Glucagon-like peptide-1 and diabetes treatment

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Abstract

Type 2 diabetes is characterized by insulin resistance, impaired glucose-induced insulin secretion and inappropriately regulated glucagon secretion, which in combination eventually result in hyperglycemia and in the longer term in microvascular and macrovascular complications affecting multiple organ systems. Traditional treatment modalities — even multidrug approaches — for type 2 diabetes are often unsatisfactory at achieving glycemic goals as the disease progresses due to a steady, relentless decline in pancreatic β-cell function. Furthermore, current treatment modalities are often limited by inconvenient dosing regimens, safety and tolerability issues, the latter including hypoglycemia, body weight gain, edema and gastrointestinal side effects.

The incretin hormones glucagon-like peptide (GLP)-1 and glucose-dependent insulinotropic polypeptide (GIP) are intestinal hormones that augment insulin secretion in response to the ingestion of nutrients. The actions of GLP-1 and GIP, which also include trophic effects on the β-cells, have attracted a lot of interest. GLP-1 also inhibits glucagon secretion and suppresses food intake and appetite. Recently, an entirely new therapeutic modality for the treatment of type 2 diabetes based on the effect of GLP-1 was introduced onto the market.

Incretin-based therapies fall into two groups: (1) GLP-1 receptor agonists, i.e. injectable peptide preparations with actions similar to the natural incretin hormone GLP-1; and (2) the incretin enhancers, which are orally available agents that inhibit the degradation of the incretin hormones and thereby increase their plasma concentrations and biological actions. This review outlines the scientific basis for the development of GLP-1 receptor agonists and incretin enhancers, assesses the clinical experience gathered so far and discusses future expectations for incretin-based therapy.

Key words:
Incretin hormones, dipeptidyl peptidase-4 (DPP-4), glucagon-like peptide-1 (GLP-1), glucose-dependent insulinotropic polypeptide (GIP), incretin mimetics, GLP-1 receptor agonists, type 2 diabetes

Incretin hormones: secretion, effect and degradation

The 'incretin effect' refers to the amplification of glucose-stimulated insulin secretion elicited by hormones secreted from the gastrointestinal tract. In the strictest sense, it is quantified by comparing insulin responses to oral and intravenous glucose administration where the intravenous infusion is adjusted so as to result in the same (isoglycemic) plasma glucose concentrations as the oral stimulus [1, 2]. In healthy subjects, oral administration causes a two- to threefold larger insulin response compared with the isoglycemic intravenous stimulus.

The incretin hormones GLP-1 and GIP are intestinal hormones that augment insulin secretion in response to the ingestion of nutrients

This discrepancy in insulin secretion between the two stimuli is due to the actions of incretin hormones, glucagon-like peptide (GLP)-1 and glucose-dependent insulinotropic polypeptide (GIP). GLP-1 is a 30-amino acid polypeptide produced in the endocrine L-cells of the intestinal epithelium as a product of glucagon gene expression. The GLP-1 moiety is liberated from proglucagon by the action of prohormone convertase 1/3 (PC1/3). This is in contrast to proglucagon processing in the pancreatic α-cells, where prohormone convertase 2 cleaves out glucagon but leaves the GLP-1 molecule embedded in a large inactive fragment [3]. The L-cells are found throughout the intestinal

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tract, but their density is highest in the ileum and parts of the colon.

GIP is produced in the endocrine K-cells, which are more frequent in the proximal small intestine [3]. As in the L-cells, PC1/3 is responsible for the processing of proGIP in endocrine K-cells. After the secretion of GIP and GLP-1, both hormones are degraded by the enzyme dipeptidyl peptidase-4 (DPP-4). This enzyme cleaves off the two N-terminal amino acids of the incretin hormones, which abolishes their insulinotropic activity.

GLP-1 has an apparent half-life of 1–2 min [4], whereas GIP is degraded more slowly, with a half-life of 7 min [5]. The truncated metabolites are eliminated through the kidneys. The intact and active forms of GLP-1 and GIP elicit significant insulin responses at plasma glucose levels above 4 mM. At higher levels (e.g. in the postprandial state) this insulinotropic power increases dramatically [6]. These insulinotropic actions are exerted by activation of specific GLP-1 and GIP receptors, respectively, on the pancreatic β-cells and consist of potentiation of glucose-induced insulin secretion [7]. At lower plasma glucose concentrations (4 mM and below) both hormones lose their insulinotropic activity completely. Therefore the incretin hormones seem to play a very important role in the regulation of, particularly, postprandial glucose clearance from the bloodstream.

GLP-1 has an apparent half-life of 1–2 min, whereas GIP is degraded more slowly, with a half-life of 7 min

The incretin hormones have several additional actions besides being 'incretins'. In addition to its insulinotropic action, GLP-1 enhances insulin biosynthesis and insulin gene expression, it has trophic and protective effects on the β-cells (stimulating β-cell growth, proliferation and neogenesis, and reducing β-cell apoptosis) [8]. Furthermore, GLP-1 strongly inhibits glucagon secretion, which, combined with its effect on insulin secretion, results in an inhibition of hepatic glucose production that contributes to the glucose-lowering effect of the hormone [9]. GLP-1 also functions as an inhibitor of upper gastrointestinal motility and secretion (reducing postprandial glucose excursions) [10], suppressing appetite and food intake. Chronic administration of GLP-1 has been shown to lead to weight loss [11]. Finally, GLP-1 appears to have beneficial actions on the cardiovascular system: enhancing myocardial performance in experimental and clinical cardiac insufficiency, reducing infarct size in experimental myocardial infarction and improving endothelial dysfunction in patients with type 2 diabetes [3]. GIP appears to have similar actions to GLP-1 on the β-cells, but its effects with regard to glucagon secretion remain relatively unclear.

Incretin hormones and type 2 diabetes mellitus

When patients with type 2 diabetes are subjected to isoglycemic oral and intravenous glucose challenges, the amplification of insulin secretion during the oral stimulus is markedly reduced unlike in healthy subjects [12]. Considering the power of the incretin effect to maintain postprandial glucose levels in healthy subjects, there can be little doubt that the attenuated incretin effect in type 2 diabetes contributes to the glucose intolerance of these patients.

When trying to explain why the incretin effect is lost, one may ask whether there is something wrong with the secretion or actions of GIP and GLP-1. Mixed-meal stimulation tests have revealed that postprandial GIP secretion is near normal or slightly impaired, whereas particularly the late phase of the GLP-1 response is significantly reduced in patients with type 2 diabetes [13]. Furthermore, in early studies of the actions of the two peptides, it was clearly demonstrated that the insulinotropic effect of GLP-1 was retained, whereas that of GIP was almost completely lost [14].

Subsequent studies of the effects of GLP-1 on glucose-stimulated insulin secretion (β-cell responsiveness to glucose) revealed that it was possible to completely normalize the glucose responsiveness with GLP-1, but also that the potency of GLP-1 in this respect was reduced in type 2 diabetic patients [15]. Further clamp studies revealed that particularly the late-phase insulin response to GIP was completely lost in type 2 diabetes [16]. Thus the loss of incretin effect in type 2 diabetes seems to be due to an impaired secretion of GLP-1 in particular and, perhaps more importantly, a loss of the insulinotropic effect of GIP and reduced insulinotropic potency of GLP-1. However, since the insulinotropic effect of high doses of GLP-1 is preserved, it should be possible to restore incretin action in patients with type 2 diabetes with supraphysiological doses of GLP-1.

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GLP-1 receptor agonists

It is fairly easy to stabilize the GLP-1 molecule against DPP-4: a substitution of alanine in position 2 with, for example, valine is sufficient and does not change the biological activity of the peptide [17]. However, the stabilized molecule is still eliminated extremely rapidly in the kidneys (with a half-life of 4–5 min), which leaves such analogues unsuitable for prolonged drug exposure.

However, exendin-4, a peptide with about 50% sequence homology to GLP-1, which was isolated from the saliva of the Gila monster (*Heloderma suspectum*) during a search for biologically active peptides, turned out to be a full agonist for the GLP-1 receptor, to be stable against DPP-4, and to be eliminated through the kidneys exclusively by glomerular filtration [18]. After subcutaneous injection of exenatide (a synthetic replica of exendin-4) in the dose selected for clinical use (10 μg), the plasma concentration is elevated into the insulinotropic range for about 5–6 h. Exenatide is therefore administered twice daily [19]. Exenatide has been developed by Amylin and Eli Lilly for the treatment of type 2 diabetes under the trade name Byetta®.

Another GLP-1 receptor agonist under clinical development is liraglutide (Novo Nordisk), which is based on the structure of human GLP-1 (97% homology with the native peptide) but modified to include an amino acid substitution and an attachment of a C16 acyl chain [20], enabling the molecule to bind to albumin, thus preventing renal elimination and degradation by DPP-4. Following subcutaneous administration, liraglutide is slowly absorbed into the bloodstream and has a plasma half-life of approximately 11–13 h [21], making it suitable for once-daily injection. Clinically, the molecule has similar actions to continuously infused GLP-1 and appears to have a similar clinical potential to that of exendin-4 [22].

Clinical studies using exenatide have demonstrated sustained beneficial effects on HbA_{1c}, body weight and β-cell function in patients with type 2 diabetes. Controlled studies comparing exenatide and placebo injections as add-on therapy to already instituted antidiabetic treatment have revealed a statistically significant decline in HbA_{1c} of approximately 1% from baseline (baseline HbA_{1c} 8.2%) [23] and significant weight loss in favour of exenatide. The weight loss was progressive, dose-dependent and with no apparent plateau by week 30 (–2.3 kg); however, it appeared to plateau after 2–3 years of treatment (with a weight loss of 5.3 kg) in completers participating in an open-label extension of the trials [24, 25]. Side effects were primarily dose-dependent nausea and vomiting, occurring in as many as 57% and 17% of participants, respectively, although nausea was generally mild to moderate and declined with time.

The clinical efficacy of exenatide in patients with type 2 diabetes over a period of 6 months has been evaluated by extractions of data from a primary care electronic medical record database [26]. In this study, weight loss among the 1785 patients was >3 kg (baseline weight 121 kg) and as many as 70% of the patients lost weight. Lowering of HbA_{1c} ranged from 0.7% to 0.9% regardless of weight loss. It was concluded that the effectiveness of exenatide in a primary care setting is similar to that observed in controlled clinical trials.

Exenatide given as twice-daily injections may not provide complete 24-h coverage, especially after midday meals, and overnight plasma concentrations seem inadequate to obtain optimal glycemic control. Therefore a long-acting release (LAR) formulation of exenatide for subcutaneous injection in patients with type 2 diabetes has recently been developed. Data from a phase-II trial (n = 295) in which exenatide LAR 2.0 mg once weekly was compared with exenatide 10 μg twice daily for 30 weeks were recently published [27]. These data showed significant reductions in HbA_{1c} (1.9% and 1.5%, respectively) and body weight (average decrease of 4 kg in both groups). The trial was extended for 22 weeks with all subjects being switched to exenatide LAR in the extension period. A sustained effect of exenatide LAR was seen after 52 weeks with improvements in both HbA_{1c} (–2%) and fasting plasma glucose (–2.6 mmol/l). Three out of four patients achieved an HbA_{1c} of 7.0% or below. The change in body weight was –4 kg by week 52 compared with baseline. Exenatide LAR was well tolerated. The most common side effect, nausea, was predominantly mild and transient during the study; during the 22-week follow-up the incidence of nausea was 7% [28]. Exenatide LAR is currently in phase-III clinical development and is expected to reach the market in 2010 (*Table I*).

The next GLP-1 receptor agonist for the treatment of type 2 diabetes to reach the market is expected to be liraglutide. Published data have demonstrated that liraglutide as monotherapy (1.9 mg once daily) is capable of decreasing fasting plasma glucose levels by 3.4 mmol/l on average in patients with type 2 diabetes when compared with placebo [22]. In the same study, a decrease in HbA_{1c} of up to 1.7% (baseline

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Table I: *The GLP-1 receptor agonists currently on the market or in clinical development.*

Company	Compound	Status of development	Formulation
Eli Lilly/Amylin	Byetta® (exenatide)	Launched	Twice daily
Novo Nordisk	Liraglutide	Filed USA/EU May 2008	Once daily
Eli Lilly/Amylin	Exenatide LAR	Phase III Expected 2010	Once weekly
Sanofi-Aventis	AVE0010(ZP10)	Phase III Expected 2011	Once daily
Roche/Ipsen	Taspoglutide (R1583)	Phase IIb–III Expected 2011	Once weekly
GlaxoSmithKline	Syncria® (albiglutide)	Phase IIb	Once weekly
ConjuChem	CJC-1134	Phase II	Once weekly
Eli Lilly	LY2405319	Phase II	Once weekly
Sanofi-Aventis	AVE0010(ZP10)	Phase II	Once weekly
Novo Nordisk	NN9535	Phase II	Once weekly

HbA_{1c} 8.0%) was observed, and almost 50% of the patients managed to reach the HbA_{1c} goal of <7%. In the highest liraglutide dose group (1.9 mg/day) the change from baseline in body weight was –3 kg (–1.2 kg compared with placebo). As for exenatide, transient and mild nausea was reported in the liraglutide-treated subjects (liraglutide 10%, placebo 3%). Data from a number of phase-III trials with liraglutide were presented at the 2008 annual scientific meetings of the American Diabetes Association (ADA) and the European Association for the Study of Diabetes (EASD). These trials, called the LEAD (Liraglutide Effect and Action in Diabetes) trials, demonstrated that liraglutide given as a once-daily injection, as monotherapy and in combination with a range of antidiabetic drugs, is associated with significant improvements in HbA_{1c} (sustained reductions of up to 1.6%), fasting plasma glucose, postprandial glucose and β-cell function [29, 30]. Furthermore, in trials of up to 1 year liraglutide showed maintained weight reduction (up to 4 kg in subjects with a high BMI) [29], minimal risk of hypoglycemia, reductions of up to 3.6 mmHg in systolic blood pressure [31], low and transient incidence of nausea and negligible antibody formation.

Many GLP-1 receptor agonists that are in their late clinical development are anticipated to have optimized pharmacokinetic profiles, fewer gastrointestinal side effects and are expected to reach the market from 2010 onwards

Very recently, liraglutide was compared with exenatide twice daily (for 26 weeks) and significant differences in glycemic control were observed (HbA_{1c} –1.1% and –0.8%, respectively), a tendency towards a more pronounced weight loss with liraglutide (–3 kg vs. –2 kg) and less nausea in favour of liraglutide (Blonde L et al. *Can J Diabetes* 2008; 32 suppl: A107). Applications for liraglutide as a new drug were filed with the authorities in both the United States and the European Union in May 2008.

Regarding adverse events of GLP-1 receptor agonists, attention has recently been drawn towards a few cases of acute pancreatitis in patients treated with exenatide. These cases have been reviewed by the US Food and Drug Administration during post-marketing. A very few cases of acute pancreatitis have also been diagnosed during treatment with liraglutide in the LEAD programme. However, it is not clear whether the incidence of acute pancreatitis in patients with type 2 diabetes treated with GLP-1 receptor agonists is higher than in a type 2 diabetic population not treated with GLP-1 receptor agonists, and, so far, the reports are few and seem to be correlated to other causes of pancreatitis (hypertriglyceridemia, alcohol abuse, gall bladder stones/operation).

Many GLP-1 receptor agonists that are in their late clinical development are anticipated to have optimized pharmacokinetic profiles and presumably fewer gastrointestinal side effects. Many of these are expected to reach the market from 2010 onwards (*Table I*).

DPP-4 inhibitors

The extremely rapid and extensive degradation of GLP-1 by DPP-4 has given rise to the proposal that inhibitors of the enzyme could be used as a therapy for patients with type 2

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Table II: *The leading DPP-4 inhibitors — all orally available — in clinical development or on the market.*

Company	Compound	Status of development
Merck	Januvia® (sitagliptin, MK-0431)	Launched 2006
Novartis	Galvus® (vildagliptin, LAF-237)	Launched 2008
Takeda	Alogliptin (SYR-322)	Filed USA 2008
Bristol-Myers Squibb	Onglyza™ (saxagliptin)	Filed USA/EU 2008
Boehringer-Ingelheim	BI-1356	Phase III
Glenmark	Melogliptin (GRC 8200)	Phase IIb
Phenomix	Dutogliptin (PHX1149)	Phase IIb
Mitsubishi Tanabe Pharma	TA-6666	Phase II
Mitsubishi Tanabe Pharma	MP-513	Phase II
Amgen/Servier	AMG 222/ALS 2-0426	Phase II
Pfizer	PF 00734200	Phase II
Takeda	SYR-472	Phase II
Abbott Laboratories	ABT-279	Phase II

diabetes by protecting and thereby enhancing the circulating levels of endogenous GLP-1 [32]. Early experiments documented that administration of an inhibitor of DPP-4 to pigs completely protected both endogenous and exogenous GLP-1 and, furthermore, greatly enhanced insulin responses to glucose [33]. In a subsequent study, DPP-4 inhibition was demonstrated also to protect GIP from degradation, again resulting in enhanced insulinotropic activity of infused GIP [34]. The idea was quickly accepted by the pharmaceutical industry and numerous companies embarked on the development of DPP-4 inhibitors for clinical use in the treatment of type 2 diabetes (*Table II*).

The rapid and extensive degradation of GLP-1 by DPP-4 suggests that inhibitors of the enzyme could be used as a therapy for type 2 diabetes by enhancing the circulating levels of endogenous GLP-1, resulting in enhanced insulinotropic activity of infused GIP

The first DPP-4 inhibitor to reach the market in 2007 was sitagliptin (Merck) [35]. Vildagliptin (Novartis) [36] was launched in the European Union in spring 2008. Both inhibitors have good oral bioavailability and a relatively long duration of action, such that once-daily (sitagliptin) or twice-daily (vildagliptin) dosing gives 70–90% inhibition of plasma DPP-4 activity over a 24-h period [37], which is sufficient to fully protect the endogenous incretin hormones from degradation. Sitagliptin and vildagliptin have significant antidiabetic effects when given in monotherapy and result in further improvements in glycemic control when given in combination with other antidiabetic agents including metformin, sulfonylurea and thiazolidinediones [37]. More than 25 studies have been published, comparing treatment with sitagliptin and vildagliptin with placebo, and in a few studies with other oral agents [23]. Most studies have been of a relatively short duration (<30 weeks) and the observed reductions in HbA_{1c} approximated 0.6–0.8% compared with placebo during the first 6 months of treatment [23].

In a recent study sitagliptin was given for 26 weeks in combination with various doses of metformin after a washout of previous medication. The combination of the highest doses (100 mg sitagliptin + 2000 mg metformin) resulted in large reductions in HbA_{1c} with 66% of the patients reaching values below 7%, considered a therapeutic target by the ADA [38]. Data from the same subjects completing a 2-year extension trial demonstrated a sustained effect on glycemic control and were recently presented at the annual meeting of the EASD [39]. The combination with metformin is of particular interest, because recent studies have indicated that metformin may increase GLP-1 biosynthesis and secretion, so that a larger increase in the concentration of active GLP-1 may be obtained with the combination compared with either agent alone [40]. If the combination treatment can be demonstrated to prevent deterioration of β-cell function with time better than the currently recommended initial treatment (metformin) it may be recommended as an initial treatment for newly diagnosed patients with type 2 diabetes.

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