

THE GLP-1 SYSTEM: BIOCHEMISTRY, PHYSIOLOGICAL EFFECTS AND THE ROLE OF DDP-IV INHIBITORS IN THE MANAGEMENT OF TYPE 2 DIABETES MELLITUS

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ABSTRACT

Type-2 diabetes *mellitus* (T2DM) is a metabolic disease characterized mainly by hyperglycemia, resulting from defects in **β -cell function and/or insulin resistance**. Treatment approaches include diet, exercise, and pharmacological agents these however can produce adverse effects such as weight gain and hypoglycemia. Glucagon-like peptide-1 (GLP-1) is a peptide hormone that controls glycaemia and preserves β -cell mass and function. Currently dipeptidyl peptidase (DDP-IV) inhibitors are a new class of anti-diabetics for the treatment of T2DM, prolonging GLP-1 action on insulin release.

KEYWORDS

GLP-1; Glucagon-like peptide-1; DDP-IV; DDP-IV inhibitors; Diabetes *mellitus* type 2.

RESUMO

A Diabetes *mellitus* tipo-2 (DMT2), é uma doença metabólica caracterizada por hiperglicemia, devido à disfunção das células- β e/ou resistência à insulina. Os tratamentos disponíveis além de dieta e exercício físico, incluem terapia farmacológica com possíveis efeitos colaterais tais como ganho de peso e hipoglicemia. O *Glucagon-like peptide-1* (GLP-1) é uma hormona peptídica que controla a glicemia, preserva e previne a perda das células- β mas é rapidamente degradada pela dipeptidyl peptidase-IV (DDP-IV). Os Inibidores da DDP-IV têm sido propostos para tratamento da DMT2, dado prolongarem a acção de GLP-1 na libertação de insulina e controle glicémico.

PALAVRAS-CHAVE

GLP-1. Glucagon-like peptide-1; DDP-IV; inibidor da DDP-IV; Diabetes *mellitus* tipo 2.

1. INTRODUCTION

The current pandemic of diabetes *mellitus* and projections for future growth in the prevalence of the disease threaten to create a global health crisis (Pratley). β -cell dysfunction in type 2 diabetes *mellitus* (T2DM) is characterized by reduced β -cell sensitivity to glucose a delay and a reduction in the meal-induced insulin secretion, a loss of the regular oscillatory insulin secretion and an excess hepatic glucose production (Verspohl). In addition, in T2DM is observed resistance to insulin action that is determined by several factors, including genetic predisposition, age, sex, obesity with resulting hyperglycaemia, elevation of blood pressure and metabolic syndrome (Sinaiko). Currently, several treatment modalities for T2DM exist, including exercise, diet and a variety of therapeutic agents (e.g., insulin, biguanides, sulfonylureas and thiazolidinediones). However, several adverse effects can be associated with the use of these agents, such as hypoglycaemia, weight gain and oedema. Thus, a new treatment concept based on incretin hormone GLP-1 action can complement the existing therapies and possibly attempt to preserve normal physiological response to meal intake. From the numerous pharmacological agents, DDP-IV inhibitors appear to be the most attractive candidates for the treatment of T2DM. Therefore this review focuses on the more recent studies of the mechanism regulating the synthesis, the biological actions and the potential therapy of the incretin GLP-1 as well as the current preclinical and clinical development of DDP-IV inhibitors and their efficacy profile in the therapeutic strategy for type 2 diabetes *mellitus*.

2. BIOCHEMICAL AND PHYSIOLOGICAL ACTIONS OF GLP-1

2.1. SYNTHESIS

GLP-1 is a product of the proglucagon gene. It is expressed in pancreatic α -cells and also in the L-cells of the intestinal mucosa (Bell et al.). During the maturation of these two cell types, proglucagon gene is activated. However, with further cell differentiation, post-translationally the 160 amino acid proglucagon protein precursor undergoes differential proteolytic processing by the secretory granules convertases at distinct dibasic residues. The α -cells cleave glucagon from the region spanning amino acids (aas) 33 to 61 and then release it along with the major proglucagon fragment (MPGF) (Holst et al.). In contrast, L-cells cleave two structurally related glucagon-like peptides (GLPs) from C-terminal located portions of the precursor molecule, namely GLP-1, from the region spanning aas 78 to 107, and GLP-2, from the region spanning aas 126 to 158 (Mojsov et al.).

2.2. SECRETION

Meal ingestion, particularly if rich in fats and carbohydrates, is the primary physiologic stimulus for GLP-1 secretion (Brubaker). Although there is a basal rate of secretion, fasting GLP-1 plasma concentrations remains very low (Ørskov et al.). Oral, but not intravenous, glucose administration stimulates GLP-1 secretion in humans (Hermann et al.). In addition, several studies have shown that the autonomic nervous system, the gastrin-releasing peptide (GRP), acetylcholine and the gastric inhibitory polypeptide (GIP, also known as glucose-dependent insulintropic peptide), all can contribute to the rapid release of GLP-1 after nutrient ingestion (Baggio and Drucker). Therefore, nutrient-generated stimulatory signals can be transmitted to L-cells either indirectly, through neural or endocrine mediators, or via direct contact, to produce the early and late phases of GLP-1 secretion, respectively. However, because L-cells seem to be present throughout the entire length of the small intestine, it is possible that early GLP-1 secretion also can occur by direct association of nutrients with L-cells located in more proximal regions of the small intestine (Theodorakis et al.).

2.3. GLP-1 RECEPTORS

The GLP-1 receptor (GLP-1R) is a specific 7-transmembrane receptor guanine nucleotide-binding G protein coupled receptor (GPCR), *i.e.*, one of the group of 15 receptors (in the human genome), including GIP and the glucagon receptors, that is activated by intermediate sized peptides (typically ~30-40 amino acid residues) (Mayo et al.). Upon GLP-1R activation, adenylyl cyclase is activated and cAMP is generated, leading, in turn, to cAMP dependent activation of second messenger, such as the protein kinase A (PKA) and Epac pathways. Hence, GLP-1 acts directly through the cAMP-dependent protein kinase A pathway to enhance and sensitize β -cells in the process of glucose-stimulated insulin secretion (Holz, Kuhlreiter and Habener).

2.4. PHYSIOLOGICAL EFFECTS

Administration of GLP-1 to patients with T2DM increases glucose-dependent stimulation of insulin release, inhibits glucagon secretion, suppresses appetite, produces weight loss and delays gastric emptying. Studies in animals have shown that GLP-1 slows apoptosis and promotes proliferation of β -cells leading to an increase in their mass (Mayo et al.). The inhibition of glucagon secretion is an effect that seems to be due to paracrine regulation of α -cells (Tornehave et al.) or due to the inhibition of somatostatin release (Fehman, Goke and Goke).

In the gastrointestinal tract, GLP-1 inhibits gastrin-induced acid secretion in humans, besides decreases and delays the gastric emptying rate by stimulating antral churning while inhibiting pyloric propulsion and duodenal peristalsis (Schirra et al.). GLP-1 suppresses appetite either by reducing gastric emptying and by inducing stomach fullness or by activating the satiety centers on the arcuate nucleus of the hypothalamus or inhibiting the solitary tract nucleus of the brain stem (Holst).

GLP-1 has been shown to produce beneficial effects on myocardial function *in vitro* and *in vivo* experiments (Bose et al.; Nikolaidis et al.; Nyström et al.). Lastly, GLP-1 improves postprandial lipidemia, probably due to delayed gastric emptying and insulin-mediated inhibition of lipolysis or due to an increase clearance or even reduced endogenous synthesis of triglycerides (Meier et al.).

3. DDP-IV ENZYME CHARACTERISTICS

The catalytic enzyme dipeptidyl peptidase IV (also called DDP-IV, DP IV, CD 26) is a 766 amino acid, membrane-associated ecto-peptidase that is widely expressed in several organs and circulates in the blood (Knudsen and Pridal). DPP-IV has substrate specificity for oligopeptides with a penultimate prolyl-, anlyl-, or seryl-, residue at their N-terminal. This enzyme is the primary inactivator of the incretin hormones GLP-1 and GIP. In the case of GLP-1, the metabolites generated, namely GLP-1 (9-36) amide from GLP-1 (7-36) and GLP-1 (9-37) from GLP-1 (7-37), are not only inactive but they may act as competitive antagonists of the intact GLP-1 at the GLP-1 receptors (Knudsen and Pridal).

4. THERAPEUTIC APPROACHES BASED ON DDP-IV INHIBITORS

Inhibition of dipeptidyl peptidase-IV is a novel oral treatment for T2DM. DPP-IV inhibitors inhibit more than 90% of the DPP-IV activity. Thus, these drugs exert their glucose-regulatory effects through prolongation of the action of GLP-1 (Figure 1) (Hansotia et al.). The DPP-IV inhibitors have been shown to improve glycaemic control, being a new promise used in the treatment of diabetes (Richter et al.).

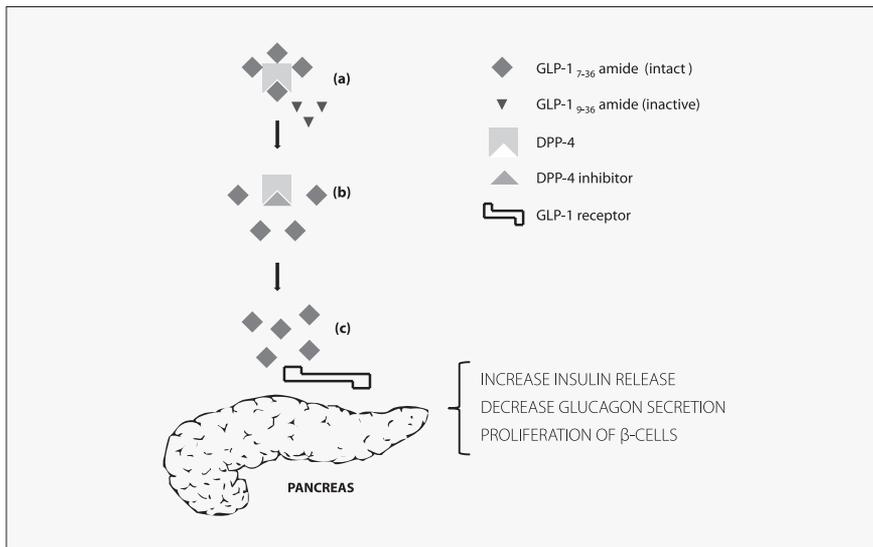


FIGURE 1 - Mechanism of action of DPP-IV inhibitors: These drugs inhibit the DPP-IV activity, preventing the enzymatic inactivation of GLP-1. a) The enzyme DPP-IV cleaves the intact GLP-1, producing inactive fragments of GLP-1; b) DPP-IV inhibitors inactivate the enzyme and c) active GLP-1 bind to incretin receptors of pancreatic β -cell, increasing the insulin release, the proliferation of β -cell and decreasing of glucagon secretion.

4.1. SITAGLIPTIN

Sitagliptin (Januvia[®]; Merck and Co, Inc.) was approved for use in a dose of 100 mg once daily, by the FDA in 2006 and subsequently in the EU, Australia and Asia, for the improvement of glycaemic control in combination with metformin (Janumet[®]; Merck), sulfonylurea and/or glitazone

(Khoo et al.; Rosenstock et al., "Efficacy and tolerability"). The beneficial effect of Sitagliptin on glycaemic control is clearly associated with a significant improvement on β -cell function and islet mass in a rodent model of T2DM (Mu et al.). Furthermore, chronic DPP-4 inhibition with Sitagliptin demonstrated superior glucose-lowering efficacy and β -cell preserving effects compared to the commonly used insulin secretagogue glipizide (Mu et al.). Greater benefits in glycaemic control were seen with Sitagliptin 100 mg daily, compared to placebo in patients with T2DM already on metformin (Goldstein et al.) and pioglitazone, (Rosenstock et al., "Efficacy and safety") and when added to sulfonylureas (with or without metformin) (Hermansen et al.). Sitagliptin is not associated with changes in body weight (Nauck et al., "Efficacy and safety of the dipeptidyl").

The most common side events include urinary tract infection, headache and nasopharyngitis (Amori, Lau and Pittas). In addition, the FDA has reported cases of pancreatitis in patients using Januvia® and Janumet® (FDA).

4.2. VILDAGLIPTIN

Vildagliptin (Galvus®; Novartis) was approved in the EU in 2008, for use in combination with metformin (Eucreas®; Novartis) and/or thiazolidinediones (50 mg twice daily), or with a sulphonylureas (50 mg once daily) for the treatment of T2DM. However, this drug is pending approval by the US Food and Drug Administration (FDA) (Khoo et al.) due to the skin lesions and kidney impairment (Electronic Medicines Compendium).

Clinically, Vildagliptin monotherapy was well-tolerated and provided similar clinical benefit (Pi-Sunyer; Scherbaum et al.; Utzschneider et al., 2008). In other trial studies, addition of Vildagliptin to a sulphonylurea (Garber et al., "Effects") or pioglitazone (Garber et al., "Vildagliptin") significantly reduced HbA_{1c}. Clinical trials indicate that Vildagliptin is weight-neutral (Bolli et al.). Vildagliptin has been shown to increase insulin and C-peptide responses to glucose by 50 and 100% respectively (D'Alessio et al.) suggesting improvements in β -cell function (Mari et al.). It's most common adverse effects include headache, nasopharyngitis, dizziness, back pain, peripheral oedema and arthralgia (Bolli et al.).

4.3. SAXAGLIPTIN

Saxagliptin (Onglyza™) is a selective inhibitor suitable for once-daily administration that has currently completed phase 3 trials and has just been approved by FDA. It is a reversible inhibitor of DDP-IV, which is 10-fold more potent than Vildagliptin and Sitagliptin. Saxagliptin is capable of decreasing the average HbA_{1c} level in patients with T2DM when used alone or in combination with metformin, a sulphonylurea, or a thiazolidinedione (Rosenstock, Ratner and Botka). Common adverse reactions reported in the clinical trials with Saxagliptin include nasopharyngitis, headache, diarrhea, upper respiratory infections, influenza, and urinary tract infection (Ravichandran et al.).

4.4. OTHER DDP-IV INHIBITORS

Alogliptin (Takeda Pharmaceutical Company) is a potent and highly selective DDP-IV inhibitor which has currently completed the phase 3 trials. Alogliptin has demonstrated more than 10,000 times more selectivity for DDP-IV than for other related proteases (Christopher et al.). Alogliptin inhibited plasma DPP-4 activity and significantly decreased postprandial plasma glucose levels when compared with placebo (Covington et al.). The most commonly reported adverse events for Alogliptin were headache, dizziness and constipation. Patients experienced

a 0.6% reduction in HbA1c level and a 1 mmol/l reduction in fasting glucose level after 26 weeks of adding Alogliptin to metformin therapy (Nauck et al., "Efficacy and safety of adding").

Linagliptin (Boehringer Ingelheim) is another oral DPP-4 inhibitor under evaluation for once daily dosing in patients with T2DM. It has been demonstrated that in 47 type 2 diabetic patients, multiple rising doses of Linagliptin (1, 2.5, 5, 10 mg) were well tolerated and resulted in significant improvements of glucose parameters (Heise et al.). The frequency of adverse events was not higher with Linagliptin (54%) than with placebo (75%).

ASP8497 is a novel selective and competitive dipeptidyl peptidase-IV inhibitor that is less likely to induce hypoglycemia and less likely to show reduced efficacy even after repeated administration (Matsuyama-Yokono et al.). The combination of ASP8497 with existing anti-diabetic drugs could be useful for correcting the postprandial hyperglycemia seen with type 2 diabetes (Tahara et al.; Someya et al.).

5. DISADVANTAGES OF DDP-IV INHIBITORS

There are several enzymes which have similar activity of DDP-IV such as DDP-2, DDP-8 and DDP-9. Therefore, inhibition of DDP-IV may interfere with the activity of these enzymes. Data have shown that inhibition of DDP-8 and DDP-9 seem to be responsible for toxic effects such as alopecia, thrombocytopenia, anemia, enlarged spleen, multiple histological pathologies and mortality (Lankas et al.).

Another limitation of DDP-IV inhibitors use is the multiple functions of this enzyme. DDP-IV is an enzyme that inactivates several neuropeptides, peptide hormones and chemokines. Thus, DDP-IV inhibitors may also prolong the action of some substances and result in potential side effects, including neurogenic inflammation, enhanced general inflammation, and allergic reactions. However, so far, side effects have not been published in preclinical animal or clinical human studies (Holst and Deacon). In addition, the DDP-IV inhibitors therapy is associated with higher cost in comparison with the use of the GLP-1 agonists (Lage et al.).

6. CONCLUSION

As the prevalence of diabetes T2DM increases worldwide, novel anti-diabetic agents have been and are being developed to treat this disease. DDP-IV inhibitors offer various benefits over commonly used drugs, including the improving the β -cells function, low risk of hypoglycemia and no weight gain. Because of these characteristics and due to their ease of use for patient, DDP-IV inhibitors play an important role in the treatment of T2DM. Several trials have demonstrated the efficacy and safety mainly of Sitagliptin and Vildagliptin. However, further clinical data will be required to show other effects such as body weight loss and β -cell protection and to elucidate the full potential of DDP-IV inhibitors as anti-diabetic agents.

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