

Insulin – History, Biochemistry, Physiology and Pharmacology



Shashank R. Joshi*, Rakesh M. Parikh**, A. K. Das***

History of Insulin

The discovery of insulin was a seminal event in both the study of diabetes and the care of diabetic patients. The development of procedures for purifying and modifying insulin took an additional 30 years. In his masterful rendition of these developments, Michael Bliss recounts the remarkable story surrounding the discovery of insulin and notes that the discovery of insulin at the University of Toronto in 1921-22 was one of the most dramatic events in the history of the treatment of disease.¹ Insulin received its name before it was discovered in 1889. In Germany, Oskar Minkowski and Joseph von Mering observed that total pancreatectomy in experimental animals leads to the development of severe diabetes mellitus and begun the speculation that a mysterious substance produced by the pancreas is responsible for metabolic control.¹ By the first decade of the Twentieth Century it was widely hypothesized that an “internal solution” of the pancreas controls carbohydrate metabolism.¹ Even so there was so much impressionistic evidence supporting the existence of pancreatic internal secretion emanating from the islet cells that in 1907 a Belgian investigator J de Meyer proposed it be named “insulin”. In 1916, Sharpey Schafer in Britain independently suggested the same name. Much truth is in the notion again clarified by hindsight, that insulin was sitting there waiting to be isolated or “discovered”. It almost certainly would have been found during the second decade of the 20th Century, but the work of Central European researchers, such as Zuelzer and the Romanian physiologist, NC Paulesco was utterly disrupted by World War I.¹

In 1920, Frederick Grant Banting a 22 years old orthopedic surgeon was attempting to launch general practice in the small Canadian city of London, Ontario. With time on his hands he accepted a demonstratorship in surgery and anatomy at London’s Western University. On Monday 31st October he had to talk to physiology students about carbohydrate metabolism, a subject with which he was not particularly familiar. Late Sunday night, as part of his preparation he read the leading article in the November issue of *Surgery, Gynecology and Obstetrics* a discussion of “The relation of the Islets of Langerhans to diabetes with special reference to cases of Pancreatic Lithiasis” by Moses Barron. Barron’s unremarkable report stimulated a train of thought in Banting’s mind that caused him, sometime after midnight, to jot down this idea: “Diabetes Ligate pancreatic ducts of dog. Keep dogs alive till acini degenerate leaving

islets. Try to isolate the internal secretion of these to relieve glycosuria”.¹

Banting enjoyed dabbling in research and returned to his alma mater, the University of Toronto and approached JJR Macleod professor of physiology, with a proposal to engage in summer research to test his “Diabetes” idea. Macleod a noted expert in carbohydrate metabolism, doubted that a novice could succeed where masters had failed however he may have seen some value in Banting’s hypothesis that the internal solution was somehow being nullified in pancreatic extracts by the action of the externally secreted digestive ferments. By ligating the pancreatic ducts Banting hoped to induce atrophication of the acinar cells and eliminate the external solution. Banting’s training as a surgeon would serve him well in such research; it also predisposed him to an interest in grafting experiments as the second stage in his work in an age before the rejection phenomenon was understood. Several experts had suggested pancreatic dissection in the search for the elusive secretion. With surplus facilities at hand in his very well - equipped laboratory Macleod agreed to give Banting spare dogs, and a student assistant for a “Summer” thing to the problem. One of the Macleod’s summer students Charles Best, reluctantly won a coin toss to see who would start work with Banting.¹

Banting began his research assisted by Best on 17th May 1921. Macleod was both the formal supervisor and an active advisor before leaving the city in mid-June. The casualty rate among Banting’s dogs was high, some depancreatized, others duct ligated. At the end of July, he and Best began intravenous injections into depancreatized animals of saline extracts of chilled atrophied pancreas. They observed a pattern of hypoglycemic effects. When Macleod returned in September, he urged Banting and Best to repeat and amplify their experiments. He discouraged Banting from returning down the grafting road and after some friction with the young doctor supplied more space and dogs.

By December, Banting and Best had accumulated further evidence that their extract reduced the blood glucose of diabetic dogs. After experiments with fetal calf pancreas and then with fresh beef pancreas, Banting found he could dispense with the cumbersome duct-ligation/atrophication procedures (though he never quite realized that in doing so he had disproven his original hypothesis of an antagonism between the pancreatic secretions). Because of Best’s inexperience Macleod and Banting decided to add JB Collip to the research team. Collip the biochemist from the University of Alberta, was visiting Toronto to work with Macleod and had expressed an interest in the pancreas work.

The first presentation of the Toronto research, read at the New Haven meeting of the American Physiological Association on 30th December 1921 was not well received.

*Dept. of Endocrinology, Seth GS Medical College and KEM & Lilavati Hospitals, Mumbai; **S.K. Soni Hospital, Jaipur; ***Additional Director of Health Services & Professor, Dept. of Medicine, JIPMER, Pondicherry

In their inexperience and haste. Banting and Best had been sloppy and muddled. Their lack of data on the side effects of their extracts (which were almost certainly pyrogenic, as others had been), meant that it was difficult to convince anyone that their findings were better than those of Killner and others. The team's recent experiments notably evidence complied by Collip on the extracts, apparent restoration of glycogen mobilization in the liver and its ability to clear ketones, may have seemed more promising.¹

Discovery of Insulin

On 11th January 1922, clinicians at Toronto General Hospital injected a 14 year old, severely diabetic boy Leonard Thompson with 15 ml of pancreatic extract made by Banting and Best. This clinical test was a failure. The injection caused only slight reductions of glycemia and glycosuria, had no effect on ketoacidosis or the patient's subjective presentation, and resulted in the formation of a sterile abscess. These results were not as encouraging as those obtained by Zuelzer in 1908, Banting later wrote treatment was immediately discontinued.¹

On January 23rd, a new series of injections began. Thompson responded immediately. His glycosuria almost disappeared, his ketonuria did disappear, his blood glucose dropped to normal. He was brighter and stronger. For the first time in history there was clear unambiguous evidence that scientists were able to replace the function impaired in diabetes. This was the demonstration of the isolation of the internal secretion of the pancreas that the world had awaited for 30 yrs.¹

It was JB Collip the biochemist who had produced the successful extract. He had developed a method of extraction that involved changing the concentrations of slightly acidic alcohol solutions of chilled beef pancreas. It is not clear which members of the research team first suggested using acid alcohol until he was able to precipitate out the active principle relatively free from toxic contaminants. It was a major improvement on Banting and Best's methods, the single most important step forward in the discovery process.¹ Banting and Best were particularly confused and self serving in their refusal to recognize their collaborators contributions to the work, as Newelyn Barker put it that "in insulin there is glory enough for all".¹

The glory came almost immediately. On 3rd May, 1922, Macleod delivered a complete summary of the Toronto work at the Washington meeting of the Association of the American Physicians. By now it had been decided to name the active principle "insulin". Macleod suggested the Latin root for islands without knowing of Meyer's and Schaeffer's earlier proposals. The audience agreed that the Toronto team had made one of the greatest breakthroughs in modern medicine and gave them a standing ovation. Eighteen months later, in area of the fastest recognitions of a medical discovery in its history the Nobel committee of the Caroline Institute awarded the 1923 Nobel Prize in Physiology or Medicine to Banting and Macleod. Banting divided his prize money equally with Best, Macleod split his with Collip. The Nobel committee was probably mistaken in not having named Collip as a co-reipient of the prize.¹ For further reading on the exiting story of insulin discovery book by Michael Bliss may be referred.²

Structure of Insulin

Like most of the other hormones, insulin is a protein comprising of 2 polypeptide chains A (with 21 amino acid residues) and B (with 30 amino acid residues) [Fig. 1]. Chains A and B are linked by disulphide bridges. In addition A-chain contains an intra-chain disulphide bridge linking residue 6 and 11. The structure of insulin is shown in the figure 1 below. C-chain, which connects A and B chains is liberated along with insulin after breakdown of proinsulin. Insulin monomers aggregate to form dimers and hexamers.³ Zn hexamer is composed of three insulin dimers associated in threefold symmetrical pattern.

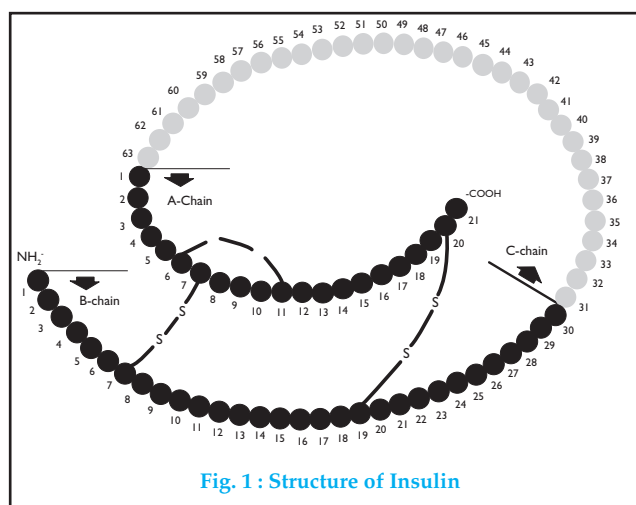


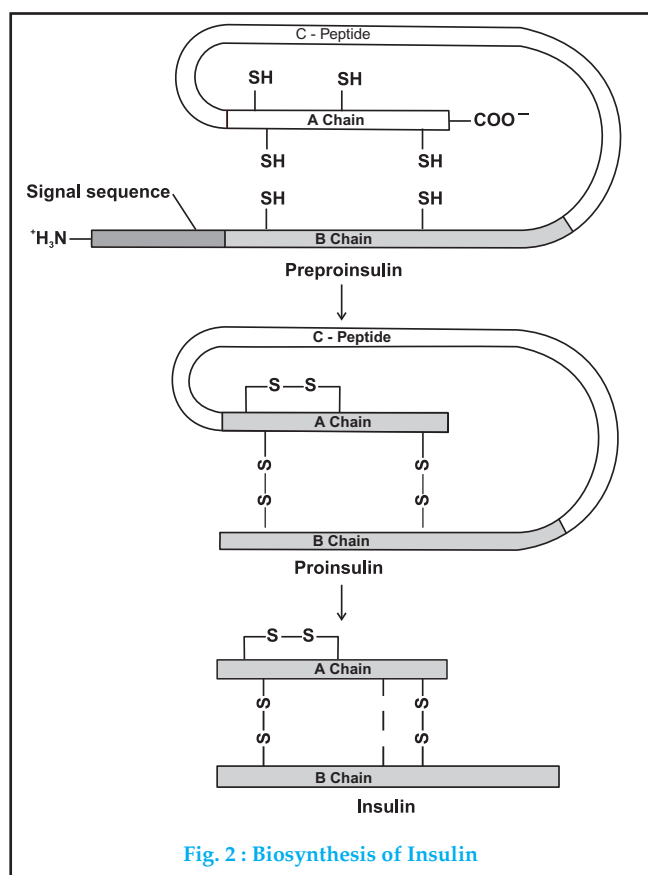
Fig. 1 : Structure of Insulin

Biosynthesis of Insulin

Insulin is synthesized in the beta cells of pancreas in the form of preproinsulin which is the ultimate precursor and gene for the same is located on chromosome 11 close to that for insulin like growth factor-2 (IGF-2) [Fig.2].² Within a minute after synthesis it is discharged into cisternal space of rough endoplasmic reticulum where it is cleaved into proinsulin by proteolytic enzymes. Proinsulin with a C (connecting) chain linking A and B chains is then transported by microvesicles to the Golgi apparatus. Proinsulin is released in vesicles. Conversion of proinsulin to insulin continues in maturing granules through the action of prohormone convertase 2 and 3 and carboxy peptidase H.⁴ Maturing granules are translocated with the help of microtubules and microfilaments.

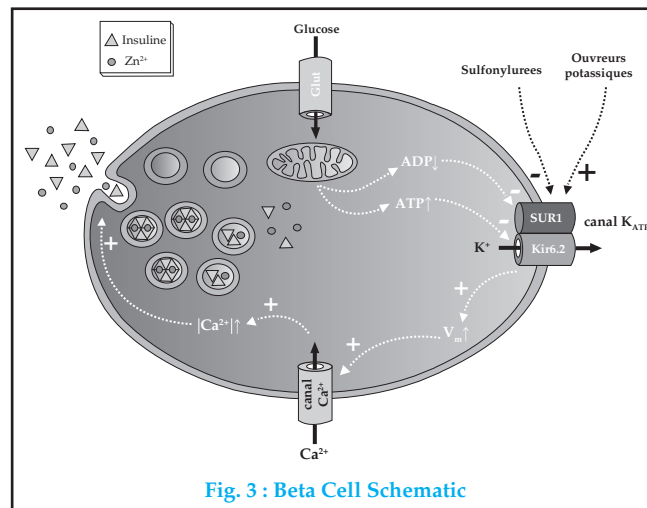
Insulin secretion

Insulin is secreted from the beta cells in response to various stimuli like glucose, arginine, sulphonylureas though physiologically glucose is the major determinant. Various neural, endocrine and pharmacological agents can also exert stimulatory effect. Glucose is taken up by beta cells through GLUT-2 receptors. After entering the beta cell, glucose is oxidized by glucokinase, which acts as a glucose sensor. Glucose concentration below 90 mg/dl do not cause any insulin release. At such substimulatory glucose concentrations, K⁺ efflux through open K_{ATP} channels keeps the β cell membrane at a negative potential at which voltage-gated Ca²⁺ channels are closed. As there is increase in plasma glucose, glucose uptake and metabolism by the β



cell is enhanced. Rise in ATP concentration result in closure of K_{ATP} channels, leading to a membrane depolarization, opening of voltage-gated Ca^{2+} channels, Ca^{2+} influx, a rise in intracellular calcium concentration, and ultimately exocytosis of insulin granules.

Structurally, the pancreatic K_{ATP} channel consists of two unrelated subunits: a sulfonylurea receptor (the SUR1 isoform) and a potassium channel subunit (Kir6.2) that forms the central ion-conducting pathway (Fig 3). The mature K_{ATP} channel exists as an octamer of Kir6.2 and SUR1 subunits in a 4:4 stoichiometry (Fig 3). A sub unit specific site specific to pancreatic K_{ATP} channel, confers glimepiride an advantage over the other sulfonylurea secretagogues. Sulfonylurea⁵,



and non-sulfonylurea drugs act as insulin secretagogues by closing K_{ATP} channels bypassing the β cell metabolism. Diazoxide is a K channel opener and inhibits insulin secretion, independent of blood glucose levels.⁶

Pharmacology of Insulin

Human insulin is now produced by recombinant DNA technology. Various companies differ in their methodology but the basic principal is introduction of human insulin or proinsulin gene into organisms like E coli or Yeast. Yeast based technology may offer physio-chemical structural and protein folding advantages though, this may not be clinically meaningful. The organisms keep on multiplying and in turn producing insulin or proinsulin which is converted to insulin by enzymatic cleavage. Dry human insulin is a microcrystalline powder with a molecular weight of 5808. Insulin precipitates at its isoelectric pH of 5.4, while it is soluble at a pH of 2-3. 1 IU of insulin corresponds to 38.5 μ g dry substance.⁷ Insulin is available in the market in the strength of 40U and 100U i.e. 40U/ml and 100 U/ml respectively. Even U500 is available in US and U10 is sometimes formulated individually for use in infants with diluents provided by manufacturer. Half life of injected insulin is about 40 min.

Insulin preparations

Porcine insulin has been withdrawn from the market globally and bovine is expected to be extinct very soon. Human insulin is available in the short acting i.e. regular and intermediate acting i.e. Neutral Protamine Hagedorn (NPH) forms. Insulin analogues which are synthetically modified with some changes in the amino acid sequence are also available. Rapid acting insulin analogues Lispro (Eli Lilly) and Aspart (Novo Nordisk) are already in the market while glulisine (Sanofi-Aventis) is to be launched shortly. Glargine (Sanofi-Aventis) and Detemir (Novo Nordisk) are the long acting analogues available. Ultralente is now withdrawn.

Regular insulin

Regular insulin is available as a clear solution at neutral pH. 0.4% of zinc is added to allow the insulin molecules to self associate into hexamers. For the prevention of growth of micro-organisms phenol or m-cresol is added. Regular insulin has its onset of action within 15-30 min after subcutaneous injection, maximum activity peaks at 120-150 min while the action lasts for 6-8 hours. In order match the peaks of glucose and insulin, subcutaneous injection is advised to be taken 30-40 min prior to meals.

NPH or isophane insulin

Isophane insulin is known as NPH insulin as it was developed in Denmark at the Hagedorn Laboratory in 1940s.⁸ In order to prolong the action of insulin, a positively charged protein, protamine is added in a molar ratio of 1:6 to regular insulin. It binds with the negatively charged insulin at neutral pH. Neutral pH value is achieved by use of phosphate buffer. Zn and m-cresol are also added. NPH insulin is slowly absorbed from subcutaneous tissue with peak at 5-7 hours and the action lasts for 12-15 hours. This insulin is most commonly used at bedtime to control fasting blood sugar.

Lente insulin

If zinc is added in excess amount (10 times that added in NPH), at neutral pH and if acetate is used as a buffer in stead

of phosphate, it forms insoluble insulin-zinc complexes.⁹ This property is exploited for the production of lente insulins. The action profile of these preparations depends upon the physical conditions of insulin. Semilente is amorphous and has biphasic absorption kinetics with short duration of action. Ultralente is long acting crystalline suspension. These insulins cannot be mixed with regular insulin due to their zinc content and are not very popular.

Premixed formulations

Regular and NPH insulin are available in a premixed formulation with 30:70, 50:50, 25:75 proportions. These preparations are very popular as there is no mixing involved. Patients of type 2 diabetes on split mix regime may be shifted to premixed preparation if they are on approximately similar proportion.

Rapid acting Analogues

Regular insulin when injected is in hexamer form, which slowly releases into monomers and is responsible for delay in the action. Analogues are synthesized by modifying the amino acid sequence so as to keep insulin molecule in monomeric form which has rapid onset of action and peak resembling physiology. After subcutaneous injection the action starts at 30 min, peaks at 60-90 min and is over by 4 hours. This action profile is very efficient in controlling post-prandial glycemic excursions without any risk of delayed hypoglycemia.^{10,11}

Long acting analogues

Long acting analogues are designed in an attempt to obtain a steady basal insulin level without any peak unlike NPH, which has a risk of late night hypoglycemia. In insulin glargine, the amino acids sequence is altered so as to change isoelectric pH of insulin to 7.4 from 5.4. It is clear and soluble at an acidic pH. After injection in subcutaneous space it gets precipitated and is released slowly, making its action last for even more than 24 hours. Most of the patients require a single dose for basal cover. Insulin detemir has a long action by virtue of a fatty acid chain attached to it.

Premixed Analogue preparations

Insulin glargine cannot be mixed with any other insulin by virtue of its acidic pH and high Zn content i.e. 30 µg/ml. Premixed preparations are available with protamine insulin. In these preparations the protamine-insulin part has to be formulated with the same insulin analogue like lispro or aspart. These preparations are available in 25:75, 30:70, 50:50 proportions.

Selection of insulin

Species

Porcine insulin is no more available. Bovine insulin, the most economical option for non-affording patients will be out of market very soon, due to ecological and environmental considerations. Human insulin should be preferred for management of GDM, diabetic women considering pregnancy, individuals with allergy or immune resistance to animal derived insulins, those initiating insulin therapy and those expected to use only intermittently. Changing insulin species and brands should be avoided as it may affect blood glucose control.

Types of insulin (Conventional) [Physical Appearance & Colour Code]

Conventional

Regular insulin – it is clear and watery, short acting.

NPH insulin – Cloudy, intermediate acting.

Semilente, Lente and Ultralente are insulin formulations with varying concentration of zinc, their actions being short, intermediate and long acting respectively.

Premixed preparations – with regular and NPH insulin mixed in fixed proportions viz. 30/70, 50/50, 25/75 are available. These combinations are not physiological. They should be used if there is doubt about patient's compliance or feasibility of mixing insulins.

Insulin Analogues

Insulin analogues have been synthesized by modifying structure of insulin so the action profile mimics physiology.

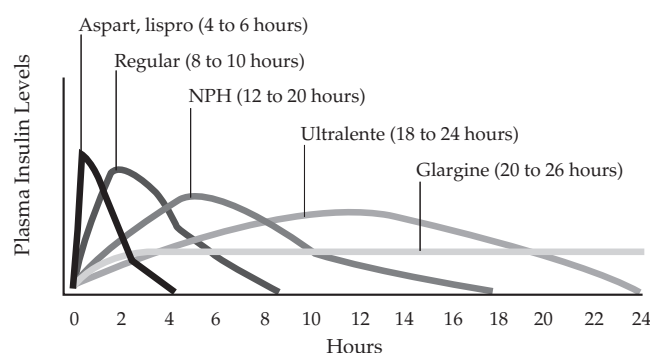
Rapid acting analogues – Aspart, Lispro, Glulisine

Long acting analogues – Glargine & Detemir

Table 1 : Time Course of Action of Human Insulin Preparations

Insulin Preparation	Onset of Action	Peak Action	Effective Duration of Action
Rapid-Acting Insulin Analogs Insulin lispro Insulin aspart Insulin glulisine	5-15 min 5-15 min 5-15 min	30-90 min 30-90 min 30-90 min	3-5 h 3-5 h 3-5 h
Short-Acting Insulin Regular	30-60 min	2-3 h	5-8 h
Intermediate-Acting Insulins NPH Lente	2-4 h 3-4 h	4-10 h 4-12 h	10-16 h 12-18 h
Long-Acting Insulins Ultralente Insulin glargine Insulin detemir	6-10 h 2-4 h 2-4 h	10-16 h Peakless 6-14 h	18-24 h 20-24 h 16-20 h
Insulin Mixtures 70/30 human mix (70% NPH, 30% regular) 75/25 lispro analog mix (75% intermediate, 25% lispro) 70/30 aspart analog mix (70% intermediate, 30% aspart) 50/50 lispro analog (50% intermediate, 50% lispro) 50/50 human mix (50% NPH, 50% regular)	30-60 min 5-15 min 5-15 min 30-60 min	Dual Dual Dual Dual	10-16 h 10-16 h 10-16 h 10-16 h

Profiles of Human Insulins and Analogues



Strengths available (U-40, U-100)

U40 i.e. 40 U/ml of insulin is the most widely used in India. U100 can be used for patients requiring very high doses. Even U500 is available in US and U10 is sometimes formulated individually for use in infants with diluents provided by manufacturer. Vials are U-40, U-100 and rarely U-10, U-500 but cartridges are always U-100. WHO recommend eventually all insulins across the globe to be U-100.

Storage

Vials & cartridges not in use should be refrigerated. Extremes of temperature $<2^{\circ}\text{C}$ and $>30^{\circ}\text{C}$ and excess agitation should be avoided to prevent loss of potency, clumping, frosting or precipitation. Vials & cartridges in use, used for >1 month may lose potency specially if not stored at room temperature. They should be kept away from direct sunlight.

Syringes (U-40, U-100)

Though syringes are available in varying capacities viz. 0.3, 0.5, 1 and 2 ml; in our country 1 ml is the most commonly used. Different syringes are available for use with 40U and 100U vials, differing in markings, though both may be of 1 ml capacity. Syringe of corresponding strength only should be used. Needles are available with thickness varying from 26 to 31 Gz. While changing from one length to another, blood sugar should be monitored. 30 or 31 Gz needle can bend in a single use. Syringes must not be shared. Bending or breaking needle should be avoided. Syringes and needles should be disposed in a puncture resistant container. Though the manufacturers recommend single use, reuse is widely practiced and supposed to be safe as most insulin preparations have bacteriostatic additives. Reusing of needles may increase risk of infection for some individuals—patients with poor hygiene, an acute concurrent illness, open wounds on the hands, or decreased resistance to infection for any reason.

Mixing Insulins

Patients who are well controlled on a particular mixed-insulin regimen should maintain their standard procedure for preparing their insulin doses. Insulin glargine should not be mixed with other forms of insulin due to the low pH of its diluent. Currently available NPH and short acting insulin formulations when mixed may be used immediately or stored for future use. Rapid-acting insulin can be mixed

with NPH, lente, and ultralente. Mixing of short-acting and lente insulins is not recommended except for patients already adequately controlled on such a mixture. If short-acting and lente mixtures are to be used, the patient should standardize the interval between mixing and injection.

Timing of injection

Regular insulin should be injected on an average 30 min prior to meals. If rapid acting analogues are used the time lag should not be more than 30 min and can be given immediately prior to or even just after meals. Similar instructions should be given if the patient is using regular or rapid acting insulin mixed with other insulins. These intervals may need to be individualized in some patients depending on site of injection, type of food, exercise etc. Intermediate acting insulin is preferable given at bed time to avoid late night hypoglycemia associated with predinner administration.

Sites

Insulin may be injected into the subcutaneous tissue of the upper arm and the anterior and lateral aspects of the thigh, buttocks, and abdomen (with the exception of a circle with a 2-inch radius around the navel). Rotation of the injection site is important to prevent lipohypertrophy or lipoatrophy. Rotating within one area is recommended (e.g., rotating injections systematically within the abdomen) rather than rotating to a different area with each injection. The abdomen has the fastest rate of absorption, followed by the arms, thighs, and buttocks (Fig. 4).

Exercise increases the rate of absorption from injection sites while; areas of lipohypertrophy usually show slower absorption. The rate of absorption is faster if given intramuscularly and, although not recommended for routine use, can be given under other circumstances (e.g., diabetic ketoacidosis or dehydration).

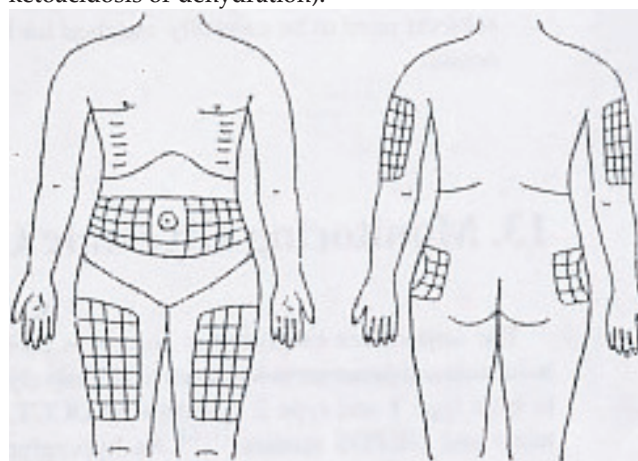


Fig. 4 : Potential Site of Insulin Injection

Insulin injection

Hands and injection site should be clean. All cloudy insulins need to be gently rolled in between the palms or shaken gently. In case of pens, pen needs to be rotated upside down for at least ten times before using. Air of volume equal to dose of insulin should be injected in vials.

While mixing insulins, clear insulin should be drawn first. Before injecting one should look for air bubbles if any by flicks of forefinger against the upright syringe. In case of pens, to prevent this, avoid leaving needle on pen between injections and prime the needle with 2 U of insulin before injection. Use of alcohol or any antiseptic at injection site is not required. On the contrary it may make injection more painful by destroying silicon coating on the needle tip. Mostly insulin injection is given in subcutaneous tissue. Patient should grasp a fold of skin, release the pinch, then inject at a 90° angle. In children and thin individual short needle may be used, or may need to pinch the skin and give injection at 45° angle. Routine aspiration is not necessary. Needle should be embedded within the skin for 5 sec after complete depression particularly with the use of insulin pens. Patient should rotate his site of injection but in the same area to prevent lipohypertrophy.

Side effects

- **Hypoglycemia:** Late night hypoglycemia is mostly attributable to night dose of intermediate acting insulin. Shifting predinner dose to bedtime or reduction in bedtime dose may be required. Postabsorptive hypoglycemia is mostly due to delayed hyperinsulinaemia while using short acting regular insulins. It can be prevented by having a snack, reducing dose of regular insulin or substituting with rapid acting analogues.
- **Weight gain:** Initial weight gain is due to correction of the catabolic state. Later patient puts on weight by fluid retention, and excessive eating attributable to hypoglycemia or fear of impending hypoglycemia.
- **Local:** Allergy, infection, injection site abscess and lipodystrophy are very rarely seen but lipohypertrophy is still common and is attributable to repeated injection of insulin at same site.
- **Anaphylaxis:** Very rarely seen and requires desensitization with gradually increasing doses of insulin.

Initiating insulin

Various regimens are available for initiating insulin therapy in a diabetic patient. The choice of regimen depends on the diagnosis, glycemic status, patient compliant and physician's choice.

Sliding Scale

Subcutaneous sliding is used in some hospitals but is not recommended. Sliding scale can be used in some situation when insulin infusion pump is available. 50 U of regular insulin is diluted in 50 ml of normal saline and the infusion is started. Blood glucose needs to be tested every hour and the rate of infusion is adjusted as per hospital glycemic protocols and depending on the blood sugar (Sliding scale for IV insulin - Insulin infusion via insulin syringe pump : <120 mg/dl - No insulin; 120 - 200 - 2 U/hr; 200 - 300 - 3U/hr; 300 - 500 - 5U/hr; > 500 - 7U/hr).

Multiple doses

Patient should be given 2 doses of NPH insulin one before breakfast (BBF) and one at bedtime and 3 doses of regular insulin half an hour before major meals. Initial dose is started

empirically and later the doses are adjusted according to Self Monitoring of Blood Glucose (SMBG). SMBG reflects the insulin action as follows

Fasting – Bed time NPH

Pre-lunch – Regular insulin BBF

Post lunch – regular insulin before lunch and NPH insulin BBF

Pre-dinner – pre-lunch regular insulin

Bed-time – pre-dinner regular insulin

Bed time dose of insulin can be preponed and mixed with predinner dose of regular to reduce number of injections. This may increase risk of Somogyi Phenomenon i.e. Late night hypoglycemia followed by early morning hyperglycemia.

Split mixing

Two doses of insulin with mixture of regular and NPH, BBF and predinner are given. Prebreakfast regular insulin is supposed to act on post breakfast glycemic excursion whereas post lunch peak is expected to coincide with peak of prebreakfast NPH. Similarly predinner regular controls the post dinner blood sugar whereas the predinner NPH acts on fasting blood sugar. Doses are adjusted using SMBG.

Bed-time insulin and daytime sulphonylurea (BIDS)

This is used in patients going in secondary OHA failure or if early initiation of insulin is considered. One single dose of NPH is given at bed time.

MDI using analogues

Analogues are designed so as to mimic physiology. Long acting analogue Glargine given once a day can provide basal insulin without any peak. For controlling post prandial excursions bolus shots of rapid acting analogue Lispro or Aspart should be given along with each major meal. Though this algorithm mimics the physiology, its use is limited in view of its cost and multiple pricks needed.

Carbohydrate counting

This is a recent development in the management of type 1 DM which allows tailor making of insulin doses as per quantum of meal the child is going to have. Extra dose of regular insulin which needs to be added to cover additional carbohydrates is calculated by using following formula.

$$500 / \text{Total daily dose of insulin} = X$$

For every X gm of carbohydrates 1 U of regular insulin should be added. In case of analogues 500 in the formula should be replaced by 450.

Rule of 1500

Many a times the premeal blood sugars are far above target. In such situation extra dose of regular insulin should be given based on following formula.

$$1500 / \text{Total daily dose of insulin} = X$$

For every X mg/dl of blood sugar above set target 1 U of regular insulin should be added.

Initial Insulin Doses and Distribution

The insulin dosage required for meticulous glycemic control in typical patients with type 1 diabetes within 20% of their ideal weight, in the absence of intercurrent infectious or other periods of instability is -0.5-1.0 units/kg body weight/day. During the relative remission (honeymoon period) early

in the course of the disease, insulin requirements generally are less. During intercurrent illness, dosage requirements may increase markedly. Dosage also increases during the adolescent growth spurt, and some adolescents may have a sustained increased dose requirement.

About 50% of the total daily insulin dose is used to provide basal insulinemia. The remainder is divided among the meals either empirically, proportionate to the carbohydrate content of the meals, or by giving -1.0 - 1.5 units insulin/10 g carbohydrate consumed.

Patients with type 2 diabetes have defects in both insulin secretion and insulin action. The impairments in insulin secretion are manifest in at least three ways:

1. Blunted or absent first-phase insulin response to glucose, so that insulin secretion is delayed and fails to restore prandial glycemic excursions in a timely manner.
2. Decreased sensitivity of insulin response to glucose, such that hyperglycemia may fail to trigger an appropriate insulin response.
3. Decreased overall insulin secretory capacity, progressive in nature with more prolonged and therefore more severe type 2 diabetes.

This impairment in insulin secretory response is not static but dynamic, such that chronic hyperglycemia may itself aggravate the impairment in insulin secretion, a phenomenon known as glucose toxicity. Thus, with decompensation of glycemic control in type 2 diabetes, there is concomitant deterioration in insulin secretory response. Moreover, and most important, when there is correction of hyperglycemia, there is some reversal of the impairment in endogenous insulin response to a meal challenge (i.e. a demonstrable improvement in insulin secretion). Thus, attainment of glucose control facilitates maintenance of glucose control.

Patients with type 2 diabetes also have impaired insulin action (insulin resistance) at target cells. This increases the overall insulin requirement. Like the defect in insulin secretion, this impairment in insulin action is not static but dynamic. Chronic hyperglycemia may aggravate the impairment of insulin action, another manifestation of glucose toxicity. Thus, with decompensation of glycemic control, insulin action is diminished. Moreover, when hyperglycemia is corrected, some reversal in the impairment of insulin action occurs.

Evaluation of patient with increasing insulin requirements

- On history rule out causes like infection, stress, puberty, pregnancy and endocrine causes like acromegaly, Cushing's Syndrome etc.
- Look for expiry date of insulin vial and any abnormality in appearance of insulin like clumping, precipitation etc.
- Confirm whether the patient is using prescribed insulin, in proper doses with proper syringe and correct technique.

- Evaluate mental condition of the patient and confirm the compliance.
- Look for lipohypertrophy at the site of injection.
- After ruling out all these, patient should be investigated for causes like occult infections or malignancies. Subcutaneous and intravenous insulin resistance is a rarely described entity.

Evaluation of patient with decreasing requirements

- Impending renal failure
- Honeymoon phase in type 1
- Hypothyroidism, Addison's Disease and Hypopituitarism particularly in patients with autoimmunity
- Remission of diabetes

Conclusions

Patient education is an important aspect of insulin therapy. Patients should be educated about storage of insulin, use of syringes, mixing of insulin, timing of injection and meals, selection of proper site and proper technique of injection. SMBG and hypoglycemia management are integral part of patient education. Patient education and proper evaluation can improve outcome in the form of better glycemic control.

References

1. Shah SN, Joshi SR, Parmar DV. History of Insulin. J Assoc Physicians Ind 1997; 45 (Suppl 1):4-9.
2. Bliss M. The discovery of Insulin. Chicago: University of Chicago Press, 1982.
3. Bell G I, Pickett RL, Rutter WJ et al. Sequence of the human insulin gene. Nature 1980; 284: 26-32
4. Hutton JC. Insulin secretory granule biogenesis and the proinsulin-processing endopeptidases. Diabetologia 1994; 37 (Suppl.2): S48-56
5. Gribble, F.M., and Reimann, F. 2003. Sulphonylurea action revisited: the post-cloning era. Diabetologia. 46:875-891.
6. Gribble, F., and Ashcroft, F.M. 2000. New windows on the mechanism of action of potassium channel openers [review]. Trends Pharmacol. Sci. 21:439-445.
7. WHO Expert Committee on Biological Standardisation. Thirty-seventh report. Geneva: World Health Organisation, 1987: 25-6.
8. Krayenbuhl C, Rosenberg T. Crystalline Protamine Insulin. Rep Steno Mem Hosp Nord Insulinlab 1946;1: 60-73
9. Hallas- Moller K, Peterson K, Schlitchkrull J. Crystalline amorphous insulin-zinc compounds with prolonged action. Science 1952;116: 394-6
10. Heise T, Weyer C, Serwas A et al. Time-action profiles of novel premixed preparations of insulin lispro and NPL insulin. Diabetes Care 1998; 21: 800-3
11. Weyer C, Heise T, Heineman L. Insulin aspart in a 30/70 premixed formulation: pharmacodynamic properties of a rapid-acting insulin analogue in stable mixture. Diabetes care 1997; 20: 1612-15