

Back to Basics: PCSK9 as a New Target for the LDL Receptor

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Introduction

Forty years after Goldstein and Brown's discovery of the low density lipoprotein receptor (LDLR)¹ and 25 years after the introduction of the statins², a cornerstone of the atherosclerotic disease management, the view of the scientific community studying the process of atherosclerosis turns once again to the LDLR through the action mechanism of proprotein convertase subtilisin/kexin type 9 (PCSK9)³.

The assessment of genetic polymorphism with unexpected results in large studies revived the hypothesis about the important role of high density lipoprotein cholesterol (HDL-C) in lipoprotein metabolism⁴; the apparent eternal debate about the contribution of triglycerides (TG) and their remnants to the phenomenon of atherogenesis⁵. Such advances have led to the re-prioritization of LDL management as a central and primary objective towards the prevention of the morbidity and mortality main cause in the Western world, atherosclerosis.

Once again, we return to the discovery that led Goldstein and Brown to a Nobel Prize in 1985, the LDLR¹; now seeking to avoid its degradation through the knowledge and understanding of PCSK9 action mechanism.

Biochemical and molecular bases

PCSK9, also called neural-apoptosis-regulated convertase 1 (NARC-1), is a serine protease, characterized by a three domain structure and a catalytic triad; it is the ninth member of the pro-protein convertase family³.

The PCSK9 gene is located on chromosome 1p32.3 and is 22-kb in length, comprising 12 exons encoding a 692 amino acid glycoprotein. This convertase is highly expressed in liver, intestine and kidneys⁶.

PCSK9 is synthesized as 74 kDa soluble zymogen (proPCSK9) which after an autocatalytic process within the endoplasmic reticulum, releases the pro-peptide (14kDa)-N terminal, resulting in a 60-kDa enzyme. This autoclavage process is necessary both towards its activation and release from the endoplasmic reticulum³.

The autocatalytic process allows the progression through the secretory pathway and thus directly interacts with the

LDLR. It is important to mention, that the catalytic activity, does not seem to be required for LDLR degradation, but only for the activation and secretion of PCSK9⁶.

This serine protease binds the epidermal growth factor-domain A (EGF-A) on the LDLR, upon which both PCSK9 and LDLR are internalized by the hepatocyte to be finally degraded within the lysosome (Figure 1)⁶.

Past, present and future of PCSK9

Ten years ago, Abifadel et al. have reported 3 families in France with familial hypercholesterolemia associated with increased functionality and expression of PCSK9, with no alteration in the LDL receptor or apo B structure⁷.

In 2005, two years later, Cohen et al. through the program/study Atherosclerosis Risk in Communities (ARIC) described the loss of PCSK9 function in African-American and Caucasian individuals. In the past, they reported a prevalence of 2.6% in deficit of function, with concomitantly reduced serum levels of low density lipoprotein cholesterol LDL-C (28% lower) and CV events, such as myocardial infarction, need for cardiac bypass surgery and coronary deaths (88%). The latter, in turn, showed a decrease in PCSK9 expression of 3.2%, with an average LDL-C reduction of 15% vs. control groups and reduction of 47% in CV events requiring cardiac surgery and mortality related to this cause. Interestingly, in the ARIC study, 50% of subjects had hypertension, 30% were smokers and 20% had diabetes⁸.

The inevitable comparison between the reduction on risk observed among individuals with a deficit of PCSK9 expression/function and the individuals evaluated in large studies with statins for 5 years (similar LDL-C decrease between the two groups with marked lower risk in patients with the genetic alteration), has led to the development of different strategies to silence this serin protease and thus, increase LDLR levels in the liver, with a consequent decrease of circulating LDL-C levels.

The early intervention might magnify the clinical efficacy of cholesterol-lowering therapy by attenuating the development and progression of atherosclerosis.

Therapeutic groups primarily involved in this strategy, would be those with familial hypercholesterolemia, statin-intolerance and perhaps patients with a very high cardiovascular risk with failure to meet the targets through the existing pharmacological armamentarium.

It also necessary to determine whether PCSK9 affects only LDL-C levels, or whether it may also exert a direct action on the vasculature and other structures. Its interaction/interplay with cholesterol ester transfer protein (CETP) and with the cholesterol efflux mediated by HDL-C is also yet to be determined.

Despite an undoubtedly interesting mechanism of action, several questions remain: the safety of PCSK9 combination

Keywords

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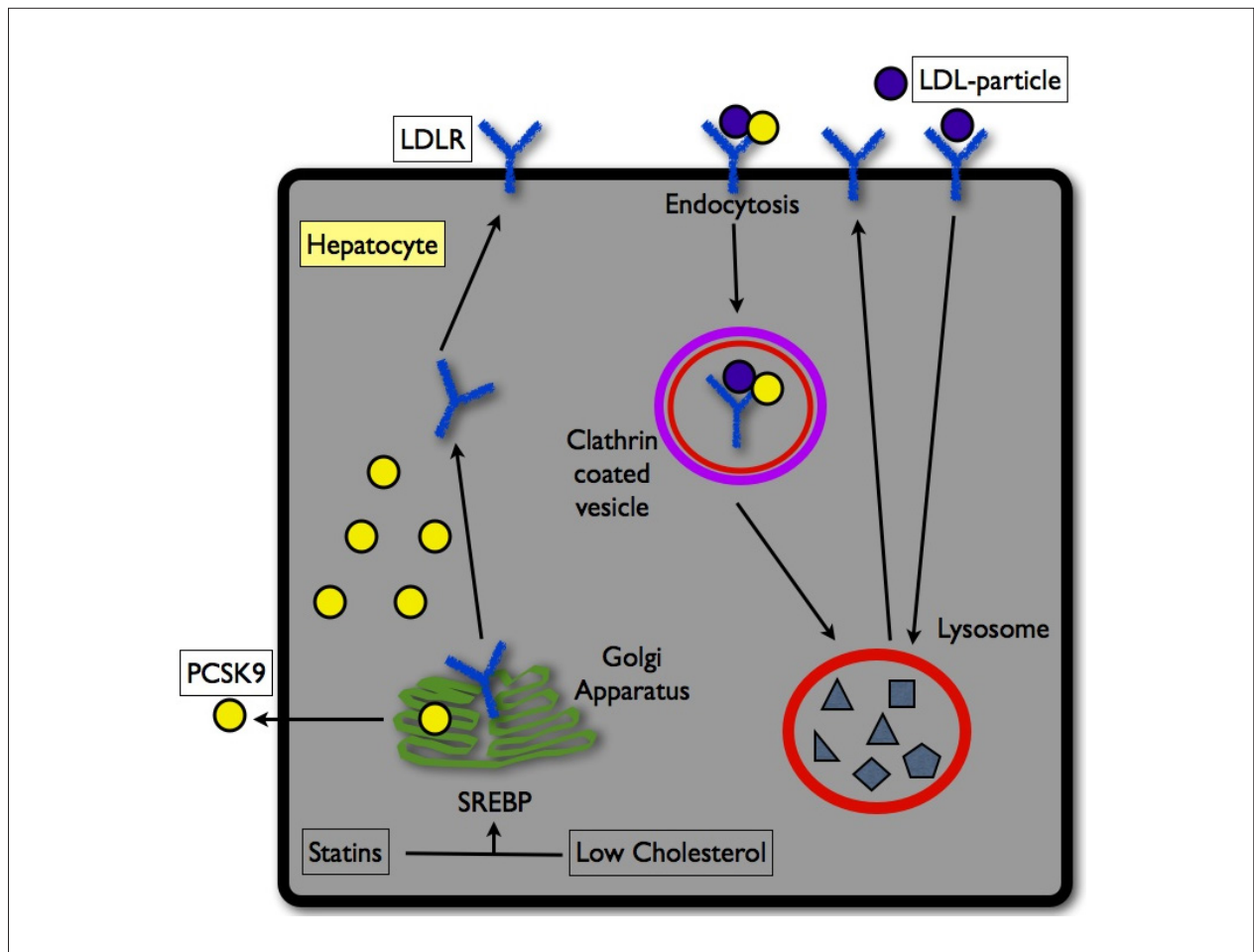


Figure 1 - PCSK9 regulates LDLR turnover through increased intracellular degradation.

with statins, the immune response to PCSK9 antibodies after a prolonged treatment, the real value of the pleiotropic effects that statins show and finally, the potential interactions with others enzymes and proteases.

Therapeutic strategies and PCSK9

Considering what was previously described, several lines of research are currently focused on PCSK9 modulators/inhibitors development in an effort to lower LDL-C mediated by an increase in the number of hepatic LDLRs.

Different pharmacological strategies are at different stages of development; from phase 3 to preclinical stages (Table 1)⁹.

Currently in phase 3, monoclonal antibodies define the direction for this therapeutic strategy, with demonstrated outcomes of safety and efficacy; peptide mimetics, small molecule inhibitors and attempts to gene silencing are some of the different strategies and approaches currently being pursued⁹.

The potential results of different pharmacological strategies, alongside the attractive hypothesis about the potential synergistic effect when administered with statins, make the PCSK9 modulation the most promising future therapy to avoid and prevent atherosclerosis process in those with functional LDLR.

Discussion

The PCSK9 discovery ten years ago, along with the recent results of studies in both intervention and genetic polymorphism around different lipoprotein fractions, open a promising future therapeutic field. The adverse effects of high doses of statins, and the logical and rational posing of the need to descend to values < 70 or 50 mg/dL LDL-C¹⁰, raised the need for alternatives in the management of this lipoprotein fraction, central to the atherosclerotic process.

Unexpected results and failures in different trials with different molecules (ezetimibe, niacin, fibrates, omega 3, protein inhibitors of cholesterol ester transfer-iCETP-) in a opposite way to the clear demonstration of statins beneficial effects in different scenarios and populations, highlighted the importance of not only decreasing LDL-C levels but also the mechanism by which this process occurs.

We cannot ignore the current controversy about the role that HDL-C plays in the atherosclerosis process; the clear results of population's studies with strong epidemiological evidence are contrasted with recent genetic analysis of polymorphism and Mendelian regression, along with the recent failures of intervention studies. Pending the results of

Table 1 - PCSK9, therapies in development

Target/Mechanism of Action	Drug/Agent	Phase
Monoclonal antibodies	SAR236553/REGN727	3
Monoclonal antibodies	AMG-145	2
Monoclonal antibodies	RN3169A/RG7652	2
Monoclonal antibodies	MPSK3169A/RG7652	2
Monoclonal antibodies	LGT209	2
siRNA Oligonucleotide	ALN-PCS02	1
Antisense Oligonucleotide	TBD	PRE-CLINICAL
Fusion protein using Alectin	BMS-962476	PRE-CLINICAL
Small peptide mimetic	SX-PCSK9	PRE-CLINICAL
Small PCSK9 modulator	TBD	PRE-CLINICAL

different studies (IMPROVE IT with ezetimibe and DEFINE with anacetrapib) the current role of the HDL-C (marker factor versus risk factor) resets, once again, the focus on LDL-C, as well as the mechanisms towards its modulation.

Statins have proven to be insurmountable compounds in their preventive and therapeutic effect on cardiovascular and cerebrovascular disease. However, 60% to 70% of patients with clinical cardiovascular events maintain persistently high LDL-C levels, even after reaching the maximum recommended doses. This clearly leads to the need of new approaches towards reducing the residual risk of cardiovascular events. Recent reports and systematic reviews of the statins effects on glycemic profiles and new cases of diabetes associated with the real dimension and importance of other adverse effects, such as statin-induced myopathy, adds more reasons to seek alternative therapies to combat atherosclerosis.

The scenario described above, directly and indirectly enhances the importance and emphasizes the PCSK9, its genetic and epidemiological evidence and potential therapeutic effect in this field.

The determination of a people's subgroup with a mutation in the gene encoding the PCSK9, low levels of LDL-C since an early age, and extremely low atherosclerotic cardiovascular risk, suggests not only the possibility of a new therapeutic strategy, but also the need to manage and maintain LDL-C at consistently lower levels and from an early age. The latter is perhaps the biggest basis from current prevention strategies, applicable today since the years 40', 50' and 60'.

In conclusion, we can say that we are facing a new potential therapeutic class, with an action mechanism in which the LDL receptor is located at the center of the process 40 years after its discovery, reaching a milestone in the history of atherosclerosis that meant to be a turning point in the prognosis and therapy of this disease, currently positioned as an undoubted public health priority.

Author contributions

Conception and design of the research, Acquisition of data, Analysis and interpretation of the data, Statistical analysis, Writing of the manuscript, Critical revision of the manuscript for intellectual content: Corral P.

Potential Conflict of Interest

No potential conflict of interest relevant to this article was reported.

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Study Association

This study is not associated with any post-graduation program.

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