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Breakthroughs and Views

Dipeptidyl peptidase IV inhibition as an approach to the treatment and prevention of type 2 diabetes: a historical perspective

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We have read with interest the recent paper by Hinke et al. [1], which appeared in the March issue of Biochemical and Biophysical Research Communications. In their paper, Hinke et al. describe the potential of inhibitors of dipeptidyl peptidase IV (DPP IV) as novel therapeutic agents to treat type 2 diabetes, and address the question of whether the mechanism of action of metformin involves inhibition of DPP IV to enhance the antidiabetic effect of the incretin hormone glucagon-like peptide-1 (GLP-1). Having read the paper, we were concerned that the authors have presented a summary of the literature which is not perfectly balanced and is, in some instances, inaccurate. The purpose of this short paper is, therefore, to present a balanced and accurate review of the studies which have led up to the current interest in developing DPP IV inhibitors as therapeutic agents in type 2 diabetes.

The discovery in the early 1980s that mammalian preproglucagon contains, in addition to glucagon, sequences of two related peptides [2], began a search for the biological function of these additional peptides, with many research groups taking up the chase, our own amongst them. With the demonstration that GLP-1 was probably the most potent insulinotropic agent hitherto known [3–5], interest grew explosively. GLP-1 was shown to be released in response to orally ingested nutrients and to act as an incretin hormone, stimulating meal-induced insulin secretion (see [6,7] for reviews). Particular excitement was aroused when it was shown in 1992 that GLP-1 was also effective in patients with type 2 diabetes [8,9], and could normalise blood glucose in these subjects when given as a continuous intravenous infusion [10–12]. However, unexpectedly, the effects of a single subcutaneous injection of GLP-1 were disappointing. Despite plasma levels of immunoreactive GLP-1 being maintained well above basal levels [13], insulin secretion rapidly returned to pre-treatment values and blood glucose concentrations were not normalised [13–15]. However, the effect of repeated subcutaneous administration on fasting blood glucose is as good as that of intravenous administration [13], and continuous subcutaneous administration is also effective [16].

The first hint of an explanation for the short-lived effectiveness of single subcutaneous injections of GLP-1 was indicated in 1992. In a meeting abstract, Buckley and Lundquist [17] described degradation of GLP-1 by plasma and the resulting formation of a metabolite truncated by two N-terminal residues, but it was Mentlein et al. [18] in 1993, who identified the enzyme responsible. In in vitro incubations, they showed that micromolar concentrations of GLP-1 (and the other related incretin hormone, glucose-dependent insulinotropic polypeptide, GIP) were N-terminally degraded by DPP IV [18]. This finding spurred research into incretin hormone metabolism in vivo. Our own group [19], first became interested in this specific area in mid 1993, resulting in the presentation of preliminary data showing that also picomolar concentrations of GLP-1 were degraded by human plasma in vitro. In these early days, research effort from our own group would appear now to have paralleled that of Pederson et al. and Demuth et al. with the first reports that GLP-1 was degraded by DPP IV in vivo appearing in 1995. Thus, we published that physiological concentrations of GLP-1 were degraded by DPP IV and, furthermore, identified the truncated metabolite as an *endogenous* circulating peptide *in man* in March 1995 [20], while Kieffer et al. were the first to show in vivo degradation of *exogenously* administered GLP-1 (and GIP) *in rats* in August 1995 [21]. This was followed one month later by our own

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demonstration that exogenous GLP-1, particularly after subcutaneous injection, was N-terminally degraded in both normal and diabetic subjects [22], and later, with the development of specific analyses, endogenous GIP was also shown to be N-terminally degraded [23]. Indirect observations in uremic patients had indicated a role for the kidney in the elimination of GLP-1 [24], and this was later confirmed in rats by Ruiz-Grande et al. [25] in 1993. Subsequently, our own group extended the observations to include the liver in 1996, and by use of region-specific radioimmunoassays showed that N-terminal degradation of GLP-1, particularly in the hepatic bed, was widespread [26]. Similar findings were later shown for GIP [27]. Taken together with the knowledge that DPP IV has a widespread distribution in the renal tubules, on hepatocytes and on capillary endothelium (reviewed in [28]) and the suggestion that the truncated metabolites may not be able to activate the respective incretin receptors [18,29,30], an important role for DPP IV in the physiological regulation of GLP-1 and GIP activity was suggested [18,20,21]. The finding that the enzyme is localised in the endothelium of capillaries actually adjacent to the GLP-1-containing L-cells [31], together with the demonstration that over half of newly synthesised (intact) GLP-1 is N-terminally degraded even before it leaves the local capillary bed to enter the systemic circulation [31], further underscores the relevance of DPP IV in incretin hormone biology.

The elucidation of the pivotal role of DPP IV in incretin hormone metabolism coupled with on-going studies from many research groups into the possibility of using GLP-1 therapeutically prompted the initiation of studies into the possibility of manipulating the in vivo metabolism of GLP-1 as a novel therapeutic approach, and the idea of using DPP IV inhibitors as a diabetes treatment has been the focus of much recent interest (academic as well as commercial). Hinke et al. [1] claim that their own research group was the first to postulate such a "link between the possible benefits of DP IV inhibition and glycemic control due to enhancement of the incretin effect" [32]. However, examination of the literature reveals this not to be the case. Our own study, published in 1995 [22], a full year before the Pauly [32] study, discussed the role of DPP IV in incretin hormone metabolism and concluded that "inhibition of dipeptidyl peptidase IV may prove a useful adjunct in the management of type 2 diabetes" because "inhibition of GLP-1 (7–36) amide degradation would ... increase the availability of the biologically active peptide," and it was additionally suggested that DPP IV-resistant GLP-1 analogues may also have therapeutic potential [22]. This was followed by a review paper [33] in which we discussed the therapeutic potential of DPP IV inhibitors in diabetes treatment. In fact, Pauly et al. [32] do not actually mention DPP IV inhibitors in relation to glycaemic control per se, but rather speculate that DPP IV inhibition "is predicted to have a profound effect on the enteroinsular axis" because of "an increased half-life of endogenously released GIP and GLP-1."

The initial suggestion in 1995, that DPP IV inhibition may influence GLP-1 metabolism in vivo leading to improvements in glucose tolerance [22], was indicated to be correct in two preliminary communications presented at the same meeting in 1996. Thus, partial inhibition of DPP IV with diprotin A (a competitive substrate of DPP IV which is not wholly effective in vivo) reduced the N-terminal degradation of exogenous GLP-1 in anaesthetised pigs [34], while the inhibitor ile–thiazolidide was observed to improve glucose tolerance in rats [35]. These preliminary observations were subsequently confirmed in full papers. Thus, in May 1998, Deacon et al. [36] demonstrated that the inhibitor valine–pyrrolidide improved the metabolic stability of intact GLP-1 3-fold, potentiating its insulinotropic and anti-hyperglycaemic effects, while Pederson et al. [37] reported in August 1998 that ile–thiazolidide improved glucose tolerance in rats. Subsequently, these studies have been corroborated by a number of acute studies all demonstrating that DPP IV inhibition is effective in animal models of impaired glucose tolerance [38–40]. Data on whether long-term DPP IV inhibition maintains its beneficial effect on glucose tolerance is only just beginning to emerge. Brand et al. [41] in a meeting abstract from 1999, reported that improved glucose tolerance was maintained after 1-week treatment with valine–pyrrolidide in Zucker obese rats, while more recently, Pospisilik et al. [42] first in a preliminary communication in 2001 and subsequently as a full paper in 2002 [43], demonstrated that 12-week treatment was also associated with sustained improvements in glucose tolerance. In fact, in the latter report [43], the effect of DPP IV inhibition actually appeared to improve with time. The preliminary results from human studies are also starting to be reported. Thus, single dose treatment with a DPP IV inhibitor reduces the glucose excursion in both healthy and diabetic subjects [44,45], while chronic treatment over 4 weeks improves metabolic control in patients with type 2 diabetes [46]. These preliminary results are encouraging, and support the hypothesis that DPP IV inhibition may be a viable approach to diabetes treatment.

In their paper Hinke et al. [1], examine whether the mechanism of action of metformin involves inhibition of DPP IV. They suggest that the only other study to examine a potential interaction between metformin, GLP-1 and DPP IV was carried out by Mannucci et al. [47], and that no studies have been carried out in diabetic subjects. Again, we find this to be inaccurate, since our own group [48] also published a study in 2001, in which the effects of metformin alone, GLP-1 alone and metformin + GLP-1 were examined in patients with type 2 diabetes. Hinke et al [1] criticise the study of Mannucci et al. [47] for failing to measure total GLP-1 in addition

to intact GLP-1 levels, yet ignore the Zander et al.'s [48] study where both these parameters were measured and found to be identical in the GLP-1 alone and metformin + GLP-1 treated groups. Indeed, in the Zander et al.'s [48] paper, it was concluded that "inhibition of GLP-1 degradation cannot explain the additive effect of GLP-1 and metformin."

It is our intention that this short chronological review should recognise all the studies which have contributed to the elucidation of incretin hormone metabolism and the subsequent research into the therapeutic potential of DPP IV inhibitors for the prevention and treatment of type 2 diabetes. It is our hope that future reports will acknowledge fairly the contributions of *all* pertinent research in this intriguing field.

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