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REVIEW ARTICLE

The Role of Proprotein Convertase Subtilisin/Kexin Type 9 (PCSK9) in Cardiovascular Homeostasis: A Non-Systematic Literature Review

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Abstract: ***Background*:** Proprotein convertase subtilisin/kexin type 9 (PCSK9) has been gaining major attention recently after the emergence of data showing the promising role of these proteins in lipid homeostasis and atherosclerosis process, glucose and blood pressure regulation.

***Materials and Methods*:** PubMed, EMBASE, Scholar and Scopus databases were searched to identify randomized controlled trials, observational studies, in-vitro trials and reviews about the role of PCSK9 in cardiovascular homeostasis.

***Results*:** PCSK9 was found to have major impact on lipid homeostasis and inflammatory process through regulation of low-density lipoprotein receptors. Furthermore, inflammation was found to stimulate the expression of PCSK9 in various cells. As for glomerular proteinuria, a positive correlation was determined between PCSK9 levels and the degree of proteinuria. Hypertension, a major cardiovascular risk factor, is likely affected by PCSK9 levels through their role on epithelial sodium channel (ENaC) surface expression. Likewise, some studies show that PCSK9 is associated with higher fasting blood glucose and plasma insulin, demonstrating a potential role of PCSK9 in glucose homeostasis. The role of PCSK9 in cardiovascular homeostasis is one that is still not completely unraveled.

Conclusion: Studies have clearly shown the implication of PCSK9 in the cardiovascular risk factors: the higher the PCSK9 levels, the higher the risk of atherosclerosis, fasting plasma glucose and insulin resistance. Inhibiting PCSK9 may therefore theoretically prove to present great benefits in diabetic patients with high cardiovascular risk.

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1. INTRODUCTION

The role of proprotein convertase subtilisin/kexin type 9 (PCSK9) recently emerged as a crucial component in lipid homeostasis after extensive investigational efforts conducted to unveil the complex mechanisms associated with familial hypercholesterolemia. In 2003, Abifadel *et al.* were the first to identify mutations in the genes encoding for PCSK9 as a cause of autosomal familial hypercholesterolemia (FH) [1]. Investigating the role of PCSK9 showed its association with low-density lipoprotein (LDL) receptor intracellular degradation [2-5]. The relationship between LDL-cholesterol (LDL-C) serum levels and atherosclerosis has been extensively studied. Higher LDL-C levels are strongly correlated with atherosclerosis and higher cardiovascular comorbidities and mortality [6]. Whereas gain-of-function mutations was associated with increased levels of LDL-C and early onset of atherosclerosis [7], loss-of-function mutations on the other hand was linked to a lower LDL-C and a subsequent decrease in cardiovascular risk [8, 9]. This prompted evaluation of the therapeutic potential of inhibiting PCSK9, where a monoclonal antibody inhibiting PCSK9, when added to conventional therapy (statins), was found to further reduce the incidence of cardiovascular events in the Open Label Study of Long Term Evaluation against LDL-C (OSLER) trials [10], as well as reduce mortality and improve outcomes in the recently published "Further Cardiovascular Outcomes Research With PCSK9 Inhibition in Subjects With Elevated Risk" (FOURIER) trial [11].

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Regarding its physiology, the PCSK9 protein is expressed in significant amounts in the liver, intestine and the kidneys [12]. In addition to its role in lipid metabolism, PCSK9 is also expressed in pancreatic insulin-secreting beta cells and has been shown to play a part in normal insulin homeostasis [13]. Besides, the role of PCSK9 extends to involve regulation of inflammation, blood pressure and carcinogenesis [14-16]. This non-systematic review summarizes the data present to date and pertaining to the emerging role of PCSK9 in the different aspect of cardiovascular homeostasis focusing on dyslipidemia, glomerular proteinuria, insulin secretion, blood pressure regulation and inflammation. In order to compile a comprehensive data for this review, PubMed, EMBASE, Scholar and Scopus databases were searched to identify randomized controlled trials, observational studies, in-vitro trials and reviews about the role of PCSK9 in cardiovascular homeostasis.

2. ROLE OF PCSK9 IN CARDIOVASCULAR HOMEOSTASIS

LDL receptors (LDL-R) play a key role in regulating plasma LDL levels, a major risk factor in the development of atherosclerosis, by internalization of the LDL/LDL-R complex into the hepatocytes resulting in LDL particle degradation and subsequent recycling of the LDL-R on the cell surface. PCSK9 is a crucial protein in LDL metabolism since it plays a pivotal role in the degradation of the LDL-R [17]. As shown in Fig. (**1**), PCSK9 causes degradation of LDL-Rs by 2 distinct mechanisms: (1) extracellularly, where it binds to LDL-R leading to its internalization and lysosomal degradation; and (2) intracellularly, where it is directly sorted to the lysosomes along with LDL-R leading to its degradation [18]. Regulation of PCSK9 appears to be mediated by endogenous proteolytic cleavage as well as hormonal and nutritional status [17, 18]. Most significantly, levels of both LDL-R and PCSK9 are co-regulated by the sterol regulatory element binding protein-2 (SREBP-2); whenever intracellular cholesterol is low, SREBP-2 will act by increasing LDL-R production to increase cholesterol uptake, while also increasing PCSK9 expression to regulate such process and prevent excessive cholesterol uptake [19]. Understanding the molecular pathways of PCSK9 functions and regulations has led to three different therapeutic approaches: gene silencing that targets both the intra- and extracellular functions of PCSK9, monoclonal antibodies exclusively targeting circulating PCSK9, and therefore its extracellular function, and mimetic peptides competitively binding to extracellular PCSK9 domains of LDL-Rs (Fig. **2**) [17, 20]. Furthermore, studies suggest that other pathways, including inflammatory pathways, may also be involved in mediating the effects of PCSK9 on vascular biology [20, 21]. The major aspects of the role of PCSK9 in cardiovascular homeostasis are summarized in Table **1**.

2.1. PCSK9 and Dyslipidemia

Atherosclerosis is a chronic, lipid-induced, inflammatory disease of the arterial wall involving intricate and multifactorial processes, such as endothelial dysfunction, inflow and alteration of LDL particles, leukocyte recruitment, foam cell formation and plaque expansion [22]. Individuals with subclinical atherosclerosis are at higher risk for overt cardiovascular disease (CVD) [22]. Hypercholesterolemia is a major cardiovascular risk factor that speeds up the process of atherosclerotic plaque development, consequently increasing the incidence of stroke and myocardial infarction [20]. Therefore, finding new plasma markers related to subclinical atherosclerosis such as PCSK9 can have important clinical implications for cardiac patients [21].

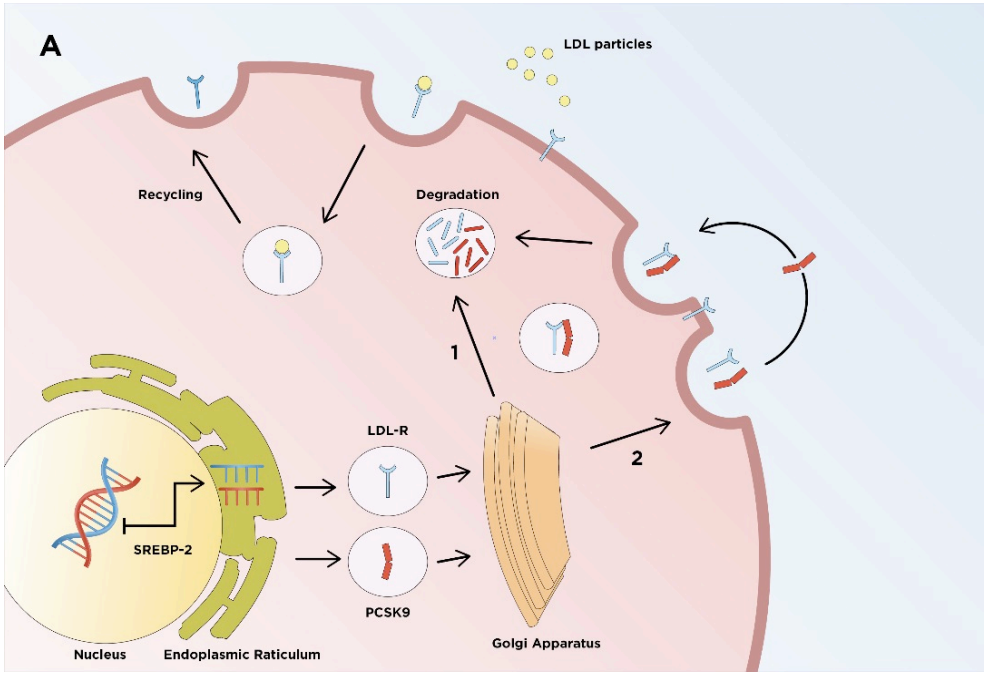

Fig. (1). Mechanism of Action of PCSK9. When their plasma levels are low, SREBP-2 stimulates production of both LDL-R and PCSK9. PCSK9 acts by 2 mechanisms: either intracellularly (**1**) by inducing lysosomal degradation of LDL-R or extracellularly (**2**) by binding to the LDL-R on the cell membrane and inducing its internalization and lysosomal degradation.

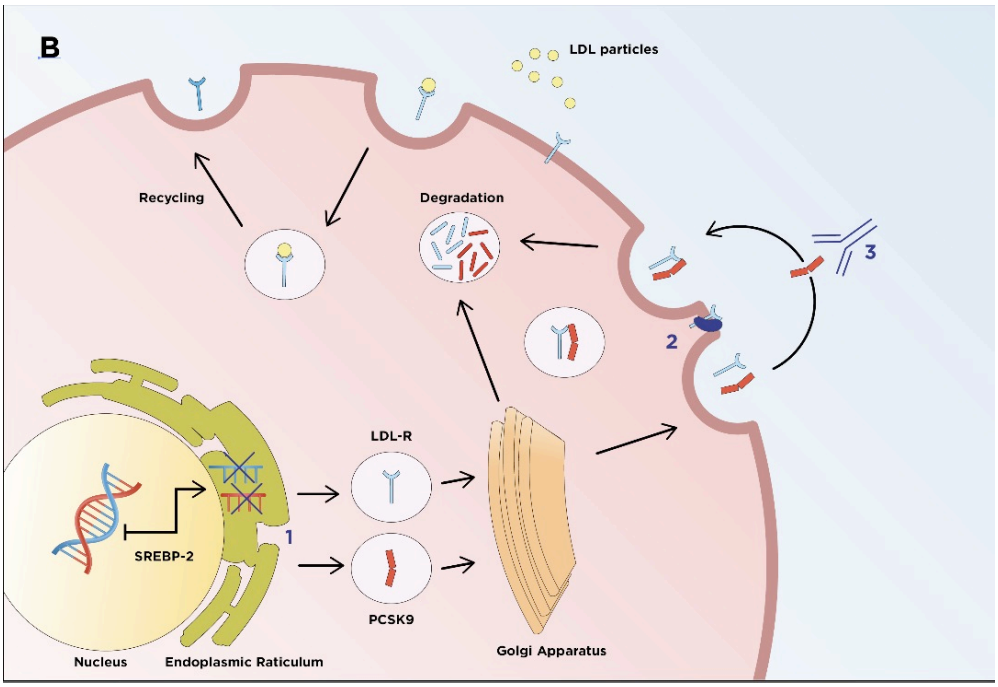

Fig. (2). Proposed mechanisms of targeting PCSK9. Silencing of PCSK9 action was investigated via 3 mechanisms: (**1**) gene silencing that targets SREBP2, (**2**) mimetic peptides competitively binding to extracellular PCSK9 domains of LDL-Rs and (**3**) monoclonal antibodies targeting circulating PCSK9 and therefore its extracellular function.

Table 1. Studies defining the role of PCSK9 in cardiovascular homeostasis.

Effect	Molecular Mechanism	References
Atherosclerosis	PCSK9 levels are positively associated with fibrinogen levels in patients in patients with CAD independently of vascular risk factors	[25]
	PCSK9 levels were linearly associated with the fraction and amount of necrotic core tissue in coronary atherosclerosis	[27]
	Statins have shown to increase PCSK9 levels as early as within 24hours and the increase is dose-dependent	[30, 31]
Inflammation	In stable CAD patients, there is a positive correlation between PCSK9 and WBC levels independently of lipid parameters	[33]
	In vitro, TNF-α increases PCSK9 levels	[34]
	There is a significant positive correlation between PCSK9 and CRP levels in women	[35]
	In vitro, resistin induces PCSK9 levels	[36]
	PCKS9 is elevated in sepsis, associated with decreased pathogen lipids and endotoxin clearance by liver and lower survival in septic patients	[38-41]
Glucose Homeostasis	Among adult and pediatric populations, epidemiologic studies showed conflicting correlations between PCSK9 levels and fasting blood glucose and insulin	[33, 58-60]
	Glucagon decreases expression of PCSK9 mRNA while insulin increases it	[57]
	PCSK9 and HbA1C are proportionally correlated in diabetic patients	[60]
	PCSK9 mRNA is expressed in pancreatic beta cells. PCK9 plays a role in beta cell function, and it protects them from oxidative stress and death induced by LDL.	[13-56]
Blood Pressure	*In vitro*, PCSK9 activity reduces ENaC surface expression thus decreasing blood pressure	[44]
	In vivo in mice, PCSK9 deficiency was not associated with increased ENaC expression	[15]
	Rare variants of PCSK9 may have an effect on diastolic and systolic blood pressure regulation, possibly by interfering with ENaC function	[52, 53]
Glomerular Proteinuria	PCSK9 levels are elevated in subjects with glomerular proteinuria and nephrotic syndrome and correlated with high LDL-C levels in those patients	[63-65]
	PCKS9 is decreased in chronic kidney disease patients on hemodialysis	[66]

Specifically, PCSK9 modulates atherosclerosis mainly via the LDL-R. In mice fed with a high-fat, high-cholesterol diet, PCSK9 gene inactivation significantly decreased aortic cholesteryl-esters formation. However, these were prominently increased by overexpression of PCSK9, leading to accelerated development of atherosclerotic plaque [23]. In LDL-R-deficient mice expressing null, normal, or high levels of PCSK9, the circulating cholesterol levels and aortic accumulation of cholesteryl esters were similar to those of wild-type mice, indicating that the harmful effect of PCSK9 on atherogenesis is mediated mainly by degradation of LDL-R. Results of this study show a direct relationship between PCSK9 and atherosclerosis in vivo: PCSK9 overexpression is pro-atherogenic, whereas its absence is protective [23].

On a clinical level, elevated PCSK9 serum concentrations were shown to predict cardiovascular events in patients receiving stating therapy and with stable coronary artery disease (CAD) and well-controlled LDL-C concentrations [24]. Serum PCSK9 concentration was also proven to be a significant predictor of carotid intima-medial wall thickness (IMT), a measure of subclinical atherosclerosis and predictor incident coronary heart disease in asymptomatic individuals [21]. A more recent study indicates that in patients with stable CAD, plasma PCSK9 concentration is independently and positively associated with fibrinogen [25], a known independent risk factor for adverse cardiac events in cardiac patients during short and long-term follow up [26]. Another study has shown that PCSK9 would be a potential therapeutic target for atherosclerosis beyond LDL-C lowering, as PCSK9 levels were linearly associated with the fraction and amount of necrotic core tissue in coronary atherosclerosis independent of LDL-C levels and statin use [27].

However, other studies have shown that PCSK9 concentration is of low clinical utility. In patients with acute coronary syndrome (ACS), high initial PCSK9 plasma levels did not predict mortality at 1 year [28]. A limitation for the use of PCSK9 as a marker for atherosclerosis and a predictive prognostic factor is the fact that most cardiac patients are on LDL-C-lowering statin therapy, as per most the current guidelines for prevention of atherosclerotic disease and [29]. In fact, it was shown that this class of drugs increases PCSK9 levels [30]. Welder *et al*. reported that high-dose atorvastatin treatment resulted in the elevation of PCSK9 protein levels, disrupting the correlation between serum levels of PCSK9 and LDL-C [30]. Similarly, Guo *et al*. (2013) conducted a single-center study on a small sample of Chinese patients to investigate the short-term impact of low-dose atorvastatin on PCSK9 in humans [31]. The study showed that treatment with atorvastatin 10 mg/day and 20 mg/day showed a trend towards an incremental increase in PCSK9 levels at 8 weeks, with both doses achieving lipid-lowering effects. Hence, the higher the statin dose, the higher is the expression of PCSK9. This could explain the non-linear statin dose-response relationship: with higher doses of the statin, clinicians witness a less than expected incremental LDL-C lowering [30]. In regards to other LDL-C lowering drug, short-term use of ezetimibe was also found to significantly increase plasma PCSK9 levels in patients with dyslipidemia [32]. Ezetimibe and lovastatin combination therapy however did not cause an additional increase in PCSK9 compared with monotherapy with either agent alone. Therefore, a combination of low-dose statin plus other lipid-lowering drugs can be an alternative to minimize the effects of statins on PCSK9 [32]. All in all, the increase in PCSK9 levels by statins can largely counteract statin-induced increases in hepatic LDL-R levels, which could limit the linear LDL-C-lowering effects of statins [31].

The genetic aspect of PCSK9 concerning statin therapy response is also well studied. Missense and loss-of function mutations in the PCSK9 gene are associated with increased statin response and substantial hypocholesterolemia, highlighting the potential benefit of PCSK9 inhibition and its potential additive effect in combination with statins [20]. Evidently, PCSK9 is shown to be strongly correlated with cholesterol-mediated atherosclerotic processes and fibrinogen levels, both strong predictive factors for cardiovascular diseases. However, the use of PCSK9 as a clinical prognostic factor for cardiovascular morbidity and mortality may be limited by the use of statins, the present cornerstone of primary and secondary prevention of cardiovascular disease.

2.2. PCSK9 and Inflammation

Inflammation stimulates the expression of PCSK9, but the mechanism by which it increases PCSK9 messenger ribonucleic acid (mRNA) levels is unknown [14]. Conversely, the expression of inflammatory genes is reduced by knockdown of PCSK9 [33]. In mice, systemic inflammation induced by administration of lipopolysaccharide led to increased expression of PCSK9 and decreased hepatic levels of LDL-R, which was associated with a significant increase in circulating LDL-C levels [14]. Furthermore, several studies tested the relationship between PCSK9 expression and inflammatory markers. In-vitro studies showed that tumor necrosis factor –alpha (TNF- a) may increase PCSK9 levels [34]. In another study, a positive correlation was described between PCSK9 levels and C-reactive protein (CRP) in women but not in men [35]. This suggests a relationship between low-grade inflammation and PCSK9 regulation. In addition, an *in vitro* study showed that PCSK9 levels were induced by resistin, a pro-inflammatory adipokine that is mainly released by macrophages [36]. However, these findings were not translated *in vivo*, whereby this association was not demonstrated when studied in lean or moderately obese subjects [37]. Consequently, further investigations are needed to test the relationship between PCSK9 and inflammatory cytokines.

Besides, PCSK9 has been shown to play a vital role in innate immune response and sepsis. Pathogen lipids such as lipopolysaccharide and endotoxins are carried in lipoprotein particles and are hepatocyte LDL-Rs, which are primarily regulated by PCSK9 [38, 39]. Since PCSK9 decreases clearance of LDL particles, it consequently decreases clearance of pathogen lipids that are carried by those LDL particles [40]. Conversely, reduction in PCSK9 function results in increased endotoxin clearance, decreased inflammatory response, and improved survival in septic mice as well as septic humans who carry loss-of-function variants of the PCSK9 gene [38]. Moreover, plasma PCSK9 are greatly increased during sepsis and are associated with multi-organ failure [41].

Inflammation has also been significantly associated with atherosclerosis and CAD [42]. This is explained by the recruitment and accumulation of inflammatory cells into the vessel wall after endothelial injury, leading to cytokine release and an increase in the lipid content of the atheromatous plaque and rendering it more vulnerable to rupture [43]. The potential role of PCSK9 in the atherosclerotic process beyond lipid homeostasis and regulation of LDL-R is supported by Li *et al*. (2014), who studied the relationship between WBC count and PCSK9 in patients with stable CAD [33]. Li reported a significant positive correlation between high WBC count, including lymphocytes, and PCSK9 levels, after adjustment for covariates of lipid parameters, but was non-significant in the female population [33]. The association between PCSK9 and WBC count is independent of concurrent dyslipidemia and expands the role of PCSK9 in the atherosclerosis process beyond cholesterol regulation. Moreover, high initial PCSK9 levels were associated with inflammation in the acute phase of ACS [28]; yet the exact mechanism linking PCSK9 and markers of inflammation in atherosclerosis and CAD remains however to be determined.

2.3. PCSK9 and Hypertension

Hypertension is a major independent risk factor for CAD for all age, race, and sex groups [44], and hypertension-associated target organ damage is attributed to diverse pathophysiological mechanisms involving various neurohormonal pathways. The epithelial sodium (Na+) channel (ENaC) plays an important role in absorption of Na+ across epithelial membranes, including those of the kidney tubules, lung, distal colon, and sweat gland ducts [15]. This channel is critical for Na+ homeostasis and blood pressure control, thus a defect in its regulation causes blood pressure alteration [45]. ENaC concentration is regulated by proteolytic cleavage by serine and other proteases such as furin [46], in a process controlled by negative feedback depending on intracellular Na+ concentration [47]. Sharotri *et al*. [44] investigated the presence of additional proteases involved in the regulatory process of ENaC, and one of the proteases found was PCSK9. In contrast to furin, which activates ENaC by proteolytic cleavage of extracellular domains, PCSK9 inhibits ENaC by decreasing its cell surface expression mainly by increasing its degradation in the biosynthetic pathway; it also decreases ENaC exocytosis and increases the rate of ENaC degradation by proteasomes.

Alterations in ENaC regulation account for most genetic forms of hypertension [48]. This implies that PCSK9 could modulate cardiovascular risk in part through its regulation of ENaC, and thus a reduction in PCSK9 activity would increase renal Na+ absorption and intravascular volume, consequently elevating the risk of hypertension and associated cardiovascular disease. Thus, despite similarities in mechanism of PCSK9 regulation of ENaC and LDL-R, it is evident that these regulations have opposing effects on cardiovascular risk. In both cases, PCSK9 reduces surface expression through a change in trafficking, culminating in increased degradation. Nonetheless, PCSK9 regulates ENaC and the LDL-R through different binding sites; hence PCSK9 mutations that disrupt LDL-R regulation may not have similar effects on ENaC.

Building on these findings, Berger *et al*. [15] wanted to assess the physiological consequences of PCSK9-deficiency on blood pressure and electrolyte homeostasis *in vivo*. However, his data did not support the model suggested by Sharotri *et al*. [44], since PCSK9-deficiency was not associated with an increased ENaC expression. Their main observation involved an increased abundance of the cleaved (i.e. active) form of ENaC in PCSK9-/- mice compared with control mice under basal conditions. PCSK9 deficiency did not alter blood pressure and sodium homeostasis in mouse models of hypertension, and the neutral effect of PCSK9 on blood pressure was observed in both salt-insensitive and salt-sensitive models. Thus, these are reassuring results in terms of PCSK9's effect on blood pressure regulation, in light of the ongoing development and use of PCSK9 inhibitors in hypercholesterolemia and cardiovascular diseases.

Hypertension is particularly more prevalent in African-Americans [49], owing in part to the genetic susceptibility of this population to hypertension-related morbidity and mortality [50, 51]. Using genomic data from the Hypertension Genetic Epidemiology Network (HyperGEN), the association of PCSK9 polymorphisms with blood pressure was tested [52]. Using genome wide association study (GWAS), a minimal effect on diastolic blood pressure (DBP) was attributed to two single-nucleotide polymorphisms (SNPs), but a significant cumulative effect on DBP of all PCSK9 rare variants was identified. . Moreover, in this study, these gene-based associations were tested in the population belonging to the "REasons for Geographic And Racial Differences in Stroke" (REGARDS) study [53], and a cumulative association with systolic blood pressure was found, indicating that rare variants in PCSK9 may play a role in blood pressure regulation, possibly through interfering with ENaC function. Overall, the role of PCSK9 in blood pressure regulation remains controversial with conflicting data in the literature. This however could be explained by the scarcity of such data and the need for further more extensive research investigating its particular role in regards to blood pressure regulation.

2.4. PCSK 9 and Type 2 Diabetes Mellitus

The recent abundant PCSK9 literature available extends beyond lipid metabolism to investigate its role in glucose homeostasis and type 2 diabetes mellitus (T2DM). Pancreatic beta cells are known to express significant amount of LDL-R; these receptors are responsible for the uptake of exogenous lipoprotein. Exposure to large amounts of LDL and VLDL could lead to oxidative stress and subsequent death of pancreatic beta cells [54, 55]. *In vitro* studies also show that certain insulin-producing beta cell lines express significant amount of PCSK9 mRNA [56]. In fact, PCSK9 plays a crucial rule in the survival and normal function of pancreatic islet beta cells. This is shown through an *in vivo* study done with PCSK9-knockout mice, which when compared to control mice, carried more LDL-R but significantly less insulin in their pancreas and thus were glucose intolerant or hyperglycemic, and their islets exhibited signs of malformation, apoptosis and inflammation, likely due to cholesterol accumulation [13]. Several studies thus investigated the physiological role of PCSK9 expression in terms of glucose homeostasis. In one study, Persson *et al*. [57] examined the association between (1) PCSK9 and insulin levels, (2)

PCSK9 and glucagon levels, and (3) PCSK9 levels and a combination of both insulin and glucagon in rats. Firstly, glucagon treatment was shown to decrease the mRNA expression of PCSK9 by 60%. Insulin treatment, however, was shown to increase the expression of PCSK9 mRNA by 170%. Likewise, the combination therapy increased the PCSK9 mRNA expression to levels seen in the control group. In another study, Kappelle *et al*. [58] examined the effect of 24-hour insulin infusion on plasma PCSK9 levels in healthy volunteers and in patients with T2DM. There was not a significant change in the levels of plasma PCSK9 levels before and after infusion in healthy subjects and in patients with T2DM.

Several epidemiological trials tried to capitalize the above *in vitro* findings to study the correlation between PSCK9 levels and glucose metabolism among adult and pediatric populations. These studies showed conflicting associations between plasma PCSK9 and fasting glucose and insulin [35, 59, 60]. The Dallas Heart Study is a large multiethnic study with more than 3000 patients recruited, which aimed at measuring the levels of PCSK9 in a population with different genetic backgrounds [35]. The study demonstrated a significantly positive correlation between the levels of PCSK9 and fasting serum glucose, insulin, and insulin resistance among both males and females. PCSK9 levels were also significantly higher among diabetic patients compared to healthy controls. On the other hand, the Cohort on Diabetes and Atherosclerosis Maastricht (CODAM) study [61] investigated the role of PCSK9 levels in the occurrence of diabetic dyslipidemia, where they compared the levels of PCSK9 among patients with a wide spectrum of glucose tolerance: normal glucose metabolism, impaired glucose metabolism and T2DM [61]. Plasma PCSK9 levels were not significantly altered in patients with T2DM or impaired glucose metabolism compared with individuals with a normal glucose metabolism. In an effort to further investigate the role of PCSK9 in diabetics, Yang *et al*. (2015) enrolled 176 patients with T2DM and 629 patients without type 2 diabetes, in a hospital-based cross-sectional study to investigate the association of PCSK9 with hemoglobin A1C (HbA1c) in patients with T2DM not receiving lipid-lowering therapy [62]. This study showed that there is no statistically significant difference in the levels of plasma PCSK9 between the two groups. However, in this study, a univariate regression analysis to clarify the association between PCSK9 and T2DM was done which showed that the concentrations of PCSK9 and both fasting blood glucose (FBG) and HbA1c were proportionally correlated in patients with T2DM, but not in patients without T2DM. In addition, a subgroup analysis was performed among patients with T2DM between well-controlled patients (HbA1c < 7%) and poorly controlled patients (HbA1c ≥7%), which showed higher PCSK9 levels in the latter group. Similarly higher PSCK9 levels were obtained in patients with T2DM with a FBG ≥ 7 mmol/L when compared to patients with T2DM but with a FBG < 7 mmol/l. This study was the first to show the positive correlation between PCSK9 and both FBG and HbA1c among diabetic patients, irrespective of traditional atherosclerotic risk factors [62]. Hence, based on these analyses, PCSK9 can be considered as a marker of poorly controlled T2DM. Eventually, PCSK9 inhibition is thought to provide positive cardiovascular outcomes among patients with T2DM, and several clinical trials are currently ongoing to investigate these outcomes with PCSK9 inhibitors (Table **2**).

2.5. PCSK9 and Glomerular Proteinuria

In addition to being an indicator of renal dysfunction, proteinuria is an independent risk factor for cardiovascular morbidity and mortality [63]. It has been hypothesized that abnormalities in the PCSK9 pathway may contribute to the pathogenesis of glomerular proteinuria-associated alterations in apolipoprotein B - containing lipoprotein metabolism [64]. Kwakernaak *et al*. (2013) first documented that plasma PCSK9 levels are proportionally elevated in subjects with increased glomerular proteinuria compared to age- and sex-matched healthy subjects, while adjusting for concurrent statin treatment, renal function, and BMI [64]. PCSK9 levels did not decrease after maximal anti-proteinuric treatment, but individual changes in total cholesterol, non-HDL cholesterol and LDL-C were positively associated with changes in PCSK9 in response to maximal anti-proteinuric treatment. Besides, in patients with nephrotic syndrome and peritoneal dialysis patients, PCSK9 levels are elevated, which may explain the high levels of LDL-C in those patients with acquired LDL-R deficiency and hypercholesterolemia [65]. However, in patients with chronic kidney disease undergoing

Table 2. Major phase III clinical trials involving PCSK9 monoclonal antibodies.

Study	Recruitment/ Estimated Completion date	Primary Objective
• ***FOURIER:*** **Further Cardiovascular Outcomes Research With PCSK9 Inhibition in Subjects With Elevated Risk**	Fully enrolled (27,564) and Completed on November 2016 Results presented at the American College of Cardiology (ACC) on March 2017, and concomitantly published in NEJM [59].	• To compare the effect of *evolocumab* with placebo on the time to cardiovascular death, myocardial infarction, hospitalization for unstable angina, stroke, or coronary revascularization whichever occurs first in patients with clinically evident cardiovascular disease and already being treated with statins
• ***ODYSSEY:*** **Outcomes Evaluation of Cardiovascular Outcomes After an Acute Coronary Syndrome During Treatment With *Alirocumab***	Recruiting (estimated enrollment: 18,600)/ February 2018	• To compare the effect of *alirocumab* with placebo on the occurrence of cardiovascular events in patients who have experienced an ACS event 4 to 52 weeks prior to randomization

hemodialysis, PCSK9 levels are decreased yet still positively correlated with LDL-C levels, suggesting the persistence of PCSK9 as major player in LDL homeostasis in those patients [66]. This further indicates that the PCSK9 pathway may play a role in atherogenic lipoprotein changes in human proteinuria, and that its inhibition could become an important treatment target in proteinuric patients with chronic kidney disease.

CONCLUSION

The role of PCSK9 in cardiovascular homeostasis is yet to be completely understood. Studies have clearly shown the implication of PCSK9 in the atherosclerotic process, a major risk factor for future cardiovascular morbidity and mortality. Data concerning PCSK9 in regards to blood pressure regulation is still scarce and contradictory, and the role of PCSK9 in blood pressure homeostasis is still poorly understood. Inhibiting PCSK9 may theoretically prove to present great benefits in diabetic patients with high cardiovascular risk. PCSK9 production and insulin levels were found to be positively correlated: the higher the PCSK9 levels, the higher were the fasting plasma glucose levels and insulin resistance, but this association was not consistent across different studies. In addition, recent studies involving monoclonal antibodies targeting PCSK9 demonstrated promising results towards reduction in cardiovascular events, which will cement the role of PCSK9 as a major key player in cardiovascular homeostasis. Such findings support the need for further clinical trials to fully unravel the therapeutic potential and cardiovascular outcomes of targeting such protein. One major trial, whose long-awaited results were presented at the American College of Cardiology (ACC) Conference (March, 2017) is the FOURIER trial [11] showed that inhibition of PCSK9 with evolocumab on a background of statin therapy lowered LDL-C levels to a median of 30 mg per deciliter (0.78 mmol per liter) and reduced the risk of cardiovascular events after a median follow-up duration of 2.2 years. Currently, ODYSSEY [67] is an ongoing phase III clinical trial (Table **2**), evaluating the cardiovascular outcomes and safety of PCSK9 inhibitor, whose results are expected to be released in early 2018. In summary, LDL is definitely one major player in the process of atherosclerosis and consequent cardiovascular morbidity and mortality, and PCSK9 is definitely a crucial player in the homeostasis LDL metabolism as well as other cardiac-related processes.

LIST OF ABBREVIATIONS

ACS	=	Acute Coronary Syndrome
CVD	=	Cardiovascular Disease
CAD	=	Coronary Artery Disease
CRP	=	C-reactive Protein
ENaC	=	Epithelial Sodium (Na+) Channel
DBP	=	Diastolic Blood Pressure
FH	=	Familial Hypercholesterolemia
FBG	=	Fasting Blood Glucose
GWAS	=	Genome Wide Association Study
Hba1C	=	Hemoglobin a1C
IMT	=	Intimal-medial Wall Thickness
LDL	=	Low-density Lipoprotein
LDL-C	=	Low-density Lipoprotein – Cholesterol
LDL-R	=	Low-density Lipoprotein – Receptor
mRNA	=	Messenger Ribonucleic Acid
PCSK9	=	Proprotein Convertase Subtilisin/Kexin Type 9
SREBP-2	=	Sterol Regulatory Element Binding Protein-2
T2DM	=	Type 2 Diabetes Mellitus
TNF-a	=	Tumor Necrosis Factor – alpha

CONSENT FOR PUBLICATION

Not applicable.

CONFLICT OF INTEREST

The authors declare no conflict of interest, financial or otherwise.

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Declared none.

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