*For debate***The pathogenesis of NIDDM involves a defective expression of the GIP receptor****J.J. Holst¹, J.Gromada², M. A. Nauck³**¹ Department of Medical Physiology, University of Copenhagen, Copenhagen, Denmark² Department of Islet Cell Physiology, Novo Nordisk A/S, Symbion Science Park, Copenhagen, Denmark³ Department of Medicine, Knappschafts-Krankenhaus, Ruhr University, Bochum, Germany

Carbohydrate ingestion causes the release of two insulinotropic intestinal hormones, glucose-dependent insulinotropic polypeptide (GIP) [1] and glucagon-like peptide-1 (GLP-1) [2]. GIP is mainly released from the duodenum, and GLP-1 from the ileum, and this probably explains why the GIP response to a moderate oral glucose load is large whereas the GLP-1 response is very small [3]: the glucose is absorbed before it reaches the GLP-1 secreting cells. The augmented secretion of insulin resulting from the insulinotropic actions of the two hormones, the so-called incretin effect [4, 5], is thought to be important for maintaining a normal glucose tolerance. For GLP-1, this has recently been confirmed with the aid of a GLP-1 receptor antagonist, exendin 9–39, the administration of which greatly reduced the insulin response to intestinal glucose in rats [6, 7]. Furthermore, targeted disruption of the GLP-1 receptor in mice resulted in glucose intolerance and basal hyperglycaemia (at least in male animals) [8]. GIP has a similar, well documented [9] incretin effect and, in a recent study administration of a GIP receptor antagonist reduced postprandial insulin release in conscious rats by 72 % [10]. It seems probable, therefore, that elimination of its insulinotropic actions would also result in glucose intolerance.

A feature that characterises patients with NIDDM is an absent or grossly impaired incretin effect, evident after administration of small to moderate amounts of glucose [11–13]. A number of studies have addressed secretion of GIP and GLP-1 in diabetes, but most agree that, if anything, secretion,

particularly of GIP, is increased rather than impaired in NIDDM [12, 14]. Secretory defects, therefore, do not explain this decreased incretin effect. Furthermore, there is no evidence that the structure of the hormones is defective in NIDDM. However, when one compares the effects of the two hormones on insulin secretion in NIDDM patients, an interesting difference emerges [15, 16]. GLP-1 is strongly insulinotropic in these patients. Indeed, in patients with moderate disease, insulin secretion (as estimated by C-peptide responses) was near normal when compared to that of healthy control subjects under the conditions of an identical hyperglycaemic clamp [15]. The peptide is so effective that its i. v. administration may completely normalise blood glucose levels [17]. This has been observed in all patients investigated in this laboratory so far, regardless of the severity or duration of disease. Part of this remarkable effect is undoubtedly due to the concomitant inhibition of glucagon secretion [17, 18] and, as for meal-induced glucose excursions, its inhibitory effect on gastric emptying [19], but in all patients, the hormone causes a significant release of insulin. In fact, GLP-1 is the most efficacious and potent insulinotropic hormone known [2, 4, 20]. Possibly, with GLP-1, one may actually gauge the secretory capacity of the residual beta-cell mass in patients with diabetes mellitus. Why, then, is the incretin effect lost in NIDDM patients? It is typical of the studies reporting a lack of incretin effect, that small or moderate oral glucose loads were administered. As already mentioned, small glucose loads represent a very weak stimulus for GLP-1 secretion. Presumably, the incretin effect of GLP-1 is preserved in NIDDM, but this is overlooked with conventional incretin analysis, simply because it does not bring about appropriate stimulation of GLP-1 secretion [3]. In contrast, it turns out that GIP has little or no effect on insulin secretion in these patients! This has been found in studies employing both

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Abbreviations: NIDDM, Non-insulin-dependent diabetes mellitus; GIP, glucose-dependent insulinotropic polypeptide; Glp-1, glucagon-like peptide-1.

porcine natural or synthetic or more recently, human GIP [15, 16, 21]. Indeed, in parallel studies of GLP-1 and GIP, the full efficacy of GLP-1 and the lack of effect of GIP has been documented in the same patients [15, 16]. Thus, the lack of effect of GIP in NIDDM could very well explain the absent incretin effect (for small glucose loads) in these patients. Conceivably, an impaired GLP-1 response to glucose as observed in some patients with NIDDM [14] and an increased activity of the GLP-1 degrading enzyme, dipeptidyl-peptidase IV [22] may contribute to an impaired incretin response in patients with NIDDM, but could hardly explain its complete absence.

Why, then, is GIP ineffective while GLP-1 retains sufficient activity to be of therapeutic value? Both hormones exert their effect on the beta cell by interaction with a specific receptor. The two receptors, including the human forms, have been cloned and have been demonstrated to belong to the family of seven transmembrane spanning G-protein coupling receptors [23, 24]. Furthermore, both couple to adenylate cyclase [19, 24, 25]. Given the ineffectiveness of GIP, but not GLP-1, in diabetic beta cells, we speculated that, by identifying differences between the intracellular actions of the two hormones, one would also reveal the site of the defect in the diabetic beta cell. Detailed attempts were, therefore, made to find such a difference. In a series of studies in single beta cells, islet cell lines or cell lines transfected with receptors, and more recently, human beta cells isolated from fresh pancreases obtained from multiorgan donors (unpublished data), we were able to study, in parallel, the effects of the two hormones using sophisticated modern techniques, including patch-clamping, direct determination of intracellular ion concentrations, transmembrane ion currents, membrane potentials, and hormone secretion by capacitance measurements [26–28]. In these studies, all effects of the two hormones were identical: they generated identical changes of membrane potential, intracellular calcium responses, membrane currents, and cAMP responses [26–28]. Moreover, the identical insulin responses to either peptide were both abolished by inhibitors of the actions of protein kinase A, activated by cAMP; similarly, the responses could be entirely mimicked by adenylate cyclase activation using forskolin, which bypasses the membrane receptors and their coupling to the enzyme. In addition, the insulinotropic effects of both peptides are similarly potentiated by sulphonylurea treatment and similarly augmented in the genetically obese (*fa/fa*) Zucker rat [29–31]. We conclude, therefore, that the two receptors activate the same intracellular machinery.

The present assessment is that the incretin effect (for small loads of glucose which mainly release GIP) is absent in NIDDM, but the secretion of GIP is normal (or increased); the intracellular machinery of the beta cells of the diabetic pancreas functions

normally when tested with GLP-1, the mechanism of action of which is identical to that of GIP. Conclusion? The diabetic beta-cell either does not express or, if it does, expresses a defective receptor for GIP. Indeed, two missense mutations in the coding region of the GIP receptor gene were recently described, one of which exhibited markedly decreased responsiveness to GIP in functional studies [32]. However, none of the mutations were associated with NIDDM in a Japanese study group. On the other hand, the many studies in Caucasians showing that GIP does not stimulate insulin secretion in NIDDM patients provide strong evidence for the hypothesis. Thus, NIDDM in Caucasians may have a different genetic background [33], or the defect may lie outside the coding region of the gene. In most discussions of the role of the beta cell in the pathogenesis of NIDDM up until now, investigators have focussed on its defective glucose responsiveness, and have therefore overlooked the possible role of the insulinotropic hormones. Indeed, GLP-1 – but in all likelihood this also applies to GIP – has been demonstrated to confer “glucose competence” on the beta cells: without GLP-1 (or GIP or glucagon, which have a similar effect) glucose cannot elicit normal secretory responses from the cells [26, 34]. Of course, it may be that additional defects are found in NIDDM, which is believed to be a polygenic disease. Furthermore, it cannot be excluded that features of the diabetic state, e.g. hyperglycaemia and changes in lipid metabolism, may impair GIP receptor function. However, it is difficult to understand why this should affect only the GIP receptor and not the homologous GLP-1 receptor. In one study [29], sulphonylurea treatment was found to generate an insulin secretion in response to a high dose of GIP under the conditions of an 18 mmol/l hyperglycaemic clamp (whereas GIP alone was ineffective). However, the insulin response amounted to, at most 10% of normal (without sulphonylurea) as opposed to the near normalisation of insulin secretion obtained with GLP-1 in patients with similar characteristics (and without sulphonylurea) [15, 16]. Presumably this means that NIDDM patients may express small amounts of a GIP receptor (suggesting that the defect might lie in the regulation of GIP receptor gene expression), or larger amounts of a defective receptor. Whichever is true, it is striking that all patients investigated so far, have had a defective insulin response to GIP. Moreover, it is similarly striking that genetic “knock out” of an incretin hormone receptor causes frank diabetes [8]. Possibly, lack of a functional incretin receptor for a lifetime may put such a strain on the body’s additional mechanisms for handling and disposing of glucose that diabetes finally results. We propose that a genetically defective expression of the GIP receptor causes this strain on the body. This may, in turn, be aggravated by a sedentary lifestyle and obesity. It may be the latter factors rather

than the GIP receptor defect that explain the apparent polygenicity of the disease! Clearly, however, proof of our hypothesis requires identification of the responsible mutation or mutations in the GIP receptor gene. It is our hope that our hypothesis may inspire the Occidental molecular geneticists to look in the direction of the GIP receptor gene or gene regulation without being discouraged by the negative Japanese study [32].

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