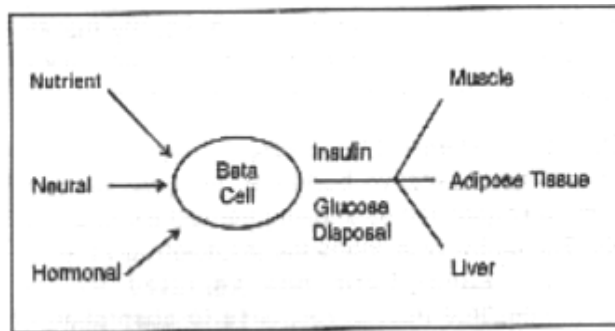


INSULIN KINETICS

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Insulin was first isolated from pancreatic tissue by Banting and Best in 1921. Insulin is the major hormonal regulator of glucose metabolism. The present article summarizes the information available about the secretion, release and its action on target tissues.



INSULIN SECRETION:

Insulin is released by the process of exocytosis (emicytosis). The stored granules move along the network of microtubules and microfilaments pathway from the golgi complex to the plasma membrane. The microtubules provide a mechanical structure along which the secretory granules move in an oriented fashion. The actin and myosin, the contractile proteins play a role in the secretory process. Actin, is a constituent of the microfilaments. This actin is said to be involved in the release of insulin. The actin and myosin act together to transport granules; the micro tubules acting as network, propelled by the interaction of actin and myosin. The daily insulin secretion is 30-40 units out of which 50% is secreted in the basal state and the rest with each meal.

Secretagogues:

A. Neural:

Pancreatic islets receive rich innervation of autonomic nerve supply. The cholinergic neurotransmitters that stimulate insulin secretion are acetylcholine, VIP and GRP. Sympathetic neurotransmitters that inhibit insulin secretion include nonadrenaline, galanin and neuropeptide Y (NPY).

Activation of parasympathetic nerves:

The autonomic innervation of islets plays an important role in the modulation of insulin release from the islet cells of Langerhans. Insulin is secreted at the smell, sight and expectation of food. This is known as the cephalic phase of insulin secretion and is due hypothalamo-entero-insular axis which is mediated by vagal nerve[1]. During feeding the parasympathetic nerves are activated and are instrumental for the cephalic phase of insulin release. The cephalic phase of insulin secretion minimizes the early rise in postprandial blood sugar[2,3]. Vagotomy and pancreatic transplantation (Islet denervation) results in early rise in postprandial blood sugar. Sympathetic activity partly mediates inhibition of insulin secretion during stress and trauma.

B. Hormonal:

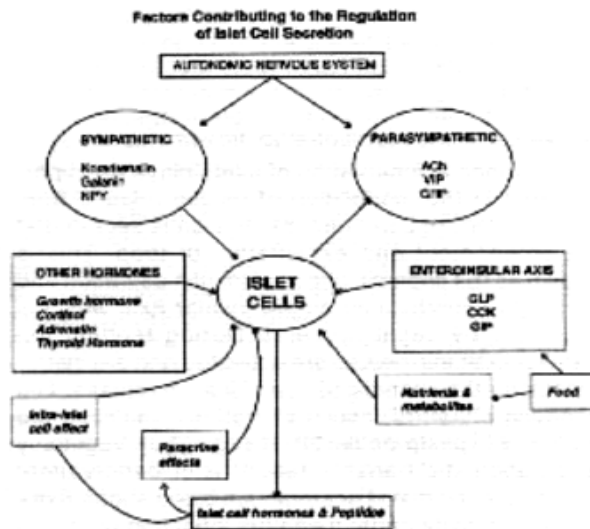
Many peptides modulate insulin release. They are co-localised in islet cells with their major secretory products. Insulin release is stimulated by TRH (Thyrotropin-releasing hormone), GHRH (Growth hormone releasing hormone), ACTH (Adrenocorticotrophic hormone) and opiodis. Insulin secretion is inhibited by many hormones including pancrestatin, islet amyloid polypeptide (IAPP), Diazepam-binding inhibitor (DBI) Peptide YY (PYY), Corticotrophin releasing factor (CRF), atrial natriuritic peptic (ANP) and biogenic amines [4,5,6,7].

Various gut peptides also affect insulin secretion and other hormones of the pancreas, namely the glucagon and somatostatin by local paracrine action. Gut hormones may mediate the 'incretin' effect. There is augmentation of insulin release following ingestion rather than parental administration, so-called enteroinsular axis. This is augmented to hormones, the gastric inhibitory polypeptide (GIP), and more recently glucagon like peptide-1 (GLP1 7-36 amide) which are shown to increase the nutrient mediated insulin secretion by increasing the intracellular cyclic AMP levels.

Thus the pathways involved in insulin biosynthesis and secretion have yielded much information about the basic biology of the pancreatic beta cell. This will form the important basis for further studies in the pathobiology or both IDDM and NIDDM.

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C. Nutrient:

Glucose availability is the major physiological determinant of insulin secretion. The principal action of nutrient secretagogues are mediated by the opening of voltage dependent calcium channels in the beta membrane. ('Voltage-gated' i.e. opened by depolarisation of the membrane). There are a number of other physiological and pharmacological agents which act as secretagogues. They are the initiators and the potentiators of insulin release. The initiators are called as primary stimulants as they can provoke the insulin release. The potentiators are agents which are ineffective by themselves but will increase the insulin release in response to glucose or amino acids. The initiators include the metabolites (glucose, arginine, lysine, ketone bodies and free fatty acids), and the drugs (sulphonylureas). The potentiators include metabolites (Cyclic AMP), neurotransmitters (Acetylcholine), and the gut peptides (glucagon, GLP17-36 amide, GIP, secretin).

Glucose stimulated insulin release is biphasic, with a rapid first phase lasting 5-10 min, followed by a prolonged second phase which persists for the duration of the high glucose stimulus. Concentrations of glucose less than 5 mmol/l does not affect the rate of release of insulin. The secretory rates occur with the extra cellular glucose concentration between 5.5 and 17 mmol/l with a half maximal stimulation at 8 mmol/l.

In the basal state ATP modulated K^+ channels remain open and voltage dependent Ca^{++} channel is closed. With the entry of nutrients (glucose) depolarization of the membrane occurs and the K^+ channel is closed and Ca^{++} releases insulin stored in

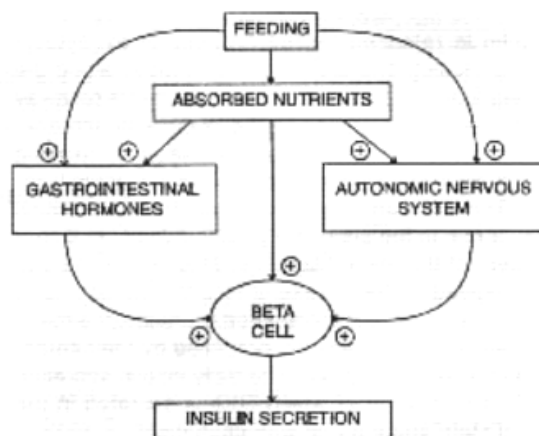
the cytosol. The released insulin enters the portal vein in seconds and half of it is cleared by the liver in the first pass.

ENTEROINSULAR AXIS AND INCRETINS:

The entero insular axis comprises of neural and hormonal pathways that are activated by feeding[8,9]. The enteroinsular axis enhances insulin secretion. The parasympathetic supply innervating the islets can be activated by sight, smell, taste and processing of food in alimentary tract and also by the absorbed nutrients from the alimentary canal. This is called as the "Cephalic phase" of insulin secretion[3].

The hormonal arm of the enteroinsular axis is composed of the putative hormone "Incretin" which is secreted during meals and augments the insulin levels. The pathway includes the insulinotropic (i.e. insulin-releasing) hormones secreted by the gastrointestinal tract in response to absorption of nutrients.

BETA CELL FUNCTION IN DIABETICS:



Enteroinsular axis – Neural & Hormonal pathways involved in insulin secretory response to feeding.

In IDDM patients there is total destruction of the beta cells. Hence they require insulin for survival. Replacement of insulin is the only treatment of choice. In NIDDM patients one of the pathophysiological events is the defective beta cell function. These abnormalities include absent first phase secretory response to intravenous glucose, delayed and blunted secretory response to mixed meals, increased concentrations of proinsulin and insulin breakdown intermediates and alterations in ultradian oscillations and rapid pulses of insulin secretion[10].

When glucose is infused at a constant rate in normal subjects, the plasma glucose concentrations do not demonstrate a constant glucose concentration. There are regular oscillations in the insulin secretion every 90-120 mts. Oscillations at this frequency are termed ultradian oscillations. The oscillations of insulin secretions lag temporarily behind the oscillation in glucose. In a normal subject when glucose is infused at a constant rate, the beta cells respond appropriately to the glucose stimulus and the periodicity of insulin secretory oscillations adjusts accordingly. The Phenomenon is termed *entertainment*. The insulin secretion from the normal beta cells can be completely entertained by the exogenous glucose. Alterations in the pattern of oscillatory insulin secretions or reduction in the ability of the insulin secretion to exogenous glucose load has been said to be seen in the patients with IGT, even when glucose tolerance test and glycosylated haemoglobin are normal. This defect is severe in patients with NIDDM and reductions in the insulin secretory response to glucose antedate the elevation is fasting plasma glucose.

INSULIN ACTION ON TARGET TISSUES:

Insulin is an anabolic and anticatabolic hormone that promotes glucose transport, glycogen synthesis, lipogenesis and protein synthesis (*anabolic*) and inhibits glycogenolysis, gluconeogenesis and lipogenesis (*anticatabolic*). The most important target organ of these metabolic events are liver, muscle and adipose tissues which have specific insulin receptors.

1. RECEPTOR EVENTS:

Insulin acts on the target cells by binding to its receptors. Insulin binds to its receptor at the extracellular surface in a specific, rapid, saturable and reversible manner. The insulin receptors are found on the insulin sensitive cells. The gene for the insulin receptor lies on chromosome 19. The insulin receptors have a structural homology with LDL, growth factor and oncogene receptor. The receptor is a glycoprotein which consists of two extra cellular alpha sub-units and two beta sub-units. The alpha sub-unit lies extracellularly and the beta sub-unit spans the cell membrane. The alpha and beta subunit are linked covalently. The alpha sub-unit is externally oriented and contains the insulin binding site, while the beta sub-unit being in the transmembrane site serves as the effector system. Insulin binds to the alpha subunit and activates the protein kinase activity present in the beta subunit. This activation process phosphorylates several

tyrosine residues on the receptor. These changes on the receptor triggers the intracellular events underlying insulin's metabolic effect. The receptor-insulin complex is then internalized by the surrounding membrane invaginating to form an endosome. Inside the cell the insulin is digested by protease and the receptors are recycled.

The insulin bound to the receptor increases with rising concentrations of free hormone and has a non-linear relationship. The non-linear response is characteristic of bimolecular dynamic interactions and are described from the two variables, *capacity* and *affinity* of the system. The total quantity of insulin capable of being bound by the preparation at infinite free insulin concentration is defined as *capacity*. From this the number of binding sites per cell can be elevated. The free insulin concentration required to produce half-maximal occupation of the receptor population by the hormone is defined as *affinity* of the binding reaction. The *insulin receptor number tunes with the changes in the circulating concentration of insulin*[11]. High ambient insulin concentrations reduce the insulin binding. This phenomenon is known as down-regulation and may be due to increased receptor internalization and turn over. Similarly the opposite process of *up-regulation* also occurs when the circulating insulin levels are low. Insulin-receptor complexes reduce the affinity of other insulin binding sites to insulin. This phenomenon is termed negative-co operativity.

CLINICAL IMPLICATIONS:

- A. *During acute fasting state the affinity for insulin receptor is increased, before an increase in number of receptor is observed.*
- B. *During chronic fasting or dieting there is a return of the number of insulin receptors towards normal level with a concomitant decrease in hyperinsulinemic state.*
- C. *There is an increase in insulin receptor affinity five hours after an oral glucose load.*
- D. *After an exercise there is an increased affinity of insulin for its receptor correlating with the fall in the plasma glucose level.*

2. POST RECEPTOR EVENTS:

Insulin lowers the cAMP concentrations by activating one or more of the membrane associated phosphodiesterase. The lowering of camp is an unique post-receptor action of insulin which

underlies a number of its anti catabolic actions, such as inhibition of gluconeogenesis and glycogenolysis in liver and inhibition of lipolysis in the adipose tissue[12]. The various other postulated mechanisms of insulin action are through the tyrosine kinase cascade, a "insulin second messenger", cGMP, small peptides and the most recently described Glycosylated Inositol Phosphate (GIP). GIP is thought to be released from the plasma membrane by an insulin stimulated phospholipase C enzyme.

Insulin increases the glucose uptake in certain tissues like the muscles and fat. Glucose influx across the membrane is catalyzed by a family of proteins called as "Glucose Transporters" (GLUT). Insulin not only increases but also recruits more of these transporters exposed on the cell surface. Insulin also modifies the intrinsic activity of these glucose transporters.

Insulin also activates several intracellular enzymes in the pathway of fatty acid and triglyceride metabolism leading onto leading onto lipogenesis. ***Insulin thus functions as an anabolic and anti-catabolic hormone.***

INSULIN RESISTANCE:

Insulin resistance is a state in which a normal amount of insulin produces a subnormal amount of insulin response. Defects in binding of insulin to its receptors due to reduction in their number of affinity would result in insulin resistance. Clinically insulin resistant is a state falls into two categories. 1) *Decreased Sensitivity* – Where normal response can be obtained with supra maximal insulin levels and 2) *Decreased responsiveness* – Even massive doses of insulin cannot bring about a normal level of response. These patients can present with diabetes or some of those patients can compensate for this defect with elevated serum levels and may have only impaired glucose tolerance and euglycaemia. At the molecular level, insulin resistance can occur a) the insulin receptor interaction site b) insulin receptor binding and kinase function and c) at the post receptor signaling pathway.

Insulin resistant states can be due to, a) genetic disorders (Type A Syndrome and its variants, leprechuanism, and lipoatrophic diabetes), b)

immune disorders (anti-insulin antibodies, and anti-insulin-receptor antibodies or the Type B syndrome) and c) endocrine and metabolic conditions (obesity), NIDDM, IDDM, physiological states like puberty, pregnancy, aging and endocrinopathies like cushings syndrome and acromegaly).

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