Harnessing the incretin system beyond glucose control: Potential cardiovascular benefits of GLP-1 receptor agonists in type 2 diabetes

B. Cariou a, b, *

a Université de Nantes, CHU de Nantes, Hôpital Guillaume et René-Laennec, boulevard Jacques-Monod, Saint-Herblain, 44093 Nantes cedex 1, France
b L’Institut du Thorax, Clinique d’Endocrinologie, Maladies Métaboliques et Nutrition Hôpital Guillaume et René-Laennec, boulevard Jacques-Monod, Saint-Herblain, 44093 Nantes cedex 1, France

Received 10 August 2011; received in revised form 14 April 2012; accepted 14 April 2012

Abstract

The management of type 2 diabetes continues to evolve as new data emerge. Although glycaemic control is still important, other risk factors – such as hypertension, dyslipidaemia and obesity – must also be addressed in order to reduce the long-term risks of cardiovascular complications and mortality. In this context, targeting the incretin system, and glucagon-like peptide-1 (GLP-1) in particular, has generated much interest. GLP-1 is released from the gut in response to food ingestion and plays a crucial role in glucose homeostasis. GLP-1 receptors are expressed in the heart and vasculature, prompting evaluation of their physiological role and pharmacological stimulation, both in healthy and disease states. These studies indicate that GLP-1 and GLP-1-based therapies appear to have direct, beneficial effects on the cardiovascular system, in addition to their glucose-lowering properties, such as modulation of blood pressure, endothelial function, and myocardial contractility. Intriguingly, some of these effects appear to be independent of GLP-1 receptor signalling. Data from clinical studies of the GLP-1 receptor agonists, exenatide and liraglutide on cardiovascular risk factors, in patients with type 2 diabetes are also promising and the results from prospective studies to assess cardiovascular outcomes are eagerly awaited.

© 2012 Elsevier Masson SAS. All rights reserved.

Keywords: Cardiovascular disease; Incretin therapy; Type 2 diabetes; GLP-1; Endothelial function; Hypertension; Myocardial function; Review

Résumé

Bénéfices cardiovasculaires potentiels des analogues du GLP-1 dans le diabète de type 2 : une action au-delà du contrôle glycémique.

La prise en charge du diabète de type 2 (DT2) a évolué avec l’arrivée des nouvelles classes d’antidiabétiques. Bien que le contrôle glycémique demeure important, les autres facteurs de risque cardiovasculaires tels que l’hypertension artérielle, la dyslipidémie et l’obésité doivent être également pris en compte. Le système incrétine et notamment le glucagon-like peptide-1 (GLP-1) a émergé ces dernières années comme une nouvelle cible pour le traitement du DT2. Le GLP-1 est sécrété par l’intestin en réponse aux repas et stimule la sécrétion d’insuline de manière glucose-dépendante. Outre les îlots de Langerhans, les récepteurs du GLP-1 sont aussi exprimés dans la paroi vasculaire et le cœur, ce qui suggère la possibilité de l’existence d’une action cardiovasculaire directe du GLP-1. Des études ont démontré que le GLP-1 endogène ou les agonistes du récepteur du GLP-1 (exenatide ou liraglutide) exerçaient plusieurs effets bénéfiques directement au niveau cardiovasculaire, comme une diminution de la pression artérielle, une amélioration de la fonction endothéliale et de la contractilité du myocarde dans des situations d’ischémie. De façon surprenante, une partie de ces effets est indépendante de la voie de signalisation du récepteur du GLP-1 et semble impliquer la forme clivée inactive du GLP-1 (GLP-1 [9–36]). Les premières données cliniques qui concernent le contrôle des facteurs de risque cardiovasculaire dans le DT2 avec l’exenatide ou le liraglutide sont prometteuses et semblent valider l’hypothèse d’un bénéfice cardiovasculaire de cette nouvelle classe thérapeutique. Néanmoins, il faut encore attendre les résultats des études de morbi-mortalité cardiovasculaire actuellement en cours pour en avoir la confirmation définitive.

© 2012 Elsevier Masson SAS. Tous droits réservés.

Mots clés : Diabète de type 2 ; Incrétines ; GLP-1 ; Hypertension artérielle ; Fonction myocardique ; Insuffisance cardiaque ; Revue générale

* Tel.: +33 2 53 48 27 07; fax: +33 2 53 48 27 08.
E-mail address: bertrand.cariou@univ-nantes.fr

1262-3636/$ – see front matter © 2012 Elsevier Masson SAS. All rights reserved.
http://dx.doi.org/10.1016/j.diabet.2012.04.003
1. Introduction

It is well established that patients with diabetes have a higher risk of cardiovascular (CV) morbidity and mortality than patients without diabetes. For example, in the Framingham Heart study, diabetes was associated with a 2 to 4-fold risk of myocardial infarction, congestive heart failure and stroke, as well as an increased risk of mortality [1,2]. In addition, in the Multiple Risk Factors Intervention Trial, the absolute risk of CV-disease was much higher for diabetic than non diabetic men, independently of the presence of other CV risk factors [3]. At the end, absolute excess CV risk for diabetic men is progressively greater than for non diabetic men with higher risk factor levels [3]. This increased diabetes-associated risk of CV-disease reflects the negative effects of chronic hyperglycaemia on the vasculature [4]. In addition, patients with diabetes often suffer from other comorbid conditions, such as hypertension, combined dyslipidaemia (high LDL-C and low HDL-C levels) and visceral obesity, which also predispose them to CV complications [5].

Glycaemic control remains fundamental to the management of diabetes, especially for preventing microvascular complications [6]. However, the benefits of tight glycaemic control for preventing CV complications are being questioned since recent prospective intervention studies failed to demonstrate a clear benefit in patients with T2DM at very high CV risk [7–9]. A more holistic approach, designed to address all diabetes-associated risk factors, is now advocated based on data from studies showing significant benefits when multiple risk factors are addressed [10–12]. This approach is exemplified in the most recent guidelines on medical care for diabetes from the American Diabetes Association [13]; i.e., that the ideal antidiabetic drug would improve glycaemic control and CV risk factors simultaneously. However, many standard antidiabetic agents are either risk factor neutral or have a detrimental effect on CV risk factors [14], and it has been suggested that this may explain, in part, the lack of effect on CV outcomes when diabetes is treated using these agents [15].

The incretin system has been the focus of intense research as a potential target for drugs with which to treat T2DM, given its key role in stimulating postprandial insulin secretion and concomitantly reducing glucagon secretion [16]. In the case of GIP, this is characterised by a marked impairment in insulinotropic activity, with relatively unchanged circulating levels of the peptide [21]. The situation with GLP-1 is more complex. Some studies have reported a decrease in GLP-1 secretion in T2DM, but the balance of evidence now suggests that GLP-1 levels are relatively unchanged, and that there is a modest impairment in insulinotropic activity [21]. However, unlike GIP, high concentrations of GLP-1 can restore the insulin response to glucose and normalise insulin secretion in T2DM [22]. In view of this, GLP-1 has become the focus of research and drug development in this area. GLP-1 has additional properties, namely inhibition of glucagon secretion and gastric emptying, which are also attractive in the context of diabetes treatment [18]. Its effects on insulin and glucagon release occur in a glucose-dependent manner, thus minimizing the risk of hypoglycaemia [16,20,22,23]. Based on these properties, GLP-1-based therapies have emerged as pivotal therapies in the pharmacological management of T2DM [24].

In addition to its roles in glycaemic control and satiety, GLP-1 appears to exert several additional effects on many tissues via the GLP-1 receptor, which is expressed not only in the pancreatic islets, but also in the lung, kidney, intestine and several regions of the central nervous system [16]. This widespread expression of the GLP-1 receptor may help to explain the range of extrapancreatic effects of GLP-1, including a potentially protective effect on the CV system as GLP-1 receptors are also found in endothelial cells, vascular smooth muscle cells, monocytes – macrophages of the vascular wall [25], and in the heart [26].

3. The physiology of native GLP-1 in the cardiovascular system

The pharmacological effects of GLP-1 on CV function (Fig. 1) have been evaluated using infusion of native GLP-1, and knockout mice lacking the GLP-1 receptor. GLP-1 is sometimes specifically referred to as GLP-1(7–36) amide and GLP-1(7–37) amide, the bioactive forms of GLP-1 [27,28], which are derived from the 37-amino acid precursor, GLP(1–37), which in turn is derived from the large proglucagon precursor (Fig. 2) [18]. GLP-1(7–36) amide constitutes 80% of circulating GLP-1 [18], to 70% of insulin secretion after food intake [16]. The incretin effect is impaired in patients with T2DM [20]. In the case of GIP, this is characterised by a marked impairment in insulinotropic activity, with relatively unchanged circulating levels of the peptide [21]. The situation with GLP-1 is more complex. Some studies have reported a decrease in GLP-1 secretion in T2DM, but the balance of evidence now suggests that GLP-1 levels are relatively unchanged, and that there is a modest impairment in insulinotropic activity [21]. However, unlike GIP, high concentrations of GLP-1 can restore the insulin response to glucose and normalise insulin secretion in T2DM [22]. In view of this, GLP-1 has become the focus of research and drug development in this area. GLP-1 has additional properties, namely inhibition of glucagon secretion and gastric emptying, which are also attractive in the context of diabetes treatment [18]. Its effects on insulin and glucagon release occur in a glucose-dependent manner, thus minimizing the risk of hypoglycaemia [16,20,22,23]. Based on these properties, GLP-1-based therapies have emerged as pivotal therapies in the pharmacological management of T2DM [24].

In addition to its roles in glycaemic control and satiety, GLP-1 appears to exert several additional effects on many tissues via the GLP-1 receptor, which is expressed not only in the pancreatic islets, but also in the lung, kidney, intestine and several regions of the central nervous system [16].
and the term GLP-1 will therefore be used to refer to this amide unless stated otherwise. Of note, circulating human GLP-1 is rapidly inactivated by DPP-4 by cleaving of two amino acids, resulting in the metabolite GLP-1(9–36) amide (Fig. 2) [29]. This GLP-1 metabolite functions as a weak GLP-1 receptor antagonist, but is mostly considered to be biologically inactive [30,31]. Furthermore, as outlined below, GLP-1 receptor agonists such as exendin-4 (exenatide) and liraglutide have also been used to evaluate the effects of GLP-1 on CV function. A truncated version of the peptide exendin-4, exendin(9–39) has been shown to have GLP-1 receptor antagonist properties [32] and is often used to assess the GLP-1 receptor selectivity of GLP-1 mediated action.

3.1. The effects of native GLP-1 on blood pressure

3.1.1. Preclinical studies

In vitro studies have consistently shown that GLP-1 is vasodilatory [33–37], and has a dose-dependent effect [33,34,37] which can be inhibited by exendin(9–39) [34]. Furthermore, an ex vivo study using aortas from rats with diabetes induced by streptozotocin/nicotinamide demonstrated that GLP-1 could normalise the altered vascular tone in these rats [35].

In vivo animal studies present a more complex picture: the effect of GLP-1 on blood pressure appears to depend on the dose, species and CV status. In most studies of ‘normal’ (non pathological status) rats, systemic or intracerebroventricular (i.c.v) administration of GLP-1 increased blood pressure in a dose-dependent manner [38–41]. However, in one study, which used a wider range of GLP-1 doses (10–10,000 ng), lower doses (10 or 100 ng) were associated with either a hypertensive or a biphasic response (an increase followed by a decrease), whereas higher doses were associated with a consistent hypertensive effect [42].

It has been proposed that the delayed hypotensive component of the biphasic response is mediated by GLP-1(9–36), a metabolite of GLP-1, which was traditionally considered to be inactive but is now considered to play a beneficial role in its CV effects (Grieve et al., 2009) [43]. GLP-1 had no effect on blood pressure in larger species such as calves and pigs [44,45], but the doses were generally lower (i.e. picomolar range) than those used in rat studies. In addition to the increase in blood pressure, GLP-1 infusion also increases heart rate in rodents [38] and calves [44], but not in pigs [45].

As mentioned above, another factor influencing the effect of GLP-1 may be the pathophysiological status of the CV system. For example, in contrast to the hypertensive effects generally seen in ‘normal’ rats, chronic treatment with recombinant GLP-1 reduced the development of hypertension in Dahl salt-sensitive rats receiving a high-salt diet [46]. In contrast, in a model of hypovolaemic and hypotensive rats, infusion of GLP-1 was found to restore healthy blood pressure through stimulation of the neurohypophysial hormones, oxytocin and vasopressin [47].

The mechanisms of action of GLP-1 on blood pressure and heart rate are complex and involve both central and peripheral pathways [43]. The finding that increases in blood pressure in rats induced by GLP-1 or exendin-4 are prevented by treatment with the GLP-1 receptor antagonist exendin suggests that these effects are mediated via the GLP-1 receptor [39]. Several studies have suggested a role for the GLP-1 receptors expressed in the central nervous system (CNS). For example, the i.c.v. administration of exendin(9–39) has been shown to block the CV response to peripheral infusion of GLP-1 [39]. Furthermore, central and peripheral administration of GLP-1 receptor agonists (i.e. exendin-4) induce c-Fos expression in the adrenal medulla and medullary catecholamines neurons, suggesting that central GLP-1 receptors may mediate neuroendocrine responses.
and modulate sympathetic outflow [40]. The mechanism behind increased heart rate is unknown but may be due to an effect on the sympathetic nervous system.

The effects of GLP-1 on renal function—and in particular its effects on sodium and water balance—may also contribute to its antihypertensive effects. In the study in Dahl salt-sensitive rats described above, treatment with recombinant GLP-1 significantly increased urine flow over the first 3 days and sodium excretion over the first day, correlating with a step change in sodium intake; the cumulative sodium balance was negative on the first day, and over 3 days it was lower than that in vehicle-treated rats [46]. Diuretic and natriuretic effects of GLP-1/GLP-1(7–36) amide have also been described in other studies in normal and obese rats [48,49].

3.1.2. Studies in humans

This potential mechanism may also inform results from studies in humans. In healthy volunteers, native recombinant GLP-1 either had no significant effect on blood pressure [49,50] or resulted in a statistically significant, but modest, increase [51]. Short- and long-term administration of GLP-1 to patients with heart failure and those undergoing bypass surgery had no detectable chronotropic or pressor effects [43,52–54]. There are a number of small studies that have evaluated the effect of GLP-1 on blood pressure in patients with T2DM [23,50,55]. None of these studies reported a significant change in blood pressure (baseline values were either not reported [23,55] or not elevated, possibly due to antihypertensive medication [50]); however, a single infusion of GLP-1 produced an increase in urinary sodium excretion in both healthy and obese insulin-resistant individuals, following sodium load [56]. In obese subjects, GLP-1 has also been shown to reduce H+ secretion and the glomerular hyperfiltration rate, suggesting potential renoprotective effects for this agent [56].

3.2. The effects of native GLP-1 on the myocardium

3.2.1. Preclinical studies

Studies of the effects of GLP-1 on the myocardium often concentrate on its effects following ischaemic injury. In both ex vivo and in vivo models using rat hearts, GLP-1 added before ischaemic injury led to significant reductions in infarct size [57]. Similarly, in ex vivo rat hearts subjected to low-flow ischaemia, GLP-1 increased myocardial glucose uptake and enhanced recovery of cardiac function, demonstrating a similar effect to insulin [58]. In a canine model, a 24-hour perfusion of GLP-1 following transient ischaemia produced significantly quicker and more complete recovery of regional wall motion, with no change in systemic haemodynamics or overall systolic function [59]. Furthermore, in dogs with advanced induced dilated cardiomyopathy, a 48-hour infusion of GLP-1 led to a significant increase in left ventricular contractility, stroke volume and cardiac output, and significant reductions in left ventricular end-diastolic pressure, heart rate and systemic vascular resistance [59]. Treatment with GLP-1 also increased myocardial insulin sensitivity and myocardial glucose uptake in this model [59].

3.2.2. Studies in humans

The beneficial effects of GLP-1 in animal models have translated into positive effects in small studies in humans. For example, in patients with acute MI and severe left ventricular dysfunction (i.e. left ventricular ejection fraction [LVEF] < 40%) who had undergone angioplasty, a 72-hour infusion of GLP-1 significantly improved LVEF and both global and regional wall motion [60]. In a small pilot study of 12 patients with chronic heart failure, 5-week infusion of GLP-1 significantly improved left ventricular function, functional status and quality of life [53]. Finally, in a study of patients with preserved left ventricular function undergoing coronary artery bypass grafting, GLP-1 treatment before and after surgery improved glycaemic control and reduced patients’ requirements for high-dose insulin or inotropes [54].

3.2.3. Molecular mechanisms

Given the effects reported from in vivo studies, as described above, a direct action of GLP-1 on the ischaemic myocardium seems likely. The mechanisms for these beneficial effects of GLP-1 on ischaemic myocardium appear to be both GLP-1 receptor-mediated and GLP-1 receptor-independent. One theory is that GLP-1 may exert an antiapoptotic effect on myocardial cells, mediated via the GLP-1 receptor and activation of survival kinases, such as PI3 kinase, p42/44 mitogen-activated protein kinase, and the ERK1/2 pathway [57,61]. A study exploring the protective effects of GLP-1 in ex vivo rodent hearts against ischaemic infarction found that any benefits conferred by pretreatment with GLP-1 were lost when the GLP-1 receptor antagonist exendin(9–39) was introduced [57,62], indicating a receptor-dependent action. Direct proof for the involvement of GLP-1 receptor signalling in cardiac function came from the characterization of GLP-1 receptor knock-out mice (GLP-1R−/−) [63]. It was shown that the cardiac structure of 5-month-old GLP-1R−/− mice demonstrated an increase in left ventricular thickness, leading to impaired left ventricular contractility and diastolic function after insulin administration [63].

The picture is more complex, however; other effects on the myocardium appear to be independent of the GLP-1 receptor, as determined by studies comparing the effects of GLP-1 in animals with or without the receptor. Notably, the GLP-1 metabolite GLP-1(9–36) amide has been shown to augment left ventricular performance during reperfusion of isolated rat hearts [62]. These effects were only partially antagonised by exendin(9–39), indicating that this effect was mediated, at least in part, by a mechanism independent of the GLP-1 receptor [62]. Pretreatment with GLP-1 also increased functional recovery and cardiomyocyte viability after ischaemia-reperfusion injury of isolated mouse hearts, and many of these actions were preserved in mice lacking the GLP-1 receptor [36]. In the same study, administration of GLP-1(9–36) during reperfusion reduced ischaemic damage and increased vasodilation and coronary flow in mice both with and without the GLP-1 receptor [36]. The addition of an inhibitor of nitric oxide synthase significantly inhibited vasodilation in response to GLP-1 and GLP-1(9–36), and inhibition of DPP-4 activity abolished...
the beneficial effects of GLP-1 on hearts of mice lacking the GLP-1 receptor [36]. This led the authors to propose a novel 2-pathway mechanism for the CV actions of GLP-1: a GLP-1-receptor-dependent pathway, which produces the inotropic action, glucose uptake, ischaemic preconditioning and mild vasodilatory actions, and a second pathway, which depends on the rapid metabolism of GLP-1 to GLP-1(9–36). This second pathway exerts a GLP-1-receptor-independent effect on the post-ischaemic recovery of cardiac function and vasodilation [36]. Thus, it may be hypothesized that pharmacological inhibition of DPP-4 might be detrimental for cardiac function, by decreasing the production of GLP-1(9–36). However, it has been demonstrated that genetic deletion (DPP-4−/−) or pharmacological inhibition of DPP-4 by sitagliptin do not alter CV function in the hearts of diabetic and non diabetic mice [64]. In contrast, as observed with GLP-1 infusion, DPP-4−/− mice exhibit increased survival and myocardial viability after myocardial ischemic injury [64].

3.3. The effects of native GLP-1 on endothelial function

The endothelium plays a major role in maintaining CV homeostasis and health [65]. It mediates organ blood flow (via vasoconstriction and vasodilatation), blood vessel growth, inflammation and haemostasis via a complex system of chemical mediators [66]. Endothelial dysfunction – a disruption of any of these mediators or functions – can result in such atherogenic changes as rolling of leukocytes, smooth-muscle growth, impaired coagulation, vascular inflammation and chemokine release, which all predispose to atherosclerosis and thrombosis [67]. Several factors associated with T2DM, such as hyperglycaemia and dyslipidaemia, contribute to endothelial dysfunction [66–68].

Endothelial dysfunction may be measured indirectly by using either blood flow and reactivity to acetylcholine infusion, or the levels of certain markers, such as endothelin-1, plasminogen activator inhibitor-1 (PAI-1) and vascular cell adhesion molecule-1 (VCAM-1). In a culture of human endothelial cells, GLP-1 was shown to reduce TNF-α-induced expression of PAI-1 mRNA and production of PAI-1 protein in the presence of a DPP-4 inhibitor, suggesting that it may have a beneficial effect on endothelial function [69]. GLP-1 also reduced the expression of VCAM-1 in human endothelial cells by inhibiting expression of the receptor for advanced glycation end products, which are thought to play an important role in the vascular complications of T2DM [70]. In vivo experiments using isolated aortic rings from Dahl-S mice found that GLP-1 significantly improved endothelial function [46].

The results from these in vitro studies are supported by in vivo studies in humans. In healthy individuals (non-, normotensive and non-smokers), GLP-1 has been observed to enhance acetylcholine-induced vasodilation in the forearm [71], supporting its role in the homeostatic control of vascular function. In patients with T2DM and stable coronary artery disease, infusion of GLP-1 significantly improved endothelial dysfunction, as measured by flow-mediated vasodilation, compared with placebo [34].

3.4. The effects of native GLP-1 on lipid metabolism

High LDL-C and low HDL-C levels are independent risk factors for CV-disease, and disturbances in lipid metabolism are thought to play a key role in T2DM. The effects of GLP-1 on lipid metabolism are, therefore, particularly relevant to patients with T2DM.

3.4.1. In vitro studies

The effect of GLP-1 on lipid metabolism is complex. With the exception of one study [72], in vitro studies using normal adipocytes from rats and humans have shown that GLP-1 has both lipolytic and lipogenic effects [73–76]. It has been suggested that this dual action may depend on the dose, with lower doses being lipogenic and higher doses being lipolytic [74]. In adipocytes from rats with streptozotocin-induced diabetes and from obese human subjects, the lipogenic effect of GLP-1 was no longer observed [75–77].

3.4.2. Studies in animals

In vivo, following infusion of fat into the duodenum of rats, GLP-1 reduced intestinal lymph flow, triglyceride absorption and synthesis of apolipoproteins B and A-IV. Both of these apolipoproteins are structural elements of chylomicrons [78], and the clinical effects of reducing their production would be to limit the release of triglycerides into the circulation after lipid-containing meals. In contrast, inhibition of endogenous GLP-1 receptor signalling by the antagonist exendin(9–39) or the ablation of endogenous GLP-1 receptor signalling in mice lacking the GLP-1 receptor enhanced lipoprotein secretion. The authors therefore concluded that endogenous GLP-1 receptor signalling is essential for the control of intestinal lipoprotein biosynthesis and secretion in hamsters and mice [79]. Another in vivo preclinical study has shown that GLP-1 reinforces the inhibitory action of insulin on hepatic VLDL production in insulin-resistant mice, with a concomitant decrease in hepatic glucose production [80]. Central administration of GLP-1 has also been shown to decrease adipocyte lipid storage in lean (but not obese) mice [81], and to shift the use of energy sources from carbohydrates to lipids in chicks [82].

3.4.3. Studies in humans

It has been demonstrated that postprandial lipaemia, reflected by non fasting TG levels, is an independent CV risk factor [83]. Interestingly, in healthy human volunteers, an infusion of recombinant GLP-1 completely abolished the postprandial increase in triglyceride levels, and also reduced plasma levels of non esterified fatty acids in both the fasting and fed states [84].

4. GLP-1 receptor agonists: a new avenue for addressing cardiovascular comorbidities in type 2 diabetes?

As mentioned above, human GLP-1 is rapidly degraded by DPP-4, which limits its therapeutic value.

The first of the two main therapeutic strategies for utilising the incretin effect in T2DM therefore involves the inhibition of
DPP-4. Currently available DPP-4 inhibitors include sitagliptin, vildaglaptin, saxaglaptin and linaglaptin, all administered orally as a once-daily (sitagliptin, and saxaglaptin, linaglaptin) or twice-daily (vildaglaptin) treatment [85]. DPP-4 inhibitors have been shown to increase active GLP-1 levels 2- to 3-fold, by providing up to 90% inhibition of plasma DPP-4 activity over 24 hours [86].

The second therapeutic strategy focuses on the direct replacement of GLP-1 using exenatide or liraglutide, currently administered through subcutaneous (s.c.) injection [87]. Synthetic exendin-4 (licensed as exenatide) is a GLP-1 receptor agonist that has 53% sequence homology with human GLP-1 [88] and is resistant to cleavage by DPP-4, resulting in a half-life in humans of approximately 3.5 to 4 hours after s.c injection [89], making it suitable for twice-daily dosing [90]. A once-weekly formulation of exenatide is now available in many parts of the world. The second GLP-1 receptor agonist currently available, liraglutide, is a human GLP-1 analogue with 97% sequence identity to native GLP-1, the only differences being an amino-acid substitution at position 34 and the addition of a fatty acid moiety linked to position 26 [91]. Liraglutide has a half-life of 11 to 13 hours after s.c. injection, so is suitable for once-daily dosing [91,92].

Although DPP-4 inhibitors and GLP-1 receptor agonists have similar glucose-regulatory mechanisms, GLP-1 receptor agonists have additional, beneficial effects on food intake and body weight. GLP-1 receptor agonists achieve more potent control of glycaemia due to more potent and sustained receptor activation [24,89,90]. Although a meta-analysis of DPP-4 inhibitor clinical trials has shown positive CV outcomes for these therapies as well [91], this review focused primarily on GLP-1 receptor agonists.

4.1. The effect of GLP-1 receptor agonists on blood pressure and heart rate

Animal and human studies have not yet provided a clear answer on the effect of exenatide on blood pressure and heart rate. For example, exenatide has been shown to increase heart rate and depress heart rate variability in mice following i.c.v. administration [92]; however, in a small, 12-week, placebo-controlled study of patients with T2DM, exenatide produced a reduction in systolic blood pressure (−3.5 mmHg) but a small non significant increase in heart rate (2.1 beats/minute) [93]. In a larger study of patients with T2DM, exenatide treatment was not associated with any significant changes in heart rate or blood pressure [94]. In reports of studies of overweight patients with T2DM, 82-weeks’ treatment with exenatide was associated with reductions in both diastolic and systolic blood pressure, although some of these changes did not reach statistical significance [95,96].

During the phase III Liraglutide Effect and Action in Diabetes (LEAD) programme, liraglutide treatment consistently resulted in reductions in systolic blood pressure. Indeed, liraglutide was shown to reduce systolic blood pressure within 2 weeks of treatment initiation, an effect that was maintained throughout the 26-week studies [97,98]. These reductions were seen before patients had experienced substantial weight loss, suggesting that these two effects were independent of each other [98]. In the LEAD-6 head-to-head trial, liraglutide and exenatide treatment resulted in comparable reductions in both systolic and diastolic blood pressure [99]. A minor increase in heart rate of 3-4 beats/min has been observed in liraglutide-treated patients compared with baseline [99–103]. The mechanism behind the dose-independent increase in heart rate noted with liraglutide is yet to be determined.

4.2. The effect of GLP-1 receptor agonists on the myocardium in animal studies

Both exenatide and liraglutide have shown to reduce infarct size in animal models of ischaemia-reperfusion injury. For example, exenatide showed a strong infarct-limiting action after ischaemia-reperfusion injury in an isolated rat heart preparation, and improved left ventricular performance [62]. In vivo, 3-days’ treatment with exenatide significantly reduced infarct size and prevented deterioration of both systolic and diastolic function in a porcine model of ischaemia-reperfusion injury [104]. Exenatide was also found to increase myocardial salvage compared to placebo ($P = 0.003$) in patients with ST segment elevation myocardial infarction when administered intravenously from 15 minutes before through 6 hours after primary percutaneous coronary intervention [105]. Another GLP-1 receptor agonist, albiglutide, has also been shown to reduce myocardial infarct size and improve cardiac function following myocardial ischaemia-reperfusion injury in rats [106].

Pretreatment of both normoglycaemic and diabetic mice with liraglutide reduced infarct size and the incidence of cardiac rupture after chronic MI [107]. The treatment was also associated with improvements in survival (28 days after ligation, perioperative mortality was 20% in mice pretreated with liraglutide vs. 77% of mice pretreated with phosphate buffered saline (PBS); $P = 0.0001$) and cardiac output in these mice. In addition, in diabetic mice, liraglutide conferred cardioprotection and survival advantages over metformin treatment, despite the fact that the two therapies had equivalent effects on glycaemic control. These beneficial effects appear to be mediated by an antiapoptotic effect on cardiomyocytes [107].

Of note, clinical trials are ongoing to validate the effect of GLP-1 receptor agonists (i.e. exenatide) on cardiac function in diabetic patients with diastolic (clinicaltrial.gov identifier: NCT00799435) and congestive (clinicaltrial.gov identifier: NCT00766857) heart failure.

4.3. The effect of GLP-1 receptor agonists on lipids and cardiovascular biomarkers

4.3.1. Liraglutide

Treatment with liraglutide has been shown to inhibit tumour-necrosis-factor-alpha or hyperglycaemia-mediated induction of PAI-1, intercellular adhesion molecule-1 and VCAM-1 mRNA and protein expression in a human vascular endothelial cell line [108]. Liraglutide has also been shown to improve the lipid profile and several biomarkers of CV risk in humans. Indeed, in
patients with T2DM, liraglutide reduced levels of PAI-1 and B-type natriuretic peptide [109,110], as well as plasma triglycerides [111]. In the LEAD-6 trial conducted in patients with T2DM, both liraglutide and exenatide produced reductions in plasma lipids [99]. The reductions in triglycerides and free fatty acids were significantly greater for liraglutide than for exenatide [99].

### 4.3.2. Exenatide

Exenatide has demonstrated beneficial effects on biomarkers of CV-disease in both animal models and patients with T2DM. Exenatide reduced the recruitment and adhesion of monocytes to the aortic endothelium in mice, possibly reducing the development of atherosclerotic plaques [25]. In hamsters and mice, exenatide reduced postprandial intestinal secretion of triacylglycerol, cholesterol and apolipoprotein B-48 [79]. In obese patients with T2DM, exenatide significantly reduced both triglycerides and C-reactive protein [112]. Similarly, in studies of overweight patients with T2DM, exenatide improved lipid parameters with a statistically significant reduction in concentrations of triglycerides and an increase in high-density lipoprotein cholesterol [95,96]. Other parameters examined, such as total cholesterol, low-density lipoprotein cholesterol and apolipoprotein B, also showed numerical improvements, although these were not statistically significant [95,96]. In patients with impaired glucose tolerance or recent onset T2DM, a single subcutaneous injection of exenatide reduced postprandial elevation of triglycerides, apolipoproteins B-48 and CIII, remnant-lipoprotein-cholesterol and – triglyceride, and non esterified fatty acids [113]. Exenatide treatment of patients with T2DM for 1 year also increased levels of adiponectin and reduced high-sensitivity C-reactive protein [114]. These changes were statistically independent of the change in total body fat mass and body weight [114].

### 4.4. The effect of GLP-1 receptor agonists on major adverse cardiovascular events

#### 4.4.1. Liraglutide

A pooled analysis of major CV events was performed on all the completed intermediate/long-term randomised trials and their open-label extensions in the clinical development programme for liraglutide [115,116]. The point estimates of the incidence ratio for death, MI or stroke associated with liraglutide treatment was less than 1.0 compared to total comparator, indicating no increased CV risk with liraglutide. The upper 95% confidence interval was less than 1.8, which is a specified upper limit in FDA guidance for new antiglycemic drugs [117]. The effect of liraglutide on CV events will further be evaluated in the LEADER™ trial, a large-scale randomised, placebo-controlled CV outcomes trial (clinicaltrial.gov identifier: NCT01179048) [118].

#### 4.4.2. Exenatide

A similar retrospective analysis of pooled data from the exenatide clinical trial programme found the relative risk of customized primary major adverse CV events was also less than 1.0 for exenatide BID vs. comparators, suggesting that exenatide use also does not increase CV risk [116,119]. As with liraglutide, the upper 95% confidence interval was less than the FDA guidance level of 1.8. These results supported a previous retrospective database analysis for exenatide conducted using the LifeLink database of medical and pharmaceutical insurance claims for June 2005 to March 2009 [120]. Patients who initiated exenatide treatment were more likely to have prior ischaemic heart disease, obesity, hyperlipidaemia, hypertension and/or other comorbidities at baseline compared with those who started treatment with other glucose-lowering therapies. Exenatide-treated patients were less likely to have a CV-disease event than non-exenatide-treated patients; they also had lower rates of CV-disease-related hospitalisation and all-cause hospitalisation [120]. The CV safety profile of the once-weekly formulation of exenatide will be investigated in the EXSCEL trial (clinicaltrial.gov identifier: NCT01144338) [121].

### 4.4.3. DPP-4 inhibitors

A meta-analysis has demonstrated that DPP-4 inhibitors may have beneficial CV effects as well [91] and studies that are also underway to collect long-term data on these therapies will provide a clearer picture (TECOS: clinicaltrial.gov identifier: NCT00790205; SAVOR-TIMI: clinicaltrial.gov identifier: NCT01107886; CAROLINA: clinicaltrial.gov identifier: NCT01243424).

### 5. Conclusions

Based primarily on animal studies, with limited human data, GLP-1 and GLP-1-based therapies appear to have direct beneficial effects on the cardiovascular system, in addition to their glucose-lowering characteristics. The potential use of GLP-1-based therapies in patients with a high CV risk requires further investigation. As the FDA requires a demonstration that any new antidiabetes agent is not associated with an unacceptable risk of CV-disease, incretin-based therapies are currently being investigated in a large cardiovascular outcomes trial. Evidence from these interventional studies may shed light on the potential clinical cardiovascular benefit of these therapies for patients with T2DM.

### Disclosure of interest

B.C. has acted as an investigator for Boehringer-Ingelheim, Eli Lilly & Co, Genfit, Jansenn Cilag, Novo Nordisk, Roche and Sanofi-Aventis and has received fees for consultancy, speaking, travel, or accommodation from Astra-Zeneca, BMS, Eli Lilly & Co, Genfit, Pierre Fabre, Novartis, Novo Nordisk, Merck, Sanofi-Aventis, Servier, and Takeda.

### Acknowledgements

The writing assistance and support of Elien Moës, PhD, Watermeadow Medical plc, UK in preparing this review article is gratefully acknowledged. The preparation of this article was funded by Novo Nordisk France.
References


[70] Ishitahi Y, Matsui T, Takeuchi M, Yamagishi S. Glucagon-like peptide-1 (GLP-1) inhibits advanced glycation end product (AGE)-induced up-regulation of VCAM-1 mRNA levels in endothelial cells by suppressing AGE receptor (RAGE) expression. Biochem Biophys Res Commun 2010;391:1405–2148.


[82] Tachibana T, Oitkawa D, Adachi N, Boswell T, Furuse M. Intracerebroven-
tricular injection of glucagon-like peptide-1 changes lipid metabolism in
[83] Nordestgaard BG, Benn M, Schnohr P, Tybjerg-Hansen A. Nonfasting
triglycerides and risk of myocardial infarction, ischemic heart disease and
Glucagon-like peptide 1 abolishes the postprandial rise in triglyceride
concentrations and lowers levels of non esterified fatty acids in humans.
[85] Duez H, Cariou B, Staels B. DPP-4 inhibitors in the treatment of type 2
[86] Deacon CF. Dipeptidyl peptidase-4 inhibitors in the treatment of type 2
[87] Gilbert MP, Pratley RE. Efficacy and safety of incretin-based therapies in
[88] Madsbad S. Exenatide and liraglutide: different approaches to develop
GLP-1 receptor agonists (incretin mimetics) – preclinical and clinical
[89] Drucker DJ, Sherman SI, Nauck M, Baehr B, Madsbad S, Buse JB.
Incretin-based therapies for the treatment of type 2 diabetes:
Liraglutide versus sitagliptin for patients with type 2 diabetes who did not
have adequate glycaemic control with metformin: a 26-week, randomised,
peptidase-4 inhibitors in type 2 diabetes: a meta-analysis of randomized
receptor stimulation depresses heart rate variability and inhibits
neurotransmission to cardiac vagal neurons. Cardiovasc Res 2011;89:
830–6.
of exenatide on heart rate and blood pressure in subjects with type 2
diabetes mellitus: a double-blind, placebo-controlled, randomised pilot
[95] Lehnberg J, Veijstrup N, Kelberg H, Batker HE, Kim WY, Mathiasen AB,
et al. Exenatide reduces reperfusion injury in patients with ST-segment
elevation myocardial infarction. Eur Heart J 2011 [Epub ahead of print]
DOI:10.1093/eurheartj/ehr309.
DR, et al. Albiglutide, a long lasting glucagon-like peptide-1 analog,
protects the rat heart against ischemia/reperfusion injury: evidence for
AM, et al. GLP-1R agonist liraglutide activates cytoprotective pathways
and improves outcomes after experimental myocardial infarction in mice.
[98] Liu H, Dear AE, Knudsen LB, Simpson RW. A long-acting glucagon-
like peptide-1 analogue attenuates induction of plasminogen activator
[99] Plutzky J, Poulter NR, Falahati A, Toft AD, Davidson MH. Plasminogen
activator inhibitor-1 is reduced by the once-daily human glucagon-like
peptide-1 analog liraglutide when used in the treatment of type 2 diabetes.
Circulation 2009;120:S397.
[100] Courrèges JP, Vilsbøll T, Zdravkovic M, Le-Thi T, Krarup T, Schmitz O,
et al. Beneficial effects of once-daily liraglutide, a human glucagon-like
peptide-1 analogue, on cardiovascular risk biomarkers in patients with
[101] Vilsbøll T, Zdravkovic M, Le-Thi T, Krarup T, Schmitz O, Courrèges JP,
et al. Liraglutide, a long-acting human glucagon-like peptide-1 analog,
given as monotherapy significantly improves glycemic control and lowers
P. Exenatide therapy in obese patients with type 2 diabetes mellitus with
[103] Schwartz EA, Koska J, Mullin MP, Syoufi I, Schwenke DC, Reaven PD.
Exenatide suppresses postprandial elevations in lipids and lipoproteins
in individuals with impaired glucose tolerance and recent onset type 2
[104] Bundt MC, Diamant M, Elinsson B, Nordestgaard BG, Janssens RAJ,
et al. Exenatide effects on circulating cardiovascular risk biomarkers
KF, et al. Cardiovascular safety of liraglutide assessed in a patients-level
diabetes: evaluation of cardiovascular outcome results (LEADER™):
rationale and study design. Diabetes 2011;60(Suppl.1), 2303. [Abstract].
L. Cardiovascular safety of exenatide BID: an integrated analysis from
