Pathophysiology of cardiovascular disease in diabetes mellitus

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Diabetes mellitus elicits cellular, epigenetic, and post-translational changes that directly or indirectly affect the biology of the vasculature and other metabolic systems resulting in the apparition of cardiovascular disease. In this review, we provide a current perspective on the most recent discoveries in this field, with particular focus on hyperglycemia- induced pathology in the cardiovascular system. We also provide perspective on the clinical importance of molecular targeting of cardiovascular and diabetes mellitus therapies to treat hyperglycemia, inflammation, thrombosis, dyslipidemia, atherosclerosis, and hypertension. ***Cardiovasc Endocrinol Metab*** **7:4–9**

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Introduction

Diabetes mellitus (DM) often coexists with cardiovascular disease (CVD) in clinical practice, but the pathophysiology of this comorbid condition could be rather confusing as the amount of scientific evidence is dispersed and has increased, especially in the last decade.

The strong link between these two diseases is evident. Patients with CVD share similar risk factors for DM onset such as unhealthy dietary and lifestyle habits, obesity, smoking, etc., or already have DM (*http://www.who.int/mediacentre/factsheets/fs317/en/*). Similarly, DM itself constitutes a risk factor for CVD and its complications, such as myocardial infarction (MI), stroke, amputation, etc. [1].

The mechanisms of the pathogenesis of CVD in diabetes are related to epigenetic, genetic, and cell-signaling defects in inter-related metabolic and inflammatory pathways. These metabolic defects (especially in the endothelium, liver, skeletal muscle, and β cells) can be triggered by various environmental factors such as high caloric intake, smoking, glycation end-products, glucose toxicity, and some medications [2]. It could be stated that the expression of both type 2 diabetes mellitus (T2DM) and CVDs is an idiosyncratic response to the environment, guided by the biological capacity of cellular systems in patients [3].

These idiosyncrasies are expressed differently among patients and populations [3]. Some patients may express clear or mixed phenotypes of hyperglycemia, dyslipidemia, hypertension, inflammation, or thrombosis, which also represent risk factors for CVD [4]. Interestingly, all of these clinical manifestations share similar cellular mechanisms and molecular abnormalities. T2DM has multiple cell-signaling pathways in cell growth, survival, and proliferation such as pAkt, endothelial nitric oxide synthase pathway, and AMP-activated protein kinase pathways that could potentiate the development of CVD. In addition to this, glucose and oxidized lipids exert important effects in tissues at the epigenetic level [5–7]. It is noteworthy that some of these epigenetic adaptations can even be passed down to several generations [8–10].

In this review, we provide a current perspective on the advances of such discoveries and for this purpose; we grouped them as CVD risk factors affecting the biology of patients with pre-existing DM. We also provide a brief overview on how cell-signaling pathology and post-translational changes in hypertension, dyslipidemia, inflammation, and hyperglycemia result in the early appearance of CVD phenotypes and the opportunity for new therapies. In an attempt to explain the major relevant pathophysiological pathways that are present in DM that are related to CVD and their therapeutic implications, we have listing them on the basis of their clinical phenotype.

Hyperglycemia

T2DM is considered a multifactorial disease that involves abnormalities in carbohydrate and lipid metabolism [11]. Its most evident manifestation is chronic hyperglycemia. Recent evidence shows that T2DM patients have defects in epigenetic and post-translational modifications of the vascular architecture [2]. The multiple endometabolic defects of diabetes impact behaviors such as overfeeding (leptin resistance or deficiency) as well as aspects of

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glucose and lipid metabolism such as cellular toxicity and insulin receptor (IR) effects of palmitic acid and other saturated fatty acids [12]. Leptin is a protein secreted by white adipocytes and has the function of stimulating satiety (postprandial) and increase energy expenditure by binding its cognate receptor (leptin receptor B) [13]. Mutations in either leptin protein (biologically inactive) or its receptor (defective activation) result in overfeeding behaviors, leading to profound obesity phenotypes with the association of peripheral insulin resistance and hyperglycemia [14].

Similarly, saturated fatty acids such as palmitic acid have been proposed as insulin desensitizers not just in peripheral tissues but also in the hypothalamus. This dual effect leads to peripheral and central insulin resistance, resulting in hyperglycemia and dysregulation of energy balance in the whole organism [14,15]. The chronicity of the insulin resistance along with the effect of saturated fatty acids, lipoproteins, leptin, and circulating proinflammatory cytokines translates into apoptosis of islet β cells [16]. As glucose is taken up poorly by cells in the organism, this causes postprandial glucose levels to remain consistently high for prolonged periods of time, resulting in glucose-related tissue toxicity [production of receptor for advanced glycation end-products (RAGE), endothelial dysfunction, histone hyperacetylation, DNA methylation, etc.] [2]. This toxicity affects microvessels and macro vessels (retinopathy, coronary arterial disease, etc.), nerves (peripheral neuropathy), and nephrons (decreased glomerular filtration and microalbuminuria), with deleterious clinical consequences. IR agonists such as chaetochromin derivatives and monoclonal antibodies with agonist activity on the IR have been reported to improve IR responsiveness and Akt activations, respectively, thus improving glucose metabolism at the cellular level in patients with peripheral insulin resistance [17,18].

Chronic inflammation and thrombosis

Glucose toxicity by aldose reductase activation initiates subsequent PKC-dependent nonosmotic nuclear factor (NF)-κB activation, resulting in the production and release of proinflammatory cytokines such as interleukin (IL)-6, IL-12, IL-10, tumor necrosis factor-α, etc. [19,20]. Similarly, inflammation in adipose tissue leads to the release of adipocytokines such as adipsin, adiponectin, leptin, tumor necrosis factor-α, and plasminogen activator inhibitor I. The vascular redox state is affected by transduction signal signals originating from inflammatory, obesity, and insulin-related pathways. Importantly, adipose tissues can modify the secretory profile when sensing paracrine signals of cardiovascular (CV) oxidative stress or injury [21]. Such inflammatory signals can transduce cellular signals in tissues such as fat, liver, muscle, heart, endothelium, etc., through toll-like receptor (TLR) signaling, which in turns activates inflammatory nuclear factors (NF-κB) feeding the chronic loop of persistent inflammation. In particular, TLR-2 and TLR-4 affect the frequency, plaque size, and lipid content of atherogenic plaque and the expression of inflammatory genes and cytokines (IL-12, monocyte chemoattractant protein-1, etc.) [22]. In this respect, colchicine, an anti-inflammatory agent, has been shown to decrease IL-1b, MI, acute coronary syndrome, and noncardioembolic stroke in phase III studies [23,24]. Canakinumab, a monoclonal antibody against IL-1b, is another perfect example of an anti-inflammatory drug that reduces recurrent CV events independent of lipid levels as shown in the CANTOS study [25].

For instance, circulating inflammatory factors can activate potentially life-threatening cell signaling such as thrombosis by platelet activation of both classical and alternative pathways [26]. Platelets are easily activated and can aggregate quite fast in response to such circulating cytokines, especially in low-flow areas such as the coronaries, the lower extremities, the brain, etc. [26,27]. The occlusion or sub occlusion of these vessels can result in infarctions or necrosis of important tissues such as the brain and the heart, increasing the risk for stroke and MIs [28–30].

In vessels that have an atherogenic lesion, in addition to the circulating inflammatory signals, local signals, along with plaque erosion, partial, or total rupture, can trigger thrombosis in the atherogenic suboccluded area or distal regions on that artery territory [29,30]. Infiltration of immune cells can be found in plaques and although these cells repair and replace tissue, its presence and the release of inflammatory chemoattractive substances worsen the thrombotic state and increase the risk of further plaque core necrosis and plaque instability, with the subsequent release of debris into the distal portions of the artery lesion, a condition that worsens under low shear stress conditions [31–33].

It is noteworthy that another less characterized player, RAGE, is implicated in deleterious effects on energy expenditure, weight gain, adipose tissue inflammation, and insulin resistance together with a high-fat diet. RAGE protects against high-fat diet-induced systemic inflammation and weight gain [34]. Also, elevated serum RAGE of more than 838.19 pg/ml can double the risk for CV events in patients with pre-existing CVD (a composite of MI, stroke, and CV death) [35].

Dyslipidemia and atherogenesis

Dysplipidemia and obesity are often present in patients with DM and can facilitate atherogenesis and atherosclerosis [36]. Fat droplets in cells, especially adipocytes, are essentially 'packed energy' that our body can use as a fuel source in times of fasting or when there is a need for extraphysical activity [37]. In contrast, carbohydrates are metabolized into energy using aerobic or anaerobic

mitochondrial pathways. If all the elementary energy requirements of the cell are met, then lipids are synthetized from carbohydrates, a process called 'de-novo lipogenesis' [38]. Lipids can be stored and converted back into burnable compounds (pyruvate) within the cell [37]. Lipids can themselves be a 'source of energy' in times of fasting that, along with their high affinity to cell membranes, can access the cells with minimum effort (vectors-exosomes) [37,39]. However, the distribution of lipids is aided by proteins as their physicochemical properties allow them to remain in the circulatory system, avoiding early absorption [40]. The synthesis of those proteins is mainly orchestrated by the liver. Those proteins are categorized according to their molecular density into very low-density lipoprotein (LDL), LDL, and high-density lipoprotein. Along with triglycerides, which are clusters of lipids, lipoproteins travel along the circulatory system to distal organs and tissues [40].

Chronic high levels of atherogenic LDL cholesterol along with increased non-high-density lipoprotein C and ApoB values in patients have been related to the progression of atherogenesis [41]. Oxidation of low-density lipoprotein (oxLDL) is an important condition that represents oxidative stress and increases the atherogenic and inflammatory properties of LDL [42]. In addition, elevated serum levels of oxLDL are associated with the incidence of coronary disease [42,43]. Therefore, a logical therapeutic target is the reduction of the LDL cholesterol by statins or by the novel proprotein convertase subtilisin/kexin type 9 inhibitors. Statins inhibit the production of cholesterol by inhibiting the transformation from hydroxymethylglutaryl-coenzyme A into mevalonic acid (primitive fatty acid) [44]. In contrast, proprotein convertase subtilisin/kexin type 9 inhibitors increase LDL-receptor density on the cell surface, facilitating LDL intake by the cell and decreasing circulating LDL, thereby facilitating plaque regression as reported recently in the GLAGOV study [45].

Concurrently, hyperglycemia contributes toward the development of atherosclerosis and arterial stiffness [46]. Chronic damage to the endothelium and the effects of inflammatory cytokines on endothelium play important roles in the genesis and stability of the plaque. The cellular mechanisms of media thickening and proliferation, presence of endothelium- adhesion molecules (vascular cell adhesion molecule 1 and intercellular adhesion molecule 1), and infiltration of macrophages in the subintima are regulated by epigenetic mechanisms and posttranslational modifications [47,48]. Hyperglycemia induces hyperacetylation of histone H3K9/K14 in 88 genes codifying for diabetes, 52 genes for hypertension, and 84 genes for CV disorders among other diseases [49]. It is particularly noteworthy that hyperacetylation of the histone H3K9/K14 in the endothelium results in the expression of important glucose metabolism and metalloproteinases regulating genes such as heme oxygenase 1 (*HMOX1*), IL-8 precursor, matrix metalloproteinase (*MMP*) protein-10, cysteine/glutamate transporter (*SLC7A11*), and *MMP1* [49]. ILs and metalloproteinases are closely related as they are both regulated by proinflammatory signals and participate in vascular remodeling, particularly in plaque progression and plaque instability [50,51]. MMP inhibitors have been used to stabilize plaques, but there is a need for more selective targeting of MMPs as broad-spectrum inhibitors exert dual effects on the plaque [51].

Hyperglycemia also induces DNA methylation of important genes for glucose metabolism, G-coupled protein receptors (GPRs), and insulin growth factor proteins such as *ABCC11*, *ADAD1*, *ADAM8*, *BCL3*, *CCDC61*, *CEP120*, *CSF1R*, *CSTL1*, *CTTNBP2NL*, *EGLN3*, *ENOX1*, *ERAS*, *FAM107A*, *FASLG*, *GADD45B*, *GNG2*, *GPR39*, *GPR62*, *GRK7*, *HMGB2*, *HNRNPL*, *HYOU1*, and *IGFBPL1* [49]. Gene expression and suppression persist for up to 6 days in the endothelium after the hyperglycemic episode *in vitro* [2]. Here is the importance of novel GPR agonists which currently are underway in an effort to improve GPR signaling in tissues and its metabolic benefits in patients with diabetes [52,53].

Other epigenetic mechanisms such as microRNAs (miR) can regulate gene expression post-transcriptionally, directly exert their effects in signal pathways, and reach other cells when included in extracellular vesicles called 'exosomes' [54]. miR-941, miR-208b, miR-197, and miR-223 have been found to have diagnostic value in predicting CV events or CV death [55–57]. miR-126-5p has been associated inversely with the complexity of CAD with low serum levels in multivessel disease and high SYNTAX scores in patients with stable angina [58]. Some epigenetic therapies are underway as potential antithrombotics such as miR-19b for use in patients with unstable angina [59]. Also, a bigger epigenetic factor, long noncoding RNAs in exosomes, such as exosomal long noncoding RNA-growth arrest-specific 5 (long noncoding RNA GAS5), can increase the apoptosis of macrophages and endothelial cells in atherosclerosis [60].

Hypertension

The renin–angiotensin–aldosterone system has been proposed as a feasible model to explain secondary hypertension as the cause of primary hypertension is unknown [61]. Inflammatory cytokines have a major impact on the endothelium by affecting the capacity of energy metabolism (mitochondrial dysfunction) and the release endothelial nitric oxide synthase, which is an important vasodilator, thus affecting vascular relaxation and inducing arterial stiffness [62–64]. These inflammatory cytokines chemoattract macrophages and lymphocytes, which can produce and release reactive oxygen species and angiotensin II (AngII) [62,63]. Reactive oxygen species activates NF-κB signaling, amplifying the vicious cycle of local inflammatory response, and AngII increases the blood flow by inducing constriction of the media of arteries, thereby increasing blood pressure [65].

As this inflammatory stage is chronic, AngII can consistently and continuously induce an increase in blood pressure. This high-flow system induces the development of media hypertrophy, reducing even more the arterial lumen, which in turn increases resting blood pressure values [66,67]. Unchecked stages of this condition may result in the onset of secondary hypertension and the need for a medical intervention with lifestyle changes and antihypertensives [67].

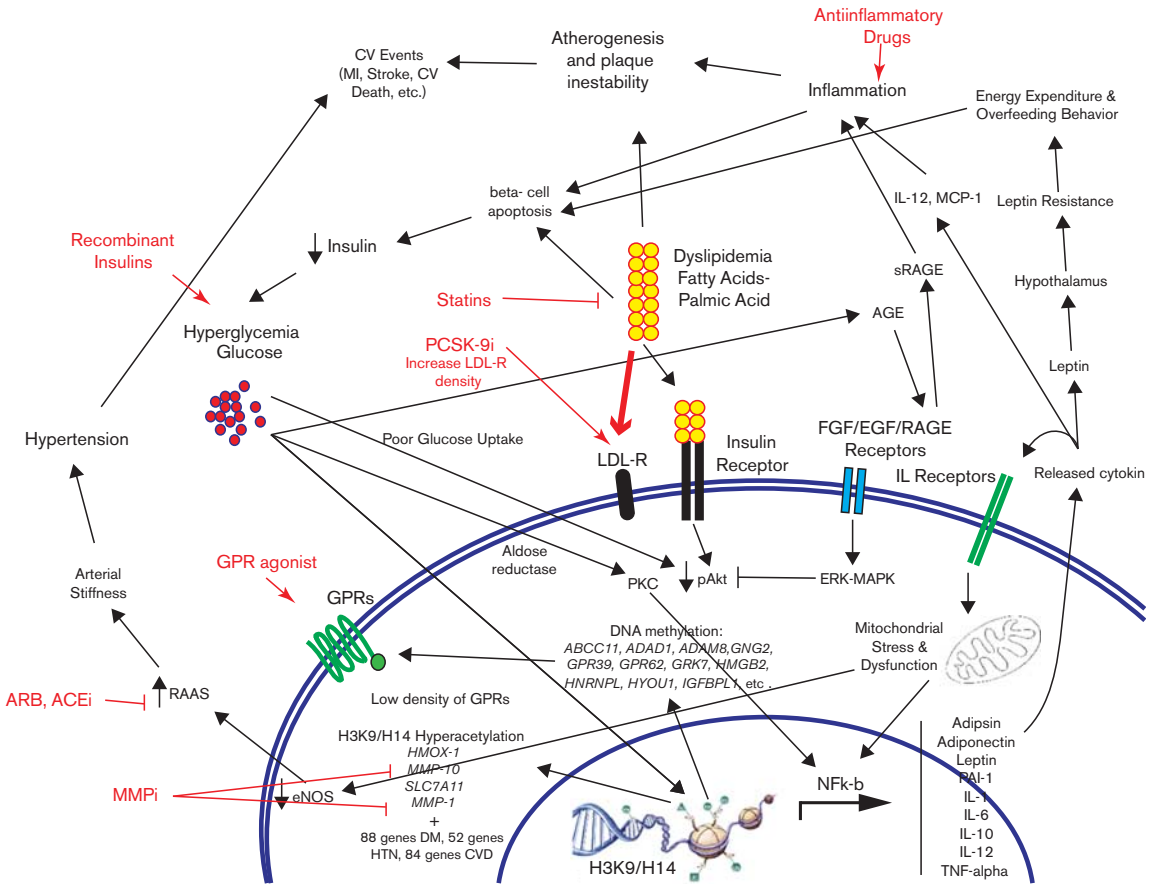
The persistence of a high-flow system together with inflammation, dyslipidemia, and hyperglycemia increases the risk of atherogenic plaque erosion or rupture, hemorrhage (especially microcirculation), and thrombosis [68].

Sympathetic regulation of blood pressure by catecholamines also plays an important role in the presence and persistence of hypertension [61,69]. Renal denervation was proposed to treat uncontrolled hypertension without relevant and consistent results in the SYMPLICITY HTN-3 trial, pointing to the utility of targeting renin–angiotensin–aldosterone system, as it may be more clinically relevant than the sympathetic pathway [70].

Discussion

The optimal balance between genes codifying for epigenetic modulators and associated proteins required for the transcription of these modulators can be affected by cellular toxic products such as glucose itself and glycation end-products leading to transcriptional stages of inflammation (oxidative stress, cytokine production, and release and apoptosis), endothelial dysfunction (decrease in nitric oxide production and release of AngII), and down-regulation of GPR density. oxLDL plays a role in the pathogenesis of CVD by desensitizing the IR pathway and IR-dependent glucose uptake, thus reinforcing hyperglycemia and its toxic effects in cells. Another CVD risk factor described in this review is hypertension, triggered by inflammatory signals, together with the inability to control vascular relaxation by nitric oxide and angiotensin, both of which are endothelium-release-dependent factors. Taken together with atherovascular lesions, this could result in a

Fig. 1



Cellular and clinical implications in DM that precipitate CVD and their importance for therapeutics.

mature hypertension phenotype and its associated increased risk for CV morbidity and/or CV death (Fig. 1).

There are still a few gaps in the understanding of these signals. For instance, soluble RAGE characterization at the epigenetic level and its inflammatory and Akt signal competition properties should be investigated further. Exosome-mediated long or short RNA information transfer and signal transductions have not been fully characterized and standardized for any ethnical or environmental variations. However, as the field advances, it is even more evident that some or most of the signal pathways are inter-related following a pattern that starts with the cellular response to high concentrations of glucose and cholesterols.

Conclusion

Diabetes is characterized by the presence of risk factors and common important epigenetic, genetic, and cellular signaling mechanisms that lead to or accelerate the development of CV disease and progression.

A better understanding of such cellular mechanisms can translate into a more selective and personalized therapy for the primary and secondary prevention of CV events in patients with diabetes.

Acknowledgements

Conflicts of interest

There are no conflicts of interest.

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