REVIEW ARTICLE

Oral Insulin Delivery: Novel Strategies

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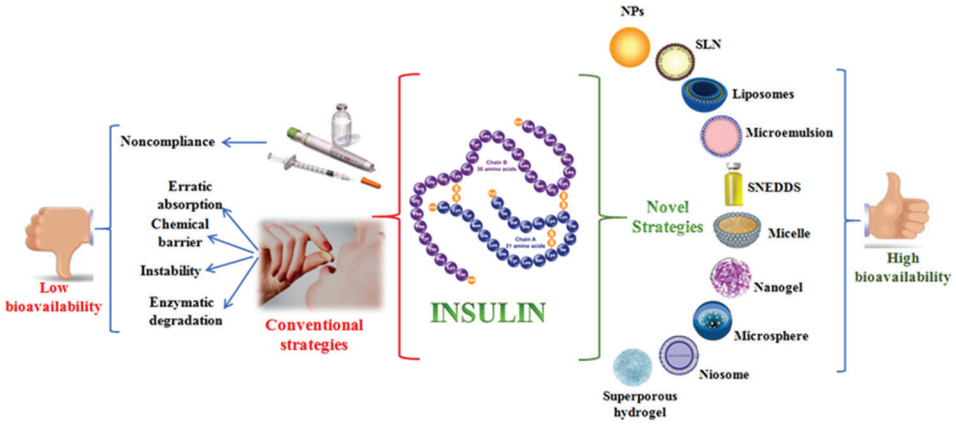
Abstract

Diabetes is a disorder related to the deficiency in the secretion or action of insulin - a peptide hormone synthesized by the β cells of islets of pancreas. Insulin is given by the subcutaneous (SC) route. Patient non-compliance is frequent with the SC route. To remove the hurdle related to oral insulin delivery various efforts have been made. Thus, oral insulin is a dream of patients. Nanotechnology is an emerging era of science which provides an innovative means to detect, diagnose and to treat a disorder. Nanocarriers have been garnering more attention recently because of their size in nano range and greater surface area. These characteristics improve their absorption in contrast to bigger carriers. This article gives an insight into different novel approaches to get better oral insulin delivery. These novel formulations improve bioavailability; absorption problems associated with insulin and give protection from enzymatic degradation. More research has been done on nanoparticles (NPs) as a carrier to deliver insulin orally. At present, researchers from both industries as well as academics are working on oral insulin. With this struggle, the dream of researcher to deliver insulin orally will turn out to be a reality in the future.

Key words: Diabetes, improved bioavailability, insulin, nanoparticles, novel approaches, oral delivery

Graphical Abstract



INTRODUCTION

Proteins, from the Greek Proteios, meaning first, is an organic compound which is present in every living cell. Proteins are one of the building blocks of the body and have played an important role in cell growth and metabolism. Proteins in the form of skin, hair, muscles, cartilage, tendons and ligaments hold together, protect, and provide a proper organization to the body of a multi-celled organism. Proteins in the form of hormones, enzymes, antibodies, and globulins, help in catalyzing and regulating the chemistry of the body. Proteins in the form of hemoglobin, myoglobin, and various lipoproteins, effectively do the transport of oxygen and other substances within an organism.[1] Due to specificity, excellent activity and effectiveness of proteins and peptides,

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they are used as biopharmaceuticals.[2] Structurally proteins are consisting of an amino acids which are considered by the $CH(NH_2)COOH$ substructure. Peptides are amino acid monomers linked by peptide (amide) bond. According to the World Health Organization (WHO), diabetes is considered as a chronic disease, which arises when sufficient amount of insulin is not produced by the pancreas (Type 1 diabetes), or insulin which is formed is not utilized properly by the body (Type 2 diabetes). This leads to an elevation of blood glucose level (hyperglycemia). It often results from over body weight and physical immobility.[3] Diabetes is the most common endocrine disorder. According to the National Diabetes Statistics Report 2014, 9.3% of the population or 29.1 million people are suffering from diabetes in the United States. Further, 21 million people are diagnosed with diabetes whereas 8.1 million people are undiagnosed.[4] Between 2010 and 2030, the count of adults with diabetes in developing countries will be 69% and in developed countries will be 20%, respectively.[5] As per WHO, during the year 2000, around 31.7 million people in India were affected by diabetes. Thus diabetes proved to be a major disease which increases the rate of morbidity and mortality.[6]

TYPES OF DIABETES MELLITUS (DM)

- Type-I or insulin dependent DM/juvenile-onset/ketone-prone diabetes
- Type-II or insulin-independent DM/adult-onset diabetes
- Gestational DM, and
- Other specific types (secondary DM).

Pancreas synthesizes insulin which allows the utilization of glucose from carbohydrate which provides energy and store for future use. It helps to control your blood sugar level and does not allow the extremes conditions such as hyperglycemia or hypoglycemia. Insulin is useful for both the types of DM. It has a molecular weight of 5808. Insulin is a collection of 51 amino acids exhibited in two chains A (21 amino acid residues) and chain B (30 amino acid residues) linked by disulfide bridges. An intrachain disulfide bridge is present in chain A which links residue 6 and 11. C-chain which connects A and B chain releases drug with insulin after the breakdown of proinsulin.[7]

A major challenge is to deliver protein drugs like insulin appropriately and effectively.[8]Administration of insulin is limited to the subcutaneous (SC) route as it requires one or more daily injections which may lead to peripheral hyperinsulinemia and hyperinsulinemia. Thus, several research studies are seeking for the progress of new formulations of insulin which can be given by another route. Oral intake of insulin mimics the typical insulin pathway within the body after endogenous secretion. Therefore, oral novel formulations of insulin will prove to be a successful key for management of insulin. However, insulin has very low oral bioavailability and work has been performed to aid insulin bowel absorption. As an outcome of this, insulin delivery and release have acquired greater interest by researchers.[9]

MECHANISM OF ACTION

Various actions of insulin are depicted in Figure 1. Insulin inhibits glycogenolysis, ketogenesis, gluconeogenesis, proteolysis, and lipolysis. Insulin allows the process of uptake of glucose by muscle and tissue, glycolysis, glycogen synthesis, and protein synthesis. These actions lead to control of glucose in the blood.

Binding of insulin to its receptor triggers inherent tyrosine kinase activity results in autophosphorylation and the recruitment of intracellular signaling molecules, such as insulin receptor substrates (IRS). Cascade of phosphorylation and dephosphorylation reactions initiated by means of IRS and other adaptor protein, leading to metabolic and mitogenic effects of insulin. As an instance, initiation of the phosphatidylinositol-3-kinase (PI-3-kinase) pathway stimulates glucose transporter (GLUT) allowing translocation of glucose (e.g., GLUT4) to the cell surface, an occasion that is vital for glucose uptake via skeletal muscle and fat.[10]

SC route is preferred conventionally for the administration of insulin with the aid of injection which is marked by insulin units. Rapid, short, intermediate, and long-acting insulin injections are given individually or combined in the same syringe. Units can be assigned either as per the size of syringe or manufacturer. Insulin syringes are accessible with 0.3-, 0.5-, 1-, and 2-ml limits with various needle lengths. Syringes should be disposable and should not be used for another person because of the hazard of getting infected with a blood-borne viral infection. 100 or 500 units/ml concentrations in which insulin is available (entitled U-100 and U-500, respectively; 1 unit equals ~36 μg of insulin).[11]

DISADVANTAGE OF THE SC ROUTE

a. Two or more injections are needed to reduce long-term complications of hyperglycemia (retinopathy, neuropathy, and nephropathy).

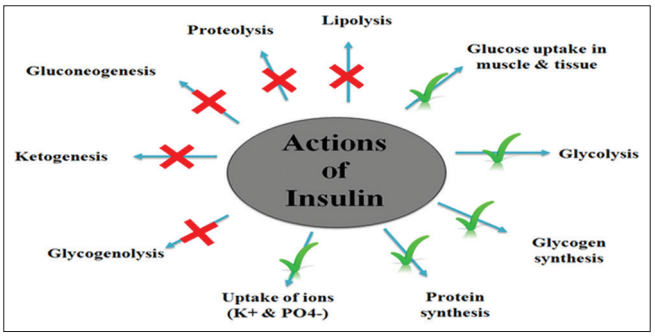

Figure 1: Actions of insulin

b. Patient non-compliance.
c. Injection is given repeatedly results in lipoatrophy (it is a term describing the loss of SC fat) or lipohypertrophy (irregular accumulation of fat under the skin surface).
d. Moreover, insulin given by SC route goes directly into general circulation which ends up in peripheral hyperinsulinemia (a condition within which the level of insulin will be highly circulating in the blood relative to the level of glucose).[12]

As compared to other routes of drug delivery; oral route has always dominated. This can be attributed to various advantages of the oral route. In spite of being the most important route of administration, it is generally not convenient to deliver every drug through the oral route. Insulin delivery via oral route is challenging as incomplete and unpredictable absorption through the gastrointestinal tract (GIT), degradation due to varying pH of the stomach and enzymatic degradation lead to poor oral bioavailability. Hence, the importance of novel drug delivery system (NDDS) gained more attention of researchers for efficient delivery of insulin.[13]

BENEFITS OF AN ORAL INSULIN DELIVERY

1. Patient compliance
2. Convenient
3. Painless
4. Easy for self-medication
5. Avoid weight gain
6. Reduces the danger of hypoglycemic incidents, immune responses and other problems associated with SC route
7. Cost effective.[2]

OBSTACLES TO ORAL INSULIN DELIVERY

Absorption through gastrointestinal membrane

Paracellular and the transcellular route is the general route for absorption of molecules. The earlier one is favored path for those hydrophilic molecules which have a mol. Wt. less than 500 Da. The molecules having high molecular weight like insulin (about 6KDa) would not enter via this route. Absorption of insulin by transcellular route is hindered by enormous molecular size, its charge, and its hydrophilicity.

Enzymes present all over in GIT

There is a group of aspartic proteases stated to as pepsin is present within the stomach. Pancreatic proteases present in small intestine comprising the serine endopeptidase (trypsin, α-chymotrypsin, elastase and exopeptidases, carboxypeptidase A, and carboxypeptidase B) results in degradation of proteins. Various other enzymes are situated at the brush–border membrane or in the enterocytes of the intestinal tract. Following is the order of enzymatic degradation of insulin in the small intestine: Duodenum > jejunum > ileum.[8,12]

Stability of insulin

The structure of insulin is very delicate. Stability is affected by component elements in addition to processing elements. Several protein degradation pathways are oxidation, photodegradation, disulfide scrambling, deamidation, aggregation, precipitation, dissociation, and fragmentation. Insulin is prone to oxidative damage through reaction of certain amino acids with oxygen radicals present in their environment. Oxidation can alter a protein's physiochemical characteristics (e.g., folding and subunit association) and lead to aggregation or fragmentation. Methionine, cysteine, histidine, tryptophan, and tyrosine are most liable to oxidation.

Deamidation is a chemical reaction in which an amide functional group is removed from an amino acid. Results include isomerization, racemization, and truncation of proteins. In acidic solution, extensive deamidation of insulin occurs at the residue AsnA21, whereas in neutral solution, less deamidation occurs at AsnB3 residue. Some processing factors which affect insulin stability need to be monitored to ensure product safety and efficacy.

Light degradation: Photo-oxidation is responsible for changing primary, secondary, and tertiary structures of proteins and lead to differences in long-term stability, bioactivity, or immunogenicity. Exposure to light can activate a chain of biochemical events that continue to affect a protein even after the light source is turned off. It depends on how much of energy imparted to a protein and the presence of environmental oxygen. Excipients and containers used can affect the oxidation of protein. Oxidation can be induced during protein processing and storage by peroxide contamination resulting from polysorbates and polyethylene glycols which are commonly used pharmaceutical excipients. Peroxide can also leach from plastic or elastomeric materials used in primary packaging container closure systems. Thus care must be taken during processing with container closure systems.

Aggregation is a common problem observed during manufacture and storage of proteins. Aggregation occurs due to mechanical stresses of agitation such as shaking, stirring, pipetting, or pumping through tubes. Freezing and thawing process also promote aggregation. Microaggregated subvisible particles formed while manufacturing process can develop into larger particles over time during storage.

Protein degradation occurs due to mechanical shearing and exposure to ultrasound. Shearing significantly augments the chances for dissolved proteins to adsorb onto air/water and

water/organic solvent interfaces, thus promoting hydrophobic interactions further leads to aggregation. The choice of the apparatus used for the preparation of the w/o emulsion has an influence on the protein stability.[14,15]

Chemical barrier

In the GIT, many proteins are prone to the pH variation. There's a difference in pH ranging from acidic in the stomach (pH 1.1-3.0) to somewhat basic in the intestine (pH 6.4-8.0). Such deviations in pH may lead to pH-induced oxidation, deamidation and hydrolysis result into an inactive product.[8,12]

ATTEMPTED ORAL INSULIN DELIVERY SYSTEMS

Enzyme inhibitors

Many approaches have been developed to improve insulin bioavailability, one of them is enzyme inhibitor. Protease inhibitors prevent the enzymatic degradation of insulin, enhances the membrane permeability or widening of tight junctions to improve absorption of insulin. Coadministration of protease inhibitors can lower the enzymatic barrier and prevent degradation of proteins and peptides in the GI tract thereby facilitating intestinal absorption. Sodium cholate in conjugation with aprotinin is used to bring out enzyme inhibition which enhanced insulin absorption in rats. Other inhibitors are leupeptin (Protease inhibitor) and FK-448 (chymotrypsin inhibitor). The impervious film formed by polymer cross-linked with azo aromatic groups protects insulin from digestion in the stomach and small intestine. Once reached to large intestine, this polymer film degrades by microflora thereby releasing the drug into colon lumen for absorption enzyme inhibitors are classified into reversible or irreversible. An irreversible inhibitor produces a stable complex with the enzyme (forms a covalent bond with the enzyme). Therefore, the enzyme is permanently inactivated or it is slowly reactivated.[16-18]

Nanocarrier-based systems for mucoadhesive drug delivery

These systems prevent degradation of entrapped drug and improve the circulation time of drug at absorption site. Polyionic polymers show mucoadhesive properties. From such polymers, alginate showed the best candidate for the intestinal mucosal system. Alginate is a nontoxic, biodegradable, and mucoadhesive polysaccharide polymer that possesses mucoadhesive properties than carboxymethylcellulose, chitosan, poly (lactic acid), and other polyionic polymers. In additionally, alginate possesses pH-sensitive swelling property. At low pH, it is insoluble and shrinks, preventing drug to escape from encapsulated matrix. At higher pH, it swells, become more porous discharging the entrapped drug. To resolve this problem, silica-alginate composites are used to shield insulin from degradation. There are two ways by which drug release from alginate matrix takes place: Diffusion from porous matrix and degradation of the polymer network.[8]

Absorption enhancers

A range of absorption enhancers is used which opens the tight junction and enables a water-soluble protein to pass. Conjugation of insulin with trans-activating transcriptional activator, a cell penetrating peptide (CPP) increases insulin transport across Caco-2 cells (human colon epithelial cancer cell). The popular example of the permeation (absorption) enhancer is mucoadhesive polymers. It proves to be safe and effective intestinal penetration enhancers. Absorption of the macromolecular drug can also be facilitated by chitosan, Pz-peptide, thiolated polymers, and others.[18]

Overcoming the mucosal barrier

To overcome the mucosal barrier, insulin is chemically conjugated with CPP. Given that CPP-mediated cell entry is independent of the receptor. So insulin bonded to a CPP should be transducible for all cell types. Due to this conjugation, quick response is observed as it crosses the tight junction of intestinal mucosa and it intently resembles normal physiologic responses. It will also give protection from the enzymes as the residence time of this conjugation reduces.[19]

NOVEL STRATEGIES FOR ORAL INSULIN DELIVERY

NDDS offer some advantages such as increased efficacy of the drug, site-specific delivery, and reduced side effects.[20] Various NDDS researched for oral insulin delivery are nanoparticles (NPs), liposomes, microemulsions (MEs), self-nanoemulsifying drug delivery systems (SNEDDS), micelles, nanogels (NGs), microspheres, niosomes, and superporous hydrogels (SPHs).

Nanoparticles (NPs)

A NP is a small entity that works as an entire unit in relation with transport and other aspects. Particle size of NPs ranges from 10 to 1000 nm. More attention has been focused on NPs because they increase the absorption of a drug due to nano-sized particle and greater surface area.[21] Furthermore, it can trigger as well as control the release of the content which may enhance the transmucosal transport and cellular uptake. Targeting will also be possible when attached to a suitable moiety.[13]

Mechanisms of the absorption of NPs

Two major pathways by which NPs pass through intestinal epithelium are paracellular (between cells) and transcellular

(through the cells). Transcellular route is the most common route of absorption. By transcellular route, NPs can be taken up by enterocytes or M cell of peyer's patches. However, macromolecules like polymeric NPs cannot pass through cells due to their big size. Thus, four mechanisms for active transport of polymeric NPs are phagocytosis, macropinocytosis, clathrin-mediated endocytosis, and caveolin-mediated endocytosis. Paracellular route is preferred for transport of hydrophilic drugs. However, it is restricted to polymeric NPs due to very small intercellular space and tight junctions between epithelial cells. Hence to improve paracellular transport, tight junctions can be opened reversibly using permeation/penetration enhancers such as cationic and anionic polymers [Figure 2 shows mechanism of absorption of NP].[22-24]

NPs can be classified into polymeric and lipid-based systems. Since literature reveals the use of polymers to a greater extent for insulin delivery, these are detailed in depth in this review.

Polymeric NPs

Biocompatible and biodegradable polymeric NPs have turned out to be an ideal carrier for delivering proteins and peptides orally. It improves oral insulin bioavailability as it supplies a steady environment for the encased drug.[2] Polymeric NPs are classified into two types. Nanospheres in which drug is distributed in the polymer matrix, and nanocapsules in which drug core is enclosed by a polymeric film.

Table 1 shows different polymers used for the preparation of polymeric NP.[12,18]

Huang *et al.* have reported preparation of novel self-assembled NPs. The core of NPs was made up of insulin and trimethyl chitosan (TMC) coated by hydrophilic coating of N-(2-hydroxypropyl) methacrylamide copolymer (pHPMA) derivative. pHPMA coating opens the tight junction of epithelial cells improving the absorption of NPs. As the NPs penetrates through mucus, the pHPMA molecules start detaching from NPs, and the TMC NP core further undergoes paracellular absorption.[25] In one study, pellets of polyethylene imine containing insulin core were obtained by extrusion and spheronization. These were further coated with 3 overlapping layers of hydroalcoholic solutions of Methocel and Eudragit along with a gastro-resistant film. These coatings were responsible for delayed release and gastro-resistant behavior.[9]

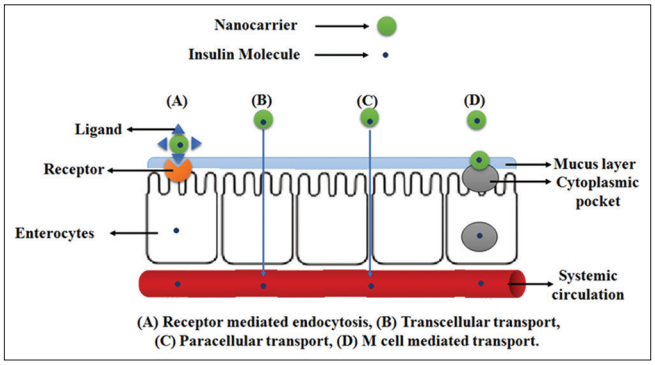

Figure 2: Mechanism of absorption of NPs

Solid lipid nanoparticles (SLNs)

One of the important colloidal carriers is SLNs, composed of physiological lipid, usually dispersed in an aqueous surfactant solution. SLN offers some advantages like nano size range and comparatively narrow size distribution, controlled release of drug over a long period of time, protection of drug against chemical degradation, nontoxic, relatively cheaper and stable, can be easily freeze or spray dried. Sarmento *et al*. have reported insulin-loaded SLNs coated with chitosan which showed benefits of mucoadhesive and absorption enhancing properties ensuring significantly lowered blood glucose up to 24 h. The chemical degradation of insulin in GIT was avoided by solid matrix of SLN whereas intestinal absorption of insulin was enhanced by chitosan coating.[26,27]

Liposomes

Liposomes are small concentric bilayer vesicles that can be formed from cholesterol and nontoxic phospholipids. Liposomes offer several advantages like nanosize, able to incorporate both hydrophilic and hydrophobic drug, improved effectiveness and therapeutic index of the drug, better stability by encapsulation, non-hazardous, compatible in biological environment, biodegradable, and nonantigenic. Liposomes are classified according to the size and number of bilayers as unilamellar vesicles (large unilamellar vesicles and small unilamellar vesicles) and multilamellar vesicles.[28]

It has been studied that biotinylated liposomes (BLPs) enhance the delivery of insulin. Biotinylation was obtained by including biotin-conjugated phospholipids into the liposome membranes. Blood glucose level was remarkably lowered, and enhanced absorption was observed. Physical stability of liposomes was increased due to biotinylation. The enhanced absorption of BLPs was confirmed because of increased cellular uptake and quick GIT.[29]

MEs

MEs are colloidal systems which are stable, isotropic, and transparent in nature and are composed of surfactant molecules organized with their hydrophilic head facing aqueous phase and their hydrophobic tails positioned toward organic phase. MEs form spontaneously above the critical micelle concentration (CMC) of the surfactant in which it forms molecular structures according to the aqueous phase content, an organic phase and the surfactant used. Various reasons are attributed for increased insulin bioavailability using MEs such as resistance offered by MEs against enzymes

Table 1: Polymers utilized for preparation of polymeric NPs[12]

Polymer	Method of preparation	Description
PLGA	Multiple emulsions, solvent evaporation technique	Aliphatic polyester. Blood glucose level decreases for up to 24 h. Complexation with sodium lauryl sulfate facilitates loading efficiency of insulin. Encapsulation efficiency of insulin reached up to 90%
PLA (polylactides)	Solvent evaporation technique	More hydrophobic than PLGA. Due to its crystalline nature, it degrades more slowly
PCL	Double emulsion method	Polyester polymer. It has viscoelastic properties and possesses easy formability. Generating less acidic environment during degradation
Poly (acrylic acid)	Complex coacervation method	Non-degradable polymers with mucoadhesive properties based on acrylic or methacrylic acid. Excellent binding efficiency on mucin and shows pH-dependent release of drug
Dextran	Prepared by polyelectrolyte complexation of oppositely charged natural polymers	Dextran sulfate is an exocellular bacterial polysaccharide. It is pH-sensitive. Vitamin B12 coated NP is used as a carrier for the oral delivery of insulin
Alginate	Ionotropic pre-gelation followed by polyelectrolyte complexation	Polysaccharide obtained from marine brown algae. It is non-toxic and biodegradable polymer
Chitosan	Ionotropic gelation with tripolyphosphate	Weak poly base. Available in high and low molecular weight chitosan. HPMCP coated chitosan NP protects insulin from harsh GIT

HPMCP: Hydroxypropylmethylcellulose phthalate, GIT: Gastrointestinal tract, PLGA: Poly lactic-co-glycolic acid, PCL: Poly-ε caprolactone

in stomach, increased retention in gut along with tremendous penetration properties across GI mucosa.

Insulin-loaded MEs were prepared by Ravi Kumar *et al.* using a low shear reverse micelle method where didodecyldimethylammonium bromide, propylene glycol, triacetin (TA), and insulin solution employed as surfactant, cosurfactant, oil phase, and aqueous phase, respectively. On the basis of multiple cloud point titrations, a ternary phase diagram was developed to emphasize the reverse micelle region. It was reported that droplet size was found to be 161.7 nm and there was a 10-fold increase in bioavailability of insulin MEs in contrast to plain solution of insulin.[30,31]

SNEDDSs

SNEDDSs are thermodynamically stable and isotropic mixture of oil, surfactant, and cosurfactant and when mixed with water, it will instantly form an O/W nanoemulsion with a range of 50 nm. This nano size is helpful in efficient absorption of the oil droplets.[32] This is a preconcentrate or anhydrous form of the nanoemulsion which can be dispensed in hard gelatin capsules. Dissolution of poorly water soluble drugs can be increased using SNEDDS leading to the development of solubilized phases from which absorption might take place.

Combination of SNEDDSs and multifunctional polymer, thiolated chitosan, has been utilized to improve oral insulin bioavailability by Shao *et al.* Insulin when incorporated into this lipid-based formulation was stable up to 4 weeks at 4°C. This was promising application for oral insulin delivery.[33]

Micelles

Micelles are self-assembled to nanosized aggregates formed by amphiphilic copolymers above the CMC. The core of micelles consists of hydrophobic moiety whereas corona consists of hydrophilic moiety. Endocytosis is the common mechanism for delivery of insulin through micelles, and this is affected by surface properties of nanocarriers. The glucose responsive micelles are developed by the complexation of a phenylboronic acid-containing block copolymer (e.g., poly [-ethylene glycol]-b-poly [aspartic acid-co-aspartamido phenyl boronic acid]) and a glycopolymer (e.g., poly [aspartic acid co-aspartglucosamine]). This complex offer some advantages such as formation of clump can be eliminated because of the poly (ethylene glycol) shell, increased sensitivity to the glucose level, and fast response to change in glucose concentration at the physiological pH.[24]

NGs

NGs are nanosized particles obtained by cross-linking polymeric networks that swell in a good solvent. Nanogels are usually 10 to 100 nm in diameter. C. Feng, and X. Chen *et al.* reported the preparation of CMCS (carboxymethyl chitosan)/CS (chitosan) nanosgel of insulin using the simple ionic gelation method. Solutions of CMCS (1 mg/mL,

pH 7.2) in water, CS (1 mg/mL, pH 4.5) in acetic acid, and insulin (1 mg/mL, pH 2) in hydrochloric acid were prepared. Aqueous solution of CMCS was premixed with insulin solution under stirring for 10 min. This insulin/CMCS mixture was added into CS solution at weight ratios of 1:4:5 (insulin: CMCS: CS) to obtain insulin: CMCS/CS-NGs (+) and 1:5:4 (insulin: CMCS: CS) to obtain insulin: CMCS/CS-NGs (−). Both positive and negative insulin: CMCS/CS-NGs have nearly the similar effect on transepithelial electrical resistance (TEER) of Caco-2 cell monolayers. However, insulin: CMCS/CS-NGs (−) exhibited a higher mucoadhesion as well as better intestinal permeability than insulin: CMCS/CS-NGs (+) in *ex vivo* intestinal studies. It was concluded that CS-based nano gels containing negative insulin have more potential to be used as a non-invasive substitute to replace the injection administration of insulin preparation.[34]

Microspheres

Microspheres are small, spherical, free-flowing powder comprising protein molecules or polymers with particle size ranging from 1 to 1000 µm. Microspheres improve oral delivery of proteins by providing protection against proteolysis, preferable crossing of intestinal mucosa and alteration of tissular distribution. In one study it is reported that administration of isobutyl 2-cynoacrylate microspheres (250-300 nm) at a dose of 100 IU/kg of insulin directly into the duodenum, jejunum, ileum, and colon led to significant decrease in serum glucose level. Reduction in blood glucose levels was reported as 65%, 50%, 50%, and 30% in ileum, duodenum, jejunum, and colon, respectively. On oral administration, this effect was initiated after 2 days and lasted up to 20 days.[35]

Niosomes

Niosomes are extensions of liposomes, in which the drug is encapsulated in a vesicle which is composed of a bilayer of non-ionic surfactant and cholesterol. Encapsulation of insulin in niosomes protects insulin against proteolytic enzymes thus improving its oral bioavailability. Insulin was entrapped in niosomes with composition of Brij 52, Brij 92, Span 60, and cholesterol. Insulin release was measured in simulated intestinal fluid and simulated gastric fluid. The protection given to entrapped insulin was measured against pepsin, α-chymotrypsin and trypsin in comparison with free insulin solution. The rate and degree of insulin release from Brij 92 and Span 60 niosomes were lower than that of Brij 52 niosomes ($P < 0.05$). Insulin gets protected when it is in niosome form as compared with free insulin solution against proteolytic enzymes ($P < 0.05$). Oral administration of Brij 92 niosome-encapsulated insulin (100 IU/kg) to animals showed significant decrease in blood glucose levels and high serum insulin levels were recorded. This proved that niosomes are suitable carrier as it enhances the bioavailability by avoiding enzymatic degradation.[36]

SPHs

SPH is a 3D network formed due to a hydrophilic polymer which shows the presence of interconnected pores which helps in absorption of more amount of water in short period of time. SPH when used as drug carriers, swell and remain in a place for a long time, releasing almost all loaded drugs. SPH facilitates protection against degradation by enzymes and exhibits swelling and deswelling mechanisms in various pH environments that control the insulin release.

SPHs are containing poly (acrylic acid-co-acrylamide)/O-carboxymethyl chitosan (O-CMC) interpenetrating polymer networks (SPH-IPNs) were prepared and tested for their probable effectiveness. The release of insulin from SPH-IPNs was pH and ionic strength dependent. As their ability to bind Ca^{2+} and to entrap enzymes, SPH-IPNs partially inactivate the enzymes such as trypsin and chymotrypsin. SPH-IPN with higher O-CMC/monomer ratio observed to be more potent. These swollen complexes adhere to the intestinal wall, giving enhanced retention properties. Insulin release was complete and rapid in a neutral medium than acidic and fast insulin release was occurred at the ionic strength of 0.1 M.

Insulin transport across rat intestine was improved around 2-3 fold after application of the SPH-IPN. Insulin-loaded SPH-IPN exhibited significant hypoglycemic effects achieving 4.1% bioavailability compared to SC injection of insulin. These prominent properties established that the SPH-IPN would be a promising carrier for oral insulin delivery.[37]

Miscellaneous

Several other novel systems are developed for oral insulin delivery such as layered double hydroxides,[38,39] carbon nanospheres,[40,41] and cyclodextrin complexed insulin.[42,43]

Oral insulin formulations in clinics

About 12 companies that are working on oral insulin formulations were identified. From that, some are on pre-clinical trials, and some of them reached to clinical trials. Following table gives the list of clinically tested oral insulin formulations [Table 2].

CONCLUSION

Oral insulin delivery is a promising approach because of enhanced disease management and improvement in patient compliance. A major problem in the oral administration of insulin is the enzymatic degradation, as well as the low intestinal permeability and consequently low oral bioavailability. Although development of novel formulations for oral insulin delivery is more challenging, it is still possible through nanotechnology. It has proved to be an emerging tool

Table 2: List of clinically tested oral insulin formulations[44]

Company	Name	Product	Action	Development phase
Biocon/Bristol-Myers Squibb	IN-105	Conjugated insulin	Short	II
Access Pharmaceuticals, Inc.	CobOral™	Insulin coated insulin-loaded nanoparticles	Short	Preclinical
Aphios Corporation	APH-0907	Nanoencapsulated insulin/ biodegradable polymer nanospheres	Short	Preclinical
Diabetology Ltd	Capsulin™ OAD	Insulin with delivery system Axcess™	Short	II
Diasome Pharmaceuticals, Inc.	HDV-Insulin	Hepatic-directed vesicle-insulin (nanocarrier)	Short	III
Emisphere Technologies, Inc.	Eligen® insulin	Insulin with chemical delivery agents (Eligen®)	Short	I
Jordanian Pharmaceutical Manufacturing Co. PLC	JPM oral insulin	Liquid delivery system with insulin-chitosan nanoparticles		I
Novo Nordisk A/S	NN1952	Insulin analog with an oral delivery system GIPET®	Short	
	OI338GT (NN1953)	Insulin analog with oral delivery system GIPET®	Long	I
	OI362GT (NN1954)	Insulin analog with oral delivery system GIPET®	Long	I
	OI287GT (NN1956)	Insulin analog with oral delivery system GIPET®		I
Oramed, Inc.	ORMD-0801	Insulin with protein oral delivery system POD™	Short	II
Oshadi Drug Administration Ltd.	Oshadi Icp.	Insulin, proinsulin, and C-peptide in Oshadi carrier	Short	II
NOD Pharmaceuticals, Inc./ Shanghai Biolaxy, Inc.	Nodlin	Insulin with bioadhesive nanoencapsulation (NOD Tech)	Intermediate	II
Transgene Biotek Ltd.	TBL1002OI	Proprietary nanotechnology Trabi-Oral™	Short	Preclinical

for developing the oral formulations of insulin. For further improvement, investigators should focus on developing simpler, safer and reliable methods for oral delivery of insulin. It is possible that these systems for insulin delivery could replace the traditional SC injections. The dream of oral insulin delivery will turn into real in the future with these efforts.

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