

Incretin Mimetics in the Treatment of Type 2 Diabetes Mellitus

a report by

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Incretins are hormones from the gut that augment the postprandial nutrient-induced insulin secretion. The incretin effect is normally quantified by comparing the insulin responses with oral and intravenous glucose administration, where the infusion is adjusted so that the plasma glucose concentrations are similar to those observed after the oral administration. The insulin response to oral glucose is normally much larger than that observed after intravenous (IV) glucose, and in healthy subjects as much as 70% of the insulin response may be attributable to the action of the incretin hormones. An illustration of the power of the incretin effect was provided by Nauck et al.,¹ who studied glucose excursions and insulin responses after increasing doses of oral glucose. It turned out that the glucose excursions were identical regardless of the amount of glucose ingested, the explanation being that incretin-induced insulin secretion was augmented progressively, thereby effectively preventing post-challenge hyperglycaemia (see *Figure 1*).

The two most important incretin hormones are glucose-dependent insulintropic polypeptide (GIP)—formerly called gastric inhibitory polypeptide—and glucagon-like peptide-1 (GLP-1). Their insulintropic effects are additive, and together the two hormones can fully explain the incretin effects.² The secretion of the incretin hormones is also proportional to the size of the ingested nutrient load.³ However, in patients with type 2 diabetes the incretin effects are severely reduced or absent⁴ and there is little doubt that the lacking incretin effect contributes significantly to the inadequate insulin secretion observed in these patients. It is therefore relevant to ask why the incretin effect is lost in type 2 diabetes. It is clear that differences in the metabolism of the GIP and GLP-1 cannot explain the deficiency.⁵ Detailed studies of meal-induced secretion of GLP-1 and GIP in the patients revealed that GIP secretion may be slightly decreased compared with healthy controls whereas GLP-1 secretion may be reduced by as much as 50% (calculated as incremental area under the curve (AUCs)),⁶ and the lower GLP-1 secretion is likely to contribute to the incretin deficiency. However, the most important finding is that the actions of both

incretin hormones are severely compromised in type 2 diabetes. Thus, the insulintropic effects of GIP as studied in hyperglycaemic clamp experiments is nearly completely lost, regardless of GIP dose.⁷ In contrast, GLP-1 is still capable of enhancing glucose-induced insulin secretion, actually to completely normal levels,^{8,9} but the amount of GLP-1 required to normalise insulin secretion is about five times higher than that required in healthy controls.¹⁰ Thus, while the diabetic beta cells are almost completely unresponsive to GIP, GLP-1 is capable of totally restoring the beta cell responsiveness to glucose, but the potency of the hormones in this respect is markedly reduced. This raises the possibility of restoring beta cell function in patients with type 2 diabetes with supraphysiological amounts of GLP-1. GIP is unlikely to be of therapeutic value, at least acutely, although GIP analogues have shown some efficacy in animal models of diabetes.¹¹ It is not known why the effect of GIP is lost in the patients, except that it seems clear that the loss is secondary to the occurrence of diabetes.¹² Therefore, it cannot be excluded that the effect of GIP can be restored with time as metabolic control is improved by other treatment. Thus, it has recently been shown that the potency of GLP-1 with respect to enhancing beta cell responsiveness to glucose can be improved by strict glycaemic control.¹³ At any rate, GLP-1 infusions have been demonstrated to completely normalise fasting glucose concentrations in patients with severe long-standing diabetes mellitus and haemoglobin A1c (HbA_{1c}) levels around 11%¹⁴ and to virtually normalise both fasting and postprandial glucose levels in patients with moderate disease.¹⁵ GLP-1 is therefore of interest as a treatment for diabetes.

However, GLP-1 has many more actions than merely enhancing glucose-induced insulin secretion.¹⁶ It also increases the biosynthesis of new insulin molecules and upregulates all the machinery required for insulin synthesis in the beta cells. Furthermore, it has been demonstrated to exert tropic actions on the beta cells in rodents. Thus, GLP-1 causes proliferation of already existing beta cells, and also promotes differentiation of new beta cells from progenitor cells in the ducts leading to the

formation of small new islets. Finally, and probably most important for humans, it strongly inhibits cytokine, lipid and glucose-induced apoptosis of beta cells including human beta cell.¹⁷ Although no data regarding this are available in humans *in vivo*, this trophic potential nevertheless holds great promise, since GLP-1 may be able to exert beta cell protective effects in our patients and possibly prevent progression of the disease.

In addition, GLP-1 inhibits glucagon secretion,¹⁴ and thereby reduces hepatic glucose production,¹⁸ an effect that is likely to contribute importantly to its glucose-lowering effects in diabetic patients. Thus, in patients with type 1 diabetes and no residual beta cell secretory capacity, GLP-1 lowers glucagon secretion and also lowers blood glucose.¹⁹ Furthermore, GLP-1 inhibits gastric motility and emptying, and thereby strongly reduces postprandial glucose excursions.^{20,21} It also significantly inhibits appetite and food intake,²² and probably acts as one of the endocrine signals from the gut that terminates meal ingestion and signals interdigestive satiation.²³ Most recently GLP-1 has also been suggested to exert protective effects on the heart and the blood vessels.^{24,25} All of these actions render GLP-1 of unusual interest in the context of diabetes treatment.

However, single subcutaneous injections of GLP-1 have little effect on blood glucose.²⁶ The explanation is that the peptide is broken down extremely rapidly in the body. The culprit is the ubiquitous enzyme, dipeptidyl-peptidase (DPP)-IV, which cleaves of the two N-terminal amino acids and renders the metabolite inactive.²⁷ This process is so rapid and extensive that just a few percent of what is injected subcutaneously survives in the intact, active form,²⁸ and leaves the peptide with a half-life in the circulation of 1–2min⁵ and a plasma clearance of two to three times the cardiac output. Clinically relevant agents GLP-1 analogues must therefore be resistant against (DPP)-IV and also exhibit less renal clearance, which is the second mechanism whereby GLP-1 is eliminated.²⁹

Proof of concept for the efficacy of GLP-1 treatment was provided by Zander et al. who administered GLP-1 by continuous subcutaneous infusion for six weeks to subjects with severe long standing diabetes. The treatment resulted in a lowering of fasting and average plasma glucose levels by 4–6mmol/l, reduction of HbA_{1c} by 1.3%, a weight loss of 2kg, a reduction of free fatty acid levels, almost a doubling of insulin sensitivity and a dramatic restoration of beta cell function.³⁰

A mere substitution of amino acid (ALA) No. 2 in the GLP-1 sequence provides resistance to DPP-

IV(31), but such analogues are still eliminated extremely rapidly by the kidneys with half lives of 3–4min. Substances with less renal clearance are therefore required. One such substance is exendin 4, a 39 amino acid peptide with 53% sequence homology to GLP-1, isolated from the saliva of the lizard, *Heloderma suspectum* (the gila monster). Exendin 4 is resistant to DPP-IV and is cleared in the kidneys exclusively by glomerular filtration (as opposed to GLP-1, which is also extracted by peritubular uptake mechanisms).³² Thereby, exendin acquires an intravenously plasma half-life of 30min,³³ but otherwise seems to share all the actions of GLP-1. There is apparently no mammalian counterpart of exendin 4, which is not the GLP-1 of the gila monster.³⁴ A subcutaneous injection of the maximally tolerated dose of exendin 4 provides an exposure in the circulation for about five hours,³⁵ and this has turned out to be sufficient for effective diabetes therapy in a twice daily administration regimen. Exendin 4 has been developed for clinical use by the Amylin Corporation (www.Amylin.com) and has patented exenatide in its synthetic form. The compound has been tested in several clinical trials, most recently in three controlled pivotal phase 3 studies comprising 1,494 patients. Exenatide was given for 30 weeks as an add-on therapy to type 2 diabetic patients inadequately treated with sulfonylureas (SU),³⁶ metformin³⁷ or a combination of metformin and SU.³⁸ After 30 weeks treatment, fasting blood glucose concentrations were significantly reduced, HbA_{1c} levels were reduced by approximately 0.8% in all groups and to or below 7% (a recommended value) in 41, 46 and 34% of the patients in the three groups. Adverse effects were mild and generally gastrointestinal. Mild hypoglycaemia was noted in 28–36% of patients also receiving SU. An important result was a significant, dose-dependent and progressive weight loss of 1.6kg (SU and SU + metformin) and 2.8kg (metformin) from baseline. In open-label extensions of these studies, exenatide has been given for a total of two years with continued effects on HbA_{1c} and body weight.³⁹ However, some patients (about 38% of patients after 30 weeks) appear to develop low titre antibodies against exenatide, and 6% developed antibodies with higher titres. In about half of these, the glucose lowering effect of exenatide appeared attenuated. Exenatide was approved by the US Food and Drug Administration (FDA) in April 2005. Information about the new drug – named Byetta – is available on the website of the company (www.byetta.com). Most recently, the Amylin Corporation has developed a slow-release formulation of exenatide.⁴⁰ Exenatide LAR (long-acting release) is a poly-lactide-glycolide microsphere suspension containing 3% exenatide that exhibits sustained dose-dependent glycaemic control in

diabetic fatty Zucker rats for up to 28 days following a single subcutaneous injection. In 30 patients with type 2 diabetes previously treated with diet/exercise and/or metformin, weekly injections of Exenatide LAR for 15 weeks was reported to reduce fasting plasma glucose by $\sim 3\text{mmol/l}$, to reduce HbA_{1c} by 1.7% and cause a weight loss of 3.8kg in the group with the highest dose.⁴¹ Notably, the enhanced efficacy was associated with a markedly reduced incidence of side effects (nausea). This supports the concept that it is essential to maintain a constant level of high GLP-1 activity for optimal treatment results. In conclusion, exenatide represents an efficacious supplement to failing conventional oral antidiabetic agents, and the sustained effect observed in the extension studies and its continued weight lowering effects must be considered very promising.

Other analogues in clinical development include modified versions of the GLP-1 molecule, which by various means, attach to albumin, and thereby acquire the pharmacokinetic profile of albumin. One such analogue is Liraglutide, produced by NovoNordisk. It consists of a slightly modified GLP-1 sequence attached a palmitoyl chain. Thereby the molecule obtains affinity for and binds to albumin and, as a result, escapes both DPP-IV and renal elimination. The plasma half-life of this compound is approximately 12 hours, and it therefore provides exposure for several days after a single injection.⁴² The compound seems to possess

all of the activities of native GLP-1.⁴³ A recent report describes treatment of 165 patients with type 2 diabetes and baseline HbA_{1c} of 8.1–8.5% for 14 weeks.⁴⁴ Subjects were washed out for four weeks and randomised to 1 of 3 doses of Liraglutide (0.65, 1.25 or 1.9mg). Liraglutide lowered fasting blood glucose and HbA_{1c} dose dependent manner (by up to 1.74% in the highest dose group) with 50% of subjects reaching values at or below 7%. Bodyweight was reduced by 3kg. Side effects were very mild. The strength of this compound seems to be its attractive pharmacokinetic profile, providing a rather stable plateau of active compound in plasma upon single daily injections. In this way, side effects (nausea, vomiting) associated with large excursions in the plasma concentration of more rapidly metabolised compounds after subcutaneous injection may be avoided. The compound does not appear to be antigenic.

In conclusion, the incretin mimetics seem to possess a spectrum of beneficial actions for patients with type 2 diabetes. Their particular virtue is their ability to produce durable improvements in glycaemic control combined with a sustained and progressive weight loss. It is so far impossible to determine their trophic potential for the beta cell in humans, but it may be that the durable glycaemic control does indeed reflect a protective action. Unfortunately, placebo-controlled studies of a sufficient duration are not yet available. ■

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