8.
13. Kshirsagar RM, Deoke S, Akhtar S. Panacea J Med Sci. 2019;9(1):23–8 10.18231.

International Journal of Diabetes in Developing Countries (2021) 41:94–100
https://doi.org/10.1007/s13410-020-00847-y

ORIGINAL ARTICLE

Foot care practices among Diabetologists in India: A descriptive study by the Diabetic Foot Research India

Vijay Viswanathan[1] · Sukanya Nachimuthu[2] · Balkhiwala Ahmed Khan[2]

Received: 13 March 2020 / Accepted: 2 July 2020 / Published online: 15 August 2020

Abstract

Background Evidence regarding the foot care practices among practitioners is still scarce in the Indian scenario. This study aimed at assessing the foot care practices among diabetologists from various parts of the country.

Methods A cross-sectional online survey was conducted among diabetologists, to understand their foot care practices across India, who were part of Diabetic Foot Research India (DFRI) and have participated in the study.

Results In a total of 89 participants, majority of the diabetologists examined patients with a history of foot infection once in 3 months (53%), 128 Hz tuning fork has been used for diagnosis of high-risk foot (31%), majority of patients were advised to use TCC by diabetologists (73%), and footwear was manufactured by qualified orthotist and prosthetics (60%). The foot problems commonly treated by diabetologists are cellulitis (26%) followed by callus (24%). Patients were also referred to other specialists (69%); almost all the participants used antibiotics (95%), and medication and wound care management (29%) were also a part of their practice.

Conclusion The diabetologists displayed a multidisciplinary approach to the treatment and management of foot care practices among patients with foot complications. It is imperative to establish effective communication between diabetologists and/or other practitioners to generate sufficient evidence to determine the foot care practices throughout the country.

Keywords Diabetic foot care · Foot care practice among diabetologists · High risk foot · Foot examination · Indian diabetologists

Background

The number of people with diabetes worldwide according to the International Diabetes Federation (IDF) is predicted to increase to 84% by 2045 in India [1]. A rapid change in these figures due to this chronic condition is envisioned to expand, raising more concern on public health and the livelihood of the community [2]. Diabetic foot accounts to nearly 35% of hospital admissions among the diabetic populations and continues to pose a challenge over management and practice among doctors, podiatrist, or any trained staff [3, 4]. It has been estimated that in developing countries like India, foot problems attribute to 40% of the use of healthcare resources [5]. Foot problems and their complications are widely seen throughout the country which can be due to inadequate knowledge, awareness, and education with an indirect influence by poverty on foot care practices prevailing in the community, importantly among patients accustomed to socio-cultural compulsion and traditional practices like barefoot walking (both indoors and outdoors) including the absence of timely access to podiatry services. These kinds of habits naturally debilitate an individual's physical quality of life among the diabetic population with peripheral neuropathy, foot ulceration, and peripheral arterial disease leading to amputation as a result of diabetic foot complications [5, 6]. According to a study, patients from developing countries such as India spent almost 32% of their total income on foot problems [7]. This presents an enormous economic burden upon the diabetic individual's family and the health system

✉ Vijay Viswanathan
drvijay@mvdiabetes.com

Sukanya Nachimuthu
epidemiology@mvdiabetes.in

Balkhiwala Ahmed Khan
physiofootresearch@mvdiabetes.in

[1] M.V. Hospital for Diabetes & Prof. M. Viswanathan Diabetes Research Centre, No. 4, West Madha Church road, Royapuram, Chennai 600 013, India

[2] Prof. M. Viswanathan Diabetes Research Centre, Chennai, India

as a result of inflated healthcare expenditure related to diabetic foot complications. Though the American Diabetes Association (ADA) recommends that people with diabetes should undergo a comprehensive foot examination once a year [8], multicentric studies conducted in India report that 65% of the patients hardly followed proper foot care practices [7, 8]. These pieces of information portray the importance of not only a doctor's role in educating a patient on foot care practices and preventing complications [9] but also their responsibility in adopting effective interventions that are tailor-made to the demands of an individual with a high-risk foot. Presently, individual practitioners, surgical specialists (podiatry, plastic surgeon, and orthopedic surgeon), endocrinologists, and primary care physicians are involved in the healthcare delivery to a patient with diabetic diabetic foot lesions [10]. And data regarding foot care practices among diabetologists is still lacking in the Indian scenario as compared with practices among patients in different study settings. Hence, this study is aimed at assessing the foot care practices among diabetologists from various parts of India to understand their diversity on management and treatment of foot complications, including the identification of key foot care practices that are indicators attributable to an improved outcome of a patient with the high-risk foot.

Materials and methods

A cross-sectional online survey was conducted among diabetologists across India who were part of Diabetic Foot Research India (DFRI) from March 2017 to August 2018. A list of diabetologists and/or members, who were registered at DFRI, was prepared by the DFRI team followed by a personal invitation for participation in the study. The cohort of diabetologists was from 18 states widely spread throughout the country with members from Gujarat, Tripura, Rajasthan, Punjab, Maharashtra, Andhra Pradesh, Telangana, Tamil Nadu, Odisha, Pondicherry, Jharkhand, Kerala, Uttar Pradesh, Himachal Pradesh, West Bengal, Assam, Bihar, and Delhi. A complete enumeration of all the available participants was carried to re-ensure a better picture of their practice. Participants who consented for participation were included in the study. An e-mail containing the link to the online survey was sent to each participant. A total of 89 diabetologists participated in the study, with a response rate of 60% among the members. Data were collected through a questionnaire (online) from diabetologists via a secured web link which could be completed at their own convenience. The questionnaire consisted of information regarding foot care practices, the examination of foot infection, treatment, type of referral, and management procedures. The completed questionnaire was downloaded at the head institution in Royapuram, Chennai, Tamil Nadu, and processed for analysis. Statistical analysis was done by using SPSS version.20. Descriptive analysis was performed, and the results were obtained regarding measures of central tendency and dispersion including distribution of foot care presented in tables and charts.

Results

The results are on diabetic foot care practices among diabetologists throughout India. Each of the variables was individually analyzed, and the result was interpreted accordingly under the given domains.

Among the 89 participants, the percentage distribution of examination practiced by the diabetologists (Table 1) showed that 53% of the diabetologists examined the patients once in 3 months. 10% of the diabetologists examined the patients once in 6 months. 6% of the diabetologists examined once in a year. And other 31% examined less frequently.

It was observed that most of the physicians (88%) used a minimum of three procedures to evaluate and diagnose a high-risk foot (Table 1). Some of the methods were tuning fork 128 Hz (31%), monofilament test (29%), biothesiometer (20%), and ankle-brachial index (ABI) (20%). This was observed in the study done by Boulton and Mayfield et al. where the tuning fork (128 Hz) and monofilament 10 g (otherwise called Semmes-Weinstein mono filament) were commonly used due to their high predictability of re-ulceration, easy to administer, portability, and cost-effectiveness [11, 12]. Figure 1 represents the conditions and/or foot problems commonly treated by the diabetologists around the country.

Cellulitis (26%) and callus (24%) were the most treated conditions followed by ingrown toe nail (19%), gangrene (17%), and bony deformities such as Charcot foot (14%). These conditions could certainly lead to other invasive procedures such as amputation unless appropriate management was implemented.

The procedures used to treat foot problem and the choice of treatment are two of the important challenges faced by any physicians (Table 1). Procedures such as wound debridement are an important aspect of wound healing which removes non-viable tissues or infection material. Surgical intervention in the presence of systemic toxicity is imperative. In this study, wound debridement (49%), toe amputation (28%), below knee (18%), and tendon Achilles lengthening (5%) were predominantly practiced by the and their team.

Table 1 Questions and the responses given by the diabetologists

S. no.	Questions	Responses	Percentage (%)
1.	Frequency of foot examination by diabetologists	A) Once in a year	6
		B) Once in 6 months	10
		C) Once in 3 months	53
		D) Others	31
2.	Method used to diagnose high-risk foot	A) 128 Hz	31
		B) Monofilament test	29
		C) Biothesiometer	20
		D) ABI	20
3.	Procedures used to treat foot problems	A) Wound debridement	49
		B) Toe amputation	28
		C) Tendon Achilles lengthening	5
		D) Below knee amputation	18
4.	Patients referred to personnel for offloading	A) Qualified orthotists and prosthetics	62
		B) Shoe maker	12
		C) Self	18
		D) Orthopedic surgeon	1.5
		E) Podiatry assistant	1.5
		F) Foot surgeon	3
		G) Others	2
5.	Patients referred to the surgeons for foot infection	A) General surgeon	40
		B) Orthopedic surgeon	16
		C) Foot surgeon	23
		D) Plastic surgeon	21
6.	Conditions on which patients referred to vascular surgeon	A) H/o claudication	3
		B) ABI < 0.8	5
		C) Both of the above	78
		D) None of the above	14

Management of foot problems

The management of a patient could be subjective to treatment compliance and importantly based on psycho-social acceptance to the management for better outcome and prevention. The current study revealed that the majority of physicians (Fig. 2) provided referral to surgeons outside their clinic/hospital (69%) to manage patients with

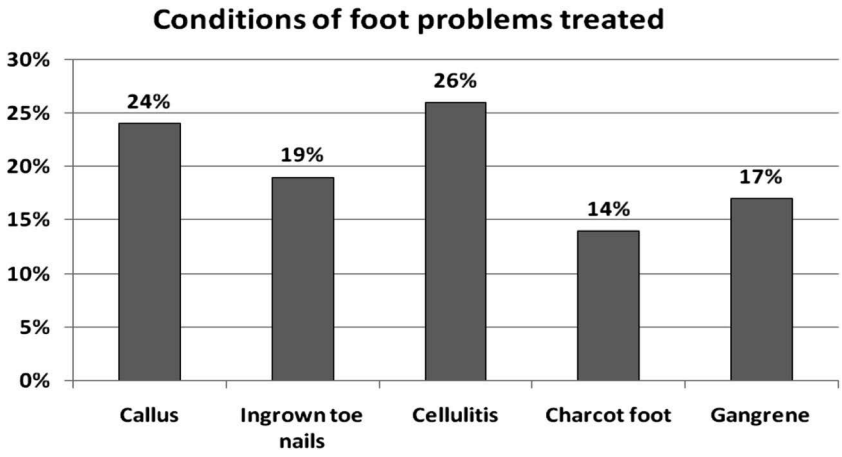


Fig. 1 Distribution of foot problems commonly treated by the diabetologists

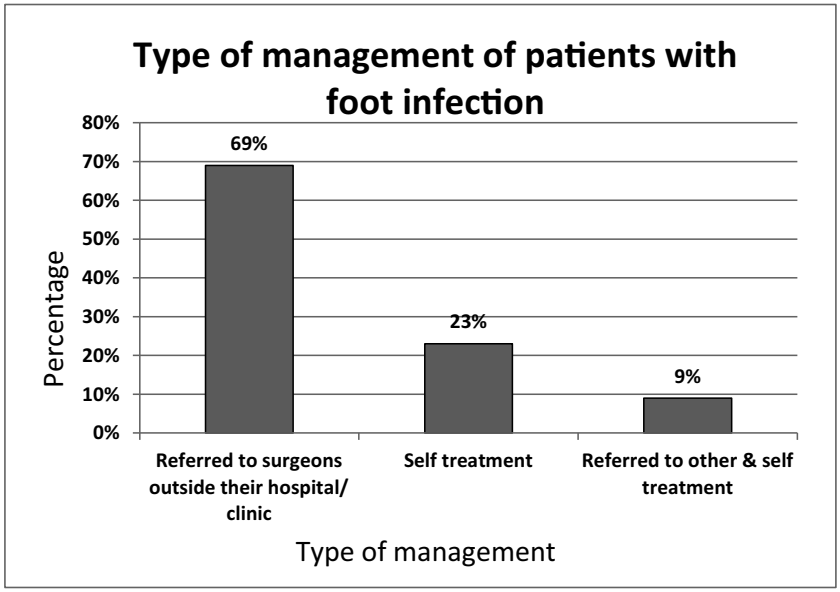


Fig. 2 Distribution on type of management of patients with foot infection

foot infection or ulcers, and around 23% of the diabetologists treated patients at their own clinic/hospital while 9% practiced both self-treatment and referred patients to other surgeons.

Among the group who referred the patients to other surgeons, nearly 40% of patients were referred to a general surgeon, 16% to an orthopedic surgeon, 23% to the foot surgeon, and 21% to the plastic surgeon (Table 1).

The patients were also referred to vascular surgeons only under specified conditions (Table 1). Among the group, 78% of the patients with both history of claudication and ankle-brachial index (< 0.8) were referred to a vascular surgeon compared with patients with only claudication history (3%) and ABI (5%). Also, they recommended/advised patients with foot ulcer on different offloading methods.

Patients were referred to other personnel for offloading methods. Nearly 62% of the physicians referred a patient to a qualified orthotist and prosthetist for offloading, and 18% have performed the offloading by themselves. Some diabetologists also referred patients to a shoe maker (12%), podiatry assistant (1%), and orthopedic and podiatry surgeon (1% and 3%) including other surgeons (2%). Apart from referrals, patients were also given appropriate offloading advices/counselling for better outcome of foot ulcer. It was observed that the majority of physicians advised patients for total contact casting (TCC): 69 (77.5%) offloading than other methods and 20 (22.5%) such as diabetic footwear, offloading sandals (customized), modified footwear, customized footwear, prefabricated walker, front and hind foot offloading, bed rest and walking stick, plaster of Paris slab, rocker bottom footwear, and POP. Therefore, total contact casting was the first choice for offloading by the physicians. In line with this finding, a study done by Giovanni et al. showed that TCC had excellent healing tendency of neuropathic foot ulcers and an average healing time of 8.5 weeks [13].

Management and treatment of high-risk foot involve a combination of therapy and medication that facilitate the process of wound healing. The usage of antibiotics (Fig. 3) according to the standard treatment guidelines for diabetic foot solely depends on the severity of the infected wound [10]. Almost 95% of the participants had used antibiotics either as an empirical therapy or regimen, while only 5% have not used any antibiotic medication. The reasons for the use of antibiotics were further quantified in the following representation (Fig. 4).

Most of the participants used empirical antibiotics (41%), opted for wound culture (33%), and 21% used for both.

Analysis on medication and performed therapy (Fig. 5) showed that 47% of the diabetologists used negative pressure wound therapy, 12% used growth factors, 29% used both the methods, and 13% of the diabetologists did not use either of them in their foot care practice. Therapies for wound healing such as negative pressure wound therapy are safe and effective in improving the wound healing rates [14].

Discussion

Although studies substantiating the practices on foot care among patients are available under diverse setups [6, 9], there is a dearth of evidence concerning diabetologists'/physicians' practice on foot care and management. Our study findings showed that majority of the participants examined patients every 3 months. This could make a better practice since according to the position statement by the ADA, earlier identification of patients with diabetes and high-risk foot could prevent the risk of foot complications that could lead to amputation [12]. The findings also noted that the method of assessment used for diagnosing a high-risk foot especially

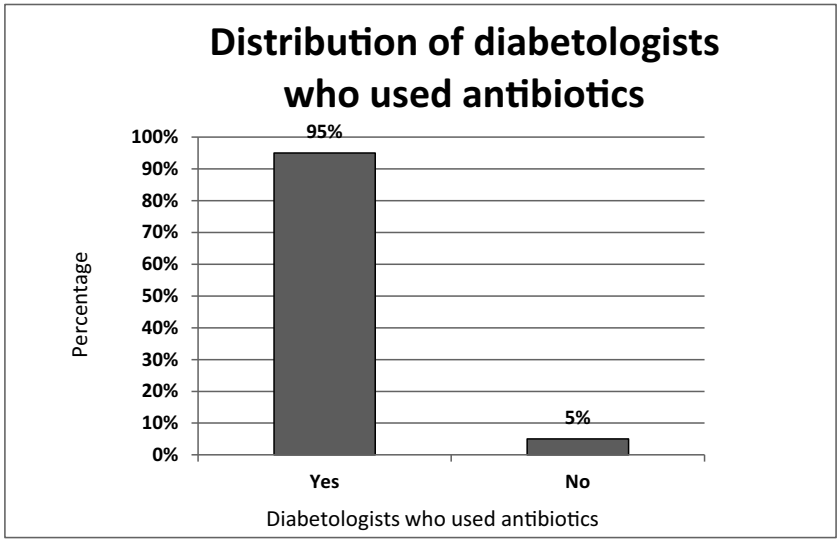


Fig. 3 Distribution of diabetologists who used antibiotic

neurological assessment like tuning fork (128 Hz), mono filament therapy, and vibration perception threshold was commonly adopted. According to Boulton et al., a comprehensive foot examination involved the selection of all three methods including ABI under vascular assessment [15, 16]. The current study noted that most of the diabetologists referred patients to a general surgeon (40%) followed by foot and plastic surgeon (23% & 21%). This was inversely related to the results obtained by De Berardis et al. where other practitioners were referred or examined less compared with diabetologists. These findings also signify the choice of a diabetologist on referral for management of the patient's foot infection [17]. Studies reveal that a team of multidisciplinary specialists should be involved in the effective management of patients, including vascular surgeons [18]. The current study also showed that vascular surgeons play an important role in the treatment of patients with a history of claudication and poor ABI. Other types of physical and clinical management (the type of offloading, usage of antibiotic, referral for offloading) were also important in the mitigation of diabetic foot complications using a comprehensive approach [19]. Perspectives on wound care and healing using growth factors and negative pressure wound therapy have proven successful in the faster wound healing process by accelerating angiogenesis and enhancing cellular proliferation [19–21]. The present study showed that almost 29% of the diabetologists used both types of management.

The variability in the healthcare system under the Indian context demands the need for a common guideline to restrict the increase of diabetic foot and its complications. Information on studies relating to standard guidelines on foot care practice is still lacking in the country including unspecified roles and responsibilities or a protocol under a common domain. This study tried to acquire a snapshot of the common practice of foot care among diabetologists and practitioners who were from different parts of the country. Many of the diabetologists

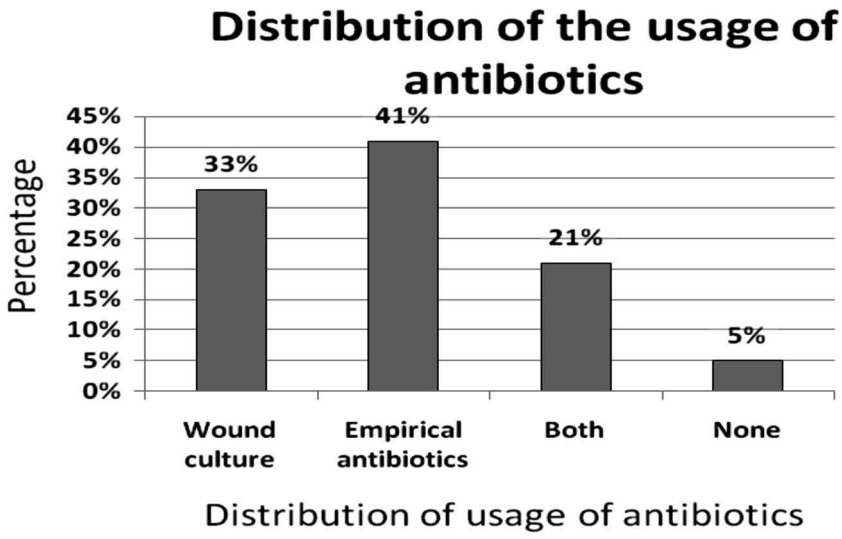


Fig. 4 Distribution on the usage of antibiotic

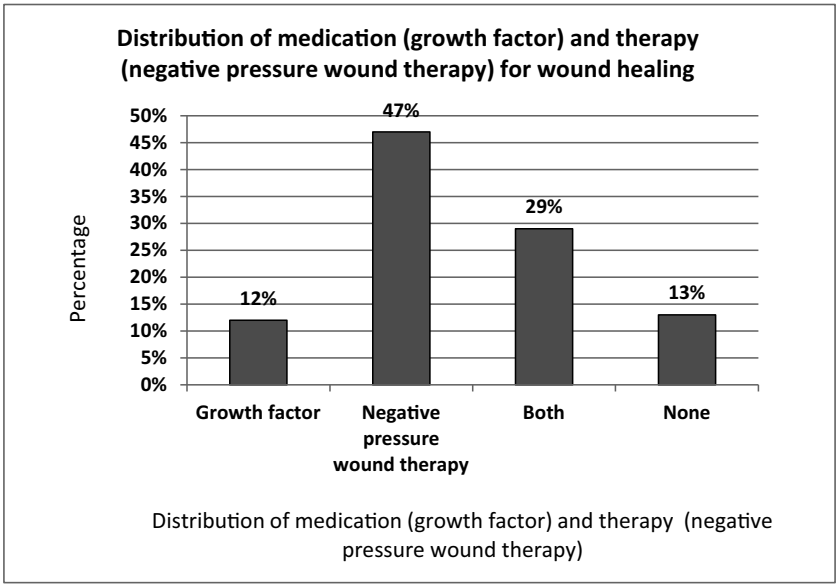


Fig. 5 Distribution of medication (growth factor) and therapy (negative pressure wound therapy) for wound healing

adopted a common approach for the treatment and management of diabetic foot complications examining diverse conditions associated with diabetic foot and complications. It is imperative to understand and establish effective communication between diabetologists and/or practitioners to generate sufficient evidence to determine their foot care practices throughout the country and help cater effective intervention for intensive patient-centered management on diabetic foot complications among practitioners, who play a vital role in preventing patient re-admission to hospitals and demonstrating prompt care for better prognosis of foot problems [22, 23].

Limitations and future research

The number of participants in this study was limited to practitioners who consented for participation. Therefore, the generalization of the study results is not possible, although results could improve with an increased sample size in the future studies by including more practitioners.

Acknowledgments This work was done with the data obtained from the following DFRI members, Dr. M. Senthil Kumar, Dr. Mohit Malhotra, Dr. M. Muralikrishna, Dr. B. RajaGanessan, Dr. Ragavendra Sthapak, Dr. Digambar Naik, Dr. Sriharee, Dr. S. Harinatha Reddy, Dr. P. Krishna Prasanthi, Dr. Divya, Dr. Mohammed Tauseef, Dr. Gaurav Chhaya, Dr. Vijayarani, Dr. Mangesh Tiwaskar, Dr. Mukul Borah, Dr. Mk. Rajendran, Dr. Jyoti G Mannari, Dr. M. Pavankumar, Dr. Bharani, Dr. Prasanth Sankar, Dr. Jugal Kishor Sharma, Dr. Sanjay Dash, Dr. Debiprasad Sarkar, Dr. Kannan, Dr. Bhavatharini Aruyerchelvan, Dr. R. Gobinath, Dr. Sangeetha, Dr. M.S. Ashraf, Dr. Rajesh Kumar Jayaram, Dr. Amitabh Sur, Dr. Shahnowaj Majumder, Dr. Anand Shankar, Dr. Nirupam Prakash, Dr. Mannepalli Ramakrishnarao, Dr. Jayaprakash Sahoo, Dr. Ravi Kant Mishra, Dr. Ajay patwari, Dr. G. Anil Kumar Reddy, Dr. S. Vinoth Kumar, Dr. Sadishkumar Kamalanathan, Dr. Prashant, Dr. Ramesh Reddy, Dr. Shiva Shankar Polavarapu, Dr. Kannammai, Dr. Basab Ghosh, Dr. Ponnusamy, Dr. G.Shanti Kumar, Dr. Kishorekumar Modi, Dr. P. Nellaippar, Dr. S.R. Ashok, Dr. Yagna narayana reddy, Dr. M.V.Jaiganeshan, Dr. L.Meenakshisundaram, Dr. Hyderi, Dr. Rajnish Saxena, Dr. Amit Naghate, Dr. Bhavesh Patel, Dr. K. Bhuvanesh, Dr. M. Muralikrishna, Dr. Rishi George, Dr. Mriganka Talukdar, Dr. Vinay Dhandhania, Dr. Sanjay Gandhi, Dr. M.V. Lakshmanan, Dr. Ragini Rohatgi, Dr. Niraj Soni, Dr. Snehal Vishnu Fegade, Dr. K.C. Mohapatra, Dr. Vijay S Shivpuje, Dr. Sivakumar Krishnamoorthy, Dr. Ashok Damir, Dr. Sujay Chanda, Dr. Laxmi Chaitanya Ramathoti, Dr. Darshak, Dr. Tirthankar Satpathi, Dr. Sandip Debashis Mishra, Dr. Ramchandra Saini, Dr. Mohammed Rennis, Dr. Mokalla Venkateswararao, Dr. Joozer Rangwala, Dr. Nitin Subrav Gade, Dr. Mukesh kapadia, Dr. R. Gopinath, Dr. S. Rajasekaran, Dr. M. Ramesh Kumar, Dr. Amar Jeet Singh Nagpal, Dr. K. Mahalakshmi, Dr. Sabeer TK, and Dr. Prakash Mohanty, who participated in the survey.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflicts of interest.

References

1. IDF DIABETES ATLAS Eighth edition 2017. http://www.diabetesatlas.org/ Access 27th November 2019.
2. Gupta, Sanjeev, Singh, Surya. Diabetic foot: a continuing challenge. Adv Exp Med Biol. 2012;771:123–38.
3. Ahmad J. The diabetic foot. Diabetes Metab Syndr. 2016;10(1):48–60. https://doi.org/10.1016/j.dsx.2015.04.002.
4. Little M, Humphries S, Patel K, Dewey C. Decoding the type 2 diabetes epidemic in rural India. Med Anthropol. 2016;36(2):96–110. https://doi.org/10.1080/01459740.2016.1231676.
5. Viswanathan V (2013) Delivering an effective foot care for people with diabetes. Book chapter. Mar 2015.
6. Taksande BA, Thote M, Jajoo UN. Knowledge, attitude, and practice of foot care in patients with diabetes at central rural India. J Fam Med Prim Care. 2017;6:284–7.
7. Abbas ZG, Viswanathan V. The diabetic foot in Africa and India. Int Diab Monit. 2007;19:8–12.
8. American Diabetes Association (ADA). Diabetes.

9. George H, Rakesh PS, Krishna M, Alex R, Abraham VJ, George K, et al. Foot care knowledge and practices and the prevalence of peripheral neuropathy among people with diabetes attending a secondary care rural hospital in southern India. J Fam Med Prim Care. 2013;2:27–32.
10. Standard Treatment Guidelines. The diabetic foot. Prevention and management in India, January 2016. http://clinicalestablishments.gov.in/WriteReadData/5381.pdf Access 27th November 2019.
11. Mayfield JA, Sugarman JR. The use of the Semmes-Weinstein monofilament and other threshold tests for preventing foot ulceration and amputation in persons with diabetes. J Fam Pract. 2000.
12. American Diabetes Association: Preventive foot care in diabetes. Diabetes Care. 2004;27(suppl 1):s63–4.
13. Matricali GA, Deroo K, Dereymaeker G. Outcome and recurrence rate of diabetic foot ulcers treated by a total contact cast: short-term follow-up. Foot Ankle Int. 2003;24(9):680–4. https://doi.org/10.1177/107110070302400905.
14. Tecilazich F, Dinh TL, Veves A. Emerging drugs for the treatment of diabetic ulcers. Expert Opin Emerg Drugs. 2013;18(2):207–17. https://doi.org/10.1517/14728214.2013.802305.
15. Boulton AJ, Armstrong DG, Albert SF, Frykberg RG, Hellman R, Kirkman M, et al. Comprehensive foot examination and risk assessment: a report of the task force of the foot care interest group of the American Diabetes Association, with endorsement by the American Association of Clinical Endocrinologists. Diabetes Care. 2008;31(8):1679–85. https://doi.org/10.2337/dc08-9021.
16. Kishore S, Upadhyay AD, Jyotsna VP. Categories of foot at risk in patients of diabetes at a tertiary care center: insights into need for foot care. Indian J Endocrinol Metab. 2015;19(3):405–10. https://doi.org/10.4103/2230-8210.152789.
17. De Berardis G, Pellegrini F, Franciosi M, Belfiglio M, Nardo BD, Greenfield S, et al. Physician attitudes toward foot care education and foot examination and their correlation with patient practice. Diabetes Care. 2004;27(1):286–7. https://doi.org/10.2337/diacare.27.1.286.
18. Chandra V, Glebova NO, Salvo NL, Wu T. Partnerships between podiatrists and vascular surgeons in building effective wound care centers. J Am Podiatr Med Assoc. 2017;66(3):902–5. https://doi.org/10.1016/j.jvs.2017.06.071.
19. Wukich DK, Armstrong DG, Attinger CE, Boulton AJ, Burns PR, Frykberg RG, et al. Inpatient management of diabetic foot disorders: a clinical guide. Diabetes Care. 2013;36(9):2862–71. https://doi.org/10.2337/dc12-2712.
20. Liu S, He CZ, Cai YT, Xing QP, Guo YZ, Chen ZL, et al. Evaluation of negative-pressure wound therapy for patients with diabetic foot ulcers: systematic review and meta-analysis. Ther Clin Risk Manag. 2017;13:533–44. https://doi.org/10.2147/TCRM.S131193.
21. IWGDF guidelines 2019. https://iwgdfguidelines.org/wp-content/uploads/2019/05/02-IWGDF-prevention-guideline-2019.pdf Access 27th November 2019.
22. Dündar C, Akıncı GE. Knowledge and practice of foot care in diabetic inpatients: a descriptive cross-sectional study. Erciyes Med J. 2017;39(4):160–4.
23. Saber HJ, Daoud AS. Knowledge and practice about the foot care and the prevalence of the neuropathy among a sample of type 2 diabetic patients in Erbil, Iraq. J Fam Med Prim Care. 2018;7:967–74.

International Journal of Diabetes in Developing Countries (2021) 41:101–107
https://doi.org/10.1007/s13410-020-00854-z

ORIGINAL ARTICLE

Prevalence of musculoskeletal complications of type-2 diabetes mellitus in population of southern Punjab, Pakistan

Muhammad Nasir Kalam[1,2] · Syed Nisar Hussain Shah[2] · Muhammad Fawad Rasool[2] · Abdul Majeed[2] · Naveed Ahmed[1] · Muhammad Saqlain[1]

Received: 4 September 2019 / Accepted: 27 July 2020 / Published online: 11 August 2020

Abstract

Background Diabetes mellitus is a major public health issue globally, and type-2 diabetes mellitus is the most prevalent form of this disease. Due to multiple factors, most diabetic patients develop functional disabilities including musculoskeletal complications.

Objective To determine the frequency of musculoskeletal complications, the relationship between these complications and different potential variables, and the status of pharmacotherapy management to treat these complications among patients with type-2 diabetes mellitus.

Materials and Methods It was a cross-sectional study and data was collected through self-administered, face-validated pro forma in the English language in outpatient clinics of two tertiary care hospitals of Multan, Pakistan.

Results Total $n = 270$ type-2 diabetic patients of both genders were assessed for musculoskeletal complications; $n = 126$ (46.7%) patients had at least one musculoskeletal complication. The most common manifestations were shoulder capsulitis or frozen shoulder (20.7%), flexor tenosynovitis (10.4%), hand stiffness syndrome (10%), and limited joint mobility (9.6%). A statistically significant association was found between frozen shoulder, limited joint mobility, and hand stiffness syndrome with the duration of diabetes (p value = 0.003, 0.0001), and overall glycemic control (p value = 0.004, 0.001) respectively. Out of 254 complaints, only 42.9% of complaints were receiving pharmacotherapy management. Main outcome measure: overall prevalence of musculoskeletal complications among patients of type-2 diabetes and level of pharmacotherapy management for them.

Conclusion Periarticular region of the joints in the hands and shoulders should be examined whenever diabetic patients present with uncontrolled diabetes, and pharmacotherapy management should be planned for these patients who will improve their quality of life.

Keywords Musculoskeletal complications · Peripheral neuropathy · Shoulder capsulitis · Pharmacotherapy management

✉ Muhammad Nasir Kalam
mnkalam@bs.qau.edu.pk

Syed Nisar Hussain Shah
nisarhussain@bzu.edu.pk

Muhammad Fawad Rasool
fawadrasool@bzu.edu.pk

Abdul Majeed
abdulmajeed@bzu.edu.pk

Naveed Ahmed
natanoli@qau.edu.pk

Muhammad Saqlain
msaqlain@bs.qau.edu.pk

[1] Department of Pharmacy, Quaid-i-Azam University, Islamabad 45320, Pakistan

[2] Pharmacy Practice Department, Faculty of Pharmacy, Bahauddin Zakariya University, Multan, Pakistan

Introduction

Diabetes mellitus (DM) is the most alarmingly growing global problem, and its associated cost to the society is continuously escalating [1]. DM is a multi-system disease characterized by persistent hyperglycemia and poor glycemic control can lead to acute and chronic complications. American Diabetes Association (ADA) classifies diabetes mellitus as type-1 diabetes mellitus (type-1 DM), TYPE-2 diabetes mellitus (type-2 DM), gestational diabetes mellitus (GDM), and other specific types [2]. The type-2 DM is the most prevalent form, as it accounts for approximately 90% of the total cases of diabetes [3]. A global emergency has to be declared to address this

global issue. Progression of this disease is rapid, and every day thousands of new individuals are diagnosed with DM [4]. Diabetes affects the lives of individuals and their families psychologically, socially, and financially. Quality of life (QoL) has been affected a lot due to this disease [5]. About 415 million adults, in age from 20 to 79 years, are living with diabetes and 318 million have impaired glucose tolerance (IGT) [1]. In comparison with microvascular and macrovascular complications associated with DM, the musculoskeletal (MSK) complications are less recognized and reported to be treated very poorly [6].

MSK complications are common endocrine arthropathies and few of them can lead to permanent disability. These complications are not only limited to joints but bones and soft tissues are affected equally. The US National Health Interview Survey (US-NHIS) reported that 58% of patients with diabetes have a functional disability [7]. The connective tissues are affected by hyperglycemia in DM which causes abnormalities of the periarticular and skeletal systems. These changes in connective tissues create various types of physical disabilities, which are classified as musculoskeletal complications [8]. It is proposed that the advanced glycation end products (AGEs) and neuropathy are underlying causes of these complications [9, 10]. Ischemia and infection of a periarticular tissue are other factors which are to be considered a risk for MSK complication and can lead to muscle infarction and foot ulcers [11]. Although these disabilities are more frequently associated with type-1 DM [7], they are also present in patients with type-2 DM [12–14].

Physical disability due to musculoskeletal complications is an emerging public health problem because its prevalence is increasing proportionally with the prevalence of DM. Previous studies showed that the prevalence of MSK complications in the hands and shoulder region was around 30% among patients with type-1 and type-2 DM [12]. These manifestations are closely linked to age [14], prolonged disease duration [15, 16], and vascular complications [17]. Most common musculoskeletal complaints associated with poorly controlled DM are diabetic cheiroarthropathy or limited joint mobility (LJM), flexor tenosynovitis (FT), carpal tunnel syndrome (CTS), Dupuytren's contracture (DC), frozen shoulder or shoulder capsulitis (SC), reflex sympathetic dystrophy (RSD), Charcot joint and diabetic muscle infarction [12–16, 18, 19]. The MSK complications associated with DM have increased the cost of illness (COI) and have decreased the quality of life (QoL) of these patients by jeopardizing their mobility. Therefore, it is the need of time to prioritize the pharmacological management of MSK complications along with other long-term complications of DM. The primary objective of this study was to estimate the frequency of MSK complications in patients with type-2 DM of Multan, Pakistan, whereas the secondary objective was to determine the relationship between the MSK complications and their associated risk factors in patients with type-2 DM.

Materials and Methods

Study design, study settings, and sample size

A cross-sectional descriptive study design was employed to accomplish the study objectives. Study settings were endocrinology outpatient clinics of two tertiary care hospitals of Multan, Pakistan. Patients were approached and enrolled in the current analysis by using a convenient sampling method based on the universal sampling technique. This technique was implemented as all patients with diabetes attending the settings during the study duration of 6 months were invited to participate in the study. A total of 327 patients were approached, evaluated on pre-defined inclusion/exclusion criteria, and 270 patients were included in the final analysis. Patients with missing data ($n = 37$) and who did not meet the inclusion criteria ($n = 20$) were excluded.

Inclusion criteria

The adult patients with diabetes, having a history of diabetes for more than 5 years with a confirmed diagnosis of MSK complication, were selected to participate in the study.

Exclusion criteria

Patients diagnosed with rheumatoid arthritis, osteoarthritis, and osteoporosis were not included in this study. The patients with a history of diabetes for less than 5 years and those who were not willing to participate in the study were excluded from the study.

Data collection method

The study was performed by following the declaration of Helsinki. Patients were invited to participate in the study and those who agreed were explained about the nature and purpose of the study, and both oral and written consents were taken from the patients. A comprehensive data collection form was designed based on an extensive literature review. Data form comprised of two parts: (A) demographic characteristics and (B) clinical characteristics. Demographic characteristics include age, gender, and body mass index. Clinical characteristics include the duration of disease (in years), type of diabetes (type-1 or type-2 DM), fasting and random blood sugar level, HbA1C, and MSK complications (carpal tunnel syndrome, frozen shoulder, flexor tenosynovitis, Dupuytren's contracture, reflex sympathetic dystrophy, Charcot joint, and

Table 1 Demographic data and the clinical characteristics of the study population

	Female	Male
Mean age (in years)	54.88 ± 8.12	54.38 ± 8.33
Weight (kg)	65.47 ± 10.04	67.83 ± 11.20
Height (cm)	157 ± 06	167 ± 06
BMI (kg/m^2)	26.35 ± 4.24	24.07 ± 3.82
Obesity	20.4%	4.4%
Overweight	21.9%	21.5%
Ideal body weight	13.3%	16.7%
Under weight	0.7%	1.1%
HbA1c (%)	8.8 ± 1.6	8.6 ± 1.5
FBS (mg/dl)	137 ± 37.7	137 ± 37.1
RBS (mg/dl)	215 ± 64.62	216 ± 63.29

diabetic muscle infarction) screening. This study considered overweight subjects with body mass index (BMI) 25–29.9 and obese with BMI ≥ 30 according to WHO classification of obesity [20].

A systematic method was used to screen the patients to access MSK complications. First, their hands, shoulders, spine, and finally the lower limbs were examined. The following criteria were used to identify all of the manifestations [18, 21]. The prayer sign test was used to evaluate limited joint mobility, in which the patients were asked to put their hands in prayer position with the fingers fanned and the wrist maximally extended. If they were unable to do this, LJM was considered to be present [18]. Patients having less shoulders joint rotation than normal in at least three planes [21], unilateral pain for more than 1 month, and unable to lie on the affected shoulder were diagnosed as adhesive capsulitis. Localized or diffused unilateral pain in limbs with vasomotor disturbance, swelling, and impaired mobility were the characteristics of RSD. The Dupuytren's contracture was diagnosed if participants had any of the following signs: nodules on palm or digits, digital or palmer skin tethering and profound thickening of palm along with deformity of 3rd to 5th digits. The locking of fingers during extension and flexion process and presence of palpating nodule were considered to be the diagnostic criteria for FT or trigger finger. The carpal tunnel syndrome was considered to be present if participants claimed nocturnal paresthesia of thumb, first and second finger with or without positive Tinel's and Phalen's tests, pain or weakness of hands and had visible signs of thenar atrophy. Deformation and swelling of knee and ankle joints without symptoms of pain were diagnostic criteria for Charcot joint or neuropathic arthropathy. It was further confirmed from X-ray radiograph with differential pieces of evidence of joint dislocation, disorganization, surface destruction, and increased bone density. The diabetic muscle infarction was a rare condition and was identified with the following signs: skeletal muscle, benign painful lumpy mass and surgical removal in the past after the diagnosis of diabetes.

Data entry and analysis

Statistical analysis was performed by using SPSS 21 (IBM Corp. Released 2012. IBM SPSS statistics for Windows, version 21.0. Armonk, NY: IBM Corp.). Descriptive statistics were calculated as frequencies and percentages for categorical variables, and mean ± standard deviation was used to describe the numerical variables. The chi-square tests were applied to find an association between MSK manifestations and

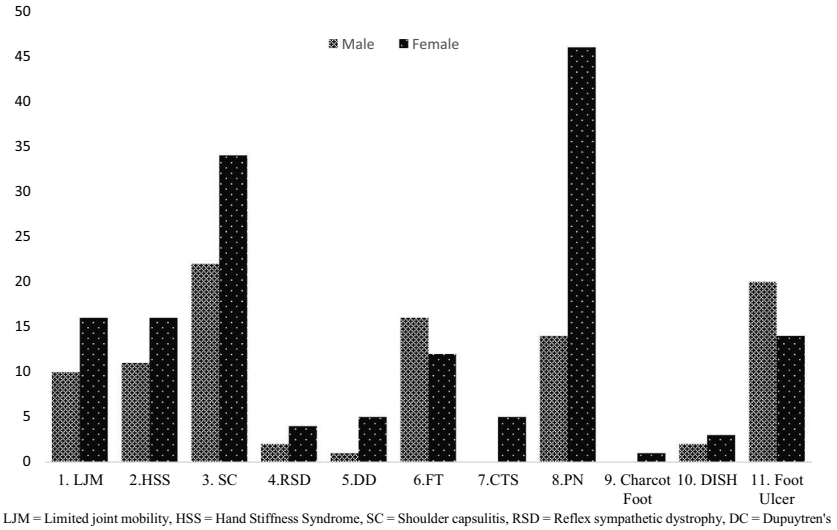


Fig. 1 Gender-wise distribution and prevalence of all musculoskeletal complications. LJM, limited joint mobility; HSS, hand stiffness syndrome; SC, shoulder capsulitis; RSD, reflex sympathetic dystrophy; DC, Dupuytren's contracture; FT, flexor tenosynovitis; CTS, Carpal tunnel syndrome; PN, peripheral neuropathy; DISH, diffuse idiopathic skeletal hyperostosis

Table 2 The difference in frequency of MSK by patient characteristics ($N = 270$)

MSK complications	Glycemic control			History of diabetes in years					Body mass index (BMI)			Gender		
	n (%)			*n* (%)					*n* (%)			*n* (%)		
	Optimal	Poor	*p* value	5–10	10–15	15–20	> 20	*p* value	Overweight	Obesity	*p* value	Male	Female	*p* value
LJM	0 (0.0)	25 (9.3)	*0.001*	4 (1.5)	9 (3.3)	10 (3.7)	2 (0.7)	*0.001*	9 (3.3)	6 (2.2)	0.812	10 (3.7)	15 (5.6)	0.695
HSS	0 (0.0)	27 (10)	*0.001*	4 (1.5)	9 (3.3)	12 (4.4)	2 (0.7)	*0.001*	10 (3.7)	7 (2.6)	0.812	11 (4.1)	16 (5.9)	0.744
SC	2 (0.7)	56 (20.7)	*0.004*	18 (6.7)	18 (6.7)	13 (4.8)	7 (2.6)	*0.003*	20 (7.4)	15 (5.6)	0.297	22 (8.1)	34 (12.6)	0.454
RSD	0 (0.0)	6 (2.2)	0.149	0 (0.0)	3 (1.10	2 (0.7)	1 (0.4)	0.088	4 (1.5)	1 (0.4)	0.707	2 (0.7)	4 (1.5)	0.599
DD	0 (0.0)	6 (2.2)	0.151	2 (0.7)	1 (0.4)	2 (0.7)	1 (0.4)	0.294	1 (0.4)	1 (0.4)	0.258	1 (0.4)	5 (1.9)	0.177
FT	0 (0.0)	27 (10)	0.116	10 (3.7)	8 (3.0)	8 (3.0)	2 (0.7)	0.153	13 (4.8)	6 (2.2)	0.840	16 (5.9)	12 (4.4)	0.130
CTS	0 (0.0)	5 (1.9)	0.208	1 (0.4)	2 (0.7)	2 (0.7)	0 (0.0)	0.342	2 0.7)	1 (0.4)	*0.026*	0 (0.0)	5 (1.9)	0.047
PN	0 (0.0)	70 (26)	*0.001*	15 (5.5)	31 (11.4)	16 (5.9)	8 (3.0)	*0.001*	14 (5.2)	6 (2.2)	0.655	24 (8.9)	46 (17.1)	0.174
Charcot Foot	0 (0.0)	1 (0.4)	0.734	0 (0.0)	0 (0.0)	1 (0.2)	0 (0.0)	0.130	0 (0.0)	0 (0.0)	0.505	0 (0.0)	1 (0.4)	0.174
DISH	0 (0.0)	5 (1.9)	0.208	0 (0.0)	3 (1.1)	2 (0.7)	0 (0.0)	0.110	2 (0.7)	1 (0.4)	0.505	2 (0.7)	3 (1.1)	0.866
Foot Ulcer	0 (0.0)	34 (12.6)	*0.001*	15 (5.6)	11 (4.1)	6 (2.2)	2 (0.7)	0.953	7 (2.6)	3 (1.1)	*0.001*	20 (7.4)	14 (5.2)	0.057

LJM limited joint mobility, *HSS* hand stiffness syndrome, *SC* shoulder capsulitis, *RSD* reflex sympathetic dystrophy, *DC* Dupuytren's contracture, *FT* flexor tenosynovitis, *CTS* Carpal tunnel syndrome, *PN* peripheral neuropathy, *DISH* diffuse idiopathic skeletal hyperostosis. A *p* value of less than 0.05 considered statistically significant

Italic values indicated significant association. Percentage values are rounded after one decimal point

demographic or clinical characteristics. A p value of less than 0.05 was considered to be significant.

Results

A total of 270 patients with type-2 DM were screened to analyze the prevalence of diabetes-related musculoskeletal complications, among which 118 (43.7%) were men and 152 (56.3%) were women. A total of 11 musculoskeletal complications were identified, and almost half of the study subjects ($n = 126$, 46.7%) had experienced at least one musculoskeletal complication. The demographic data and clinical characteristics of the study population can be seen in Table 1.

The most frequent MSK complications were LJM (9.6%), HSS (10%,), and the frozen shoulder or SC (20.7%). More than one-quarter of the patients (26.0%) were having complaints of peripheral neuropathy. The details of MSK complications can be seen in Fig. 1.

Findings demonstrated that occurrence of LJM, HSS, and PN was significantly related to history of diabetes ($p = 0.001$), and glycemic control ($p = 0.001$). Frozen shoulder or SC was also significantly associated with a history of diabetes ($p = 0.003$), and overall glycemic control ($p = 0.004$). There was no significant association of any MSK complaint with gender and BMI except foot ulcers or gangrenes, which was significantly associated with BMI levels ($p = 0.001$) and overall glycemic control ($p = 0.001$) (Table 2).

A total of 254 MSK complications were reported among 126 participants of this cross-sectional study; i.e., one patient was suffering from more than one MSK complications. Among these MSK complications, only 109 (42.9%) complaints were addressed by giving a pharmacological therapy while the remaining 145 (57.1%) were not given any pharmacological therapy. A significant association was present between MSK complaints and pharmacological care ($p = 0.0001$). Status of pharmacotherapy management among all these patients for each musculoskeletal complaint has been explained in Table 3.

Discussion

Musculoskeletal complications are the most under-reported manifestations associated with both type-1 and type-2 DM [6, 18, 22]. A lot of research has been done on the acute and chronic complications of DM [6, 18, 22], but to date the data related to MSK complications is very limited. There are very few published studies that have focused on MSK complications among patients with type-2 DM [13, 23, 24]. The International Diabetes Federation (IDF) and ADA have issued guidelines for the management and prevention of long-term complications with diabetes, but no instructions or measures are reported for MSK complications [6]. MSK complications are non-vascular and are responsible for the development of physical disabilities among patients with diabetes. Physical disabilities and muscular deformities can be easily linked with aging. Microvascular complications, poor glycemic control, and duration of diabetes are other contributing factors in the development of these complications [22]. More than 50% of patients with diabetes have a diabetes-related physical disability at any stage of their life [25]. It was reported that patients with diabetes have more mobility-related problems, and they experience greater difficulties in performing basic activities of daily living (ADL), when compared with the same age group of persons without DM [25]. The MSK complications have been associated with an increased frequency of hospital-related visits, admissions, stay, and mortality rate [25, 26]. Long-term morbidity can be avoided by early identification of these complications because when diagnosed at initial stages, most of them can be self-limiting [22].

Table 3 Pharmacotherapy status of patients with musculoskeletal complications

MSK complications	Yes	%	No	%	Total	%
1. Limited joint mobility (LJM)	6	2.4%	20	7.9%	26	10.2%
2. Hand stiffness syndrome (HSS)	7	2.8%	20	7.9%	27	10.6%
3. Shoulder capsulitis (SC)	19	7.5%	37	14.6%	56	22.0%
4. Reflex sympathetic dystrophy(RSD)	4	1.6%	2	0.8%	6	2.4%
5. Dupuytren's contracture (DC)	2	0.8%	4	1.6%	6	2.4%
6. Flexor tenosynovitis (FT)	9	3.5%	19	7.5%	28	11.0%
7. Carpal tunnel syndrome (CTS)	1	0.4%	4	1.6%	5	2.0%
8. Peripheral neuropathy (PN)	23	9.1%	37	14.6%	60	23.6%
9. Charcot foot	1	0.4%	0	0.0%	1	0.4%
10. Diffuse idiopathic skeletal hyperostosis (DISH)	3	1.2%	2	0.8%	5	2.0%
11. Foot ulcer/gangrene	34	13.4%	0	0.0%	34	13.4%
Total	109	42.9%	145	57.1%	254	100.0%

To best of our knowledge, this is the first study aimed to explore the prevalence and risk factors associated with MSK complications among Pakistani patients with diabetes. It also assesses the standard of pharmacological management to treat these complications. Frozen shoulder or SC (20.7%) was the most frequent MSK complication reported in the present study. In contrast to this finding, a similar cohort study reported less than 20% of study participants experienced shoulder capsulitis [27]. Frequencies of FT (10.4%) and LJM (9.6%) reported in the current study were lower than those in a similar study that states the frequency of these MSK as 20% and 58% respectively [26]. Like previous studies, the percentages of cases of DC and CTS were also not much higher in the present study [28, 29]. Peripheral neuropathy was the most common (22%) MSK complication of diabetes reported in the current study. It may be the cause of the development of HSS, DC, and CTS because previous studies showed a significant association between PN and these MSK complications [8, 22, 29–31]. In the current study, the women were reported to be more susceptible to the MSK complications than men. This variation may be due to poor glycemic control, less physical activity, and poor adherence to therapy than men [22, 31, 32]. One of the major findings of this study was the association of these complications with glycemic control and duration of disease. It was well-established evidence that a systematic process of care plays an important role to empower the patients to control their diabetes [31]. Severe cases of these complications require treatment either surgically or medically, but unfortunately, the present study explored that more than 50% of the population was receiving neither pharmacological care nor non-pharmacological interventions. Establishment of the pharmaceutical care plan and working in a multidisciplinary healthcare team can reduce not only symptoms of these disabilities but also the psychological stress in these patients [33]. Diabetes is a chronic condition and patients also need to understand it as a situation, not a disease. They have to adapt their lifestyle according to their individual needs. Most of the patients of diabetes are unaware of diabetes care and undesirable consequences of persistent hyperglycemia and are nonadherent to therapy or missing their doses [31]. To prevent the life-threatening complications of diabetes, patient's education about the management and care of disease is very important. Continuous adjustment in the medication regimen is required according to the glycemic control of patients. Most of the patients are taking the same medication for a longer period without consulting their healthcare professionals. As diabetes is a progressive disease, patients need to monitor their blood glucose level regularly to get the idea of their glycemic control. Health care professionals must motivate the patients for the management of their disease. Patients are to be asked to visit their physician regularly, at least once in 6 months, to evaluate the development of these rare complications of DM.

Conclusion

MSK complications are not commonly recognized as a complication of DM as they were present in 46.7% of the participants of this study. Results suggest that examination of the periarticular region of hand and shoulder joints should be done whenever a patient with diabetes visits and complains pain of joint. Many of these complications are potentially self-limiting and treatable, especially if diagnosed early. These are the most negligible nonvascular complications, but they affect the quality of life of patients a lot. To improve the quality of life (QoL), prescribed pharmacotherapy should be planned for these patients.

Compliance with ethical standards The ethical approval for this study was obtained from the ethical committee of the Department of Pharmacy, Bahuddin Zakariya University, Multan (Reference No. 029/PHP/16 dated 04-01-2016). The study was performed by following the Declaration of Helsinki. Patients were invited to participate in the study, and those who agreed were explained about the nature and purpose of the study, and both oral and written consents were taken from the patients.

Conflict of interest The authors declare that they have no conflict of interest.

References

1. Ogurtsova K, da Rocha FJ, Huang Y, Linnenkamp U, Guariguata L, Cho N, et al. IDF diabetes atlas: global estimates for the prevalence of diabetes for 2015 and 2040. Diabetes Res Clin Pract. 2017;128:40–50.
2. Gavin JR III, Alberti K, Davidson MB, DeFronzo RA. Report of the expert committee on the diagnosis and classification of diabetes mellitus. Diabetes Care. 1997;20(7):1183.
3. DeFronzo RA, Ferrannini E, Groop L, Henry RR, Herman WH, Holst JJ, et al. Type 2 diabetes mellitus. Nat Rev Dis Prim. 2015;1:nrdp201519.
4. Wild SH, Roglic G, Green A, Sicree R, King H. Global prevalence of diabetes: estimates for the year 2000 and projections for 2030: response to Rathman and Giani. Diabetes Care. 2004;27(10):2569.
5. Chan JC, Cho NH, Tajima N, Shaw J. Diabetes in the Western Pacific Region—past, present and future. Diabetes Res Clin Pract. 2014;103(2):244–55.
6. Larkin ME, Barnie A, Braffett BH, Cleary PA, Diminick L, Harth J, et al. Musculoskeletal complications in type 1 diabetes. Diabetes Care. 2014;37(7):1863–9.
7. Egede LE. Diabetes, major depression, and functional disability among US adults. Diabetes Care. 2004;27(2):421–8.
8. Arkkila P, editor. Hand and shoulder abnormalities in diabetic patients: association with diabetes-related complications and diseases. ANNALES-UNIVERSITATIS TURKUENSIS SERIES D; 1996: Turun yliopisto.
9. Brownlee M. Glycation products and the pathogenesis of diabetic complications. Diabetes Care. 1992;15(12):1835–43.
10. Nathan DM. The pathophysiology of diabetic complications: how much does the glucose hypothesis explain? Ann Intern Med. 1996;124(1_Part_2):86–9.
11. Wyatt LH, Ferrance RJ. The musculoskeletal effects of diabetes mellitus. J Can Chiropr Assoc. 2006;50(1):43–50.

12. Douloumpakas I, Pyrpasopoulou A, Triantafyllou A, Sampanis C, Aslanidis S. Prevalence of musculoskeletal disorders in patients with type 2 diabetes mellitus: a pilot study. Hippokratia. 2007;11(4):216–8.
13. Kidwai SS, Wahid L, Siddiqi SA, Ghauri I, Sheikh I. Upper limb musculoskeletal abnormalities in type 2 diabetic patients in low socioeconomic strata in Pakistan. BMC Re Notes. 2013;6(1):16.
14. Mathew AJ, Nair JB, Pillai SS. Rheumatic-musculoskeletal manifestations in type 2 diabetes mellitus patients in South India. Int J Rheum Dis. 2011;14(1):55–60.
15. Fasika S, Abebe SM, Kebede AG. The prevalence of shoulder and hand complications and associated factors among diabetic patients at University of Gondar Teaching Referral Hospital in Northwest Ethiopia. J Diabetes Res Clin Metab. 2013;2(1):8.
16. Serban A, Udrea G. Rheumatic manifestations in diabetic patients. J Med Life. 2012;5(3):252–7.
17. Kuzuya T, Nakagawa S, Satoh J, Kanazawa Y, Iwamoto Y, Kobayashi M, et al. Report of the Committee on the classification and diagnostic criteria of diabetes mellitus. Diabetes Res Clin Pract. 2002;55(1):65–85.
18. Cagliero E, Apruzzese W, Perlmutter GS, Nathan DM. Musculoskeletal disorders of the hand and shoulder in patients with diabetes mellitus. Am J Med. 2002;112(6):487–90.
19. Markenson JA. Rheumatic manifestations of endocrine diseases. Curr Opin Rheumatol. 2010;22(1):64–71.
20. Xavier P-SF. Obesity: criteria and classification. Proc Nutr Soc. 2000;59(04):505–9.
21. Ewald A. Adhesive capsulitis: a review. Am Fam Physician. 2011;83(4):417–22.
22. Mustafa KN, Khader YS, Bsoul AK, Ajlouni K. Musculoskeletal disorders of the hand in type 2 diabetes mellitus: prevalence and its associated factors. Int J Rheum Dis. 2016;19(7):730–5.
23. Molsted S, Tribler J, Snorgaard O. Musculoskeletal pain in patients with type 2 diabetes. Diabetes Res Clin Pract. 2012;96(2):135–40.
24. Arkkila P, Kantola IM, Viikari J, Rönnemaa T. Shoulder capsulitis in type I and II diabetic patients: association with diabetic complications and related diseases. Ann Rheum Dis. 1996;55(12):907–14.
25. Sarkar P, Pain S, Sarkar R, Ghosal R, Mandal S, Banerjee R. Rheumatological manifestations in diabetes mellitus. J Indian Med Assoc. 2008;106(9):593–4.
26. Banon S, Isenberg D. Rheumatological manifestations occurring in patients with diabetes mellitus. Scand J Rheumatol. 2013;42(1):1–10.
27. Robinson C, Seah KM, Chee Y, Hindle P, Murray I. Frozen shoulder. J Bone Joint Surg Br Vol. 2012;94(1):1–9.
28. Vessey M, Villard-Mackintosh L, Yeates D. Epidemiology of carpal tunnel syndrome in women of childbearing age. Findings in a large cohort study. Int J Epidemiol. 1990;19(3):655–9.
29. Chammas M, Bousquet P, Renard E, Poirier J-L, Jaffiol C, Allieu Y. Dupuytren's disease, carpal tunnel syndrome, trigger finger, and diabetes mellitus. J Hand Surg. 1995;20(1):109–14.
30. Mehuys E, Van Bortel L, De Bolle L, Van Tongelen I, Annemans L, Remon JP, et al. Effectiveness of a community pharmacist intervention in diabetes care: a randomized controlled trial. J Clin Pharm Ther. 2011;36(5):602–13.
31. Butt M, Ali AM, Bakry MM, Mustafa N. Impact of a pharmacist led diabetes mellitus intervention on HbA1c, medication adherence and quality of life: a randomised controlled study. Saudi Pharm J. 2016;24(1):40–8.
32. Mazroui A, Rashid N, Kamal MM, Ghabash NM, Yacout TA, Kole PL, et al. Influence of pharmaceutical care on health outcomes in patients with type 2 diabetes mellitus. Br J Clin Pharmacol. 2009;67(5):547–57.
33. Franklin BD, Van Mil J. Defining clinical pharmacy and pharmaceutical care. Int J Clin Pharm. 2005;27(3):137.

International Journal of Diabetes in Developing Countries (2021) 41:108–113
https://doi.org/10.1007/s13410-020-00844-1

ORIGINAL ARTICLE

Determinants of gestational diabetes mellitus: a hospital-based case–control study in coastal South India

Archana Ganapathy[1,2] · Ramesh Holla[3,4] · B. B. Darshan[3,4] · Nithin Kumar[3,4] · Vaman Kulkarni[3,4] · Bhaskaran Unnikrishnan[3,4] · Rekha Thapar[3,4] · Prasanna Mithra[3,4] · Avinash Kumar[3,4]

Received: 13 April 2019 / Accepted: 13 June 2020 / Published online: 23 July 2020

Abstract

Background A public health problem that has been on the rise in the twenty-first century is gestational diabetes mellitus (GDM). There are serious adverse effects on both maternal and fetal health following GDM. Potential complications can be reduced by early detection of risk factors, which predispose women to GDM.

Objectives This study aims to identify the risk factors associated with GDM.

Methods A case–control study was carried out among antenatal women admitted to hospitals affiliated to Kasturba Medical College, Mangalore. The study population consisted of cases, who were GDM patients, and controls, who were age-matched, non-GDM patients. Statistical Package for Social Sciences (SPSS) version 25.0 was used for entering and analysing data. Both univariate and multivariate analysis was done for determining the factors responsible for GDM.

Results The mean age of cases was 29.54 (± 4.3) years and of controls was also 29.54 (± 4.2). There was no significant difference while comparing the socioeconomic status across the study groups. Irregular menstrual cycle (OR = 2.78, CI = 0.94–08.4, $P = 0.06$) and history of type 2 diabetes mellitus in first-degree relatives (OR = 5.26, CI = 2.13–12.99, $P \leq 0.001$) were found to be significant risk factors.

Conclusion It was found in our study that irregular menstrual history, history of GDM in previous pregnancy, history of type 2 diabetes mellitus in first-degree relative and history of GDM in first-degree relative are all independent risk factors of GDM.

Keywords Gestational diabetes mellitus · Case–control study · Risk factors · Multivariate analysis

✉ Ramesh Holla
ramesh.holla@manipal.edu

Archana Ganapathy
archanag2694@gmail.com

B. B. Darshan
darshan.bb@manipal.edu

Nithin Kumar
nithin.gatty@manipal.edu

Vaman Kulkarni
vaman.kulkarni@manipal.edu

Bhaskaran Unnikrishnan
unnikrishnan.b@manipal.edu

Rekha Thapar
rekha.thapar@manipal.edu

Prasanna Mithra
prasanna.mithra@manipal.edu

Avinash Kumar
avinash.kumar@manipal.edu

[1] Kasturba Medical College, Mangalore, India

[2] Manipal Academy of Higher Education, Manipal, India

[3] Department of Community Medicine, Kasturba Medical College, Mangalore, India

[4] Faculty of Health Sciences, Manipal Academy of Higher Education, Manipal, India

Introduction

A public health problem that has been on the rise in the twenty-first century is gestational diabetes mellitus (GDM). It can be defined as any degree of glucose intolerance with onset or first recognition during pregnancy [1]. It can be classified into G1, where women have normal blood glucose levels but impaired glucose tolerance and G2, where women in both fasting and postprandial states have hyperglycemia [2]. The pathophysiology behind this disease is the decreased action of insulin due to above-average levels of insulin resistance that sets in as a result of placental hormone secretion during pregnancy [2].

Globally, the median estimate of GDM is at approximately 6–13% of all pregnancies [3]. In India, the prevalence is expected to rise from 40.9 million in 2009 to an alarming 69.9 million in 2025 [2] while in South India a rise has already been noted from 1% in 1998 [4] to 14.6% in 2016 [5].

There is a pressing need that has arisen to address the cause of this rise in GDM and a few risk factors, both modifiable and non-modifiable, have been identified. Non-modifiable risk factors include advanced maternal age, family history of diabetes mellitus in a first-degree relative and a personal history of GDM. Modifiable risk factors are body mass index (BMI), diet, physical activity and cigarette smoking [4]. Other factors are history of stillbirth, treatment for infertility, unexplained neonatal death, polyhydramnios, delivery of a large infant (> 4 kg), prematurity and pre-eclampsia in a multipara [1].

There are serious adverse effects on both maternal and fetal health following GDM. The maternal complications are increased risk for preeclampsia, cardiovascular complications, maternal infections [6], premature birth, increased risk of cesarean section [3] and also greater chances (20–50%) of developing type 2 diabetes mellitus in 5 years after delivery [6]. In fetal complications, there is a likelihood of infants developing diabetes mellitus, metabolic syndrome, obesity, cardiovascular disease increase and furthermore, they may have higher chances of developing childhood leukemia [4]. These potential complications can be reduced by early detection of risk factors, which predispose women to GDM. Currently, the screening for this is done around 24–28 weeks of gestational age [1].

In developing countries, there has been a shift of pattern from communicable to non-communicable diseases so as much research needs to be done in order to study the pattern of these diseases and accordingly manage them. Thus, this current study was carried out in one of the coastal districts of Southern India to identify the risk factors involved in GDM.

Materials and methods

Design

A case–control study was carried out among antenatal women admitted to tertiary care teaching hospitals affiliated to Kasturba Medical College, Mangalore over a period of 5 months.

Setting

The study population consisted of cases, who were GDM patients, and controls, who were age-matched, non-GDM patients. Participants from both groups were taken from the outpatient department of the aforementioned hospitals and gave their informed consent to participate in the study. In these hospital settings, women were diagnosed to be GDM based on the American College of Obstetrics and Gynecologists (ACOG) guidelines. A glucose challenge test was performed at 24 weeks, and if the blood glucose levels at 2 h following consumption of 50 g glucose (irrespective of fasting state) exceeded > 140 mg/dl, an oral glucose tolerance test was performed next. In this, the patient consumed 75 g glucose after an overnight fast of 8–10 h and blood glucose levels were measured in a fasting state, at 1 h, 2 h and 3 h following consumption. If any two of the four values (fasting ≥ 95 mg/dl, 1-h blood glucose ≥ 180 mg/dl, 2-h blood glucose ≥ 155 mg/dl, 3-h blood glucose ≥ 140 mg/dl) were seen as abnormal (as per the Carpenter and Coustan criteria), the woman was diagnosed to have GDM [7].

The calculation of the sample size was based on the findings of a previous case–control study wherein 37.9% of the cases and 14.3% of the controls had a pre-pregnant BMI of ≥ 25 with an odds ratio of 3.7 [1]. Taking an alpha error of 5% and a power of 90% with 1:1 ratio of cases and controls, the sample size was calculated to be 70 in each arm.

Data collection and analysis

The tool used for data collection was a questionnaire prepared after referring to the published literature on GDM and the medical records of GDM patients admitted to the hospitals. In this, section 1 consisted of all the sociodemographic details and the socioeconomic status was classified based on a modified B.G. Prasad Scale [8]. Section 2 of the questionnaire consisted of menstrual history, obstetric details and anthropometric measurements, while section 3 consisted of the patient's family history and any other details. During the study, an interview was conducted using this pretested questionnaire and the necessary anthropometric measurements were done.

Statistical Package for Social Sciences (SPSS) version 25.0 was used for entering and analysing data. Descriptive statistics like proportion, mean, median, and standard deviation was

used for expressing the results. χ^2 test was done for comparing the qualitative data between cases and controls. Both univariate and multivariate analysis (binary logistics regression) was done for determining the factors responsible for GDM. p value < 0.05 was considered as statistically significant. Only the factors found to be significant on univariate analysis were then considered for multivariate analysis. Unadjusted and adjusted odds ratio (OR) and corresponding 95% confidence intervals (CI) were also reported.

Results

While comparing the baseline characteristics as shown in Table 1, the mean age of cases was 29.54 (± 4.3) years and of controls was also 29.54 (± 4.2) years. The proportion of Hindus, Muslims and Christians among the cases and controls was almost the same and majority of the women in both cases (81.4%) and controls (94.2%) were engaged as unskilled labour. There was no significant difference while comparing the socioeconomic status across the study groups. The literacy rate among the cases and controls was comparable and showed no noteworthy variance.

Table 2 shows that on univariate analysis, irregular menstrual cycle ($p = 0.04$), history of exercise ($p = 0.01$), history of GDM in previous pregnancy ($p < 0.001$), history of type 2 diabetes mellitus ($p < 0.001$) and history of GDM ($p = 0.01$) in first-degree relatives were all found to be significant determinants contributing to GDM.

With respect to the regularity of the menstrual cycle, 21.4% of the cases presented with irregularity in menstrual cycle compared to a meagre 8.6% in the controls. It is also interesting to note that while 17.1% of the cases had a history of previous GDM and 10% of them gave a history of GDM among their first-degree relatives, none of the controls had such a history in both categories. On further comparing the family history among the two groups, 44.3% of the cases had a history of type 2 diabetes mellitus in the first-degree relative while only 11.4% of the controls had such a history.

In terms of exercise, 18.6% of the cases exercised while only 2.9% of the controls did so, but it is worth noting that the questions asked were regarding their current and not previous exercise regime, if any.

On further multivariate analysis, as depicted in Table 3, irregular menstrual cycle (OR = 2.78, CI = 0.94–08.4, p = 0.06) and history of type 2 diabetes mellitus in first-degree relatives (OR = 5.26, CI = 2.13–12.99, p = <0.001) were found to be significant risk factors for GDM.

Other independent risk factors which were found to be significant determinants of GDM are history of GDM in previous pregnancy ($p < 0.001$) and history of GDM ($p = 0.01$) in first-degree relative.

Table 1 Baseline characteristics of study participants ($N = 140$)

Baseline characteristics	Cases *n* (%)	Controls *n* (%)	*p* value
Age (years)			
20–24	08 (11.4)	08 (11.4)	
25–29	28 (40.0)	28 (40.0)	
30–34	22 (31.4)	22 (31.4)	
≥ 35	12 (17.1)	12 (17.1)	
Mean age (years)	29.54 (±4.3)	29.54 (±4.2)	
Religion			0.93
Hindu	52 (74.3)	52(74.3)	
Muslim	14 (20.0)	13(18.6)	
Christian	04 (05.7)	05 (07.1)	
Occupation			0.14
Unskilled	57(81.4))	66 (94.2)	
Semiskilled	07 (10.0)	02(02.9)	
Skilled	06 (08.57)	02 (02.9)	
Education			0.24
Illiterate	05 (07.2)	02 (02.9)	
Literate	65 (92.8)	68 (97.1)	

Discussion

Gestational diabetes mellitus is an upcoming problem which has been on the rise in India during the past few years. Considering the implications of GDM on both the mother and the child, this study hopes to identify the risk factors of GDM which may aid in implementing early interventional measures that may in turn reduce its incidence.

One of the important independent risk factors for GDM that we found from our study was the history of irregular menstrual cycle (O.R. = 2.799, C.I. = 0.938–8.353, p = 0.065) where 21.4% of the cases presented with irregular menstrual cycles compared to 8.6% among the controls. The University of Texas conducted a case–control study which corroborates this result as it showed how irregular menstrual cycle is an independent risk factor for GDM (24% vs. 7%, p = 0.006) [9]. Another possible explanation for this association could be that irregular menstrual cycles are more prevalent among women with PCOS [10, 11]. PCOS in turn has been seen as a potential risk factor for GDM in various studies [12, 13]. The pathophysiology underlying this is the state of insulin resistance induced by PCOS which affects up to 25–70% of women affected by the disease. During pregnancy, these women are further predisposed to insulin resistance, thus increasing their likelihood of developing GDM [14].

Another significant variable which was found to be a risk factor for developing GDM is the previous history of GDM ($p < 0.001$). This result can be substantiated with another age-matched case–control study conducted in Sri Avittom Thirumal Hospital where the history of previous

Table 2 Univariate analysis showing determinants of gestational diabetes mellitus ($N = 140$)

Variables	Study groups		Unadjusted OR (95% CI)	p value
	Cases ($n = 70$)	Controls ($n = 54$)		
Pre-pregnant BMI				
< 25	61 (87.1)	65 (92.9)	1	0.27
≥ 25	9 (12.9)	5 (7.1)	1.92 (0.60–6.04)	
*Menstrual cycle				
Regular	55 (78.6)	64 (91.4)	1	0.04
Irregular	15 (21.4)	6 (8.6)	2.91 (1.06–8.01)	
Gravida				
Primi	31 (44.3)	36 (51.4)	1	0.39
Multi	39 (55.7)	34 (48.6)	1.33 (0.68–2.59)	
*History of exercise				
Yes	13 (18.6)	2 (2.9)	1	0.01
No	57 (81.4)	68 (97.1)	0.12 (0.03–0.59)	
Past abortion				
Yes	10 (14.3)	10 (14.3)	1 (0.388–2.58)	1.0
No	60 (85.7)	60 (85.7)	1	
History of stillbirths				
Yes	01 (1.4)	2 (02.9)	0.493 (0.04–5.56)	0.57
No	69 (98.6)	68 (97.1)	1	
Infertility treatment				
Yes	2 (2.9)	1 (01.4)	2.03 (0.18–22.90)	0.57
No	68 (97.1)	69 (98.6)	1	
*History of GDM in previous pregnancies				
Yes	12 (17.1)	00	–	< 0.001
No	58 (82.9)	70 (100)	–	
Premature delivery in previous pregnancies				
Yes	4 (05.7)	5 (7.1)	0.79 (0.20–3.07)	0.73
No	66 (94.3)	65 (92.9)	1	
Recurrent UTI in previous pregnancy				
Yes	3 (4.3)	2 (2.9)	1.52 (0.25–9.40)	0.651
No	67 (95.7)	68 (97.1)	1	
*History of type 2 diabetes mellitus in first-degree relative				
Yes	31 (44.3)	8 (11.4)	6.16 (2.57–14.77)	< 0.001
No	39 (55.7)	62 (88.6)	1	
*History of GDM in first-degree relative				
Yes	7 (10)	0	–	0.01
No	63 (90)	70 (100)	–	

*Determinants which contribute to gestational diabetes mellitus (p values < 0.05)

GDM ($p = 0.035$) proved to be a significant determinant for developing GDM in the current pregnancy [1]. Interestingly, another retrospective study [15] and a case–control study [16] conducted among the Chinese-Asian populations also showed similar results with a high recurrence rate of 73.1% and 55.0%, respectively, among pregnant women who were diagnosed with GDM in their first pregnancy. Various other studies conducted in South

Table 3 Multivariate analysis showing risk factors for gestational diabetes mellitus ($N = 140$)

Variables	Unadjusted OR (95% CI)	p value	Adjusted OR (95% CI)	p value
Menstrual cycle	2.91 (1.06–8.01)	0.04	2.78 (0.94–08.4)	0.06
History of exercise	0.12 (0.03–0.59)	0.01	0.15 (0.03–0.73)	0.02
History of type 2 diabetes mellitus in first-degree relatives	6.16 (2.57–14.77)	< 0.001	5.26 (2.13–12.99)	< 0.001

Indian states, that is, in Karnataka [17] and Kerala [18], have similar findings. This recurrence of GDM could be due to a shared risk factor in repeated pregnancies [19] and other additive factors in between two pregnancies such as increased inter-pregnancy weight gain [15].

History of type 2 diabetes mellitus in a first-degree relative was found to be an independent risk factor for GDM with 44.3% of the cases but only 11.4% of controls having such a history (OR = 6.160, CI=2.13-12.99, $p = 0.000$). A cross-sectional study in Peru showed similar results where women with a family history of diabetes were 1.5 times more prone for developing GDM (OR = 1.51, 95% CI = 1.10–2.07) than those women who did not [3]. Other studies in Kerala (37.3% cases vs. 12.0% controls having history of GDM in first-degree relative) [1] and Tamil Nadu (positive family history of diabetes in 57.87% of women with GDM) [6] also show the same. Furthermore, a history of GDM in the first-degree relative ($p = 0.007$) was also found to be associated with GDM in current pregnancy as can be seen in another study in Kollam, Kerala. Here, the prevalence of a family history of GDM was 41% among the GDM women [20]. In fact, while 10% of the cases in our study had a family history of GDM in their first-degree relative, none of the controls had the same. Thus, there seems to be a strong genetic component in the development of GDM with both family history of GDM and type 2 Diabetes mellitus in first-degree relative playing a role as risk factors. This could be due to the inheritance of a genetic insulin secretory defect which increases the predisposition for developing GDM [19].

The importance of lifestyle interventions such as exercise in reducing the incidence of GDM has been established in various studies. In fact, The Royal College of Obstetricians and Gynecologists recommend 30 min of aerobic exercise at least four times a week [21]. Inversely, lack of exercise is an important risk factor for GDM which has been highlighted in studies like Kollam where among the GDM women 83.33% were not engaging in any exercises regularly [20]. An unmatched case–control study in Ethiopia showed how the risk of developing GDM decreased by 97% following routine physical exercise [19]. Another study published in the American Diabetic Association showed that increased physical activity before or during early pregnancy was related to a significantly lower risk of GDM [22]. This is possibly because exercise increases the stimulation for insulin secretion [19, 21]. In our study, however, there was an inverse relationship between exercise and history of GDM where 18.6% of the cases exercised while only 2.9% of the controls did so. This might be because most of the cases obtained from the wards had already been diagnosed with GDM and admitted, so they were recommended by the clinicians to perform mild exercises like walking immediately after meals in order to reduce their post-prandial glucose levels [18]. Thus, many cases adopted the practice of exercising while the controls did not, hence exercise was seen as associated with GDM.

It is interesting to note that pre-pregnancy BMI, which has been established as a risk factor for GDM in several studies [15, 23], showed no significant association in our research. A study was conducted in Urban Tanzania among 743 participants to find out if the risk factors for type 2 diabetes well established among the Western populations also applied to a population where majority fall in the lower socioeconomic strata (Urban Tanzania) [24]. It was found that BMI was not an independent risk factor for developing type 2 DM here. Furthermore, a cross-sectional study conducted with 5100 North Indian women from Punjab [25] and a prospective study conducted with 240 Thai women [26] showed that pre-pregnancy BMI was not a risk factor for developing GDM. This could be explained by evaluating the pathophysiology behind the development of GDM among these lower socioeconomic groups. A genetically decreased beta-cell function or hepatic insulin resistance, rather than increased peripheral insulin resistance associated with obesity [24], could be leading to the development of GDM. This is in concordance with our study where majority of the population belonged to the lower socioeconomic strata, and a high pre-pregnancy BMI was not found to be a risk factor for developing GDM.

Conclusion

It was found in our study that irregular menstrual history, history of GDM in previous pregnancy, history of type 2 diabetes mellitus in first-degree relative and history of GDM in first-degree relative are all independent risk factors of GDM.

Author contribution 1.2. conceptualized the study design and carried out the experiment, including data acquisition. 1.2.3. Carried out the data analysis and contributed to interpretation of the results. 6. Conceived the original idea and supervised the project. 1.2.3 wrote the manuscript and 4.5.6.7.8.9. Reviewed and edited the manuscript. 1.2.3.4.5.6.7.8.9 have seen and approved the final version of the manuscript and all the subsequent versions.

Compliance with ethical standards Approval was sought from the Institutional Ethics Committee of Kasturba Medical College before commencement of the study. During the study, informed written consent was taken from all participants after clearly explaining the study objectives in a language known to them. If the participant was unable to read/write, the informed consent was taken from a Legally Authorized representative or next of kin.

Ethical approval Prior to the commencement of the study, the study protocol was submitted for approval to the Institutional Ethics Committee (IEC) of Kasturba Medical College. Thereafter, permission was obtained from the Medical Superintendents of the concerned hospitals for conduction of the study. Study objectives were clearly explained to the participants in a language familiar to them. Anonymity and

discretion of the information given by the patients were maintained with utmost care and a written informed consent was obtained from the participants.

Conflict of interest The authors declare that they have no competing interests.

References

1. Bhat M, Ramesha KN, Sarma SP, Menon S, Sowmini CV, Ganesh Kumar S. Determinants of gestational diabetes mellitus: a case control study in a district tertiary care hospital in South India. Int J Diabetes Dev Ctries. 2010;30(2):91–6.
2. Lakshmi D, Felix AJW, Devi R, Manobharathi M. Study on knowledge about gestational diabetes mellitus and its risk factors among antenatal mothers attending care, urban Chidambaram. Int J Commun Med Public Health. 2018;5(10):4388–92.
3. Larrabure Torrealva GT, Martinez S, Luque Fernandez MA, Sanchez SE, Mascaro PA, Ingar HA, et al. Prevalence and risk factors of gestational diabetes mellitus: findings from a universal screening feasibility program in Lima, Peru. BMC Pregnancy Childbirth. 2018;18(1):303.
4. Carey Rubin R 2016 Gestational diabetes mellitus—risk factors and screening. today's dietitian, [online] available at: <https://www.todaysdietitian.com/pdf/courses/CareyGDM.pdf> [Accessed 19 Mar 2020].
5. Agrawal S, Das V, Agarwal A, Pandet A, Namrata. Prevalence of gestational glucose intolerance and gestational diabetes in a tertiary care centre in Northern India. J Clin Diagn Res. 2018;12(8):QC04–6.
6. Sivakumar V, Rajasekeran AM, Vijayakumar A. Assessment of risk factors for the early detection of gestational diabetes mellitus. Int J Pharm Sci Res. 2014;5:114–7.
7. Karagiannis T, Bekiari E, Manolopoulos K, Paletas K, Tsapas A. Gestational diabetes mellitus: why screen and how to diagnose. Hippokratia. 2010;14(3):151–4.
8. Mangal A, Kumar V, Panesar S, Talwar R, Raut D, Singh S. Updated BG Prasad socioeconomic classification, 2014: a commentary. Indian J Public Health. 2015;59:42–2.
9. Haver MC, Locksmith GJ, Emmet E. Irregular menses: an independent risk factor for gestational diabetes mellitus. Am J Obstet Gynecol. 2003;188:1189–91.
10. Norman RJ, Dewailly D, Legro RS, Hickey TE. Polycystic ovary syndrome. Lancet. 2007;370(9588):685–97.
11. Waldman IN, Legro RS. Chapter 26—polycystic ovary syndrome. The Ovary (Third Edition); 2019. p. 415–35.
12. Khomami MB, Joham AE, Boyle JA, Piltonen T, Silagy M, Arora C, et al. Increased maternal pregnancy complications in polycystic ovary syndrome appear to be independent of obesity—a systematic review, meta-analysis, and meta-regression. Obes Rev. 2019;20(5):659–74.
13. Oviya C, Mohanraj U, Sathya N, Deeksha N. The study of gestational diabetes mellitus among pregnant women with and without polycystic ovary syndrome—cohort study. Paripex - Indian J Res. 2019;8(4).
14. Boomsma CM, Fauser BCJM, Macklon NS. Pregnancy complications in women with polycystic ovary syndrome. Semin Reprod Med. 2008;26(1):072–84.
15. Barnes RA, Wong T, Ross GP, Griffiths MM, Smart CE, Collins CE, et al. Excessive weight gain before and during gestational diabetes mellitus management: what is the impact? Diabetes Care. 2019;43(1):74–81.
16. Wang YY, Liu Y, Li C, Lin J, Liu XM, Sheng JZ, et al. Frequency and risk factors for recurrent gestational diabetes mellitus in primiparous women: a case control study. BMC Endocr Disord. 2019;19:22.
17. Gangadhara Goud T, Pavan Kumar K, Ramesh K. Risk factors of gestational diabetes in Karnataka. Int J Curr Res Acad Rev. 2014;2(9):286–91.
18. Alexander B. After-meals walks may help control diabetes, study suggests. NBC news [Internet]. 2013 12 [cited Oct 13 2016]. Available from: http://www.nbcnews.com/health/after-meal-walks-may-help-control-diabetes-study-suggests-6C10287692. Accessed 27 Mar 2018.
19. Feleke BE. Determinants of gestational diabetes mellitus: a case–control study. J Matern Fetal Neonatal Med. 2018;31(19):2584–9.
20. Sreekanthan K, Belicita A, Rajendran K, Vijayakumar A. Prevalence of gestational diabetes mellitus in a medical college in South India: a pilot study. Indian J Clin Pract. 2014;25(4).
21. Ming WK, Ding W, CJP Z, Zhong L, Long Y, Li Z, et al. The effect of exercise during pregnancy on gestational diabetes mellitus in normal-weight women: a systematic review and meta-analysis. BMC Pregnancy Childbirth. 2018;18(440).
22. Tobias DK, Zhang C, van Dam RM, Bowers K, Hu FB. Physical activity before and during pregnancy and risk of gestational diabetes mellitus. Diabetes Care. 2011;34(1):223–9.
23. Muller PS, Nirmala M. Effects of pre-pregnancy maternal body mass index on gestational diabetes mellitus. Int J Eng Technol. 2018;7(1.9):279–82.
24. Faurholt-Jepsen D, Range N, PrayGod G, Jeremiah K, Faurholt-Jepsen M, Aabye MG, et al. The association between conventional risk factors and diabetes is weak among urban Tanzanians. Diabetes Care. 2014;37(1):e5–6.
25. Arora GP, Thaman RG, Prasad RB, Almgren P, Brøns C, Groop LC, et al. Prevalence and risk factors of gestational diabetes in Punjab, North India: results from a population screening program. Eur J Endocrinol. 2015;173(2):257–67.
26. Kongubol A, Phupong V. Prepregnancy obesity and the risk of gestational diabetes mellitus. BMC Pregnancy Childbirth. 2011;11:59.

International Journal of Diabetes in Developing Countries (2021) 41:114–120
https://doi.org/10.1007/s13410-020-00859-8

ORIGINAL ARTICLE

Maternal age at pregnancy and risk for gestational diabetes mellitus among Chinese women with singleton pregnancies

Yating Han[1] · Mingkun Tong[2,3] · Lei Jin[4] · Jinhui Yu[2,3] · Wenying Meng[4] · Aiguo Ren[2,3] · Lei Jin[2,3]

Received: 23 September 2019 / Accepted: 4 August 2020 / Published online: 22 August 2020

Abstract

Objectives Maternal age at pregnancy is one of the most important risk factors for gestational diabetes mellitus (GDM); the particulars of the association vary by racial origin. Women less than 25 years old are considered to have low risk by the American Diabetes Association, but there are little data to support this among Chinese women. The aim of this study was to explore the relationship of maternal age and the incidence of GDM.

Methods The data were drawn from a prenatal healthcare system and clinical record: 15,668 singleton pregnancies in women who had no pre-pregnancy diabetes and who became pregnant naturally. The relationships of age and GDM incidence were examined using χ^2 and logistic regression models.

Results The overall incidence of GDM was 22.72% (95% CI, 22.07–23.38). The incidence increased from 10.21% (95% CI, 8.18–12.14) in the age group of 18–22 years to 37.10% (95% CI, 33.71–40.49) in the age group of 36–49 years. The risk of GDM increased by an average of 8% for every 1 year of maternal age, and within each age group, the risk of GDM was 5% higher in primiparas than in pluriparas, in the range of age of 22 and 35 years.

Conclusions for practice The incidence of GDM increased with maternal age. Women who got pregnant younger than 23 years had the lowest risk, followed by those who were younger than 30 years. The incidence GDM was especially high in women who were primiparas and were older than 30 years.

Keywords Pregnancy · Woman · Gestational diabetes mellitus (GDM) · Age · Parity · Primiparas · Pluriparas

Yating Han and Mingkun Tong have equal authorship.

Significance Previous studies have found that maternal age at pregnancy is one of the most important risk factors for GDM. Women less than 25 years old are considered to have low risk by the American Diabetes Association, but there are little data to support this among Chinese women. We explored the association between maternal age and risk for GDM in China and found: the incidence of GDM increased with maternal age at pregnancy, and the risk for GDM increased by an average of 8% for every year of maternal age between 23 and 36 years old; for Chinese women, reproduction at a younger age may reduce the incidence of GDM; for prevention GDM in China, primiparity should be encouraged at younger ages than 23 years old or at least not older than 30 years old.

✉ Lei Jin
jinlei@bjmu.edu.cn

[1] Peking University People's Hospital, Beijing, China

[2] Institute of Reproductive and Child Health/National Health Commission Key Laboratory of Reproductive Health, Peking University, No. 38 Xueyuan Road, Haidian District, Beijing 100191, China

[3] Department of Epidemiology and Biostatistics, School of Public Health, Peking University, Beijing, China

[4] Tongzhou Maternal and Child Health Hospital, Tongzhou District, Beijing, China

Introduction

Gestational diabetes mellitus (GDM) is defined as glucose intolerance that first occurs or is first identified during pregnancy [1]. Maternal GDM is related to short-term and long-term adverse outcomes for both mothers and their offspring [2]. The incidence of GDM has been increasing worldwide [3, 4], including in China [5]. One out of every five pregnant women in Beijing had either GDM or diabetes during pregnancy in 2013, based on data from 15 hospitals [6].

Maternal age at pregnancy is one of the most important risk factors for GDM [7–9]. In a previous study, rates of GDM were greater in older women born in all of the regions studied [10]. Women less than 25 years old are considered to have low risk by the American Diabetes Association [11], but there are little data to support this among Chinese women. A prospective population-based study conducted in China in 2010–2012 found that older age at pregnancy was associated with increasing incidence of GDM [12].

In 2016, Chinese family planning policy was changed from a one-child to a two-child policy, which may lead to more advanced maternal ages and larger numbers of pluriparas. The association between maternal age and risk for GDM should be examined to determine whether the incidence of GDM will increase further as the new family policy takes effect, allowing women to be able to choose the optimal age for becoming pregnant, in terms of GDM prevention.

We explored the incidence of GDM for different age groups and the difference between GDM incidence in primiparas and pluriparas to be able to inform the public of the optimal age for women to become pregnant, with the aim of lowering GDM risk.

Methods

Data sources

The data were drawn from the prenatal healthcare system and the clinical record at Tongzhou Maternal and Child Health Hospital and three community hospitals in the Tongzhou District of Beijing. The prenatal healthcare system is held on a computer network that connects community clinics and birthing hospitals in the district. Women who live in the district are advised to visit any hospital for a clinical pregnancy diagnosis 6 to 12 weeks after last menstrual period (LMP). If a pregnancy is confirmed, the pregnant woman must go to a community clinic to register within 30 days of the diagnosis. At registration, a unique 14-digit prenatal healthcare code is given to her for the pregnancy. A maternal booklet is issued with the prenatal code and the ID numbers of the woman and the husband (if available). Information on demographic characteristics, use of folic acid supplements, and type of supplement (whether folic acid only or multivitamins containing folic acid) used during the peri-conceptional period, personal obstetric and gynecological history, and weight before LMP are collected and recorded to be entered into the network system. Height and weight are measured in the clinic as well. Later, the woman can choose her birthing hospital and complete her prenatal care there through delivery. Information on prenatal care is entered into the system in real time at the birthing hospital. Using the network, community doctors can check each pregnant woman's status each day. When the woman is found to have delivered at a hospital, they visit her and her child at their home within 7 days of delivery. The statistical year in the healthcare system is defined as extending from October 1 to September 30.

Participants

In 2015 and 2016, 16,041 women received prenatal examinations and gave birth at Tongzhou Maternal and Child Hospital and three community hospitals in Tongzhou. Among these, 202 women had twins, 95 became pregnant using assisted reproductive technology, 53 had pre-pregnancy diabetes, and 23 were under 18 years old. Thus, 15,668 pregnancies in total were included in the study (Fig. 1).

Diagnostic criteria for GDM

According to the Chinese guidelines for pre-prenatal and prenatal care published in 2011 [13], all pregnant women registered in the prenatal healthcare system should receive fasting blood glucose tests at their first prenatal healthcare examination and a 75-g oral glucose tolerance test (OGTT) at 24–28 gestational weeks. GDM was diagnosed when any of the following criteria were met, according to the July 2011 guidelines of the Chinese Ministry of Health [13]: fasting plasma glucose ≥ 5.1 mmol/L, at 1 h ≥ 10.0 mmol/L, or at 2 h ≥ 8.5 mmol/L. Pre-pregnancy diabetes mellitus was diagnosed when a diagnosis of diabetes mellitus had been made before the beginning of the current pregnancy or when blood glucose levels were > 7.0 mmol/L at the first health examination (before 12 weeks of gestation), ≥ 11.1 mmol/L at 2 h after consumption of glucose in a 75-g OGTT, or when casual blood glucose levels were ≥ 11.1 mmol/L, with typical manifestations of high glucose levels [14].

Statistical analysis

GDM incidence and its 95% confidence interval (CI) were calculated using a binomial distribution model. The relationships between age or parity and GDM incidence were tested using χ^2 tests and χ^2 trend tests. ORs and their 95% CIs were used to estimate the association between age or parity and risk for GDM in a univariate logistic regression model.

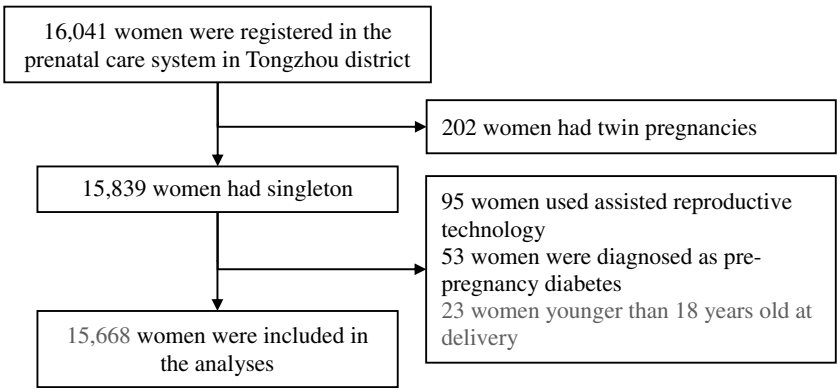


Fig. 1 Flow chart showing inclusion and exclusion of participants. Pregnant women were registered in the perinatal healthcare system and received medical care at a maternal and child hospital and three community hospitals in Tongzhou of Beijing, China, 2015–2016

Multivariable logistic regression models were used to control potential confounders (including ethnicity, education level, occupation, pre-pregnancy BMI, and folic acid or multiple micronutrients that included folic acid supplementation). The predicted incidence of GDM was calculated using a multivariate logistic model for each age group. For ORs and predicted incidence in each age group, age was used as a category variable. The rate ratio was calculated using the incidence rate in the current year in the age group divided by the incidence rate in the previous year of the age group to indicate changes in GDM incidence by year of age. The average rate ratio was $\exp.(\hat{a})^{-1}$, and the $\exp.(\hat{a})$ of the measurement variable was obtained using a univariate or multivariate logistic regression model.

Results

Participant characteristics

During the statistical years 2015 and 2016, a total of 16,041 pregnant women were registered and gave birth at Tongzhou Maternal and Child Health Hospital and the three other township hospitals being studied. After 202 women with twins, 95 women who used assisted reproductive technology, and 53 women with pre-pregnancy diagnoses of diabetes, and 23 women under 18 years old were excluded, 15,668 women were incorporated into the study.

The characteristics of the women in the study are shown in Table 1.

Incidence of GDM and crude OR

The overall incidence of GDM was 22.72% (95% CI, 22.07–23.38) (Table 2). The rate ratio for each age was between 1.02 and 1.67, with the exception of 27-year-olds, whose rate ratio was 0.98.

The crude ORs between maternal age and GDM incidence are presented in Table 2. The incidence of GDM increased with age (trend $\chi^2 = 259.11, p = 0.00$). Compared with women younger than 23 years old, the ORs of 23 years or older

Table 1 Characteristics of the women in the study

Characteristics	*n* (%)
Delivery year	
2015	7102 (45.3)
2016	8566 (54.7)
Age	
18–24	2514 (16.0)
25–29	7479 (47.7)
30–34	4424 (28.2)
35–49	1251 (8.0)
Ethnicity	
Han	14,765 (94.2)
Others	903 (5.8)
Education level[a]	
Middle school or less	5593 (35.7)
High school	9733 (63.4)
Occupation[b]	
Office work	1623 (10.4)
Managers or professional skill worker	2864 (18.3)
Commerce	2568 (16.4)
Others	4640 (29.6)
Unemployed	3243 (20.7)
Pre-pregnancy BMI[c]	
< 18.5	1720 (11.0)
18.5–23.9	8793 (56.1)
24–27.9	2524 (16.1)
≥ 28	848 (5.4)
Parity[d]	
0	10,139 (64.7)
≥ 1	5525 (35.3)
Folic acid or multiple micronutrient supplementation	
Yes	13,949 (89.0)
No	1719 (11.0)

[a] Missing 342 cases; [b] missing 730 cases; [c] missing 1783 cases; [d] missing 4 cases

Table 2 Incidence of GDM by age and ORs for GDM in age groups compared with the 16- to 22-year-old age group

Age	*N*	GDM				OR	
		n	Incidence	95% CI (lower–upper)	Rate ratio[a]	OR	95% CI (lower–upper)
18–22	852	87	10.21	8.18–12.24	1.00	1.00	
23	712	121	16.99	14.24–19.75	1.66	1.80	1.34–2.42
24	950	167	17.58	15.16–20.00	1.03	1.88	1.42–2.48
25	1436	278	19.36	17.32–21.40	1.12	2.11	1.63–2.73
26	1581	331	20.94	18.93–22.94	1.08	2.33	1.81–3.00
27	1620	331	20.43	18.47–22.40	0.98	2.26	1.75–2.91
28	1617	360	22.26	20.24–24.29	1.09	2.52	1.96–3.24
29	1225	280	22.86	20.51–25.21	1.03	2.61	2.01–3.37
30	1090	254	23.30	20.79–25.81	1.02	2.67	2.05–3.47
31	1039	269	25.89	23.23–28.55	1.11	3.07	2.36–3.99
32	891	237	26.60	23.70–29.50	1.03	3.19	2.44–4.16
33	800	223	27.88	24.77–30.98	1.05	3.40	2.59–4.45
34	604	178	29.47	25.83–33.11	1.06	3.67	2.77–4.87
35	472	155	32.84	28.60–37.08	1.11	4.30	3.21–5.77
36–49	779	289	37.10	33.71–40.49	1.13	5.19	3.98–6.76
Total	15,668	3560	22.72	22.07–23.38		1.08	1.07–1.09

[a] The rate ratio is the incidence rate in the current year of age divided by the incidence rate in the previous age group. $\chi^2 = 281.48$, $p < 0.001$; $\chi^2_{\text{trend}} = 259.11$, $p < 0.001$. The rate ratio was calculated using the incidence rate in the year of age divided by the incidence rate in the previous year of age to indicate the changes in GDM incidence per year of age. The average rate ratio was $\exp(\hat{a})^{-1}$, and the $\exp(\hat{a})$ of the measurement variable was obtained using a univariate or multivariate logistic regression model

women changed from 1.80 (95% CI, 1.34–2.42) to 5.19 (95% CI, 3.98–6.76).

The overall rates of GDM among primiparas and pluriparas were 21.88% (95% CI, 21.07–22.68) and 24.29% (95%CI, 23.16–25.42), respectively (Table 2). Compared with primiparas women, the crude OR of pluriparas was 1.15 (95% CI, 1.06–1.24).

Adjusted ORs and the predicted incidence of GDM

The adjusted ORs for maternal age and risk for GDM incidence are shown in Table 3. Compared with women younger than 23 years old, the adjusted ORs were 1.70 (95% CI, 1.23–2.35) for the 23-year-old and 4.45 (95% CI, 3.29–6.01) for the 36- to 49-year-old age group.

The predicted overall incidence of GDM was 23.54% (95% CI, 22.37–23.71). Its incidence increased with age from 10.98% (95% CI, 10.60–11.36) to 37.11% (95% CI, 33.30–34.46) after adjusting for potential confounders (Table 3).

Compared with primiparas women, the adjusted OR was 0.81 (95% CI, 0.73–0.89) for pluriparas women (Table 3). The rates were higher (by about 5%) for primiparas than for pluriparas within each age group, particularly for those who became pregnant at 30–35 years old (Fig. 2).

Comments

The incidence of GDM found in this study was close to the incidence recently found for Beijing in general [6] and lower than that for the USA [15], Scotland [9], and Korea [16]. Global country-level estimates of the prevalence of hyperglycemia and total diabetes have indicated that the prevalence of all types of diabetes in pregnancy in China is only 7.0% and China is not considered a high-incidence region [17]. This may be due to the time of data collection, screening strategies, or diagnostic criteria [18].

Maternal age at pregnancy has been studied as a factor for GDM in studies of Chinese [6] and other [9, 15, 19] populations. In two previous studies, the ORs for age at pregnancy and incidence of GDM in northern Chinese women were 1.05 to 1.1 for each year increase in age, and the risk for GDM increased by 5.3% to 12% for the same age interval [6, 12]. In this study, we found that from the 23-year-old to the 36- to 49-year-old age group, the average rate ratio of predicted GDM incidence was 1.08; after adjusting for confounders, the risk for GDM increased by 8.0% per year of age, similar to the findings of other studies on Chinese women [6, 12].

Few studies have explored the optimal primiparas age for lowering the incidence of GDM in Chinese women. We found a rate ratio of 1.70 for the 23-year-old compared with the 18-

Table 3 Adjusted predicted incidence of GDM by age group and ORs for GDM in age groups compared with the 18- to 22-year-old age group

	N	GDM women	Adjusted OR[a]		Predicted incidence		
			OR	95%CI (lower–upper)	Incidence	95% CI (lower–upper)	Rate ratio[b]
Age							
18–22	692	76	1.00	–	10.98	10.60–11.36	1.00
23	575	107	1.70	1.23–2.35	18.61	17.97–19.24	1.69
24	752	137	1.65	1.21–2.24	18.22	17.69–18.75	0.98
25	1223	245	1.88	1.42–2.50	20.03	19.59–20.47	1.10
26	1316	285	2.04	1.54–2.70	21.66	21.22–22.09	1.08
27	1364	284	1.95	1.47–2.58	20.82	20.40–21.24	0.96
28	1388	319	2.21	1.67–2.92	22.98	22.55–23.42	1.10
29	1063	247	2.22	1.66–2.96	23.24	20.70–25.78	1.01
30	914	226	2.44	1.82–3.27	24.73	22.74–23.73	1.06
31	831	216	2.65	1.96–3.57	25.99	24.18–25.82	1.05
32	768	204	2.61	1.93–3.52	26.56	25.38–26.61	1.02
33	694	207	3.21	2.37–4.35	29.83	25.92–37.21	1.12
34	516	162	3.45	2.51–4.74	31.40	29.10–30.55	1.05
35	414	138	3.58	2.57–54.99	33.33	30.54–32.55	1.06
36–49	679	252	4.45	3.29–6.01	37.11	32.30–34.36	1.11
Total	13,189	3105	1.08	1.07–1.09	23.54	22.37–23.71	1.07[c]
Parity							
0	10,139	2218	1	–	–	–	–
≥ 1	5525	1342	0.81	0.73–0.89	–	–	–

[a] Multivariate logistic analyses were used to adjust for confounders including age, ethnicity, occupation, education, pre-pregnancy BMI, parity, and folic acid supplementation

[b] The rate ratio was calculated using the incidence rate in the current year of age divided by the incidence rate in the previous year to indicate the changes in GDM incidence year by year. The average rate ratio was $\exp.(\hat{a})^{-1}$, and the $\exp.(\hat{a})$ of the measurement variable was obtained using univariate or multivariate logistic regression models

[c] Average predicted GDM incidence rate ratio for each age from 23 to 36 years old, after adjusting for confounders

to 22-year-old age group in our study population, which is much greater than the ratios among other age groups. The

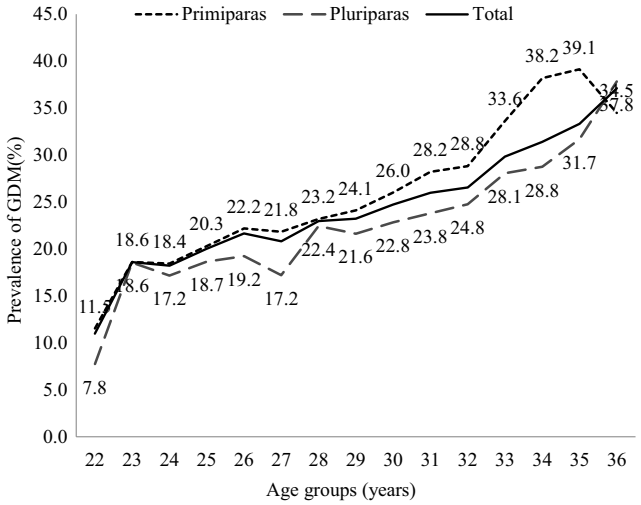


Fig. 2 The incidence of gestational diabetes (GDM) within each age group by parity

ORs for other age groups relative to the 18- to 22-year-old age group extended from 1.65 to 4.45. We found that the incidence of GDM changed with age at pregnancy and a difference between primiparas and pluriparas was observed, namely, the rate was higher (by about 5%) for primiparas than for pluriparas within each age group, particularly in the 30- to 35-year-old age group. Taken together, these data may indicate that the optimal age for Chinese primiparas is younger than 23 years old. A study in Finland also suggested that primiparity should be encouraged at a younger age [20].

During pregnancy, the development of the fetus depends on the appropriate nutrient flow from the mother to the fetus through the placenta, including the flow of glucose [21, 22]. Young β cells are highly proliferative and increase rapidly in number from the prenatal phase until 30 years of age [23]. Nevertheless, adult β cell proliferation can increase during pregnancy [24]. As maternal age advances, insulin secretion normally decreases at a rate of 0.7% per year [25]. The pathogenesis of GDM in aging maternal women may thus be due to age-related insulin resistance and β cell dysfunction, and as

a result, the needs of the fetus and mother cannot be met during the pregnancy.

The incidence of GDM was higher for primiparas than for pluriparas in each age group. We were unable to find a published study that focused specifically on the relationship between maternal parity and risk for GDM. Studies that have touched on the topic have drawn inconsistent conclusions, with most indicating that maternal parity is a protective factor [26, 27]. However, one study found maternal parity to be a risk factor (AOR = 1.78; 95% CI, 1.3–2.49) [28], and another found an association between maternal parity and risk for GDM (OR = 1.27; 95% CI, 0.99–1.64) using a multivariate model [29]. The sample sizes in the three studies listed above were not as large as those of this study, and parity could be different in different populations. Chinese women, because of family planning policy, have a smaller proportion of pluriparas than other populations. In this study, around 65% of women were primiparas, and less than 35% had one or more previous births. Other countries have higher proportions of pluriparas.

Since the Chinese family planning policy was adjusted in 2016, more women of advanced age have become pregnant, making it important to clarify the relationship between maternal age at pregnancy and incidence of GDM. This study focused on the relationship between GDM incidence and maternal age or parity within a large sample in Beijing. The quality of perinatal data from the healthcare system is high, and the GDM screening strategy and process is unified. However, certain factors, such as age of maternal menarche, genetic factors (family diabetes history), dietary habits, and weight gain during the gestational period, were not considered in the multivariate model because of the limitations in the routine data used in this study.

In summary, the incidence of GDM increased with maternal age at pregnancy, and the risk for GDM increased by an average of 8% for every year of maternal age between 23 and 36 years old. For Chinese women, reproduction at a younger age may reduce the incidence of GDM. For prevention, GDM primiparity should be encouraged at younger ages than 23 years old or at least not older than 30 years old.

Funding information The study received funding from the National Key Research and Development Program, Ministry of Science and Technology of the People's Republic of China (Grant Nos. 2016YFC1000500 and 2016YFC1000501). The contents of this manuscript are solely the responsibility of the authors and do not necessarily represent the official views of the Ministry of Science and Technology. The funding agents of the study played no role in its design; in the collection, analysis, or interpretation of data; in the writing of the report; or in the decision to submit the article for publication. The researchers are independent from the funding agents, and all authors, external and internal, had full access to all study data (including statistical reports and tables) and can take responsibility for the integrity of the data and the accuracy of the analyses.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Informed consent The data we used for research was from regular healthcare, for which we did not need the informed consent. The study was approved by the Institutional Review Board of Peking University (IRB00001052-18010).

References

1. World Health Organization. Diagnostic criteria and classification of hyperglycemia first detected in pregnancy. 2nd ed. Geneva: World Health Organization WHO/NMH/MND/13; 2013.
2. Herath H, Herath R, Wickremasinghe R. Gestational diabetes mellitus and risk of type 2 diabetes 10 years after the index pregnancy in Sri Lankan women-a community based retrospective cohort study. PLoS One. 2017;12(6):e0179647. https://doi.org/10.1371/journal.pone.0179647.
3. Chan JC, Cho NH, Tajima N, Shaw J. Diabetes in the Western Pacific Region–past, present and future. Diabetes Res Clin Pract. 2014;103(2):244–55. https://doi.org/10.1016/j.diabres.2013.11.012.
4. Bhat M, Ramesha KN, Sarma SP, Menon S, Sowmini CV, Ganesh Kumar S. Determinants of gestational diabetes mellitus: a case control study in a district tertiary care hospital in South India. Int J Diab Dev Countries. 2010;30(2):91–6. https://doi.org/10.4103/0973-3930.62599.
5. Zhu Y, Zhang C. Prevalence of gestational diabetes and risk of progression to type 2 diabetes: a global perspective. Curr Diab Rep. 2016;16(1):7. https://doi.org/10.1007/s11892-015-0699-x.
6. Zhu WW, Yang HX, Wang C, Su RN, Feng H, Kapur A. High prevalence of gestational diabetes mellitus in Beijing: effect of maternal birth weight and other risk factors. Chin Med J. 2017;130(9):1019–25. https://doi.org/10.4103/0366-6999.204930.
7. Bardenheier BH, Elixhauser A, Imperatore G, Devlin HM, Kuklina EV, Geiss LS, et al. Variation in prevalence of gestational diabetes mellitus among hospital discharges for obstetric delivery across 23 states in the United States. Diabetes Care. 2013;36(5):1209–14. https://doi.org/10.2337/dc12-0901.
8. Wu L, Han L, Zhan Y, Cui L, Chen W, Ma L, et al. Prevalence of gestational diabetes mellitus and associated risk factors in pregnant Chinese women: a cross-sectional study in Huangdao, Qingdao, China. Asia Pac J Clin Nutr. 2018;27(2):383–8. https://doi.org/10.6133/apjcn.032017.03.
9. Collier A, Abraham EC, Armstrong J, Godwin J, Monteath K, Lindsay R. Reported prevalence of gestational diabetes in Scotland: the relationship with obesity, age, socioeconomic status, smoking and macrosomia, and how many are we missing? J Diabetes Investig. 2017;8(2):161–7. https://doi.org/10.1111/jdi.12552.
10. Carolan M, Davey MA, Biro MA, Kealy M. Maternal age, ethnicity and gestational diabetes mellitus. Midwifery. 2012;28(6):778–83. https://doi.org/10.1016/j.midw.2011.08.014.
11. American Diabetes Association. Gestational diabetes mellitus. Diabetes Care. 2004;27(Supplement):S88–90.
12. Leng J, Shao P, Zhang C, Tian H, Zhang F, Zhang S, et al. Prevalence of gestational diabetes mellitus and its risk factors in Chinese pregnant women: a prospective population-based study in Tianjin, China. PLoS One. 2015;10(3):e0121029. https://doi.org/10.1371/journal.pone.0121029.

13. Yang HX. Diagnostic criteria for gestational diabetes mellitus (WS 331-2011). Chin Med J. 2012;125(7):1212–3.
14. Liu X, Chen Y, Zhou Q, Shi H, Cheng WW. Utilization of International Association of Diabetes and Pregnancy Study Groups criteria vs. a two-step approach to screening for gestational diabetes mellitus in Chinese women with twin pregnancies. Diabet Med. 2015;32(3):367–73. https://doi.org/10.1111/dme.12636.
15. Lavery JA, Friedman AM, Keyes KM, Wright JD, Ananth CV. Gestational diabetes in the United States: temporal changes in prevalence rates between 1979 and 2010. BJOG. 2017;124(5):804–13. https://doi.org/10.1111/1471-0528.14236.
16. Chernausek SD, Arslanian S, Caprio S, Copeland KC, El Ghormli L, Kelsey MM, et al. Relationship between parental diabetes and presentation of metabolic and glycemic function in youth with type 2 diabetes: baseline findings from the TODAY trial. Diabetes Care. 2016;39(1):110–7. https://doi.org/10.2337/dc15-1557.
17. Guariguata L, Linnenkamp U, Beagley J, Whiting DR, Cho NH. Global estimates of the prevalence of hyperglycaemia in pregnancy. Diabetes Res Clin Pract. 2014;103(2):176–85. https://doi.org/10.1016/j.diabres.2013.11.003.
18. Liao S, Mei J, Song W, Liu Y, Tan YD, Chi S, et al. The impact of the International Association of Diabetes and Pregnancy Study Groups (IADPSG) fasting glucose diagnostic criterion on the prevalence and outcomes of gestational diabetes mellitus in Han Chinese women. Diabet Med. 2014;31(3):341–51. https://doi.org/10.1111/dme.12349.
19. Karcaaltincaba D, Calis P, Ocal N, Ozek A, Altug Inan M, Bayram M. Prevalence of gestational diabetes mellitus evaluated by universal screening with a 75-g, 2-hour oral glucose tolerance test and IADPSG criteria. Int J Gynaecol Obstet. 2017;138(2):148–51. https://doi.org/10.1002/ijgo.12205.
20. Laine MK, Kautiainen H, Gissler M, Raina M, Aahos I, Jarvinen K, et al. Gestational diabetes in primiparous women-impact of age and adiposity: a register-based cohort study. Acta Obstet Gynecol Scand. 2018;97(2):187–94. https://doi.org/10.1111/aogs.13271.
21. Baeyens L, Hindi S, Sorenson RL, German MS. beta-Cell adaptation in pregnancy. Diabetes Obes Metab. 2016;18(Suppl 1):63–70. https://doi.org/10.1111/dom.12716.
22. Banerjee RR. Piecing together the puzzle of pancreatic islet adaptation in pregnancy. Ann N Y Acad Sci. 2018;1411(1):120–39. https://doi.org/10.1111/nyas.13552.
23. Perl S, Kushner JA, Buchholz BA, Meeker AK, Stein GM, Hsieh M, et al. Significant human beta-cell turnover is limited to the first three decades of life as determined by in vivo thymidine analog incorporation and radiocarbon dating. J Clin Endocrinol Metab. 2010;95(10):E234–9. https://doi.org/10.1210/jc.2010-0932.
24. Butler AE, Cao-Minh L, Galasso R, Rizza RA, Corradin A, Cobelli C, et al. Adaptive changes in pancreatic beta cell fractional area and beta cell turnover in human pregnancy. Diabetologia. 2010;53(10):2167–76. https://doi.org/10.1007/s00125-010-1809-6.
25. Szoke E, Shrayyef MZ, Messing S, Woerle HJ, van Haeften TW, Meyer C, et al. Effect of aging on glucose homeostasis: accelerated deterioration of beta-cell function in individuals with impaired glucose tolerance. Diabetes Care. 2008;31(3):539–43. https://doi.org/10.2337/dc07-1443.
26. Sweeting AN, Wong J, Appelblom H, Ross GP, Kouru H, Williams PF, et al. A novel early pregnancy risk prediction model for gestational diabetes mellitus. Fetal Diagn Ther. 2019;45(2):76–84. https://doi.org/10.1159/000486853.
27. Nombo AP, Mwanri AW, Brouwer-Brolsma EM, Ramaiya KL, Feskens EJM. Gestational diabetes mellitus risk score: a practical tool to predict gestational diabetes mellitus risk in Tanzania. Diabetes Res Clin Pract. 2018;145:130–7. https://doi.org/10.1016/j.diabres.2018.05.001.
28. Feleke BE. Determinants of gestational diabetes mellitus: a case-control study. J Matern Fetal Neonatal Med. 2018;31(19):2584–9. https://doi.org/10.1080/14767058.2017.1347923.
29. Hao M, Lin L. Fasting plasma glucose and body mass index during the first trimester of pregnancy as predictors of gestational diabetes mellitus in a Chinese.

International Journal of Diabetes in Developing Countries (2021) 41:121–126
https://doi.org/10.1007/s13410-020-00845-0

ORIGINAL ARTICLE

Mediating effects of body composition at the first trimester on the occurrence of GDM at the early stage of the third trimester during the advanced maternal age

Liu Jing[1,2] · Zheng Dan[1,2] · Wang Huiqun[3] · Liu Yanping[4] · Lu Yang[1,2] · Yin Xuan[1,2] · Zheng Mengmou[5] · Lou Didong[1]

Received: 23 January 2020 / Accepted: 13 June 2020 / Published online: 28 July 2020

Abstract

Objective To retrospectively analyze the mediating effects of body composition during the first trimester on the occurrence of gestational diabetes mellitus (GDM) in early third trimester in advanced maternal age (AMA).

Methods From December 2017 to May 2018, 1635 singleton pregnant women who underwent delivery in the Guiyang Maternal and Child Health Hospital were selected. The body composition data was measured by using the INBODY770 body composition instrument in the first trimester (mainly at a gestational age of 10–12 weeks). The blood glucose levels at fasting for one night and the cutoff values for them at 1 and 2 h after administration during the 75 g oral glucose tolerance test at gestational age of 24–28 weeks (the early stage of the 3rd trimester) were also collected. According to the Baron and Kenny's criteria (the basic principle of interpreting the intermediary role), multivariate logistic regression was used to analyze the mediating effects of body composition (body fat percentage and protein percentage) between AMA and GDM occurrence.

Results In total, 1635 subjects aged 30.93 ± 4.80 years were selected, and there were 376 women with AMA (23%). There were 983 pregnant women (39.88%) with a body fat percentage greater than 30% and 1331 (81.71%) pregnant women with a protein percentage less than 15%. The results showed significant differences in height, weight, lean body weight, body fat, body fat percentage, and GDM prevalence between AMA and low-age groups ($p < 0.05$). After adjusting height, parity, and AMA, the body fat percentage showed association with GDM (odds ratio, OR 1.85 (1.38 ~ 2.48); confidence interval, CI 95%, $p < 0.05$). After adjusting height, parity, and body weight percentage, the AMA showed association with GDM (OR (CI95%) 2.38 (1.68 ~ 3.37)). Similarly, the protein percentage showed association with GDM (OR (CI 95%) 0.50 (0.31 ~ 0.81), $p < 0.05$).

Conclusion These results confirmed the mediating effects of body fat percentage and protein percentage on AMA and the occurrence of GDM. Advanced age is directly associated with increased risk of GDM, which in turn increases with higher percentage of body fat and lower percentage of protein.

Keywords Gestational diabetes mellitus · Advanced maternal age · Body composition · Body fat · Protein

Introduction

Gestational diabetes mellitus (GDM) refers to abnormal glucose metabolism during pregnancy, but this excludes pre-pregnancy diabetes, Type I or Type II diabetes mellitus [1]. This might in turn lead to adverse pregnancy outcomes, such as cesarean section and hypoglycemia in newborns, as well as increased long-term risks of obesity, type II diabetes, and

Liu Jing and Zheng Dan contributed equally to this work.

✉ Lou Didong
15761879@qq.com

[1] School of Basic Medicine, Guizhou University of Traditional Chinese Medicine, Guiyang 550000, Guizhou, People's Republic of China

[2] Guiyang Maternal and Child Health Hospital, Guizhou, People's Republic of China

[3] Department of Nutrition and Food Hygiene, Guizhou Medical University, Guizhou, People's Republic of China

[4] Peking Union Medical College Hospital, Beijing, People's Republic of China

[5] Department of Preventive Medicine, Sichuan University, Sichuan, People's Republic of China

metabolic syndrome in filial cells [2]. Advanced maternal age (AMA) was defined as pregnant women of age ≥ 35 [3]. AMA is regarded as a risk factor of GDM [4], and the proportion of it has been gradually increased with the issuance of two-child policy in China [5]. Several clinical observations have revealed that AMA population is associated with a relatively higher body fat percentage and lower protein percentage, and so it is assumed that the AMA population is associated with higher risk of GDM. Therefore, this study aimed to explore the mediating effects of body composition, such as body fat percentage and protein percentage, in the first trimester of AMA population and the occurrence of GDM at early third trimester by analyzing the clinical data and body composition data in singleton pregnant women.

Subjects and methods

Subjects

Ethical clearance and informed consent

This study was conducted under the surveillance of Guizhou University of Chinese Traditional Medicine and Women Healthcare Department of Guiyang Maternal and Child Health Hospital, in the People's Republic of China. The study protocol was approved by the ethics committee of Guiyang Maternal and Child Health Hospital. Informed consent was obtained from women who participated.

Singleton pregnant women who underwent delivery at Guiyang Maternal and Child Health Hospital from December 2017 to May 2018 were selected for this study, to investigate the basic information of age, height, body weight, lean body weight (LBW), body fat, body fat percentage, protein percentage, and GDM.

Inclusion criteria were as follows: women (1) with singleton pregnancy, also including a history of gynecologic oncology, (2) who voluntarily participated for body composition testing, and (3) who underwent 75 g oral glucose tolerance test (OGTT) at the early stage of the third trimester.

Exclusion criteria were as follows: women (1) with type I or type II diabetes before pregnancy, (2) who lacked the body composition data tested in the first trimester, and (3) who lacked the data of 75 g OGTT.

Instrument

The body composition analyzer (including height and weight measurement) used was the INBODY770 (Biospace Co, Korea).

Data acquisition and methods

Clinical data

The basic information of age and height during the first trimester.

Instrument testing

The body composition was measured in strict accordance with the operating procedure of the INBODY770 body composition analyzer. Pregnant women with empty stomach were instructed to wear casual clothes and followed by emptying of urine, taking off their shoes and socks, and wiping their hands and feet with electrolytic wet tissue. The body weight analyzer that comes with the INBODY770 body composition analyzer was used for measuring the weight. The pregnant women were then instructed to stand on the foot-shaped electrode, holding the handle, with the thumb on the upper part of the handle and the remaining fingers on the lower part of the handle, keeping their arms straight and away at a certain distance from their torso, and this posture was followed by the instruction of the measurement. The following indicators such as the body weight, lean body weight, body fat, and protein were measured.

GDM diagnostic criteria

GDM was diagnosed according to the criteria established by the Obstetrics and Gynecology (9th edition) [6]. Firstly, blood glucose levels of the pregnant women with fasting for one night at the early stage of the third trimester were measured, then underwent OGTT (75 g glucose was dissolved in 250 ml warm water, drank within 5 min).The cutoff values for blood glucose levels at 1 h and 2 h after drinking were measured. If any values met or exceeded items of the blood glucose value standards, 5.1 (empty stomach), 10.0 (at 1 h), and 8.5 (at 2 h) mmol/l, respectively, they could be diagnosed as GDM.

Criteria of mediating effect

According to the Baron and Kenny's criteria [7], there exist mediating effects if the following four conditions are met: (1) dependent variables are altered with independent variables; (2) independent variables are related to mediators; (3) dependent variables are altered with mediators after adjusting independent variables; and (4) dependent variables are still altered with independent variables after adjusting the mediators. AMA was considered an independent variable, body fat percentage and protein percentage were considered as mediators, and GDM occurrence was considered dependent variable.

Calculation of indicators

Body fat percentage = body fat weight/body weight*100%, protein percentage = protein weight/body weight*100%.

Statistical analysis

Statistical analysis was performed using SAS9.4 software. The height, weight, lean body weight (LBW), and body fat were normally distributed and expressed as means ± standard deviation. The chi-square test was used to compare the differences in the constituent ratios or rates (of dependent variables) for independent variables. A multivariate logistic regression model was used to analyze the mediating effects of body composition on AMA and GDM. The effects of independent variable on dependent variable and on the mediators were firstly analyzed, and then the independent variable and the mediators were included in the regression model to analyze their effects on the dependent variable. A difference of $p < 0.05$ was considered to be statistically significant.

Results

Basic information

A total of 1635 subjects were enrolled, with age 30.93 ± 4.80 years, height 158.55 ± 5.03 cm, body weight 55.23 ± 8.25 kg, LBW 39.26 ± 4.43 kg, and body fat 15.98 ± 5.18 kg, in the first trimester. There were 983 pregnant women (39.88%) with a body fat percentage of greater than 30%, 1331 pregnant women (81.71%) with a protein percentage of less than 15%, 721 women of multipara (44.10%), 27 women with a history of gynecologic oncology (1.65%), and 214 women with GDM (13.09%), (Table 1).

Comparisons of the basic body information between different age groups

There were 1259 pregnant women (77.00%) who are younger than 35 years, and 376 pregnant women (23.00%) who are over 35 years. There were significant differences in height, weight, LBW, body fat, body fat percentage, protein percentage, and GDM prevalence between the two groups ($p < 0.05$), (Table 1 and Fig. 1).

The mediating effect of the body fat percentage on AMA and GDM

The AMA showed association with GDM (OR (95% CI) 2.63(1.94 ~ 2.56), $p < 0.05$) after adjusting height;

the AMA showed association with body fat percentage (OR (95% CI) 1.52 (1.21 ~ 1.92), $p < 0.05$) after adjusting height;

the body fat percentage showed association with GDM (OR (95% CI) 1.85(1.38 ~ 2.48), $p < 0.05$) after adjusting height, parity and AMA;

the AMA showed association with GDM (OR (95% CI) 2.38 (1.68 ~ 3.37), $p < 0.05$) after adjusting height, parity, and body fat percentage. Therefore, the body's high fat percentage plays a risky mediating effect on AMA and GDM.

The mediating effect of protein percentage on AMA and GDM

The AMA showed association with GDM (OR (95% CI) 2.63 (1.94 ~ 2.56), $p < 0.05$) after adjusting height;

the AMA showed association with protein percentage (OR (95% CI) 0.49 (0.35 ~ 0.70), $p < 0.05$) after adjusting height;

the protein percentage showed association with GDM (OR (95% CI) 0.50 (0.31 ~ 0.81), $p < 0.05$) after adjusting height, parity, and AMA;

the AMA showed association with GDM (OR (95% CI) 2.39 (1.69 ~ 3.38), $p < 0.05$) after adjusting height, parity, and protein percentage. Therefore, the low-protein percentage plays a risky mediating effect on AMA and GDM.

Discussion

This is a retrospective study. A total of 1635 cases from pregnant women, who had regular examinations in Guiyang City Maternal and Child Health Hospital (our hospital) in 2017, were selected as the research subjects. To ensure the accuracy and reliability of experimental study result, we analyzed the quantity of data statistically based on classic Baron and Kenny criteria of mediating effect. From the survey, we recorded pregnant women's body composition in the first trimester of pregnancy and blood glucose levels at the early stage of the third trimester. Authentic and credible, the blood glucose level was measured with 75 g oral glucose tolerance test (OGTT) by specialists of clinical laboratory. According to the observation of body components in the first trimester of pregnancy, we hope to alert the occurrence of gestational diabetes mellitus (GDM) early and to guide obstetricians and gynecologists to conduct nutritional interventions.

There are two types of GDM, one has been diagnosed before pregnancy called pregnancy with diabetes, the type we had listed as exclusion criteria. Another refers to abnormal glucose metabolism or underlying impaired glucose tolerance during pregnancy. Probably, owing to the changes of body

Table 1 Basic informations of the subjects

Variable	Sum n=1635	age<35 years n=1259	age>35 years n=376	t/X^2	*P*
Height, cm, M±SD	158.55 ±5.03	158.83±5.04	157.62±4.91	4.10	<.0001
Weight, kg, M±SD	55.23±8.25	54.69±8.11	57.01±8.49	-4.80	<.0001
LBW, kg, M±SD	39.26±4.43	39.09±4.32	39.81±4.74	-2.79	0.0053
Body fat, kg, M±SD	15.98±5.18	15.62±5.16	17.16±5.06	-5.09	<.0001
Body fat percentage, n(%)					
<30%	983(60.12)	788(62.59)	195(51.86)	13.90	0.0002
>30%	652(39.88)	471(37.41)	181(48.14)		
Protein, kg, M±SD	28.88±3.27	28.76±3.19	29.31±3.50	-2.88	0.004
Protein percentage, n(%)					
<15%	1336(81.71)	1002(79.59)	334(88.83)	16.55	<.0001
>15%	299(18.29)	257(20.41)	42(11.17)		
Parity					
Primipara	914(55.90)	851(67.59)	63(16.76)	303.55	<.0001
Multipara	721(44.10)	408(32.41)	313(83.24)		
History of gynecologic oncology, n(%)					
No	1608(98.35)	1242(98.65)	366(97.34)	3.06	0.0804
Yes	27(1.65)	17(1.35)	10(2.66)		
GDM, n(%)					
No	1421(86.91)	1132(89.91)	289(76.86)	43.35	<.0001
Yes	214(13.09)	127(10.09)	87(23.14)		

structure of women in pregnancy, or the level of placental lactogenin, estrogen, progesterone, cortisol, and placental insulinase in the middle and third trimester of pregnancy, that the insulin receptors in their bodies gradually lose the sensitivity to insulin with the pregnancy progresses, as a result, their blood glucose increases. However, most of the glucose metabolism can be restored to normal after delivery. The main object of this study is such kind of pregnant women with restorable glucose metabolism. Before the two-child policy was issued years ago, the incidence of GDM in China was 1 ~ 5% and appears to a rising trend nowadays as a result of the increasing number of advanced maternal age (AMA) [8]. Compared with the normal pregnant women, GDM patients are more likely to suffer from type 2 diabetes after delivery. During the childbirth, both GDM puerperas and newborns associate with high risk factors such as eclampsia. Also, they

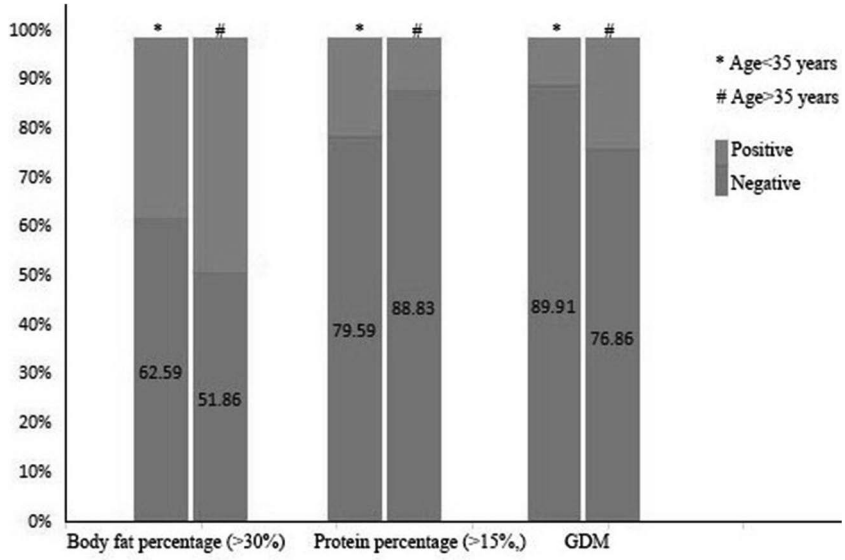


Fig. 1 The conditions of body fat percentage, protein percentage, and GDM prevalence between different age groups

will develop near-term or long-term complications in serious possibility. Therefore, it requires extensive attention to decrease the incidence of GDM by detecting early warning indexes, and proceeding timely nutritional and lifestyle intervention in the first trimester [9].

Traditionally, pre-pregnancy overweight and obesity were treated as the risk factors for the occurrence of GDM. Body mass index (BMI) is the most commonly used to measure the degree of human obesity [10–14]. Once a pregnant woman's BMI is rising above the standard, just like she may be diagnosed with GDM, it will affect her, even the newborn's quality of life. For example, Segura MT et al. [15] believe that BMI is closely related to GDM, demonstrating that high pre-pregnancy BMI or GDM independently alters mRNA expression levels of genes involved in fatty acid (FA) uptake and metabolism and the placental FA profile, which could affect fetal development and long-term health. Nevertheless, there may be diagnosis deviation for muscular obesity, and latent obesity is difficult to be distinguished by BMI solely. The more accurately body fat percentage can be detected, the more accurately we can evaluate obesity of pregnant women. Analysis of body composition data can make up for this deficiency. There have been many researches on body composition analysis, including protein percentage and body fat percentage. A variety of common data about bodies' composition and proportion, especially muscle, bone, fat, water, and minerals, are detected and recorded by a particular instrument [16, 17]. Through these indicators, we can evaluate the proportion of the internal structure of the body. There are researches also supporting that body composition analysis can better reflect the body's relevance between obesity and health [16, 17]. Wang Y et al. [16] have found that the measurement of body composition can be used to identify the gestational diabetes mellitus (GDM) risk in pregnant women. In their retrospective case-control study, they reviewed the medical records of 3965 pregnant women who had body composition measurement, finding that percentage body fat was the strongest risk factor for gestational diabetes after adjusting pre-pregnancy body mass index (BMI). Through the assessment of body composition, it may provide important guidance to identify gestational diabetes in pregnant women with low gestational diabetes risk.

So many instruments have been put to use; among which, our hospital has introduced the INBODY770 to measure body composition of pregnant women [18]. With the benefits as easy operation, non-invasive measurement, and high accuracy, this body composition analyzer utilizes multi-frequency bioelectrical impedance [19].Through these auxiliary means, our clinicians gradually took steps to manage the weight of pregnant women, up to about 12,000 people per year.

Owing to the release of two-child policy, the increase of AMA prompted the incidence of diabetes to a concerning level. Through the body component analysis at first trimester, it is beneficial to supplement an early warning index of GDM. Basically consistent with the policy, our retrospective study showed a significantly increased GDM. The incidence of pregnant women aged above 35 was double as those aged below 35, supported by statistical analysis that AMA was notably correlated with GDM.

Among the two groups, one with pregnant women aged above 35 and another below, there was a significant difference in body fat and body fat percentage, probably associated with GDM. Conducted by multivariate logistic regression, the body fat percentage was correlated with GDM after adjusting for height, parity, history of tumor, and AMA, with the OR (95% CI) value 1.85. AMA was correlated with GDM after adjusting for height, parity, history of tumor, and body fat percentage, with the OR (95% CI) value 2.38. This positive correlation indicates that AMA is a risk factor for GDM, mediated by body fat percentage. Higher body fat percentage is a risk factor for GDM, because of $OR > 1$.

It also seemed a difference in protein percentages between the two groups. The protein percentage was correlated with GDM, after adjusting for height, parity, history of tumor, and AMA, with the OR (95% CI) value 0.50. AMA was correlated with GDM after adjusting for height, parity, history of tumor, and protein percentage, with the OR (95% CI) value 2.38. Indicating another mediating effect between AMA and GDM; lower protein percentage is a risk factor for GDM, because of $OR < 1$.

Through the regression analysis, the history of gynecologic tumor had no correlation with the dependent variable. Therefore, gynecologic tumor was not excluded from this study. This study of the intermediate effect between AMA and GDM indicated that the examination of body composition could be seemed as a supplement to early factor in screening GDM among AMA. High body fat percentage and low-protein percentage mediate the occurrence of GDM among AMA; however, the relevant mechanism is still indistinct. Here comes an assumption that decompensated islet β cells caused by fat are incapable to regulate the blood sugar adequately, which leads to GDM. As the fat tissue synthesis is a decisive effect in the process of insulin resistance, and that women's body fat percentage increases while protein percentage decreases with age [20, 21]. From both domestic and overseas, some researches have pointed out a correlation between elder age and GDM [22–27]. Similarly, some have suggested that AMA is associated with obesity [27]. Nevertheless, few studies have focused on the mediating effect between AMA and GDM, especially the one mediated by body composition. The results of this study could be seemed as a supplementary warning signal for the risk factors of AMA and GDM.

Here is a limitation for the mediation analysis that explains the whole pathway partially. If more relevant factors are obtained, the relationship and effect between variables could be explored more comprehensively through the pathway analysis.

In conclusion, for a better prevention and control of GDM in the future, it is necessary to highlight the education of exercise

and health to AMA, according to which they will realize the adverse effect of high body fat percentage during pregnancy. For AMA, it might be an effective measure to reduce body fat percentage and increase protein percentage by sufficient physical activity and intake of protein before and during pregnancy.

Acknowledgments We thank Guizhou University of Chinese Traditional Medicine and Guiyang Maternal and Child Health Hospital for their help and support. We also thank the women who participated in this study for their contribution and time.

Funding information This work was financed by grants from the Natural Science Foundation of China (81760336) and Guizhou Health Commission (Gzwjkj2018-1-079).

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval The study protocol was approved by the ethics committee of Guiyang Maternal and Child Health Hospital.

Consent to participate Informed consent was obtained from women who participated.

References

1. Chen Q, Carbone ET. Functionality, implementation, impact, and the role of health literacy in mobile phone apps for gestational diabetes: scoping review. JMIR Diabetes. 2017;2(2):e25.
2. Kapur A, Seshiah V. Women & diabetes: our right to a healthy future. Indian J Med Res. 2017;146(5):553–6.
3. Laopaiboon M, Lumbiganon P, Intarut N, Mori R, Ganchimeg T, Vogel JP, et al. Advanced maternal age and pregnancy outcomes: a multicountry assessment. BJOG. 2014;121(Suppl 1):49–56.
4. Kahveci B, Melekoqlu R, Evruke IC, Cetin C. The effect of advanced maternal age on perinatal outcomes in nulliparous singleton pregnancies. BMC Pregnancy Childbirth. 2018;18(1):343.
5. Yin O, Woods A, Koos B, DeVore G, Afshar Y. Central hemodynamics are associated with fetal outcomes in pregnancies of advanced maternal age. Pregnancy Hypertens. 2019;19:67–73.
6. Xue X, Kong BH, Duan T. Obstetrics and gynecology. 9th ed: People's Health Publishing House; 2018. (In Chinese)
7. Baron RM, Kenny DA. The moderator-mediator variable distinction in social psychological research: conceptual, strategic, and statistical considerations. J Pers Soc Psychol. 1986;51(6):1173–82.
8. He Z, Xie H, Liang S, Tang Y, Ding W, Wu Y, et al. Influence of different diagnostic criteria on gestational diabetes mellitus incidence and medical expenditures in China. J Diabetes Investig. 2019;10(5):1347–57.
9. Waters TP, Dyer AR, Scholtens DM, Dooley SL, Herer E, Lowe LP, et al. Maternal and neonatal morbidity for women who would be added to the diagnosis of GDM using IADPSG criteria: a secondary analysis of the hyperglycemia and adverse pregnancy outcome study. Diabetes Care. 2016;39(12):2204–10.
10. Collier A, Abraham EC, Armstrong J, Godwin J, Monteath K, Lindsay R. Reported prevalence of gestational diabetes in Scotland: the relationship with obesity, age, socioeconomic status, smoking and macrosomia, and how many are we missing? J Diabetes Investig. 2017;8(2):161–7.
11. Hildén K, Hanson U, Persson M, Magnuson A, Simmons D, Fadl H. Gestational diabetes and adiposity are independent risk factors for perinatal outcomes: a population based cohort study in Sweden. Diabet Med. 2019;36(2):151–7.
12. Kouhkan A, Khamseh ME, Moini A, Pirjani R, Arabipoor A, Zolfaghari Z, et al. Diagnostic accuracy of body mass index and fasting glucose for the prediction of gestational diabetes mellitus after assisted reproductive technology. Int J Fertil Steril. 2019;13(1):32–7.
13. Najafi F, Hasani J, Izadi N, Hashemi-Nazari SS, Namvar Z, Mohammadi S, et al. The effect of prepregnancy body mass index on the risk of gestational diabetes mellitus: a systematic review and dose-response meta-analysis. Obes Rev. 2019;20(3):472–86.
14. Radzicka S, Pietryga M, Iciek R, Brązert J. The role of visfatin in pathogenesis of gestational diabetes (GDM). Ginekol Pol. 2018;89(9):518–21.
15. Segura MT, Demmelmair H, Krauss-Etschmann S, Nathan P, Dehmel S, Padilla MC, et al. Maternal BMI and gestational diabetes alter placental lipid transporters and fatty acid composition. Placenta. 2017;57:144–51.
16. Wang Y, Luo BR. The association of body composition with the risk of gestational diabetes mellitus in Chinese pregnant women: a case-control study. Medicine (Baltimore). 2019;98(42):e17576.
17. Andersson-Hall UK, Järvinen EAJ, Bosaeus MH, Gustavsson CE, Hårsmar EJ, Niklasson CA, et al. Maternal obesity and gestational diabetes mellitus affect body composition through infancy: the PONCH study. Pediatr Res. 2019;85(3):369–77.
18. Chavarrias M, Carlos-Vivas J, Barrantes-Martín B, Pérez-Gómez J. Effects of 8-week of fitness classes on blood pressure, body composition, and physical fitness. J Sports Med Phys Fitness. 2019c;59(12):2066–74.
19. Kuriyan R. Body composition techniques. Indian J Med Res. 2018;148(5):648–58.
20. Vrachnis N, Belitsos P, Sifakis S, Dafopoulos K, Siristatidis C, Pappa KI, et al. Role of adipokines and other inflammatory mediators in gestational diabetes mellitus and previous gestational diabetes mellitus. Int J Endocrinol. 2012;2012:549748.
21. Hashim M, Radwan H, Hasan H, Obaid RS, Al Ghazal H, Al Hilali M, et al. Gestational weight gain and gestational diabetes among Emirati and Arab women in the United Arab Emirates: results from the MISC cohort. BMC Pregnancy Childbirth. 2019;19(1):463.
22. Carolan M, Davey MA, Biro MA, Kealy M. Maternal age, ethnicity and gestational diabetes mellitus. Midwifery. 2012;28(6):778–83.
23. Meek CL, Devoy B, Simmons D, Patient CJ, Aiken AR, Murphy HR. Seasonal variations in incidence and maternal-fetal outcomes of gestational diabetes. Diabet Med. 2020;37(4):674–80.
24. Erem C, Kuzu UB, Deger O, Can G. Prevalence of gestational diabetes mellitus and associated risk factors in Turkish women: the Trabzon GDM study. Arch Med Sci. 2015;11(4):724–35.
25. Laine MK, Kautiainen H, Gissler M, Raina M, Aahos I, Järvinen K, et al. Gestational diabetes in primiparous women-impact of age and adiposity: a register-based cohort study. Acta Obstet Gynecol Scand. 2018;97(2):187–94.
26. Aydın H, Çelik Ö, Yazıcı D, Altunok Ç, Tarçın Ö, Deyneli O, et al. Prevalence and predictors of gestational diabetes mellitus: a nationwide multicentre prospective study. Diabet Med. 2019;36(2):221–7.
27. Heslehurst N, Ells LJ, Simpson H, Batterham A, Wilkinson J, Summerbell CD. Trends in maternal obesity incidence rates, demographic predictors, and health inequalities in 36 821 women over a 15-year period. BJOG. 2007;114(2):187–94.

International Journal of Diabetes in Developing Countries (2021) 41:127–135
https://doi.org/10.1007/s13410-020-00853-0

ORIGINAL ARTICLE

Barriers to postpartum follow-up of mothers with gestational diabetes mellitus and its implications: a mixed method study

Ninu P. Mathew[1] · Arathi P. Rao[1] · Prakash Narayanan[1]

Received: 13 May 2020 / Accepted: 27 July 2020 / Published online: 19 August 2020

Abstract

Aims To explore the barriers that reduce postpartum follow-up of mothers with gestational diabetes mellitus.

Settings and design This is a mixed method study with a qualitative study followed by a detailed narrative review conducted in Karnataka. The subjects were selected from Dr. T M A Pai Rotary Hospital, Karkala.

Methods and material A total of 6 in-depth interviews were conducted out of 9 patients who met the inclusion and exclusion criteria. The contact number of the patients was obtained and they were contacted over the phone. Interviews were conducted at a place and time of the women's choice.

Narrative review Thematic analysis approach was used for analysis. The codes were formed from the transcripts and then categories were formed. A narrative review was also conducted to explore the existing evidence on the topic. In total, 62 articles were reviewed including various types of studies.

Results The key findings are categorized in general as barriers, then subthemes were formed. The most common barriers were lack of prior appointment for mothers, lack of adequate hospital facilities, baby and concerns, priority and lack of knowledge or health education.

Conclusions The overall study findings reveal that healthcare facilities must be strengthened to avoid long waiting time and overcrowding to improve the screening after delivery. Educating the women regarding the need for dietary restrictions and exercise and giving priority to their own health as well as reminder systems is necessary to ensure the postpartum follow-up.

Keywords Barriers · Postpartum follow-up · Gestational diabetes mellitus

Introduction

Gestational diabetes is a common condition that occurs during pregnancy. Globally, there are 204 million women (20–79 years) living with diabetes and this number is projected to increase to 308 million by 2045. Half of women with a history of GDM progress into type 2 diabetes within 5 to 10 years after delivery [1]. According to Diabetes in Pregnancy Study group India (DIPSI), gestational diabetic women require follow-up with glucose tolerance test with 75 g oral glucose after 6 weeks of delivery and that must be repeated after 6 months if necessary and then every year to determine whether the glucose tolerance has returned to normal or has progressed [2]. However, the screening rates during postpartum are very low which ranges from 17 to 24% [3, 4], especially in low and middle-income countries where the vast majority of cases of GDM occur and access to maternal care is limited [1]. It is shown that although there is an increase in the rates of postpartum screening since the 1990s, it reached only up to 53.8% in 2006 [5].

The existing knowledge regarding the barriers for postpartum follow-up of women who were diagnosed with gestational diabetes mellitus is very limited. Very few qualitative studies have been conducted especially in a country like India where the burden of diabetes is very high. There exist some barriers on a personal level and in the health system level that leads to a missed opportunity for follow-up of mothers who had gestational diabetes mellitus. Therefore, understanding these barriers is necessary to improve the postpartum screening and preventive behavior/s. This study is undertaken among the women in the postpartum period up to 1 and half years in order to explore the barriers gap for postpartum follow-up. This study aims to explore the barriers for postpartum follow-up of mothers with history of GDM.

Key messages Given the evidence of gestational diabetes mellitus and its progression to type 2 diabetes mellitus, we have to promote postpartum follow-up.

✉ Ninu P. Mathew
ninupm@gmail.com

[1] Prasanna School of Public Health, Manipal Academy of Higher Education, Manipal, Udupi, Karnataka 576104, India

Subjects and methods

The present study was an exploratory study using a qualitative research design followed by a detailed narrative review. It was conducted in Karkala taluk of Udupi District and included the women who were diagnosed with GDM and are within 18 months from the date of delivery and women who were not coming for postpartum follow-up. Ethical clearance was obtained from the Institutional Ethical Committee, Kasturba Medical College and Kasturba Hospitals, MAHE. The list of patients who were diagnosed with GDM was obtained from the hospital after obtaining permission from the hospital authorities. In total, there were 36 women who were diagnosed with GDM and out of which 27 women were not coming for follow-up screening. These 27 women were contacted and a short description regarding the study was given. Out of these 27 women contacted, 14 women were relocated, 4 of them were not willing to participate and 3 could not be contacted due to inappropriate contact details. The interview was conducted among 6 women who were willing to participate. The methodology used for data collection is explained in Fig. 1. A convenient time, place and date of the interview were fixed upon participants' willingness to take part in the study. Home visit was done by the principal investigator. Informed consent was obtained. The duration of the interview ranged from 20 to 40 min and audio recording of the interview was done. A validated semi-structured interview guide was used to collect data which included the details of sociodemographics and other 4 domains focusing on barriers for postpartum follow-up. The domains included antenatal period, screening and treatment, medical care and service and postpartum follow-up. After interviewing the participants, the transcripts of voice-recorded interviews were prepared and translation was done. The data were read thoroughly to develop codes and then categories. Descriptive statistics were used to describe the sample in terms of sociodemographics details; thereafter, this information was used to frame a comprehensive note of barriers to mothers who had GDM.

In order to compare the results of these 6 interviews, a narrative review was conducted to find out the existing evidence on barriers faced in postpartum period for follow-up by women who had GDM. Studies that gave an insight on the different barriers that the patients face in their personal life as well as related to healthcare in postpartum follow-up were

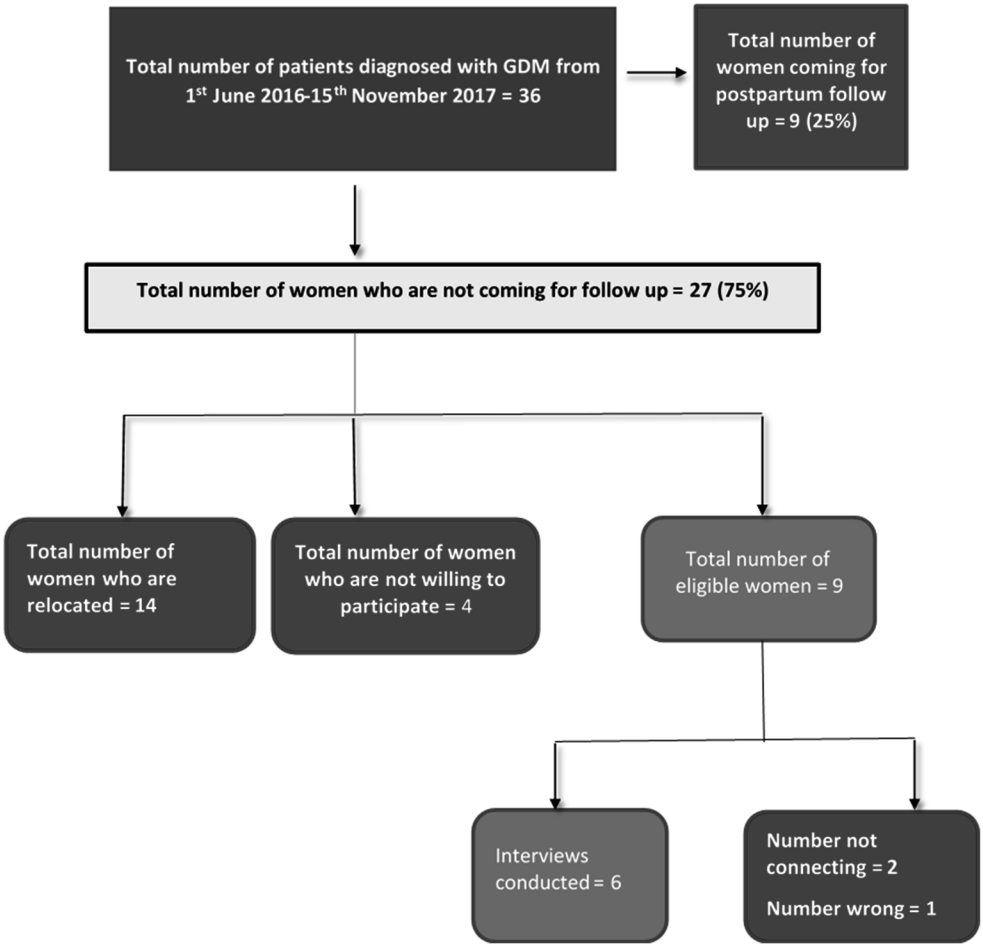


Fig. 1 Diagram of the methodology used for data collection

included. Key search terms like "Gestational Diabetes Mellitus", "post-partum", "barriers", "screening", "post-partum follow-up", "lifestyle", "physical activity" and "challenges" were used and conducted searches in electronic databases like PubMed, Medline and references of selected articles to find relevant material for barriers. All types of study designs have been included to identify all the barriers in postpartum follow-up. A total of 62 articles were identified for the review. The types and number of studies identified were qualitative studies [22], quantitative studies [32] and reviews [8] from India and other parts of the world. The articles reviewed include 32 full-text articles and 30 abstracts. The data in this review is analyzed by using a thematic analysis approach. The findings were segregated into different themes as barriers in general and subthemes were extracted.

Results

Table 1 shows the sociodemographics details of the respondents. The results of the study are categorized as barriers. The problems faced by women on the health system level or healthcare level on a personal level for follow-up have been identified as barriers. The codes and verbatim are presented in Table 2.

Barriers

There were different barriers for the postpartum screening and lifestyle modification. Those included various obstacles they faced in their personal level and health system level.

Waiting time and inconvenience to go early in the morning

Majority of the women shared that they had experienced long waiting time and also expressed regarding inconvenience to go early morning for testing (blood sugar) during their antenatal period. They also expressed the inconvenience they had due to long waiting hours to meet the doctor after testing.

Need for prior appointment

Majority of women expressed that it would be good if there is a system of prior appointment especially for mothers. This would help mothers to avoid long waiting hours.

Need for hospital facilities and services

There were other challenges that this study elicited such as overcrowding in the hospital, staying far away from the hospital and different instances where they had bad experience from the hospital which lead to dissatisfaction in the hospital facilities and services.

Table 1 Sociodemographic details of the participants

Demographic variable		Frequency (%)
Age	27	1 (16.6%)
	28	1 (16.6%)
	30	1 (16.6%)
	33	1 (16.6%)
	34	1 (16.6%)
	48	1 (16.6%)
Religion	Christian	2 (33.3%)
	Hindu	4 (66.6%)
Marital status	Married	6 (100%)
Education	Graduate	1 (16.6%)
	Higher secondary	1 (16.6%)
	PG	3 (50%)
	Primary	1 (16.6%)
Occupation	Housewife	4 (66.6%)
	Working	2 (33.3%)
Age at marriage	24	2 (33.3%)
	25	1 (16.6%)
	27	2 (33.3%)
	28	1 (16.6%)
Parity	1	4(66.6%)
	2	1 (16.6%)
	3	1 (16.6%)
Type of present delivery	Caesarian	4 (66.66%)
	Normal	2 (33.3%)
Income	200,000	2 (33.3)
	300,000	1 (16.6%)
	400,000	1 (16.6%)
	60,000	1 (16.6%)
	600,000	1 (16.6%)

Vomiting related to the test

Two of the women expressed their experience of having vomiting sensation after drinking glucose. They said that it tasted sweet and made them vomit. One of them said "sweet and vomiting was there" and it was one of the reasons for avoiding the test.

Asymptomatic and perceived normality

Majority of the women had perception of being normal. Many of them thought they are normal since the blood test before discharge from the hospital showed normal blood sugar.

Table 2 Codes and verbatim

Codes	Verbatim
Waiting	"we need to wait, we have to go early and then go to registration wait for a long time, do the tests and meet the doctor, give blood then wait for two hours, if there is a system for prior appointment, we do not need to wait for a long time".
Go early morning	"We have to go really early at around 6 o'clock in the morning and if we go early then we can get the turn, otherwise no. So, if I go in the morning, at least by afternoon we will get a turn."
Prior appointment	"Yes, that's the main reason why I did not go to hospital. And also, I will be shifting to Dubai soon and I thought I will take an appointment with the doctor there meet the doctor at the time of appointment and meet them. No need to wait, here we do not have that facility. we do not get a prior appointment and then we meet the doctor: that means when we are here we do not feel like going. There we can go on time and get an appointment"
Hospital facilities and care	"doctor was very good, now that is too far for me know that's why am going here. I did not go because I shifted here know "But somehow, I feel that there is much crowd, there is crowd like anything, it is one difficulty I found. Because as I am a lecturer I have to come back otherwise I have to take leave for a whole day, for that I feel some of inconvenience. Getting time to go and we used to go in the morning only." "I had a bad experience during caesarian section. I could feel the baby coming out and the whole that is happening in my stomach. I had some difficulties related to that. I had some problems related to the stitch, the stitch took some long time to heal".
Vomiting related to test	"sweet and vomiting was there"
Asymptomatic and perceived normality	"doctor said it was normal so I thought it is okay. I am normal and I did not go."
Time constraints and busy schedules	"since am working and that makes me busy" and another woman said "I do not have enough time to take care of herself"
Baby and concerns	"I have to go with the baby and she is very small, she is small, it is very difficult. Actually, we have to go for check up in the morning and the baby would cry, it was a reason"
Priority	"Yes, it was like that, I had thyroid issues before pregnancy and after delivery I had some symptoms so I focused on treating that, I gave priority to that"
Inadequate health education	"No no nothing, doctor did not give me dietary advice"
Lack of decision-making power and family support	"I was interested to go but money was not there So they told me no, so I did not go"
Forgot	"sometimes I thought I must go but in between I forgot and I forgot going to hospital in between the busy schedules".
Being frustrated	"I feel I'm doing wrong, sometimes I feel frustrated because in this age only I got diabetes. I will get tension and sometimes I do deceive myself eating when I go outside though I know I should not".
Financial issues	"Actually this house was built so financial issues were there. I did not go for checkup primarily because of the financial problem"

Majority of them were waiting for symptoms to appear and then take up the screening test.

Time constraints and busy schedules

Lack of time was a major barrier for most of the women. They were either busy with the baby or with work. They also expressed their busy schedules as one of the barriers in going for the test and also for following lifestyle behaviour.

Baby and concerns

As reported by majority of the women, concerns related to the baby made them to miss postpartum screening and adopting lifestyle behavior/s like diet and exercise. Women missed the postpartum screening test because their baby would cry if waited for a long time.

Priority

Majority of the women did not take this screening and lifestyle modification as important. They gave priority for other things like their children and work. One woman had financial issue and she gave priority to her sick child rather than herself. Most of them took an effort to go for baby's vaccination and ignored the screening test for themselves.

Inadequate health education

Though majority of the woman received education regarding the screening and chances for developing type 2 diabetes, they told that there is lack of education or information regarding diet and exercise after delivery. They perceived that since the blood sugar was normal immediately after delivery, there was no need for dietary changes anymore.

Lack of decision-making power and family support

Lack of decision-making power in the family was another barrier for follow-up as explained by the women. One woman from a rural area expressed her interest in going for the test but she did not go because her husband and his family said not to do so. Other two women wanted support of their family so that they can go for testing or follow the diet and exercise.

Forgot

Four of the women said that they forgot about the test and one of them said that she remembers it as 6 months and does not even remember what doctor had advised and quoted "I remember it as 6 months. May be doctor had told me 6 weeks".

Being frustrated

One of the main barriers to following exercise and diet control for the woman who had already developed type 2 diabetes was that she felt frustrated because she still has abnormal blood glucose level in spite of all the efforts for follow-up from her side.

Financial difficulties

One woman from the rural area said that she had experienced financial difficulties during the postpartum period which was a reason for not going for the test.

Results of the narrative review

Barriers

Inconvenience associated with diagnostic test and treatment

The test used for diagnosis in all the articles for type 2 diabetes screening was OGTT recommended by the American Diabetes Association. The literature shows that there are major challenges related to diagnostic test like poor laboratory conditions [6], difficulty in completing the OGTT [7], inconvenience associated with 2- to 3-h test [8, 9, 10]. Treatment-related barriers point out two contrasting ideas, i.e. treatment with insulin during pregnancy was associated with non-adherence to postpartum screening [11] and also higher adherence rate to postpartum testing in another study [12].

Lack of healthcare facilities and care

Importance and need for adequate, accessible and responsible healthcare facilities were important barriers that were more commonly found in many of the articles. In low- and middle-income countries (LMIC), the barriers include lack of trained healthcare providers, high staff turnover, lack of standard protocols, lack of consumables and equipment, financing of health services and treatment, lack of or poor referral systems, feedback mechanisms and follow-up systems, distance to health facility [13], inadequate supply of glucose [14] and lack of nearby health facility in the locality [15].

Barriers in healthcare in high-income countries included perception of doctors about the timing of screening in postpartum, not giving priority to postpartum screening, lack of awareness and adherence to postpartum screening guidelines [16–18] and lack of consensus about the type of screening test [19]. Clinicians' perception that "screening guidelines are inconsistent" was also identified as a barrier from the clinicians' side [10].

Inadequate health education, knowledge and awareness

Other important barriers to the postpartum follow-up were lack of information about type 2 diabetes, lack of education [20, 21, 22] and need for written detailed information and repeating the information in the future visits [23, 24]. Lack of knowledge [25], inadequate education from healthcare staff regarding developing diabetes after delivery and postpartum screening [25] were also elicited in few articles as barriers. Few intervention studies have shown an increase in the postpartum testing after diabetes education either in the ante partum period or postpartum period [26] and also following introduction of the counselling [27, 28].

Findings from another study indicated a range of gaps in the women's understanding of how lifestyle and diet can reduce the development of type 2 diabetes. Those included an incomplete understanding of lifestyle risk factors [28], lack of awareness of the need to attend screening and lifestyle modification [8, 15, 29] and the lack of awareness about the future development of type 2 diabetes [30, 31, 32].

Barriers for lifestyle modification (diet and exercise)

The common barriers to both diet and exercise were difficulty in following diet and exercise, lack of information [21, 23], lack of time and financial constraints, childcare duties, lack of motivation, fatigue and [33, 34] limited knowledge [35]. Other barriers include frustration over controlled diet [36], difficulties finding food alternatives, too busy or tired and wanting to have favourite foods after pregnancy restrictions [21]. The tendency for the women to tolerate restrictions during pregnancy for the sake of the baby to maximize fetal health and finding it difficult after the birth was found in a study [37]. Food preferences of children and family and [14, 21] different cultural practices [38] also influenced the dietary habits. In a study done on implementers from World Diabetes Foundation (WDF), one respondent from India noted that "it

is customary to encourage pregnant women to eat sweets and certain calorie dense, high fat snacks in order for them to have enough energy, and people bring such food as gifts when they visit. Thus, being on diet where such things are banned can be a damper on the celebrations of the pregnancy and childbirth within the family and raise issues about the health of the young woman thereby curbing her motivation to eat healthy" [13]. Confidence and skills in cooking healthy foods, along with family food preferences and time pressures, were also important influences on eating habits [39].

Major barriers to regular postpartum exercise included lack of advice regarding regular exercise postpartum [15], lack of time [29, 35] and lack of child care [29, 33, 37]. Other constraints were lack of partner support [30], lack of transportation to get to a gym to exercise [21] and concerns about cold or hot weather and household chores [37].

Baby and concerns

Baby and concerns were major barriers identified from the literature review. Prioritizing the child's and family's needs above their own and [40] being occupied with the baby were the major barriers under this theme [21, 22]. Another important constraint found for attending postpartum screening was the need to attend screening with infants and young children [8]. While tiredness, maternal attachment and childcare demands were prominent barriers in the early post-natal months, concerns on child development became more significant barriers in the later stage says another study [41].

Financial barriers

Financial barrier emerged as a major barrier in many of the articles. Financial concerns about buying healthy food [21]. Diabetes screening in postpartum was also associated with having a higher income as well as [42] private insurance Medicaid in high-income countries like the USA [43, 44]. While this is the case in countries with Medicaid or private insurance, the low- and middle-income countries face other barriers due to out of pocket expenditure on healthcare and lack of health financing. This is a barrier to access care and treatment for many women with GDM in these countries. The cost of medication and even the cost of following dietary recommendations are also defined as challenge in the study [45, 46].

Self-perception

Self-perception of women regarding the screening and postpartum health behavior/s were found as barriers in postpartum follow-up. Perceiving postpartum diabetes screening as difficult [47], perceiving themselves as healthy and not taking the visit as serious [16] and feeling testing as unnecessary [48] were major identified barriers for attending screening. Believing that GDM would disappear after delivery and diabetes screening as a foreign idea was one among the reasons for not attending screening [22]. Women perceiving that they lived a sound life and did not feel that they should change anything in their lifestyle [23] were also found under this theme.

Lack of family, social and psychological support

Lack of support from the family and society including the psychological support was found to be one of the barriers for postpartum follow-up. The societal and family barriers were more prominent in LMIC in India. Practices related to pregnant women's diet, lack of decision-making power among women regarding their own health, societal negligence of women's health, fear of stigmatization, role of women in society and expectations that the pregnant woman move to her maternal home for delivery were found to be societal barriers in LMIC for gestational diabetes care [13, 15]. A study from south India states that "In India, the growing concern of family and friends for a pregnant woman and her baby is a major factor. Almost everyone within the family has an opinion. This can confuse and complicate the pregnant woman's decisions to follow the health-care professional's advice". [46]

Family-related issues such as time constraints due to childcare and unhealthy family eating habits were mentioned as barriers to diabetes prevention [21], and personal and family adjustments due to new baby [49] and women consistently placing the needs of their families before their own [29] were also major barriers. A need for psychological support was found because women were more likely to experience shock, fear or anxiety, abandonment and uncertainty during pregnancy and in postpartum [29].

Time constraints and busy schedules

Different studies have shown that time pressure and busy schedules as barriers for the follow-up in postpartum period. Time and busy schedule as the most frequently talked about barrier which was due to child care and work responsibilities [7], while other studies show that time constraints of the physicians or feeling too busy for GDM education plays an important role in the follow-up [44].

Discussion

Major findings of the study explore the barriers for postpartum follow-up of mothers who had GDM. The most common barriers were lack of prior appointment for mothers, lack of adequate hospital facilities, baby and concerns, priority and lack

of knowledge or health education. Other observations of the study show that 75% of the women who had GDM were not coming for follow-up and only 25% of the total women with GDM came for follow-up screening in the postpartum period (9/36). 62.9% of the women were lost to follow-up due to relocation (51.8%) and loss of contact (11. 1%).Out of the total 36 women who had GDM, one woman developed type 2 diabetes within 1 year of the postpartum period. The various barriers emerged from the narrative review for postpartum follow-up included inconvenience associated with the test and lack of healthcare facilities. While barriers explored from the qualitative study are long waiting time for the test, there are also lack of prior appointments and lack of adequate hospital facilities. The study also identified certain barriers like being asymptomatic or perceived normality, which was also found in the narrative review under the theme self-perception, the women perceived themselves as normal and this was found among the top three reasons for skipping postpartum screening in a study conducted by Chang et al. in China. The same reasons were mentioned by the study participants in the qualitative study [23]. Another barrier was time constraints and busy schedules. The findings from the narrative review also point out these as barriers experienced by women from other countries as well.

The barriers to diet and exercise were lack of education and awareness about the importance of these preventive behavior/s and the review also shows that the most common barriers for undertaking dietary restrictions were lack of information, lack of advice and lack of specific written information which were found in three studies from across the world and out of which one is from India.

One main important barrier was baby and concerns which were also similar to the results of the narrative review. The qualitative study showed that being busy with the baby and carrying baby to the hospital were reasons for skipping postpartum screening. In the narrative review, concerns about child care, child development, busy in caring the baby, the health of the baby and attending the screening with the baby were major barriers found under this theme.

The results of the qualitative study also support the evidence from the review that family support and lack of decision-making power among Indian women were also a barrier to postpartum screening. The most common barriers were lack of prior appointment for mothers, lack of adequate hospital facilities, baby and concerns, priority and lack of knowledge or health education.

The overall study findings reveal that healthcare facilities must be strengthened to avoid long waiting time and overcrowding to improve the screening after delivery. Educating the women regarding the need for dietary restrictions and exercise and giving priority to their own health as well as reminder systems to ensure the follow-up is necessary to improve postpartum follow-up.

The strengths of the study are: it is a purely qualitative study that exclusively followed the women who were not coming for postpartum follow-up and 5 out of six participants in the study have not gone for follow-up at 6 weeks postpartum. The study has done 6 face to face interviews and more than half of the eligible women were relocated. More barriers could be extrapolated if more number of interviews could be conducted. A multi-centric study might have been appropriate to get enough power to the study and the findings. A focus on a quantitative analysis using a validated standardized questionnaire could have been done to get an objective picture on the barriers.

Code availability Not applicable.

Authors' contributions All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by Ninu P Mathew. The first draft of the manuscript was written by Ninu P Mathew and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript. Each author has contributed to the following areas of the study.

Conceptualization: Dr. P Arathi Rao, Ninu P Mathew, Dr. Prakash Narayanan

Methodology: Dr. Arathi Rao, Ninu P Mathew, Dr. Prakash Narayanan

Formal analysis and investigation: Ninu P Mathew

Writing - original draft preparation: Ninu P Mathew

Writing - review and editing: Dr. Arathi Rao, Ninu P Mathew, Dr. Prakash Narayanan

Funding acquisition: Not applicable

Resources: Dr. Arathi Rao, Ninu P Mathew, Dr. Prakash Narayanan

Supervision: Dr. P Arathi Rao and Dr. Prakash Narayanan

Data availability All data and materials as well as software application or custom code support published claims and comply with field standards.

Compliance with ethical standards

Ethical clearance was obtained from the Institutional Ethical Committee, Kasturba Medical College and Kasturba Hospitals, MAHE.

Conflict of interest The authors declare that they have no conflict of interest.

References

1. International Diabetes Foundation (2018) IDF diabetes atlas - Home. [online] Available at: http://www.diabetesatlas.org/. Accessed 15 Nov 2017.
2. Ratner RE, Christophi CA, Metzger BE, Dabelea D, Bennett PH, Pi-Sunyer X et al. Prevention of diabetes in women with a history of gestational diabetes: effects of metformin and lifestyle interventions. J Clin Endocrinol Metab. 2008; 93:4774.
3. Mahalakshmi MM, Bhavadharini B, Maheswari K, Anjana RM, Jebarani S, Ninov L, et al. Current practices in the diagnosis and management of gestational diabetes mellitus in India (WINGS-5). Indian J Endocrinol Metab. 2016;20:364.
4. Stage E, Ronneby H, Damm P. Lifestyle change after gestational diabetes. Diabetes Res Clin Pract. 2004;63:67–72.

5. Ferrara A, Peng T, Kim C. Trends in postpartum diabetes screening and subsequent diabetes and impaired fasting glucose among women with histories of gestational diabetes mellitus: a report from the Translating Research Into Action for Diabetes (TRIAD) Study. Diabetes Care. 2009;32:269–74.
6. Rafii F, Rahimparvar SFV, Mehrdad, Keramat. Barriers to postpartum screening for type 2 diabetes: a qualitative study of women with previous gestational diabetes. Pan Afr Med J. 2017;1:54.
7. Keely ECH, Karovitch A, Graham I. Screening for type 2 diabetes following gestational diabetes: family physician and patient perspectives. Can Fam Phys. 2010;56:558–63 .
8. Sterne V, Logan T, Palmer MA. Factors affecting attendance at postpartum diabetes screening in women with gestational diabetes. Pract Diabetes Int. 2011;28:64–8.
9. Morrison MK, Collins CE, Lowe JM. Postnatal testing for diabetes in Australian women following gestational diabetes mellitus. Aust N Z J Obstet Gynaecol. 2009;49:494–8.
10. Tovar A, Chasan-Taber L, Eggleston E, Oken E. Postpartum screening for diabetes among women with a history of gestational diabetes mellitus. Prev Chronic Dis. 2011;8:A124 .
11. Kwong S, Mitchell RS, Chik CL. Postpartum diabetes screening adherence rate and the performance of fasting plasma glucose versus oral glucose tolerance test. Diabetes Care. 2009;32:2242–4.
12. Cabizuca CA, Rocha PS, Marques JV, Costa TFLR, Santos ASN, Schröder AL, et al. Postpartum follow up of gestational diabetes in a tertiary care center. Diabet Metab Syndr. 2018;10:2–6.
13. Nielsen KK, Courten M, Kapur A. Health system and societal barriers for gestational diabetes mellitus (GDM) services - lessons from World Diabetes Foundation supported GDM projects. BMC Int Health Hum Rights. 2012;12:33.
14. Doran F, Davis K. Gestational diabetes mellitus in Tonga: insights from healthcare professionals and women who experienced gestational diabetes mellitus. N Z Med J. 2010;123:59–67.
15. Sakeena K, Sundari RTK. The "missing window of opportunity" for preventing diabetes: a mixed method study on postpartum screening for diabetes among women with gestational diabetes mellitus in Kerala, India. Int J Non-Commun Dis. 2017;2:78–84.
16. McGovern A, Butler L, Jones S, Vlymen J, Sadek K, Munro N, et al. Diabetes screening after gestational diabetes in England: a quantitative retrospective cohort study. Br J Gen Pract. 2014;64: e17–23.
17. Hunsberger ML, Donatelle RJ, Lindsay K, Rosenberg KD. Physician care patterns and adherence to postpartum glucose testing after gestational diabetes mellitus in Oregon. PLoS One. 2012;7:e47052. https://doi.org/10.1371/journal.pone.0047052.
18. Stuebe A, Ecker J, Bates DW, Zera C, Bentley-Lewis R, Seely E. Barriers to follow-up for women with a history of gestational diabetes. Am J Perinatol. 2010;27:705–10.
19. Henderson CE, Kavookjian J, Leitstein H, McKoy JM, Murage MJ, Lipman RD, et al. Window of opportunity: postpartum screening of women with gestational diabetes for early detection of prediabetes and type 2 diabetes. Open Diab J. 2012;25:25–8.
20. Van Ryswyk E, Middleton P, Shute E, Hague W, Crowther C. Women's views and knowledge regarding healthcare seeking for gestational diabetes in the postpartum period: a systematic review of qualitative/survey studies. Diabetes Res Clin Pract. 2015;110: 109–22.
21. Bieda J. Perceptions of risk for the development of type 2 diabetes in African-American women with gestational diabetes [PhD]. University of Michigan; 2009.
22. Chang Y, Chen X, Cui H, Zhang Z, Cheng L. Follow-up of postpartum women with gestational diabetes mellitus (GDM). Diabetes Res Clin Pract. 2014;106:236–40.
23. Lindmark A, Smide B, Leksell J. Perception of healthy lifestyle information in women with gestational diabetes: a pilot study before and after delivery. Eur Diab Nurs Spring. 2010;7:16–8.
24. Hirst JE, Tran TS, Do MA, Rowena F, Morris JM, Jeffery HE. Women with gestational diabetes in Vietnam: a qualitative study to determine attitudes and health behaviours. BMC Pregnancy ChildB. 2012;9:81.
25. Hoedjes M, Berks D, Vogel I, Franx A, Duvekot JJ, Oenema A. Motivators and barriers to a healthy postpartum lifestyle in women at increased cardiovascular and metabolic risk: a focus-group study. Hypertens Pregnancy. 2012;31:147–55.
26. Stasenko M, Liddell J, Cheng YW, Sparks TN, Killion M, Caughey AB. Patient counseling increases postpartum follow-up in women with gestational diabetes mellitus. Am J Obstet Gynecol. 2011;204: 522–6.
27. Capula CCE, Vero A, Foti DP, Brunetti A, Vero R. Prevalence and predictors of postpartum glucose intolerance in Italian women with gestational diabetes mellitus. Diabetes Res Clin Pract. 2014;105: 223–30.
28. Poth M, Carolan M. Pregnant women's knowledge about the prevention of gestational diabetes mellitus: a qualitative study. Br J Midwifery. 2013;2:692–700.
29. Graco M, Garrard J, Jasper AE. Participation in physical activity perceptions of women with a previous history of gestational diabetes mellitus. Health Promot J Austr. 2009;20:20–5.
30. Collier AS, Mulholland C, Williams J, Mersereau P, Turay K, Prue C. A qualitative study of perceived barriers to management of diabetes among women with a history of diabetes during pregnancy. J Women's Health. 2011;20:1333–9.
31. Keely E. An opportunity not to be missed – how do we improve postpartum screening rates for women with gestational diabetes? Diabetes Metab Res Rev. 2012;28:312–6.
32. Rayanagoudar G, Moore M, Zamora J, Hanson P, Huda M, Hitman G. Postpartum care of women with gestational diabetes: survey of healthcare professionals. Eur J Obstet Gynecol Reprod Biol. 2015;194:236–40.
33. Nicklas JMZC, Seely EW, Abdul-Rahim ZS, Rudloff ND, Levkoff SE. Identifying postpartum intervention approaches to prevent type 2 diabetes in women with a history of gestational diabetes. BMC Pregnancy Childbirth. 2011;11:1–8.
34. Tang JW, Pumarino J, Ackermann RT, Cameron KA. Perspectives on prevention of type 2 diabetes after gestational diabetes: a qualitative study of Hispanic, African-American and White women. Matern Child Health J. 2015;19:1526–34.
35. Koning SH, et al. Postpartum glucose follow-up and lifestyle management after gestational diabetes mellitus: general practitioner and patient perspectives. Diabetes Metab Syndr. 2016;15:56 .
36. Neufeld HT. Food perceptions and concerns of aboriginal women coping with gestational diabetes in Winnipeg, Manitoba. J Nutr Educ Behav. 2011;43:482–91.
37. Bandyopadhyay M, Small R, Davey MA, Oats JJ, Forster DA, Aylward A. Lived experience of gestational diabetes mellitus among immigrant South Asian women in Australia. Aust N Z J ObstetGynaecol. 2011;51:360–4.
38. Kim S, Sappenfield W, Sharma A, Wilson H, Bish C, Salihu H, et al. Racial/ethnic differences in the prevalence of gestational diabetes mellitus and maternal overweight and obesity, by nativity, Florida, 2004-2007. Obesity. 2013;21:33–40.
39. Zehle K, Smith BJ, Chey T, McLean M, Bauman AE, Cheung NW. Psychosocial factors related to diet among women with recent gestational diabetes: opportunities for intervention. Diabetes Educ. 2008;34:807–14.
40. Parsons J, Ismail K, Amiel S, Forbes A. Perceptions among women with gestational diabetes. Qual Health Res. 2014;24:575–85.
41. Lie M, Hayes L, Lewis-Barned N, May C, White M, Bell R. Preventing type 2 diabetes after gestational diabetes: women's experiences and implications for diabetes prevention interventions. Diabet Med. 2013;30:986–93.

42. Man B, Turyk M, Kominiarek M, Xia Y, Gerber B. Diabetes screening in US women with a history of gestational diabetes, National Health and Nutrition Examination Survey, 2007–2012. Prev Chronic Dis. 2016;13:e-124.
43. Hale NL, Probst JC, Liu J, Martin AB, Bennett KJ, Glover S. Postpartum screening for diabetes among Medicaid-eligible South Carolina women with gestational diabetes. Womens Health Issues. 2012;22:e163–9.
44. Russell M, Phipps M, Olson C, Welch H, Carpenter M. Rates of postpartum glucose testing after gestational diabetes mellitus. Obstet Gynecol. 2006;108:1456–62.
45. Nielsen KK, Kapur A, Damm P, de Courten M, Bygbjerg IC. From screening to postpartum follow-up – the determinants and barriers for gestational diabetes mellitus (GDM) services, a systematic review. BMC Pregnancy Childbirth. 2014;14:41.
46. Morampudi S, Balasubramanian G, Gowda A, Zomorodi B, Patil AS. The challenges and recommendations for gestational diabetes mellitus care in India: a review. Front Endocrinol. 2017;8:56.
47. Morrison MK, Lowe JM, Collins CE. Australian women's experiences of living with gestational diabetes mellitus. Women Birth. 2014;27:52–7.
48. Hamel MS, Werner EF. Interventions to improve rate of diabetes testing postpartum in women with gestational diabetes mellitus. Curr Diab Rep. 2017;17:1–7.
49. Bennett WL, Ennen CS, Carrese JA, et al. Barriers to and facilitators of postpartum follow-up care in women with recent gestational diabetes mellitus: a qualitative study. J Women's Health. 2011;20:239–45.

International Journal of Diabetes in Developing Countries (2021) 41:136–140
https://doi.org/10.1007/s13410-020-00843-2

ORIGINAL ARTICLE

Ocular surface assessment in maturity-onset diabetes of the young

Osman Celikay[1] · Mustafa Kosker[1] · Sinan Çalışkan[1] · Erman Cakal[2] · Ferda Alparslan Pinarli[3] · Canan Gurdal[1]

Received: 1 March 2020 / Accepted: 13 June 2020 / Published online: 4 July 2020

Abstract

Aim To examine ocular surface changes in patients with maturity-onset diabetes of young (MODY).

Material and methods Fifty patients with MODY who were transferred to Genetic Diagnosis Department of Diskapi Yildirim Beyazit Training and Research Hospital have been included. All patients were subjected to Ocular Surface Illness Index (OSDI) survey, shirmer test, invasive and non-invasive tear break up time (TBUT), cornea fluorescein, and Lissamine green dye tests. Lastly, meibography measurements have been completed using Sirius Scheimpflug camera (Sirius, CSO, Florence, Italy).

Results All 50 patients had a glucokinase (GCK) mutation. MODY diagnosis time was 5.0 ± 5.5 years (range: 0–20 years) on an average. While the average HBa1c levels were 5.5 (range: 4.8–6.5, reference range: 4–6), none of the patients were diagnosed with diabetic retinopathy in fundus examination. There was no significant difference between MODY and control group in terms of OSDI score, Schirmer 1 test, meibomian gland loss, average BUT, TBUT ((*p*)0.05).

Discussion Our work is the first one which specifically evaluates ocular surface in MODY patients. Contrary to the common concept that "dry eye is frequently seen in type 1 and type 2 DM," we did not come across any significant difference between MODY and control group in ocular surface tests. It might be linked to the fact that not enough time had passed for DR or DES symptoms to be detected. Our patients' average diabetes diagnosis length might be a short time span for clinical symptoms like DR and DES to show up.

Keywords Ocular surface · Cataract · Retinopathy

Introduction

Cataract and retinopathy are well-known complications of diabetes. Recently, problems related to ocular surface—especially dry eye syndrome (DES)—have been reported in patients with diabetes mellitus (DM) [1–4]. While The Canadian Dry Eye Epidemiology Study detected that 37% of the patients with DM had DES; Seifart et al. in Germany reported that 52.8% of the patients with DM had DES [1–2].

Jin et al. reported that patients with type 2 DM had DES [3]. On the other hand, Akinci et al. reported that symptoms of dry eye syndrome are more common while lacrimal secretion and lacrima film stability are lower in children with type 1 DM [4]. Although many papers have illustrated the correlation between DES and type 1 and 2 DM, there is no specific work showing the correlation between maturity-onset diabetes of young (MODY) and DES in the literature [1–4].

Ten percent of type 2 DM originates from MODY which arises from monogenetic defect [5–6]. It is estimated that MODY constitutes 20% of all diabetes cases especially in developed countries [6]. The most common presentation of MODY, which is generally detected under 25 years and commonly, childhood or adolescence period, is DM in consecutive generations and asymptomatic hyperglycemia in non-obese children. DM history in three or more generations, DM presentation in early age and non-being of obesity are thought to be MODY more likely than type 2 DM [6].

The purpose of our study was to examine ocular surface changes in 50 patients who were diagnosed with MODY. Our work is the first one which specifically evaluates ocular surface in MODY patients.

✉ Osman Celikay
ocelikay66@gmail.com

[1] Department of Ophthalmology, Diskapi Yildirim Beyazit Training and Research Hospital, Ankara, Turkey

[2] Department of Endocrinology, Diskapi Yildirim Beyazit Training and Research Hospital, Ankara, Turkey

[3] Genetic Diagnostic Center, Diskapi Yildirim Beyazit Training and Research Hospital, Ankara, Turkey

Material and method

This research has been done to evaluate the ocular surfaces of MODY patients who were transferred to Genetic Diagnosis Department of Diskapi Yildirim Beyazit Training and Research Hospital between January 2014 and January 2015. While MODY has been diagnosed by endocrine specialists (EC) and medical genetics specialists (FAP), DES has been diagnosed by ophthalmologists (OC, MK, CG). Fifty patients with MODY have been included in the research. Control group has been formed of patients who applied because of refractive errors, without any other ocular or systemic diseases, compatible to age and sex requirements. Patients with ocular surgery history, smokers, and users of contact lenses were not included in the research. Demographic data including age, sex, MODY diagnosis period, HBa1c levels, MODY gene mutations, ocular and systemic history has been collected. Only the right eyes of the patients have been examined. Inspections and measurements have been completed between 8.30 and 12.30. Fundus was examined by binocular indirect ophthalmoscopy.

Patients have been subjected to Ocular Surface Illness Index (OSDI) survey which composed of 12 questions (Table 1) [7]. Following, all the patients were subjected to a complete ophthalmologic inspection involving best corrected visual acuity (BCVA), shirmer test, invasive and non-invasive tear break up time (TBUT), cornea fluorescein, and Lissamine green dye tests. Lastly, mebiography measurements have been completed using Sirius Scheimpflug camera (Sirius, CSO, Florence, Italy).

OSDI is a survey composed of 12 questions and developed to evaluate the ocular irritation symptoms in compliance with dry eye and correlation between these symptoms and vision in a fast manner [8]. This survey consists of a scale from 0 to 100 and is proven to be a distinguishing test in terms of sensing the specifications between normal cases and cases with ocular surface problem. Patients were needed to specify the influence on them on a scale from 0 (never) to 4 (always). In our research, each patient's OSDI score has been calculated as:

OSDI = [(total score of the all answered questions) × 100] / [(number of questions) × 4].

According to the OSDI score, between 0 and 12 points were taken as normal, between 13 and 22 were light, 23–32 were moderate and 33–100 were heavy ocular surface disease [8].

Meibografi Sirius Scheimpflug Camera (Sirius, CSO, Florence, Italy) is used to evaluate Meibomian gland (MG) morphology with Phoenix-Meibography Monitoring Method which provides non-contact images. Digital analysis of Phoenix meibography module provides a classification of gland losses. Loss rates of meibomian glands in lower and upper palpebra of right eyes of all patients have been measured in meibography. Loss rate in meibomian gland is considered grade 0 if it is lower than 15%, grade 1 if between 15 and 33%, grade 2 if between 33 and 66%, grade 3 if higher than 66%.

Schirmer test has been completed without anesthesia and after conjunctival bladder attached to lower palpebra, the conjunction of one-third of the midline attached to strip, the eye has been kept closed for 5 min. After 5 min, the quantity of

Table 1 Ocular Surface Disease Index questions

	Never (0)	Rarely (1)	Sometimes (2)	Regularly (3)	Usually (4)
(1) Have you experienced any of the following conditions during the last week?					
Sensitivity to light in eyes					
Feeling of something rubbing against eye					
Pain or sensitivity in eye					
Blurry vision					
Weak sight					
(2) Have you experienced any problems related with eye while doing following activities during the last week?					
Reading					
Driving at night					
Working on computer/ Using ATM					
Watching TV					
(3) Have you experienced any discomfort in your eyes in the following situations during the last week?					
During windy weather					
Being places where the air is dry (low moisture)					
Unventilated spaces					

wetting in the strip was recorded in millimeters. Five millimeters/5 min and lower values of Schirmer test were considered as abnormal.

For the TBUT measurements, strips soaked of 1 mg of fluorescein (Visimed, İzmir, Turkey) was wetted and located on the lower palpebra's one third of lateral. After that, three measurements have been recorded between the first appearance of first corneal black spot and dyed tear film with full blinking. Averages of the measurements were calculated and measurements under 10 s were considered as abnormal. At the same time, TBUT has been evaluated as non-invasive with Meibography Sirius Scheimpflug camera (Sirius, CSO, Florence, Italy).

Cornea fluorescein dye has been evaluated under blue filter after tear break up time measurements. A staging system has been done using the instructions from the National Eye Institute Workshop [9]. The cornea has been split up to five zones: central, upper, temporal, nasal, and lower. Each zone has been evaluated according to its dyeing score from 0 to 3: 0 = normal; 1 = light or surface dotted; 2 = moderate severity or point dyeing with surface cornea abrasion; and 3 = severe abrasion or deep cornea abrasion. In that way, the maximum score has been decided to be 15.

Conjunctiva dyeing by lissamine green has been evaluated after a drop of 1% sodium lissamine. In conjunctiva dyeing, classification method identified by Bijsterveld is used: for scale between 0 to 3, three dials have been decided (temporal, central and nasal) [10–11]. According to the dyeing intensity, every dial has been given a score between 0 and 3. In that way, the maximum score has been determined to be 9.

Gene mutation analysis

Using the DNA Isolation Kit for Mammalian Blood (Roche Diagnostics, Indianapolis, USA), genomic DNA was isolated from the peripheric venous blood sample of patients. Exons and exon-intron neighboring zones in glucokinase (GCK), hepatocyte nuclear factor-1 alpha (HNF1A), hepatocyte nuclear factor-4 alpha (HNF4A), and hepatocyte nuclear factor-1 beta (HNF1B) genes were multiplicated using specific oligonucleotide primer sets in PCR. PCR products were sequenced directly and two-sided using Sanger sequence method in ABI Prism 3130XL Genetic Analyzer (ABI Applied Biosystems, Foster City, CA), according to the manufacturer's directions [12].

Statistical analysis

In the statistical analysis of the data, SPSS 18 for Windows (SPSS Inc., Chicago, ABD) package software was used. Continuous variables were designated as average ± standard deviation. For the pairwise comparison, the Mann-Whitney *U* test or independent sample *t* test was used. In the evaluation of the change of categoric variables, Ki-square test was used. Evaluations were completed within the 95% reliability range, and values of p lower than 0.05 were considered as statistically significant.

Results

A total of 50 patients were involved in the research. All of the patients had a glucokinase (GCK) mutation. While the average age of the 50 MODY patients was 25.6 ± 14.6 years (range: 8–58 years), the average age of the 50 patients in the control group was 26.6 ± 12.7 years (range: 8–59 years). MODY patients were composed of 13 males and 37 females. In the control group, 14 male and 36 females were included in the research ($p = 0.71$). There was no significant statistical difference in age and sex patterns between the two groups. The mean duration of diabetes after the diagnosis in the MODY group was 5.0 ± 5.5 years (range: 0–20 years). The mean diagnosis time was 25.6 years (range: 8–45 years).

While the average HBa1c levels were 5.5 (range: 4.8–6.5, reference range: 4–6) in MODY group, 4.9 (range: 4.4–5.6, reference range: 4–6) in the control group (Table 2). None of the patients in MODY group were diagnosed with diabetic retinopathy in fundus examination. Although none of the patients had been (were) on insülin, 19 of them had been (were) on oral hypoglycemic agents (OHA).

Cornea sensitivity of all the patients was found to be normal after an inspection conducted by a cotton-tip stick. According to the OSDI staging, it was found that 31 of them (62%) were in normal range, 10 of them (20%) were light, 3 of them (6%) were moderate, and 6 of them (12%) were severe in terms of ocular surface disease among MODY patients. In the control group, 20 of them (40%) were in normal range, 20 of them (40%) were light, 8 of them (16%) were moderate, and 2 of them (4%) were severe in terms of ocular surface disease. In the OSDI score, there was no significant difference between MODY patients and control group patients (13.41 ± 13.09 and 14.58 ± 10.34) ($p = 0.76$).

In the MODY group, while 13 of the patients had stage 0, 25 of them have 1, 12 of them had stage 2 MG loss on the upper palpebra, none of them has stage 3 MG loss. In the control group, 33 of the patients had stage 0, 11 of them had

Table 2 The baseline characteristics of the participants with MODY and the control group

	MODY group	Control group
Age (years)	25.6 ± 14.6 (range: 8–58)	26.6 ± 12.7 (range: 8–59)
Sex (W/M)	13/37	14/36
Mean HBa1c	5.5 (range: 4.8–6.5)	4.9 (range: 4.4–5.6)

MODY maturity onset diabetes of the young

stage 1, 4 of them had stage 2, and none of the patients had stage 3 MG loss on the upper palpebra. In the mebiography, MODY group had 12.04 ± 10.41 and control group had 9.3% ± 7.5 MG loss on upper palpebra on an average was not a significant statistical difference ($p = 0.16$) (Table 3).

There is no significant difference between MODY and control group in terms of Schirmer 1 test (29.7 ± 7.9 and 26 ± 8 mm, respectively) ($p = 0.16$). No patients had lower than 5 mm in Schirmer 1 test in both groups. While MODY group had 15 patients with TBUT result lower than 10 s or lower, control group had 6 patients lower than 10 s or lower. In MODY group, average TBUT was 10.56 ± 4.1 (range: 2–15) while the average was 12 ± 2.78 (range: 5–15) in control group ($p = 0.36$). Non-invasive tear break up time was 11.6 (range: 1.9–16.8) in MODY group, whereas in control group, it was 11.2 (range: 1.4–17.8) ($p = 0.83$) (Table 3).

Average score of fluorescein staining was 1.57 ± 2.89 in MODY group, while 0.17 ± 0.39 ($p = 0.09$) in control group. Average Lissamine green staining score was 0.63 ± 1.02 in MODY group; in control group, it was 0.22 ± 0.52 ($p = 0.18$).

Discussion

DM has been reported to be a risk factor for DES, and the prevalence rate for DES in DM has been reported as 54.3% [13]. DES decreases life quality because it leads to ocular discomfort and distorted vision. DES may lead to complications such as cornea epithelial defect, corneal erosion, and ulcers. This is why it is a continuously growing public health problem that needs to be diagnosed and treated early [13]. Jin et al. found that TBUT in type 2 DM patients was significantly short in comparison to the control group [3]. Goebels et al. found that the production of tears in DM patients through Schirmer test was significantly low [14]. Jain et al. did not detect any predisposing diseases except for DM in 80 (20%) out of 400 patients admitted to tertiary eye hospitals [15]. Contrary to the common concept that "dry eye is frequently seen in type 1 and type 2 DM," [14–16] we did not come across any significant difference between MODY and control group patients in ocular surface tests.

GCK, *HNF1A*, *HNF4A*, and *HNF1B* genes make up 95% of MODY patients whose genetic causes have been clarified. This is why we routinely looked at these four genes in patients who were pre-diagnosed with MODY. *GCK* has been detected to be the most mutating gene. In our series, all of the mutations have been detected in the *GCK* gene. Although it is proposed in the literature that patients with MODY caused by the GCK gene has low risk of micro and macro-vascular complications, there is no study that investigates the phenotype and genotype relationship in terms of eye symptoms [17–21]. We were not able to look at the phenotype-genotype relationship because all of the patients in our series had the GCK gene mutation.

In studies, decreased corneal sensitivity and increase in dry eye have been found to be correlated with DR. [22] Saito et al. reported that corneal sensitivity in DM patients has been decreased and this decrease was correlated with the DR phase [22]. For this reason, the more severe DR was, the more likely the patient was to have dry eye [23]. In our study, the fact that none of the patients had DR and that corneal sensitivity tested with cotton swabs was normal were in line with the normal ocular surface tests of MODY patients. It can be said that MODY progresses more moderately compared to other types of diabetes because of the fact that MODY patients did not have DR and that their dry eye tests were normal. It also can be linked to the fact that not enough time had passed for DR or DES symptoms to be detected.

When Kaiserman et al. compared the teardrop usage in 22,382 DM patients with non-DM population, they found teardrop usage in DM patients to be significantly high. A total of 20.6% of patients with diabetes and 13.8% of patients without diabetes used teardrops for DES [24]. In our study, the average age of MODY patients was 25.6 ± 14.6 years (interval: 8–58 years). Unlike other studies, the absence of dry eye symptoms in MODY patients in our study can be linked to the fact that MODY patients were young and, therefore, the clinical symptoms of diabetes had not started to show up yet.

Table 3 Test results of patients with young adult diabetes and control group patients

	YAD group 0	Control group	*P* value
OSDI score	13.41 ± 13.09	14.58 ± 10.34	$p = 0.76$
Mebiografi score (upper palpebra)	12,04 ± 10,41	10,43 ± 7.5	$p = 0,21$
Mebiografi scoru (lower palpebra)	11,3 ± 11.58	9,3 ± 7,5	$p = 0,16$
Schirmer 1 test	29,7 ± 7,9	26 ± 8,0	$p = 0.16$
TBUT	10.56 ± 4,1	12 ± 2,78	$p = 0.36$
Non-invasive TBUT	11.6 ± 2,1	11.2 ± 2,5	$p = 0.83$
Cornea dyeing with florescein	1,57 ± 2,89	0,17 ± 0,39	$p = 0.09$
Lisamin green dyeing score	0.63 ± 1.02	0,22 ± 0,52	$p = 0.18$

OSDI Ocular Surface Illness Index, *TBUT* tear break up time

When Seifart et al. compared type 1 and type 2 DM patients between the ages of 7 and 69 with a healthy control group, they found that 52.8% of DM patients and 9.3% of those healthy had DES. Also, they asserted that it is important to regularly check up on DM patients and regulate their blood sugar levels in order to prevent DES and retinopathy caused by diabetes [2]. In our study, patients had an average HBa1c of 5.5 (4.8–6.5; reference interval: 4–6). So, the fact that the MODY patients in our study had well diabetes regulation could be another reason why they did not show any dry eye symptoms. Apart from the regulation of diabetes, how long the patient has had diabetes is another important point. Because the prevalence of DR in patients who has had DM for less than 5 years was 28.8%, whereas the rate for those who had DM for more than 15 years was 78.8% [25]. Our patients' average diabetes diagnosis length [5.0 ± 5.5 years (0–20 years)] might be a short time span for clinical symptoms like DR and DES to show up.

Although no effect of MODY on ocular surface was detected in our study, since an increase of DES has been detected in type 1 and type 2 DM in the literature, DES symptoms such as burning and stinging need to be examined and dry eye tests must be performed. Since DM patients with decreased cornea sensitivity might have less dry eye symptoms than normal, we think that periodically performing dry eye tests will be useful.

As a conclusion, our study is important for being the first in the literature. Studies with patients who had been diagnosed with MODY for long term and with more patients should be done in order to better understand the effects of MODY on the ocular surface.

Compliance with ethical standards

Ethical approval Approval has been taken from the local ethics committee and research has been carried out in compliance with the Helsinki Declaration. All the patients have been informed about the research and their informed consent has been received.

Conflict of interest The authors declare that they have no conflict of interest.

References

1. Caffery BE, Richter D, Simpson T, Fonn D, Doughty M, Gordon K. The Canadian dry eye epidemiology study. Adv Exp Med Biol. 1998;438:805–6.
2. Seifart U, Strempel I. The dry eye and diabetes mellitus. Ophthalmologe. 1994;91:235–9.
3. Jin J, Chen LH, Liu XL, Jin GS, Lou SX, Fang FN. Tear film function in non insulin dependent diabetics. Zhonghua Yan Ke Za Zhi. 2003;39:10–3.
4. Akinci A, Cetinkaya E, Aycan Z. Dry eye syndrome in diabetic children. Eur J Ophthalmol. 2007;17:873–8.
5. Pajans SS. GED: a model for understanding the pathogeneses and natural history of type II diabetes. Horm Metab Res. 1987;19:591–9.
6. Ledermann HM. Maturity-onset diabetes of the young (GED) at least ten times more common in Europe than previously assumed? Diabetologia. 1995;138:1482.
7. Schiffman RM, Christianson MD, Jacobsen G, Hirsch JD, Reis BL. Reliability and validity of the Ocular Surface Disease Index. Arch Ophthalmol. 2000;118:615–21.
8. Miller KL, Walt JG, Mink DR, Satram-Hoang S, Wilson SE, Perry HD, et al. Minimal clinically important difference for the ocular surface disease index. Arch Ophthalmol. 2010;128:94–101.
9. Lemp MA. Report of the National Eye Institute/Industry workshop on clinical trials in dry eyes. CLAO J. 1995;21:221–32.
10. Berntsen DA, Mitchell GL, Nichols JJ. Reliability of grading lissamine green conjunctival staining. Cornea. 2006;25:695–700.
11. Van Bijsterveld OP. Diagnostic tests in the sicca syndromes. Arch Ophthalmol. 1969;82:10–4.
12. Sanger F, Nicklen S, Coulson AR. DNA sequencing with chain-terminating inhibitors. Proc Natl Acad Sci. 1977;74:5463–7.
13. ManaviatMR RM, Afkhami-Ardekani M, Shoja MR. Prevalence of dry eye syndrome and diabetic retinopathy in type 2 diabetic patients. BMC Ophthalmol. 2008;8:10.
14. Goebbels M. Tear secretion and tear film function in insulin dependent diabetics. Br J Ophtalmol. 2000;84:19–21.
15. Jain S. Dry eyes in diabetes. Diabetes Care. 1998;21:1364–82.
16. Uyanik FY, Cetin EN, Yaylali V, Avunduk AM, Yildirim C. Diyabetik Hastalarda Oküler Yüzey Sağlığı ve Kuru Göz. Turkiye Klinikleri J Ophthalmol. 2011;20:204–11.
17. Matschinsky FM, Randle PJ. Evolution of the glucokinase glucose sensor paradigm for pancreatic beta cells. Diabetologia. 1993;36:1215–7.
18. Froguel P, Zouali H, Vionnet N, Velho G, Vaxillaire M, Sun F, et al. Familial hyperglycemia due to mutations in glucokinase: definition of a subtype of diabetes. N Engl J Med. 1993;328:697–702.
19. Cerf ME. Transcription factors regulating beta-cell function. Eur J Endocrinol. 2006;155:671–9.
20. Kapoor RR, Locke J, Colclough K, Wales J, Conn JJ, Hattersley AT, et al. Persistent hyperinsulinemic hypoglycaemia and maturity-onset diabetes of the young due to heterozygous HNF4A mutations. Diabetes. 2008;57:1659–63.
21. Bellanné-Chantelot C, Chauveau D, Gautier JF, Dubois-Laforgue D, Clauin S, Beaufils S, et al. Clinical spectrum associated with hepatocyte nuclear factor-1beta mutations. Ann Intern Med. 2004;140:510–7.
22. Saito J, Enoki M, Hara M, Morishige N, Chikama T, Nishida T. Correlation of corneal sensation, but not of basal or reflex tear secretion, with the stage of diabetic retinopathy. Cornea. 2003;22:15–8.
23. Ozdemir M, Ozdemir G, Buyukbese MA, Cetinkaya A. Diyabetes Mellitusta Gözyaşı Fonksiyonu ve Oküler Yüzey Değişiklikleri. Turk J Ophthalmol. 2004;33:291–4.
24. Kaiserman I, Kaiserman N, Nakar S, Vinker S. Dry eye in diabetic patients. Am J Ophthalmol. 2005;139:498–503.
25. Klein R, Klein BE, Moss SE, Davis ME, DeMets DL. The Wisconsin epidemiologic study of diabetic retinopathy. III. Prevalence and risk of diabetic retinopathy when age at diagnosis is 30 or more years. Arch Ophtalmol. 1984;102:4527–32.

International Journal of Diabetes in Developing Countries (2021) 41:141–147
https://doi.org/10.1007/s13410-020-00855-y

ORIGINAL ARTICLE

Assessment of nutritional status and quality of life in individuals with and without diabetes over 65 years of age

Gulseren Pamuk[1] · Gulsah Kaner[2] · Esra Meltem Koc[1] · Tuncay Toşur[1] · Sumeyra Dasdelen[1] · Saliha Aksun[3] · Baris Önder Pamuk[4]

Received: 16 February 2020 / Accepted: 27 July 2020 / Published online: 17 August 2020

Abstract

Objective The aim of this study is to evaluate the nutritional status of individuals with and without diabetes over the age of 65 and its effects on the quality of life.

Materials and methods A total of 188 subjects, 94 diabetic individuals and 94 non-diabetic individuals as the control group, were included in the study. Both groups were surveyed using the mini nutritional assessment short form (MNA-SF) scale, Europen quality of life 5 dimensions-3 level version (EQ-5D-3L) quality of life scale and socio-demographic data questionnaire. Height, weight, and body mass index (BMI) were taken for anthropometric measurements. The levels of biochemical parameters were measured.

Results The mean age of the participants was 70.34 ± 4.63 years, and 64.9% were female. In total, 4.3% of patients with diabetes had malnutrition and 37.2% were at risk of malnutrition. While malnutrition was not detected in patients without diabetes, 26.6% were at risk. In terms of malnutrition, a statistically significant difference was found between the two groups ($p = 0.027$). The EQ-5D-3L quality of life index score was lower in patients with diabetes ($p = 0.025$).

Conclusion Identifying and following up on malnutrition status in elderly diabetes patients are of importance with regard to disease control and quality of life.

Keywords Diabetes · Nutrition · Quality of life · Malnutrition

Introduction

Today, with the growing elderly population, healthy aging is gaining gradually increasing importance. While the population over 65 years of age constituted 5% of the world population in 1960, in 2018, this ratio increased to 9%. In the projection for 2050, the population over 65 is estimated to constitute 16% of the world population [1].

Together with that, diabetes in aging populations is a growing public health problem. Elderly individuals form one of the fastest-growing fractions of the diabetes population. With life expectancy on the rise, the management of diabetes in elderly

✉ Gulseren Pamuk
gulseren.pamuk@ikc.edu.tr

Gulsah Kaner
dytgulsah@gmail.com; kanergulsah@gmail.com

Esra Meltem Koc
meltemnuzumlali@yahoo.com

Tuncay Toşur
tuncaytosur@mynet.com

Sumeyra Dasdelen
sumeyra.dasdelen@saglik.gov.tr

Saliha Aksun
salihaaksun@yahoo.com

Baris Önder Pamuk
bopamuk@gmail.com

[1] Faculty of Medicine, Department of Family Medicine, İzmir Katip Çelebi University, İzmir, Turkey

[2] Health Sciences Faculty, Department of Nutrition and Dietetics, İzmir Katip Çelebi University, İzmir, Turkey

[3] Faculty of Medicine, Department of Medical Biochemistry, İzmir Katip Çelebi University, İzmir, Turkey

[4] Faculty of Medicine, Department of Endocrinology, İzmir Katip Çelebi University, İzmir, Turkey

individuals has become increasingly critical. Diabetes is associated with malnutrition and other geriatric syndromes according to the medical literature [2]. Assessment of nutritional status in the elderly has significance in terms of early diagnosis of nutritional deficiency and its complications, as well as the prognosis of existing diseases. Early symptoms of malnutrition are nonspecific and develop slowly. Since the effects of malnutrition can easily be confused with signs of aging, the condition is difficult to diagnose initially [3].

Diabetes in young adults often co-exists with obesity. Therefore, in addition to medication, a change in the quality of life, including low-calorie diets, is recommended in the treatment of the disease [4].

In the literature, the incidence of malnutrition in elderly people with diabetes ranges from 3.7 to 21.2% [2–5]. Low-calorie and diabetes-focused diets prescribed only for the treatment of diabetes can lead to an increased risk of malnutrition, especially in the elderly. Therefore, malnutrition status should be checked up in elderly individuals with diabetes [2]. In the literature, there are a limited number of studies investigating malnutrition in elderly individuals with diabetes, and the majority of these studies have been conducted with hospitalized patients. Also, there are few publications in the literature that link the presence of malnutrition with adverse effects on the quality of life [6]. As an important health indicator, quality of life defines the subjective perception of one's own health in the sociocultural environment in which one lives. The presence of nutrition disorders, old age, and chronic diseases such as diabetes are factors affecting the quality of life [7]. In this respect, the evaluation of elderly and diabetic individuals regarding the nutritional disorders and malnutrition is important in preserving the well-being and quality of life of elderly individuals.

Materials and methods

This descriptive cross-sectional study was conducted between November 2017 and September 2018 at the Endocrinology Outpatient Clinic of İzmir Katip Çelebi University Faculty of Medicine Hospital. The sample size was calculated using the Open Source Epidemiologic Statistics for Public Health (OpenEpi) sample size calculator. When we accepted malnutrition frequency as 21% among diabetic patients over 65 and 6% in the general elderly population, a total sample size of 188 people, with a minimum number of 94 people for each group, was calculated with 95% confidence interval, 5% margin of error, and 80% power.

The study included volunteers over 65 years of age, who applied to the hospital for any reason, with type 2 diabetes diagnosis for at least 1 year, and gave verbal and written consent to participate in the study, and individuals of similar age and characteristics without diabetes. Individuals with advanced dementia that causes communication problems, those who have severe psychological disorders and difficulties in understanding and listening, and those who have other chronic diseases than diabetes that may cause malnutrition such as infection and malignancy were excluded from this study.

All subjects included in the study were surveyed using the face-to-face interview technique. The participants were given three forms: a socio-demographic data questionnaire prepared by researchers evaluating their general characteristics (age, gender, educational status, marital status, family history, etc.), diseases, treatments (medication, insulin, vitamin–mineral), mini nutritional assessment short form (MNA-SF) scale evaluating their nutritional status, and Europen quality of life 5 dimensions-3 level version (EQ-5D-3L) quality of life scale to evaluate the overall quality of life. Anthropometric measurements (height, body weight, body mass index (BMI) calculation) of the participants were conducted by the researchers. Blood samples were collected in order to test the fasting plasma glucose (FPG), serum albumin, prealbumin, calcium (Ca), hemoglobin A_{1c} (HbA_{1c}), the lipids (total cholesterol, triglyceride, high-density lipoproteins (HDL), low-density lipoproteins (LDL)).

Assessment of nutritional status

The MNA-SF scale is a screening tool to help identify elderly patients who are malnourished or at risk of malnutrition. The MNA-SF scale consists of six parts to assess food intake loss, weight loss, mobility, physical stress or acute illness, cognitive status, and BMI. If BMI could not be determined, it was replaced with calf circumference [8].

Each question is scored from zero to two or three, for a maximum score of 14. A total score of MNA-SF 7 and lower was considered as malnutrition, 8 to 11 as the risk of malnutrition, and 12 to 14 as a normal nutritional status [9, 10].

Assessment of quality of life

The researchers used EQ-5D-3L to measure the patients' overall quality of life. The EQ-5D-3L questionnaire is a self-reported measure developed by EuroQoL that evaluates five dimensions with five questions. These five dimensions are mobility, self-care, usual activities, pain/discomfort, and anxiety/depression. Each dimension provides a range for measurement: mobility from walking around to being confined to bed, self-care from taking care of oneself to being unable to wash or dress oneself, usual activities from performing usual activities to being unable to perform, pain/discomfort from no pain or discomfort to extreme pain or discomfort, and anxiety/depression from not feeling anxious or depressed to feeling extremely anxious or stressed. Each item is evaluated using a 3-point Likert scale: no problems, some

problems, and extreme problems (labeled 1–3). The respondent is asked to indicate her/his health state by checking the box against the most appropriate statement in each of the five dimension. The health benefit index score can be calculated for each dimension, in addition to obtaining the response distribution to these dimensions [11].

There is a visual analog scale in the questionnaire which allows for a response from 0 to 100, that is, from "the worst imaginable health" to "the best imaginable health" [11].

The study of determining reliability and validity and norms of society for Turkish was performed by Süt et al. [11]. Süt et al. performed the evaluation for the reliability and validity of the Turkish version of the instrument and the norm determination study [12].

Anthropometric measurements and bioimpedance analysis

Patients' weight was measured without shoes or excessive clothing, using a Tanita SC 330 bioimpedance analysis (BIA) scale. The individuals were asked to fast and to limit the consumption of the liquids prior to measurement (at least for 4 h) and to remove all the metals in contact with their skin.

Height was measured with a stadiometer while the participants were standing against the wall, feet together on the floor, with head in the Frankfurt horizontal plane (the lower border of the eye orbit in line with the auditory meatus and parallel to the floor) [13]. Lower leg length was measured for the old participants with spinal deformity. The calculation of body mass index (kg/m^2) for all participants was made using the weight/height^2 equation [11]. The values for BMI were evaluated with cut-off values recommended by the World Health Organization (WHO) [14].

Biochemical tests

A total of 8–10 mL of venous blood was collected from the participants and centrifuged at 3000 rpm for 10 min under sterile conditions to separate the serums. The serums were stored in clean and dry Eppendorf tubes at − 20 °C in the freezer until the time of the study. The spectrophotometric method was utilized to analyze the FPG, albumin, prealbumin, Ca, lipids (total cholesterol, triglyceride, HDL, LDL), and HbA_{1c} levels. The study also included hemolyzed and lipemic sample.

Statistical method

Statistical analysis of the study was performed using Statistical Package for the Social Sciences (SPSS) 16.0. Descriptive analyses were presented with mean ± standard deviation; median (the minimum and maximum value) for numerical variables; and numbers, ratios, and percentages for categorical variables. The normality of the distribution was tested by visual (histogram and probability) and analytical methods (Kolmogorov–Smirnov test). Chi-square and Mann–Whitney *U* tests were used in accordance with the variables for between-group comparisons. The association between the groups was analyzed with Spearman's correlation and multiple linear regression methods. A p value lower than 0.05 was considered as statistically significant.

Results

A total of 188 people participated in the study, 94 of whom were diagnosed with diabetes. The mean age of the participants was 70.34 ± 4.63 years (min: 62, max: 87), and 64.9% ($n = 122$) were female. The average age of men was found to be statistically higher than that of women ($p = 0.045$; female 69.84 ± 4.57, male 71.27 ± 4.62). In terms of age, there was no difference between those with diabetes and those without ($p > 0.05$). The socio-demographic characteristics of the participants stratified by the presence of diabetes are summarized in Table 1.

In the study, 64.9% ($n = 61$) of the participants with diabetes had the disease for more than 10 years, and the average age of being diagnosed with diabetes was 57.03 ± 9.23. In total, 46.8% ($n = 44$) of the participants with diabetes received oral anti-diabetic, 29.8% ($n = 28$) insulin, and 20.2% ($n = 19$) both insulin and oral anti-diabetic, while 3.2% ($n = 3$) applied lifestyle changes as a treatment option.

A total of 85.6% ($n = 161$) of the participants were obese and overweight (BMI > 25), while 57.1% ($n = 107$) had BMI ≥ 30. There was no difference in BMI between those with and without diabetes ($p > 0.05$)

Nutritional status

According to the MNA-SF scale, 66% ($n = 124$) of the participants had a normal nutritional status, 31.9% ($n = 60$) were under risk, and 2.1% ($n = 4$) had malnutrition.

Among individuals with diabetes, 4.3% of participants had malnutrition and 37.2% were under risk. No case of malnutrition was detected in participants without diabetes, while 26.6% of them were at risk of malnutrition. In terms of malnutrition status, a statistically significant difference was found between the two groups ($p = 0.027$) (Table 2).

While 64.9% ($n = 61$) of patients with diabetes were diagnosed more than 10 years ago, 4.9% ($n = 3$) of them had malnutrition and 39.3% ($n = 24$) were under risk. On the other hand, 3% ($n = 1$) of the patients with diabetes for less than 10 years were malnourished, and 33.3% were under risk ($n = 11$). There was no statistically significant difference between diabetes duration and malnutrition status ($p > 0.05$).

According to MNA-SF scale scores, there was no significant relationship between age, gender, level of education,

Table 1 Socio-demographic characteristics of participants based on diabetes status

	Individuals with diabetes	Individuals without diabetes	*p*
Age (mean ± SD)	70.26 ± 4.40	70.42 ± 4.86	0.898
Gender *n* (%)			
Female	55 (58.5)	67 (71.3)	0.067
Male	39 (41.5)	27 (28.7)	
Marital status *n* (%)			
Married	61 (64.9)	61 (64.9)	0.068
Divorced/widow	27 (28.7)	28 (29.8)	
Single	6 (6.4)	5 (5.3)	
Level of education *n* (%)			
Illiterate	12 (12.8)	22 (23.4)	0.947
Elementary	50 (43.2)	48 (51.1)	
Secondary	13 (13.8)	4 (4.3)	
High school	9 (9.69)	10 (10.6)	
University	10 (10.6)	10 (10.6)	
Place of residence *n* (%)			
Alone	20 (21.3)	20 (21.3)	0.887
With family	74 (78.7)	74 (78.7)	

marital status, and living alone or with family ($p > 0.05$). There was no correlation between age and malnutrition in both groups with and without diabetes (Spearman's correlation: diabetic r: -0.036, $p > 0.05$; non-diabetic r: -0.090, $p > 0.05$) (Table 3).

Quality of life

The comparison of the EQ-5D-3L index score between the participants with and without diabetes determined that the quality of life of patients with diabetes was lower. When each parameter of the EQ-5D-3L scale is compared between the two groups, the quality of life is found to be statistically significantly lower in the group with diabetes ($p < 0.05$) with respect to the scores of self-care, usual activities, and anxiety/depression, while there was no significant difference in terms of mobility and pain/discomfort score despite the lower scores in the diabetic ($p > 0.05$) (Table 4). There was no significant relationship between the EQ-5D-3l scale and the duration of diabetes found ($p > 0.05$).

When the relationship between age variable and quality of life was evaluated, there was a low-level, significant correlation between EQ-5D-3L index, self-care score, usual activities score, and anxiety/depression score, indicating decreased quality of life as age increased ($p < 0.05$). According to EQ-5D-3L index scores, there was no significant relationship between age, gender, level of education, marital status, and living alone or with family ($p > 0.05$).

The relationship between nutritional status and quality of life

The evaluation of the association between the MNA-SF score and the EQ-5D-3L index scale determined a weak positive correlation ($p < 0.001$, $r = 0.356$). Similarly, when the MNA-SF score was evaluated with each parameter of the EQ-5D-3L scale, the correlation was weak ($p < 0.001$) (Table 5).

In order to assess the effect of nutrition on quality of life, multiple linear regression analysis was performed, and the relationship between MNA-SF scores and EQ-5D-3L index scores was evaluated by adjusting the model by age, gender, and presence of diabetes. Accordingly, nutritional status has a positive effect on the quality of life, $\beta = 0.037$ (95% CI: 0.018–0.057, $p < 0.001$). Diabetes mellitus (DM) had a negative effect on quality of life, $\beta = -0.079$ (95% GA: -0.159–0.000, $p = 0.049$) for the presence of diabetes (Table 6).

Table 2 Evaluation of malnutrition with the MNA-SF scale in groups with and without diabetes

MNA-SF scale	Individuals with diabetes *n* (%)	Individuals without diabetes *n* (%)	*p*
Normal nutrition	55 (58.5)	69 (73.4)	0.027
Risk of malnutrition	35 (37.2)	25 (26.6)	
Malnutrition	4 (4.3)	0 (0)	

Table 3 The relationship between the clinical characteristics of individuals with diabetes and their malnutrition status

Individuals with diabetes over 65 years of age	Malnutrition	Risk of malnutrition	Normal nutritional status	*p*
Age (mean ± SD)	74.25 ± 6.55	70.88 ± 4.92	69.58 ± 3.73	0.242
Gender female, *n* (%)	2 (3.6)	21 (38.2)	32 (58.2)	0.926
Diabetes duration (mean ± SD)	11.75 ± 8.05	13.48 ± 8.40	13.18 ± 8.59	0.948
BMI (> 30) (mean ± SD)	23.93 ± 5.58	30.72 ± 4.98	30.49 ± 4.64	0.153
Waist circumference (mean ± SD)	94.33 ± 13.65	105.46 ± 8.13	105.15 ± 13.90	0.303
FPG (mean ± SD)	84.00 ± 42.79	143.71 ± 80.04	136.89 ± 84.06	0.502
Albumin (mean ± SD)	3.86 ± 0.75	4.19 ± 0.47	4.37 ± 0.38	0.131
Prealbumin (mean ± SD)	19.00 ± 8.67	19.97 ± 5.03	22.69 ± 5.18	0.190
Calcium (mean ± SD)	9.06 ± 0.87	9.34 ± 0.60	9.42 ± 0.68	0.629
HbA1c (mean ± SD)	7.17 ± 1.41	7.37 ± 1.80	7.24 ± 1.82	0.934

Discussion

The incidence of malnutrition increases in individuals over the age of 65 due to diabetes and diabetes-related complications [5]. In this study, the malnutrition status in elderly diabetic individuals and the relationship between malnutrition and quality of life were investigated. As a result of our study, the risk and incidence of malnutrition in diabetic individuals were higher compared with that in the control group. Another outcome of the study was that malnutrition and diabetes had a negative effect on the quality of life.

A total of 66% of the participants ($n = 124$) were in normal nutritional status, 2.1% ($n = 4$) were malnourished, and 31.9% ($n = 60$) were under risk of malnutrition. In total, 4.3% of diabetic patients were malnourished, and 37.2% were under risk, whereas in non-diabetic individuals, no incidence of malnutrition was found, and 26.6% were at risk. According to a study conducted in Spain using the MNA-SF scale, 1014 elderly diabetic individuals admitted to an inpatient clinic had a mean age of 77.9, and the incidence of malnutrition was 6.7% [15]. In a study conducted by Liu et al., including 302 diabetic individuals with the average age of 80 years who received inpatient treatment, the incidence of malnutrition was 18.5% and the risk of malnutrition was 33.1% [16]. In both of these studies, hospitalization duration was closely associated with nutritional status. In another study, conducted by Alfonso-Rosa et al. with 42 individuals with diabetes, 14% were malnourished, and 78% were at risk [17]. Similarly, in a study conducted by Kanwa et al. with 200 diabetic individuals, malnutrition incidence was found to be 14% and the risk of malnutrition was 33.5% [18]. These results display that the incidence of malnutrition increases in individuals over the age of 65 due to diabetes and diabetes-related complications [16]. The comparison of our results with the studies in the literature showed that the presence of malnutrition was lower in our study, even though similar results were obtained in terms of malnutrition risk. This may be due to the fact that these studies have been conducted with individuals under inpatient treatment in a hospital setting. Additionally, many of these studies had a higher mean age compared with our study, and they reported that the incidence of malnutrition escalates as the age increases. Indeed, Yıldırım et al. conducted a study involving 104 diabetes patients with an average age of 65 years who received inpatient treatment due to poor glycemic control, and they obtained results similar to ours in that the incidence of malnutrition was 4.8% and the risk of malnutrition 24% [19]. Similarly, in a study conducted with 246 diabetic individuals, malnutrition incidence was found to be 3.7% and the risk of malnutrition was 15.9% [5].

Table 4 Comparison of EQ-5D-3L quality of life scale scores of the groups with and without diabetes

EQ-5D-3L overall quality of life score	Individuals with diabetes (mean ± SD)	Individuals without diabetes (mean ± SD)	*p*
Mobility score	1.62 ± 0.48	1.52 ± 0.50	0.14
Self-care score	1.34 ± 0.49	1.15 ± 0.36	0.02
Usual activities score	1.42 ± 0.53	1.25 ± 0.43	0.05
Pain/discomfort score	1.72 ± 0.61	1.68 ± 0.55	0.47
Anxiety/depression score	1.54 ± 0.66	1.34 ± 0.49	0.02
VAS score	56.96 ± 13.78	61.01 ± 15.78	0.082
EQ-5D-3L index score	0.63 ± 0.32	0.74 ± 0.21	0.025

Table 5 The association between MNA-SF scale scores and the EQ-5D-3L index

	MNA-SF score	
	r	*p*
VAS score	0.332	< 0.001
Mobility score	− 0.275	< 0.001
Self-care score	− 0.299	< 0.001
Usual activities score	− 0.274	< 0.001
Pain/discomfort score	− 0.237	0.01
Anxiety/depression score	− 0.293	< 0.001
EQ-5D-3L index score	0.356	< 0.001

r, Spearman's correlation coefficient

$p < 0.05$

In total, 34% of the participants in our study were at nutritional risk. In their study conducted with 2327 elderly individuals using the MNA-SF scale, Ülger et al. found the risk of malnutrition to be 28%, as they considered the scores below 11 to be a risk of malnutrition. In the same study, the incidence of diabetes in individuals with and without nutritional risk was respectively 28.7% and 24.1% [20]. In our study, 60.9% of those at nutritional risk had diabetes, while 44.4% of those who were not at nutritional risk had the disease, and the difference between the groups with and without diabetes was significant. The reason why the figures in our study were higher could be due to the fact that 94 participants in our study were diabetes patients followed up by endocrinology outpatient clinics.

Many of the studies investigating the relationship between diabetes and malnutrition in the literature have evaluated nutritional status only in groups with diabetes. There are a limited number of studies conducted with a control group. In our study, the presence of malnutrition and the risk of malnutrition were evaluated together, and the frequency was 41.5% and 26.6% respectively in individuals with and without diabetes. A study by Turnbull et al. on individuals with and without diabetes found significant differences between the two groups, similar to our study [21]. In another study involving a total of 100 people with and without diabetes, malnutrition risk and malnutrition were evaluated together, and similar to our results, malnutrition incidence was 58% in people with diabetes and 22% in those without diabetes [22].

Table 6 Linear regression analysis with respect to the factors affecting quality of life

Variables	β	95% confidence interval		*p*
		Lower	Upper	
Age	− 0.007	− 0.015	0.002	0.120
Gender	− 0.032	− 0.112	0.049	0.443
DM	− 0.064	− 0.143	0.014	0.108
MNA-SF score	0.037	0.018	0.056	0.001

Our study obtained significant results that are similar to other studies in the literature in terms of malnutrition risk and malnutrition in diabetic individuals, and showed the importance of malnutrition in individuals with diabetes. On the other hand, some studies in the literature determined the incidence of malnutrition in individuals with diabetes higher than our study. However, many of these studies involved diabetic individuals who received inpatient treatment, and the duration of hospitalization was shown to negatively affect nutritional status. We believe that the incidence of malnutrition was lower than that in the literature because the participants in our study were individuals who visited the outpatient clinic. In addition, many studies have reported an increased risk of malnutrition as age increases. In numerous studies analyzing the relationship between diabetes and malnutrition, the mean age of the participants is higher than that of the individuals involved in our study. This may have resulted in a lower incidence of malnutrition compared with other studies [15, 16].

When we evaluated the relationship between diabetes and quality of life, we found a significant correlation indicating a lower quality of life in patients with diabetes. Many studies in the literature reported a lower quality of life in individuals with diabetes in concordance with our study [23, 24].

When we examined the MNA-SF score and EQ-5D-3L quality of life, a significant correlation indicated a lower quality of life for individuals with malnutrition. Likewise, Alfonso-Rosa et al. found a similar correlation in their study with individuals with first-stage diabetes [17].

In another study where malnutrition status was evaluated in individuals with and without diabetes, MNA-SF scores and quality of life were found to be significantly correlated, similar to our study [21]. In addition, when we evaluated the factors affecting the quality of life, we determined that the presence of malnutrition and diabetes negatively affected the quality of life.

Conclusion

Diabetes is a chronic disease frequently observed in older people, so malnutrition and its negative consequences are quite common in elderly diabetic individuals. For this reason, it is possible to improve the quality of life of diabetic patients by early assessment of malnutrition status and necessary measures.

Limitations

The cross-sectional nature of the study caused limitations in interpreting the causes of malnutrition. Therefore, incorporating diabetes-related complications may constitute superior evidence in evaluating this situation.

Funding information The study was funded by the İzmir Katip Çelebi University Scientific Research Projects Unit.

Compliance with ethical standards

The İzmir Katip Çelebi University Non-Interventional Clinical Research Ethics Committee granted the ethics committee approval with the approval date 17.11.2016 and number 122.

Conflict of interest The authors declare that they have no conflict of interest.

References

1. World Population Data (2018) http://www.worldpopdata.org/
2. Sanz-París A, García-Almeida JM, Gomez-Candela C, Burgos-Pelaez R, Martín-Palmero A, Matía-Martin P, et al. Malnütrition prevalance in hospitalized elderly diabetic patients. Nutr Hosp. 2013;28(3):592–9.
3. Rakicioğlu N. The approaches of medıcal nutrıtıonal therapy for dıabetıc elderly. Turk J Geriatr. 2006;9(1):52–9.
4. Standards of Medical Care in Diabetes—2018 Diabetes Care 2018 Jan; 41(Supplement 1): S1-S2.https://doi.org/10.2337/dc18-Sint01
5. Saintrain MVL, Sandrin RLSP, Bezerra CB, Lima AOP, Nobre MA, Braga DRA. Nutritional assessment of older adults with diabetes mellitus. Diab Res Clin Pract. 2019;155:107819.
6. Bakker MH, Vissink A, Spoorenberg SLW, Jager-Wittenaar H, Wynia K, Visser A. Are edentulousness, oral health problems and poor health-related quality of life associated with malnutrition in community-dwelling elderly (aged 75 years and over)? A cross-sectional study. Nutrients. 2018;10(12):E1965. https://doi.org/10.3390/nu10121965.
7. Grandy S, Fox KM, SHIELD Study Group. Change in health status (EQ-5D) over 5 years among individuals with and without type 2 diabetes mellitus in the SHIELD longitudinal study. Health Qual Life Outcomes. 2012;10:99. https://doi.org/10.1186/1477-7525-10-99.
8. Rubenstein LZ, Harker JO, Salvà A, et al. Screening for undernutrition in geriatric practice: developing the short-form mini-nutritional assessment (MNA-SF). J Gerontol A Biol Sci Med Sci. 2001;56(6):M366–72.
9. Kaiser MJ, Bauer JM, Ramsch C, Uter W, Guigoz Y, Cederholm T, et al. Validation of the mini nutritional assessment short form (MNA-SF): a practical tool for identification of nutritional status. J Nutr Health Aging. 2009;13:782–8.
10. Guigoz Y. The mini nutritional assessment (MNA) review of the literature-what does it tell us? J Nutr Health Aging. 2006;10:466–85 Discussion 485-467.
11. The EuroQol group. EQ-5D ınstruments. https://euroqol.org/eq-5d-instruments/eq-5d-5l-about/
12. Sut KH, Unsal S. Is EQ-5D a valid quality of life instrument in patients with acute coronary syndrome? The Anatolion. J Cardiol. 2011;1:156–62.
13. Baysal A, Aksoy M, Besler HT, Bozkurt N, Keçecioğlu S, Kutluay Merdol T, Pekcan G, Mercanlıgil SM, Yıldız (Haz.) E (2008) Diyet El Kitabı (yenilenmiş 5. bs.) (s67-141). Ankara: Hatipoğlu Yayınevi
14. Global Database on Body Mass Index. http://apps.who.int/bmi/index.jsp
15. Sanz-París A, Gomez-Candela C, Martín-Palmero A, García-Almeida JM, Burgos-Pelaez R, Matía-Martin P, et al. Application of the new ESPEN definition of malnutrition in geriatric diabetic patients during hospitalization: a multicentric study. Clinical Nutr. 2016;35(6):1564–7.
16. Liu G-X, Chen Y, Yang Y-X, Yang K, Liang J, Wang S, et al. Pilot study of the mini nutritional assessment on predicting outcomes in older adults with type 2 diabetes. Geriatr Gerontol Int. 2017;17:2485–92.
17. Alfonso-Rosa RM, Del Pozo-Cruz B, Del Pozo-Cruz J, Del Pozo-Cruz JT, Sañudo B. The relatıonshıp between nutrıtıonal status, functıonal capacıty, and health-related qualıty of lıfe in older adults with type 2 dıabetes: a pılot explanatory study. J Nutr, Health Aging. 2013;17(4):315–21.
18. Kanwa S, Qidwai W, Nanji K. Relationship of nutritional status and functional capacity in elderly patients visiting outpatient clinics of a tertiary care hospital. J Coll Phys Surg Pak. 2018;28(7):509–13.
19. Yildirim ZG, Mehmet UM, Ozge Telci Caklili OT, Hasan Huseyin Mutlu HH, Oguz A. Malnutrition rate among hospitalized patients with type 2 diabetes mellitus. Progr Nutr. 2018;20(2):183–8.
20. Ulger Z, Halil M, Kalan I, Yavuz BB, Cankurtaran M, Gungor E, et al. Comprehensive assessment of malnutrition risk and related factors in a large group of community-dwelling older adults. Clin Nutr. 2010;29(4):507–11.
21. Turnbull PJ, Sinclair AJ. Evaluation of nutritional status and its relationship with functional status in older citizens with diabetes mellitus using the mini nutritional assessment (MNA) tool-a preliminary investigation. J Nutr Health Aging. 2002;6(3):185–9.
22. Ellinger V, Carbonell A. More to look into the nutrition of elderly diabetic patients. J Diab Metab Disord Control. 2014;1(3):47.
23. Ayman A, Hayek A, Robert AA, Al Saeed A, Alzaid AA, Al Sabaan FS. Factors associated with health-related quality of life among Saudi patients with type 2 diabetes mellitus: a cross-sectional survey. Diab Metab J. 2014;38:220–9.
24. Saleh F, Ara F, Mumu SJ, Hafez MA. Assessment of health-related quality of life of Bangladeshi patients with type 2 diabetes using the EQ-5D: a cross-sectional study. BMC Res Notes. 2015;8:497.

International Journal of Diabetes in Developing Countries (2021) 41:148–155
https://doi.org/10.1007/s13410-020-00848-x

ORIGINAL ARTICLE

Geriatric syndromes and the cumulative impacts on quality of life in older people with type 2 diabetes mellitus

Ting Liu[1] · Xiaoyun Man[2] · Xiuxin Miao[1]

Received: 15 March 2020 / Accepted: 2 July 2020 / Published online: 21 July 2020

Abstract

Objective Older people with diabetes commonly experience geriatric syndromes that affect health. Little is known about the occurrence of geriatric syndromes and the impacts on quality of life in Chinese older people with type 2 diabetes mellitus (T2DM). This study aimed to understand geriatric syndrome and its association with quality of life among this population.

Methods A descriptive, cross-sectional study design was used. A total of 397 older people with diabetes were consecutively recruited from geriatric and endocrinology departments of a tertiary hospital using convenience sampling. Data on seven specific geriatric syndromes and quality of life were collected through checking medical records and distributing questionnaires.

Results The participants had 2.81 ± 1.38 geriatric syndromes on an average, with prevalence of 94.7%. The presence of each geriatric syndrome had significantly negative association with the physical component summary and mental component summary of quality of life. The number of geriatric syndromes, co-morbidity, and diabetes duration explained 29.1% of the total variance in physical component summary. The number of geriatric syndromes and co-morbidity explained 23.4% of the total variation in mental component summary.

Conclusions The number of geriatric syndromes was the major contributing factor to lower quality of life compared with socio-demographics factors and diabetic conditions among older people with T2DM.

Keywords Diabetes mellitus · Geriatric syndrome · Older people · Quality of life

Introduction

Population aging is one of the most prominent demographic problems worldwide. China holds the largest and a rapidly growing older population in the world. In 2018, there were 249 million people aged 60 years and above in China, accounting for 17.88% of total population [1]. Older people commonly experience age-associated diseases including chronic non-communicable diseases such as diabetes and hypertension [2]. Moreover, they suffer non-disease-specific conditions termed geriatric syndromes that have significant impacts on health [3, 4]. The geriatric syndrome refers to a series of different manifestations of multiple organ function impairment due to aging and age-related disease in the older population, including poly-pharmacy, malnutrition, dementia, frailty, urinary incontinence, sleep disorders, chronic pain, and falls [5, 6].

Studies show that geriatric syndromes are highly prevalent in older people including those with diabetes. Older people with diabetes in the USA and Japan developed a higher occurrence of geriatric syndromes than their counterparts without related condition [7, 8]. China is the country with the highest diabetes prevalence in Asia and with the largest disease burden of diabetes in the world. The prevalence of diabetes among Chinese people aged 60 to 69 years was 22.5% and 23.5% among people aged 70 and older in 2010, higher than younger age group [9]. However, the prevalence of geriatric syndrome is rarely reported.

Geriatric syndromes predicted increased risk of incident disabilities in US older women [10], mortality in older Koreans [11], greater likelihood of hospitalization, and higher nursing home admission in community-dwelling older adults from the USA, Canada, Australia, France, and Finland [12]. For older people with diabetes, geriatric syndromes were

✉ Xiuxin Miao
xiuxinmiaoqd@163.com

[1] School of Nursing, Qingdao University, No. 38 Dengzhou Road, Qingdao 266021, Shandong Province, China

[2] The affiliated Qingdao Municipal Hospital of Qingdao University, No. 1 Jiaozhou Road, Qingdao 266011, Shandong Province, China

associated with adverse health outcomes including mortality, disability, and impaired quality of life (QOL) [13]. Previous studies generally focused on the outcomes of a single geriatric syndrome, such as depression [14], poly-pharmacy [15], and frailty [16]. A few studies investigated the combined effects of multiple syndromes on QOL in older people with diabetes [4, 17]. For instance, Laiteerapong et al. [4] found that geriatric syndromes including chronic pain, depression, falls, underweight, and urinary incontinence contributed to lower QOL to a comparable degree as diabetes complications in the USA. Zhang et al. [17] assessed the combined impacts of two symptoms (i.e., poor sleep quality and depressive symptoms) on reduced QOL in Chinese community-dwelling patients with T2DM. However, few studies have explored the cumulative effects of multiple geriatric syndromes on QOL among older people with diabetes in China. This study was conducted to (1) investigate the occurrence of multiple geriatric syndromes among hospitalized older people with T2DM in China and (2) examine the relationship between geriatric syndromes and QOL in this population.

Material and methods

Participants

Older people with T2DM were consecutively recruited from geriatric and endocrinology departments of a tertiary hospital in north China by convenience sampling method from September 2015 to March 2016. The inclusion criteria included the following: (1) diagnosis of T2DM; (2) age ≥ 60 years; (3) able to or have a guardian to provide consent; and (4) able to or have a primary caregiver to provide information.

Data collection

The data were collected by means of reviewing the hospital medical records and a face-to-face structured interview of the participant or his/her caregiver. The measurements include socio-demographic and disease information, geriatric syndromes, and quality of life, which are further described below.

The socio-demographic information included age, gender, religion, marital status, highest education level, and income. The disease data of participants included diabetes duration, glycated hemoglobin A1c (HbA1c), insulin use, and comorbid diseases.

Based on the literature, seven geriatric syndromes were included in this study that were highly prevalent in older people with diabetes [7, 8] and expected to be intervened through clinical practice. They were chronic pain, chronic constipation, fall, poly-pharmacy, urinary incontinence, malnutrition, and sleep disorder.

Chronic pain is defined as continuous or intermittent continuous pain lasting more than 3 months [18]. Chronic constipation is defined as individual having difficulty to defecate or having a sensation of incomplete evacuation, and the symptom persists for over 6 months [19]. A fall is defined as an event which results in a person coming to rest inadvertently on the ground or floor or other lower level [20]. Poly-pharmacy is defined as the current use of five or more oral prescription drugs in this study [15]. These four geriatric syndromes were assessed by checking the medical records or by asking the participant whether related symptoms were present. The other three geriatric syndromes were evaluated by using questionnaires that have good reliability and validity. Urinary incontinence was assessed by employing the Chinese version of the International Consultation on Incontinence Questionnaire Short Form (ICIQ-SF) [21]. The ICIQ-SF has 4 items, with total score ranging from 0 to 21. A score being greater than 7 indicates the presence of urinary incontinence. Nutrition status was measured by using the Chinese version of Mini-Nutritional Assessment Short Form (MNA-SF) [22]. The total score of MNA-SF ranges from 0 to 14, with a score being below 11 indicating malnutrition or at risk of malnutrition [23]. Sleep quality was evaluated by using the Chinese version of Pittsburgh Sleep Quality Index (CPSQI) [24]. The total score ranges from 0 to 21, with a score being greater than 7 indicating poor sleep quality.

Quality of life was assessed by using the Chinese version of the Medical Outcomes Study 36-item short form health survey (SF-36) [25]. The SF-36 consists of 36 items in eight subscales: physical functioning (PF), role limitations due to physical problems (RP), bodily pain (BP), general health (GH), vitality (VT), social functioning (SF), role limitations due to emotional problems (RE), and mental health (MH). The former four subscales comprise the physical component summary (PCS), and the remaining four constitute the mental component summary (MCS). The score of each subscale ranges from 0 to 100, with higher scores signifying better QOL.

Statistical analyses

The Statistical Package for the Social Sciences (SPSS) 21.0 (SPSS Inc., Chicago, IL, USA) was used for all statistical data analysis. The data were described by means (M) and standard deviation (SD), or median and interquartile range (IQR) depending on variable distribution. Independent sample t test and one-way analysis of variance (ANOVA) followed with post hoc analysis by least-significant difference (LSD) t test were used to examine differences in QOL among groups of participants with different characteristics. Spearman rank correlation analysis was used to examine the correlation between diabetes-related conditions, geriatric syndromes, and QOL. Multiple linear stepwise regression analysis was conducted

to identify factors associated with QOL. Statistical significance was set at $p < 0.05$.

Results

Participants' characteristics and the association with quality of life

A total of 420 questionnaires were distributed, while 410 questionnaires were returned. Finally, 13 questionnaires were excluded due to missing data, and thus, 397 questionnaires were included for data analysis. The participants were aged 65–91 years (M = 74.2 years, SD = 5.4), with slightly more males (212, 53.4%). The majority of the participants had a partner (74.6%) and completed secondary education (66.3%). Of the participants, 241 (60.7%) were treated with insulin. The median of diabetes duration, number of comorbid diseases, and HbA1c were 13 years (IQR 8–18; range 1–27), 3 (IQR 2–4; range 0–10), and 8.0% (IQR 7.4–9.6%; range 5.4–14.9%), respectively. Table 1 presents other characteristics of the participants.

The means of PCS and MCS scores were 277.10 ± 64.30 and 307.00 ± 60.45, respectively. The lowest and highest ratings of QOL were on GH (63.60 ± 19.32) and RE (81.44 ± 28.03) subscales. The average scores of the other six subscales (i.e., RP, PF, VT, BP, SF, and MH) were 67.76 ± 35.62, 71.52 ± 19.19, 72.64 ± 17.22, 74.23 ± 18.81, 76.41 ± 19.97, and 76.50 ± 15.94, respectively.

Group comparison by *t* test and ANOVA showed that age ($p = 0.008$), diabetes duration ($p = 0.048$), and number of comorbid diseases ($p < 0.001$) were significantly associated with PCS of QOL. Age ($p = 0.008$), marital status ($p = 0.013$), educational level ($p = 0.031$), and number of comorbid diseases ($p < 0.001$) were significantly related to MCS (Table 1). Specifically, LSD *t* test showed that participants aged 60–69 years reported both greater PCS ($p = 0.003$) and MCS ($p = 0.004$) than participants aged 70–79 years. Participants with shorter duration of diabetes (0–5 years and 6–10 years) reported greater PCS ($p = 0.038$; $p = 0.026$) than those with

Table 1 Characteristics of participants according to PCS and MCS ($n = 397$)

Variables		*n* (%)	PCS			MCS		
			M (SD)	*t/F*	*p*	M (SD)	*t/F*	*p*
Age (year)	Young old (60–69)	100 (25.2)	294.37 (67.29)	4.911	0.008	323.08 (61.15)	4.853	0.008
	Middle old (70–79)	229 (57.7)	271.43 (63.81)			302.04 (60.60)		
	Very old (≥ 80)	68 (17.1)	270.87 (57.40)			300.05 (55.24)		
Gender	Male	212 (53.4)	282.81 (61.34)	1.896	0.059	308.90 (60.32)	0.672	0.502
	Female	185 (46.6)	270.58 (67.11)			304.82 (60.68)		
Religion	Yes	28 (7.1)	264.79 (62.35)	− 1.052	0.293	295.88 (60.85)	− 1.009	0.313
	No	369 (92.9)	278.05 (64.43)			307.84 (60.42)		
Marital status	Married or living with a partner	296 (74.6)	280.61 (63.09)	1.860	0.064	311.39 (58.85)	2.496	0.013
	Living without a partner[a]	101 (25.4)	266.87 (67.00)			294.12 (63.47)		
Educational level	Primary school or below	64(16.1)	258.92 (59.48)	2.171	0.091	289.28 (54.59)	2.996	0.031
	Junior middle school	167 (42.1)	282.66 (65.38)			312.81 (60.55)		
	High school	96 (24.2)	278.91 (63.49)			302.66 (59.79)		
	College or above	70 (17.6)	278.06 (65.39)			315.29 (63.54)		
Monthly income (CNY)	< 1000	10 (2.5)	248.00 (83.77)	1.991	0.115	299.42 (53.58)	2.080	0.102
	1001–2000	26 (6.5)	254.27 (53.71)			280.34 (56.70)		
	2001–3000	96 (24.2)	278.05 (65.47)			312.96 (57.60)		
	> 3000	265 (66.8)	280.11 (64.30)			307.74 (61.63)		
Diabetes duration (years)	0–5	54 (13.6)	290.06 (70.93)	2.653	0.048	317.37 (72.73)	1.187	0.314
	6–10	100 (25.2)	287.47 (61.70)			310.39 (63.55)		
	11–15	94 (23.7)	271.55 (63.87)			307.67 (52.07)		
	> 15	149 (37.5)	268.93 (62.72)			300.62 (58.39)		
Insulin use	Yes	241 (60.7)	275.17 (64.04)	− 0.746	0.456	306.60 (59.38)	− 0.162	0.871
	No	156 (29.3)	280.11 (64.79)			307.61 (62.24)		
No. of comorbid diseases	0	8 (2.0)	326.75 (35.88)	11.512	< 0.0001	333.69 (48.62)	10.663	< 0.001
	1–2	134 (33.8)	294.63 (63.03)			324.75 (54.68)		
	≥ 3	255 (64.2)	266.35 (63.04)			296.83 (61.43)		

PCS, physical component summary; *MCS*, mental component summary; *M*, mean; *SD*, standard deviation

[a] Including separated, divorced, widowed, or never married

diabetes longer than 15 years. Participants with higher education (junior middle school, and with college or above) reported greater MCS ($p = 0.008$; $p = 0.013$) than less educated older people. Other characteristics including gender, religion, income, and insulin use showed no statistically significant difference in both PCS and MCS.

Geriatric syndromes and quality of life

The participants had 2.81 ± 1.38 geriatric syndromes on an average, with prevalence of 94.7%. Among the participants, 325 (81.9%) had two or more syndromes. Poly-pharmacy was the most prevalent condition (70.5%), followed by sleep disorder (56.4%), chronic pain (44.1%), chronic constipation (31.5%), fall (31.2%), urinary incontinence (26.4%), and malnutrition (20.9%) (Table 2).

Independent sample *t* test showed that participants with any of the seven geriatric syndromes reported significantly lower PCS and MCS scores than those without specific geriatric syndrome (all $p < 0.001$ except $p = 0.029$ for sleep disorders for PCS). ANOVA showed that the difference among participants with varying numbers of geriatric syndromes was significant ($p < 0.001$). Fig. 1 illustrates no significant difference in PCS score between participants with 1 or no geriatric syndrome by using LSD *t* test, and no significant difference in MCS score among participants with no, 1, and 2 geriatric syndromes. Participants with 2, 3, and ≥ 4 geriatric syndromes reported lower PCS score than those with 1 or no geriatric syndrome (all $p < 0.001$). Participants with 3 and ≥ 4 geriatric syndromes reported lower MSC score than those with 1 or no geriatric syndrome (all $p < 0.001$).

As Table 3 shows, Spearman correlation analysis indicated that HbA1c, diabetes duration, the number of comorbid diseases, and number of geriatric syndromes were negatively correlated with both PCS and MCS of QOL (all $p < 0.01$), and the number of geriatric syndromes was moderately correlated with lower QOL. In addition, HbA1c, diabetes duration, and the number of comorbid diseases were positively correlated with the number of geriatric syndromes.

Multiple regression analysis on quality of life

With independent variables identified as being statistically associated with PCS and MCS by the abovementioned univariate analysis, multiple stepwise regression analysis was conducted. The analysis found that the number of geriatric syndromes was the major contributing factor to lower scores of both PCS ($\beta = -0.44$, $p < 0.001$) and MCS ($\beta = -0.41$, $p < 0.001$). For the PCS, the most significant influencing factor was the number of geriatric syndromes (adjusted $R^2 = 0.252$, $p < 0.001$), followed by the number of comorbid diseases (adjusted $R^2 = 0.036$, $p < 0.001$), and diabetes duration (adjusted $R^2 = 0.009$, $p < 0.001$). These three variables together explained 29.1% of the total variance in PCS ($p < 0.001$).

Table 2 Comparisons of PCS and MCS according to geriatric syndromes ($n = 397$)

Variables		*n* (%)	PCS			MCS		
			M (SD)	*t*/*F*	*p*	M (SD)	*t*/*F*	*p*
Poly-pharmacy	Present	280 (70.5)	267.5 (61.8)	−4.71	< 0.001	297.6 (59.9)	−4.95	< 0.001
	Absent	117 (29.5)	300.0 (64.7)			329.6(55.9)		
Sleep disorder	Present	224 (56.4)	261.0(63.0)	−5.94	0.029	293.5(60.3)	−5.21	<0.001
	Absent	173 (43.6)	298.0(59.9)			324.4(56.1)		
Chronic constipation	Present	125 (31.5)	260.1 (65.8)	−3.63	< 0.001	289.4 (60.9)	−4.00	< 0.001
	Absent	272 (68.5)	284.9 (62.2)			315.1 (58.6)		
Chronic pain	Present	175 (44.1)	257.9 (62.5)	−5.47	< 0.001	294.2 (60.8)	−3.81	< 0.001
	Absent	222 (55.9)	292.2 (61.7)			317.1 (58.4)		
Fall	Present	124 (31.2)	255.8 (60.2)	−4.56	< 0.001	290.7 (58.3)	−3.67	< 0.001
	Absent	273 (68.8)	286.8 (63.9)			314.4 (60.1)		
Urinary incontinence	Present	105 (26.4)	256.8 (58.5)	−3.84	< 0.001	295.3 (58.2)	−2.33	< 0.001
	Absent	292 (73.6)	284.4 (64.8)			311.2 (60.8)		
Malnutrition	Present	83 (20.9)	263.4 (58.7)	−2.19	< 0.001	287.7 (61.8)	−3.31	< 0.001
	Absent	314 (79.1)	280.7 (65.3)			312.1 (59.1)		
No. of geriatric syndromes	0	21 (5.3)	344.3 (47.7)	32.83	< 0.001	358.7 (58.5)	29.07	< 0.001
	1	51 (12.9)	326.1 (41.5)			344.7 (50.0)		
	2	90 (22.7)	294.0 (64.7)			334.0 (37.6)		
	3	103 (25.9)	269.1 (54.3)			291.8 (53.9)		
	≥ 4	132 (33.2)	242.2 (56.8)			277.7 (61.8)		

PCS, physical component summary; *MCS*, mental component summary

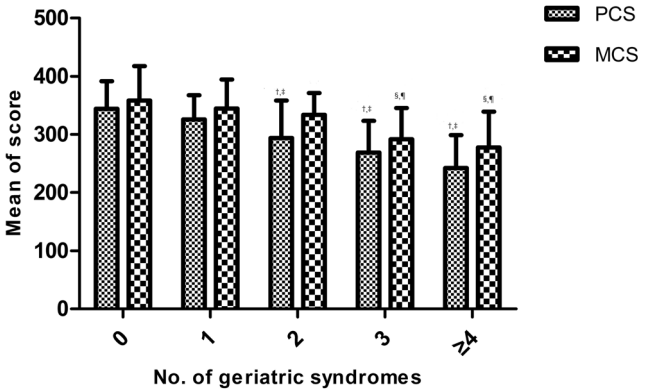


Fig. 1 Comparison of PCS and MCS according to the number of geriatric syndromes by post hoc LSD-t tests. †Compared with participants without geriatric syndrome, participants with 2, 3, and ≥4 reported significantly lower PCS score (all $p < 0.001$). ‡Compared with participants with 1 geriatric syndrome, participants with 2, 3, and ≥4 reported significantly lower PCS score (all $p < 0.001$). §Compared with participants without geriatric syndrome, participants with 3 and ≥4 reported significantly lower MCS score (all $p < 0.001$). ¶Compared with participants with 1 geriatric syndrome, participants with 3 and ≥4 reported significantly lower MCS score (all $p < 0.001$). There was no significant difference in PCS score between participants without and with 1 geriatric syndrome, and no significant difference in MCS score among participants without, with 1, and 2 geriatric syndromes. PCS, physical component summary; MCS, mental component summary

For the MCS, the most significant influencing factor was also the number of geriatric syndromes (adjusted $R^2 = 0.203$, $p < 0.001$), followed by the number of comorbid diseases (adjusted $R^2 = 0.034$, $p < 0.001$). The two factors accounted for 23.4% of the total variance in MCS ($p < 0.001$) (Table 4).

Discussion

The study found that the older people with T2DM had low QOL, in which the score of general health (GH) subscale was the lowest and role-emotional (RE) was the highest. In SF-36, GH assesses participants' self-assessment of current health condition and treatment perspectives, and RE reflects the degree of emotional problems impeding daily activities [25]. The lower score of GH in this study may be closely associated with aging and diabetes conditions such as presence of cardiovascular disease and hypoglycemia that affect physical function [7]. Older people with T2DM reporting relatively high RE scores in this study may suggest that mental health was not severely affected to extent that physical health was impaired. These findings are consistent with that of a previous study in which community-dwelling older Japanese with diabetes reported significant impairment in PCS of QOL, yet no difference in mental health and social functioning compared with those without diabetes [8].

With regard to socio-demographic factors, this study showed significant difference in PCS score by age, and significant difference in MCS score by age, marital status, and educational levels while no difference by gender and income. These findings partially correspond with recent studies in China [3] and other parts of the world [26]. Patients with younger age, with a partner, and higher educational level may better deal with emotional stress associated with having T2DM and thus had higher MCS score [27]. In contrast, some studies found that female patients and those with lower income reported lower QOL [14, 28]. The inconsistence may be attributed to different characteristics of the study populations such as in wide age ranges or living in varied sociocultural contexts.

Older people with T2DM in this study reported high prevalence of geriatric syndromes, and over one-third of them had 4 or more geriatric syndromes. Specifically, the occurrence of poly-pharmacy, chronic pain, and sleep disorder in this population was higher than their counterparts in Italy (57.1%) [15], in the USA (41%) [4], and communities living in China (33.6%) [17], respectively. These findings indicate that prevalence of geriatric syndromes increased in older people with diabetes, in line with the results of previous research [7, 8, 29]. The high prevalence of geriatric syndromes in older people with T2DM could be a result of the interactions between symptoms of T2DM, adverse effects of treatment, and

Table 3 Correlation analysis of diabetes-related conditions, comorbidity, geriatric syndromes, and QOL

Variables	r_s					
	1	2	3	4	5	6
HbA1c (1)	–					
Diabetes duration (2)	0.042	–				
No. of comorbid diseases (3)	0.042	0.043	–			
No. of geriatric syndromes (4)	0.238**	0.131*	0.248**	–		
PCS (5)	− 0.188**	− 0.170*	− 0.342**	− 0.519**	–	
MCS (6)	− 0.172*	− 0.132*	− 0.299**	− 0.489**	0.615**	–

HbA1c, glycated hemoglobin A1c; *PCS*, physical component summary; *MCS*, mental component summary; *QOL*, quality of life; r_s, Spearman rank correlation coefficient

*$p < 0.01$; **$p < 0.001$

Table 4 Multiple stepwise regression analysis on PCS and MCS ($n = 397$)

		Constant	*B*	β	*t*	Adjusted R^2 (R^2)	F	*p*
PCS	Model 1	342.659				0.250 (0.252)	133.061	< 0.001
	No. of geriatric syndromes		− 23.317	− 0.502	− 11.535			< 0.001
	Model 2	362.405				0.284 (0.288)	79.704	< 0.001
	No. of geriatric syndromes		− 21.148	− 0.455	− 10.401			< 0.001
	No. of comorbid diseases		− 8.123	− 0.196	− 4.468			< 0.001
	Model 3	372.222				0.291 (0.297)	55.260	< 0.001
	No. of geriatric syndrome		− 20.478	− 0.441	− 10.007			< 0.001
	No. of comorbid diseases		− 8.115	− 0.195	− 4.485			< 0.001
	Diabetes duration		− 0.975	− 0.094	− 2.197			0.029
MCS	Model 1	362.355				0.201 (0.203)	100.840	< 0.001
	No. of geriatric syndromes		− 19.692	− 0.451	− 10.042			< 0.001
	Model 2	380.389				0.234 (0.237)	61.332	< 0.001
	No. of geriatric syndromes		− 17.711	− 0.406	− 8.953			< 0.001
	No. of comorbid diseases		− 7.419	− 0.190	− 4.194			< 0.001

For PCS, compared model 2 with model 1, with no. of comorbid disease entering into the regression model, adjusted R^2 was increased from 0.252 to 0.288, indicating that no. of comorbid diseases could explain 0.036 (adjusted $R^2 = 0.036$, obtained by 0.288 minus 0.252) of the variance. Compared model 3 with model 2, diabetes duration could explain 0.009 of the variance adjusted ($R^2 = 0.009$, obtained by 0.297 minus 0.288). For MCS, compared model 2 with model 1, no. of comorbid diseases could explain 0.034 of the variance adjusted ($R^2 = 0.034$, obtained by 0.237 minus 0.203)

PCS, physical component summary; *MCS*, mental component summary; *B*, unstandardized coefficient; *β*, standardized coefficient beta

underlying vulnerability [7, 29]. For example, patients with T2DM are prone to suffer from some co-morbidities and complications including peripheral neuropathy, hyperglycemia, and hypoglycemia that can result in persistent pain, impaired gait balance, and vision. These comorbid conditions could subsequently increase the risk of falls, urinary incontinence, and sleep disorder [7]. In addition, treatment for glycemic control often requires patients to take medications and dietary restriction, potentially resulting in poly-pharmacy, chronic constipation, and risk of malnutrition [30]. Correlation analysis in this study revealed that HbA1c, diabetes duration, and the number of comorbid diseases were positively correlated with the number of geriatric syndromes. These diabetes-related conditions perhaps are more serious for the hospitalized patients in this study than community-dwelling older patients that may consequently contribute to the high prevalence of geriatric syndromes.

The presence of each geriatric syndrome (i.e., chronic pain, chronic constipation, urinary incontinence, fall, malnutrition, sleep disorders, and poly-pharmacy) assessed in this study was found to be correlated with reduced QOL. Geriatric syndromes represent common yet serious conditions for older persons, which may affect self-care abilities and thus are associated with adverse outcomes and low QOL [29]. Other studies consistently found that presence of geriatric syndromes including poor sleep quality [3], falls [4], and chronic pain [31] was correlated with reduced QOL in older people with diabetes. In addition, participants with 1 geriatric syndrome in this study may be relatively healthy and thus reported no difference in QOL than patients without geriatric syndrome.

Diabetes-related conditions including HbA1c, duration, and the number of comorbid diseases were reversely related with QOL, in line with findings of previous studies [26, 28]. However, a study among older Japanese patients with diabetes revealed no significant associations between QOL and disease-specific factors including duration of diabetes [8]. The discrepancy may lie in that the participants in Japan were community dwellers with relatively better diabetic status compared with the hospitalized patients in the current study. Comorbidities can directly impair QOL in patients with diabetes as they affect patients' functional status [4]. A study showed that the presence of co-morbidity increased the burden of geriatric syndromes for older people with diabetes [32] that may further impair QOL.

Further multiple regression analysis found that the number of geriatric syndromes rather than diabetic conditions was the major contributing factor closely related to lower PCS and MCS of QOL in older people with T2DM. This suggests the cumulative and predominant effects of multiple geriatric syndromes on reduced QOL for this study population. The findings support previous work demonstrating that the combination of geriatric syndromes instead of chronic diseases were the most prominent predictors of poor health outcomes in US older people [33].

Overall, this study showed that geriatric syndromes were prevalent and significantly contributed to impaired QOL in the older people with T2DM. The findings provide important

implications for clinical practice and education. The geriatric conditions imply complex health needs of older people with T2DM and raise distinct challenges for health professionals. As suggested by other scholars [29], care guidelines and tools such as the comprehensive geriatric assessment for effectively assessing and managing the co-occurrence of T2DM and geriatric syndromes for older people are warranted. Furthermore, intervention programs led by interdisciplinary health providers including exercise training and diet therapy are beneficial to prevent certain geriatric syndromes including fall, malnutrition, chronic constipation, and urinary incontinence [29]. Therefore, healthcare professionals including nurses in China should consider integrating geriatric syndrome assessment into clinical practice and transitional care, along with taking effective interventions to improve QOL for older people with T2DM.

Strengths and limitations

The study focused on hospitalized older people with T2DM, explored the prevalence of geriatric syndromes, and found the significantly negative impacts on their quality of life. However, several limitations need to be recognized. First, we surveyed hospitalized older people with a relatively small sample size. For community-dwelling older people with T2DM, the prevalence of geriatric syndromes and its association with QOL may be different. Second, we only examined 7 geriatric syndromes and thus cannot reveal the overall prevalence of geriatric syndrome among older people with T2DM. Other geriatric syndromes such as cognitive impairment and depression were not included in the survey. Older patients with cognitive impairment may not be able to perform self-care tasks such as compliance with dietary plan and medication, leading to persistent hyperglycemia and increased risk of diabetic complications, which in turn worsen their QOL [34]. Third, this study is cross-sectional descriptive and thus cannot examine the causal relationship between variables. Further large-scale and multi-institutional studies with inclusion of mental state assessment are needed to explore the complex interactions between geriatric syndromes, diabetes conditions, and QOL.

In addition, the regression analysis in this study showed that number of geriatric syndromes, comorbid diseases, and diabetic conditions accounted for only a small part of the total variance in QOL. This indicates other factors besides the variables included in this study affect QOL. For instance, previous research reported that activities of daily life, physical activity, self-efficacy, family function, and social support were predictors of QOL for older people [28]. Family support could improve patients' diabetes self-management and prognosis, and thus was associated with a low sense of diabetes distress and a strong mental health-related QOL [35]. Moreover, some of these factors such as lack of physical activity and social support (e.g., living alone) increased the risk of geriatric syndromes [36]. Therefore, a potential limitation of this study is that we did not collect data about living situation (living alone or with someone) of older patients and thus could not evaluate the impacts on geriatric syndrome and QOL. Future studies are required to further explore predictors of geriatric syndromes and QOL in this population through adding more variables in personal and family domains.

Conclusions

Chinese older people with T2DM had multiple geriatric syndromes. The number of geriatric syndromes was the major contributing factor closely related to lower QOL, along with the number of comorbid diseases and diabetes duration. Therefore, screening for and managing geriatric syndromes need to be considered as a priority in nursing practice to improve QOL of older people with diabetes. Interventions that target geriatric syndromes in patients with T2DM may help to prevent the aggravation of geriatric conditions and thus to reduce the detrimental impacts on QOL.

Acknowledgments The authors would like to sincerely thank the patients with type 2 diabetes mellitus for their participation in this study. We also wish to thank the staff of geriatric and endocrinology departments who allowed us to recruit patients from their department.

Compliance with ethical standards The study was approved by the Ethics Review Committee of Faculty of Medicine at Qingdao University where the study was conducted. All the participants signed informed consent prior to their participation.

Conflict of interest The authors declare that they have no conflict of interest.

References

1. National Bureau of Statistics of China. China statistical yearbook 2019. http://www.stats.gov.cn/tjsj/ndsj/2019/indexch.htm. Accessed 20 Feburary 2020.
2. Yang GWY, Zeng Y, Gao GF, Liang X, Zhou M, Wan X, et al. Rapid health transition in China, 1990–2010: findings from the Global Burden of Disease Study 2010. Lancet. 2013;381(9882): 1987–2015. https://doi.org/10.1016/S0140-6736(13)61097-1.
3. Lou P, Qin Y, Zhang P, Chen P, Zhang L, Chang G, et al. Association of sleep quality and quality of life in type 2 diabetes mellitus: a cross-sectional study in China. Diabetes Res Clin Pract. 2015;107(1):69–76. https://doi.org/10.1016/j.diabres.2014.09.060.
4. Laiteerapong N, Karter AJ, Liu JY, Moffet HH, Sudore R, Schillinger D, et al. Correlates of quality of life in older adults with diabetes: the diabetes & aging study. Diabetes Care. 2011;34(8): 1749–53.
5. McRae PJ, Peel NM, Walker PJ, de Looze JW, Mudge AM. Geriatric syndromes in individuals admitted to vascular and urology surgical units. J Am Geriatr Soc. 2014;62(6):1105–9. https://doi.org/10.1111/jgs.12827.

6. Inouye SK, Studenski S, Tinetti ME, Kuchel GA. Geriatric syndromes: clinical, research, and policy implications of a core geriatric concept. J Am Geriatr Soc. 2007;55(5):780–91.
7. Kirkman MS, Briscoe VJ, Clark N, Florez H, Haas LB, Halter JB, et al. Diabetes in older adults: a consensus report. J Am Geriatr Soc. 2012;60(12):2342–56. https://doi.org/10.1111/jgs.12035.
8. Nezu S, Okamoto N, Morikawa M, Saeki K, Obayashi K, Tomioka K, et al. Health-related quality of life (HRQOL) decreases independently of chronic conditions and geriatric syndromes in older adults with diabetes: the Fujiwara-kyo study. J Epidemiol. 2014;24(4):259–66. https://doi.org/10.2188/jea.JE20130131.
9. Xu Y. Prevalence and control of diabetes in Chinese adults. J Am Med Assoc. 2013;310(9):948–59. https://doi.org/10.1001/jama.2013.168118.
10. Rosso AL, Eaton CB, Wallace R, Gold R, Stefanick ML, Ockene JK, et al. Geriatric syndromes and incident disability in older women: results from the women's health initiative observational study. J Am Geriatr Soc. 2013;61(3):371–9. https://doi.org/10.1111/jgs.12147.
11. Kim S, Park JH, Won CW. Combined effects of four major geriatric syndromes on adverse outcomes based on Korean National Health Insurance claims data. Geriatr Gerontol Int. 2018;18(10):1463–8. https://doi.org/10.1111/ggi.13513.
12. Wang S-Y, Shamliyan TA, Talley KMC, Ramakrishnan R, Kane RL. Not just specific diseases: systematic review of the association of geriatric syndromes with hospitalization or nursing home admission. Arch Gerontol Geriatr. 2013;57(1):16–26. https://doi.org/10.1016/j.archger.2013.03.007.
13. Sinclair A, Dunning T, Rodriguez-Mañas L. Diabetes in older people: new insights and remaining challenges. Lancet Diabetes Endocrinol. 2015;3(4):275–85. https://doi.org/10.1016/S2213-8587(14)70176-7.
14. Kim H, Kim K. Health-related quality-of-life and diabetes self-care activity in elderly patients with diabetes in Korea. J Community Health. 2017;42(5):998–1007. https://doi.org/10.1007/s10900-017-0347-2.
15. Noale M, Veronese N, Cavallo Perin P, Pilotto A, Tiengo A, Crepaldi G, et al. Polypharmacy in elderly patients with type 2 diabetes receiving oral antidiabetic treatment. Acta Diabetol. 2016;53(2):323–30. https://doi.org/10.1007/s00592-015-0790-4.
16. Umegaki H. Sarcopenia and frailty in older patients with diabetes mellitus. Geriatr Gerontol Int. 2016;16(3):293–9. https://doi.org/10.1111/ggi.12688.
17. Zhang P, Lou P, Chang G, Chen P, Zhang L, Li T, et al. Combined effects of sleep quality and depression on quality of life in patients with type 2 diabetes. BMC Fam Pract. 2016;17(1):40. https://doi.org/10.1186/s12875-016-0435-x.
18. Merskey H. Classification of chronic pain: descriptions of chronic pain syndromes and definitions of pain terms. Pain Suppl. 1995;3(2):S1.
19. Longstreth G, Thompson W, Chey W, Houghton L, Mearin F, Spiller R. Functional bowel disorders. Gastroenterology. 2006;130(5):1480–91. https://doi.org/10.1053/j.gastro.2005.11.061.
20. World Health Organization. Falls. 2012. http://www.who.int/news-room/fact-sheets/detail/falls. Accessed 1 January 2018.
21. Z-b C, Y-q L, Q-d C, S-l S, B-t S, Z-c G. The relevance study of Chinese Short Form of International Consultation on Incontinence questionnaire (ICIQ-SF) and urodynamics. J Mod Urol. 2011;16(9):403–5.
22. He X, Liu X. Evaluation of reliability and validity of mini-nutritional assessment and Chinese nutrition screen. Nurs J Chin People's Lib Army. 2010;27(6B):894–6.
23. Bauer J, Kaiser M, Anthony P, Guigoz Y, Sieber C. The mini nutritional assessment-its history, today's practice, and future perspectives. Nutr Clin Pract. 2008;23(4):388–96. https://doi.org/10.1177/0884533608321132.
24. Tsai P-S, Wang S-Y, Wang M-Y, Su C-T, Yang T-T, Huang C-J, et al. Psychometric evaluation of the Chinese version of the Pittsburgh Sleep Quality Index (CPSQI) in primary insomnia and control subjects. Qual Life Res. 2005;14(8):1943–52. https://doi.org/10.1007/s11136-005-4346-x.
25. Li L, Wang H, Shen Y. Development and psychometric tests of a Chinese version of the SF-36 Health Survey Scales. Chin J Prev Med. 2002;36(2):109–13.
26. Dogan H, Harman E, Kocoglu H, Sargin G. Can metabolic control variables of diabetic patients predict their quality of life? J Am Soc Hypertens. 2016;10(1):81–8. https://doi.org/10.1016/j.jash.2015.11.014.
27. Bowen PG, Clay OJ, Lee LT, Vice J, Ovalle F, Crowe M. Associations of social support and self-efficacy with quality of life in older adults with diabetes. J Gerontol Nurs. 2015;41(12):21–9; **quiz 30-1**. https://doi.org/10.3928/00989134-20151008-44.
28. Werfalli M, Kassanjee R, Kalula S, Kowal P, Phaswana-Mafuya N, Levitt NS. Diabetes in South African older adults: prevalence and impact on quality of life and functional disability – as assessed using SAGE Wave 1 data. Glob Health Action. 2018;11(1):1449924. https://doi.org/10.1080/16549716.2018.1449924.
29. Araki A, Ito H. Diabetes mellitus and geriatric syndromes. Geriatr Gerontol Int. 2009;9(2):105–14. https://doi.org/10.1111/j.1447-0594.2008.00495.x.
30. Okayasu S, Kitaichi K, Hori A, Suwa T, Horikawa Y, Yamamoto M, et al. The evaluation of risk factors associated with adverse drug reactions by metformin in type 2 diabetes mellitus. Biol Pharm Bull. 2012;35(6):933–7.
31. Liberman O, Peleg R, Shvartzman P. Chronic pain in type 2 diabetic patients: a cross-sectional study in primary care setting. Eur J Gen Pract. 2014;20(4):260–7. https://doi.org/10.3109/13814788.2014.887674.
32. Vetrano DL, Foebel AD, Marengoni A, Brandi V, Collamati A, Heckman GA, et al. Chronic diseases and geriatric syndromes: the different weight of comorbidity. Eur J Intern Med. 2016;27:62–7. https://doi.org/10.1016/j.ejim.2015.10.025.
33. Koroukian SM, Schiltz N, Warner DF, Sun J, Bakaki PM, Smyth KA, et al. Combinations of chronic conditions, functional limitations, and geriatric syndromes that predict health outcomes. J Gen Intern Med. 2016;31(6):630–7. https://doi.org/10.1007/s11606-016-3590-9.
34. Abdelhafiz AH, Sinclair AJ. Diabetes in the elderly. Medicine. 2019;47(2):119–22. https://doi.org/10.1016/j.mpmed.2018.11.007.
35. Bennich BB, Munch L, Egerod I, Konradsen H, Ladelund S, Knop FK, et al. Patient assessment of family function, glycemic control and quality of life in adult patients with type 2 diabetes and incipient complications. Can J Diabetes. 2019;43(3):193–200. https://doi.org/10.1016/j.jcjd.2018.09.002.
36. Rausch C, Liang Y, Bultmann U, de Rooij SE, Johnell K, Laflamme L, et al. Social position and geriatric syndromes among Swedish older people: a population-based study. BMC Geriatr. 2019;19(1):267. https://doi.org/10.1186/s12877-019-1295-8.

International Journal of Diabetes in Developing Countries (2021) 41:156–163
https://doi.org/10.1007/s13410-020-00828-1

ORIGINAL ARTICLE

Assessment of a group-based comprehensive diabetes management program to improve glycemic control, quality of life and self-care behavior in patients with type 2 diabetes mellitus in a primary healthcare setting of a metropolitan city in India: CDMP MUM Trial

Mandar Padmakar Baviskar[1] · Sumitra Rangari[2] · Sandeep Mishra[3] · Biswendu Sundar Mohanta[4]

Received: 26 September 2019 / Revised: 4 January 2020 / Accepted: 24 April 2020 / Published online: 6 October 2020

Abstract

Background Resource-poor primary health settings in an urban slum presented with special challenges in diabetes care. The study evaluated a need-based, patient-friendly, acceptable, appropriate package of care and its implementation in primary healthcare setup.

Material and Methods An open-label two-arm parallel randomized controlled trial with 40 patients in the control group who received medical management as prescribed by physician and patients in the intervention group receiving usual medical care + group-based comprehensive diabetes management program. HbA1c, weight, BMI, Quality of Life Instrument for Indian Diabetes Patients (QOLID) scores, and audit of Self Care Inventory-Revised Version (SCI-R) were assessed at baseline and 6 months.

Result There was a significant reduction in mean HbA1c levels in the intervention arm (8.44, SD = 1.802 to 7.56, SD = 1.87) as compared with the control arm (8.4, SD = 1.87 to 8.19, SD = 1.77). Multiple linear regression model ($R^2 = 0.886$, ANOVA F (7, 72) = 79.733, $p < 0.001$) found a fall in blood sugar levels ($\beta = -0.511$, $p < 0.001$), improvement in QOLID scores ($\beta = 0.221$, p = 0.004), increase in physical activity ($\beta = 0.198$, p = 0.006), and fall in BMI ($\beta = 0.153$, $p = 0.009$) to be predictive of change in HbA1c.

Discussion The patient profile consisted predominantly of homemakers, with low formal education and belonging to lower socio-economic strata. They depended primarily on the public health system for their health care needs and could benefit most from the implementation of the program. There was an improvement in knowledge, regular follow-up, compliance to medication, diet, and physical activity along with improvement in glycemic control, self-care behavior and quality of life of patients in the intervention arm.

Keywords CDMP MUM · Diabetes mellitus · Urban slum · Group-based comprehensive care

Introduction

India is presently home to 62 million diabetics, and by 2030, diabetes numbers are expected to cross the 100 million mark [1]. Adequate care of diabetics requires a formal and more structured involvement of the primary care sector, together with the diabetes units and diabetes centres [2]. Comprehensive programs have been developed to treat and prevent chronic diseases such as diabetes in western countries. The diversity in Indian population paired with lack of resources makes diabetes care especially challenging.

Studies from India reveal a huge gap between the recommended and actual diabetes care, resulting in poor health outcomes [3–7]. Lack of knowledge, poor treatment adherence and lack of self-care behavior all mar treatment outcome and lead to poor glycemic control which in turn leads to a poor quality of life for the patients.

✉ Mandar Padmakar Baviskar
baviskar.mandar@gmail.com

Sumitra Rangari
sumitrarangari@gmail.com

Sandeep Mishra
drsandipamishra@gmail.com

Biswendu Sundar Mohanta
bisswa@gmail.com

[1] Department of Community Medicine, Pravara Institute of Medical Sciences (DU), Loni, Tal-Rahata, Ahmednagar, Maharashtra, India

[2] ESIS, Mumbai, India

[3] Seth GS Medical College & KEM Hospital, Mumbai, India

[4] Divisional Railway Hospital, Malda, India

Adoption of non-pharmacological interventions remains unmonitored in primary care settings. The authors endeavoured to assess the role of a group-based comprehensive Diabetes care program to improve glycemic control, quality of life and self-care behaviors in patients with type 2 diabetes mellitus. The study integrates the recommended program interventions in a structured manner. It has been designed as need-based, patient-friendly, acceptable, appropriate package. This study gives practical insights regarding mainstreaming of comprehensive diabetes management in primary health settings.

Aim

To assess the role of a group-based comprehensive diabetes management program to improve glycemic control, quality of life and self-care behaviors in patients with type 2 diabetes mellitus.

Objectives

1. Study the profile and health needs in confirmed type 2 diabetes patients with reference to barriers to glycemic control, quality of life and self-care behavior.
2. To evolve and implement a feasible behavioral intervention program along with usual medical care of type 2 diabetes mellitus based on assessment of study subjects.
3. To compare outcomes in study and control subjects.

Material and methods

The study was designed as an open-label two-arm parallel randomized controlled trial with patients in the control group receiving usual medical care (medical management as prescribed by the physician) and those in intervention group receiving usual medical care + group-based comprehensive diabetes management program.

Study area

Study was conducted at Malavni, Urban Health Training Centre situated in an urban slum of Mumbai. A chronic disease OPD is run by the department of Community Medicine where patients from surrounding region come to avail treatment.

Recruitment

Among the patients registered and availing treatment at chronic diseases OPD, eligible patients who met the inclusion criteria and gave a written informed consent were recruited.

Sample size estimation

A pilot study was conducted in 20 randomly selected patients meeting the inclusion criteria. The standard deviation of Fasting HbA1c was found to be 1.5. The targeted sample size sufficient to detect an estimated net intervention to control difference of 1% in fasting HbA_1c levels at 6 months after randomisation with $\alpha = 0.05$, $\beta = 0.2$ and one-tailed significance test was included in each arm. Similar studies have shown an attrition rate ranging from 11 to 17% [8–11]. A further addition of 15% to the calculated sample size was done considering the attrition rate based on previous studies. The intervention to control ratio was 1:1. A total of 40 patients were recruited in each group.

Data collection and study instruments

Data collection was carried out by one on one structured interview with the patients. Help of medical social worker was taken in counselling the patients and their families. Baseline data included socio-demographic profile, clinical history, nutritional assessment, weight, height and BMI. Quality of Life Instrument for Indian Diabetes Patients (QOLID) scale containing 8 domains and 34 questions was used to assess the quality of life in patients with diabetes. It has been found reliable and valid tool for assessment of quality of life of Indian patients with diabetes (Cronbach's alpha = 0.894) [12] Self Care Inventory-Revised Version (SCI-R), a 13-item scale used to find the level of self-care among diabetic patients (alpha = 0.84) [13]. Three items that are not relevant in the Indian scenario were dropped, and the audited 10-item scale was administered. Glycosylated hemoglobin (HbA1c), and fasting blood glucose were estimated at baseline and at the end of 6 months.

Randomisation

Since all the patients to be recruited in the study were already registered in the OPD (i.e. all participants had already been identified), proportionate stratified randomized allocation method was used from the two arms of study.

This was done to control for the possible influence of covariates that would jeopardize the conclusions of the research. This scheme also ensured 1:1 ratio of intervention and control arms.

Stratified randomisation was achieved by generating a separate block for each combination of covariates to be controlled. After all subjects had been identified and assigned into blocks, simple randomisation was performed within each block to assign subjects to one of the two groups [14].

Blinding An open-label design was chosen, since being a behavioral intervention study, it was impossible to blind the patients or the investigators.

Prevention of spillover In order to avoid spillover from intervention arm to control arm, the patients in control arm and intervention arm were followed up on separate (alternate) weeks to minimize interaction.

Intervention

Grouping The patients in the intervention arm were divided into peer groups. The group setting aimed to provide social interaction and peer support. The participants were encouraged to bring their spouses or significant others with them for counselling.

We adopted a framework, that is (i.e.) an amalgamation of various theoretical models of behavior change, ensuring validity and generalizability of intervention in the target population [15]. The process involved:

1. Identifying problem areas**:** These included lack of diet control, lack of physical activity, lack of motivation and lack of social support.
2. Education about self-care: The education included basic information about diabetes in simple language which was given to patients by investigator and Medical Social Worker. Information regarding proper diet, proper exercise, foot care, taking timely medication and regular checkups was given. Patient education material developed in local language by the patient education cell was used for this purpose. Further, group discussions were conducted to address any doubts that patients had.
3. Collaborative goal setting: The groups were interviewed and motivated to set specific time bound goals that they could attain. A dietary plan was given, and information regarding food groups was given to participants and their family members to ensure compliance. Preparing an exercise schedule or having evening walks in the local park for at least half an hour for 5 days a week was encouraged. Family members were encouraged to actively participate in ensuring compliance by patients. The goals were reviewed during follow-ups.
4. Maintaining and revising goals: Progress was assessed at every follow-up meeting. New goals were set once old goals were achieved. Patients were kept motivated to sustain the change already achieved. In case of relapse in behavior, the cycle was repeated again.

Key components of group-based comprehensive diabetes care program

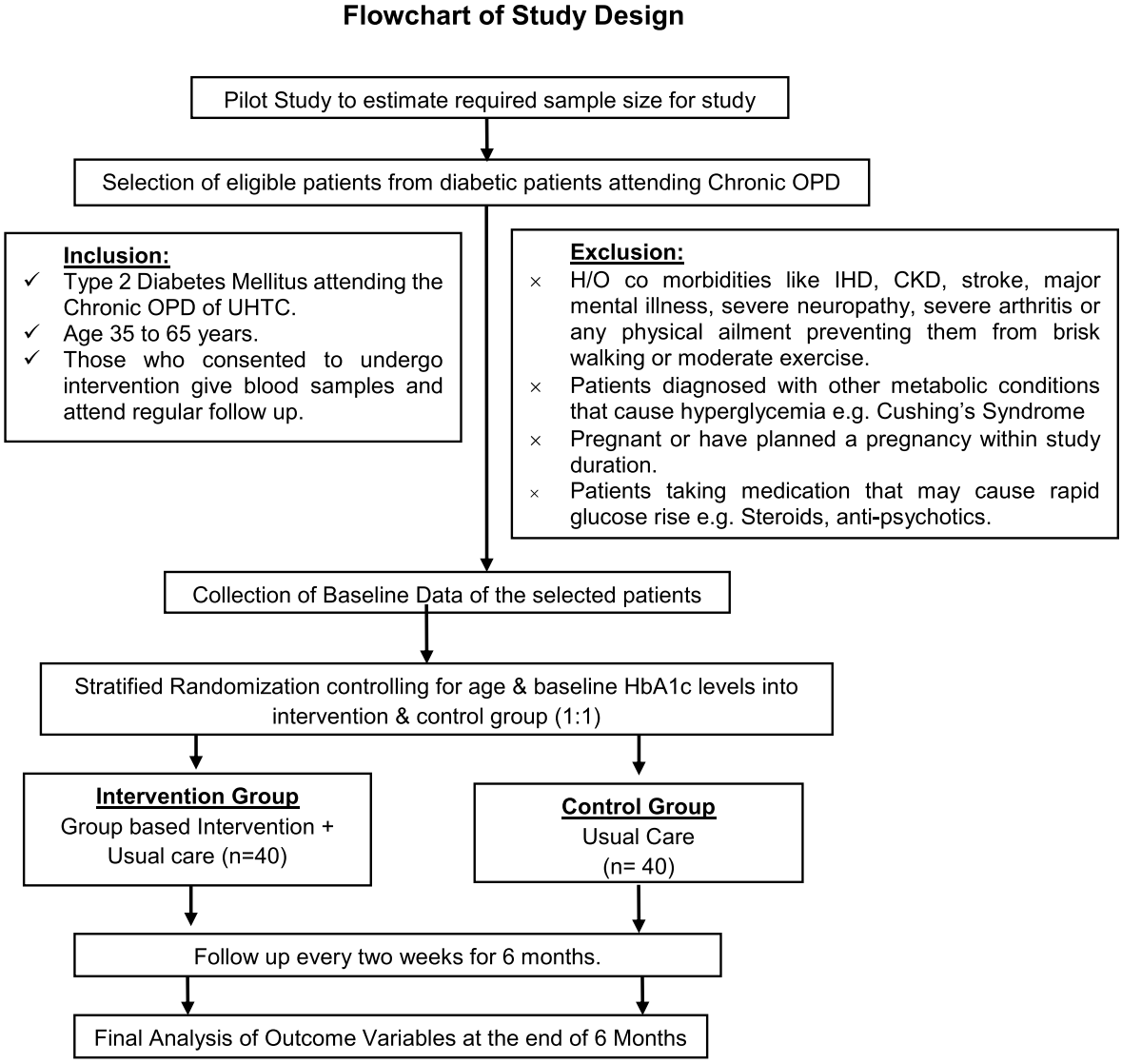
Knowledge	Diet	Self-care and timely medications	Physical activity	Follow-up
Basic information about diabetes in simple language regarding proper diet, proper exercise, foot care, taking timely medication and regular checkups was given.	Advice regarding reducing food portions, frequent small meals, foods to avoid and foods to eat was given after ensuring the cultural appropriateness, acceptability and feasibility of the designed diet plan. Counselling along with a family member was done wherever necessary to ensure compliance with the diet plan. Help of medical social worker (MSW) was taken while counselling and follow-up.	The patients were asked to follow-up regularly at the chronic OPD. Blood sugar levels were checked free of cost at this OPD. Timely intake of medication was advocated, and improvements were assessed by self-care inventory at 6 months.	A moderate physical activity like 30-min brisk walk 5 times a week has been found to be useful in weight maintenance in diabetic patients. Patients were encouraged to form peer groups and walk for 30 min in the community park near the UHC in the evening. They were advised to slowly increase their activity level. An activity chart was maintained.	While taking baseline data of patients, their complete address and phone number were noted. In case the patient did not have telephone, the number of a close relative or a neighbour was noted. Follow-up at UHC every 2 weeks. In case of loss to follow-up, the patient was contacted. Reasons for loss to follow-up were noted.

Outcome variables

- Primary outcome:
  1. Change in level of glycosylated hemoglobin (HbA_{1c})
- Secondary outcomes:
  1. Anthropometric measures: weight and BMI
  2. Changes in fasting blood glucose level
  3. Changes in quality of life as seen by Quality of Life Instrument for Indian Diabetes Patients (QOLID)
  4. Changes in self-care behavior as seen by audit of Self Care Inventory-Revised Version (SCI-R)

Statistical analysis

Descriptive statistical analysis was done at baseline as well as at the end of follow-up. Chi-square test was used to compare



baseline characteristics among intervention and control groups (Table 1). Unpaired *t* test was used to compare continuous variables in control and intervention group. Mann-Whitney *U* test was applied to compare scores of Quality of Life Instrument for Indian Diabetes Patients (QOLID) and audit of Self Care Inventory-Revised Version (SCI-R) in control and intervention group (Table 2). Linear regression model was also applied to find predictors.

The study was approved by Institutional ethics committee of Seth GS Medical College & KEM Hospital.

Results

Both intervention and control groups had comparable socio-demographic profile. Multiple linear regression model was applied ($R^2 = 0.886$, ANOVA $F\ (7, 72) = 79.733$, $p < 0.001$). Fall in blood sugar levels ($\beta = -0.511$. $p < 0.001$), improvement in quality of life-QOLID scores ($\beta = 0.221$, $p = 0.004$), increase in physical activity ($\beta = 0.198$, $p = 0.006$) and fall in BMI ($\beta = 0.153$, $p = 0.009$) were all predictive of change in the dependent variable HbA1c.

Discussion

The patient profile consisted predominantly of females who were homemakers, with low formal education and belonging to lower socio-economic strata. The study population depended primarily on public health system for their health care needs and could benefit most from implementation of the program.

Most patients (71.25%) were in overweight category, while 25% had normal BMI and only 3 were obese and none was morbidly obese. This is in keeping with the Indian scenario. In the intervention group, the mean weight reduced from 62.35 kg (SD = 7.04) at baseline to 59.7 kg (SD = 6.39) at end of 6-month

Table 1 Baseline characteristics of patients in intervention & control arm

Characteristic	Total *n*= 80 (%)	Intervention Arm *n*= 40 (%)	Control Arm *n*= 40 (%)	Chi Square Test
Sex				χ^2= 0.287
Female	62 (77.5)	32 (80)	30 (70)	df= 1
Male	18 (22.5)	8 (20)	10 (30)	*p*=0.592
Marital Status				χ^2= 1.067
Married	60 (75)	32 (80)	28 (70)	df=1
Widow / Widower	20 (25)	8 (20)	12 (30)	*p*=0.302
Religion				
Hindu	21 (26.2)	8 (20)	13 (32.5)	χ^2= 1.653
Muslim	50 (62.5)	27 (67.5)	23 (57.5)	df=3
Buddhist	7 (8.8)	4 (10)	3 (7.5)	*p*=0.647
Christian	2 (2.5)	1 (2.5)	1 (2.5)	
Type of Family				
Nuclear	47 (58.8)	22 (55)	25 (62.5)	χ^2= 0.487
Joint	29 (36.2)	16 (40)	13 (32.5)	df=1
Three Generation	4 (5)	2 (5)	2 (5)	*p*=0.485
Education				
Illiterate	39 (48.8)	20 (50)	19 (47.5)	χ^2= 0.117
Literate but Uneducated	30 (37.5)	15 (37.5)	15 (37.5)	df=2
Primary (till 5th std.)	9 (11.2)	4 (10)	5 (12.5)	*p*=0.943
Middle School (5th-10th)	2 (2.5)	1 (2.5)	1 (2.5)	
Occupation				
Homemaker	60 (75)	30 (75)	30 (75)	χ^2= 2.269
Unemployed	4 (5)	2 (5)	2 (5)	df=2
Unskilled Labour	14 (17.4)	6 (15)	8 (20)	*p*=0.319
Skilled Labour	1 (1.3)	1 (2.5)	-	
Shop Owner	1 (1.3)	1 (2.5)	-	
Socio Economic Status				
Lower Class	7 (8.7)	3 (7.5)	4 (10)	χ^2= 1.886
Upper Lower Class	63 (78.7)	30 (75)	33 (82.5)	df=2
Lower Middle Class	10 (12.6)	7 (17.5)	3 (7.5)	*p*=0.390
Detection of Diabetes				
1 year or less	17 (21.2)	9 (22.5)	8 (20)	χ^2= 0.149
1-5 years	52 (65)	26 (65)	26 (65)	df=2
5-10 years	11 (13.8)	5 (12.5)	6(15)	*p*=0.927
Duration of Treatment				
1 year or less	17 (21.2)	9 (22.5)	8 (20)	χ^2= 0.075
1-5 years	52 (65)	26 (65)	26 (65)	df=1
5-10 years	11 (13.8)	5 (12.5)	6(15)	*p*=0.785
Regularity of follow up & medication				χ^2= 0.053
Regular	49 (61.2)	24 (60)	24 (62.5)	df=1
Irregular	31 (38.8)	16 (40)	15 (37.5)	*p*=0.818
Type of Diet				χ^2= 0.738
Vegetarian	15 (18.75)	6 (15)	9 (22.5)	df=1
Mixed	65 (81.25)	34 (85)	31 (77.5)	*p*=0.390
Medications				
Metformin	47 (58.8)	24 (60)	23 (57.5)	χ^2= 0.052
Metformin + Glibenclamide	24 (30)	11 (27.5)	13 (32.5)	df=1
Oral Hypoglycemics + Anti-Hypertensives	9 (11.2)	5 (12.5)	4 (10)	*p*=0.820

Table 1 (continued)

Characteristic	Total *n*= 80 (%)	Intervention Arm *n*= 40 (%)	Control Arm *n*= 40 (%)	Chi Square Test
H/o Addiction	7 (8.8)	5 (12.5)	2 (5)	*p=0.215
H/o Hypertension	9 (11.3)	5 (12.5)	4 (10)	*p=0.5

Note: Rows had to be suitably clubbed to ensure applicability of test in some cases.*Fisher Exact test used

follow-up. Agurs-Collins et al. reported a mean BMI of 33.9 (SD = 5.1) and mean weight of 93.3 (SD = 18.6) [9]. Our patients showed lower BMI values at baseline and post intervention as compared to western studies. Snehalatha et al. have found that Asian Indians tend to have more visceral adipose tissue, causing higher insulin resistance, despite having lean BMI [16]. It is also possible that people with higher BMI may have been excluded due the exclusion criteria of study.

At baseline, the mean HbA1c value was 8.42%, which was higher than the WHO recommended value of 6.5%. The baseline data of A_1chieve Study by Viswanathan Mohan et al. (2013) in 20,554 Indian type 2 DM patients showed mean Hba1c 9.2% [17]. This indicates poor glycemic control among patients at baseline. Both groups show reduction in HbA1c levels after 6 months of follow-up as compared to baseline with intervention group showing highly significant ($p < 0.01$) reduction in HbA1c levels in intervention arm. Both groups show reduction in fasting blood sugar levels after 6 months of follow-up. There was a highly significant ($p < 0.01$) reduction in fasting blood sugar levels in intervention arm as compared to control arm.

We found that the median scores of Quality of Life Questionnaire for Indian Diabetics (QOLID) among intervention and control group are comparable at baseline with the exception of perceived Role Limitation Due to Physical Health in which control group fared better. There was a highly significant improvement in intervention arm as compared to control arm in all the domains of the scale with the exception of financial worries. Addressing financial worries was beyond the scope of our intervention. Nevertheless, our intervention succeeded in improving general health, treatment satisfaction symptom botherness, diet satisfaction and quality of life as a whole among the intervention arm. Separate measures need to be taken to address financial worries of the patients.

One of the major objectives of study was to assess improvement in self-care behavior. At baseline, both the groups displayed similar self-care behavior (Table 3). People rarely got blood glucose checked regularly; the awareness regarding proper and timely diet, regular medications, management of hypoglycemia and exercise was low among both groups. Education regarding improvement in self-care behavior was given with help of patient education material in local language. Post intervention, there was a significant improvement in self-care behavior in intervention group. The scores of Intervention arm show a highly significant improvement as compared to control arm after 6 months of intervention ($p < 0.001$) (Table 4).

Table 2 Quality of life at baseline and after 6 months of Intervention

Items	Median QOLID Score At baseline (95% CI)		Mann-Whitney *U*	Median QOLID Score At 6 months (95% CI)		Mann-Whitney *U*
	Intervention	Control	*p* value	Intervention	Control	*p* value
Role limitation due to physical health	16 (13.05–17)	17 (15–17.9)	*0.008*	21 (16–24)	17 (15–19)	*0.001*
Physical endurance	19 (16–22)	19 (16–22)	0.965	21 (18–25.95)	20 (16–22.95)	*0.001*
General health	7 (5–8)	7 (5–8)	0.642	9 (6–10)	8 (5–9)	*0.001*
Treatment satisfaction	8 (6–9)	7.5 (6–9)	0.762	15 (9–17.95)	8 (6–10)	*0.001*
Symptom botherness	9 (6–11)	9 (6–11)	0.852	12 (8–14.95)	9 (7–11)	*0.001*
Financial worries	9 (7–11.95)	9 (7–11.95)	1.000	9 (8–11.95)	9 (7–12)	0.964
Emotional and mental satisfaction	13 (9–15)	13 (9–15)	0.871	17 (10–19)	13 (9–15)	*0.001*
Diet satisfaction	8.50 (6–9)	8 (7–9)	1.000	11 (9–13)	8.5 (7–10)	*0.001*
Total score	89 (77.05–5.95)	88.45 (79–95)	0.497	116 (90.05–128.8)	91.5 (82.05–98.95)	*0.001*

At 6 months except for financial worries there was a significant improvement in all otherdomains of QOLID score in intervention arm as compared to control arm

Table 3 Comparison of Self-care Inventory–Revised scores at baseline and after 6 months

Items	Median SCI R Score At baseline		Median SCI R Score After 6 months	
	Intervention	Control	Intervention	Control
Get blood glucose checked regularly	2	2	4	2
Take the correct dose of diabetes pills or insulin	4	4	5	4
Take diabetes pills or insulin at the right time	3	3	5	3
Eat the correct food portions	2	2	4	2
Eat meals/snacks on time	2	2	4	2
Keep food records	1	1	3	1
Treat low blood glucose with just the recommended amount of carbohydrate	2	2	4	2
Carry quick acting sugar to treat low blood glucose	1	1	5	2
Come in for clinic appointments	3	3	5	4
Exercise	2	2	4	2
Total score	21.5 (19–24)	21.5 (19–24)	41.5 (38–44)	24 (21–34.8)

Applying Mann-Whitney *U* test, the difference in SCI R scores was not significant at baseline($p = 0.833$) but became significant at 6 months ($p < 0.01$)

Improvement in regularly blood sugar checkup, taking medications on time, reduced food proportions and consumption of frequent small meals, managing hypoglycemia and regular exercise was evident. Peer support played a key role in improving physical activity. The patients also attended appointments regularly. This has led to a highly significant improvement in an overall self-care behavior. The only behavior that did not show marked improvement was the maintaining of diet records. This could be attributed to the lack of conceptual understanding of nutrition and the absence of a trained dietician or low level of education among the participants. In studies conducted in the west, most of the respondents are often literate and this was not the condition in the current study. Thirty-five out of 40 patients in intervention arm attended all 12 follow-ups while only 21 of control patients attended all follow-ups. There were no dropouts from any of the groups. Intervention arm showed better follow-up as compared to control arm.

Conclusion

It is possible to overcome the barriers of providing diabetes care at resource poor primary settings by developing need-based comprehensive programs. There was good patient response to all 4 key areas of intervention that the program sought to encompass (namely knowledge, regular follow-up and medication, diet and physical activity). There was improvement in glycemic control, self-care behavior and quality of life of diabetic patients in the intervention arm.

Table 4 Comparison of lab and anthropometric findings at baseline and 6 months

	Baseline			After 6 months of intervention		
	Intervention arm	Control arm	*p* value	Intervention arm	Control arm	*p* value
Mean Weight (SD)	62.35 (7.04)	64.33 (6.91)	0.723	59.7 (6.39)	63.35 (6.8)	*0.016*
Mean BMI (SD)	25.96 (1.9)	26.42 (1.7)	0.23	24.9 (1.77)	26.02 (1.6)	*0.003*
Mean HbA1c level (SD)	8.44 (1.802)	8.4 (1.87)	0.937	7.56 (1.67)	8.19 (1.77)	*0.001*
Mean fasting BSL (SD)	184.78 (37.447)	188.03 (44)	0.723	158.13 (28.83)	179.03 (35.18)	*0.005*

p values for unpaired *t* test

There was statistically significant difference in Mean Weight, Mean BMI, Mean HbA1c and Mean fasting BSL values in intervention arm at 6 month as compared to the control arm

Acknowledgments Late Dr. Ratnendra Shinde, Former Professor & Head, Community Medicine and the Department of Community Medicine Seth GS Medical College and KEM Hospital, Mumbai.

Compliance with ethical standards

The study was approved by Institutional ethics committee of Seth GS Medical College & KEM Hospital.

Conflict of interest The authors declare that they have no conflict of interest.

References

1. Aguiree F, Brown A, Cho NH, Dahlquist G, Dodd S, Dunning T, et al. IDF diabetes atlas: International Diabetes Federation; 2013.
2. Sathish T, Williams ED, Pasricha N, Absetz P, Lorgelly P, Wolfe R, et al. Cluster randomised controlled trial of a peer-led lifestyle intervention program: study protocol for the Kerala diabetes prevention program. BMC Public Health. 2013;13(1):1035.
3. Venkataraman K, Kannan AT, Mohan V. Challenges in diabetes management with particular reference to India. Int J Diabetes Dev Ctries. 2009;29(3):103–9.
4. Anand K, Shah B, Yadav K, Singh R, Mathur P, Paul E, et al. Are the urban poor vulnerable to non-communicable diseases? A survey of risk factors for non-communicable diseases in urban slums of Faridabad. Natl Med J India. 2007;20:115–20.
5. Nagpal J, Bhartia A. Quality of diabetes care in the middle- and high-income group populace: the Delhi diabetes community (DEDICOM) survey. Diabetes Care. 2006;29:2341–8.
6. Ramachandran A, Mary S, Sathish CK, Selvam S, Seeli AC, Muruganandam M, et al. Population based study of quality of diabetes care in southern India. JAPI. 2008;56:513–6.
7. Shobhana R, Begum R, Snehalatha C, Vijay V, Ramachandran A. Patients' adherence to diabetes treatment. J Assoc Physicians India. 1999;47:1173–5.
8. Weinger K, Beverly EA, Lee Y, Sitnokov L, Ganda OP, Caballero AE. The effect of a structured behavioral intervention on poorly controlled diabetes: a randomized controlled trial. Arch Intern Med. 2011;171(22):1990–9.
9. Agurs-Collins TD, Kumanyika SK, Ten Have TR, Adams-Campbell LL. A randomized controlled trial of weight reduction and exercise for diabetes management in older African-American subjects. Diabetes Care. 1997;20(10):1503–11.
10. Gucciardi E, DeMelo M, Offenheim A, Stewart DE. Factors contributing to attrition behavior in diabetes self-management programs: a mixed method approach. BMC Health Serv Res. 2008;8(1):33.
11. Balagopal P, Kamalamma N, Patel TG, Misra R. A community-based diabetes prevention and management education program in a rural village in India. Diabetes Care. 2008;31(6):1097–104.
12. Nagpal J, Kumar A, Kakar S, Bhartia A. The development of quality of life instrument for Indian diabetes patients (QOLID): a validation and reliability study in middle and higher income groups. J Assoc Physicians India. 2010;58:295–304.
13. Weinger K, Butler HA, Welch GW, La Greca AM. Measuring diabetes self-care a psychometric analysis of the self-care inventory-revised with adults. Diabetes Care. 2005;28(6):1346–52.
14. Peyrot M, Rubin RR. Behavioral and psychosocial interventions in diabetes a conceptual review. Diabetes Care. 2007;30(10):2433–40.
15. Kernan WN, Viscoli CM, Makuch RW, Brass LM, Horwitz RI. Stratified randomization for clinical trials. J Clin Epidemiol. 1999;52(1):19–26.
16. Snehalatha C, Viswanathan V, Ramachandran A. Cutoff values for normal anthropometric variables in Asian Indian adults. Diabetes Care. 2003;26(5):1380–4.
17. Mohan V, Shah S, Saboo B. Current glycemic status and diabetes related complications among type 2 diabetes patients in India: data from the A1chieve study. J Assoc Physicians India. 2013;61(1 Suppl):12–5.

Mandar Padmakar Baviskar Formerly Resident Doctor, Community Medicine, Seth GS Medical College & KEM Hospital, Mumbai

Sumitra Rangari Formerly Resident Doctor, Community Medicine, Seth GS Medical College & KEM Hospital, Mumbai

Biswendu Sundar Mohanta Formerly Resident Doctor, Community Medicine, Seth GS Medical College & KEM Hospital, Mumbai

International Journal of Diabetes in Developing Countries (2021) 41:164–171
https://doi.org/10.1007/s13410-020-00842-3

ORIGINAL ARTICLE

The predictors of perceived stress in patients with type 2 diabetes in Turkey: styles of coping with stress and metabolic variables

Gulcan Bakan[1] · Fadime Hatice Inci[2]

Received: 3 November 2019 / Accepted: 13 June 2020 / Published online: 19 August 2020

Abstract

Background The study was conducted to examine the relationship between metabolic control variables and the coping strategies of type 2 DM patients with stress perception.

Methods The study design was a descriptive and cross-sectional survey. The study sample consisted of 153 patients who reported to the Internal Medicine Outpatient Clinic. Data were collected using the descriptive characteristic form, perceived stress scale, and the coping styles inventory.

Results Patients' mean BMI was 28.82 ± 7.14, and the mean HbA1c level was 10.31 ± 2.75. The prevalence of metabolic syndrome identified in patients with type 2 DM was 68%. A negative correlation was determined between perceived stress and age and the problem-focused coping method, whereas a positive correlation was found between BMI, number of accompanying chronic diseases, insulin use period, waist circumference, and emotion-focused coping method and perceived stress.

Conclusions The variable that most affects the stress perception levels of patients with diabetes mellitus is problem-focused coping. Following these results, teaching patients how to use efficient techniques for stress coping and providing support for psychosocial care is necessary.

Keywords Diabetes mellitus · Metabolic variables · Perceived stress · Coping

Introduction

Diabetes mellitus (DM) is one of the most common metabolic disorders in the world [1–4]. According to the Diabetes Atlas (2017) of the International Diabetes Federation (IDF), the prevalence of DM among the adult population (20–79 years) worldwide is 8.8% [5]. According to the TURDEP-II study of Turkey Diabetes Epidemiology, type 2 diabetes in Turkish adults (13.7%) is above the global average [6]. DM-caused metabolic disorders may negatively influence both the mood and cognitive functions of individuals [7, 8]. Perceived stress level is a factor that influences the metabolic control level [1, 4, 9–11]. While stress increases glucose and HbA1c levels in DM patients, diabetes and its complications increase stress levels. For this reason, stress is a problem that plays an important role in DM patients as both the cause and the effect of the disease [12]. Once the disease is perceived as a threat by affected individuals, their anxiety and stress level will increase [13]. Cortisol, which is secreted due to stress, may cause hormonal abnormalities and obesity [14, 15]. Earlier research has also demonstrated that many people with type 2 diabetes experience high levels of emotional distress stemming from concerns and worries associated with their diabetes, its management, and the experience of more complications [10, 16, 17]. According to previous studies, living with type 2 diabetes over a long period and using insulin may increase psychological distress [18–22]. Worries about increased BMI may be a source of higher levels of distress [21]. In the literature, more intensive diabetes treatment, long disease duration, increased numbers of accompanying chronic diseases, the number of diabetes-related complications, increased BMI, and coping methods are all important indicators of diabetes-related strains that may influence the

✉ Gulcan Bakan
gbakan@pau.edu.tr

Fadime Hatice Inci
hemel@pau.edu.tr

[1] Internal Medicine Nursing Department, Faculty of Health Sciences, Pamukkale University, Kinikli Campus, Universite Street. Number: 11, 20160 Pamukkale, Denizli, Turkey

[2] Department of Public Health Nursing, Faculty of Health Sciences, Pamukkale University, Kinikli Campus, Universite Street. Number: 11, 20160 Pamukkale, Denizli, Turkey

level of distress among adults with type 2 diabetes [1, 10, 11, 16, 18, 20, 21, 23–25]. Coping methods are pictured as a reaction set of behavioral and cognitive responses against stressful circumstances [25–27]. To overcome the stress that manifests due to diabetes and other stress factors, DM patients use problem-focused and emotion-focused methods [2, 28]. The patient uses direct coping behaviors that change the person-environment relationship in the problem-focused method, whereas in the emotion-focused method, the change in the person-environment relationship manifests in the form of the patient's interpretation of the environment [21]. Studies have demonstrated that during the clinical course of patients, using the problem-focused coping method loses less ground, and patients using the emotion-focused coping method have poor metabolic control [9, 29]. Using the problem-focused coping method is the most efficacious solution to cope with the stress, which exerts negative effects on the disease and treatment [25].

In the literature of our country, there is no study on the relationship between metabolic control and coping styles and perception of stress in DM patients. The aim of this research is to assess the perception of stress predictors of type 2 diabetic patients in Turkey and the relationship between them.

Methods

The study design was a descriptive and cross-sectional survey.

Sample and setting

The population of the research included adults aged 19 and older having type 2 DM who were admitted to the Internal Medicine Outpatient Clinic of the State Hospital. A total of 153 patients admitted to the Internal Medicine Outpatient Clinic between March 2015 and March 2016 formed the sample. Applying the multiple regression R^2 (deviation from zero) test in the G*Power program, a power analysis was conducted at the end of the research. In the retrospective analysis, the effect size (f^2) was found to be 0.37 and the power of the study was 0.99 in the calculation performed by taking the sample size as 153, the squared multiple correlation (R^2) as 0.272, and the significance level (p) as 0.05. Data were obtained from adults aged 18 years or older who participated in the research of their own free will, were already diagnosed with Type 2 DM at least 6 months prior, and had no deficiency in Turkish reading or comprehension.

Data collection tools

Data were collected using the descriptive characteristic form, perceived stress scale (PSS), and coping styles inventory (CSI).

Descriptive characteristic form

Prepared by the researchers, this form includes questions gathering data about the sociodemographic characteristics of the patients such as sex, educational level, marital status, employment, family income, health insurance, and other information (body mass index, duration of diabetes, treatment used, chronic diseases accompanying the diabetes, and metabolic syndrome status).

Metabolic syndrome status According to the revised NCEP ATP III criteria for the classification of metabolic syndrome, study participants should have at least three of the five following components, which were categorized as "normal" or "altered:" large waist circumference (102 cm in men or 88 cm in women); elevated triglycerides (150 mg/dL) or on drug treatment for elevated triglycerides; reduced HDL-C (40 mg/dL in men and 50 mg/dL in women) or on drug treatment for reduced HDL-C; elevated blood pressure (130 mmHg systolic blood pressure or 85 mmHg diastolic blood pressure) or on antihypertensive drug treatment in a patient with a history of hypertension; and elevated fasting glucose (100 mg/dL) or on drug treatment for elevated glucose.

Body mass index

Anthropometric measurements were measured by researchers following standard procedures. Body mass index (BMI) was defined as weight in kilograms divided by height in meters squared [30]. BMI was categorized as underweight (< 18.50), normal (18.50–24.99), overweight (25.00–29.99), and obese (≥ 30.00).

Perceived stress scale

Perceived stress scale was developed by Cohen et al. in 1983 [31]. In addition to the 14-item long-form, ASO has two other forms with 10 and 14 items. Form with 10 items has been used in this research. The responses were measured using a five-point Likert-type scale (0 = never to 4 = very often). The items 4, 5, 7, and 8 of the scale are reversely scored. The possible score range was from 0 to 40. A higher score indicates higher perceived stress. The internal consistency of the original scale was 0.78. The scale was adapted into Turkish by Eskin et al. Cronbach's α of the Turkish version was 0.82 [32]. The internal consistency was found to be 0.69 in the present study.

The coping styles inventory

The scale was developed by Folkman and Lazarus [33]. It was adapted into Turkish by Sahin and Durak in 1995 [34]. It

contains 30 questions and uses a 4-point Likertscale (between 0 and 3). The items 1 and 9 of the scale are reversely scored. Coping styles inventory (CSI) consists of five subscales as self-confidence, helpless, optimistic, submissive, and seeking of social support approaches. The scale measures two major styles of coping with stress. These are problem-focused/effective and emotion-focused/ineffective styles. Problem-focused styles are the subscales of seeking of social support, optimistic approach, and self-confident approach, whereas emotion-focused styles are the subscales of helpless approach and submissive approach. The score of subscale is obtained by dividing total score obtained from the relevant subscale into the number of items. The high score obtained from the subscale signifies that the subscale is mainly used by individuals. Cronbach's Alpha values vary between 0.45 and 0.80 for the mentioned five factors [34].

Data analysis

Data were analyzed using the Statistical Package for Social Sciences (SPSS). For the intergroup comparison, an independent sample *t* test and Kruskal Wallis test were used. To determine the link between dependent variables and independent variables, Pearson's correlation and stepwise multiple regression were used. The significance level was defined as $p < 0.05$.

Results

The average age of the participants was 57.03 ± 13.87; 47.1% of the patients were women, 66.7% were primary school graduates, 73.9% were married, 39.9% were employed, 48.4% were in the middle-income level, and almost all had social security. The average BMI of the patients was 28.82 ± 7.14, and the average HbA1c level was 10.31 ± 2.75 (Table 1).

The prevalence of metabolic syndrome determined in type 2 DM patients was 68% in this study. Based on the distribution of prevalence of components of metabolic syndrome, the levels of FBS, BP, TG, and WC were high in 94.10%, 58.8%, 56.90%, and 46.40% of the sample, respectively. In contrast, HDL was low in 35.30% of patients, as shown in Fig. 1. Meanwhile, 40.5%, 25.5%, and 2% of the patients met three, four, and five of the metabolic syndrome criteria, respectively.

The specifications, illness, and treatment characteristics of the DM patients were analyzed from the point of the stress perceived. There was a significant difference between the stress perception of insulin users and nonusers. When the coping methods were investigated with regard to the stress perception, the stress perception of the patients experiencing emotion-focused coping methods was found to be higher, and the difference was significant (Table 2).

The correlation results exhibit a significant link between some variables and perceived stress (Table 3). A negative correlation seemed to exist between perceived stress and age and problem-focused coping methods; meanwhile, positive correlations were found between BMI, the number of accompanying chronic diseases, the insulin use period, the waist circumference, and the emotion-focused coping method and the perceived stress. No significant link was found between other variables and perceived stress.

When the R^2 values were analyzed, the problem-focused coping method encompassed 11% of the total variance in the first phase. In the second stage, with the addition of emotion-focused coping methods into the predictive model, the total variance increased to 20%; in the third stage, with the number of accompanying chronic diseases variable, the total variance increased to 22%; in the fourth stage, with the age variable, the total variance increased to 25%; and in the final stage, with the insulin period variable, the total variance increased to 27% ($R^2 = 0.272$, $R = 0.522$, $p < 0.01$) (Table 4).

The stepwise multiple regression analysis found that the most important predictor was the problem-focused coping method ($\beta = -0.284$), followed by the emotion-focused coping method ($\beta = 0.288$), the number of accompanying chronic diseases ($\beta = 0.219$), the age ($\beta = -0.206$), and the insulin use period ($\beta = 0.146$).

When the aspects of the relationships between the variables were examined, the problem-focused coping and age were negatively correlated with the perceived stress level, whereas the emotion-focused coping method, number of accompanying chronic diseases, and insulin use period were positively correlated with the perceived stress level.

In conclusion, as a result of the analyses conducted with regard to the regression assumptions, the use of the model obtained was significant in explaining the DM patients' stress perception levels, encountering no multiple correlations and homoscedasticity problems and exhibiting normally distributed error terms.

Discussion

Our study investigated predictions of the stress perception variables of DM patients in Turkey. The most predictive variable was the coping methods used by the patients followed by some metabolic variables. Conducting such a study is essential, as defining the factors and effect levels in relation to DM patients' stress perception will identify the cases that need to be addressed first. Long-term stress and inadequate stress-coping mechanisms lead to mental, emotional, and physiological problems in individuals [35, 36].

DM patients' metabolic control variables were investigated, and 68% of the patients met the metabolic syndrome criteria per the National Cholesterol Education Program

Table 1 Descriptive characteristics of patients with diabetes mellitus

Variables	Frequency (%)	$\bar{x}$(SD)
Age		57.03 (13.87)
Gender		
Female	72 (47.1)	
Male	81 (52.9)	
Educational level		
Illiterate-literate	20 (13.1)	
Primary school	102 (66.7)	
Middle school	20 (13.1)	
High school/university	11 (7.2)	
Marital status		
Married	113 (73.9)	
Single	40 (26.1)	
Employment		
Working	61 (39.9)	
Not working	92 (60.1)	
Family income		
High	6 (3.9)	
Moderate	74 (48.4)	
Low	73 (47.7)	
Body mass index		28.82 ± 7.14
HbA1c		10.31 ± 2.75

(NCEP). Many studies have been conducted by using the NCEP ATP III criteria in DM patients. In the studies performed in type 2 diabetes patients in Turkey, metabolic syndrome prevalence was determined to be 70.5% [37] and 80.4% [38]. The metabolic syndrome prevalence of type 2 DM patients in some studies performed outside Turkey varies between 45.0% and 91.9% [39–44]. Among these studies, some results were similar to our research findings [45] and some were different. The reason for this may be because the average age, sex, and duration of DM in the countries in which the studies were performed differ from each other.

The HbA1c level of the patients was found to be 10.31% in our study, but no significant relationship was discovered between this and the perceived stress. Similarly, no relationship was found between psychometric variables and HbA1c in some studies conducted [21, 46, 47]. In a study conducted in Malaysia, a significant relationship was not found between depression, anxiety, and stress and glycemic control in patients diagnosed with type II DM with an average age of 61 [48], while in a study conducted in a younger population in the same country, anxiety, and stress were significantly correlated with higher HbA1c levels [49]. In accordance with some other study results, there is a significant association between HbA1c and perceived stress and coping methods [1, 9–11]. In a review study which is included 22 different studies results from different countries, there was a significant relationship between HbA1c and stress factors and coping methods whereas according to other study results in the same review study, there was no relationship as in our study results [50]. These differences are thought to be due to the studies being performed in patients of different races, ages, sexes, and HbA1c levels [1, 10, 46, 50].

In our study, 70% of the patients were overweight and obese, the average BMI was 28.82, and the average waist circumference was 94.16. We found that the higher the BMI value of the DM patients, the more their perception of stress increased, and the patients with increased stress perception benefit from emotion-focused coping methods much more. In the studies performed, obese people were found to use both methods, but they used emotion-focused coping methods much more [21]. Once they perceive the situation as irrevocable, these kinds of patients tend to accept and submit to it, and then, they begin to use emotion-focused coping methods in particular. Patients should be encouraged to lose weight and be oriented to benefit from more active coping methods, which are problem-focused coping methods.

At the end of the study, problem-focused coping, emotion-focused coping, and number of accompanying chronic

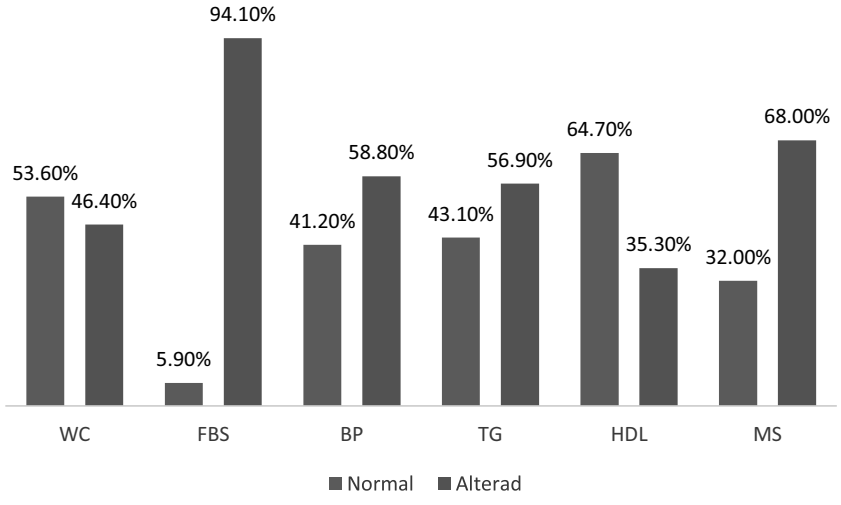


Fig. 1 Distribution of the components of metabolic syndrome in patients with diabetes mellitus. *WC* waist circumference, *FBS* fasting blood sugar, *BP* blood pressure, *TG* triglycerides, *HDL* high-density lipoprotein cholesterol, *MS* metabolic syndrome

Table 2 Descriptive characteristics of the patients, average stress score distribution according to disease and treatment-related characteristics

Variables	*N*	Mean	Std. deviation	*t* or KW	*p*
Gender					
Male	72	19.68	6.71	− 1.284	0.201
Female	81	21.06	6.58		
Educational level					
Illiterate-literate	20	19.70	6.01		
Primary school	102	20.63	6.59	0.281	0.963
Middle school	20	20.30	7.85		
High school/university	11	19.91	6.86		
Marital status					
Married	113	19.88	6.69	− 1.655	0.100
Single	40	21.90	6.39		
Employment					
Working	61	21.15	7.27	1.115	.267
Not working	92	19.92	6.20		
Family income					
High	6	16.83	8.95		
Moderate	74	20.08	6.28	2.344	0.310
Low	73	21.04	6.80		
Chronic diseases accompanying the diabetes					
No	59	19.56	6.97	− 1.258	0.210
Yes	94	20.95	6.42		
Insulin use					
Yes	112	21.38	6.55	3.039	0.003
No	41	17.78	6.29		
Regular drug use					
Yes	140	20.36	6.71	− 0.332	0.740
No	13	21.00	6.28		
Metabolic syndrome					
Yes	104	19.89	6.90	1.406	0.162
No	49	21.51	6.02		
Stress coping style					
Problem-focused	119	19.08	6.32	− 4.987	0.001
Emotion-focused	34	24.88	5.87		

t Student *t* test, *KW* Kruskall Wallis

Table 3 Relationship between perceived stress and other variables

	PSS *r*
Age	− 0.165*
BMI	0.185*
Duration of diabetes	0.043
Number of chronic diseases accompanying	0.160*
Insulin use period (year)	0.191*
Metabolic variables	
HbA1c	0.098
Waist circumference	0.199*
Fasting blood sugar	0.048
Blood pressure systolic	0.040
Blood pressure diastolic	0.137
Triglycerides	− 0.049
High-density lipoprotein	− 0.013
Problem-focused styles	− 0.324**
Emotion-focused styles	0.289**

***PSS* perceived stress, *r* correlation coefficient, $p < 0.05$, $p < 0.01$

diseases, age, and duration of insulin use, which are stress-overcoming strategies, were found to be predictor variables of perceived stress level. The analyses showed that the five above mentioned variables had a 27% explanatory stress perception, and the relationship between them was at a level of 0.52. The variable that contributed the most to the stress perception levels of DM patients who participated in this study was problem-focused coping. As the problem-focused coping levels and age increased, the patients experienced lower stress perception; as the emotional coping levels, year of insulin use, and the number of accompanying chronic diseases increased, the patients experienced higher stress perception. There is no way to completely separate life from stress; what matters is how stress is interpreted and addressed. It will be useful to have DM patients recognize their stresses and to teach them how to cope with stress [7].

In our research, the problem-focused coping method was the best predictive variable of the stress perception; the higher its level, the lower the stress perception. The problem-focused coping method is an active coping method and includes rational responses that were concentrated on the data and deliberate actions against stress sources [21, 27]. It subsumes actions that change the environment or the way people behave [27]. In the studies conducted, good glycemic results have been obtained in DM patients using problem-focused coping methods, compliance with treatment increased, and symptoms of depression were seen less often [11, 23, 26, 46]. Stimulation of the benefits from problem-focused coping may hinder the progression of stress in patients. In addition to being a physical disease, DM also has psychological and psychosocial bearings. Individuals with a diagnosis of DM should be considered holistically while maintaining the metabolic control on the one hand, and the individual's mood should be supported by interventions on the other hand [7]. In our study, type 2 DM patients were found to experience lower stress perception as they grew older. In other studies, a negative relationship between age and stress has been found [1, 10]. Elderly DM patients are more inclined to cope while young people have more difficulty in perceiving the burden related to diabetes management and coping with the disease [10, 21]. The retirement of older patients and the lack of work stress along with a more stable and regular life may be among the reasons that lower the perception of stress. In line with

Table 4 Results of stepwise multiple regression analyses found using the perceived stress as a dependent variable

Models	Variables	*B*	SE	*β*	*R*	R^2		
Model 1	Problem-focused styles	− 3.930	0.933	− 0.324**	0.324	0.105	17.734	0.000
Model 2	Problem-focused styles	− 4.087	0.888	− 0.337**	0.474	0.197	18.380	0.000
	Emotion-focused styles	3.817	0.922	0.303**				
Model 3	Problem-focused styles	− 4.010	0.876	− 0.331**	0.503	0.224	14.372	0.000
	Emotion-focused styles	3.947	0.911	0.313**				
	Number of accompanying chronic diseases	1.033	0.449	0.167*				
Model 4	Problem-focused styles	− 3.769	0.869	− 0.311**	0.444	.253	12.503	0.000
	Emotion-focused styles	3.848	0.898	0.306**				
	Number of accompanying chronic diseases	1.334	0.460	0.215**				
	Age	− 0.084	0.036	− 0.176*				
Model 5	Problem-focused styles	− 3.440	0.876	− 0.284**	0.522	0.272	10.993	0.000
	Emotion-focused styles	3.629	0.896	0.288**				
	Number of accompanying chronic diseases	1.361	0.456	0.219**				
	Age	− 0.099	0.036	− 0.206**				
	Insulin use period (year)	0.134	0.067	0.146*				

these findings, patients should be taught and supported in problem-oriented coping methods starting from a younger age.

In our study, as the emotion-focused coping levels increased, the patients experienced much higher stress perception. During the coping process, individuals with high levels of perceived stress tend to use emotion-focused coping methods that include helplessness [16, 23]. Studies have shown that treatment compliance problems, poor glycemic control, and psychosocial problems related to illness are more common in DM patients using emotion-focused coping methods [16, 23, 25, 46]. In particular, newly diagnosed DM patients experience more anxiety and depression and believe that diabetes will negatively affect their future and thus use emotion-focused coping methods [24, 51]. Benefiting from the inert coping methods for chronic diseases such as DM may have critical adverse results on the physical health and sanity of individuals [9]. If patients do not resist the threats to their individual integrity, especially on a mental level, the accumulated and intensifying effects of stresses that they cannot cope with reverberate in their behaviors [36]. With regard to Folkman and Lazarus, in the problem-focused coping method cognitive strategies are used, whereas the emotion-focused coping method targets diminishing the emotional effects of stress factors [33]. In line with these results, patients should be taught methods of coping, and support should be provided to use problem-oriented coping methods rather than emotion-based coping methods. In our study, the stress perception of DM patients using insulin was found to be higher than that of patients who did not use insulin, and their stress perception increased as the years of insulin use increased while the problem-focused coping levels decreased as their stress perception increased. In studies conducted on DM patients, insulin use has been reported to increase stress due to the need for self-injection and blood glucose monitoring and the fear of hypoglycemia [18–22, 52]. For patients, insulin treatment creates a perception that the disease is worsening. In our research, as the number of chronic diseases accompanying DM increased, the patients were found to experience higher stress perception. As they do not know how to handle more than one illness, they start to experience more stress. The symptoms and treatment process in chronic diseases lead to preventing daily life activities of the individual, decreased capacity of individual coping, changes in perception of identity, sacrifices in important goals, financial loss to the family and society, and increased level of individual dependence and anxiety [53].

The results of our study found that the use of the problem-oriented coping method was the variable that predicted the stress perception positively. The fact that patients use emotion-oriented coping methods increases their perception of stress. In line with these results, diabetic patients should be taught at the earliest age to handle stress and be encouraged to use effective methods. Healthcare team members, using their education and counseling knowledge and skills, provide basic knowledge and skills to DM patients and their families, check this knowledge and these skills periodically and evaluate patients physiologically and psychologically, to enable early detection of problems and thus early treatment.

Acknowledgments The authors wish to thank all participant in this research.

Funding information This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical considerations The study was approved by the ethics committees of the university hospital (60116787 – 020/28462). The patients were informed about the participation in the study, and their consent was obtained. The patients were informed that their data would be kept confidential by the researchers and that, even if the patients had already provided consent, during the process, they could leave the study at any time.

References

1. Hara Y, Hisatomi M, Ito H, Nakao M, Tsuboi K, Ishihara Y. Effects of gender, age, family support, and treatment on perceived stress and coping of patients with type 2 diabetes mellitus. Biopsychosoc Med. 2014;8(1):1–11.
2. Davies M Psychological aspects of diabetes management. Medicine (Baltimore) [Internet] 2015;43(1):57–59. Available from: https://doi.org/10.1136/bmj.39474.
3. Schinckus L, Dangoisse F, Van den Broucke S, Mikolajczak M. When knowing is not enough: emotional distress and depression reduce the positive effects of health literacy on diabetes self-management. Patient Educ Couns. 2018;101(2):324–30.
4. Gahlan D, Rajput R, Gehlawat P, Gupta R. Prevalence and determinants of diabetes distress in patients of diabetes mellitus in a tertiary care centre. Diabetes Metab Syndr Clin Res Rev [Internet]. 2018;12(3):333–336. Available from: https://doi.org/10.1016/j.dsx.2017.12.024.
5. International Diabetes Federation. IDF DIABETES ATLAS 2017 [Internet]. Eighth edi. IDF Diabetes Atlas, 8th edition. 2017. 1–150 p. Available from: www.diabetesatlas.org
6. Satman I, Imamoglu S, Yılmaz C, Ayvaz G, Comlekci A. Diabetes in the world and Turkey. Turkish J Endocrinol Metab. 2012;16(1):1–56.
7. Kumcagız H, Özenoglu A, Avcı IA, Ugurlu S. Anxiety levels in patients with type 2 diabetes mellitus and coping with stress. Cumhuriyet Med J. 2009;31(2):122–9.
8. Tareen RS, Tareen K. Psychosocial aspects of diabetes management: dilemma of diabetes distress. Transl Pediatr. 2017;6(4):383–96.
9. Yi JP, Yi JC, Vitaliano PP, Weinger K. How does anger coping style affect glycemic control in diabetes patients? Int J Behav Med. 2008;15(3):167–72.
10. Pintaudi B, Lucisano G, Gentile S, Bulotta A, Skovlund SE, Vespasiani G, et al. Correlates of diabetes-related distress in type 2 diabetes: findings from the benchmarking network for clinical and humanistic outcomes in diabetes (BENCH-D) study. J Psychosom Res. 2015;79(5):348–54.
11. Dyke ML, Cuffee YL, Halanych JH, McManus RH, Curtin C, Allison JJ. The relationship between coping styles in response to unfair treatment and understanding of diabetes self-care. Diabetes Educ. 2013;39(6):848–55.
12. Zamani-Alavijeh F, Araban M, Koohestani HR, Karimy M. The effectiveness of stress management training on blood glucose control in patients with type 2 diabetes. Diabetol Metab Syndr. 2018;10(39):1–9.
13. Schneiderman N, Ironson G, Siegel SD. Stress and health: psychological, behavioral, and biological determinants. Annu Rev Clin Psychol. 2005;1(1):607–28.
14. Falco G, Pirro PS, Castellano E, Anfossi M, Borretta G, Gianotti L. The relationship between stress and diabetes mellitus. J Neurol Psychol. 2015;3(1):1–7.
15. Önem Akcakaya R, Celik ES. Stres and psychiatric approach in coping with stress. Turkish Fam Physician. 2014;5(2):18–25.
16. Kokoszka A. Treatment adherence in patients with type 2 diabetes mellitus correlates with different coping styles, low perception of self-influence on disease, and depressive symptoms. Patient Prefer Adherence. 2017;11:587–95.
17. Roy K, Iqbal S, Gadag V, Bavington B. Relationship between psychosocial factors and glucose control in adults with type 2 diabetes. Can J Diabetes [Internet]. 2020;1–7. Available from: https://doi.org/10.1016/j.jcjd.2020.01.005.
18. Gonzalez JS, Esbitt SA, Schneider HE, Osborne PJ, Kupperman EG. Psychological issues in adults with type 2 diabetes. In: Pagoto S, editor. Psychological co-morbidities of physical illness: a behavioral medicine perspective. New York: Springer; 2011. p. 73–121.
19. Tuncay T, Musabak I, Gok DE, Kutlu M. The relationship between anxiety, coping strategies and characteristics of patients with diabetes. Health Qual Life Outcomes. 2008;6:79.
20. Iversen MM, Nefs G, Tell GS, Espehaug B, Midthjell K, Graue M, et al. Anxiety, depression and timing of insulin treatment among people with type 2 diabetes: nine-year follow-up of the Nord-Trøndelag Health Study, Norway. J Psychosom Res. 2015;79(4):309–15.
21. Karlsen B, Oftedal B, Bru E. The relationship between clinical indicators, coping styles, perceived support and diabetes-related distress among adults with type 2 diabetes. J Adv Nurs. 2012;68(2):391–401.
22. Parsa S, Aghamohammadi M, Abazari M. Diabetes distress and its clinical determinants in patients with type II diabetes. Diabetes Metab Syndr Clin Res Rev. 2019;13(2):1275–9.
23. Gois C, Dias VV, Raposo JF, Do Carmo I, Barbosa A. Vulnerability to stress, anxiety and depressive symptoms and metabolic control in type 2 diabetes. BMC Res Notes. 2012;5(271):1–6.
24. Madhu SV, Siddiqui A, Desai NG, Sharma SB, Bansal AK. Chronic stress, sense of coherence and risk of type 2 diabetes mellitus. Diabetes Metab Syndr Clin Res Rev. 2019;13(1):18–23.
25. Burns RJ, Deschênes SS, Schmitz N. Associations between coping strategies and mental health in individuals with type 2 diabetes: prospective analyses. Health Psychol. 2016;35(1):78–86.
26. Zhang CX, Chen YM, Chen WQ. Association of psychosocial factors with anxiety and depressive symptoms in Chinese patients with type 2 diabetes. Diabetes Res Clin Pract. 2008;79(3):523–30.
27. Lazarus RS. Cognition and emotion from the ret viewpoint. J Ration Cogn Ther. 1995;13(1):29–54.
28. Chew B-H. Psychological aspects of diabetes care: effecting behavioral change in patients. World J Diabetes. 2014;5(6):796–808.
29. Folkman S, Chesney M, McKusick L, Ironson G, Johnson DS, Coates TJ. Translating coping theory into an intervention. In: Eckenrode J, editor. The social context of coping. New York: Springer; 1991. p. 239–60.
30. Nuttall FQ. Body mass index: obesity, BMI, and health: a critical review. Nutr Today. 2015;50(3):117–28.
31. Cohen S, Kamarck T, Mermelstein R. A global measure of perceived stress. J Health Soc Behav. 1983;24(4):385–96.
32. Eskin M, Harlak H, Demirkiran F, Dereboy C. The adaptation of the perceived stress scale into Turkish: a reliability and validity analysis. New Symp J. 2013;51(3):132–40.
33. Folkman S, Lazarus RS. An analysis of coping in a middle-aged community sample. J Health Soc Behav. 1980;21(3):219–39.
34. Şahin NH, Durak A. A brief coping styles inventory for university students. Turk J Psychol. 1995;10(34):56–73.

35. Orzechowska A, Zajaczkowska M, Talarowska M, Gałecki P. Depression and ways of coping with stress: a preliminary study. Med Sci Monit. 2013;19:1050–6.
36. Jordan TR, Khubchandani J, Wiblishauser M. The impact of perceived stress and coping adequacy on the health of nurses: a pilot investigation. Nurs Res Pract. 2016;2016:1–11.
37. Serkan İ. Tip 2 Diyabetli Hastalarda Metabolik Sendrom Prevalansı. Turgut Özal Tıp Merk Derg. 2015;15(1):29–33.
38. Oktay M, Oktay G, Haydar EY, Sönmez Cemil I, Turan SF. The evaluation of metabolic syndrome frequency and its components in newly diagnosed type-2 diabetic patients. Ankara Med J. 2012;12(4):174–7.
39. Lira Neto JCG, de Almeida Xavier M, Borges JWP, de Araújo MFM, Damasceno MMC, de Freitas RWJF. Prevalence of metabolic syndrome in individuals with type 2 diabetes mellitus. Rev Bras Enferm. 2017;70(2):265–70.
40. Lone S, Lone K, Khan S, Pampori RA. Assessment of metabolic syndrome in Kashmiri population with type 2 diabetes employing the standard criteria's given by WHO, NCEPATP III and IDF. J Epidemiol Glob Health [Internet]. 2017;7(4):235–239. Available from: https://doi.org/10.1016/j.jegh.2017.07.004.
41. Yadav D, Mahajan S, Subramanian SK, Bisen PS, Chung CH, Prasad GB. Prevalence of metabolic syndrome in type 2 diabetes mellitus using NCEP-ATPIII, IDF and WHO definition and its agreement in Gwalior Chambal region of Central India. Global J Health Sci. 2013;5(6):142–55.
42. Ahmed A, Khan TE, Yasmeen T, Awan S, Islam N. Metabolic syndrome in type 2 diabetes: comparison of WHO, modified ATPIII & IDF criteria. J Pak Med Assoc. 2012;62(8):569–74.
43. Rodríguez Bernardino Á, García Polavieja P, Reviriego Fernández J, Serrano RM. Prevalence of metabolic syndrome and consistency in its diagnosis in type 2 diabetic patients in Spain. Endocrinol Nutr. 2010;57(2):60–70.
44. Pokharel DR, Khadka D, Sigdel M, Yadav NK, Acharya S, Kafle RC, et al. Prevalence of metabolic syndrome in Nepalese type 2 diabetic patients according to WHO, NCEP ATP III, IDF and harmonized criteria. J Diabetes Metab Disord. 2014;13(1):1–13.
45. Ipek S. Prevalence of metabolic syndrome in type 2 diabetic patients. J Turgut Ozal Med Cent. 2008;15(1):29–33.
46. Parildar H, Cigerli O, Demirag NG. Depression, coping strategies, glycemic control and patient compliance in type 2 diabetic patients in an endocrine outpatient clinic. Pakistan J Med Sci [Internet]. 2014;31(1):19–24 Available from: http://pjms.com.pk/index.php/pjms/article/view/6011.
47. Wang X, Hardin HK, Zhou L, Fang L, Shi P, Robinson KM. Implementation and evaluation of the chronic-disease self-management program among Chinese immigrant older adults in the US. Geriatr Nurs (Minneap) [Internet]. 2014;35(6):448–450. Available from: https://doi.org/10.1016/j.gerinurse.2014.07.002
48. Nini Shuhaida MH, Siti Suhaila MY, Azidah KA, Norhayati NM, Nani D, Juliawati M. Depression, anxiety, stress and sociodemographic factors for poor glycaemic control in patients with type II diabetes. J Taibah Univ Med Sci. 2019;14(3):268–76.
49. Kaur G, Tee GH, Ariaratnam S, Krishnapillai AS, China K. Depression, anxiety and stress symptoms among diabetics in Malaysia: a cross sectional study in an urban primary care setting. BMC Fam Pract [Internet]. 2013;14(1):1. Available from: BMC Family Practice.
50. McCoy MA, Theeke LA. A systematic review of the relationships among psychosocial factors and coping in adults with type 2 diabetes mellitus. Int J Nurs Sci [Internet]. 2019;6(4):468–477. Available from: https://doi.org/10.1016/j.ijnss.2019.09.003.
51. Rane K, Wajngot A, Wändell PE, Gåfvels C. Psychosocial problems in patients with newly diagnosed diabetes: number and characteristics. Diabetes Res Clin Pract. 2011;93(3):371–8.
52. Yorulmaz H, Tatar A, Saltukoğlu G, Soylu G. Analysis of the factors influencing illness perception in patients with diabetes. J Humanit Soc Sci. 2013;2:367–87.
53. Clarke DM, Currie KC. Depression, anxiety and their relationship with chronic diseases: a review of the epidemiology, risk and treatment evidence. Med J Aust. 2009;190(7):1326–77.

International Journal of Diabetes in Developing Countries (2021) 41:172–174
https://doi.org/10.1007/s13410-020-00856-x

CASE REPORT

Concomitant diabetic ketocacidosis and renal tubular acidosis in a type 1 diabetes mellitus patient

Khushboo Agarwal[1] · Dhruv Thakur[1] · Alok Gupta[1]

Received: 27 April 2020 / Accepted: 29 July 2020 / Published online: 11 August 2020

Abstract

Background Both RTA and T1DM have been associated with autoimmunity but they have been reported together rarely.

Case description A 23-year-old female presented with DKA persisted to have acidosis after normalization of sugars. She was diagnosed with concurrent RTA on urine analysis without any underlying cause. The simultaneous occurrence of these two conditions is challenging because of the need for bicarbonate therapy in RTA; and its relative contraindication in DKA. Management of hypokalemia also becomes challenging in this condition.

Conclusion Both RTA and DKA have a similar clinical presentation and, therefore, require a higher degree of clinical suspicion, especially in cases of non-resolving acidosis.

Keywords Type 1 diabetes · Diabetic ketoacidosis · Renal tubular acidosis · Treatment dilemma

Introduction

Type 1 diabetes mellitus frequently presents with diabetic ketoacidosis (DKA), a potentially life-threatening condition whose incidence remains high even in countries with a highly developed medical care system [1]. Our case represents concurrent diabetic ketoacidosis with distal renal tubular acidosis (RTA) in a patient of type 1 diabetes mellitus (T1DM) as its first presentation. We want to highlight the treatment dilemma posed by the simultaneous occurrence of these two conditions and the need to screen for RTA in non-resolving acidosis.

Case report

A 23-year-old female was presented to Emergency with a history of decreased appetite and abdominal pain for 2 days. There was no history of diarrhea, blood in the stool, or burning micturition. She had been lethargic for 3 days with a low-grade fever. The patient did not have a history of any kidney disease like nephrocalcinosis or renal osteodystrophy. There was no family or gestational history of diabetes and the patient was not on any known medication. On examination, she was dehydrated, lethargic with a Glasgow Coma Scale of 13 (E3V4M6). Her pupils were sluggishly reacting, plantar bilateral flexor, and there was no neck rigidity. There were no lateralizing neurological signs and the rest of the physical examination was normal. Her BP was 100/70 mmHg and she had a heart rate of 142 beats/min. She was hyperventilating with a respiratory rate of 35 and SpO_2 100%.

Dipstick blood glucose gave a reading as HI (measurable up to 600 mg/dL) and blood ketones of 7.2. The patient was admitted to ICU with a diagnosis of DKA and the blood investigations were done (Table 1). ECG and chest X-ray were normal. Initial ABG revealed metabolic acidosis with hypokalemia. She was treated with 0.9% saline and potassium initially followed by IV insulin infusion 0.1 U/kg/h with concurrent potassium. During the treatment, her blood sugar normalized and hydration improved but the patient continued to be drowsy. CT scan did not reveal any edema. The patient's consciousness improved after 3 days but the acid-base imbalance with low potassium persisted (Table 2). So, a cause for acidosis other than DKA was sought. Urine analysis revealed high PH with high urine anion gap (26 mEq/L) and low potassium (Table 3).

The diagnosis of distal RTA was made in view of the normal serum anion gap. As the patient became conscious and

✉ Khushboo Agarwal
khushboo.agrwl@gmail.com

[1] Department of Medicine, Dr. SN Medical College, Jodhpur, India

Table 1 Blood investigations

Investigation	Day 1	Day 6
Hemoglobin (g/dL)	14.6	13.6
Total leucocyte count (/μL)	13,500	5950
Differential leucocyte count	N73L20.8M5.5	N84L6.6M7.6
Platelet (/μL)	196,000	140,000
Blood sugar (mg/dL)	696	123
Blood urea (mg/dL)	44	67
Serum Creatinine (mg/dL)	1.38	1.2
Total bilirubin (mg/dL)	0.39	0.57
Direct bilirubin (mg/dL)	0.18	0.25
SGOT (IU/L)	38	22
SGPT (IU/L)	32	18
Alkaline phosphatase (IU/L)	232	214
Total protein (g/dL)	6.9	7.44
Serum albumin (g/dL)	4.15	3.7
TSH (μIU/L)	2.92	
fT3 (pmol/L)	1.88	
fT4 (pmol/L)	16	

started taking food, she was shifted on subcutaneous insulin along with sodium bicarbonate and potassium replacement. USG revealed no kidney pathology such as stones or nephrocalcinosis and markers of autoimmunity were negative. Her thyroid hormones and renal function tests were normal. She was discharged on insulin along with oral bicarbonate and potassium supplements.

Discussion

Type 1 DM is a chronic autoimmune disease of young caused by the destruction of beta cells. Distal RTA—hereditary or autoimmune—is a syndrome of systemic normal anion gap hyperchloremic acidosis with alkaline urine pH, hypokalemia, hypocitraturia, and hypercalciuria due to reduced secretion of H+ ions by the cells of the collecting tubules [2].

Table 3 Urine investigations

	Day 3	Day 6
pH	8	7
Sodium (mmol/L)	70	
Potassium (mmol/L)	18	
Calcium (mg/dL)	8	
Chloride (mmol/L)	62	
Phosphorous (mg/dL)	3.22	
Glucose	++++	Nil
ketones	++++	Nil
Albumin	+	+

Both RTA and T1DM have been associated with autoimmunity but they have been reported together only in a few case reports [3]. Whether this coexistence is purely coincidental or is because of common autoimmune pathogenesis is unclear. In addition to being reported together in association with autoimmune diseases such as Sjogren syndrome [4] or in association with a renal pathology [5], additional cases have been reported without discernible cause [3, 6].

The primary etiology of acidemia in patients with DKA is ketoacidosis, with the contribution of lactic acidosis and renal dysfunction. In most countries, the treatment of DKA follows standard protocols of intravenous fluids and insulin infusion. After metabolism during the recovery phase, bicarbonate is regenerated and aids the restoration of acidosis. The mean time to achieve pH > 7.3 and bicarbonate > 15 is 10–14 h [7]. Rosenbloom [8] defines persistent acidosis as bicarbonate < 10 mmol/L after 8–10 h treatment. It can be due to inadequate insulin dose or renal impairment secondary to severe dehydration.

Our patient had persistent acidosis for more than 5 days, even after normalization of blood glucose. Our patient had a normal renal function and the insulin preparation and dose (0.1 U/kg/h) were appropriate. Infections were also ruled out. Persistent acidosis with alkaline urine and hypokalemia and normal anion gap were consistent with type 1 renal tubular acidosis. The clinical spectrum of distal RTA in T1DM is

Table 2 ABG analysis of the patient

Investigation	Day 1	Day 2	Day 3	Day 4	Day 5	Day 6
pH	6.9	7.252	7.255	7.267	7.325	7.336
Sodium (mmol/L)	158.8	159.7	155.7	155.1	150.1	2.43
Potassium (mmol/L)	2.28	1.97	1.88	2.88	2.6	2.43
Bicarbonate (mmol/L)	3.5	10.3	9.3	11.1	10.9	9.4
Anion gap (mmol/L)	8.4	5.3	17.5	1.9	11.8	13
Ionic calcium (mmol/L)	0.655	0.717	0.806	0.965	1.012	1.157
Chloride (mmol/L)	149.1	146.1	127.3	145.9	131.0	130.7

similar to that of diabetic ketoacidosis complicating T1DM, except for urine pH > 5.5 in the former and presence of ketonuria in the latter. Concurrent occurrence of both ketoacidosis and renal tubular acidosis is challenging for the physician as the treatment is contrary in both. Bicarbonate therapy is associated with poor prognosis and cerebral edema in children in diabetic ketoacidosis treatment [9], whereas bicarbonate is essential for the management of hypokalemia in distal RTA. It is complicated by the fact that without correction of hypokalemia, the use of insulin is contraindicated in diabetic ketoacidosis. We did not initiate bicarbonate treatment until she had regained consciousness.

The further management of such a patient also is important. They should be managed on a nutritionally balanced diet with a greater proportion of base-producing components like fruits and vegetables. They also require lifelong potassium and alkali replacement at the rate of 1 to 2 mmol/kg per 24 h [10]. Strict correction of the hypercalciuria and appropriate supplementation of calcium and vitamin D is required to prevent osteomalacia and nephrocalcinosis. These patients are more prone to end-stage renal disease than either RTA or T1DM alone, making adequate management with proper doses of insulin vital.

Conclusion

This case describes the challenges in managing two seemingly contrary conditions with similar symptoms together. It also emphasizes the importance to document the presence or absence of ketones in diabetic patients presenting with acidosis, especially if persisting despite aggressive treatment. There is a need to explore the possible mechanisms involved in distal RTA in T1DM and the plan treatment guidelines for these patients.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval This article does not contain any studies with animals performed by any of the authors.

Informed consent Informed consent was obtained from the patient for publication of the case report.

References

1. Fritsch M, Rosenbauer J, Schober E, Neu A, Placzek K, Holl R. Predictors of diabetic ketoacidosis in children and adolescents with type 1. Experience from a large multicenter database. Pediatr Diabetes. 2011;12:307–12.
2. Hess B. Acid–base metabolism: implications for kidney stones formation. Urol Res. 2006;34(2):134–8.
3. Dymot JA, McKay GA. Type 1 (distal) renal tubular acidosis in a patient with type 1 diabetes mellitus–not all cases of metabolic acidosis in type 1 diabetes mellitus are due to diabetic ketoacidosis. Diabet Med. 2008;25:114–5.
4. Raddatz V, Alvo M, Durruty P, Orellana L, Garcia de los Rios M. Decompensated diabetes mellitus and hyperchloremic metabolic acidosis: a case with both pathologies. Rev Med Chil. 1998;126:1224–8.
5. Garg M, Kant R. Type 1diabetes Mellitus presenting as distal renal tubular acidosis (RTA type 1). J Diab Endo Assoc Nepal. 2019;3(2):49–52.
6. Abdul-Rasoul M, AlTerkait A, Merza J. Persistent acidosis at presentation in a patient with type 1 diabetes: concomitant diagnosis of type 1 distal renal tubular acidosis. J Diabetes Mellitus. 2012;2:258–60.
7. Chua H, Schneider A, Belloma R. Bicarbonate in diabetic ketoacidosis—a systemic review. Ann Intensive Care. 2011;1:23–35. https://doi.org/10.1186/2110-5820-1-23.
8. Rosenbloom A. The management of diabetic ketoacidosis in children. Diabetes Ther. 2010;2010:103–20.
9. Hekkala A, Reunanen A, Koski M, Knipp M, Veijola R. Age-related differences in the frequency of ketoacidosis at diagnosis of type 1 diabetes in children and adolescents. Diabetes Care. 2010;33:1500–2. https://doi.org/10.2337/dc09-2344.
10. Soriano JR. Renal tubular acidosis: the clinical entity. JASN. 2002;13(8):2160–70.

International Journal of Diabetes in Developing Countries (2021) 41:175–176
https://doi.org/10.1007/s13410-020-00895-4

LETTER TO THE EDITOR

Missing the wood for the trees: cardiocentricity in current diabetes guidelines

Sanjay Kalra[1] · Banshi Saboo[2] · Nagendra Kumar Singh[3] · Sarita Bajaj[4]

Received: 21 June 2020 / Accepted: 7 November 2020 / Published online: 17 November 2020

Diabetes is a complex syndrome, and guidelines are expected to simplify the process of clinical decision making. The consensus by the American Diabetes Association (ADA and the European Association for the Study of Diabetes (EASD [1]) stratifies persons with type 2 diabetes based on established atherosclerotic cardiovascular disease (ASCVD)/chronic kidney disease (CKD) and supports use of glucagon-like peptide 1 receptor agonists (GLP1RA) and sodium glucose cotransporter 2 inhibitors (SGLT2i). Better-established drug classes, supported by experience and evidence [2], have been relegated to third-line status.

This consensus, by focusing on vascular disease as the main determinant of glucose-lowering medication, has missed the wood for the trees. Diabetes is equivalent to a dense tropical jungle, full of heterogenous flora and fauna. Obstacles encountered in such an environment necessitate individualization of coping strategies at every bend and corner. This is absolutely true for diabetes care as well.

The term "established ASCVD" is difficult to understand. Many people with diabetes have subclinical ASCVD [3]. Where these individuals will fit in the current algorithm is uncertain. A similar dilemma occurs in atypical CVD, or anginal equivalent symptoms. Single-minded emphasis on established ASCVD, without acknowledging subclinical and atypical CVD, is as limited in relevance as the glucocentricity of earlier guidelines.

CKD offers similar challenges for the practicing diabetes care provider. eGFR-based triage does make pharmacodynamic sense, but deflects attention from other aspects of renal health, such as albuminuria.

Current guidance relegates adverse events and patient satisfaction to the sidelines. Adverse events that occur with GLP1RA and SGLT2i are not insignificant, and should be considered before issuing blanket recommendation for their use.

Persons with symptomatic severe hyperglycemia, or those who are "sick," are not right candidates for GLP1RA and SGLT2i. For example, how does one classify a person with recent acute coronary syndrome, heart failure, stroke, or acute kidney injury? Should preexisting GLP1RA or SGLT2i be continued during the acute illness? What time lag should be allowed to lapse before prescribing GLP1RA or SGLT2i [4, 5]?

Medicine is built upon physiology and biochemistry. The consensus makes no mention of the pathophysiologic basis of diabetes, or the need to assess insulin resistance or secretory defect while treating the syndrome.

Ideal management must take the entire glucophenotype of the individual into account. The current consensus may be used, with a caveat: it does not apply to severe hyperglycemia, symptomatic diabetes, sick persons (with acute comorbidity special situations such as pregnancy, hepatic, renal impairment), and susceptibility to side effects (e.g., gastrointestinal discomfort with GLP1RA; genital infections with SGLT2i) (Table 1).

The "glycemic personality" model responds to the mnemonic ASSURE (Associated complications and comorbid

Table 1 Caveats for the applicability of current ADA/EASD consensus

Does not apply to
• Severe hyperglycemia
• Symptomatic diabetes
• "Sick" diabetes
• Special situations
• Special situations
Downplays, for certain drug classes
• Contraindications
• Potential adverse effects

✉ Sanjay Kalra
brideknl@gmail.com

[1] Department of Endocrinology, Bharti Hospital, Karnal, India

[2] Department of Diabetology, DiaCare Hospitals, Ahmedabad, India

[3] Department of Diabetology, Diabetes and Heart Research Centre, Dhanbad, India

[4] Department of Medicine, MLN Medical College, Allahabad, India

Table 2 The glycemic personality: ASSURE

A Associated complications and comorbid conditions
S Sensitivity and social realities
S Severity and style of hyperglycemia
U Utility and urgency of glucose lowering
R Relative risk of hypoglycemia
E Expected efficacy of various glucose lowering therapies

conditions, Sensitivity and social reality, Severity and urgency of glucose lowering (in both short and long term), Relative risk of hypoglycemia, and Expected efficacy of various glucose-lowering therapies should be considered while planning treatment for diabetes) (Table 2).

Inclusion of the caveats and considerations mentioned above will help improve the utility of the ADA/EASD guidance.

References

1. Davies MJ, D'Alessio DA, Fradkin J, Kernan WN, Mathieu C, Mingrone G, et al. A consensus report by the American Diabetes Association (ADA) and the European Association for the Study of Diabetes (EASD). Diabetologia. 2018;2018:2461–98. https://doi.org/10.1007/s00125-018-4729-5.
2. Kalra S, Aamir AH, Raza A, Das AK, Azad Khan AK, Shrestha D, et al. Place of sulfonylureas in the management of type 2 diabetes mellitus in South Asia: a consensus statement. Indian J Endocrinol Metab. 2015;19(5):577–96.
3. Volgman AS, Palaniappan LS, Aggarwal NT, Gupta M, Khandelwal A, Krishnan AV, et al. Atherosclerotic cardiovascular disease in South Asians in the United States: epidemiology, risk factors, and treatments: a scientific statement from the American Heart Association. Circulation. 2018;138(1):e1–34.
4. Diabetes in special situations: glucagon-like peptide-1 receptor agonist use in acute myocardial infarction. Available at: https://diabetes.medicinematters.com/en-GB/cardiovasculardisorders/glp-1-agonists/diabetes-in-special-situations-glucagon-like-peptide-1-receptor/15153012?searchResult=10.Kalra&searchBackButton=true. Last accessed on 7 June 2020.
5. Diabetes in special situations: sodium-glucose co-transporter-2 inhibitors in acute coronary syndrome. Available at: https://diabetes.medicinematters.com/en-GB/cardiovasculardisorders/sglt2-inhibitors/diabetes-in-special-situations-sodium-glucose-co-transporter2/15199584?searchResult=9.Kalra&searchBackButton=true. Last accessed on 7 June 2020.

VISION STATEMENT

To be recognized as a global leader for clinical care, education, training, research, advocacy and capacity building in the field of diabetes.

MISSION STATEMENT

1. Promotion of excellence in diabetes care to make India the Diabetes Care Capital
2. Empowerment of persons living with diabetes
3. Support for diabetes research
4. Dissemination of information and knowledge in diabetes care
5. Advocacy for the cause of diabetology

NEW EXECUTIVE COMMITTEE AND OFFICE BEARERS 2021

Patrons of RSSDI

Dr. H.B. Chandalia, Mumbai
Dr. C. Munichhoodappa, Bengaluru
Dr. Ashok K. Das, Puducherry
Dr. Binode K. Sahay, Hyderabad
Dr. O.P. Gupta, Ahmedabad
Dr. V. Seshiah, Chennai
Dr. P.V Rao, Hyderabad
Dr. Jitendra Singh, New Delhi
Dr. V Mohan, Chennai
Dr. Vinod Kumar, New Delhi

President

Dr. Banshi Saboo, Ahmedabad

President Elect

Dr. Ch.Vasanth Kumar, Hyderabad

Immediate Past President

Dr. Rajeev Chawla, New Delhi

Secretary

Dr. Sanjay Agarwal, Pune

Vice-President

Dr. Anuj Maheshwari, Lucknow

Vice-President

Dr. Vijay Viswanathan, Chennai

Joint Secretary

Dr. Sujoy Ghosh, Kolkata

Treasurer

Dr. Sunil Gupta, Nagpur

Executive Committee

Dr. C R Anand Moses, Chennai
Dr. Sudhir Bhandari, Jaipur
Dr. J K Sharma, New Delhi
Dr. Bikash Bhattacharjee, Guwahati
Dr. Pratap Jethwani, Rajkot
Dr. L. Sreenivasa Murthy, Bengaluru
Dr. Sanjay Reddy, Bangaluru
Dr. Shalini Jaggi, New Delhi

Co-opted

Dr. Vijay Panikar, Mumbai
Dr. Brij Makkar, New Delhi
Dr. Rakesh Sahay, Hyderabad

TRAINEE GRANTS (Up to 10 grants)

Research Grants upto INR 200000 to support outstanding thesis/ research work by first year MD/DNB/ PHD students/Research fellows from India.

Eligibility Criteria
All Postgraduates in First year MD, DM /DNB from any of the institutions in the country are eligible to apply

How to apply?
Send in your Research proposals by email to the RSSDI Secy/ Chairman research committee by email/ apply directly on web site.

Research proposal should have following proofs-

1. A supporting letter from your guide/ head of department stating that this is a bonafide project for your thesis and also mentioning the dates of you joining the program and expected date of graduation. The guide must also state that he/she will stand guarantee for the work done
2. A detailed budget
3. Thesis proposal approved by the department/appropriate institutional authority
4. Approval by the ethics committee

Selection Process
Proposals will be reviewed by the research committee of the RSSDI.

Disbursement of Grant
A minimum of 50% of the grant amount will be disbursed initially. Further disbursement will be done annually based on submission of progress reports on the work done and utilisation of sanctioned amount. These reports must be filed to the secretary of the RSSDI

Responsibility:
All grant awardees are expected to present their work at RSSDI Annual Conference during research presentation's session. Failure to file progress reports annually and when requested by the RSSDI and failure to present progress at RSSDI Annual conf may result in the forfeiture of the grant.

All awardees are expected to follow the tenets of responsible and ethical conduct of research. Unethical or fraudulent use of RSSDI research funds will warrant adverse action from the society including

forfeiture of grant, black listing in the society's databases and other legal recourses that are available to the society.

Publication
The RSSDI expects that the grant source be acknowledged in all publications and submissions made with regards to the research done with the grant.

All awardees are encouraged to submit their work to the RSDDI Journal IJDDC

CALL for RESEARCH PROPOSALS for GRANTS (up to 5 lacs)

Research proposals are invited from Indian scientists, who are members of RSSDI interested in conducting research in the field of Diabetes, Endocrinology& Metabolism, for funding by RSSDI

The proposals may of clinical or translational research importance. A maximum grant amount of INR 5 Lakhs will be sanctioned. All grants will be reviewed by the research committee.

The detailed proposals should include the following:
Title, names of principal and co investigators, summary, introduction/ background, review of literature, aims, methodology, study design and detailed plan of work & bibliography.

Brief biodata of principal investigator and other co-investigators.

Importance of work
Detailed Budget sought along with full justification/ proposed utilization, of funding sought from RSSDI

Whether the project is being partly funded from any other source? If yes, please mention the source and the amount received.

Ethics Committee clearance of the Institution or other bonafide body.

How to apply
All applications should be addressed to:
1. The Secretary , RSSDI
2. Soft copy of the research proposal should be sent to Secretary, RSSDI

When to apply
Proposals will be accepted Twice a year. Once between 1st Jan - 31st April & then July 1st to 30th Nov.

All research proposals will be reviewed by Research committee over a period of 4-6 weeks & approved proposals will be provided Research Grant after fulfilling all documentation by 30th June & then 31st December of each year.

MAJOR RESEARCH GRANT PROPOSALS- usually not more than one at a given time.

Above 10 Lacs upto a total amount of 50 Lacs will be Granted to RSSDI initiated, owned, multi-centric, clinical or translational research, having long term application of scientific and clinical findings, which can translate into strategies for improving health-care delivery, patient outcomes, and community health in India.

Such research proposals will be carried out in only centres with research capabilities across India.

TRAVEL GRANTS FOR YOUNG DIABETES RESEARCHERS TO ATTEND INTERNATIONAL CONFERENCES

Criteria for the travel grant are as follows:

- Applicant should apply 2 months in advance.
- Travel Grant is open only to the RSSDI members.
- Applicant should submit Oral paper / Poster acceptance document to RSSDI Secretariat.
- Applicant should submit Declaration that he/she has not receiving grant from any other agency / Organization – In case of receiving grant from any other Organization, RSSDI shall pay only the exceeding amount not covered by that agency.

ADVANCED CERTIFICATE COURSE IN DIABETOLOGY

(IN ASSOCIATION WITH JAIPUR NATIONAL UNIVERSITY)

Research Society for the Study of Diabetes in India (RSSDI) was founded by Prof. M.M.S. Ahuja in 1972. RSSDI is the largest body of professional doctors and researchers in Asia, working in the area of Diabetes & is the National Body recognized by IDF (International Diabetes Federation). One of the key areas of focus is to train doctors at all levels to better manage Diabetes and its complications. RSSDI recognizes this problem and runs a well-structured, full time, residential "Advanced Certificate Course in Diabetology". This two-year course is like any other post graduate course and has immensely helped doctors to practice better diabetes care. RSSDI has

List of RSSDI Accredited Centres

Sl. No	Institute Name	Institute Location
1.	Diacon Hospital	Bangalore, Karnataka
2.	North Delhi Diabetes Centre	New Delhi, Delhi
3.	Prithvi Hospital	Tumkur, Karnataka
4.	Total Diabetes Hormone Institute	Indore, Madhya Pradesh
5.	Dia Care - A Complete Diabetes Care Centre	Ahemdabad, Gujarat
6.	Sonal Diabetes Hospital	Surat, Gujarat
7.	Jothydev's Diabetes and Research Center	Trivandrum, Kerala
8.	Advanced Endocrine & Diabetes Hospital	Hyderabad, Telangana
9.	Sunil's Diabetes Care N' Research Centre	Nagpur, Maharashtra
10.	Marwari Hospital and Research Centre	Guwahati, Assam
11.	Down Town Hospital	Guwahati, Assam
12.	St.Theresa's Hospital	Hyderabad, Telangana
13.	Aegle Clinic	Pune, Maharashtra
14.	Lilavati Hospital & Research Centre	Bandra West, Mumbai
15.	Srajan Hospital	Udaipur, Rajasthan
16.	Endeavour Clinics & Dr. Sambit's Centre of Diabetes and Endocrinology	Bhubaneswar, Odisha
17.	ILS Hospital, Salt Lake	Salt Lake City, Kolkata
18.	Belle Vue Clinic	Dr. U N Brahmacahri Sreet, Kolkata
19.	Arthur Asirvatham Hospital	Mdurai, Tamil Nadu
20.	M V Hospital for Diabetes	Chennai, Tamilnadu
21.	Sarvodaya Hospital and Research Centre	Faridabad, Uttar Pradesh
22.	GALAXY SPECIALITY CENTRE	Sodala, Jaipur

carefully looked into all aspects of this course & has accredited & recognized 17 centres across India at present and more centers are being inspected for accreditation. National Faculties and experts of RSSDI chosen from Academia visit these centers from time to time to ensure high standards. Now this Advanced Certificate Course has Dual Accreditation from RSSDI and Jaipur National University.

COURSE DETAILS

Name of the Course: Advanced Certificate Course in Diabetology

Duration: 2 Years – Post MBBS & 1 Year - Post MD / DNB (Gen - Medicine)* (Full Time) Educational.

Qualification: A candidate must possess MBBS degree from ANY of the recognized university approved by Medical Council of India (*The duration of the course is 1 Year for those with MD/ DNB in Internal Medicine. Candidates having MD degree in other specialties will have to do the course over 2 Years).

Number of seats: 2 seats per year for every eligible teacher as per rules of Medical Council of India (MCI).

Selection of Candidates: Selection for the Certificate course is through a performance evaluation by Theory test for 90 marks (90 minutes duration) which is conducted at all accredited centres. The result is displayed WITHIN 3 days on the Web site of JNU and RSSDI. Post MD (Internal Medicine) will be given !

COURSE FEES:

- Rs 30000/- (for post MD/DNB (internal medicine), 1 year program)
- Rs. 50000/- (for post MBBS, MD in other branches, 2 years program)

Session: Two sessions are run annually, in January and in July. Prospectus details are available on the RSSDI website. All applications must be sent to Jaipur National University.

Dates for Admission in ACCD 2021

1) Last date of submission of Application Form - 20th February 2021
2) Screening Interview - 27th February 2021
3) Declaration of Exam Result - 3rd March 2021
4) Last date of payment of course fee - 8th March 2021
5) Commencement of course - 10th March 2021

ANNOUNCEMENTS

Dear Member,

Please update your Membership details like Complete Postal Address, Email Id, Pan No. & Mobile no. after log in your membership area on our website www.rssdi.in under sub heading Membership corner, so that we can send you RSSDI Newsletter & Journals.

49th Annual Conference of RSSDI-RSSDI 2021

Date: 11th - 14th November 2021

Venue: Mumbai

Acknowledgement

We are grateful to the following reviewers for reviewing articles for International Journal in Diabetes in Developing Countries for the year 2020.

A AR
Abdelaziz, Phd H
Abdelnasser Z
Abdulrazzaq DA
Abed A
Acharya L
Adisa R
Adriana R
Agarwal K
Agarwal N
Agoramoorthy G
Agrawal T
Agrawal S
Agrawal A
Ahenkorah Fondjo L
Ahmed C
ahmed Mphil d
AJEET J
Akram M
Aktas G
Alam, DM S
Alare k
Ali W
Ali, PhD S
Al-Lami F
Al-Nimer M
Al-Sayyed PhD, H
Amr N
AR K
Arora R
Arora V
Aslam M
Atwa H
Ayatollahi H
Ayatollahi H
Ayyagari U
Ayyagari M
Bachani D
Baid L
Bajaj S
Balachandran K
Balbaa M
Baloch Z
Barzilay J.
Bashir A
Behl A
Bekele BB
Bello A
Bhadada SK
Bhat S
Bhati P
Bhattacharjee R
Bhattacharya S
Bhattacharya C
BHATTACHARYA G
BHAVANI N
Brink S
Censi, MD S
Chakravarthy DK
Challa S
Chaudhuri A
Chawla M
Chawla R
Chiefari E
Chowdhury S
Chutia H
Coppell K
D A
D.S P
Dabas A
Daradka PhD H
Das S
Dayal D
desai Kv
Devaraj NK
Dhanunjaya Y
Dolatkhah N
Dorairaj P
Dudekula AB
Dutta D
Eshghinia S
Ezenwaka C
Fasanmade MBBS, FWACP O
Fekry A
Fernandes, PhD GW
G Seshadri, MD K
Gaddam A
Ganesan M
George L
Gharesouran J
Ghosh A
Ghosh A
GHOSH U
Girei, MD B
Gnana Sam K
GODI S
Gokulakrishnan K
Golboni F
Golboni F
Golla NK LG
gopal j
GOPALAKRISHNAN VK
Gossain V
Govil A
Gowda V
Gowda, MD G
Goyal G
Goyal V
Gupta A
GUPTA S
Gupta S
Gupta S
Gupta V
Gupta, MD, DM Y
Harati-Sadegh M
Harish R
Herath M
Hian TK
Hilawe EH
Hu D
Huri HZ
HUSSAIN T
Ichhpujani P
Jagadeesha A
Jaggi S
Jain N
Jethwani P
Jha S
Joshi A
juttada u
K Darwish R
K.M r
Kadam DB.
Kale M
Kandi CM
Kannappan S
Kant S
Kapoor N
Kapur S
Kar S
karmakar S
Karmakar PhD, PDF S
Karoli R
Kasnavi Yazdi h
Kaul N
Kesavadev J
Khan AM

Kır Biçer E
krishna s
krishna s
Krishnan K
Kucukardalı Y
Kulshreshtha B
Kumar V
kumpatla S
L R
L S
Lakshminarayanan S
Laway B
M N
M R
M G R
Madhu K
Maheshwari S
Maheshwari, MD, FICP, FIACM, FACP, FACE A
Mahilmaran A
Maisnam I
Majaliwa E
MALIK RA.
Manivel, MD Physiology R
Masuzaki R
Mateti UV
MATHUR G
Mathur S
Mehendale, PhD N
Mehra NK.
MERT M
Meseri R
Mishra B
Mishra, PhD M
Mittal V
Mittal G
Mittal P
Mohammadifard N
Mohan V
Mohtasham-Amiri Z
Mongkolsomlit S
Monroy A
Mukherjee S
MUNICHOODAPPA CS
Nachimuthu S
Naik D
Nair M
Narayan, MD, DM P
Narouei-nejad M
Nayak M
Nayak S
Obirikorang C
Olamoyegun M
Ozgu-Erdinc A.S
P R A
Pandey H
Pandya H
Panicker S
Pankaj P
Pant V
Papanas N
Pasupula D
PATEL M
Patil M
Patil A
Paul B
Paul S
Pendsey S
Prabhu S
prakash s
R C
Raina RS
Raina S
Raizada N
raj v
Rajagopal, MD J
Rajalakshmi S
Rajsekar S
Ram U
RAMACHANDRAN S
Ramchandani G D
Rao R
Rao, PhD PV
Rastogi A
Rathod MD N
RaviKumar .
Reang T
RJ S
ROTTE A
Rouhani, PhD M
Roy B
Saboo B
salahuddin a
Saluja M
salwa h
Sankaranarayanan, MD, DM Dr G
Santos C
Saraf A
SARAN S
Saraswathy R
Sarna M
Saxena S
Selvan C
Selvaraj B
Serban M-C
Seshiah V.
Sharma A
Sharma A
Sharma BB
Sharma P
SHARMA N
Sharma, MD, DM D
Shenoy MD, M
Shetty S
Shriraam V
siddiqi, PhD HS
Silver B
Singh A
Singh A
Singh N.K.
Singh S.K.
SINGH S
Singhal N
Singla M
SINHA A
sinha Dr.N
Śliwińska-Mossoń M
SN N
Snabboon T
Srivastava A
SRIVASTAVA S
Subramanian B
Sudhir P
Sulania A
Surendran S
Suri J
swelam PhD M
Swem F
Tak S
talla v
talla v
Tarantino G
Thomas SC
Thoomati, M.S S
Tiwaskar M
Umapathy D
Upadhyay D
upendra s
Uppal B
V A
Vahdat Shariatpanahi, Ph.D Z
Valliyot Dr. B
venkataiah v
Venkateswarlu K
Verma K
Vikram N
Virmani MD DNB A
VP S
Wadhwa AP
Waisundara V
Wang Y
Werida R
Yadav S
Yang Ph.D m
Yurtcu, PhD E
Zhang W

Registered with Registrar of News Papers, India vide no. RNI-43019/85

International Journal of Diabetes in Developing Countries

Volume 41 | Issue 1 | January – March 2021

Springer