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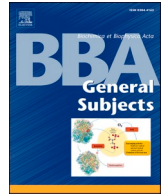
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Role of platelet-derived growth factor c on endothelial dysfunction in cardiovascular diseases

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ABSTRACT

Loss of endothelial function is a common feature to all cardiovascular diseases (CVDs). One of the risk factors associated with the development of CVDs is the hyperglycaemia that occurs in patients with metabolic disorders such as Type 1 and Type 2 diabetes mellitus. Hyperglycaemia causes endothelial dysfunction through increased production of reactive oxygen species (ROS) from different cellular sources leading to oxidative stress. Vascular endothelial growth factor (VEGF) is essential in the stimulation and maintenance of endothelial functional aspects and, although it can mitigate the impact of ROS, VEGF-mediated signalling is partially inhibited in diabetes mellitus. The search for therapeutic strategies that preserve, protect and improve the functions of the endothelium is of great relevance in the investigation of CVDs associated with hyperglycaemia. Platelet-derived growth factor C (PDGF-C) is a peptide with angiogenic properties, independent of VEGF, that stimulates angiogenesis and revascularization of ischemic tissue. In a diabetic mouse model, PDGF-C stimulates mature endothelial cell migration, angiogenesis, endothelial progenitor cell mobilization, and increased neovascularization, and protects blood vessels in a retinal degeneration model activating anti-apoptosis and proliferation signalling pathways in endothelial cells. This review summarizes the information on the damage that high D-glucose causes on endothelial function and the beneficial effects that PDGF-CC could exert in this condition.

Abbreviations: Akt, Protein kinase B; BH4, Tetrahydrobiopterin; Bis I, Bisindolylmaleimide I; cDNA, Complementary desoxyribonucleic acid; CUB, Complement subcomponents C1r/C1s, urchin epidermal growth factor (EGF)-like protein and bone morphogenic protein 1; CVDs, Cardiovascular diseases; eNOS, Endothelial nitric oxide synthase; ERK, Extracellular regulated protein kinases; ET-1, Endothelin 1; ETC, Electron transport chain; FGF, Fibroblast growth factor; GSH, Glutathione; H₂O₂, Hydrogen peroxide; HAECs, Human aortic endothelial cells; HDL, High density lipoprotein; HMOX-1, Heme oxygenase 1; HMVECs, Human microvascular endothelial cells; HUVECs, Human umbilical vein endothelial cells; IL- β , Interleukin β ; JAK, Janus kinase; LDL, Low density lipoprotein; MAPK, Mitogen-activated protein kinases; Mn-SOD, Manganese-dependent superoxide dismutase; NADPH, Nicotinamide adenine dinucleotide phosphate; NO, Nitric oxide; NOX, Nicotinamide adenine dinucleotide phosphate oxidase; O₂, Oxygen; O₂⁻, Superoxide radical; ONOO⁻, Peroxynitrite; PAECs, Porcine aortic endothelial cells; PDGF, Platelet derived growth factor; PECAM-1, Platelet endothelial cell adhesion molecule 1; PI3K, Phosphatidylinositol 3-kinase; PKC, Protein kinase C; PLC, Phospholipase C; Prx, Peroxiredoxin; RNS, Reactive nitrogen species; ROS, Reactive oxygen species; SCID, Severe combined immunodeficiency; sFlt, Soluble vascular endothelial growth factor receptor 1; SH2, Src homology 2; shRNA, Short hairpin ribonucleic acid; SOD, Superoxide dismutase; STAT, Signal transducer and activator of translation; STZ, Streptozotocin; T1DM, Type I diabetes mellitus; T2DM, Type II diabetes mellitus; TGF β , Transforming growth factor β ; tPA, Tissue plasminogen activator; Trx, Thioredoxin; uPA, Urokinase plasminogen activator; VE, Vascular endothelium; VEGF, Vascular endothelial growth factor; VEGFR, Vascular endothelial growth factor receptor; VSMCs, Vascular smooth muscle cells; α -SMA, α -Smooth muscle actin.

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1. Introduction

Cardiovascular diseases (CVDs) cause microvascular and macrovascular complications [1–6]. Risk factors for CVDs include age, gender, high plasma level of total cholesterol and low-density lipoprotein (LDL), low plasma level of high-density lipoprotein (HDL), high diastolic or systolic blood pressure, smoking, and metabolic diseases such as Type 1 (T1DM) and Type 2 (T2DM) diabetes mellitus [7–9]. Patients with diabetes mellitus have 2 to 3-fold higher risk of developing CVDs than healthy people [8,9]. CVDs show with vascular complications [10–13], particularly regarding abnormal regulation of the vascular tone due to imbalanced synthesis of endothelium-derived vasodilators and vasoconstrictors [14–18]. The latter is referred to as endothelial dysfunction [16,19,20], which contributes to the pathogenesis of CVDs associated with diabetes mellitus [21].

Hyperglycaemia targets the vascular endothelium [22] and the function of molecules regulating the endothelial function [23–25]. One of these molecules is the vascular endothelial growth factor (VEGF) [24,26,27] whose capacity to activate VEGF receptors (VEGFR) is reduced in monocytes from T2DM patients [24,28] and in experimental animal models of diabetes [29–31]. Another molecule is the platelet-derived growth factor C (PDGF-C) which shows VEGF-independent angiogenic properties with a protective effect on the vasculature [32,33]. Nevertheless, the mechanisms of PDGF-C vascular actions are not fully understood. This review addresses the potential beneficial effects of PDGF-C in the cardiovascular damage caused by a condition of hyperglycaemia as seen in patients with diabetes mellitus.

2. Endothelial dysfunction in diabetes mellitus

Diabetes mellitus is a complex and multifactorial metabolic disease that associates with hyperglycaemia [34–36]. The American Diabetes Association (ADA) guideline recommend diabetes mellitus diagnosis when fasting glucose level is ≥ 126 mg/dL (7 mmol/L) or when glycaemia in an oral glucose tolerance test with 75 g glucose is ≥ 200 mg/dL (11.1 mmol/L) or aleatory glucose values are ≥ 200 mg/dL [37]. According to its aetiology, T1DM refers to an early-onset autoimmune form, and T2DM corresponds to a late-onset non-autoimmune form [34,38]. Patients with diabetes mellitus show endothelial dysfunction attributed to insulin resistance and hyperglycaemia [11,12]. In the endothelium, insulin activates the phosphatidylinositol 3-kinase (PI3K)/protein kinase B (Akt) signalling pathway involved in regulating the generation of NO because of the phosphorylation at Ser¹¹⁷⁷ in the endothelial NO synthase (eNOS) [39]. Furthermore, insulin stimulates the release of the potent vasoconstrictor endothelin 1 (ET-1), which requires activation of the mitogen-activated protein kinases (MAPK) signalling pathway [40,41]. Lower activation of the PI3K/Akt pathway and higher MAPK activation results in reduced vasodilation [40,42,43] addressing the association between cardiovascular and metabolic diseases seen in patients with diabetes mellitus.

Hyperglycaemia induces oxidative stress which is defined as a misbalance between the production of oxidant molecules, such as reactive oxygen species (ROS), and antioxidant cell defence systems including scavenger enzymes such as superoxide dismutases (SOD), catalases, peroxidases, peroxiredoxins (Prx), thioredoxin (Trx) [44,45]. Increased ROS generation by the endothelium, particularly the superoxide anion ($O_2^{\cdot -}$) [12,16,19,46], reduces the bioavailability of NO, therefore limiting its biological activity. The $O_2^{\cdot -}$ reacts with NO to form peroxynitrite ($ONOO^-$), a highly unstable and reactive nitrogen specie (RNS) that damages nucleic acids, lipids, and proteins, leading to a predominant state of vasoconstriction [16]. Increased ROS generation results from upregulation of nicotinamide adenine dinucleotide phosphate (NADPH) oxidases (NOX), eNOS uncoupling, and higher activity of the mitochondrial electron transport chain (ETC) [16,18,19]. NOX are protein complexes formed by cytosolic factors and a redox transmembrane core [47]. The NOX4 isoform is the most abundant in the

endothelium [48] and its unique function is to catalyse the reduction of oxygen to hydrogen peroxide (H_2O_2) and $O_2^{\cdot -}$ by transferring electrons donated by NADPH [45,47,48]. eNOS catalyses the formation of NO from the cationic amino acid L-arginine requiring tetrahydrobiopterin (BH_4) and NADPH [16]. In diabetes mellitus, eNOS reduces O_2 to $O_2^{\cdot -}$ due to eNOS uncoupling caused by BH_4 reduced bioavailability [49].

Diabetic vascular complications may also result from lower generation or activity of growth factors, including PDGF, VEGF and fibroblast growth factor (FGF) [50]. Growth factors increase angiogenesis [51–53], although patients with diabetes mellitus showed lower angiogenic response to growth factors leading to deficient wound healing and low peripheral and coronary circulation [29,31,52]. PDGF is expressed in vasculature pericytes, smooth muscle cells (VSMCs), and endothelial cells [54,55] and seems to accelerate the growth of VSMCs in aortas of patients with T2DM [56]. However, low levels of PDGF relate with cardiovascular complications leading to deficient angiogenesis and impaired wound healing in patients with T2DM [56–58]. VEGF is essential to maintain a functional endothelium [24,26,27]. VEGF may mitigate the impact of ROS by inducing the expression of the mitochondrial-derived antioxidant manganese SOD (Mn-SOD) [59]. Nevertheless, hyperglycaemia-induced ROS formation results in activation of the non-receptor tyrosine kinases Src. The latter results in phosphorylation of the intracellular VEGF receptor 2 (VEGFR2) reducing its availability at the plasma membrane, therefore limiting the signalling mechanisms induced by exogenous VEGF [29].

The angiogenic potential of VEGF and FGF has been assayed in animal models of coronary and peripheral ischemia [26,53,60,61]. The results show increased blood flow and vascular function in response to these growth factors. However, the action of VEGF and FGF gene transfer in subjects with peripheral arterial disease and lower extremity ischemia associated with diabetes mellitus showed no positive biological effects. An explanation for these results is that angiogenesis is induced by multiple factors [8,30,62,63]. Thus, searching for other growth factors hopefully overcoming these deficiencies becomes a therapeutic strategy worth exploring.

3. PDGF

3.1. PDGF forms

The PDGF/VEGF family belongs to the cystine-knot growth factors superfamily, which characterizes by formation of intra and interchain disulfide bonds [64]. There are four structurally related inactive PDGF monomer chains. The active forms of PDGF are synthesised in platelets, VSMCs, activated macrophages, and endothelial cells, forming homo or heterodimers of two of the A, B, C, or D chains [64–66]. Four PDGF homodimers (PDGF-AA, PDGF-BB, PDGF-CC, PDGF-DD) and one heterodimer (PDGF-AB) have been described [55,67].

PDGF-A and PDGF-B contain short amino acid extensions located at the C-terminus and N-terminus, are activated by proteolysis, and involved in cell matrix adhesion. PDGF-C and PDGF-D do not have amino acid extensions in C-terminus but have a complement C1r/C1s, Uegf, Bmp1 (CUB) domain in the N-terminus which binds to the extracellular matrix to prevent their diffusion [68]. Proteolytic cleavage of the CUB domain sets free the growth factor domain for binding and activation of PDGF receptors [69]. Active PDGF dimers act through three receptors made up of five immunoglobulin-like extracellular domains, one transmembrane domain, and one intracellular domain with tyrosine kinase activity [70,71]. The three biologically active receptors are formed for homo or heterodimerization of α and β subunits [72,73].

Binding of PDGF ligands to one monomeric subunit receptor leads to the recruitment of a second monomeric subunit receptor, their dimerization, and autophosphorylation on intracellular domain tyrosine residues [74]. The five PDGF ligands show different specificity to receptors binding, *viz*, PDGF-AA, PDGF-BB, PDGF-AB, and PDGF-CC activate PDGF receptor α (PDGFR α), PDGF-BB and PDGF-DD activate PDGFR β ,

and PDGF-AB, PDGF-BB and PDGF-CC activate PDGFR $\alpha\beta$ [55,70,74]. These interactions create coupling sites for proteins containing Src homology 2 (SH2) domains involved in signal transduction mainly through PI3K/Akt, MAPK, and phospholipase C (PLC γ). Activation of these receptors associate with cell survival, growth, differentiation, and proliferation [54,74], and induce mitogenic signals in fibroblasts, VSMCs, glial cells, stem/progenitor cells, neuronal cells, vascular endothelial and inflammatory cells [69] (Fig. 1).

Dysregulation of expression of all PDGF ligands is involve in CVDs. PDGF-A, PDGF-B, PDGF-C and PDGF-D are overexpressed in vascular atherogenic wall and mainly released by endothelial cells, macrophages and VSMCs [68,75]. PDGF-A and PDGF-D are highly expressed in infarcted myocardium while PDGF-B and PDGF-C are diminished in non-infarcted area [75–77]. The phenotypic change of fibroblasts to myofibroblasts is leading by PDGF-A and PDGF-B resulting in fibrosis. PDGFR β is the principal receptor involved in vascular pathology acting on fibroblasts and VSMCs, whereas signalling mediated by PDGFR α regulates fibrotic process in mesenchymal cells and in fibroblasts [68].

PDGFR β is overexpressed in VSMCs and pericytes in atherosclerotic injuries and cardiac fibrosis [74] and leads to leukocyte migration to the intima [68], whereas PDGFR α is highly expressed in endothelial cells and macrophages only under proinflammatory cytokines like interleukin 1 β (IL-1 β) and transforming growth factor β (TGF- β) stimulation [74]. In myocardial infarction, PDGFR $\alpha\alpha$ and PDGFR $\beta\beta$ are highly expressed [76,77]. PDGFR β leads the development of VSMCs synthetic phenotype in restenosis [68]. PDGFR α and PDGFR β activation is involved in the disruption of blood brain barrier increasing the development of stroke [64]. These findings suggest that expression of PDGF ligands and receptors is linked to cardiovascular pathologies, either due to excess or defect. Although most of PDGF-target therapies associated to cardiovascular complications in diabetes mellitus are addressed to the inhibition of its effects [64], PDGF-C-based studies particularly seem to be conducted to stimulate its angiogenic, proliferative, neuroprotective, and anti-apoptotic properties [64,78].

3.2. PDGF-C

PDGF-C is a peptide of 32 amino acids with a predicted molecular weight of 36.7 kDa [64,66]. The gene encoding this protein is found in the q31-q32 region of the long arm of chromosome 4 [66]. In addition to

the C-terminal domain, the N-terminal CUB domain of PDGF-C is proteolytically removed by tissue plasminogen activator (tPA), plasmin, matriptase, and urokinase plasminogen activator (UPA) [69,79]. Binding of PDGF-C to PDGFR $\alpha\alpha$ and PDGFR $\alpha\beta$ but not PDGFR $\beta\beta$ is of high-affinity [80]. Interaction of PDGF-C with PDGFR α and PDGFR β initiates PLC γ , Janus kinase (JAK)/ signal transducer and activator of translation (STAT), PI3K/Akt and Ras-MAPK signalling pathways involved in proliferation, angiogenesis, inhibition of apoptosis, and differentiation, respectively [64,65,79]. Additionally, active PDGFR β initiates rapidly accelerated fibrosarcoma (Raf)/MAPK1/2 pathway leading to fibrosis [65]. Remarkably, PDGF-C induces angiogenesis in ischemic tissues [58] even in diabetes state [30], independent of VEGF, although the mechanisms of its actions are undescribed [30,58].

PDGF-C is highly expressed in the heart, liver, kidney, pancreas, ovary, VSMCs, and macrophages but expression is lower in the brain, placenta, lung, skeletal muscle, thymus, prostate, testis, and small intestine [55,69,79]. PDGF-C is also expressed in different types of endothelial cells including the human umbilical vein endothelial cells (HUVECs), human aortic endothelial cells (HAECs) [69] and human glomerular microvascular endothelial cells (HMVECs) [81]. However, PDGFR expression level in these cell types is lower compared with VSMCs or pericytes [55]. Consequently, only few studies addressing the effect of PDGFs in endothelial cells are available.

Some publications propose PDGF-C as a new growth factor with independent VEGF properties that could be of clinical use. As mentioned, most of the positive effects mediated by PDGF-C at the level of vasculature are related to angiogenesis [58,82,83]. PDGF-C stimulated the growth of microvessels, increased neovascularization with high-density vessels, and increased the formation of new branches and vessel sprouts in *in vivo* assays for embryonic development. In the same study, vascular corneal tissue was stimulated by PDGF-C creating a large, robust number of capillaries and recruiting wall cells. To identify whether the effects of PDGF-C were mediated by PDGFR α and PDGFR β , phosphorylation of these receptors in porcine aortic endothelial cells (PAECs) over-expressing these receptors was analysed [82]. The results show that PDGF-C activated both receptors highlighting the critical role played by PDGF-C in blood vessel growth and maturation and its capacity to induce angiogenesis.

PDGF-C increased the expression of PDGFR α and stimulated the formation of functional vessels as well as VSMCs covering in a model of

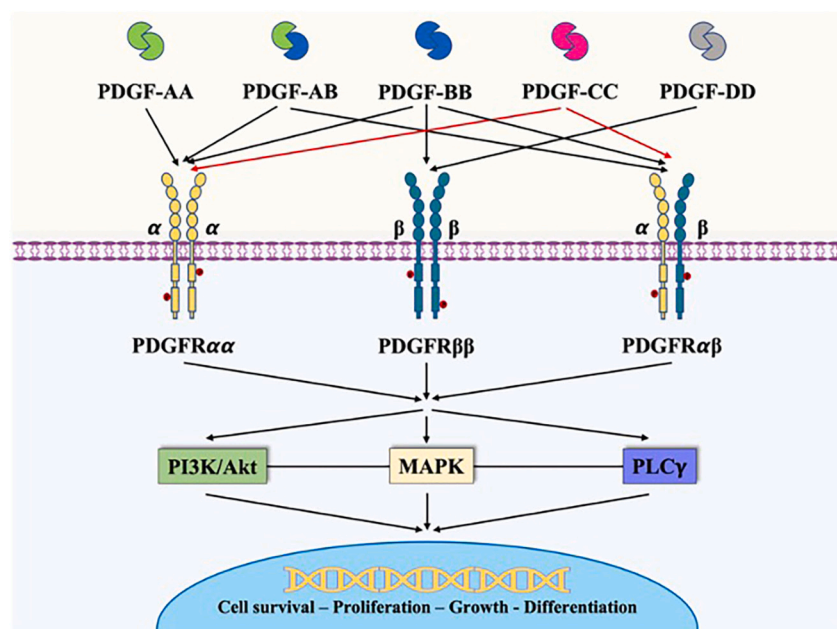


Fig. 1. Signalling pathways and biological outcomes mediated by interactions between PDGF ligands and receptors. Four homodimers (PDGF-AA, BB, CC, and DD) and one heterodimer (PDGF-AB) of platelet-derived growth factors are expressed in humans. These isoforms differentially activate at least three PDGF receptors, viz, PDGFR $\alpha\alpha$, PDGFR $\beta\beta$, and PDGFR $\alpha\beta$, leading to activation of phosphatidylinositol 3 kinase/protein kinase B (PI3K/Akt), mitogen-activated protein kinases (MAPK), and phospholipase C γ (PLC γ). Activation of PDGF receptors results in increased cell survival, proliferation, growth, and differentiation.

coronary artery ligation. Thus, PDGF-C could be key in ischemic myocardial revascularization [58]. Using a model of hind limb ischemia, researchers found that PDGF-C induced the formation of new and mature vessels and increased the mobilization of endothelial progenitor cells. PDGF-C also induced differentiation of bone marrow progenitors to endothelium, confirmed by positive expression of vascular endothelial (VE)-cadherin/platelet endothelial cell adhesion molecule 1 (PECAM-1). Also, PDGF-C enhanced the migration of HMVECs, HUVECs, bovine aortic endothelial cells (BAECs), and PAECs evaluated *in vitro* by the scrape wounding assay in isolated cultures, suggesting that the angiogenic effect of this growth factor may result from a direct action on the endothelial cells. Nevertheless, PDGF-C effects in *in vivo* studies may be mediated by an indirect upregulation of others growth factors such as VEGF, given that endothelial progenitor cells (EPCs), VSMCs, fibroblasts, and tumour cells release VEGF in response to PDGF-C [58]. These results show direct and indirect roles that PDGF-C may play in the treatment of ischemic myocardial and extremities revascularization after ischemia in patients with CVDs. Furthermore, PDGF-C protects blood vessels by acting on vascular endothelium and VSMCs in mouse models of retina degeneration and retinitis pigmentosa gene. The proposed mechanisms involve the activation of PDGFR α and PDGFR β and up-regulated expression of *HMOX1* coding for the potent antioxidant heme oxygenase-1 [83].

The role of PDGF-C on blood vessels permeability was addressed in functional B and T lymphocytes deficient SCID mice. Animals implanted with U87 glioma cells overexpressing PDGF-C (U87-C) showed tumours where the blood vessels were less permeable showing more perivascular cells than those implanted with cells where PDGF-C expression was inhibited by shRNA (U87RNAi) [84]. These results suggest that PDGF-C is involved in the recruitment and differentiation of perivascular cells resulting in basement membrane integrity and a mature and stable vascular wall with lower permeability [84]. Another study performed in a PDGF-C-deficient C57BL/6 mouse model revealed that although the extracranial vessels had high VSMCs coverage compared to wild-type controls, these cells did not show normal organization and were partially separated from the abluminal endothelial cell surface. The latter phenomenon resulted in subcutaneous oedema formation and haemorrhage showing PDGF-C-lacking abnormal permeability [85].

Vascular complications of diabetes mellitus are explained in part by VEGF resistance and reduced angiogenesis in ischemic extremities and heart [24,29,31]. Some studies focused in finding different growth factors with VEGF-like activity [30,32,63,84]. It is reported that the expression of *Pdgfc* (for PDGF-C) was downregulated, but expression of the *Pdgfra* (for PDGFR α) was unaltered in ischemic limbs of STZ-induced diabetes mice compared to non-diabetic animals. These findings suggest that STZ-induced diabetes may result in lower effectiveness of PDGF-C due to lower bioavailability of this growth factor rather than a reduced biological action because a lower number of receptors [30].

After hind limb surgery-associated ischemia *Pdgfc* and *Pdgfra* genes expression was downregulated, and recovery of blood flow was impaired in diabetic compared to non-diabetic mice. Insertion of a vector encoding PDGF-C resulted in efficient revascularization and flow recovery. Thus, a decreased expression of *Pdgfc* and subsequent lower level of PDGF-C may be involved in the deficient angiogenesis observed in diabetic tissues. Since the introduction of PDGF-C was sufficient to restore blood flow in ischemic tissues of diabetic mice and VEGF expression was not altered in this condition, PDGF-C might be a therapeutic option independent of VEGF for ischemic CVDs in diabetes mellitus [30].

Assays using HUVECs and HMVECs exposed *in vitro* to high D-glucose conditions (30 mmol/L) were done trying to elucidate the mechanism(s) associated with the actions of PDGF-C. Exposure to high D-glucose (30 mmol/L for 5 days) reduced the cell proliferation and viability compared to cells incubated in normal D-glucose (5.5 mmol/L). Interestingly, even when these cell types show substantial phenotypic differences, v.g. macro versus microvascular and foetal versus adult endothelium, the

high extracellular D-glucose effect was similar [86]. These findings contrast with those described in other study where angiogenesis increased in the microvasculature in response to hyperglycaemia and a diabetic environment [87]. The involved mechanism included increased release of VEGF and reduced level of the soluble VEGF receptor 1 (sFlt) [88]. However, the potential role of PDGF-C and PDGFR α in this phenomenon was not addressed.

HUVECs and HMVECs exposed to high D-glucose show reduced expression of *Pdgfra* and its transcript and there was no change in *Pdgfc* regarding cells exposed to normal D-glucose concentrations. It was proposed that the effect of D-glucose associated with upregulation of protein kinase C α (PKC α). Furthermore, inhibition of PKC α with bisindolylmaleimide I (Bis I) resulted in higher PDGF-C-increased activation and phosphorylation of Akt and 44 and 42 kDa MAPK (p44/42^{mapk}), signalling pathways that are essential to maintain the endothelial cell function [86]. These findings agree with results showing that primary cultures of HUVECs from normal pregnancies exposed to high D-glucose (25 mmol/L, 30 min) show reduced cell proliferation involving activation of PKA, PKC and PKG [89]. Interestingly, incubation of HUVECs and HMVECs with the PKC α inhibitor Bis I resulted in a potentiation of the PDGF-C-induced tube formation *in vitro*. Thus, PKC α along with an involvement of PKC and PKG is crucial for PDGF-C stimulated angiogenesis *in vitro* [86].

4. Concluding remarks

PDGF-C may be a therapeutic target in CVDs and its association with metabolic diseases including diabetes mellitus (Fig. 2). Strong evidence supports the VEGF-independent angiogenic properties of PDGF-C in ischemic tissues, which constitutes an alternative therapy under conditions of diabetes mellitus or high extracellular D-glucose concentrations, where resistance to VEGF is persistent. However, there is a lack of research to clarify the role and action mechanisms of PDGF-C in the involvement of endothelial cells in regulating the inflammatory response, the control of vascular tone, and coagulation. Since PDGF-C upregulates several genes involved in modulating oxidative stress and inflammation, such as *HMOX1*, it is crucial to evaluate the effect of this growth factor on oxidative stress generated by an environment containing high D-glucose levels such as in diabetes mellitus with and uncontrolled and sustained hyperglycaemia condition. Characterizing the mechanisms involved in the modulatory effect of PDGF-C on the glycaemia control and therefore its angiogenic properties in patients with diabetes mellitus is still under development. The characterization of these mechanisms might help to improve or identify therapeutic targets for treating patients with CVDs.

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CRedit authorship contribution statement

Adriana Grimaldo: Conceptualization, Methodology,

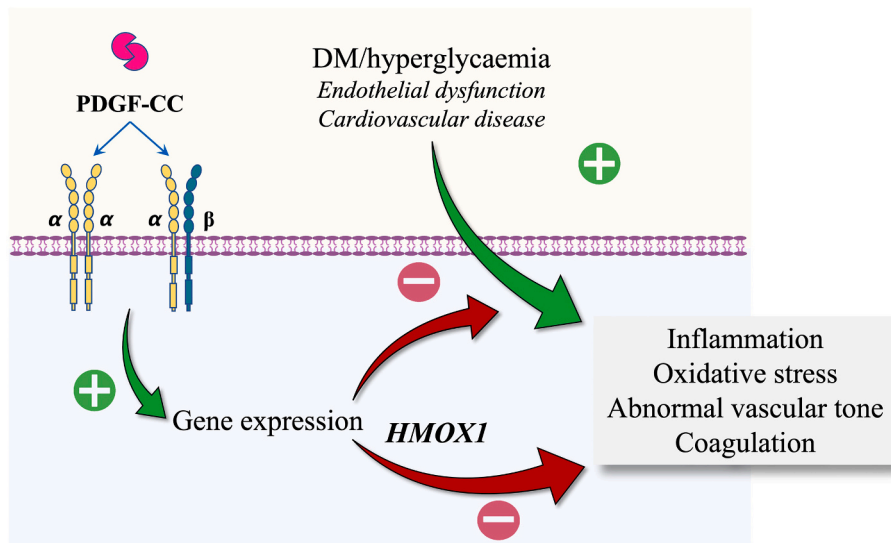


Fig. 2. Possible therapeutic actions of PDGF-C on the development of cardiovascular diseases caused by hyperglycaemia and/or diabetes mellitus (DM). Platelet-derived growth factor isoform C (PDGF-C) activates the PDGF homodimer receptor α (α,α) and heterodimer receptor $\alpha\beta$ (α,β). PDGF-C increases (+) the expression of a variety of genes, including heme oxygenase 1 (HMOX-1) which reduces (–) the inflammation, oxidative stress, coagulation, and vasoconstriction observed in endothelial dysfunction associated with diabetes mellitus and hyperglycaemia. Increased expression of other genes may counteract (–) the effect of high glucose on cardiovascular diseases.

Investigation, Writing – original draft, Writing – review & editing, Visualization. **Luis Sobrevia:** Conceptualization, Methodology, Investigation, Writing – original draft, Writing – review & editing, Visualization, Supervision. **Ludis Morales:** Conceptualization, Methodology, Investigation, Writing – original draft, Writing – review & editing, Visualization, Supervision.

Declaration of Competing Interest

None.

Data availability

No data was used for the research described in the article.

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