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Inclisiran in lipid management: A Literature overview and future perspectives

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ABSTRACT

Primary and secondary prevention protocols aim at reducing the plasma levels of lipids - with particular reference to low-density lipoprotein cholesterol (LDL-C) plasma concentrations – in order to improve the overall survival and reduce the occurrence of major adverse cardiovascular events. The use of statins has been widely considered as the first-line approach in lipids management as they can dramatically impact on the cardiovascular risk profile of individuals. The introduction of ezetimibe and proprotein convertase subtilisin-kexin type 9 (PCSK9) inhibitors overcame the adverse effects of statins and ameliorate the achievement of the target lipids levels. Indeed, advances in therapies promote the use of specific molecules – i.e. short strands of RNA named small-interfering RNAs (siRNAs) – to suppress the transcription of genes related to lipids metabolism. Recently, the inclisiran has been developed: this is a siRNA able to block the mRNA of the *PCSK9* gene. About 50% reduction in low-density lipoprotein cholesterol levels have been observed in randomized controlled trials with inclisiran. The aim of this review was to summarize the literature regarding inclisiran and its possible role in the general management of patients with lipid disorders and/or in primary/secondary prevention protocols.

1. Introduction

The management of lipid levels – with particular reference to low-density lipoprotein cholesterol (LDL-C) plasma concentrations – in primary and secondary prevention follows the principle "the lower, the better" [1,2].

LDL-C plays a central role in the pathogenesis of atherosclerotic disease, since it enhances vascular alterations and endothelial dysfunction [3].

The Framingham study outlined a 12/13-fold increase in the occurrence of acute myocardial infarction events in patients with higher total cholesterol levels (>260 mg/dL) and lower high-density lipoprotein cholesterol (HDL-C) levels (<40 mg/dL) [4]. With regard to LDL-C, higher plasma concentrations have been associated with a 50–70% increase in the development of atherosclerotic plaques in patients with or without overt cardiovascular risk factors [5,6].

Therefore, the need for a tight monitoring of lipid plasma levels in the general population is the cornerstone for an improvement of the overall cardiovascular risk profile of individuals.

Antilipemic agents promote the best therapeutic approach for the management of lipid disorders, since a 20 mg/dL decrease in cholesterol levels prevents 2–3 coronary heart disease (CHD) mortal events every 1000 [7].

Statins have been widely accepted as the first-line treatment in lipid disorders and in the primary/secondary prevention of cardiovascular diseases [8–16]. The JUPITER (*Justification for the Use of Statins in Prevention: an Intervention Trial Evaluating Rosuvastatin*) trial highlighted the need to administer statins to maximally reduce the LDL-C levels in order

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to prevent all-cause mortality and/or major adverse cardiac events (MACE) in primary prevention [8,9].

Secondary prevention trials outlined the role of statins in the prevention of further cardiovascular events in patients who reached the predetermined LDL-C target levels [13–16].

However, the REALITY (*Return on Expenditure Achieved for Lipid Therapy*) study pointed out that 60% of European patients on active lipid-lowering therapies did not reach the pre-specified lipid plasma concentrations goal [17]. Similar results were produced by a recent Danish study, which enrolled 3040 patients with previous myocardial infarction: only 43.4% and 47.7% of these subjects achieved the LDL-C target values at the 6- and 12-month follow-up, respectively, despite an optimized lipid-lowering therapy at the maximum tolerated dose [18].

The DA VINCI study [19] revealed a low number of patients on primary or secondary prevention programs who were treated with a combination therapy with ezetimibe (only 9%) and/or proprotein convertase subtilisin-kexin type 9 (PCSK9) inhibitors (1%), thus accounting for the great gaps in the daily application of clinical guidelines on the management of lipid disorders.

Inclisiran is a new pharmacological compound recently introduced as antilipemic agent for reducing the LDL-C levels. The aim of this review is to summarize the literature regarding inclisiran and its possible role in the general management of patients with lipid disorders and/or in primary/secondary prevention protocols.

1.1. Search strategy

We performed an extensive literature review, searching the main international electronic databases – PubMed/Medline, PubMed CENTRAL, Scopus, and ClinicalTrials.gov – until September 1, 2021. We used the term "inclisiran", "ALN-60212″, "ALN-PCSsc", "PCSK9si", "ALN-PCSSC", "ALN-60212″, "ORION", "VICTORION", and "siRNA" in order to identify and evaluate all the publications dealing with the molecule and describing the biochemical structure, pharmacokinetics, pharmacodynamics, and preclinical/clinical results from international randomized controlled trials (RCTs) and/or observational studies.

2. Proprotein convertase subtilisin-kexin type 9 in lipid disorders: genetic and protein expression

Advances in the comprehension of lipid disorders and the progress of pharmaceutical technology for new drug delivery led to the identification of PCSK9 as a further target for counteracting the increase in plasma concentration of lipids.

PCSK9 is a member of a specific family of human enzymes which – acting as proteases – are able to activate secretory proteins through their proteolytic action, i.e. the proprotein convertase (PC) family [20].

Eight PCs had been previously identified: PC1, PC2, furin, PC4, PC5, paired basic amino acid cleaving enzyme 4 (PACE4), PC7, and subtilisin kexin isozyme 1 (SKI-1), which act at the Golgi level to process proteins into granules [21–23]. In 2003, *Sedha* et al. identified the ninth component of the family, i.e. the *PCSK9* gene, which was located on chromosome 1 (1p32.3) [24].

PCSK9 is a 22-kb gene composed by 12 exons that is subjected to a high number of polymorphisms, which promote gain-/loss-of-function and alterations in the expression of the related protein [25–27].

Such mutations are responsible for severe syndromes. In particular 2.3% of the cases of autosomal dominant familial hypercholesterolemia (FH) are mediated by mutations in the *PCSK9* gene [27]. Specifically, it was shown that mutations leading to loss-of-function were related to hypocholesterolemic syndromes, while gain-of-function mutations resulted in FH [27].

PCSK9 is mainly expressed in the liver, gut, and kidney [28]. Genome-wide association studies revealed different determinants able to affect the transcription of PCSK9 [29]. Statin therapy may increase the transcription of PCSK9: the increase in sterol regulatory element-binding protein 2 (SREBP-2) and the decrease in plasma concentrations of cholesterol are the main conditions able to up-regulate the nuclear transcription of the *PCSK9* gene [29–32]. The decrease in intracellular cholesterol accumulation can improve the translocation of the SREBP2 into the nucleus of the liver cells, where the protein binds the regulatory structure for the final transcription of the mRNA of the *PCSK9* gene [32].

Indeed, the adiponectin/adiponectin receptor (AdipoR) pathway is a further determinant of the transcription process of the *PCSK9* gene. *Sun* et al. [33] found that the increase in adiponectin and the activation of its receptor reduce the expression of *PCSK9*. Beyond the SREBP-2, the activation of the peroxisome proliferator-activated receptor-γ (PPAR-γ) by means of the liver AdipoR-2 seems to promote the transcription of the mRNA of the *PCSK9* gene [33].

A recent research from *Hu* et al. [34] shed light on possible new insights in the regulation of the transcription of the mRNA of the *PCSK9* gene. The carbohydrate-responsive element-binding protein (ChREBP) can modulate the glucose and triglyceride metabolism, thus exerting a crucial role in metabolic diseases. Mutations able to promote loss-of-function of the ChREBP are related to an improvement in the overall cardiovascular risk profile of individuals [35]. The increase in intracellular glucose/metabolites levels can activate ChREBP and induce the translocation of this protein to the nucleus. ChREBP is now able to interact with the putative E-box ChREBP-binding site (ChoRE-like sequence) at the promoter region of the human *PCSK9* gene, which induces the transcription of the mRNA of the *PCSK9* gene itself [35].

The mRNA induces the production of the PCSK9 protein from the endoplasmic reticulum (ER). A 74-kDa precursor (proPCSK9) is the early product: its autocatalytic activity promotes the cleavage of the final PCSK9 protein, which is still inhibited by a pro-segment [28]. The complex PCSK9/inhibitory segment is packaged into vesicles, which go through the *cis*-Golgi, and are then excreted from the cell. Outside the cell, the PCSK9 binds to the low-density lipoprotein receptor (LDLR) complex: this bond leads the entire complex to be included into lysosomes for the final degradation of the LDLR [28].

Therefore, the modulation of the transcription of the *PCSK9* gene can effectively interfere with the LDLR metabolism, thus providing possible solutions for an increase in the re-utilization of the LDLR and the reduction of LDL-C in the bloodstream. Recently, the introduction of monoclonal antibodies able to capture the extracellular PCSK9 protein and to avoid its internalization with LDLR into lysosomes produced a revolutionary approach to the specific targeting of lipid metabolism [36]. Indeed, further advances in pharmacological treatments have overcome this strategy in lipids management [37]. Inclisiran is the molecule representing this new approach.

3. Inclisiran: pharmacodynamics and pharmacokinetics

Inclisiran is a new drug able to reduce the circulating plasma levels of the PCSK9 protein by interfering with the transcription of its related gene [38].

The pharmacodynamics of the drug is related to the RNA interference (RNAi) regulatory process, i.e. the use of short strands of RNA – defined as small-interfering RNAs (siRNAs) – which can suppress the transcription of genes [39].

The rationale of this action is a post-transcriptional regulatory process able to prevent the definite creation of the protein. Specifically, siRNAs are two-strand sequences with 21–23 bases which overhang each other: one is responsible for the identification of the target-gene, the other is the passenger, which allow the interaction with the RNA-induced silencing complex (RISC) [40]. After entering the cytoplasm, the passenger is degraded, while the guide interacts with the RISC: the siRNA/RISC complex links to the corresponding messenger-RNA (mRNA) of the target-protein by means of the complementary guide strand [38,40]. Once connected, a specific protein component of the RISC complex – the Argonaute-2 (Ago2) protein – is activated, and

promotes the cleavage of the target mRNA, thus inhibiting the final translation of the mRNA into the definite protein [38,40].

In order to allow the delivery of the siRNA to the corresponding regions – thus avoiding their activation in cells other than the target ones – dedicated carriers or modifications of the siRNAs should be managed [38]. The bio-conjugation process consists in linking the siRNA to specific molecules that can promote the direct interaction between the siRNA and the target cells [38]. The addition of the N-acetylgalactosamine (GalNAc) sugar triggers a bio-conjugation process which allows the siRNA to interact with the asialoglycoprotein receptor (ASGPR), i.e. a receptor highly expressed on the liver cells, which promotes the entry of the siRNA into the latter [38].

These biochemical and pharmacological principles are summarized within the inclisiran molecule. Inclisiran is a double-strand siRNA which is properly conjugated with the triantennary GalNAc in order to reach the liver cells and introduce itself into their cytoplasm (Fig. 1) [41]. The sense strand is formed by 21 bases, while the antisense strand is composed by 23 bases, one overhanging the other.

The antisense strand is formed by nucleotides whose sequence is modeled to specifically correspond to the human Proprotein Convertase Subtilisin/Kexin Type 9 (PCSK9) mRNA.

Since the siRNAs usually undergo a fast degradation in the plasma, due to the action of exonucleases and/or endonuclease, further biochemical modifications should be considered in order to improve the stability of the compound [42]. The inclusion of 2′-fluoro and 2′-O-methyl modifications, as well as the addition of four phosphorothioates to the inclisiran are responsible for reducing the degradation of the molecule in the bloodstream [42].

Inclisiran mostly travels into the bloodstream as linked to proteins (87%). It showed a higher specificity to liver cells, while no significant interactions were demonstrated with cytochrome P450.

The triantennary GalNAc is linked at the 3′ end of the sense strand and guides the inclusion of the molecule into the liver cells via ASGPR (Fig. 1) [43].

ASGPR, also called the “Ashwell-Morell Receptor”, is a receptor usually present on the baso-lateral side of the membrane of the hepatocytes. Its function is still not clear [44,45]. Although peritoneal macrophages, rat and human testis, human-sperm, human intestinal epithelial cells and peripheral blood monocytes may show the presence of this receptor on their membrane, the liver accounts for the highest quantity of this receptor [44–46].

ASGPR is a transmembrane receptor which is formed by two subunits (46 and 50 kDa, respectively). The outer domain of the receptor showed a carboxylic group (COOH) which is able to interact with GalNAc using calcium (Ca^{2+}) binding domains [45,46].

The GalNAc residues of inclisiran interact with the outer part of the ASGPR via the Ca^{2+} binding domains: the connection between the molecule and the receptor leads to the modification of the structure of the membrane of the hepatocyte. The lipid bilayer of the membrane includes ASGPR/inclisiran into a clathrin-coated vesicle which undergoes endocytosis. Heat shock proteins release clathrin, which can turn towards the membrane, while the remaining vesicles promote the creation of endosomes [44–47]. Then, inclisiran is released into the cytoplasm of the liver cell, while ASGPR is re-directed towards the cell membrane, in order to be re-used.

The antisense strand of inclisiran interacts with the mRNA of PCSK9: the activation of the RISC promotes the breakdown of the nucleotide sequence of the mRNA of PCSK9, thus avoiding the translation of the RNA sequence of the gene in the Golgi apparatus for the creation of the final products, i.e. PCSK9 (Fig. 2) [43]. Therefore, the lack of production of PCSK9 leads to the reduced degradation of the receptor of the low-density lipoproteins (LDLs), thus favoring the increase in the uptake of LDL-cholesterol (LDL-C) by the liver cell (Fig. 2) [48].

After completing the action, the strands of inclisiran undergo

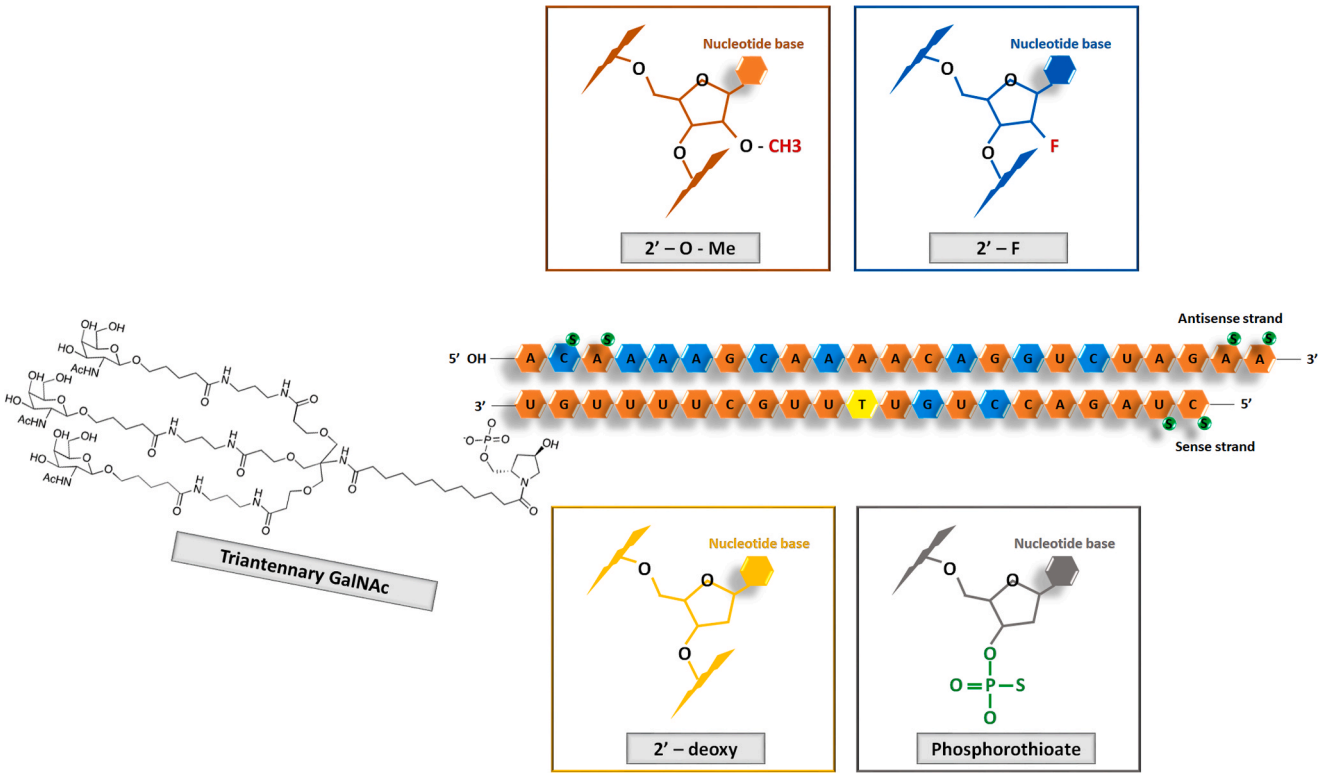

Fig. 1. Simplified biochemical representation of inclisiran. Inclisiran is a small interfering RNA (siRNA) composed of two strands. The antisense strand is composed of 23 nucleotide bases, the sense strand of 21; the former overhangs the latter. The 3′ end of the sense strand is linked to triantennary N-acetylgalactosamine (GalNAc) which allows the molecule to enter the hepatocytes by interacting with the asialoglycoprotein. Abbreviations: A: adenosine; C: cytidine; G: guanosine; GalNAc: N-acetylgalactosamine; Me: Metil; O: oxygen; S: phosphorothioate; U: uridine; T: thymidine.

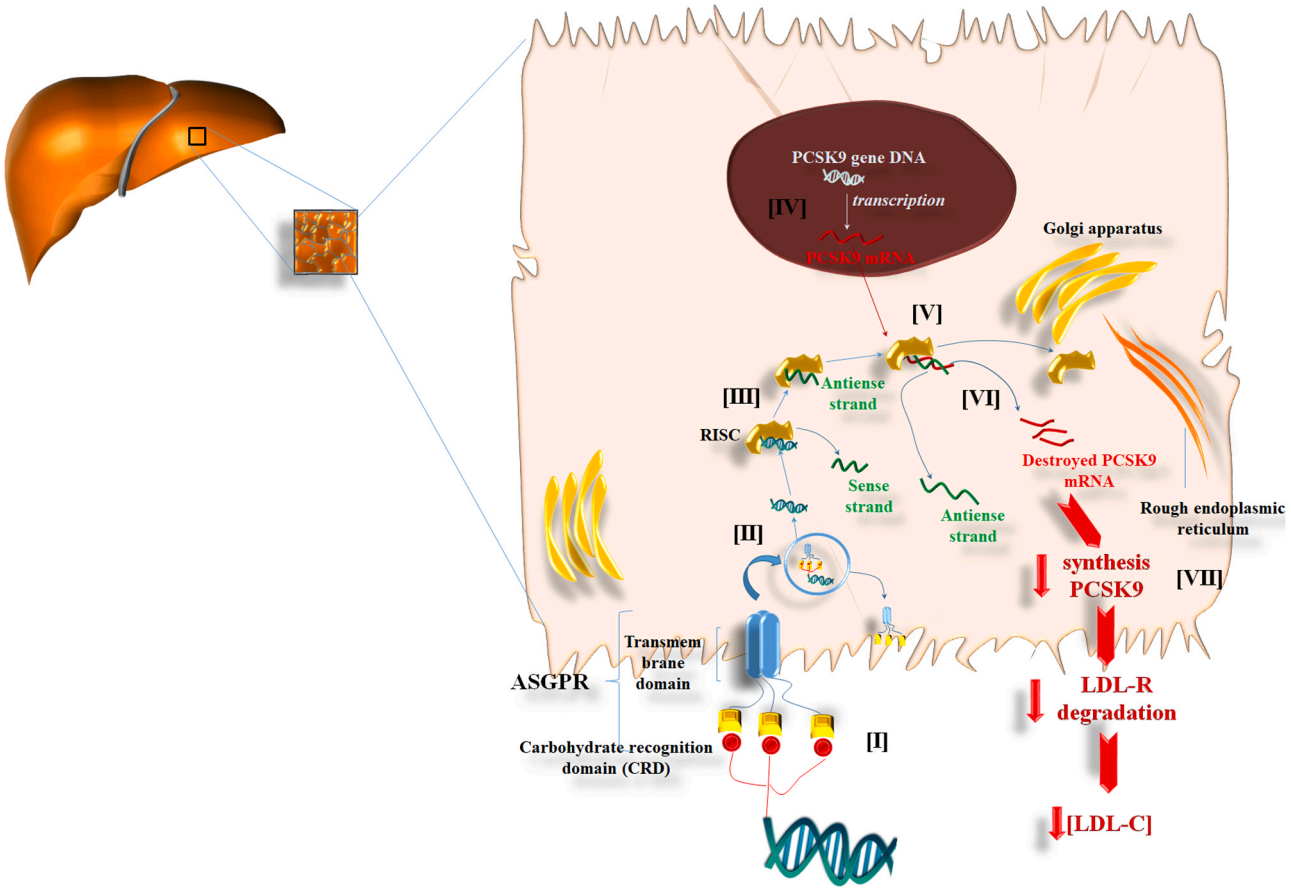

Fig. 2. Schematic representation of the mechanism of action of inclisiran. The small interfering RNA (siRNA) called inclisiran is a double strand RNA with tri-antennary N-acetylgalactosamine (GalNAc) at its 3′ end of the sense strand. The mechanism of action is represented by: [1] the interaction between the GalNAc and the asialoglycoprotein receptor (ASGPR) at the baso-lateral side of the membrane of the hepatocytes; [II] the inclusion of the ASGPR/inclisiran complex into endosome, where the double strand of inclisiran is released into the cytoplasm of the liver cell; [III] the free double strand of the inclisiran is caught by the RNA-induced silencing complex (RISC): the Argonaute-2 protein released the sense strand; [IV] the messenger-RNA (mRNA) of the proprotein convertase subtilisin-kexin type 9 (PCSK9) enters the cytoplasm, where it interacts with the corresponding antisense strand of inclisiran [V]; [VI] the RISC destroys the mRNA of the PCSK9, thus reducing the translation of the corresponding protein and, therefore, the degradation of the receptor of the low-density lipoproteins (LDL-R) and, consequently, the concentrations of LDL-cholesterol (LDL-C) in the bloodstream.

degradation. Data from rats and monkeys revealed that the sense strands are mostly eliminated unaltered or, at least, deprived of sugars, while the antisense strand can be removed as it stands or as a strand with deletion of one nucleotide at 5′ or 3′-ends [43]. Most of the antisense strands of inclisiran are eliminated by the liver (82.5%), the rest from the kidneys (Table 1). Studies on rats and monkeys revealed that about 29% and 32% of inclisiran, respectively, are primarily excreted from the kidneys over a timeframe of 7-days, while 23% and 1.6% through the faeces [43]. The final half-life elimination time of the drugs is 9 h, while no significant accumulation can be detected in the organism, even after the administration of subcutaneous multiple doses [43]. No interactions were observed between inclisiran and cytochrome P450 (CytP450), which accounts for the relatively poor interactions with other drugs metabolized by CytP450 [43]. The long-term action of the inclisiran is mostly due to the activation of the RISC complex: despite the 9 h mean elimination rate, inclisiran promotes a long-term reduction of the PCSK9 mRNA synthesis, due to its action on the RISC complex (Table 1) [43]. Therefore, the fast rate of elimination from the plasma and the long action via RISC complex activation may account for the higher impact of the drug on the lipid metabolism and the lower rate of adverse events on liver, muscles and kidneys.

4. Preclinical and clinical studies

The application of siRNAs in the management of lipid levels was firstly considered by *Frank-Kamenetsky* et al. [49] who included a siRNA in lipidoid nanoparticles (LNPs) in order to improve the liver-specific targeting of the compound. LNP is a formulation composed by a cation component, cholesterol, and poly-(ethylene glycol)-lipid, which improve the addressing of the siRNA to the specific target [49]. The LNP-PCS-A2 compound by *Frank-Kamenetsky* et al. [49] was able to reduce the transcription of the *PCSK9* gene by 60–70% in mice, when administered at the dose of 5 mg/kg, while the administration of the same compound in rats at a dose of 1–5 mg/kg was able to reduce the transcription of PCSK9 mRNA by 50–60% and, in parallel, promote a 50–60% decrease in total cholesterol plasma levels [49]. These effects persisted for three weeks and were observed also in case of concomitant administration of HMG-CoA reductase inhibitors.

Fitzgerald et al. [50] were the first to administer a siRNA (ALN-PCS) able to block the mRNA of PCSK9 into humans. In this randomized, single-blind, placebo-controlled, phase I clinical trial, 32 healthy adults with serum LDL-C > 116 mg/dL (3.00 mmol/L) were randomized 3:1 to receive one EV dose of ALN-PCS (dose range: 0.015–0.400 mg/kg) or placebo over 1 h. Specifically, 6 dosages were assessed: 0.015 (3 patients), 0.045 (3 patients), 0.090 (3 patients), 0.150 (3 patients), 0.250 (6 patients), and 0.400 mg/kg (6 patients) [50]. The mean percentage

Table 1
Chemical and physical characteristics of inclisiran.

Inclisiran	
Characteristics	Values
Name	RNA, (Am-sp-(2′-deoxy-2′-fuoro)C-sp-Am-(2′-deoxy-2′-fuoro)A-(2′-deoxy-2′-fuoro)A-(2′-deoxy-2′-fuoro) A-Gm-(2′-deoxy-2′-fuoro)C-Am-(2′-deoxy-2′-fuoro)A-Am-(2′-deoxy-2′-fuoro)A-Cm-(2′-deoxy-2′-fuoro) A-Gm-(2′-deoxy-2′-fuoro)G-Um-(2′-deoxy-2′-fuoro)C-Um-Am-Gm-sp-Am-sp-Am), complex with RNA (Cm-sp-Um-sp-Am-Gm-Am–Cm-(2′-deoxy-2′-fuoro)C-Um-(2′-deoxy-2′-fuoro)G-Um-dT-Um-Um-Gm-CmUm-Um–Um-Um-Gm-Um) 3′-(((2 S,4 R)−1-(29-((2-(acetylamino)− 2-deoxy-beta-D-galactopyranosyl)oxy)-14,14-bis((3-((3-((5-((2-(acetylamino)−2-deoxy-beta-D-galactopyranosyl)oxy)− 1-oxopentyl) amino)propyl) amino)−3-oxopropoxy)methyl)−1,12,19,25-tetraoxo-16-oxa-13,20,24-triazanonacos-1-yl)−4-hydroxy-2-pyrrolidinyl)methyl hydrogen phosphate) (1:1)
Molecular form	C_{529} H_{664} F_{12} N_{176} Na_{43} O_{316} P_{43} S_6
Type of compound	Small interfering RNA (siRNA) with two strands: sense and antisense strands overhanging each other
Molecular weight	17,284.75 g/mol
Recommended dose	300 mg (284 mg inclisiran free acid) single dose to be administered as following: first dose, second dose after 3 months, then every 6 months.
Administration	Subcutaneous
Mean half-life	Overall mean half-life: 9.58 h 1. Injection site: 526 h 2. Liver: 270 h 3. Kidney: 360 h
Plasma protein binding	87.4%
Median Tmax	6 h (0.5–12 h)
Cmax	509 ng/mL
AUC	7980 ng/mL*h
Volume of distribution	500 L
Clearance	38.1 L/hour
Elimination	• Liver: 82.5% • Kidney: 15.9%
Interaction with cytochrome P450 isoforms	None declared
Pharmacodynamics	After the release of the sense strand by the RISC, the antisense strand interacts with the mRNA of PCSK9 in the cytoplasm. The RISC can destroy the mRNA of the PCSK9, thus reducing the translation of the corresponding protein and, therefore, the degradation of LDL-R. This allows the reduction in LDL-C.
EMA indications	Adults with primary hypercholesterolemia (heterozygous familial and non-familial) or mixed dyslipidemia: • in combination with a statin or statin + other lipid-lowering agents when target LDL-C was not reached with the maximum tolerated dose of statin or, • • alone or in combination with other lipid-lowering therapies in statin-intolerant or statin-contraindicated patients.

Abbreviations: EMA: European Medicines Agency; LDL-C: low density lipoprotein cholesterol; LDL-R: low density lipoprotein receptor; mRNA: messenger RNA; PCSK9: proprotein convertase subtilisin-kexin type 9; RISC: RNA-induced silencing complex.

change in fasting plasma PCSK9 levels from baseline versus placebo was 69.2% and 69.7%, with the 0.250 and 0.400 mg/kg doses, respectively ($p < 0.0001$), which corresponded to a mean 27.8% and 40.1% change in fasting serum LDL-C from baseline versus placebo, respectively ($p < 0.01$) [50]. The drug showed no significant adverse effects versus placebo, except a transient, mild, macular, erythematous rash. The evolution of ALN-PCS (which was then named inclisiran) was the subcutaneous injection of the drug. *Fitzgerald* et al. [51] performed a phase I trial including healthy individuals with serum LDL-C > 100 mg/dL and fasting triglyceride levels < 400 mg/dL, who were randomized 3:1 to inclisiran in a single ascending dose phase (25, 100, 300, 500, or 800 mg) or in a multiple-dose phase (125 mg weekly for four doses, 250 mg every other week for two doses, or 300 or 500 mg monthly for two doses, with or without concurrent statin therapy). Inclisiran at 300 mg or higher was able to promote an overall reduction in PCSK9 levels, from a 69.3% to a 73.9% reduction, versus placebo and, in parallel, a corresponding decrease in LDL-C (from 32.5% to 39.7%), total cholesterol (from 22.7% to 26.5%), non-HDL-C (from 25.3% to 37.2%), and apolipoprotein B (from 22.2% to 31.8%) [51]. The evaluation of serum levels within 84 days after the first dose, with or without statins, revealed a mean reduction in PCSK9 levels ranging from 62.1% to 100.7% versus placebo, while the LDL-C reduction was from 45% to 59.7% [51].

The administration of inclisiran to patients at high risk for cardiovascular disease (with or without a history of ASCVD or equivalent risk) with elevated LDL-C levels in the ORION-1 trial (501 patients on lipid lowering therapies randomized to a single dose of placebo or 200, 300, or 500 mg of inclisiran or two doses of placebo or 100, 200, or 300 mg of inclisiran [days 1 and 90]) revealed a maximum reduction in LDL-C of − 41.9% (500 mg one dose) or − 52.6% (300 mg two-dose) at 180 days [52]. Indeed, at a dose of 300/500 mg inclisiran was able to promote a statistical significant reduction in non-HDL-C, triglycerides, apolipoprotein B, and lipoprotein (a) [52].

At the one-year follow-up, maximum LDL-C reduction was 38.7% and 46.4%, respectively, with the two-dose 300 mg regimen providing the most powerful reduction in LDL-C [53].

The ORION-3 trial [54], the extension arm of the ORION-1 study, will provide insights in the long-term (up to 4 years) management of lipids levels with inclisiran. Interestingly, patients will be divided into two groups: one with inclisiran only (300 mg every 180 days up to 4 years) and the other with evolocumab 140 mg every 14 days up to Day 336, then inclisiran 300 mg every 180 days up to 4 years [54]. The main outcome is the measurement of the percentage change in LDL-C during the follow-up.

The ORION-10 and ORION-11 trials were phase III studies conducted to evaluate the percentage change in LDL-C at the 540-day follow-up in patients with atherosclerotic cardiovascular disease (ORION-10, no. of patients: 1561) and patients with atherosclerotic cardiovascular disease, or an atherosclerotic cardiovascular disease risk equivalent (ORION-11, no. of patients: 1617), respectively [55]. Inclisiran 284 mg or placebo were administered subcutaneously on Day 1, Day 90, and then every 6 months until the end of the follow-up. Inclisiran was able to reduce LDL-C by 52.3% (ORION-10) and 49.9% (ORION-11) at the 510-day follow-up. Such results were consistent throughout the follow-up period. No differences were found with regard to the incidence of serious adverse events. Although an exploratory analysis was performed, in order to evaluate the impact of inclisiran on the cardiovascular endpoints, no definite analysis could be performed, due to the small number of events [55].

The ORION-4 trial [56] has been designed to evaluate the impact of inclisiran on MACE by enrolling about 15,000 individuals with pre-existing atherosclerotic cardiovascular disease randomized to inclisiran 300 mg every 180 days or placebo. This is going to be a cornerstone trial in the ORION program because of the definite inclusion of inclisiran in the clinical practice (Table 2). The ORION-4 trial will include patients older than 55 years with at least one of the following

Table 2
The development of the Inclisiran: a rappraisal of the ORION/VICTORION scheduled program.

Trial	Clinical Phase	n. pts	Patients' characteristics	Intervention/ treatment	Timing administration	Follow-up	Outcomes	Status
ORION-1	II	501	ASCVD or ASCVD Risk-Equivalents and Elevated LDL-C	Inclisiran at single or multiple s.c. injections	200 mg day 1 (bi-annual) vs 300 mg day 1 (bi-annual) vs 500 mg day 1 (bi-annual) vs 100 mg day 1 and 90 (quarterly) vs 200 mg day 1 and 90 (quarterly) vs 300 mg day 1 and 90 (quarterly) vs placebo	180 days	Primary: % change LDL-C Secondary • N pts with LDL-C reduction > 80% • N pts with LDL-C reduction > 50% • % Change PCSK9 and other Lipids, Apolipoproteins, and inflammatory markers. • N pts who attained Targets for Level of ASCVD	Completed
ORION-2	II	9	HoHF	Inclisiran sodium 300 mg s.c.	Day 1. If PCSK9 levels not suppressed by > 70% at Day 60 or Day 90, then second dose at Day 90 or Day 104, respectively	180 days	Primary: % Change LDL-C Secondary: Absolute and % change LDL-C, PCSK9, Total-C, TG, HDL-C, Non-HDL-C, VLDL-C, ApoA1, ApoB, Lp(a)	Completed
ORION-3	II	490	ASCVD or ASCVD Risk-Equivalents and Elevated LDL-C who completed ORION-1	Inclisiran sodium 300 mg s.c. vs evolocumab 140 mg	Day 1, Day 90, and then every 6 months. Evolocumab group will receive inclisiran 300 mg on Day 360 and every 180 days thereafter for up to 4 years.	4 years	Primary: % Change LDL-C at Day 210 vs Day 1 of the ORION-1 Study Secondary: • % Change LDL-C, PCSK9, Total-C, TG, HDL-C, non-HDL-C, VLDL-C, Apo-A1, Apo-B, and Lp(a) till 4 yrs • % pts in the Inclisiran Group and the Evolocumab Group Who Attain Targets for ASCVD Risk • % pts in Evolocumab Group Who Attain Targets for ASCVD Risk • % pts (Inclisiran vs Evolocumab) with ≥ 50% LDL-C Reduction • N. pts (Inclisiran vs Evolocumab) Reaching LDL-C Levels of < 25 mg/dL, < 50 mg/dL, < 70 mg/dL, and < 100 mg/dL	Active, not recruiting
ORION-4	III	15,000	ASCVD	Inclisiran sodium 300 mg s.c. vs Placebo	Day 1, Day 90, and then every 6 months.	5 years	Primary: MACE [CHD death; MI; Fatal or non-fatal ischemic stroke; Urgent coronary revascularization procedure] Secondary: • MACE in high-intensity statin at baseline • Composite CHD death or MI • CV death	Recruiting
ORION-5	III	56	HoHF	Inclisiran sodium 300 mg s.c. vs Placebo	Part 1: 6-month double-blind period inclisiran or placebo on Days 1 and 90 Part 2: 18-month open-label follow-up period where placebo-treated subjects from Part 1 will be transitioned to inclisiran on Day 180	720 days	Primary: % Change LDL-C Secondary: • Absolute and % change LDL-C, PCSK9, Total-C, non-HDL C, HDL-C, VLDL-C, apoB, Apo-A1, Lp(a), hsCRP • Number of pts reaching on treatment LDL-C levels of	Active, not recruiting

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Table 2 (*continued*)

Trial	Clinical Phase	n. pts	Patients' characteristics	Intervention/ treatment	Timing administration	Follow-up	Outcomes	Status
							< 25 mg/dL, < 50 mg/ dL, < 70 mg/dL, and < 100 mg/dL at Days 150, 180, 330, 510, 690, and 720 • LDL-C reduction ≥ 20% or ≥ 30%	
ORION-6	I	24	Hepatic impairment	/	/	/	Primary: Pharmacokinetics at different stage of hepatic function	Completed
ORION-7	I	31	Renal impairment	Inclisiran sodium 300 mg s.c.	Day 1	60 days	Primary: Pharmacokinetics at different stage of renal function Secondary: • Change In Lipids And Lipoproteins At Day 60 • Change In PCSK9 At Day 60	Completed
ORION-8	III	2991	HeHF, HoHF, ASCVD or ASCVD Risk-Equivalents and Elevated LDL-C open label, long term extension study ORION-9, ORION-10, ORION-11, and ORION-5	Inclisiran sodium 300 mg s.c. vs Placebo	Day 1 (except ORION-5), Day 90, and then every 6 months to day 990.	1080 days	Primary: • % pts reaching LDL-C targets of < 70 mg/dL; < 100 mg/dL • % pts reaching global lipid targets Secondary: Effect of inclisiran on LDL-C levels, Total-C, TG, and HDL-C Other Outcome: Safety assessment	Active, not recruiting
ORION-9	III	482	HeHF and Elevated LDL-C	Inclisiran sodium 300 mg s.c. vs Placebo	Day 1, Day 90, and then every 6 months.	540 days	Primary: % Change in LDL-C Secondary: • Absolute Change LDL-C • % Change PCSK9, Total-C, ApoB, Non-HDL-C	Completed
ORION-10	III	1561	ASCVD and Elevated LDL-C	Inclisiran sodium 300 mg s.c. vs Placebo	Day 1, Day 90, and then every 6 months.	540 days	Primary: % Change in LDL-C Secondary: • Absolute Change LDL-C • % Change PCSK9, Total-C, ApoB, Non-HDL-C	Completed
ORION-11	III	1617	ASCVD or ASCVD Risk-Equivalents and Elevated LDL-C	Inclisiran sodium 300 mg s.c. vs Placebo	Day 1, Day 90, and then every 6 months.	540 days	Primary: % Change in LDL-C Secondary: • Absolute Change LDL-C • % Change PCSK9, Total-C, ApoB, Non-HDL-C	Completed
ORION-12	I	48	Healthy volunteers	/	/	/	ECG modifications	/
ORION-13	III	15	Adolescents (12 to Less Than 18 years) with HoFH	Inclisiran sodium 300 mg s.c. vs Placebo	Year 1 - inclisiran at Days 1, 90, and 270 vs - placebo Year 2 - inclisiran at Days 450 and 630 also in placebo group	720 days	Primary: % change LDL-C Secondary: • Absolute and % change LDL-C, PCSK9, other lipoprotein, and lipid parameters	Recruiting
ORION-14	I	40	Chinese with elevated LDL-C despite lowering therapies	Inclisiran sodium 300 mg vs Inclisiran sodium 100 mg vs placebo	Day 1. Evaluation of parameters at days 30, 60 and 90.	90	Evaluate Pharmacokinetics and Pharmacodynamics of Inclisiran Treatment in Chinese	Recruiting
ORION-15	II	308	Japanese ASCVD and Elevated LDL-C	Inclisiran sodium 300 mg vs Inclisiran sodium 200 mg vs Inclisiran sodium 100 mg vs placebo	Day 1, 90, and 270.	180	Evaluate Pharmacokinetics and Pharmacodynamics of Inclisiran Treatment in Japanese	Recruiting
ORION-16	III	150				720 days		Recruiting

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Table 2 (*continued*)

Trial	Clinical Phase	n. pts	Patients' characteristics	Intervention/ treatment	Timing administration	Follow-up	Outcomes	Status
			Adolescents (12 to Less Than 18 years) with HeFH	Inclisiran sodium 300 mg s.c. vs Placebo	Year 1 - inclisiran at Days 1, 90, and 270 vs - placebo Year 2 - inclisiran at Days 450 and 630 also in placebo group		Primary: % change LDL-C Secondary: • Absolute and % change LDL-C, PCSK9, other lipoprotein, and lipid parameters	
ORION-17	III	40,000	Primary prevention trial	/	/	/	/	/
ORION-18	III	320	Asians with ASCVD or ASCVD Risk-Equivalents and Elevated LDL-C	Inclisiran sodium 300 mg s.c. vs Placebo	Day 1, Day 90, and then every 6 months.	360 days (extension to 3 years)	Primary: % Change in LDL-C Extension: safety and tolerability of inclisiran at 3 yrs Secondary: • Absolute change LDL-C • Absolute and % change PCSK9, Total-C, ApoB, non-HDL-C, ApoA1, HDL-C, Lp(a), and TG • % participants reaching LDL-C levels of < 25 mg/dL, < 50 mg/dL, < 70 mg/dL, and < 100 mg/dL • % participants in each group with $\geq 50\%$ LDL-C reduction • % participants attaining lipid targets	Recruiting
VICTORION-INITIATE	III	444	ASCVD and Elevated LDL-C (> 70 mg/dL)	Inclisiran sodium 300 mg s.c. + usual care vs usual care	/	330 days	Primary: • % change LDL-C • Discontinuation statin therapy Secondary: • Absolute change LDL-C • Absolute and % change LDL-C levels to each post-baseline visit • Achieving $\geq 50\%$ reduction from baseline in LDL-C (yes, no) • Achieving LDL-C < 100 mg/dL, < 70 mg/dL, < 55 mg/dL • Absolute and % change apoB, non-HDL-C, VLDL-C, Total-C, Lp(a), HDL-C, TG • Dose variations in lipid lowering therapy • Proportion of days covered	Recruiting
VICTORION-INCEPTION	III	384	Recent (within 5 weeks) Acute Coronary Syndrome	Inclisiran sodium 300 mg s.c. + usual care vs usual care	/	360	Primary: • % change LDL-C • % achieving LDL-C < 70 mg/dl Secondary: • Absolute change LDL-C • Achieving $\geq 50\%$ reduction LDL-C • % achieving LDL-C targets • Absolute and % change apoB, VLDL-C, HDL-C, Lp (a), non-HDL-C, and Total-C • Dose variations in lipid lowering therapy • % discontinuing statin therapy	Recruiting

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Table 2 (*continued*)

Trial	Clinical Phase	n. pts	Patients' characteristics	Intervention/ treatment	Timing administration	Follow-up	Outcomes	Status
VICTORION-2 PREVENT	III	15,000	Established cardiovascular disease	Inclisiran sodium 300 mg s.c. vs Placebo	Day 1, Day 90, and then every 6 months.	6 years	Primary: Time to First Occurrence of 3 P-MACE (CV death, non-fatal MI and non-fatal ischemic stroke) Secondary: • Time to Occurrence of CV Death • Time to First Occurrence of 4 P-MACE (CV death, non-fatal MI, non-fatal ischemic stroke and urgent coronary revascularization) • Time to occurrence of all-cause death.	Not yet recruiting

Abbreviations: apo: apolipoprotein; ASCVD: atherosclerotic cardiovascular disease; CHD: Coronary heart disease; CV: cardiovascular; HDL-C: high density lipoprotein cholesterol; HeFH: Heterozygous familial hypercholesterolemia; HoHF: Homozygous Familial Hypercholesterolemia; hsCRP: high sensitivity C-reactive protein; LDL-C: low density lipoprotein cholesterol; Lp(a): lipoprotein (a); MACE: major adverse cardiovascular events; MI: Myocardial infarction; PCSK9: Proprotein convertase subtilisin/kexin type 9; pts: patients; TG: triglycerides; Total-C: Total cholesterol; VLDL-C: very low density lipoprotein cholesterol

criteria: prior myocardial infarction, ischemic stroke, or peripheral artery disease (i.e. prior lower extremity artery revascularization or aortic aneurysm repair). No patient with recent (< 4 weeks before enrollment) acute coronary syndrome or stroke, or planned coronary revascularization, known chronic liver disease, dialysis/transplantation, and previous exposure to inclisiran/on treatment with PCSK9 inhibitors will be included. The trial has been planned for evaluating 5-years occurrence of MACE (coronary heart disease [CHD] death; myocardial infarction [MI]; fatal or non-fatal ischemic stroke; urgent coronary revascularization procedure) as primary endpoint, and MACE in high-intensity statin at baseline, composite of CHD death or MI, or cardiovascular death as secondary outcomes. ORION-4 is currently enrolling patients and results would be released by December 2024 or earlier depending on when the endpoints are met [56].

The inhibition of PCSK9 is a key point also in the management of patients with familial hypercholesterolemia (FH): the recycling of LDL-R is crucial in conditions where most of the receptors are unavailable due to genetics. Inclisiran may play a further role in this context.

Four patients with homozygous familial hypercholesterolemia (HoFH) received inclisiran 300 mg on top of high-intensity statins and ezetimibe in the phase II ORION-2 trial [57]: PCSK9 transduction was persistently reduced. The phase III ORION-5 trial is an ongoing study which will involve 45 patients with HoFH treated with inclisiran 300 mg on Days 1 and 90 versus placebo: the results will attempt to provide more insights in the pharmacological management of this rare disease [58].

A phase III clinical trial (ORION-9) was performed to evaluate the role of inclisiran in heterozygous familial hypercholesterolemia (HeFH) [59]. The trial enrolled 482 HeHF patients, who were randomized to inclisiran (300 mg) or placebo: inclisiran was able to promote a 47.9% reduction in LDL-C versus placebo, with similar adverse events [59].

ORION-8 – i.e. the extension of the ORION-9, −10, and −11 studies – will give more information about the long-term (up to three years) effects of inclisiran in patients with atherosclerotic cardiovascular disease and/or FH [60].

Although trial outcomes are still lacking – and the ongoing studies will hopefully deal with this issue – attempts for evaluating the impact of inclisiran on MACE have been made by including data from the literature. A recent meta-analysis from *Asbeutah* et al. [61] on RCTs, comparing at least 2 doses of 284 mg or more of inclisiran with placebo did not detect any improvement in the main cardiovascular outcomes (fatal and non-fatal myocardial infarction, fatal and non-fatal stroke, and cardiovascular mortality). *Khan* et al. [62] gathered data from the ORION-9, −10, and −11 clinical studies: inclisiran was able to reduce MACE by 24% as compared to placebo. However, the three trials were underpowered for the evaluation of cardiovascular outcomes. Interestingly, no significant adverse events were recorded [62]. Further studies are needed in order to better evaluate the impact of inclisiran on the outcomes.

5. Therapeutic role

The need for keeping LDL-C plasma levels lower is at the core of the international guidelines on the management of lipids levels and cardiovascular prevention [1,2,63].

The inclusion of inclisiran as a further pharmacological compound able to promote the reduction in LDL-C plasma levels increases the chances to reduce the overall cardiovascular risk profile of individuals (Table 3). The *European Medicines Agency* (EMA) authorized inclisiran throughout in all European Countries on December 2020 in patients with primary hypercholesterolemia (heterozygous familial and non-familial) or mixed dyslipidemia, on top of maximum tolerated doses of lipid-lowering drugs, either alone or with further lipid-lowering agents, in patients who showed intolerance or contraindications to statins [64,65]. Delays in the approval process are in compliance with the provisions of the US FDA (*Food and Drug Administration*) [65].

The need to include inclisiran in the general management of patients with dyslipidemia deserves more considerations. Despite the lack of data on primary outcomes, inclisiran may be part of the treatment of patients with dyslipidemia both in primary and secondary prevention programs.

Statins still remain the cornerstone of the management of dyslipidemias: they showed a number needed-to-treat (NNT) between 3 and 61, according to the overall individual cardiovascular risk profile and baseline LDL-C levels, as well as the lowest cost among antilipemic drugs [66]. The absolute frequencies of adverse events (AE) of the statin therapy (number needed-to-harm [NNH]: 190 for any AEs and 209 for non-serious AEs, excluding rhabdomyolysis and CPK >10-fold ULN [67]) are responsible for the risk of withdrawal, and therefore for the increase in the risk of cardiovascular events above all in secondary prevention protocols [68].

Indeed, data from the EUROASPIRE V registry showed that 48% of patients on statin therapy at the maximum tolerated dose were unable to reach the LDL-C target [69]. The DA VINCI study [19] outlined the impressive gap between guidelines recommendations and clinical

Table 3
Reappraisal of the main features of clinical studies about the inclisiran performance on lipid levels.

Study	N. of pts	Type of pts	Type of study	Design	Approach	Inclisiran	Follow-up	Results
Fitzgerald et al. [48]	32	Healthy adult volunteers with LDL-C > 3 mmol/L	Phase I	Randomized, single-blind, placebo-controlled, dose-escalation study	Random assignment 3:1 ratio	3 pts 0.015 mg/kg 3 pts 0.045 mg/kg 3 pts 0.090 mg/kg 3 pts 0.150 mg/kg 6 pts 0.250 mg/kg 6 pts 0.400 mg/kg	180 days	*Fasting plasma PCSK9 levels* Mean percentage change from baseline relative to placebo 0.015 mg/kg: −30.9%; 0.045 mg/kg: −63.9%; 0.090 mg/kg: −48.7%; 0.150 mg/kg: −64.9%; 0.250 mg/kg: −69.2%; 0.400 mg/kg: −69.7% *Fasting plasma LDL-C levels* Mean percentage change from baseline relative to placebo 0.015 mg/kg: −6.2%; 0.045 mg/kg: −11.4%; 0.090 mg/kg: −24.7%; 0.150 mg/kg: −22.2%; 0.250 mg/kg: −27.8%; 0.400 mg/kg: −40.1%
Fitzgerald et al [49]	24	Healthy adult volunteers with LDL-C > 100 mg/dL	Phase I	Randomized, single-blind, placebo-controlled, dose-escalation study	Random assignment 3:1 ratio	*Single-dose Phase* 18 patients subcutaneous injection in a single-ascending-dose phase (25, 100, 300, 500, or 800 mg)	84 days	*Differences to placebo:* PCSK9 levels: from −31.4% (100 mg) to −73.9% (300 mg) LDL-C levels: from −10.5% (25 mg) to −39.7% (500 mg) Total Cholesterol: from −7.6% (25 mg) to −26.5% (300 mg) Non HDL-C: from − 8.1% (25 mg) to − 37.2% (300 mg) ApoB: from − 4.0% (25 mg) to − 31.8% (300 mg) Lp(a): from − 17.6% (25 mg) to − 48.1% (300 mg)
Fitzgerald et al. [49]	43	Healthy adult volunteers with LDL-C > 100 mg/dL	Phase I	Randomized, single-blind, placebo-controlled, dose-escalation study	Random assignment 3:1 ratio	*Multiple-dose Phase* 32 patients on multiple dose (125 mg weekly for four doses, 250 mg every other week for two doses, or 300 or 500 mg monthly for two doses, with or without statins)	84 days	*Differences to placebo:* PCSK9 levels: from − 62.1% (125 mg without statin) to − 100.7% (500 mg with statin) LDL-C levels: from − 30.9% (300 mg with statin) to − 49.3% (125 mg without statin) Total Cholesterol: from − 17.3% (300 mg with statin) to − 32.8% (300 mg without statin) Non HDL-C: from − 25.1% (300 mg with statin) to − 46.3% (300 mg without statin) ApoB: from − 24.4% (300 mg with statin) to − 39.6% (300 mg without statin) Lp(a): from − 13.2% (300 mg without statin) to − 36.7% (500 mg with statin)
Ray et al. [50]**ORION 1**	501	LDL-C > 70 mg/dL (pts with history atherosclerotic CVD) or > 100 mg/dL (pts without history atherosclerotic CVD)	Phase II	Multicenter, double-blind, placebo-controlled, multiple ascending-dose trial	Random assignment 3:1 ratio	*Single-dose Phase* 64 pts placebo or 200 mg (60 pts), 300 mg (60 pts), or 500 mg (60 pts) of inclisiran	180 days	*Differences to baseline:* PCSK9 levels: from − 47.9% (200 mg) to –59.3% (500 mg) LDL-C levels: from

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Table 3 (*continued*)

Study	N. of pts	Type of pts	Type of study	Design	Approach	Inclisiran	Follow-up	Results
								− 27.9% (200 mg) to −− 41.9% (500 mg) Total Cholesterol: from − 17.6% (200 mg) to –26.6% (500 mg) Non HDL-C: from − 25.1% (200 mg) to –36.9% (500 mg) ApoB: from − 22.9% (200 mg) to –33.1% (500 mg) Lp(a): from − 14.3% (200 mg) to –18.2% (500 mg)
Ray et al. [50]**ORION 1**	501	LDL-C > 70 mg/dL (pts with history atherosclerotic CVD) or > 100 mg/dL (pts without history atherosclerotic CVD)	Phase II	Multicenter, double-blind, placebo-controlled, multiple ascending-dose trial	Random assignment 3:1 ratio	*Two doses phase* (at days 1 and 90) 61 pts placebo or 100 mg (59 pts), 200 mg (60 pts), or 300 mg (59 pts) of inclisiran.	180 days	*Differences to baseline:* PCSK9 levels: from − 53.2% (200 mg) to –69.1% (500 mg) LDL-C levels: from − 35.5% (200 mg) to −− 52.6% (500 mg) Total Cholesterol: from − 22.4% (200 mg) to –33.2% (500 mg) Non HDL-C: from − 31.7% (200 mg) to –46.0% (500 mg) ApoB: from − 27.8% (200 mg) to –40.9% (500 mg) Lp(a): from − 14.9% (200 mg) to –25.6% (500 mg)
Ray et al. [51]**ORION 1 one year follow-up**	501	LDL-C > 70 mg/dL (pts with history atherosclerotic CVD) or > 100 mg/dL (pts without history atherosclerotic CVD)	Phase II	Multicenter, double-blind, placebo-controlled, multiple ascending-dose trial	Random assignment 3:1 ratio	*Single-dose Phase* 64 pts placebo or 200 mg (60 pts), 300 mg (60 pts), or 500 mg (60 pts) of inclisiran	360 days	*Differences to baseline:* PCSK9 levels: from − 44.5% to –55.9% LDL-C levels: from − 29.5% to −− 38.7%
Ray et al. [51]**ORION 1 one year follow-up**	501	LDL-C > 70 mg/dL (pts with history atherosclerotic CVD) or > 100 mg/dL (pts without history atherosclerotic CVD)	Phase II	Multicenter, double-blind, placebo-controlled, multiple ascending-dose trial	Random assignment 3:1 ratio	*Two doses phase* (at days 1 and 90) 61 pts placebo or 100 mg (59 pts), 200 mg (60 pts), or 300 mg (59 pts) of inclisiran.	360 days	*Differences to baseline:* PCSK9 levels: from − 43.1% to –60.5%LDL-C levels: from − 29.9% to −− 46.4%
Hovingh et al. [55] **ORION-2**	4	Pts with genetic or clinical diagnosis HoFH on high-intensity statins and ezetimibe	Phase II	Pilot cohort study	/	3 pts received inclisiran 300 mg injections on days 1 and 90; 1 pts only on day 1	360 days	*PCSK9 levels* Day 90: from − 48.7% to − 83.6% Day 180: from − 40.2% to − 80.5% *LDL-C levels* Day 90: from − 11.7% to − 33.1% Day 180: from − 17.5% to − 37.0%
Raal et al. [57] **ORION-9**	482	Pts with genetic or clinical diagnosis HeFH on maximally tolerated dose statins with or without ezetimibe	Phase III	Double-blind, randomized trial	Random assignment 1:1 ratio	Subcutaneous injections inclisiran 300 mg or placebo on days 1, 90, 270, and 450.	510 days	Percent change in LDLC: − 39.7% inclisiran vs + 8.2% placebo The time-averaged percent change in LDL-C_ − 38.1% inclisiran vs + 6.2% placebo
Ray et al. [53]**ORION 10**	1561	Pts with atherosclerotic CVD and ↑ LDL-C despite statin therapy at the maximum tolerated dose	Phase III	Randomized, double-blind, placebo-controlled, parallel-group trial	Random assignment 1:1 ratio	Subcutaneous injections inclisiran (284 mg) or placebo on day 1, 90, and every 6 months over a period of 540 days.	510 days	LDL-C: − 52.3% Total cholesterol: − 33.6% ApoB: − 44.8% Non-HDL-C: − 47.4%
Ray et al. [53]**ORION 11**	1617	Pts with atherosclerotic CVD or an atherosclerotic CVD risk equivalent and ↑ LDL-C despite statin therapy at the maximum tolerated dose	Phase III	Randomized, double-blind, placebo-controlled, parallel-group trial	Random assignment 1:1 ratio	Subcutaneous injections inclisiran (284 mg) or placebo on day 1, 90, and every 6 months over a period of 540 days.	510 days	LDL-C: − 49.9% Total cholesterol: − 28.0% ApoB: − 38.2% Non-HDL-C: − 41.2%

Abbreviations: ApoB: apolipoprotein B; CVD: cardiovascular diseases; LDL-C: low density lipoprotein cholesterol; Lp (a): lipoprotein (a); Non HDL-C: non high density lipoprotein cholesterol; PCSK9: proprotein convertase subtilisin-kexin type 9; Pts: patients

application of lipid lowering therapies. High-intensity statin monotherapy was administered in 20% and 38% of very high-risk primary and secondary prevention patients, respectively, while lower percentages were for the combination therapies with ezetimibe and/or further lipid-lowering therapies (PCSK9 inhibitors for example) [19]. Therefore, the attainment of the lipid targets according to international guidelines [1] was about 17% (high-risk primary prevention with high intensity statins), 22% (secondary prevention with high intensity statins), 9% (ezetimibe combination), and 58% (PCSK9 inhibitors combination) [19]. The Hyperlipidaemia Therapy in tERtiary Cardiological cEnTer (TERCET) Registry [70] further outlined the lowest percentages of patients with (STEMI) or without (NSTEMI) ST-segment elevation myocardial infarction/unstable angina who reached the LDL-C goal < 70 mg/dL (32.4%, 29.9%, and 27.8%, respectively) at one-year follow-up.

Ezetimibe allows the improvement in LDL-C levels in those who did not reach their target despite optimal medical therapy, or in case of statin-intolerance/contraindications [71]. A meta-analysis from *Guyton* et al. [72] outlined that a further 27% of patients at a higher cardiovascular risk achieved the LDL-C target of < 70 mg/dL, while 70% reached the LDL-C target of < 100 mg/dL. Those who still do not achieve the goals of their antilipemic treatment might be eligible for therapy with PCSK9-inhibitors, which demonstrated to positively impact the MACE [73,74].

Inclisiran could be included in the context of lipid management as a further option for reducing LDL-C plasma levels. The literature emphasized the need for combining different compounds for the treatment of dyslipidemia and the improvement of cardiovascular outcomes [75]. Specifically, international guidelines are promoting the need for early combination therapies in the management of lipid disorders, above all in very high risk and extremely high risk patients: as the combination of statins and inclisiran may reduce up to 75% the plasma levels of LDL-C, the early inclusion of inclisiran in daily practice would be beneficial in patients at higher risk of cardiovascular events. The ORION/VICTORION program will shed light on possible application of this "dual" or "triple" anti-lipemic therapy [76].

Statin intolerance or contraindications are at present the best indications for the application of this drug. Inclisiran showed no serious adverse events as compared to standard statin therapies for lipid management as demonstrated in completed clinical trials [52,53,55,59]. The occurrence rate of liver impairment and/or muscular lesions in patients treated with inclisiran were equal to placebo-treated patients, thus demonstrating the safety of inclisiran in daily clinical practice [52,53,55,59]. Therefore, inclisiran may be safely applied to statin-intolerant patients or those with contra-indication to statins.

Furthermore, interesting insights are about the role of inclisiran in liver and kidney impairment [43,77]. The lack of dramatically higher increase in liver enzymes and the evaluation of the drug in patients with liver impairment [43] revealed a possible, safe application of this compound in patients suffering with liver diseases. The lipid management of patients with kidney impairment would also benefit from the use of inclisiran. Results from the ORION-1 and ORION-7 confirmed the 50% reduction in LDL-C in patients with severe renal impairment (creatinine clearance level: 15–29 mL/min) [77]. Despite the increase in plasma concentrations, no influence was in the pharmacodynamics as well as in the incidence of adverse events rate, thus justifying the safe application of inclisiran even in patients with advanced renal failure without any need for dose-reduction [77].

Data from the ORION trials did not identify any impact on the platelet count or other hematological parameters [78]. The fear for antidrug antibodies might be related to the modified sugar backbone or the subcutaneous route of administration: indeed, data from ORION-1 did not show any significant percentage in antidrug antibodies, thus promoting the long-acting action of inclisiran [52,53,78]. ORION-10 and ORION-11 [55] found comparable adverse events rates between the inclisiran and the placebo arms, except for the reactions in the injection-site.

Lipid-lowering drugs usually show reduced adherence upon longer follow-ups, in particular when high-intensity regimens are administrated [79,80]. *Waßmuth* et al. [81] observed a 15.7% and 46.6% reduction in the adherence to the therapies with statins and ezetimibe, respectively. The reduced persistence on therapy negatively impacts the outcomes, both in primary and secondary prevention programs [82]. The biannual administration of inclisiran, combined with the lower rates of adverse events, may account for a higher long-standing persistence on therapy of patients. The role of general practitioners is fundamental in this context: the higher prevalence in CVD and CV risk factors should force them to actively promote cardiovascular prevention protocols based on the most advanced therapies [83]. Their efforts will certainly improve the adherence to advanced protocols also within the field of lipid management. The bi-annual administration of inclisiran and the preventive protocols adopted from general practitioners are the most intriguing proposal for the next level of lipid management.

The safety of inclisiran will promote the use of the drug in primary prevention protocols, above all in patients with ASCVD risk equivalent. A large number of trials with inclisiran involved patients with ASCVD risk equivalent (ORION-1, ORION-3, ORION-8, ORION-11, and ORION-18) (Table 2). The ORION-17 has been planned for the evaluation of inclisiran in a dedicated primary prevention program. This will promote the widespread of the drug in the general population.

The company drug is performing a complex, detailed scientific program for promoting the Inclisiran in different clinical settings. The ORION program – and the VICTORION panel of studies – will define the role of inclisiran in clinical practice, from the primary prevention context (ORION-17), to adolescents (12–16 years old) with HeFH/HoHF (ORION-13 and ORION-16) or individuals with recent acute coronary syndromes (VICTORION-INCEPTION), to secondary prevention and outcomes evaluation (ORION-4 and VICTORION-2 PREVENT) (Table 2). The development of these ongoing trials will provide more insights on the use of them in the daily clinical practice.

In clinical practice – and based on the main information gathered from the literature – the use of inclisiran can be considered compliant with the EMA indications for patients with hypercholesterolemia who do not reach the target levels of LDL-C despite the use of lipid-lowering drugs at the maximum tolerated dose. Recently, the Polish national guidelines on the diagnosis and therapy of lipid disorders proposed recommendation for the inclusion of inclisiran in clinical practice [84]. Specifically, the authors suggested to consider inclisiran: 1. in patients with ASCVD and / or FH who failed to achieve their target at the maximum tolerated dose of statin and ezetimibe (class IIb level of evidence B); 2. in individuals who did not tolerate statin regimen at any dose (even after rechallenge) (class IIb level of evidence C); 3. in those at very high-risk in primary or secondary prevention patients who do not adhere to or are not willing to use lipid-lowering therapy (class IIb level of evidence C) [84]. The results of the scientific development program of inclisiran will try to improve actual recommendations for inclisiran administration in clinical practice.

6. Conclusions

The pharmacological treatment of dyslipidemia is a challenging process. The achievement of target LDL-C plasma levels is critical in order to improve the cardiovascular outcomes, both within primary and secondary prevention programs. Inclisiran is a new drug based on the innovative inhibition of the transduction of the mRNA of the *PCSK9* gene. The long-standing action of inclisiran and its valuable ability to reduce LDL-C allow for its possible inclusion in clinical practice for the overall management of patients with dyslipidemia. Further trials – dealing above all with cardiovascular outcomes – will shed light on the effective potential of this pharmaceutical compound on the health of individuals.

CRediT authorship contribution statement

Pietro Scicchitano: Conceptualization, Methodology, Validation, Formal analysis, Investigation, Resources, Data curation, Writing – original draft, Writing – review & editing, Visualization, Supervision, Project administration, Funding Acquisition; **Michele Milo:** Conceptualization, Methodology, Validation, Formal analysis, Investigation, Data curation, Writing – original draft, Writing – review & editing, Visualization, Supervision; **Rosanna Mallamaci:** Methodology, Validation, Data curation, Writing – original draft, Writing – review & editing, Visualization, Supervision; **Micaela De Palo:** Conceptualization, Validation, Formal analysis, Investigation, Data curation, Writing – original draft, Writing – review & editing, Visualization, Supervision; **Pasquale Caldarola:** Conceptualization, Methodology, Validation, Formal analysis, Investigation, Writing – original draft, Writing – review & editing, Visualization, Supervision. **Francesco Massari:** Methodology, Validation, Formal analysis, Investigation, Writing – original draft, Writing – review & editing, Visualization, Supervision; **Domenico Gabrielli:** Methodology, Validation, Investigation, Data curation, Writing – review & editing, Visualization, Supervision, Funding acquisition. **Furio Colivicchi:** Methodology, Validation, Investigation, Data curation, Writing – review & editing, Visualization, Supervision, Funding acquisition; **Marco Matteo Ciccone:** Conceptualization, Methodology, Validation, Formal Analysis, Investigation, Resources, Data curation, Writing – original draft, Writing – review & editing, Visualization, Supervision, Project administration, Funding acquisition.

Conflict of interest statement

The authors declare no conflict of interest.

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