New Genic agents to the Treatment of Dyslipidemia

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Abstract:

Several studies have shown a high correlation between concentration and development of low-density lipoprotein cholesterol (LDL-C) and the evolution of atherosclerosis and cardiovascular disease. Therefore, the reduction of LDL-C levels through lifestyle modification and/or pharmacological interventions has universally shown a decrease in cardiovascular events and mortality. In most cases, elevated blood lipid levels may be caused by alterations in certain genes encoding proteins involved in LDL metabolism, such as those associated with loss of function of the LDLr receptor gene, loss of function of the apoB gene or increased function of a PCSK9 protein .Family hypercholesterolemia is a hereditary disease in which a genetic alteration causes an increase in blood cholesterol.
Therapy is based on dietary control and drugs such as statins, ezetimibe or PCSK9 inhibitory monoclonal antibodies. An important scientific breakthrough in recent years is the ability to identify the genetic basis of diseases and possibly correct the defective gene by interfering with small interfering RNA (siRNA) or antisense oligonucleotides (ASO). The technologies of antisense oligonucleotides (ASOs) and small interfering RNAs (siRNAs) have also been developed for the treatment of hypercholesterolemia with the aim of controlling the expression of specific genes that play key roles in lipid metabolism. Anti-sense oligonucleotides have been developed to target apolipoprotein B, the main structural protein of VLDL, LDL and chylomicrons, apolipoprotein CIII or angiopoietin 3, both of which play a role in the regulation of triglycerides or apo(a). The siRNA approach works on the expression of PCSK9, a key modulator in LDL receptor catabolism. The purpose of this review is to present and discuss current clinical and scientific data on therapeutic evidence for new gene therapies in the treatment of hypercholesterolemia.

Introduction

Cardiovascular disease (CVD) is the leading cause of death in the world. According to the World Health Organization, an estimated 17.9 million people died from CVD in 2016, or 31% of all global deaths. Of these deaths, 5% are related to myocardial infarction and stroke. Dyslipidemia, characterized by high levels of atherogenic lipoproteins, including low-density lipoprotein cholesterol (LDLC), is known as the major risk factor for atherosclerotic cardiovascular disease (ASCVD). In recent decades, statins have become the cornerstone of medical treatment for dyslipidaemia. Many large randomized trials have validated the efficacy of statins as monotherapy or in combination with other drugs such as ezetimibe in reducing the main clinical manifestations of atherosclerosis, including coronary events, ischemic stroke and mortality from cardiovascular disease. In addition, many patients with hypercholesterolemia are unable to adequately reduce LDL levels with statin treatment, or are unable to tolerate these drugs due to adverse reactions such as myopathy, myalgia or hepatotoxicity. In recent years, studies have focused on optimizing the treatment of patients at higher risk of ASCVD events worldwide. An important therapeutic contribution came from PCSK9 inhibitory monoclonal antibodies, drugs characterized by good therapeutic efficacy and excellent tolerability profile, also derived from post-marketing safety data. In the last period, also in the treatment of dyslipidemias, there has been a significant development of the approach based on gene silencing with the possibility to use single-stranded antisense oligonucleotides or short strands of double-stranded RNA to "switch off" the expression of a specific gene. The advantage of this approach is that the molecules act within the cell,

preventing the translation of mRNA into the corresponding protein, while other types of treatments such as monoclonal antibodies can only act on the already mature and circulating protein. The reduction of harmful protein levels through RNA interference is applicable to all molecular targets, including those that are difficult to link with traditional approaches to pharmacological treatments based on small molecules or proteins. This review aims to provide an overview of the current status of new gene therapies for the treatment of dyslipidemia, in particular through the technique of antisense oligonucleotide (ASO), small interfering RNA (siRNA) and short palindromic repetitions at regular intervals (CRISPR)/CRISPR-associated 9 (Cas9) based on genome editing. [1-4]

APOB

Apolipoprotein B-100 (also known as Apolipoprotein B or Apo B) is a protein implicated in the lipid metabolism and is the principal component of the very low-density lipoproteins (VLDL) and low-density lipoprotein (LDL, the "bad cholesterol"). The APOB gene codifies apolipoprotein B (apoB). Agents designed to limit liver production of apoB can reduce the concentrations of all these atherogenic lipoproteins, such as LDL and VLDL.

PCSK9

Proprotein subtilisin/kexin convertase type 9 (PCSK9) is an enzyme codified by the PCSK9 gene in humans on chromosome 1. This protein plays a key regulatory role in homeostasis of cholesterol metabolism, primarily by reducing LDLr levels on the plasma membrane of hepatocytes. The reduction of LDLr levels on the hepatocyte membranes can lead to a reduction in LDL catabolism and an increase in blood flow. Gene dysfunction affecting the PCSK9 protein may reduce or increase circulating cholesterol. LDLs are removed from the blood by LDLRs on the surface of the hepatocytes and are internalized. When PCSK9 binds to an LDLR, the receptor is destroyed along with the LDL particle. If the PCSK9 does not function, the receptor returns to the surface of the cell and can continue to remove the LDL particles from the bloodstream. Some mutations increase its activity, reducing LDLR levels and increasing LDL circulation. New evidence indicates that PCSK9 is also highly expressed in arterial walls such as endothelium, with a local effect that can regulate vascular homeostasis and atherosclerosis. Consequently, PCSK9 has pro-atherosclerotic effects. In addition to the synthetic and pro-atherosclerotic effects of lipoproteins, PCSK9 is also involved in glucose metabolism and obesity and the regulation of sodium reabsorption in the kidney, which is relevant in hypertension.

ANGPTL3

Angiopoietin 3, also known as ANGPTL3, is a protein that is codified in humans by the ANGPTL3 gene. ANGPTL3 prevents hydrolysis and clearance of Triglicerides. The interaction of ANGPTL3 with LPL-1 depends on the formation of a complex with ANGPTL8. Humans with loss of function mutations in ANGPTL3 clinically present hypolipidemia.
This phenotype includes a reduction in total plasma cholesterol, TG and LDL-C, a reduction in coronary plaque load and a lower risk of ASCVD. The reduction in lifetime risk of CVD among patients with ANGPTL3 variants with loss of function has been estimated at 41%16.[7-8;11-15]

LPA

The LPA gene encodes apo (a), an inactive serine proteinase with procoagulant activity, through inhibition of tissue-type plasminogen activator. In circulation, apo (a) is present as part of lipoprotein (a) (Lp[a]), a macromolecular structure with similarity to LDL that includes a lipid cargo and an apo (a) molecule covalently bound to apoB.
The apo (a) has a structural homology remarkable with plasminogen for which it has an important role in the process of atherosclerosis and thrombogenesis. Lp (a) has a pro-atherogenic effect, intervening in the development of atherosclerosis and a pro-thrombotic effect, interfering with the fibrinolytic process as a competitor of the plasminogen.

APO III

Apolipoprotein C-III also known as apo-CIII is a protein that in humans is encoded by the APOC3 gene. Apo-CIII is secreted by the liver as well as the small intestine, and is found on triglyceride-rich lipoproteins such as chylomicrons, very low density lipoprotein (VLDL), and remnant cholesterol. APOC3 inhibits lipoprotein lipase and liver lipase and is thought to inhibit hepatic absorption of triglyceride-rich particles. An increase in apoC-III levels induces the development of hypertriglyceridemia. Recent evidence suggests an intracellular role for Apo-CIII in promoting the assembly and secretion of triglyceride-rich VLDL particles from liver cells under lipid-rich conditions. In vitro studies have documented that the ApoC-III activates the monocytes by increasing the expression of β1-integrins, molecules that play an important role in promoting the adhesion of monocytes to endothelium and therefore their migration in the tissues.
Furthermore apoC-III induces the expression of adhesion molecules on the endothelium, which facilitate the recruitment of leukocytes and inflammatory cells.[9;10;16]

NEW GENIC AGENTS THERAPEUTIC

Antisense oligonucleotide

ASOs are single-stranded nucleic acid molecules (mainly DNA or modified DNA) that bind to cellular mRNAs and induces their degradation or prevents their translation. In this way ASO can reduce the synthesis of a target protein. ASOs generally consist of a single strand of 12-20 long nucleotides antisense and complementary to the target mRNA tract. Several chemical modifications can be inserted in the ASO nucleotide tract, to increase its stability or increase absorption cellular, or to reduce the possibility of attack by extracellular nuclease enzymes. Once inside the cell, ASO binds specifically to high selectivity target mRNA regions.
Therapies based on the ASO technique accumulated in the hepatocytes, where the greatest amount of APO is synthesized, bind the mRNA target and are metabolized by a path completely independent of the cytochrome CYP450, this makes the therapy advantageous since the probability of liver damage or drug interactions in polytherapy patients is almost zero. The latest generation ASOs show greater resistance to attack by extracellular degradation enzymes and greater affinity with the target mRNA.[17-21]

Mipomersen

Mipomersen is a second generation nucleotide ASO 20, directed against mRNA for apoB. In clinical trials Mipomersen demonstrated modest reduction in LDL-C of 21% with a concomitant reduction of 24% in apoB. However, the main problem encountered was the enormous inter-individual variability in response to treatment, with very large response changes. The most common adverse

effects of Mipomersen reported are injection site reactions, flu-like symptoms and clinically relevant increases in liver enzymes. Most studies have also documented significant increases in fat content in the liver (in line with the mechanism of the drug inhibition of the production and secretion of lipoproteins by the liver.) A study of liver biopsies from chronically patients treated with Mipomersen showed severe hepatic steatosis in most patients, although without histological signs of nonalcoholic steatohepatitis. CVD events occurred in 9.6% of patients during the 24 months following initiation of treatment with Mipomersen, compared to 61.5% in the 24 months preceding treatment with mipomersen. Therefore, the use of Mipomersen has been associated with a 95% lower probability of CVD events.[22-29]

Volanesorsen

Volanesorsen (or ISIS 304801) is a second generation ASO complementary to the APOC3 mRNA. This ASO blocks the production and hepatic secretion of apoC-III, improving the removal of circulating atherogenic lipoproteins. The selective binding of Volanesorsen with ribonucleic acid messenger (mRNA) of apoC-III within the 3' untranslated region at base position 489-508, causes the degradation of mRNA.
This binding selected the translation of the apoCIII protein, thus removing a triglyceride clearance inhibitor and checking the metabolism through an LPL-independent route. Pre-registration studies have shown dose-dependent reductions in apoC-III plasma, which have resulted in a 31.3-70.9% drop in fasting TG plasma. The huge advantage identified with Volanesorsen is that the response to treatment demonstrates response linearity between subjects, in contrast to the great variability of subject dependent response found with other ASOs. In APPROACH, the Phase 3 Volanesorsen clinical trial reduced triglycerides, total cholesterol, non-HDL cholesterol, apoC-III, apoB-48.

ISIS-APO(a)Rx

It is a specific ASO that specifically binds a splice site between exons 24 and 25 of the apo(a) mRNA. The agent was selected from a screen of over 2200 ASO complementary to multiple sites across the human apo(a) transcript. The ISIS-APO(a)Rx Phase 2 study (n=65) enrolled Cohort A (Lp(a) ≥125 nmol/L, ≥˜50 mg/dL) and Cohort B (≥437 nmol/L, ≥˜175 mg/dL), representing the 80th and >99th percentile of Lp(a). They were dosed subcutaneously (sc) 1X/wk with 100, 200 and 300 mg for 4 wks at each dose or placebo. In the ISIS-APO(a)Rx study, baseline Lp(a) levels in Cohorts A and B were 255 nmol/L and 488 nmol/L, respectively. In the combined cohorts, Lp(a) was reduced by mean 72% (up to 94%) with ISIS-APO(a)Rx vs mean reduction of 3% in placebo ($p<0.001$ vs placebo), an effect independent of baseline Lp(a) levels. The mean LDL-C level was reduced by 13% in cohort A and 23% in cohort B. 37

IONIS-ANGPTL3-LRx

Is a second-generation, N-acetyl galactosamine-conjugated ASO against ANGPTL3. The ISIS-APO(a)-LRx Phase 1 study enrolled volunteers (n=40) with elevated Lp(a) ≥75 nmol/L, ≥˜30 mg/dL ULN). They were given sc single doses of 10, 20, 40, or 80 mg or multiple doses (6 doses over 22 days) of 10 or 20 mg. In the single dose groups ISIS-APO(a)-LRx resulted in dose dependent reductions, with mean reduction of 79% (up to 97%) in the 80 mg group. In the multiple dose groups, baseline Lp(a) levels were 143 nmol/L in the 10 mg group and 165 nmol/L in the 20 mg multiple dose group, and Lp(a) was reduced by mean of 66% and 80% (up to 99%), ($p=0.004$ vs. placebo), respectively. 15/16 (94%) subjects reached levels <75 nmol/L 37

SMALL, INTERFERING RNAS (siRNAs)

In recent years, one of the most significant advances in therapeutic research has been the discovery of small (20-30 nucleotides) non-coding RNAs that regulate genes and genomes. This regulation can occur at some of the most important levels of genomic function, including RNA processing, chromatin structure, RNA stability, chromosomal segregation, transcription and translation In general, three main categories have been identified: RNA interacting with piwi (piRNA), RNA with short interferences (siRNA), and microRNA (miRNA). The siRNAs can be artificially introduced from the outside through specific transfection methods, to induce silencing of specific genes. Theoretically, any gene whose sequence is known can be chosen as the target of an siRNA. This made siRNA an important tool for gene function studies and new drug development. siRNAs are involved primarily in the pathway of RNA interference, which leads to the interference of the expression of specific genes with complementary nucleotide sequences, degrading the mRNA after transcription, so as not to allow translation to take place. The siRNAs have a well-defined structure, which consists of a short double-stranded RNA (RNAds), usually composed of 21 nucleotides, with two nucleotides protruding at each of the two ends. This structure is the result of the processing of the Dicer enzyme, which converts long RNAds or shRNA molecules (RNA molecules that form a hairpin) into siRNA. Transfection of an exogenous siRNA can be problematic, since gene silencing is only transient, especially in fast-growing cells. One way to overcome this problem is to introduce a vector (such as a plasmid) into the target cell capable of expressing the desired siRNA molecule. Since pathological processes in hypercholesterolemia usually depend on the unregulated expression of several genes, siRNA administration is expected to be able to turn off this expression (via RNAi).[5-6]

Inclisiran

Inclisiran is a synthetic siRNA directed against PCSK9 mRNA, conjugated with N-acetylgalactosamine (GalNAc) that bind the liver express asialoglycoprotein receptors with high affinity. This wording brings efficient and targeted uptake of inclisiran by hepatocytes. Because the GalNAc platform allows precise targeting of the drug for the liver organ of interest, all to the advantage of greater tissue selectivity and a reduction in the probability of adverse events. Clinical studies have shown a 51% reduction in LDL-C from baseline to ~ 22 months, with minimal side effects. In addition, studies have been carried out to evaluate pharmacokinetics, pharmacodynamics, and the safety of Inclisiran in subjects with hepatic impairment and with mild, moderate and severe kidney failure. Study showed no difference in LDL-C reductions, PCSK9 reductions, and safety/tolerability between subjects. Though siRNAs and PCSK9 mAbs both ultimately lower plasma LDL-C concentrations, there are several differences that should be noted. First, siRNAs have a substantially different pharmacodynamic profile to the mAbs, leading to an extended duration of action with sustained lowering of LDL-C over time. Treatment with siRNAs can lead to reductions in PCSK9 and LDL C, possibly enabling a twice-yearly dosing regimen, with little variation over the 6-month period after receipt of the first injection. Thus, Inclisiran has the potential to provide effective LDL-C lowering with administration every 6 months compared with once or twice a month with the mAbs, which may facilitate adherence to therapy. Second, whereas the mAbs bind to extracellular PCSK9 and prevent binding with the LDLR, siRNAs are targeted specifically to the liver and inhibit hepatic synthesis of PCSK9. Theoretically, this could lead to higher atheroma concentrations of PCSK9 in those treated with the siRNAs versus the mAbs, though the clinical implications of this are unclear.

Third, siRNA-induced lowering of PCSK9 reflects true levels of reduced protein, as opposed to mAbs, which binds to PCSK9 protein and interferes with its ability to bind with the LDLR, thus increasing its concentration in the plasma. Clinically, elevations in plasma PCSK9 levels can be used to confirm adherence to therapy or optimal injection technique in patients who do not show the

expected LDL-C-lowering response to PCSK9i therapy, compared to decreases in plasma PCSK9 levels which would be expected in patients using siRNA therapy. The next chapter of this has been the entry of a new approach to antagonize PCSK9, namely silencing its production. Inclisiran, an siRNA complementary to PCSK9 mRNA, demonstrates sustained reductions in PCSK9 and LDL-C. Though Inclisiran acts on the same pathway as the therapeutic mAbs, siRNA exhibits a more sustained duration of action requiring less frequent injections, which serve to overcome some of the known barriers to adherence. It is conceivable that patients with hypercholesterolemia and/or ASCVD treated with this therapy may only require one to two injections per year. We appear to be at the dawn of a new anti-PCSK9 therapy. Furthermore, Inclisiran appears to have a relatively benign side effect profile, as shown in the ORION-1 trial. There were only rare symptoms of immune activation, such as flu-like symptoms, which is often a concern with RNA-targeting therapies. In addition, Inclisiran did not adversely affect platelet levels, in contrast to other recent reports from studies of antisense oligonucleotides and other siRNA molecules. However, the ongoing ORION-11 trial is expected to better define the long-term side effect profile of Inclisiran.
Currently there are four phase 3 clinical trials running. The ORION-4 is assessing the effects of Inclisiran on clinical outcomes among patients with atherosclerotic cardiovascular disease, the ORION-9 is enrolling patients with HeFH and elevated LDL-C to evaluate the efficacy, safety, and tolerability of subcutaneous Inclisiran., the ORION-10 is enrolling participants with atherosclerotic cardiovascular disease and elevated low-density lipoprotein cholesterol, and the ORION-11 includes subjects with atherosclerotic cardiovascular disease (ASCVD) or ACSVD risk-equivalents and elevated low-density lipoprotein cholesterol (ClinicalTrials.gov: NCT03400800).
Not with standing, it is not yet ascertained that the marked LDL-C reductions attained with Inclisiran would definitely translate into a reduction in CVD risk, and thus, large outcome trials would need to be conducted in the future for that reason.
Other potential future therapeutic strategies targeting PCSK9, which are currently in the initial stages of development, include small molecule inhibitors that disrupt the processing of PCSK9, the use of adnectins, which block the binding of PCSK9 to the LDL receptor, as well as the AT04A vaccine, which is currently being tested in a phase 1 clinical trial and the future is bright. [30-36]

CRISPR/CRISPR associated 9 (cas9)-based investigational therapies

The CRISPR-Cas9 (clustered regularly interspaced short palindromic repeats-associated protein 9) system, is a part of the adaptive immune system present in archaea and bacteria for defending against invasive nucleic acids from phages and plasmids. This system uses a single guide RNA (sgRNA) that recognizes its target sequence in the genome, and the Cas9 nuclease acts as a pair of scissors to cleave the double strands of DNA.
Short fragments of the foreign DNA, also named protospacers, are integrated as new spacers into the CRISPR array. The search of protospacers from the foreign DNA is based on the protospacer adjacent motif (PAM).
Subsequent the CRISPR array is transcribed to pre-CRISPR RNA (pre-crRNA) and this precursor-CRISPR transcripts through endonucleolytic cleavage yield short mature CRISPR RNAs (crRNAs) Each crRNA contains a conserved repeat sequence and a transcribed spacer, which is complementary to the foreign DNA. Using single-guide RNA (sgRNA) libraries can target multiple gene elements because each crRNA corresponds to an foreign sequence. Therefore this system can be leveraged to identify drug-target or disease-resistance genes, such as novel tumor suppressors or oncogenes, and to quickly assess drug targets. The CRISPR–Cas9-mediated genome engineering holds immense promise to treat or even cure genetic disorders, including many forms of cancer and neurodegeneration, as well as sickle cell anemia, immunological disorders and cardiovascular diseases.[38-44]

SAFETY

The safety profile of a medicinal product has always enormous importance throughout the lyfecicle, from pre-registration studies to the post marketig phase with repercussions also on regulatory aspects. Several cases of induced thrombocytopenia have been observed during ASO treatment in both preclinical animal studies and pre-registration human clinical studies. And in particular, two forms of thrombocytopenia have been observed. The most common form is mild, transient and dose-dependent thrombocytopenia, which is typically observed at high doses in the animal model. In humans, thrombocytopenia has been reported in studies on the use of ASO in neoplastic diseases, and in particular with a series of first generation ASO and rarely with second generation ASO (such as Mipomersen). In general, the reduction in the number of platelet counts is mild, reversible, dose-dependent and of a non-serious type such as to cause bleeding episodes. The second form of thrombocytopenia found is more severe, and has been observed in preclinical studies after repeated exposures. In animal studies, there have been cases of multiple bleeding and lethargy that required stopping. The appearance of severe thrombocytopenia, although rare, has raised serious concerns about this adverse event with ASO treatment. To date, no drug-induced severe thrombocytopenia has been reported preclinically or clinically with siRNA drugs. But it is important to note that ASO drugs have a longer history. Therefore, we recognize that there is insufficient information to draw definitive conclusions about the overall strength, weakness or superiority of one platform over the other in relation to safety or efficacy in humans. Cases of neuropathy have also been registered with the use of siRNA. Despite intense interest and investigations, the mechanisms underlying ASO-induced thrombocytopenia or siRNA-induced peripheral neuropathy are not yet understood. Mechanistic knowledge of ASO thrombocytopenia could increase thanks to post marketing safety data. Mipomersen therapy is more likely discontinued and associated with increased risk of injection-site reactions, hepatic steatosis, hepatic enzyme elevation, and flu-like symptoms. On safety, Inclisiran considering data of ORION-11 study was numerically superior on several measures compared to placebo. Serious treatment-emergent adverse events occurred in 22.5% of control-arm subjects and 22.3% of patients receiving study drug. Rates of death (1.9% versus 1.7%) and malignancies (2.5% versus 2%) also were higher among those who received placebo. Liver and renal function tests favored Inclisiran over placebo as well. The treatment with Volanesorsen in the pivotal Phase 3 study in patients with FCS (the APPROACH study), confirmed reductions in platelet counts to below normal (140 x 109/L) were observed in 75% of FCS patients treated with volanesorsen and 24% of placebo patients; confirmed reductions to below 100 x 109/L were observed in 47% of patients treated with Volanesorsen compared with no placebo patients. In APPROACH and its open-label extension (CS7), patients discontinuing therapy due to platelet levels included 3 patients with platelet counts <25 x 109/L, 2 with platelet counts between 25 x 109/L and 50 x 109/L, and 5 with platelet counts between 50 x 109/L and 75 x 109/L. None of these patients had any major bleeding events and all recovered to normal platelet count following drug discontinuation and administration of glucocorticosteroids where medically indicated. In the Phase 3 clinical studies (CS16 and APPROACH), 16% and 30% of Volanesorsen-treated patients tested positive for anti-drug antibodies during 6-month and 12-month treatment, respectively.

No evidence of altered safety profile or clinical response was associated with presence of anti-drug antibodies; however this is based on the limited long-term data injection site reactions defined as any local cutaneous reaction at the injection site persisting more than 2 days occurred in 82% of Volanesorsen-treated patients in the APPROACH study and its openlabel extension (CS7). These local reactions were mostly mild and typically consisted of 1 or more of the following: erythema, pain, pruritus, or local swelling. Injection site reactions did not occur with all injections and

resulted in discontinuation for 1 patient in the APPROACH study. No injection site reactions, flu-like symptoms or laboratory abnormalities were observed with ISIS-APO(a)-Lrx. Or ISIS-APO(a)-Rx. During phase 1 and 2 study respectively in conclusion there is not enough information and data to draw a general safety profile of the ASO and siRNA. additional data from post marketing studies and their use in clinical practice may be of greater help. however, the problem of severe thrombocytopenia for ASOs and a recent relationship of peripheral neuropathy with a siRNA seems to have been a class adverse reaction, although the mechanistic bases to date are still poorly understood.[45-47]

CONCLUSION

The gene silencing approach with ASO or with siRNA established a decisive step towards the development of a personalized therapy also in the field of dyslipidemia. The development of this new class of therapies led to development of molecules with an excellent pharmacokinetics profile which is expected to reduce the frequency of administration and improve patient compliance, and an excellent efficacy and safety profile, even if post marketing data are still lacking.
Several trials have shown the effectiveness of these new therapies in reducing circulating lipid or lipoprotein levels however it is need to wait for the results of the studies long-term follow-up to understand if these new therapies reduce the risk of cardiovascular diseases. In recent years, the pharmacological class of choice in the treatment of hyperlipidemia has been that of statins, even if, with an excellent efficacy profile, a large number of patients are intolerant to this therapy for various causes (myopathies, liver disease, etc.), the introduction of drugs aimed at inhibiting the PCSK9 protein (alirocumab, ecolocumab, bococizumab) has given a very valid new therapeutic alternative to these patients, however the introduction of new therapeutic options such as new gene agents can provide important therapeutic alternatives, even if for the latter expect post marketing security data. With the birth of PCSK9 inhibitory antibodies and the introduction of new gene therapeutic agents, the beginning of a new post-statin era can be define.[48]

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