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REVIEW

Selective targeting of glucagon-like peptide-1 signalling as a novel therapeutic approach for cardiovascular disease in diabetes

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Glucagon-like peptide-1 (GLP-1) is an incretin hormone whose glucose-dependent insulinotropic actions have been harnessed as a novel therapy for glycaemic control in type 2 diabetes. Although it has been known for some time that the GLP-1 receptor is expressed in the CVS where it mediates important physiological actions, it is only recently that specific cardiovascular effects of GLP-1 in the setting of diabetes have been described. GLP-1 confers indirect benefits in cardiovascular disease (CVD) under both normal and hyperglycaemic conditions via reducing established risk factors, such as hypertension, dyslipidaemia and obesity, which are markedly increased in diabetes. Emerging evidence indicates that GLP-1 also exerts direct effects on specific aspects of diabetic CVD, such as endothelial dysfunction, inflammation, angiogenesis and adverse cardiac remodelling. However, the majority of studies have employed experimental models of diabetic CVD and information on the effects of GLP-1 in the clinical setting is limited, although several large-scale trials are ongoing. It is clearly important to gain a detailed knowledge of the cardiovascular actions of GLP-1 in diabetes given the large number of patients currently receiving GLP-1-based therapies. This review will therefore discuss current understanding of the effects of GLP-1 on both cardiovascular risk factors in diabetes and direct actions on the heart and vasculature in this setting and the evidence implicating specific targeting of GLP-1 as a novel therapy for CVD in diabetes.

Abbreviations

ANP, atrial natriuretic peptide; CAD, coronary artery disease; CVD, cardiovascular disease; DPP-4, dipeptidyl peptidase-4; eNOS, endothelial NOS; GLP-1, glucagon-like peptide-1; hs-CRP, high sensitivity C-reactive protein; ICAM-1, intercellular adhesion molecule-1; MI, myocardial infarction; PAI-1, plasminogen activator inhibitor-1; STZ, streptozotocin; T1DM, type 1 diabetes mellitus; T2DM, type 2 diabetes mellitus; TLR, toll-like receptor; UKPDS, United Kingdom Prospective Diabetes Study; VCAM-1, vascular cell adhesion molecule-1

Tables of Links

TARGETS	
GPCRs[a]	**Enzymes**[e]
β_1-adrenoceptor	Akt (PKB)
β_2-adrenoceptor	DPP-4
GLP-1 receptor	eNOS
Ion channels[b]	ERK1/2
K_{ATP} channel	MMP-2
Catalytic receptors[c]	MMP-9
Toll-like receptor-2	p38-MAPK
Toll-like receptor-4	p42-MAPK
Other protein target	p44-MAPK
TNF-α	PI3K
Transporters[d]	PKA
Ca^{2+} ATPase	Src

LIGANDS	
ACh	Insulin
Adiponectin	Leptin
Alogliptin	Linagliptin
ANP	Liraglutide
Byetta	Lixisenatide
cAMP	Metformin
Exenatide	Nitric oxide (NO)
Exendin-4	Rosiglitazone
Glimepiride	Saxagliptin
Glipizide	Sitagliptin
GLP-1	Stromal cell-derived factor-1α
GLP-1(9-36)	VCAM-1
Glyburide	VEGF-A
ICAM-1	Victoza
IL-6	Vildagliptin

These Tables list key protein targets and ligands in this article which are hyperlinked to corresponding entries in http://www.guidetopharmacology.org, the common portal for data from the IUPHAR/BPS Guide to PHARMACOLOGY (Pawson *et al.*, 2014) and are permanently archived in the Concise Guide to PHARMACOLOGY 2013/14 ([a,b,c,d,e]Alexander *et al.*, 2013a,b,c,d,e).

Introduction

The prevalence of type 2 diabetes mellitus (T2DM) is increasing alarmingly with the 2013 figure of 382 million estimated to rise to 592 million by 2035 (International Diabetes Federation, 2014). A change in lifestyle coupled with an increase in obesity has led to a global epidemic, with diabetics typically carrying a fivefold greater mortality risk as a result of cardiovascular disease (CVD) compared with non-diabetics (Stamler *et al.*, 1993), and coronary artery disease (CAD) being the leading underlying cause (Bertoni *et al.*, 2004). It is well established that hyperglycaemia plays a central role in development and progression of CVD associated with diabetes (Nathan, 1996). Indeed, two long-term clinical trials, the Diabetes Control and Complications Trial/Epidemiology of Diabetes Interventions and Complications study and the United Kingdom Prospective Diabetes Study (UKPDS), have demonstrated that intensive glucose-lowering strategies are effective in markedly reducing the incidence of microvascular (e.g. retinopathy, nephropathy) and macrovascular (e.g. CAD, stroke) complications in both type 1 diabetes mellitus (T1DM) and T2DM (Holman *et al.*, 2008), although several similar large-scale trials have reported limited benefits (Action to Control Cardiovascular Risk in Diabetes Study Group, 2008; Duckworth *et al.*, 2009; Ginsberg, 2011). Nonetheless, there remains a significant incidence of CVD even in optimally treated diabetic patients, so it is clear that more effective strategies are required. In this regard, the incretin peptide hormone, glucagon-like peptide-1 (GLP-1), has received considerable recent attention.

The incretin effect is responsible for augmenting insulin secretion following nutrient ingestion and GLP-1 together with its sister hormone, gastrointestinal peptide, account for up to 60% of post-prandial insulin secretion, leading to rapid blood glucose reduction (Nauck *et al.*, 1986; Drucker *et al.*, 1987). Furthermore, they possess an inherent ability to reduce glucagon secretion (Kreymann *et al.*, 1987), delay gastric emptying (Näslund *et al.*, 1998a) and promote satiety (Flint *et al.*, 1998). The metabolic actions of GLP-1 are mediated by GLP-1 receptor activation and stimulation of cAMP and several downstream kinases, including ERK1/2, PI3K and PKA. Under physiological conditions, GLP-1 has a short half-life (~2 min) as it is rapidly degraded by its endogenous inhibitor, dipeptidyl peptidase-4 (DPP-4) (Deacon *et al.*, 1995), resulting in cleavage of two amino acids from native GLP-1(7-36) to produce GLP-1(9-36), which acts as a weak GLP-1 receptor antagonist lacking insulinotropic activity (Green *et al.*, 2004). However, emerging evidence suggests that 'metabolically inactive' GLP-1(9-36) may itself be an important signalling molecule (Ban *et al.*, 2010; Gardiner *et al.*, 2010). More detailed information on GLP-1 biology and signalling is provided by recent review articles (Grieve *et al.*, 2009; Donnelly, 2012; Pabreja *et al.*, 2013).

The unique ability of GLP-1 to promote insulin secretion in a glucose-dependent manner has been harnessed for treatment of T2DM, with GLP-1 receptor agonists resistant to DPP-4 (exenatide, Byetta®; liraglutide, Victoza®), and DPP-4 inhibitors (e.g. sitagliptin, vildagliptin) now widely used for effective glycaemic control. Interestingly, it is well recognized that GLP-1 exerts wide-ranging extra-pancreatic actions occurring independently of its established metabolic effects. Indeed, GLP-1 signalling is reported to play several important roles in the CVS in both health and disease (Grieve *et al.*, 2009), although it appears that the GLP-1 receptor may not

be as widely expressed as previously thought. For example, recent work suggests that cardiac GLP-1 receptor expression may be localized to atrial tissue, sinoatrial node and vasculature, with some species variation (Kim *et al.*, 2013; Pyke *et al.*, 2014; Richards *et al.*, 2014), and that earlier reports of more ubiquitous expression may be questionable because of poor antibody selectivity and sensitivity (Panjwani *et al.*, 2012). Nonetheless, it is clear that GLP-1 exerts important cardiovascular actions, although it is only recently that its effects in the setting of diabetes, a condition synonymous with micro/macrovascular complications, have been explored. This is clearly important because of the large number of patients receiving GLP-1-based therapies, in which its cardiovascular actions are largely unknown. This review will therefore discuss the current understanding of the effects of GLP-1 on both cardiovascular risk factors in diabetes and direct actions on the heart/vasculature in this setting and the evidence implicating specific targeting of GLP-1 as a novel therapy for CVD in diabetes, with a primary focus on the role of GLP-1 receptor agonists. More detailed discussion of the pleiotropic actions of DPP-4 inhibitors in this setting is provided by recent review articles specifically focused on this important aspect of cardiovascular GLP-1 signalling (Scheen, 2013; Aroor *et al.*, 2014).

Influence of GLP-1 on cardiovascular risk factors in diabetes

BP and hypertension

Increased BP is an established risk factor for CVD in both normoglycaemia and T2DM (Turner *et al.*, 1998; Vasan *et al.*, 2001). Notably, therapeutic reductions in BP and circulating glucose have an additive effect in decreasing cardiovascular complications in T2DM patients, as highlighted by the UKPDS (Stratton *et al.*, 2006). Indeed, in an experimental setting, chronic GLP-1 infusion inhibits development of hypertension in Dahl salt-sensitive rats, as well as reducing cardiac fibrosis and hypertrophy, effects which appear to occur via a natriuretic/diuretic mechanism independently of blood glucose (Yu *et al.*, 2003), suggesting that GLP-1 may confer additional benefits which could be harnessed for the treatment of hypertension associated with T2DM. Consistent with an indirect BP-lowering effect, it was recently reported that liraglutide-stimulated reduction of angiotensin II-induced hypertension in mice was blocked by the natriuretic peptide receptor antagonist, anantin, in a GLP-1 receptor-dependent manner, but unaltered by the NOS inhibitor, N^G-monomethyl-L-arginine, and that liraglutide induced rapid increases in atrial natriuretic peptide (ANP) secretion both *in vivo* and in isolated perfused hearts, suggesting that observed BP reduction occurred at least partly via direct activation of cardiac ANP (Kim *et al.*, 2013). Importantly, in the context of diabetes, the GLP-1 mimetic, exendin-4, inhibited development of both spontaneous and high salt-induced hypertension in obese db/db mice via beneficial actions on renal sodium handling (Hirata *et al.*, 2009). Furthermore, it was recently reported that treatment of insulin-resistant Zucker rats with the DPP-4 inhibitor, linagliptin, for 8 weeks reduced BP and improved diastolic function (Aroor *et al.*, 2013).

Interestingly, although chronic administration of GLP-1 may prevent development of hypertension, it is widely reported that acute GLP-1 exposure is associated with increased BP and heart rate, which predisposes to CVD. For example, acute infusion of GLP-1(7-36) increased systolic/diastolic BP and heart rate in both normal and insulin-deficient streptozotocin (STZ)-induced T1DM rats (Barragán *et al.*, 1994), with the same group reporting that exendin-4-induced increases in BP and heart rate were reversed by the GLP-1 receptor antagonist, exendin(9-39) (Barragán *et al.*, 1996), suggesting that these effects occurred via an insulin-independent mechanism but involving GLP-1 receptor activation. Although similar increases in BP and heart rate after short-term GLP-1 administration have been reported in various experimental models (Grieve *et al.*, 2009), the data from clinical studies are less clear. For example, GLP-1 infusion in a small number of T2DM patients with or without CAD for 105 min and 48 h, respectively, had no effect on heart rate or systolic/diastolic BP (Toft-Nielsen *et al.*, 1999; Nyström *et al.*, 2004), whereas 48 h GLP-1 infusion in patients with ischaemic heart failure (Halbirk *et al.*, 2010) and treatment of T2DM patients with an exendin-transferrin fusion protein or exenatide for 7 or 10 days, respectively (Kothare *et al.*, 2008; Gustavson *et al.*, 2011), resulted in elevated heart rate and diastolic BP. However, the data from longer-term GLP-1 clinical trials are more consistent, with the majority reporting decreased BP and minimal effects on heart rate. For example, the Liraglutide Effect and Action in Diabetes (LEAD)-4 study, investigating 26 week liraglutide treatment in combination with metformin in T2DM patients, reported a modest reduction in systolic BP of 6 mmHg compared with placebo (1.1 mmHg) (Zinman *et al.*, 2009). Similarly, the LEAD-2 study, assessing liraglutide combination therapy, reported a 2–3 mmHg decrease in systolic BP versus a small increase (0.4 mmHg) in the glimepiride control group (Nauck *et al.*, 2009). Furthermore, a 30 week trial comparing once-weekly versus twice-daily exenatide injection in drug-naïve T2DM patients observed a significant decrease in systolic/diastolic BP compared with baseline (Drucker *et al.*, 2008), while another 20 week trial in obese patients reported reduced systolic/diastolic BP in response to liraglutide, which persisted for the 2 year follow-up period (Astrup *et al.*, 2012). Interestingly, meta-analysis of six clinical trials comprising 2171 T2DM patients found that exenatide treatment for 6 months produced maximal systolic BP reduction in individuals with abnormally high baseline levels, whereas no effects were observed in normotensive subjects (Okerson *et al.*, 2010). It should be noted that BP reduction is positively correlated with weight loss (Neter *et al.*, 2003), so it is possible that the observed changes after chronic GLP-1 treatment may occur secondary to its metabolic effects. However, although beneficial effects of GLP-1 on body weight are associated with improved hypertension, it is clear that this cannot solely account for its vascular effects as several studies have reported a BP reduction prior to weight loss. For example, a combined meta-analysis of three 26 week liraglutide trials reported decreased BP after only 2 weeks, while maximal weight loss did not occur until 8 weeks (Gallwitz *et al.*, 2010). Indeed, heart rate, which is not linked to body weight, was increased by chronic administration of both liraglutide and exenatide in T2DM patients in parallel with reduced systolic BP (Garber

et al., 2009; Gill *et al.*, 2010). Interestingly, it was recently reported that GLP-1 secretory function increases with age and is negatively correlated with systolic BP, suggesting that this may represent an adaptive response (Yoshihara *et al.*, 2013).

Dyslipidaemia

The pathophysiology of diabetes is commonly considered largely in terms of associated hyperglycaemia. However, it is increasingly apparent that dyslipidaemia is equally important and represents a significant risk factor for CVD in diabetic patients (Reiner *et al.*, 2011). It is likely that impaired insulin sensitivity contributes to dyslipidaemia in T2DM, which is associated with reduced GLP-1 secretion. Indeed, in addition to their established glycaemic actions, GLP-1 receptor activation and DPP-4 inhibition are reported to improve lipid profiles in both experimental and clinical diabetes. For example, short-term infusion of GLP-1 in normoglycaemic Syrian golden hamsters decreased lipid absorption and triglyceride levels, an effect potentiated by oral glucose (Hein *et al.*, 2013), suggesting that its incretin action may inhibit intestinal production of chylomicrons, which are strongly linked to atherosclerosis (Nakano *et al.*, 2008). Similarly, circulating triglycerides and fat pad mass in rats with diet-induced obesity were reduced after 4 week treatment with liraglutide (Madsen *et al.*, 2010), while 40 day administration of exendin-4 ameliorated systemic and cardiac insulin resistance and dyslipidaemia in both genetic KKAy and diet-induced T2DM mouse models (Monji *et al.*, 2013). Furthermore, chronic GLP-1 receptor activation with both the GLP-1 analogue, CNTO3649, and exendin-4 in apolipoprotein E3-Leiden transgenic mice, which develop severe hypercholesterolaemia after high-fat feeding, resulted in reduced very-low-density lipoprotein (VLDL)-triglyceride and apolipoprotein B synthesis in parallel with decreased hepatic triglyceride, cholesterol and phospholipids and lipogenesis gene expression (Parlevliet *et al.*, 2012). The long-acting DPP-4 inhibitor, teneligliptin, is also reported to decrease circulating triglyceride and free fatty acid levels in insulin-resistant Zucker fatty rats after 2 week treatment (Fukuda-Tsuru *et al.*, 2012).

Importantly, the majority of clinical studies have also demonstrated beneficial outcomes of GLP-1 administration on lipid metabolism in T2DM, such as reduced circulating triglycerides and low-density lipoprotein (LDL) cholesterol (Flock *et al.*, 2007; Drucker *et al.*, 2008; Tremblay *et al.*, 2011), although one study in which patients were treated with exenatide for 24 weeks reported a similar plasma lipid profile versus controls (Moretto *et al.*, 2008). For example, decreased circulating levels of atherogenic triglyceride-rich lipoproteins were observed following 4 week vildagliptin monotherapy in drug-naïve T2DM patients, characterized by specific reductions in total plasma and chylomicron triglycerides, together with apolipoprotein B-48 and cholesterol in the chylomicron subfraction (Matikainen *et al.*, 2006). Furthermore, the LEAD-4 study found that 26 week liraglutide treatment in combination with metformin and rosiglitazone decreased circulating LDL cholesterol, triglycerides and free fatty acids in T2DM patients compared with placebo controls, although it is interesting to note that these changes were greater in response to low-dose treatment (Zinman *et al.*, 2009). Indeed, similar results are reported for DPP-4 inhibitors, which produce much lower circulating levels of GLP-1. For example, twice-daily sitagliptin led to a significant reduction in circulating triglycerides and free fatty acids in a large number of T2DM patients compared with placebo and glipizide control groups, despite similar decreases in fasting plasma glucose and HbA1c levels (Scott *et al.*, 2007). Interestingly, a single injection of exenatide was shown to attenuate postprandial increases in triglycerides, apolipoprotein B-48 and CIII/remnant lipoprotein cholesterol for up to 8 h in patients with impaired glucose tolerance and recent-onset T2DM (Schwartz *et al.*, 2010), suggesting that such lipid profile benefits may not be explained solely by chronic changes in body weight, glucose levels and insulin resistance. Indeed, it was recently reported that exendin-4 completely reverses hepatic steatosis in mice fed a high-fat diet via a GLP-1 receptor-dependent mechanism resulting in reduced numbers/size of circulating VLDL-triglyceride and VLDL-apolipoprotein B particles (Parlevliet *et al.*, 2012), suggesting that GLP-1 may exert direct effects on dyslipidaemia in diabetes.

Obesity

Although it is well known that obesity significantly increases the risk of T2DM (Willett *et al.*, 1999), and both are independent risk factors for CVD (Hubert *et al.*, 1983), many established diabetes therapies, including sulfonylureas and thiazolidinediones, may increase body weight. However, GLP-1 reduces body weight because of beneficial effects on glucagon secretion, gastric emptying and satiety (Kreymann *et al.*, 1987; Flint *et al.*, 1998; Näslund *et al.*, 1998a), so it seems likely that impaired GLP-1 secretion observed in non-diabetic obese individuals (Holst *et al.*, 1982; Näslund *et al.*, 1998b) may at least partly account for their increased body weight. Indeed, weight loss improves the postprandial GLP-1 response in severely obese patients (Verdich *et al.*, 2001), suggesting that the two are interlinked. Furthermore, a 20 week treatment with liraglutide was reported to cause significant weight loss in obese individuals and to reduce the incidence of prediabetes (Astrup *et al.*, 2009), confirming an apparent role for GLP-1 in weight control which may be harnessed for therapeutic benefit. This assertion is supported by the LEAD trials which have consistently reported a reduction in body weight in T2DM patients following liraglutide treatment (Moretto *et al.*, 2008; Buse *et al.*, 2009; Garber *et al.*, 2009; Nauck *et al.*, 2009; 2013; Zinman *et al.*, 2009). For example, in the LEAD-2 trial, 26 week combination therapy of liraglutide with metformin in T2DM patients resulted in increased weight loss compared with metformin alone (Nauck *et al.*, 2013). Importantly, weight loss associated with both liraglutide and exenatide treatment is reported to be linked to improved cardiovascular risk factors, such as HbA1c and BP, and to persist for at least 2 years (Klonoff *et al.*, 2007; Astrup *et al.*, 2009; 2012), highlighting important benefits of GLP-1 which may not be related to its insulinotropic actions. The long-term effects of GLP-1 on weight loss may be particularly important as conventional weight loss is typically poorly maintained in T2DM patients. Despite GLP-1 receptor agonists promoting weight loss in both diabetic and non-diabetic obese subjects, DPP-4 inhibitors appear to be weight-neutral (Amori *et al.*, 2007), suggesting that GLP-1 receptor agonists may exert direct gastrointestinal effects in addition to improving insulin resistance (Rask *et al.*, 2001), although this

could simply be due to differences in circulating GLP-1 levels. However, postprandial GLP-1 levels are reported to be increased immediately after gastric bypass surgery, despite patients remaining obese, indicating that GLP-1 may regulate appetite and food intake directly (Morinigo *et al.*, 2006). Indeed, in T2DM patients, GLP-1 promotes satiety, thereby reducing energy consumption (Gutzwiller *et al.*, 1999), while in healthy individuals i.v. administration of GLP-1(7-36) slows gastric emptying in a dose-dependent manner (Nauck *et al.*, 1997).

GLP-1 and vascular disease

Vascular function

Impaired endothelial and vascular function are established as key initiating factors underlying the development of microvascular and macrovascular complications associated with diabetes. Indeed, it has been known for some time that native GLP-1(7-36) induces *ex vivo* dose-dependent vasodilatation in a number of isolated rodent vessels, including aorta (Golpon *et al.*, 2001; Green *et al.*, 2008), pulmonary artery (Richter *et al.*, 1993; Golpon *et al.*, 2001), femoral artery (Nyström *et al.*, 2005) and mesenteric artery (Ban *et al.*, 2008), although several different mechanisms have been proposed. For example, some studies indicate that the vasorelaxant actions of GLP-1 are dependent upon endothelium-derived NO (Golpon *et al.*, 2001; Ban *et al.*, 2008; Gaspari *et al.*, 2011), whereas others have proposed endothelium-independent mechanisms involving mediators such as K_{ATP} channels, cAMP and β_2-adrenoceptor activation (Nyström *et al.*, 2005; Gardiner *et al.*, 2008; Green *et al.*, 2008). Interestingly, although several studies suggest that the vascular actions of GLP-1 are dependent upon the GLP-1 receptor (Gaspari *et al.*, 2011; Chai *et al.*, 2012), it appears that they may also be mediated, at least partly, by its truncated metabolite, GLP-1(9-36), which induces dose-dependent relaxation in both isolated mouse mesenteric artery (Ban *et al.*, 2008) and rat aorta (Green *et al.*, 2008). It should be noted that although the synthetic GLP-1 mimetic, exendin-4, exerts similar actions in rat aorta (Golpon *et al.*, 2001; Green *et al.*, 2008), they are of reduced magnitude compared with GLP-1(7-36) and are absent in mouse mesenteric artery (Ban *et al.*, 2008). Importantly, the vasorelaxant actions of GLP-1 are also reported *in vivo*. For example, systemic administration of GLP-1(7-36) by both bolus dose and short-term infusion in rats induced hindquarters vasodilatation (Gardiner *et al.*, 2010). Interestingly, however, GLP-1 promoted vasoconstriction in both mesenteric and renal arteries, while exendin-4 exerted similar vasoconstriction in mesenteric artery but induced vasodilatation in both hindquarters and renal artery (Gardiner *et al.*, 2008), suggesting differential vascular effects. Indeed, a similar study demonstrated that GLP-1(7-36) infusion acutely increased muscle microvascular blood volume in the absence of changes in microvascular blood flow velocity or femoral blood flow, in association with increased plasma NO, muscle insulin clearance/uptake, hindlimb glucose extraction and muscle interstitial oxygen saturation (Chai *et al.*, 2012). It should be noted that in contrast to the *ex vivo* studies, GLP-1(9-36) failed to modulate vascular function in rats *in vivo* when given as either a bolus dose or via short-term infusion, which together with the fact that DPP-4 inhibitors prolonged the vascular actions of native GLP-1(7-36) in this setting (Gardiner *et al.*, 2010), suggest that the actions of this 'inactive' metabolite may not be significant *in vivo*.

Importantly, it appears that the vascular effects of GLP-1 are also evident in the setting of diabetes, where they are reported to promote beneficial actions. Chronic treatment of STZ/nicotinamide T1DM rats with either GLP-1(7-36) or exendin-4 was shown to prevent endothelial dysfunction in parallel with reduction of blood glucose (Özyazgan *et al.*, 2005), effects which may be mediated via activation of endothelial NOS (eNOS) (Goyal *et al.*, 2010). Although similar studies have not been performed in the setting of overt T2DM, it was recently reported that chronic treatment of insulin-resistant Zucker rats with the DPP-4 inhibitor, linagliptin, resulted in reduced hypertension in parallel with increased expression of total/phosphorylated eNOS (Aroor *et al.*, 2013). Furthermore, high-fat fed apolipoprotein E-deficient mice treated with a different DPP-4 inhibitor, des-fluoro-sitagliptin, demonstrated attenuation of endothelial dysfunction in parallel with eNOS activation and improved glucose tolerance (Matsubara *et al.*, 2012). Interestingly, however, endothelial dysfunction in rat femoral artery induced by short-term triglyceride exposure was not affected by exendin-4 (Nathanson *et al.*, 2009), suggesting that the reported *in vivo* protective actions may occur via indirect mechanisms. In this regard, it is important to note that the vascular actions of GLP-1 in diabetes are likely to occur, at least partly, secondary to stimulation of insulin, which induces vascular relaxation via Ca^{2+}-dependent activation of eNOS (Han *et al.*, 1995; Kahn *et al.*, 1998).

In addition to the data supporting important vascular actions of GLP-1 in experimental diabetes, several studies have reported beneficial functional effects in the clinical setting. For example, in T2DM patients with stable CAD, acute GLP-1 administration improved brachial artery flow-mediated vasodilatation, an effect not observed in healthy individuals (Nyström *et al.*, 2004). Comparable effects were observed in insulin-resistant patients with obesity-related metabolic syndrome, where acute treatment with GLP-1(7-36) enhanced insulin-mediated forearm blood flow responses to both ACh and sodium nitroprusside in the absence of changes in forearm glucose extraction/uptake, while GLP-1(9-36) did not affect vascular function (Tesauro *et al.*, 2013). Similarly, in patients with T1DM, brachial artery endothelial dysfunction induced by acute blood glucose modulation was counteracted by simultaneous infusion of GLP-1 (Ceriello *et al.*, 2013). Interestingly, GLP-1-induced enhancement of endothelium-dependent peripheral vasodilatation observed in non-diabetic individuals is differentially modulated by sulphonylureas, with glyburide abolishing GLP-1-induced ACh-mediated responses which are unaltered by glimepiride (Basu *et al.*, 2007). Furthermore exenatide, which is commonly used for hyperglycaemic control in T2DM, also increased postprandial endothelial function assessed by peripheral arterial tonometry in patients with recent-onset disease when given as a single dose, largely secondary to a reduction in circulating triglycerides (Koska *et al.*, 2010), although chronic treatment for 3 months in obese prediabetic patients had no additional effect when compared with

those receiving metformin (Kelly *et al.*, 2012). While it appears that clinical GLP-1 administration exerts acute vascular effects in T2DM, data on its chronic actions in this setting are variable. T2DM patients who received exenatide for a period of 4 months as an adjunct to standard metformin therapy demonstrated improved brachial artery flow-mediated dilatation, indicated by elevated peak dilatation and shear rate which are reflective of improved macrovascular and microvascular function respectively (Irace *et al.*, 2013). Notably, enhanced vasodilatation in exenatide-treated patients in this study was significantly greater than that observed in those receiving glimepiride as an add-on to metformin therapy. Furthermore, liraglutide treatment for 12 weeks in T2DM patients well controlled on metformin monotherapy resulted in an improvement in both circulating markers of vascular function [asymmetric dimethylarginine, plasminogen activator inhibitor-1 (PAI-1), E-selectin] and retinal microvascular endothelial function (Forst *et al.*, 2012). However, a similar study in a small number of severely obese T2DM patients chronically treated with GLP-1 receptor agonists for 6 months reported that neither exenatide or liraglutide had any effect on brachial artery endothelial-dependent flow-mediated dilation (Hopkins *et al.*, 2013), indicating potential for other confounding factors in this setting which may need to be considered. Furthermore, 6 week treatment with sitagliptin or alogliptin significantly reduced flow-mediated dilatation in male T2DM patients (Ayaori *et al.*, 2013), suggesting that chronically increased physiological levels of GLP-1 may exert unfavourable vascular actions, although it is possible that this could be a class-specific effect of DPP-4 inhibitors warranting further investigation.

Inflammation and atherosclerosis

It is well known that the incidence and progression of endothelial dysfunction is exacerbated in T2DM, secondary to established risk factors, such as insulin resistance, dyslipidaemia and hyperglycaemia. Endothelial dysfunction in this setting is characterized by an elevation in circulating adhesion molecules, such as intercellular adhesion molecule-1 (ICAM-1) and vascular cell adhesion molecule-1 (VCAM-1), and an increased propensity to develop atherosclerosis which is typified by inflammatory cell infiltration and plaque formation (Van Gaal *et al.*, 2006). Interestingly, several recent clinical and experimental studies appear to indicate that GLP-1 exerts both anti-inflammatory and anti-atherogenic actions. For example, GLP-1 treatment in T2DM patients is associated with beneficial effects on a number of established CVD biomarkers, including high sensitivity C-reactive protein (hs-CRP) and PAI-1, which are important in atherosclerosis development (Haffner, 2006). Similarly, 14 week treatment of T2DM patients with liraglutide resulted in significantly reduced PAI-1 levels, and a dose-dependent decrease in plasma hs-CRP levels (Vilsbøll *et al.*, 2007; Courrèges *et al.*, 2008), an effect that was also observed after 26 week treatment with exenatide (Bergenstal *et al.*, 2010) and was over and above that seen in patients treated with insulin glargine (Diamant *et al.*, 2010). Importantly, the beneficial effects of exenatide on circulating hs-CRP appear to persist at 1 year treatment in T2DM patients receiving standard metformin therapy, resulting in reduced levels of both hs-CRP and leptin (Bunck *et al.*, 2010). DPP-4 inhibitors seem to exert similar effects as T2DM patients receiving sitagliptin for 6 months also demonstrated significant reductions in plasma hs-CRP, together with VCAM-1 and associated albuminuria, which may attenuate glucose excursion and inhibit vascular inflammation (Horváth *et al.*, 2009; Hattori, 2010). Interestingly, a recent study demonstrated that cessation of exenatide treatment resulted in the reversal of benefits on circulating hs-CRP within 6 months (Varanasi *et al.*, 2011). It should be noted that although these studies support the suggestion that GLP-1 may protect against inflammation and atherosclerosis in the clinical setting, it is not possible to draw clear conclusions because of the absence of longer-term studies specifically assessing effects on disease development. Interestingly, a recent study has reported a positive correlation between circulating GLP-1 levels and CAD in both diabetic and non-diabetic patients undergoing angiography because of typical or atypical chest pain, highlighting the possibility that GLP-1 may exert detrimental effects in this setting (Piotrowski *et al.*, 2013).

Nonetheless, the majority of clinical data are broadly supportive of the anti-inflammatory actions of GLP-1, which persist for up to 12 weeks in obese T2DM patients after a single exenatide injection (Chaudhuri *et al.*, 2011). Indeed, in this study, GLP-1 was associated with a specific reduction of several inflammatory mediators, including TNF-α, toll-like receptor-2 (TLR-2) and TLR-4, in parallel with suppression of NF-κB signalling and MMP-9 activity, which are key initiating factors of atherosclerosis. Furthermore, in obese T2DM patients, 8 week liraglutide treatment is reported to decrease levels of the inflammatory macrophage activation molecule, sCD163, and pro-inflammatory cytokines, TNF-α and IL-6, while increasing levels of the anti-inflammatory adipokine, adiponectin (Hogan *et al.*, 2014). Importantly, these clinical observations are supported by a number of experimental studies which have specifically assessed the effects of GLP-1 on development and progression of atherosclerosis. For example, continuous infusion of exendin-4 in both wild-type and apolipoprotein E-deficient normoglycaemic mice was reported to decrease monocyte adhesion and development of atherosclerotic lesions in thoracic aorta, effects proposed to occur via cAMP/PKA-dependent suppression of inflammation (Arakawa *et al.*, 2010). These findings were confirmed by a different group who demonstrated reduced aortic macrophage recruitment, foam cell formation and atherosclerotic lesion development in apolipoprotein E-deficient mice after GLP-1 infusion (Nagashima *et al.*, 2011). Furthermore, chemokine-induced migration of $CD4^+$ lymphocytes is inhibited by both GLP-1(7-36) and exendin-4 in a GLP-1 receptor-dependent manner (Marx *et al.*, 2010), while liraglutide suppresses NF-κB signalling in HUVECs and THP-1 monocyte adhesion in human aortic endothelial cells via downstream activation of several proinflammatory and cell adhesion molecules, including TNF-α, VCAM-1 and E-selectin (Shiraki *et al.*, 2012; Krasner *et al.*, 2014). Indeed, liraglutide inhibits TNF-α in human vascular endothelial cells and reduces hyperglycaemia-mediated PAI-1, ICAM-1 and VCAM-1 activation, which is associated with endothelial dysfunction and accelerated atherogenesis in T2DM (Liu *et al.*, 2009). Furthermore, the DPP-4 inhibitor, des-fluoro-sitagliptin, is reported to exert cAMP-dependent anti-inflammatory actions in cultured human macrophages by increasing GLP-1 levels and to

reduce atherosclerotic lesion formation in apolipoprotein E-deficient mice (Matsubara *et al.*, 2012), while alogliptin inhibits vascular monocyte/macrophage recruitment and reduces atherosclerotic burden in high-fat diet-fed, LDL receptor-deficient mice in association with improvement of metabolic indices (Shah *et al.*, 2011). Taken together, these experimental data clearly support an important role for GLP-1 in protecting against vascular inflammation and atherogenesis, effects which are borne out by the reported clinical benefits of GLP-1 treatment on circulating inflammatory mediators and CVD biomarkers. However, it is evident that long-term studies specifically investigating effects on atherosclerotic disease development and progression are required to ascertain whether the apparent protective effects of GLP-1 under both normoglycaemic and diabetic conditions may translate to the clinical setting.

Angiogenesis

Abnormal angiogenesis is a hallmark of CVD which is exacerbated by diabetes, with impaired neovascularization contributing significantly to progression of ischaemic disease associated with peripheral and coronary arteries. Interestingly, it is becoming apparent that GLP-1 may modulate angiogenesis suggesting that such actions may underlie some of its reported beneficial cardiovascular effects. For example, exendin-4 stimulates proliferation of human coronary artery endothelial cells in a GLP-1 receptor-dependent manner via downstream activation of eNOS, PKA and PI3K/Akt signalling (Erdogdu *et al.*, 2010) and promotes *in vitro* HUVEC migration, *ex vivo* aortic sprouting angiogenesis and *in vivo* blood vessel formation in Matrigel plugs (Kang *et al.*, 2013), while native GLP-1(7-36) stimulates *in vitro* angiogenesis in HUVECs via Akt, Src and PKC-dependent pathways (Aronis *et al.*, 2013), suggesting that GLP-1 may directly modulate neovascularization. Importantly, these effects appear to translate to the pathological situation, with several recent studies reporting that GLP-1 can promote the pro-angiogenic actions of mesenchymal stem cells in different disease settings. Intracoronary artery delivery of GLP-1 eluting encapsulated human mesenchymal stem cells in a porcine model of experimental myocardial infarction (MI) resulted in improved left ventricular function and remodelling which was associated with increased infarct zone angiogenesis (Wright *et al.*, 2012), while peri-adventitial treatment of porcine vein grafts with these cells inhibited neointima formation in parallel with accelerated adventitial angiogenesis (Huang *et al.*, 2013). Furthermore, the addition of GLP-1 to encapsulated human mesenchymal cells significantly improved blood flow recovery and foot salvage in a mouse model of hindlimb ischaemia via increased capillary and arteriole formation secondary to paracrine activation of VEGF-A (Katare *et al.*, 2013). Although the majority of work investigating the pro-angiogenic actions of GLP-1 has been performed in normoglycaemic models, a recent study reported similar beneficial effects in the setting of diabetes. Impaired myocardial angiogenesis in STZ-treated T1DM rats and associated fibrosis and diastolic dysfunction were reversed by genetic deletion of DPP-4 or pharmacological inhibition with vildagliptin (Shigeta *et al.*, 2012). Interestingly, this study identified DPP-4 as being membrane-bound and localized to the cardiac capillary endothelium with increased expression in diabetes, which together with a report of increased binding affinity of GLP-1 to the coronary endothelium but not cardiomyocytes in isolated perfused T1DM rat hearts (Barakat *et al.*, 2011), supports a key endothelial-specific role of GLP-1 in this setting. Although these data provide supportive evidence for pro-angiogenic actions of GLP-1 in diabetes, it is clear that additional mechanistic studies are required using different CVD models in order to define its precise role. Furthermore, it is important to assess the effects of GLP-1 therapy in diabetic patients in order to investigate whether the apparent pro-angiogenic effects of GLP-1 translate to the clinical setting and are of functional relevance.

GLP-1 and the diabetic myocardium

The heart is one of the major organ targets of GLP-1 and an increasing number of studies have investigated the actions of native GLP-1(7-36), GLP-1 receptor agonists and DPP-4 inhibitors in the context of cardioprotection. The majority of experimental studies have focused on the effects of GLP-1 in cardiac ischaemia and its apparent ability to protect against acute myocardial damage. Indeed, it is well established that GLP-1 pretreatment and chronic DPP-4 inhibition reduce infarct size after experimental ischaemia in both small and large animal models, which is associated with increased survival and improved cardiac function (Bose *et al.*, 2005; Ban *et al.*, 2008; 2010; Timmers *et al.*, 2009). Interestingly, a recent study employing a rabbit model of ischaemia/reperfusion injury reported protective actions of transferrin-stabilized GLP-1, when given both 12 h prior to ischaemia and immediately upon reperfusion, suggesting that GLP-1 may limit infarct size and contractile dysfunction directly, rather than by preconditioning the heart against ischaemia, as suggested by previous reports (Matsubara *et al.*, 2011). In addition to its established beneficial actions against acute ischaemic myocardial damage, GLP-1 also confers protection against contractile dysfunction associated with experimental chronic post-MI remodelling, dilated cardiomyopathy and hypertensive heart failure (Nikolaidis *et al.*, 2004a; Poornima *et al.*, 2008; Liu *et al.*, 2010), with similar results reported in the clinical setting in response to short-term GLP-1 treatment (Nikolaidis *et al.*, 2004b; Sokos *et al.*, 2006).

Until recently, only limited data were available on the cardiac actions of GLP-1 in diabetes, but it is becoming increasingly apparent that GLP-1 also plays a key cardioprotective role in this setting. This is important as it is well known that hyperglycaemia is associated with increased susceptibility to cardiac disease and poor outcomes in both humans and experimental models (Shiomi *et al.*, 2003; Liu *et al.*, 2005; Greer *et al.*, 2006; Vergès *et al.*, 2007). Chronic DPP-4 inhibition with linagliptin improves obesity-related diastolic dysfunction in insulin-resistant Zucker rats, but has no effect on cardiomyocyte hypertrophy and fibrosis (Aroor *et al.*, 2013). Indeed, exendin-4 has been reported to directly protect isolated rat cardiomyocytes from high glucose-induced apoptosis via inhibition of endoplasmic reticulum stress and activation of sarcoplasmic reticulum Ca^{2+} ATPase 2a (Younce *et al.*, 2013). GLP-1 also appears to protect against diabetic cardiomyopathy, which is defined as cardiac dysfunction in the absence of, or disproportionate to, associated

hypertension and CAD and is characterized by marked collagen accumulation and impaired diastolic function (Bugger and Abel, 2014). Both GLP-1 receptor activation and DPP-4 inhibition attenuate development of cardiac dysfunction, extracellular matrix remodelling, cardiomyocyte hypertrophy and apoptosis in experimental models of T1DM and T2DM, with various mechanisms proposed including reduction of lipid accumulation, oxidative stress and myocardial inflammation, and modulation of the MMP-2/tissue inhibitor of MMP-2 axis, endoplasmic reticulum stress and microvascular barrier function (Shigeta *et al.*, 2012; Liu *et al.*, 2013; Monji *et al.*, 2013; Picatoste *et al.*, 2013; Wang *et al.*, 2013). Furthermore, it appears that GLP-1 also confers infarct-reducing actions in diabetes, which is associated with increased susceptibility to myocardial ischaemia. For example, mice made diabetic by a combination of STZ

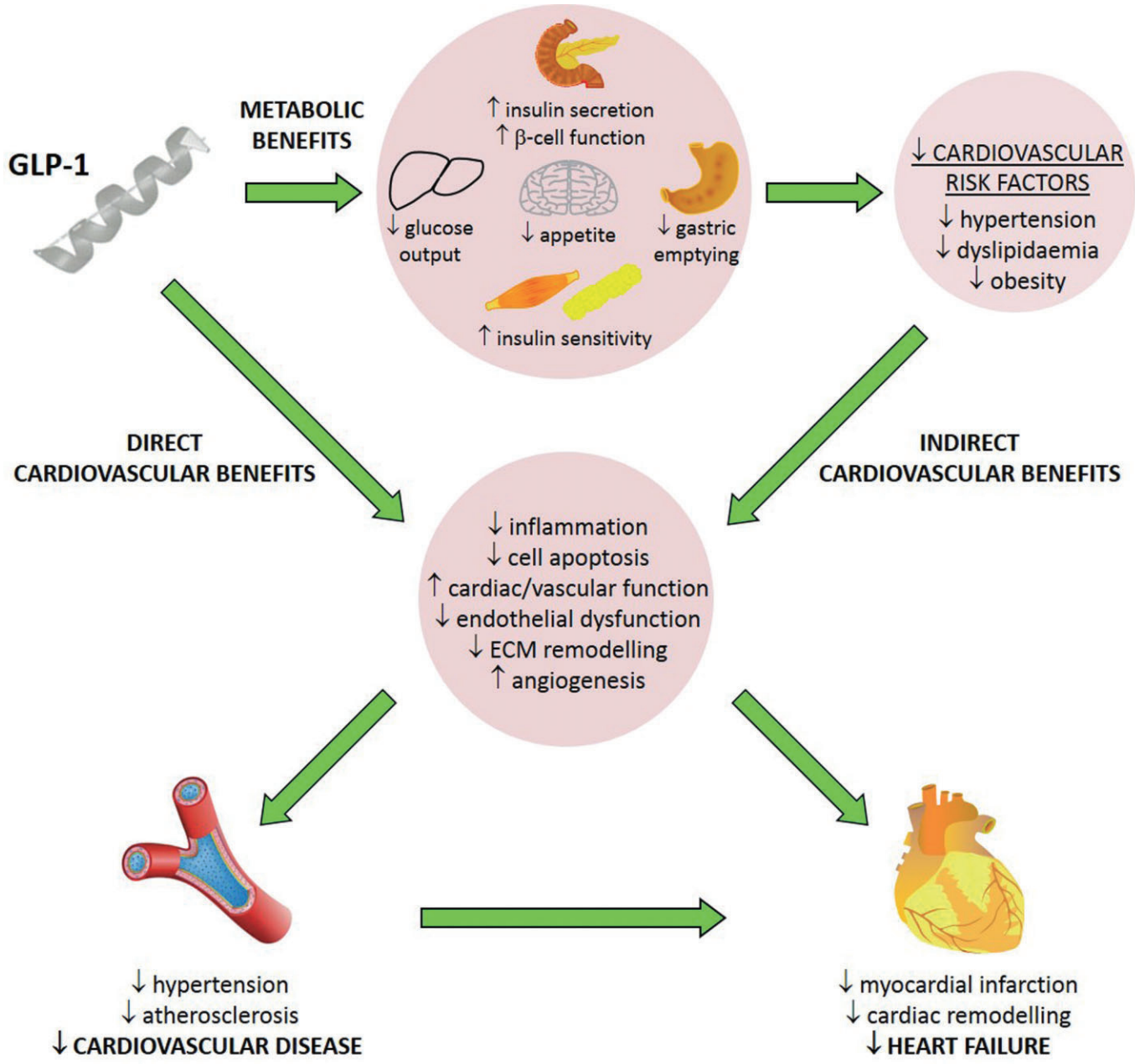

Figure 1

Summary of the cardiovascular actions of GLP-1 in diabetes. GLP-1 exerts indirect cardiovascular benefits in diabetes secondary to its established metabolic actions and subsequent reduction of cardiovascular risk factors. In addition, GLP-1 promotes direct cardiovascular benefits which confer protection against CVD and heart failure, the latter of which may occur via direct myocardial actions or secondary to reduced hypertension and coronary atherosclerosis.

injection and high-fat feeding and treated with the GLP-1 receptor agonist, liraglutide, prior to coronary artery ligation, demonstrated reduced infarct development and improved survival compared with those treated with the glucose-lowering drug, metformin, suggesting that the observed effects occurred via direct actions on the heart and not secondary to reduced blood glucose (Noyan-Ashraf *et al.*, 2009). Similar cardioprotective effects have been reported with DPP-4 inhibition in experimental diet-induced obesity (Huisamen *et al.*, 2013), while the infarct-limiting effects of exendin-4 in mice with T2DM were shown to be mediated by cAMP-induced PKA activation (Ye *et al.*, 2013). Interestingly, it has recently been suggested that the infarct-reducing actions of DPP-4 inhibitors may be glucose-dependent, as both sitagliptin and vildagliptin were found to only decrease infarct size in isolated rat hearts subjected to ischaemia-reperfusion injury when they were perfused with elevated glucose concentrations ≥7 mmol L^{-1}, with similar results observed *in vivo* in diabetic, but not normoglycaemic rats (Hausenloy *et al.*, 2013). This raises the intriguing possibility that glucose-lowering may counteract the cardioprotective actions of GLP-1 and explain why several large-scale clinical trials focused on intensive glucose control in T2DM have failed to demonstrate significant cardiovascular benefits (Giorgino *et al.*, 2013). Furthermore, it appears that at least part of the observed beneficial actions of DPP-4 inhibitors against ischaemia-reperfusion injury may be mediated by the chemokine, stromal cell-derived factor 1α in a GLP-1-independent manner (Bromage *et al.*, 2014).

In addition to the experimental data highlighting a protective role for GLP-1 in the diabetic heart, importantly, a small number of studies have assessed its cardiac actions in patients with diabetes. It has been known for some time that short-term GLP-1 treatment exerts beneficial effects in clinical heart failure in both normoglycaemic and diabetic patients. For example, in a small number of heart failure patients (New York Heart Association class III/IV), 5 week infusion with GLP-1 plus standard therapy improved left

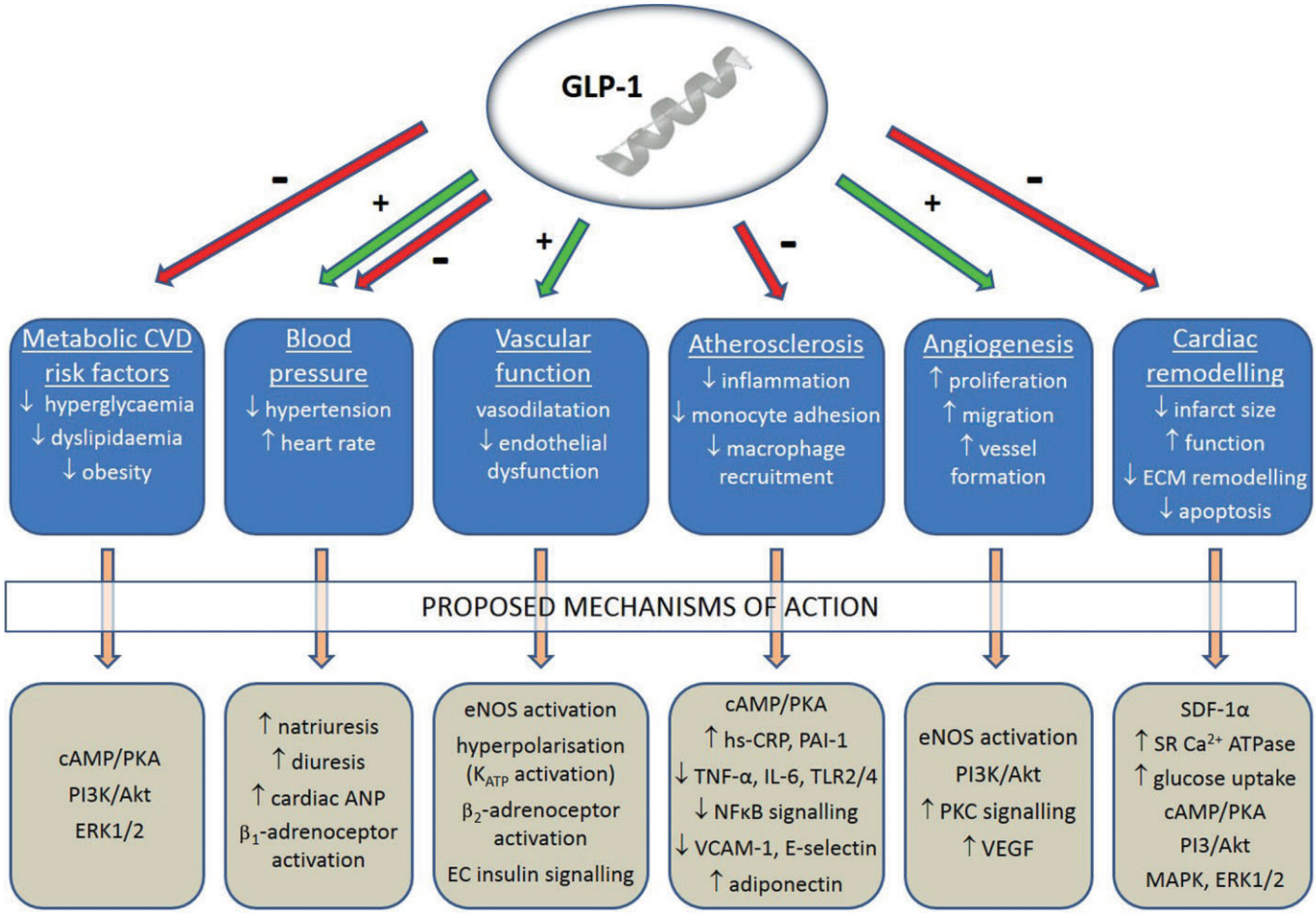

Figure 2

Proposed mechanisms underlying the reported cardiovascular actions of GLP-1. Although it is well established that GLP-1 exerts several beneficial effects on the CVS relevant to diabetes, such as reduction of metabolic CVD risk factors, BP modulation, improved vascular function, decreased atherosclerosis, promotion of angiogenesis and attenuation of adverse cardiac remodelling, the precise mechanisms are yet to be established, although several pathways have been proposed which are the focus of further investigation. EC, endothelial cell; ECM, extracellular matrix; SDF, stromal cell-derived factor 1α; SR, sarcoplasmic reticulum.

ventricular ejection fraction and myocardial oxygen consumption compared with those receiving standard therapy alone, effects that were seen in both diabetic and non-diabetic patients (Sokos *et al.*, 2006). Furthermore, a small non-randomized trial of 72 h GLP-1 infusion following primary angioplasty after acute MI led to improved cardiac function in both non-diabetic and diabetic patients which was still evident upon 120 day follow-up (Nikolaidis *et al.*, 2004b). More recently, a larger randomized trial in patients presenting with ST-segment elevation MI reported that exenatide infusion for 15 min prior to primary angioplasty continued until 6 h post-reperfusion resulted in improved myocardial salvage at 3 months although no functional benefits were observed (Lønborg *et al.*, 2012). Indeed, two current clinical trials are assessing the potential of using exenatide as a post-conditioning agent to reduce reperfusion injury following percutaneous coronary intervention (Effect of Additional Treatment With EXenatide in Patients With an Acute Myocardial Infarction, the EXAMI trial, NCT01254123; Pharmacological Postconditioning to Reduce Infarct Size Following Primary PCI, POSTCON II, NCT00835848). Interestingly, in patients with left ventricular diastolic dysfunction, DPP-4 activity in the coronary sinus and peripheral circulation is reported to be negatively correlated with diastolic function and increased by co-morbid diabetes (Shigeta *et al.*, 2012), suggesting that reduced GLP-1 levels in diabetes may underlie the associated cardiac dysfunction. Exenatide has also been found to modulate myocardial glucose transport and uptake in T2DM patients dependent upon the degree of insulin resistance (Gejl *et al.*, 2012), although a similar study reported that GLP-1-induced increases in resting myocardial glucose uptake in lean individuals were absent in obese T2DM patients, with parallel studies in pigs suggesting that this was due to impaired p38-MAPK signalling (Moberly *et al.*, 2013). Interestingly, a recent experimental study found that exendin-4 reduced contractile function and was unable to stimulate glucose utilization in normal rat hearts in the presence of fatty acids (Nguyen *et al.*, 2013), despite previous reports of increased myocardial glucose uptake in response to GLP-1 in experimental myocardial ischaemia and dilated cardiomyopathy (Nikolaidis *et al.*, 2005; Zhao *et al.*, 2006; Bhashyam *et al.*, 2010). Such findings highlight the need for detailed investigation of the effects of GLP-1 on altered myocardial metabolism in diabetic patients both with and without cardiac complications, in which the effects of GLP-1 may diverge.

Although these clinical and experimental data are clearly supportive of an important role for GLP-1 signalling in the diabetic heart, they are largely descriptive with limited focus on underlying mechanisms. Previous studies in experimental models of heart failure have highlighted several pathways which may mediate the cardioprotective effects of GLP-1, including cAMP/PKA, PI3K/Akt, p44/p42MAPK, ERK1/2 (Bose *et al.*, 2005; Timmers *et al.*, 2009; Ravassa *et al.*, 2011), together with suggestions of GLP-1 receptor-independent signalling (Nikolaidis *et al.*, 2005; Sonne *et al.*, 2008). However, the precise mechanisms underlying the apparent protective actions of GLP-1 in the diabetic heart, in which GLP-1 signalling is likely to be different, remain unknown and clearly need to be defined in order to fully assess the therapeutic potential of GLP-1 in this setting.

Table 1

Current long-term clinical trials of GLP-1-based therapies on cardiovascular outcomes

Study name	Drug	Administration	Estimated patient enrollment	Primary outcome(s)	Expected end date	NCT identifier
EXSCEL	Exenatide	s.c. injection, 2 mg, once weekly	14 000	Rate of CV death, non-fatal MI or non-fatal stroke	December 2017	NCT01144338
LEADER	Liraglutide	s.c. injection, 1.8 mg, once daily	9 340	Time to CV death, non-fatal MI or non-fatal stroke	October 2015	NCT01179048
ELIXA	Lixisenatide	s.c. injection, 20 μg, once daily	6 000	Time to first primary CV event	January 2015	NCT01147250
REWIND	Dulaglutide	s.c. injection, 1.5 mg, once weekly	9622	Time to CV death, non-fatal MI or non-fatal stroke	August 2019	NCT01394952
TECOS	Sitagliptin	p.o., 50 or 100 mg, once daily	14 000	Time to first CV event	December 2014	NCT00790205
CAROLINA	Linagliptin	Not specified	8 300	Time to CV death, non-fatal MI, non-fatal stroke and hospitalization for unstable angina pectoris	January 2018	NCT01897532
CARMELINA	Linagliptin	p.o., 5 mg, once daily	6 000	Time to CV death, non-fatal MI, non-fatal stroke and hospitalization for unstable angina pectoris	September 2018	NCT01243424

CV, cardiovascular; MI, myocardial infarction; NCT, National Clinical Trial.

Summary and future perspective

Over recent years, it has become clear that GLP-1 exerts important actions on the CVS in both health and disease in addition to its prototypic effects on glycaemic control (Grieve *et al.*, 2009) and also confers beneficial effects on the cardiovascular risk profile in diabetic patients. However, emerging evidence now strongly suggests that GLP-1 exerts specific cardiovascular actions in diabetes and may attenuate the development and progression of associated cardiovascular complications (summarized in Figure 1), although the precise mechanisms are yet to be established with several candidate pathways proposed (see Figure 2). Nonetheless, it is important to note that the majority of data pointing towards such beneficial effects are largely experimental and that equivalent information on the cardiovascular actions of GLP-1 in the clinical setting of diabetes is somewhat lacking, particularly in relation to its chronic effects. In this regard, the first large-scale GLP-1 clinical trials to assess cardiovascular outcomes after long-term treatment (SAVOR-TIMI 53, EXAMINE) have recently reported that chronic DPP-4 inhibition with either saxagliptin or apogliptin had no significant effects on their primary composite end points of cardiovascular death, or non-fatal MI/stroke (Scirica *et al.*, 2013; White *et al.*, 2013), although it should be noted that SAVOR-TIMI 53 reported a 27% increase in hospitalization for heart failure. However, until the results of several ongoing clinical trials investigating the chronic cardiovascular effects of GLP-1 receptor agonists are known (summarized in Table 1 together with further DPP-4 inhibitor trials), in which circulating GLP-1 levels will be much higher than those achieved using DPP-4 inhibitors, no conclusions can be drawn on either the potential clinical cardiovascular benefits or safety of GLP-1-based therapies in diabetes. Nonetheless, the apparent complexity of cardiovascular GLP-1 signalling under both normal and diabetic conditions clearly suggests that it is likely that selective targeting of specific aspects of CVD may be required in order to realize the optimal benefits of GLP-1 targeting in this setting.

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Conflict of interest

None.

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