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Review Article

LONG-AWAITED DREAM OF ORAL INSULIN: WHERE DID WE REACH?

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ABSTRACT

Exogenous insulin administration has been a revolution in the management of diabetes mellitus, but also carried with it the distress of daily subcutaneous injection and its complications. Insulin tablet or capsule has remained a long-awaited dream for the mankind. Exhaustive worldwide efforts in search of an effective delivery system for oral insulin have brought us quite closer to the dream. This journey has been interesting due to novel concepts of liposomes, microspheres, oral strip, and oral spray. Incorporation of insulin into mucoadhesive polymer-based nanoparticles has been proving to be one of the most promising approaches for successful oral insulin delivery. Translating the basic science technology to the commercial application has also been challenging, and the task has been accomplished to some extent to date. The oncoming years shall be interesting in the form of more oral insulin preparations actually becoming available and affordable to those thousands who are awaiting a relief from the stress and distress of daily insulin injections.

Key words: delivery system, diabetes, mucoadhesive, nanoparticle, oral insulin, polymer

INTRODUCTION

Subcutaneous injection of insulin is associated with inherent difficulties, distress, and complications. The scientists have been in search of alternate routes of insulin administration including inhalation. Oral route of administration obviously has remained the most exciting option. This involves two obstacles. One, to be able to formulate insulin in such a way that it would cross the barriers and reach its destination for effective function, and two, translating this technology to industrial success for designing an oral insulin preparation to meet the goals of treatment. The first oral insulin preparations were tried with poor results, by Joslin in 1920s. Since then the scientists have worked hard and found numerous ways for incorporation of insulin with various substances and in different forms so as to overcome the barriers of degradation and permeation[1].

Nanoparticles, microparticles, microspheres, liposomes, hydrogel, buccal films, oral strips, capsules, tablets, and film patches are designed to deliver insulin orally. They are largely formulated with substances like polymers, mucoadhesives, protease inhibitors, insulin aggregation inhibitors, and functional excipients to induce transcellular, paracellular, Peyer patch-mediated or receptor-mediated transport of insulin in gastrointestinal tract. Superporous matrix, intestinal patches, and charged-coupled micromagnet microparticles are some of the recent formulation strategies to promote oral insulin absorption. Overview of various strategies applied in the design of oral insulin delivery denotes the significance of mucoadhesiveness, whereby prolonged retention of dosage form in intestinal tract translates to cumulative insulin release and absorption, to overcome the intestinal transport capacity limit. Synthesis and use of mucoadhesive excipients, chemical modification of insulin to promote its physicochemical and biological stability for encapsulation in dosage form with prolonged retention characteristics, and identification of potential insulin adjuncts have been some key factors which accelerate the speed of obtaining a functional oral insulin delivery system[2]. Many researchers have made attempts to look for possible advantages or demerits of oral insulin. Various formulations of oral insulin are today under different stages of development. The exciting process of oral insulin search has inspired the purpose of this article to take note of remarkable efforts by scientists to bring to reality the dream of oral insulin delivery.

SEARCH FOR EFFECTS OR ADVANTAGES ORAL INSULIN

Effects of oral insulin were explored in relation to various parameters, and the efforts still continue to look for the advantages of using oral insulin.

Beta cell protection and rest

Mesiha and El-Bitar studied the hypoglycemic effect of oral insulin by using preparations containing Brij 35, 52, 58 or 92 and stearic acid[3]. Oral administration of insulin to female nonobese diabetic mice was shown to inhibit diabetes and insulitis on histopathological islet study. Immunohistochemistry showed oral insulin to induce the expression of Fas ligand on islets of Langerhans and to play a role in protecting pancreatic beta cells from autoimmune destruction[4]. It was shown that oral insulin is likely to provide an opportunity for beta cell rest, and may improve the beta cell function[5].

Avoiding weight gain

Oral insulin is able to achieve a high porto-systemic gradient, as it is delivered to the liver from the gastrointestinal tract. This reduces systemic insulin exposure, and hence it was suggested that it may obviate the excessive weight gain sometimes seen with subcutaneous insulin[6].

Correction of blunting of first-phase release

It was reported that oral insulin may also be able to correct the blunting of first-phase release of insulin, which is difficult with conventional subcutaneous insulin[6,7].

Early onset, early peak, and short duration with oral insulin spray

A randomized, five-way, cross-over study was conducted with seven healthy volunteers involving assessment under euglycemic clamp and receiving four different doses of oral spray and one dose of subcutaneous regular insulin to evaluate the pharmacodynamic and pharmacokinetic properties and the dose-ranging effects of an oral insulin spray in comparison with subcutaneous regular insulin. Time to maximum insulin concentration was shorter for oral insulin than for subcutaneous insulin. Maximum serum insulin levels were comparable between the subcutaneous and 20 puffs of oral insulin, and the area under curve proved a dose-response relationship for the three doses of oral insulin. Oral insulin had an earlier onset of action, earlier peak, and a shorter duration of action compared with subcutaneous insulin. A dose-response relationship was noted between the metabolic effect and absorption of oral insulin spray[8,9].

Does it have a role in diabetes prevention?

It was suggested that induction of oral tolerance or immune modulating effect produced by oral delivery of insulin is likely to help in prevention of diabetes[10]. However, when effect of oral insulin in relatives of patients with type 1 diabetes was studied, it was found that the oral insulin did not delay or prevent type 1 diabetes[11].

Effect of oral insulin on insulin autoantibody (IAA) levels was studied in 372 relatives of subjects with type 1 diabetes who were positive for IAA, and was compared with placebo. The results suggested that IAA levels over time were not influenced by oral insulin in subjects already positive for IAA at the start of treatment[12]. Long-term intervention effects of oral insulin on the development of type 1 diabetes were evaluated. Among individuals meeting the original criteria for insulin autoantibodies (IAAs), the overall benefit of oral insulin remained significant. However, once the therapy was stopped, the rate of developing diabetes in the oral insulin group increased to a rate similar to that in the placebo group[13]. With all these findings, the question mark still remains for the possible role of oral insulin in delaying diabetes.

BARRIERS TO ORAL INSULIN DELIVERY

The gastrointestinal tract has physiological barriers which prevent optimal delivery of oral insulin. Physiological function of gut enzymes is to break large "active" proteins into smaller "inactive" amino acids so that they can overcome the second absorption barrier of "tight epithelium" in the gastrointestinal tract. These two essential barriers have been created by Mother Nature to prevent the body from potentially dangerous proteins. Researchers have been trying to selectively break this natural defense mechanism so that helpful large "active protein" drugs can cross this barrier and produce desired pharmacological effects[14].

Enzymes such as pepsin, trypsin, chymotrypsin, carboxypeptidase, and pancreatin break the proteins such as insulin into amino acids with great efficiency[15]. The insulin that survives faces a barrier to absorption as well. The intestine has tightly bound columnar cells with hydrophobic proteins called occludins, and a thick layer of mucin, and these two prevent absorption of insulin[16, 17].

Further to these barriers the absorbed insulin has to go through the first pass metabolism at liver before reaching the peripheral sites of action (unlike the subcutaneously administered insulin which directly reaches peripheral sites). The direct entry of insulin into liver has been postulated as also being of physiological advantage by several authors; however, long-terms effects of this phenomenon are not known[15, 18].

APPROACHES TO ORAL DELIVERY OF INSULIN

One of the most fascinating approaches for insulin delivery is the development of nanoparticles that enhance oral bioavailability by facilitating insulin uptake via transcellular or paracellular pathways[19]. Various approaches employed by the researchers are inter-related and overlapping; however, they are described below under suitable headings for better understanding of the minor detail associated with each approach.

Carriers, permeation or absorption enhancers, and protease inhibitors

Obviously, oral insulin needs to be attached to substances that will carry it across the mucous membrane, help permeation or facilitate absorption, and prevent breakdown. One of the most exciting options has been tried in various studies. In these, the oral insulin was combined with 400 mg of a drug carrier molecule called monosodium N-(4-chlorosalicyloyl)-4-aminobutyrate (4-CNAB), which binds noncovalently to insulin, and improves gastrointestinal absorption[20, 21, and 22]. A single center open label, randomized, two period cross over, isoglycemic glucose clamp study was performed on 10 male insulin-naive patients with type 2 diabetes who received 300 units of oral insulin and 15 units of human regular insulin on two separate days. Oral insulin led to early enhanced pharmacokinetic and pharmacodynamic responses with a higher and faster peak of plasma insulin concentration as compared to subcutaneous insulin[22]. Rapid and short duration of action of oral insulin was demonstrated by this approach.

A similar randomized open label, cross over study has been performed in 16 patients with type 2 diabetes, using 150 and 300 U oral insulin (Capsulin) and 12 IU regular insulin. The hypoglycemic effect of 150 and 300 U of this oral insulin was similar to each other, and was 50% of subcutaneous insulin[23, 24]. This oral insulin is packaged in an enteric-coated capsule, which dissolves in jejunum, and contains excipients that enhance its absorption. Permeation enhancers like Zonula occludens toxin (ZOT)[25], monosodium N-(4-chlorosalicyloyl)-4-aminobutyrate (4-CNAB)[22], and fatty acid salts[21,26] have been studied. But it was also suggested that these permeation enhancers may lead to local inflammation, and may predispose to gastrointestinal infections.

Insulin has also been co-administered with protease inhibitors such as bacitracin, sodium glycocholate, and camostat mesilate[27], which improved the absorption rate. However, the long-term effects of this approach are not known. As per John Raymond Murlin, the physiologist at the University of Rochester, insulin could be compounded in tablet form with hexylresorcinol. Hexylresorcinol neutralizes pepsin and acid, and emulsifies fat, and thus helps insulin absorption[28].

PEGlyation

Insulin can also be modified by PEGylation (adding polyethylene glycol), which changes the pharmacokinetics, prevents enzymatic degradation, and increases absorption[29].

Liposomal drug delivery system

There have been attempts of developing liposomal drug delivery systems containing glycocholate as an enzyme inhibitor and permeation enhancer for oral insulin delivery. In one of the studies, liposomes containing sodium glycocholate were prepared by a reversed-phase evaporation method followed by homogenization. The particle size and entrapment efficiency of recombinant human insulin (rhINS)-loaded sodium glycocholate liposomes was adjusted by tuning the homogenization parameters, phospholipid:sodium glycocholate ratio, insulin:phospholipid ratio, water:ether volume ratio, interior water phase pH, and the hydration buffer pH. The optimal formulation showed an insulin entrapment efficiency of 30% ± 2% and a particle size of 154 ± 18 nm. A conformational study by circular dichroism spectroscopy and a bioactivity study confirmed the preserved integrity of rhINS against preparative stress. Transmission electron micrographs revealed a nearly spherical and deformed structure with discernable lamella for sodium glycocholate liposomes. Sodium glycocholate liposomes showed better protection of insulin against enzymatic degradation by pepsin, trypsin, and alpha chymotrypsin than liposomes containing the bile salt counterparts of sodium taurocholate and sodium deoxycholate[30].

Liposomes with hepatic directed vesicles (HDV)

Another novel delivery of oral insulin was tried with the help of liposomes loaded with insulin, using hepatic directed vesicles (HDV-1). These were liposomes with diameter less than 150 nm, and contained insulin-attached specific proprietary hepatocyte-targeting molecule (HTM). It was shown that HDV-1 was stable at low pH in blood, was able to avoid enzymatic degradation, and had high biopotency (exhibited by its low dose of 5 U)[14,31]. Studies have been performed in type 1 and type 2 diabetes, with larger studies in progress; however, the metabolic control was suggested to be poor with oral insulin as compared to subcutaneous insulin.

Influence of polymer architecture on insulin transport

Nanotechnology with use of polymers has been one of the most novel approaches. Formulating insulin in nanoparticles has been shown to increase its cellular uptake and transport across Caco-2 cells. This was done with novel amphiphilic polyeletrolyte-insulin nanocomplexes, thus suggesting the influence of polymer architecture on insulin uptake and transport. Polyallylamine (PAA) (15 kDa) (kDa means Dalton) was grafted with palmitoyl chains (Pa) and subsequently modified with quaternary ammonium moieties (QPa). These two amphiphilic polyelectrolytes (APs) were tagged with rhodamine, and their uptake by Caco-2 cells or their polyelectrolyte complexes (PECs) with fluorescein isothiocyanate-insulin (FITC-insulin) uptake was investigated using fluorescence microscopy. Integrity of the monolayer was determined by measurement of transepithelial electrical resistance (TEER), and the insulin transport across the monolayers was determined. It was

found that palmitoyl chains (Pa) and insulin were co-localized in cell membranes, while quaternary ammonium (QPa) complexes were found within the cytoplasm. Both polymers opened tight junctions reversibly, and the insulin transport through monolayers increased when QPa or Pa was used[32].

Insulin polymeric capsules

Polymeric capsules were designed by researchers of Chemical Faculty, Lomonosov Moscow State University to protect insulin from destructive effects of digestive juices and preserve its functional ability[28]. The polymeric capsules are acid stable and gradually excrete insulin in a neutral medium. The two polymers used are positive protamine and negative dextran sulfate. They form layers in series one upon the other according to the "plus towards minus" principle and make a multilayer covering around the insulin filling, which makes up to 85% of the entire microparticle. Insulin covered by protective capsule is stable in acidic medium of pH from 1.7 to 5 units. When pH increases to a level above 5 units, insulin gets released. Further pH increase of up to 8 units results in accelerated protein release rate. This happens because at pH higher than 5.5 units, insulin acquires a negative charge and its bond with the negatively charged polymer of the first layer dextran sulfate gets destroyed. Such pH dependence of protective polymeric capsules provides fundamental capability to create insulin in pills.

Encapsulation in nanoparticles by mucoadhesive polymers

Insulin has been encapsulated in nanoparticles by mucoadhesive polymers such as chitosan, poly-lactic-co-glycolic acid (PLGA), and alginate[33,34,35,36,37,38], which prevent enzymatic degradation, and allow absorption across epithelial layer in Peyer patches[39]. This approach depends on absorption of insulin in the colon, which is too delayed to correct first phase insulin secretion deficiency. Cyclodextrin-complexed insulin-encapsulated mucoadhesive nanoparticles also were found to be good candidates for oral insulin delivery. In this study, hydroxypropyl beta cyclodextrin-insulin (HPbetaCD-1) complex-encapsulated polymethacrylic acid-chitosan-polyether (polyethylene glycol-polypropylene glycol copolymer) (PMCP) nanoparticles were used[40].

Poly (N-vinylcaprolactam-co-methacrylic acid) hydrogel microparticles

Oral delivery of insulin was evaluated with pH-sensitive co-polymeric hydrogels prepared from N-vinylcaprolactam and methacrylic acid monomers by free radical polymerization, and was found to offer 52% encapsulation efficiency. The *in vitro* experiments performed on insulin-loaded microparticles in pH 1.2 media (stomach condition) demonstrated no release of insulin in the first 2 hours, but almost 100% insulin was released in pH 7.4 media (intestinal condition) in 6 hours. The carrier was characterized by Fourier transform infrared, differential scanning calorimeter, thermogravimetry, and nuclear magnetic resonance techniques to confirm the formation of co-polymer, while scanning electron microscopy was used to assess the morphology of hydrogel microparticles. The *in vivo* experiments in alloxan-induced diabetic rats showed the biological inhibition up to 50% and glucose tolerance tests exhibited 44% inhibition. The formulations of this study were suggested to be promising carriers for oral delivery of insulin[41]. Another work comprised of preparing enteric microspheres (EMS) of insulin, using hydroxy propyl methyl cellulose acetate succinate as enteric polymer. Bacitracin was used as a protease inhibitor and sodium oleate as an absorption enhancer. The *in vitro* drug release studies determined that almost no drug was released in hydrochloric acid (pH 1.2) for 2 hours and then maximum amount of drug was released within 70 minutes in phosphate buffer (pH 7.4). The *in vivo* studies on male wistar rats confirmed a remarkable decrease in blood glucose level after 2 hours of administration of insulin EMS[42].

Glutamine conjugated chitosan (GC) microparticles

Chitosan at physiological pH lacks positive charge, which affects the mucoadhesiveness and permeation enhancing capacity. Therefore glutamine conjugated chitosan (GC) was developed to enhance the protonation of chitosan at intestinal pH. Particles were prepared by sodium tripolyphosphate ionic crosslinking and were evaluated *in vitro* for its application toward oral insulin delivery. The particles had high positive charge of 35.6 ± 7.3 mV at physiological pH and a size of 4.434 µm. The mucoadhesive capacity was established *in vitro* using rat intestinal tissue. Transepithelial electrical resistance (TEER) and confocal microscopy studies proved the ability of particles in opening the tight junctions in Caco-2 monolayers. The permeation of fluorescent dextran (FD4) (molecular weight 4000) across intestinal tissue was evaluated using Franz diffusion apparatus. It was observed that the GC particles enhanced the permeation by 1.52 fold in comparison with native chitosan (NC) particles[43].

Solid lipid nanoparticles (SLN)

Cetyl palmitate-based solid lipid nanoparticles (SLN) containing insulin were prepared, and the potential of these colloidal carriers was evaluated for oral administration of insulin to diabetic rats. Solid lipid nanoparticles were prepared by a modified solvent emulsification evaporation method based on w/o/w double emulsion. The particle size and zeta potential of unloaded and insulin-loaded SLN were found to be around 350 nm and negatively charged respectively. The insulin association efficiency was found to be over 43%. After oral administration of insulin-loaded SLN to diabetic rats, a considerable hypoglycemic effect was observed during 24 hours. These results demonstrated that SLN promoted the oral absorption of insulin[44].

Eudragit microspheres for pH-sensitive oral insulin delivery systems

Eudragit L100, Eudragit RS100, and their blend systems with pH-sensitive microspheres were prepared by double emulsion-solvent evaporation technique for oral delivery of insulin. Of the three systems developed, Eudragit L100 was chosen for preclinical studies. Insulin was encapsulated, and *in vitro* experiments performed on insulin-loaded microspheres in pH 1.2 media did not release insulin during the first 2 hours, but maximum insulin was released in pH 7.4 buffer media from 4 to 6 hours. The microspheres were characterized by scanning electron microscopy to understand particle size, shape, and surface morphology. The size of microspheres ranged between 1 and 40 µm. Circular dichroism spectra indicated the structural integrity of insulin during encapsulation as well as after its release in pH 7.4 buffer media. The *in vivo* release studies on diabetic-induced rat models exhibited maximum inhibition of up to 86%, suggesting absorption of insulin in the intestine[45].

TRANSLATING TECHNOLOGY TO REALITY

Based on the knowledge obtained on various approaches to effective oral insulin delivery, many commercial formulations of oral insulin have been undergoing extensive investigation and are under various stages of development. Translation of the scientific knowledge to successful synthesis of a commercial oral insulin preparation is also not an easy task and involves tedious steps in drug development. Some of the remarkable efforts to this direction, the possible techniques employed, the results of studies, and the current status are described here with the chronology of events in case of each of these formulations.

IN-105 (Biocon India)

This product is based on the technology developed by Nobex Corporation. It is based on HIM-2 (hexyl insulin monoconjugate 2) in which a single short-chain amphiphilic oligomer is covalently linked to the free amino acid group on the Lys-ß 29 residues of recombinant human insulin via a non-hydrolysable amide bond. This alters the physicochemical characteristics, leading to enhanced stability and resistance to intestinal degradation of ingested insulin. Oral HIM-2 is safe and reproduces the physiological pathway of insulin secreted by pancreas[28]. IN-105 based on this technology is a tablet form of oral insulin. This was shown to improve the solubility, stability, and systemic absorption[46]. Solubility was increased by PEGylation, while stability was thought to be due to steric hindrance. IN-105 has insulin receptor binding and metabolic activity similar to

that of human insulin, while exhibiting much lower (25-30%) insulin-like growth factor, receptor binding, and mitogenic activity[46].

IN-105 was studied in an open label, sequential ascending dose, multicentric study in 20 subjects with type 2 diabetes poorly controlled on stable metformin monotherapy, to assess the dose requirement of the molecule. The average maximum fall in plasma glucose levels was 18.1, 26.1, 29.0 and 30.8% after administration of 10, 15, 20 and 30 mg doses. The duration of fall of glucose increased with increasing doses. Five subjects experienced six mild episodes of hypoglycemia, all of which occurred between 30 and 60 minutes of administration of varying doses (15 to 30 mg) of IN-105. Other adverse events were hypertriglyceridemia, dizziness, and hyperhydrosis. This study suggested a dose dependent decrease in post prandial glucose. The data published by manufactures indicated that IN-105 is an oral insulin that acts rapidly, which could possibly indicate its role in control of postprandial hyperglycemia. Long term effects on hemoglobin A1c (HbA1c) reduction and beta cell function were yet to be seen. It was told to be through phase III trial in type 2 diabetes as well as phase I trial in type 1 diabetes, and was claimed to be in the most advanced developmental stage, wherein a total of 264 patients were enrolled at multiple centers across India in the double-blind placebo-controlled trial[46].

However, as far the actual progress of IN-105, the manufacturers had announced in October 2010 that this product needed to be subjected to more pre-launch trials. What could be inferred from this announcement is, until October 2010 the drug development had not reached the stage of launching. Then came an announcement from the manufacturer in January 2011 saying that during the late-stage trials in India, IN-105 did not meet its primary target of lowering HbA1c levels in patients with type 2 diabetes. The announcement also said that the patients were poorly controlled on the drug when compared to a placebo. However, it was told that the drug had met the secondary targets on efficacy and safety, and that the manufacturer was planning to start looking for a global pharmaceutical partner to continue the drug development[47, 48].

NN1952 (Novo Nordisk)

NN1952 by Novo Nordisk entered phase I studies in 2009[49]. The objectives were to study the safety, tolerability, pharmacokinetics, and pharmacodynamics in healthy subjects and subjects with type 1 and type 2 diabetes. The trial consisted of two parts. Part 1 consisted of single escalating doses of NN1952, placebo or insulin aspart to healthy volunteers. Part 2 involved single doses of NN1952 (with or without a meal), insulin aspart, and placebo to subjects with type 1 or type 2 diabetes. Some details of the study were available in May 2010[22]. It will be interesting to see the completion of studies for NN1952, and the further information is awaited.

ORMD-0801 (Oramed Pharmaceuticals)

ORMD-801 by Oramed Pharmaceuticals is another oral insulin under development, for treatment of type 1 and type 2 diabetes. The results of a single-blind, open-label, single-center, Phase IIa study in 8 male subjects with type 1 diabetes mellitus (ages 24-41 years; diabetics for 2-28 years, HbA1c 6.63-8.63%), regularly treated with no-peak insulin, were recently presented. Two capsules of ORMD-0801 (8 mg insulin each) were orally administered to fasting subjects. A standard 400-kcal meal was served at 10, 45 or 90 min thereafter. Blood samples were collected over the 6-hour post-insulin administration to monitor insulin levels. Significant increases in insulin levels were detected in 61% of the treatment sessions (T_{max} of 40-180 minutes), irrespective of timing of meals. In all cases, insulin levels returned to baseline within 45-300 minutes of peak recordings, demonstrating full clearance from the bloodstream. Maximum glucose concentration (C_{max}) was reached at an approximate 100-minute lag from start of meal (range 60-150 minutes), which in 17 out of 23 cases returned to basal levels before the end of monitoring session. No serious adverse events were recorded throughout the study. This study apparently showed some promise for control of postprandial hyperglycemia in type 1 diabetes patients[50].In May 2010, the Company reported results for the recently completed Phase IIb non-FDA clinical trial of ORMD-0801[51,52]. This was a randomized, double-blind, placebo-controlled, multi-centered 6-week study conducted in South Africa, which evaluated responses of 29 type 2 diabetes patients to ORMD-0801. Insulin-loaded or placebo capsules were administered to patients who were closely monitored throughout the 6-week study period. Safety, tolerability, and efficacy parameters were assessed. The results substantiated the safety and tolerability of ORMD-0801 and demonstrated that ORMD-0801 had a relevant clinical impact at the tested dose. The results stated clinically relevant reductions in insulin, C-peptide, fasting blood glucose and HbA1c in the ORMD-0801 cohort, when compared to the placebo; however, it was said that the complete results were not yet out.

In a recent press release, the company developing ORMD-0801 announced successful completion of an exploratory clinical trial testing the effectiveness of its oral insulin capsule in type 1 diabetes patients suffering from uncontrolled diabetes; however, the study is not yet complete[53].

Pharmfilm (MonoSol Rx and Midatech)

MonoSol Rx and Midatech Group Ltd. announced in January 2011, the positive results from preclinical studies in Rhesus monkey for oral insulin delivery, and said they hope to begin human trials in 2011. The two companies have been developing an edible insulin PharmFilm under a partnership that leverages MonoSol Rx's PharmFilm and Midatech's biocompatible nanoparticles to develop orally bioavailable formulations of peptides and drugs. The transbuccal film (oral strip) is similar in size, shape, and thickness to a postage stamp or smaller, is similar to a breath-strip, and can deliver precise doses of insulin in varying quantities. Midatech has "nanosized" the medication which delivers it in a way that is easy to swallow without water[54].

The transbuccal Pharmfilm delivery of insulin in the Rhesus monkey study represents probably the most advanced proof-of-concept to date for any investigative oral buccal insulin formulation, and confirms previously announced positive results in two porcine studies. Primates in the study were administered insulin PharmFilm transbuccally, or "inside the cheek", with data confirming delivery of an active therapeutic dose of insulin[55,56]. The company was to plan to begin human trials in Europe by late 2011.

Oral-lyn (Generex Biotechnology Corporation)

Generex Biotechnology Corporation (GNBT) (Generex) designed a liquid formulation of regular recombinant human insulin in the form of an oral spray for transbuccal delivery of insulin directly in systemic circulation, for prandial (meal-time) use. The drug is delivered to the buccal cavity as a fine spray by use of a special "rapidmist" device. This formulation has a rapid absorption (15 minutes), reaching maximum insulin concentration in blood at approximately 30 minutes, and returning to baseline in approximately 2 hours. One spray is approximately equal to 1 injectable insulin unit. It is designed for "split-dose" fashion, which means half of the dose is to be administered immediately prior to the meal and half the dose immediately after the meal. The formulation is known as "Oral-lyn", and the Generex Biotechnology Corporation (GNBT) (Generex) announced positive preliminary results from two Phase III clinical trials testing its effectiveness. The trials included 463 type 1 diabetes patients, and the other trial included 31 obese patients with impaired glucose tolerance (a symptom of prediabetes). Taken at meal-time, Oral-lyn reduced the rise in blood glucose by one third at 2 hours and one fourth at 3 hours. Oral-lyn also showed promising results on other key endpoints. The hope was expressed that Oral-lyn would help the patients with type 1 as well as type 2 diabetes, and would also help in the condition of prediabetes as an additional tool to control postprandial glucose, delay the clinical onset of type 2 diabetes, and reduce the risk for complications. It was said that the drug candidate was "not associated with serious hypoglycemia or other significant adverse events, body weight changes, or development of insulin antibodies." Oral-lyn has so far been approved in Ecuador, India, Lebanon, and Algeria, and is available to patients in the US and Canada, prior to regulatory approval, as part of special access programs[57]. By this time, it has been administered to a large number of subjects in over 30 clinical trials.

Use of Oral-lyn did not demonstrate its deposition into the lungs. It was also shown to not generate insulin antibodies, which compared it better than the inhalable insulin in terms of development of insulin resistance. Oral-lyn comprises of human insulin and GRAS (Generally Regarded as Safe by the FDA) excipients, and hence is likely to be more naturally accepted by the body, which is the likely cause of diminished insulin resistance. Insulin resistance has not been demonstrated for the type 1 subjects receiving Oral-lyn. Type 1 patients using Oral-lyn in Phase III studies demonstrated no weight gain and had on average an actual decrease in the body mass index. These results compared favorably to control subjects in the study using injectable insulin who had on average weight gain and an increase in body mass index. When compared with injectable or inhalable insulin for hypoglycemic events, Oral-lyn was observed to have a better adverse event profile[58].

CONCLUSION

Insulin incorporation into complex, multilayered, polymer-based, mucoadhesive, biodegradable, biocompatible, and acid-protected nanoparticles has been the most promising approach till date in oral insulin delivery[59]. Nanotechnology and polymer-based systems have strengthened the hopes for effective insulin delivery. The strategies devised to overcome the physiologic and morphologic barriers to oral insulin absorption include – inhibition of acidic and enzymatic degradation, enhancement of membrane permeability or widening of tight junctions, chemical modification of insulin, and the formulation of carrier systems[60]. Translation of findings to a commercial production of insulin has been accepted as a challenge by the scientists. Worldwide search for oral insulin preparations is in various stages of drug development, with at least one formulation currently making its appearance on the horizon. More formulations by different manufacturers are on the way to enter the list of oral insulin preparations. With hopes of the future availability of better oral insulin formulations, the future is also waiting with challenges of more effective and sustained insulin delivery, proof of optimum efficacy in human beings, safety and tolerability issues, patient affordability, and long-term effects of the oral insulin formulations.

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REFERENCES

1. Berger M. Oral insulin 1922-1992: The history of continuous ambition and failure. In: Berger M, Gries FA, editors. Frontiers in insulin pharmacology. Germany: Thieme Publishing Group; 1993. p. 144-8.
2. Wong TW. Design of oral insulin delivery. J Drug Target. 2010; 18(2):79-92.
3. Mesiha MS, El-Bitar HI. Hypoglycaemic effect of oral insulin preparations containing Brij 35, 52, 58 or 92 and stearic acid. Journal of Pharmacy and Pharmacology 1981; 33(1): 733-4. Article first published online: 12 April 2011. DOI: 10.1111/j.2042-7158.1981.tb13917.x
4. Wang H, Li YX, Sun Q, Yang GH. Oral administration of insulin to female nonobese diabetic mice inhibited diabetes and induced Fas ligand expression on islets of Langerhans. Chinese Med J 2000;113(5):433-36.
5. Wajchenberg BL. Beta-cell failure in diabetes and preservation by clinical treatment. Endocr Rev 2007; 28:187-218.
6. Kalra S, Kalra B, Agrawal N. Oral Insulin. Diabetology and Metabolic Syndrome 2010;2:66 DOI:10.1186/1758-5996-2-66
7. Stratton IM, Adler Al, Neil HAW, Matthews DR, Manley SE, Cull CA, et al. Association of glycemia with macrovascular and microvascular complications of type 2 diabetes (UKPDS 35): prospective observational study. BMJ 2000; 321: 405-12. DOI: 10.1136/bmj.321.7258.405
8. Cernea S, Kidron M, Wohlgelernter J, Modi P, Raz I. Dose-response relationship of oral insulin spray in healthy subjects. Diabetes Care 2005; 28(6):1353-7.
9. Cernea S, Kidron M, Wohlgelernter J, Raz I. Dose-response relationship of an oral insulin spray in six patients with type 1 diabetes: a single-center, randomized, single-blind, 5-way crossover study. Clin Ther. 2005; 27(10):1562-70.
10. Bergerot I, Arreaza GA, Cameron MJ, Burdick MD, Strieter RM, Chensue SW et al. Insulin B-chain reactive CD4+ regulatory T-cells induced by oral insulin treatment protect from type 1 diabetes by blocking the cytokine secretion and pancreatic infiltration of diabetogenic effector T-cells. Diabetes 1999; 48:1720-9.
11. Skyler JS, Krischer JP, Wolfsdorf J, Cowie C, Palmer JP, Greenbaum C, et al. Effects of oral insulin in relatives of patients with type 1 diabetes: the diabetes prevention trial-type 1. Diabetes Care 2005; 28(5):1068-76. DOI:10.2337/diacare.28.5.1068
12. Barker JM, McFann KK, Orban T. Effect of oral insulin on insulin autoantibody levels in the Diabetes Prevention Trial Type 1 oral insulin study. Diabetologica 2007; 50(8):1603-6.
13. Vehik K, Cuthbertson D, Ruhlig H, Schatz DA, Peakman M, Krischer JP. Long-Term Outcome of Individuals Treated With Oral Insulin - Diabetes Prevention Trial–Type 1 (DPT-1) oral insulin trial. Diabetes care 2011. Published online before print. May 24, 2011. DOI 10.2337/dc11-0523 Retrieved from www.Care.diabetesjournals.org
14. Heinemann L and Jacques Y. Oral insulin and buccal insulin: a critical reappraisal. J Diabetes Sci Technol 2009; 3:568-84.
15. Iyer H, Khedkar A, Verma M. Oral insulin - a review of current status. Diabetes Obes Metab 2010; 12:179-85.
16. Wang W. Oral protein delivery. J Drug Target 1996; 4:195-232.
17. Carino GP, Mathiowitz E. Oral insulin delivery. Adv Drug Deliv Rev 1999; 35:249-57.
18. Arbit E. The physiological rationale for oral insulin administration. Diabetes Technol Ther 2004; 6:510-7.
19. Ramesan RM and Sharma CP. Challenges and advances in nanoparticle-based oral insulin delivery. Expert Rev Med Devices 2009; 6(6):665-76.
20. Scarlett JA, Gray RS, Griffin J, Olefsky JM, Kolterman OG. Insulin treatment reverses the insulin resistance of type 2 diabetes mellitus. Diabetes Care 1982; 5:353-63.
21. Zhao L, Sun D, Cao F, Yin T, Wang H. Can insulin resistance be reversed by insulin therapy? *Med Hypotheses* 2009; 72:34-5.
22. Kaptiza C, Zijlstra E, Heinemann L, Castelli MC, Riley G, Heise T. Oral insulin: A comparison with subcutaneous regular human insulin in patients with type 2. Diabetes Care 2010; 33(6):1288-90.
23. Phillips J, Russel-Jones DL, Wright J, Brackenridge A, New R, Bansal G. Early evaluation of a novel oral insulin delivery system in healthy volunteers. Diabetes 2004; 53(Suppl. 2):A113.
24. Luzio S, Dunseath G, Lockett A, Broke-Smith TP, New RR, Owens D. Comparison of an oral insulin (Capsulin) and Actrapid during an isoglycemic clamp study in persons with type 2 diabetes. Diabetes 2008; 56(Suppl. 1):A10.
25. Fasano A, Uzzau S. Modulation of intestinal tight junctions zona occludens toxin permits enteral administration of insulin and other macromolecules in an animal model. J Clin Invest 1997; 99:1158-64.
26. Palin KJ, Phillips AJ, Ning A. The oral absorption of cefoxitin from oil and emulsion vehicles in rats. Int J Pharm 1986; 33:99-104.
27. Yamamoto A, Taniguchi T, Rikyuu K, Tsuji T, Fujita T, Murakami M et al. Effect of various protease inhibitors on the intestinal absorption and degradation of insulin in rats. Pharm Res 1994; 11:1496-1500.
28. Nikam KR, Pawar MG, Salunkhe KS, Bachhav DG. Recent trends in insulin drug delivery system. 2011. http://www.pharmatutor.org/articles/recent-trends-in-insulin-drug-delivery-system?utm_s... [Accessed May 28, 2011].

29. Clement S, Still JG, Kosutic G, McAllister RG. Oral insulin product hexyl-insulin monoconjugate 2 (HIM2) in type 1 diabetes mellitus: the glucose stabilization effects of HIM2. Diabetes Technol Ther 2002; 4(4):459-66.
30. Niu M, Lu Y, Hovgaard L, Wu W. Liposomes containing glycocholate as potential oral insulin delivery systems: preparation, in vitro characterization, and improved protection against enzymatic degradation. Int J Nanomedicine 2011; 6:1155-66. DOI: http://dx.doi.org/10.2147/IJN.S19917
31. Schwartz S, Geho B, Rosenberg L, Lau J. A 2-week randomized active comparator study of two HDV-Insulin routes (SC and oral) and SC human insulin in patients with type 1 diabetes. Diabetes 2008, 57(Suppl. 1):A124.
32. Thompson C, Cheng WP, Gadad P, Skene K, Smith M, Smith G, et al. Uptake and transport of novel amphiphilic polyelectrolyte-insulin nanocomplexes by Caco-2 cells--towards oral insulin. Pharm Res 2011; 28(4):886-96.
33. Aspden TJ, Mason JD, Jones NS. Chitosan as a nasal delivery system: the effect of chitosan solutions on in vitro and in vivo mucociliary transport rates in human turbinates and volunteers. J Pharm Sci 1997; 86:509-13.
34. Mathiowitz E, Jacob JS, Jong YS, Carino GP, Chickering DE, Charturvedi P, et al. Biologically erodible microspheres as potential oral drug delivery systems. Nature 1997; 386:410-4.
35. Damge C, Michael C, Aprahamian M, Couvreur P. New approach for oral administration of insulin with polyalkylcyanoacrylate nanocapsules as oral carrier. Diabetes 1988; 37:247-51.
36. Takka S, Acarturk F. Calcium alginate microparticles for oral administration. I: effect of sodium alginate type on drug release and drug entrapment efficiency. J Microencapsul: Micro Nano Carriers 1999; 16:275-90.
37. Rowsen ML, Dileep KJ, Sharma CP. Beta cyclodextrin-insulin encapsulated chitosan/alginate matrix: oral delivery system. J Appl Polym Sci 2000; 75:1089-96.
38. Dapergolas G, Gregoriadis G. Hypoglycaemic effect of liposome-entrapped insulin administered intragastrically into rats. Lancet 1976; 2:824-7.
39. Pappo J, Ermak TH. Uptake and translocation of fluorescent latex particles by rabbit Peyer's patch follicle epithelium: a quantitative model for M cell uptake. Clin Exp Immunol 1989; 76:144-8.
40. Sajeesh S and Sharma CP. Cyclodextrin-insulin complex encapsulated polymethacrylic acid based nanoparticles for oral insulin delivery. Int J Pharm 2006; 325(1-2):147-54. Epub 2006 June 18.
41. Mundargi RC, Rangaswamy V, Aminabhavi TM. Poly(N-vinylcaprolactam-co-methacrylic acid) hydrogel microparticles for oral insulin delivery. J Microencapsul 2011;28(5):384-94.
42. Jindal SK, Singh M, Goswami M. Formulation and evaluation of insulin enteric microspheres for oral drug delivery. Acta Pharmaceutica Sciencia 2009; 51: 121-7.
43. Rekha MR and Sharma CP. Glutamine-chitosan microparticles as oral insulin delivery matrix: *In vitro* characterization. J Appl Polym Sci 2011; 122:2374–82. DOI: 10.1002/app.34315
44. Sarmento B, Martins S, Ferreira D, Souto EB. Oral insulin delivery by means of solid lipid nanoparticles. Int J Nanomedicine 2007; 2(4):743-9. DOI: http://dx.doi.org/10.2147/IJN.S
45. Mundargi RC, Rangaswamy V, Aminabhavi TM. pH-sensitive oral insulin delivery systems using Eudragit microspheres. Drug Dev Ind Pharm 2011; 37(8): 977-85.
46. Khedkar A, Iyer H, Anand A, Verma M, Krishnamurthy S, Savale S, et al. A dose range finding study of novel oral insulin (IN-105) under fed conditions in type 2 diabetes mellitus subjects. Diabetes Obes Metab 2010; 12(8):659-64.
47. Biocon: oral insulin fails to meet primary target. 2011. Reuters US Edition, Jan 11, 2011 Bangalore, India http://www.reuters.com/article/2011/01/11/biocon-insulin-trials-idUSSGE70A02M20110111 [Accessed Oct 21, 2011].
48. Biocon oral insulin to see more pre-launch tests. 2010. Oct 23, 2010. http://www.business-standard. com/india/ news/biocons-oral-insulin-to-see-more-pre-launch-tests /412417/...19/07/2011 [Accessed Oct 21, 2011].
49. Novo Nordisk. A two part trial investigating NN1952 in healthy subjects and subjects with type 1 and type 2 diabetes. 2009. Retrieved from http://www.novonordisk-trials.com/Website/pdf/registry/nn19523646.pdf [Accessed on October 27, 2011].
50. Eldor R, Arbit E, Miteva Y, Freier R, Kidron M. Oral Insulin:Type 1 Diabetes (T1DM) Patient Response Upon Preprandial Administration. Poster, ADA, Orlando 2010.
51. Oral insulin company Oramed gets good Phase IIb results. 2010. Published by Globes (online), Israel Business News. http://www.globes-online.com/ on May 10, 2010. Retrieved from http://www.globes.co.il/serveen/globes/docview.asp?did=1000558541&fid=1725 [Accessed Oct 21, 2010].
52. ORMD 0801 Oral insulin capsule. May 2010. http://www.oramed.com/index.php?page=14 [Accessed Oct 21, 2011].
53. A Two Part Trial Investigating NN1952 in Healthy Subjects and Subjects With Type 1 and Type 2 Diabetes This study has been completed [http:// clinicaltrials .gov/ ct2/show/study/NCT01028404] Study NCT01028404 .
54. New Product: Oral Insulin Strip. Diabetes in control.com. Issue 552. Dec 17, 2010. http:// www.diabetesincontrol. com/articles/newproduct/10224-new-product-oral-insulin-strip [Accessed Oct 21, 2011].
55. Oral Insulin. Monosol Rx, Midatech. 2011. (Positive Results for Oral Insulin Delivery in Pre-Clinical Trials. January 4, 2011) Page 1-9. Available from: http://asweetlife.org/a-sweet-life-staff/in-the-news/insulin-pumps-in-the-news/positive-re... [Accessed on July 19, 2011].
56. Gilles Gary. Oral Insulin Research Update. 2011 About.com Type 1 diabetes. Sept 17, 2011. http://type1diabetes.about.com/b/2011/09/17/oral-insulin-research-update.htm [Accessed Oct 21, 2011].
57. Donnelly JM. 2011. Generex advances oral insulin product – July 11, 2011. Boston Medical Journal. Available at http://www.bizjournals.com/boston/news/2011/07/11 /generex-avances-oral-insulin.html 19/07/2011 [Accessed July 19, 2011].
58. Steffens R. Aug 16, 2010. Good news for Generex: Positive data on oral insulin coming in September. http://seekingalpha.com/article/220670-good-news-for-generex-positive-data-on-oral-ins... [Accessed July 24, 2011].
59. Card JW, Magnuson BA. A review of efficacy and safety of nanoparticle-based insulin delivery systems. Am J Physiol Gastrointest Liver Physiol 2011; Sep 15 [Epub Ahead of print] PMID 21921287 [Pubmed – as supplied by Publisher]. [Accessed October 28, 2011].
60. Woitiski CB, Carvalho RA, Ribeiro AJ, Neufeld RJ, Veiga F. Strategies toward the improved oral delivery of insulin nanoparticles via gastrointestinal uptake and translocation. BioDrugs. 2008; 22(4):223-37.