EAS

GESTIONE DELLE DISLIPIDEMIE: COSA C'È DI NUOVO?

Prof. Giuseppe Danilo Norata
Università degli Studi di Milano

EAS

FONDAZIONE S.I.S.A.

per la promozione della ricerca sulle malattie da arteriosclerosi

Gestione delle dislipidemie, cosa c'è di nuovo

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Il sottoscritto Giuseppe Danilo Norata
Ai sensi dell'art. 76 sul Conflitto di Interessi dell'Accordo Stato-Regioni del 02 febbraio 2017 in materia di ECM
Dichiara

che negli ultimi due anni ha avuto i seguenti rapporti con soggetti portatori di interessi commerciali in campo sanitario:

Amgen

Pfizer

Novartis

Sanofi

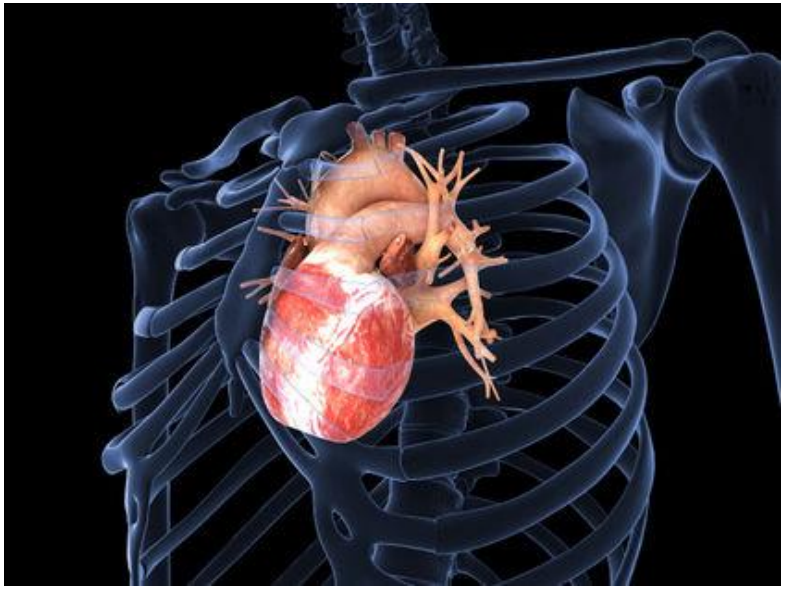
Eli Lilly

-New frontiers in hypercholesterolaemia
PCSK9 (Beyond Monoclonal antibodies)
ACLY (Bempedoic acid)
ANGPTL3 (Evinacumab, Vupanorsen, ARO-ANG3)

-Targeting trygliceridesrich lipoproteins
Icopentanyl EPA
APO CIII (Volanesorsen)
Pemafibrato
ANGPTL3

-Targeting Lp(a)
Apo(a) Gene silencing

-Targeting Inflammation
IL1beta (Canakinumab)
Colchicine



-New frontiers in hypercholesterolaemia

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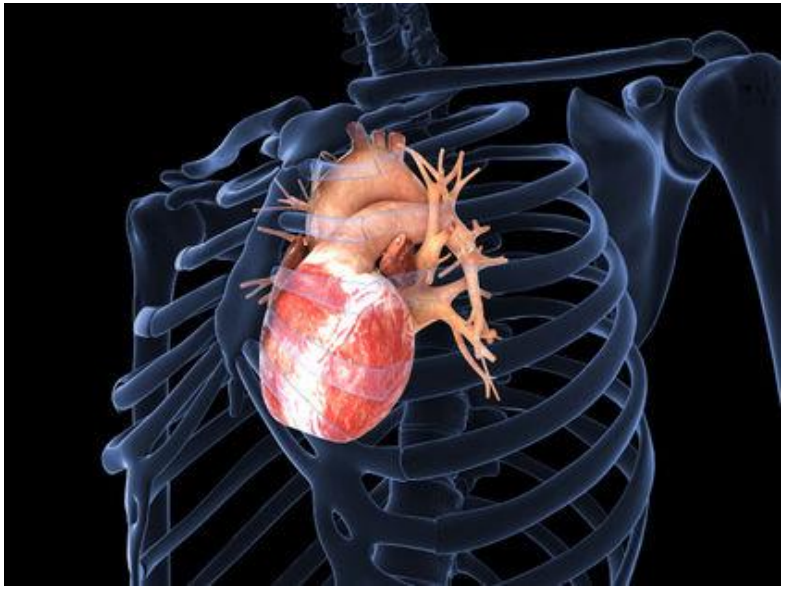
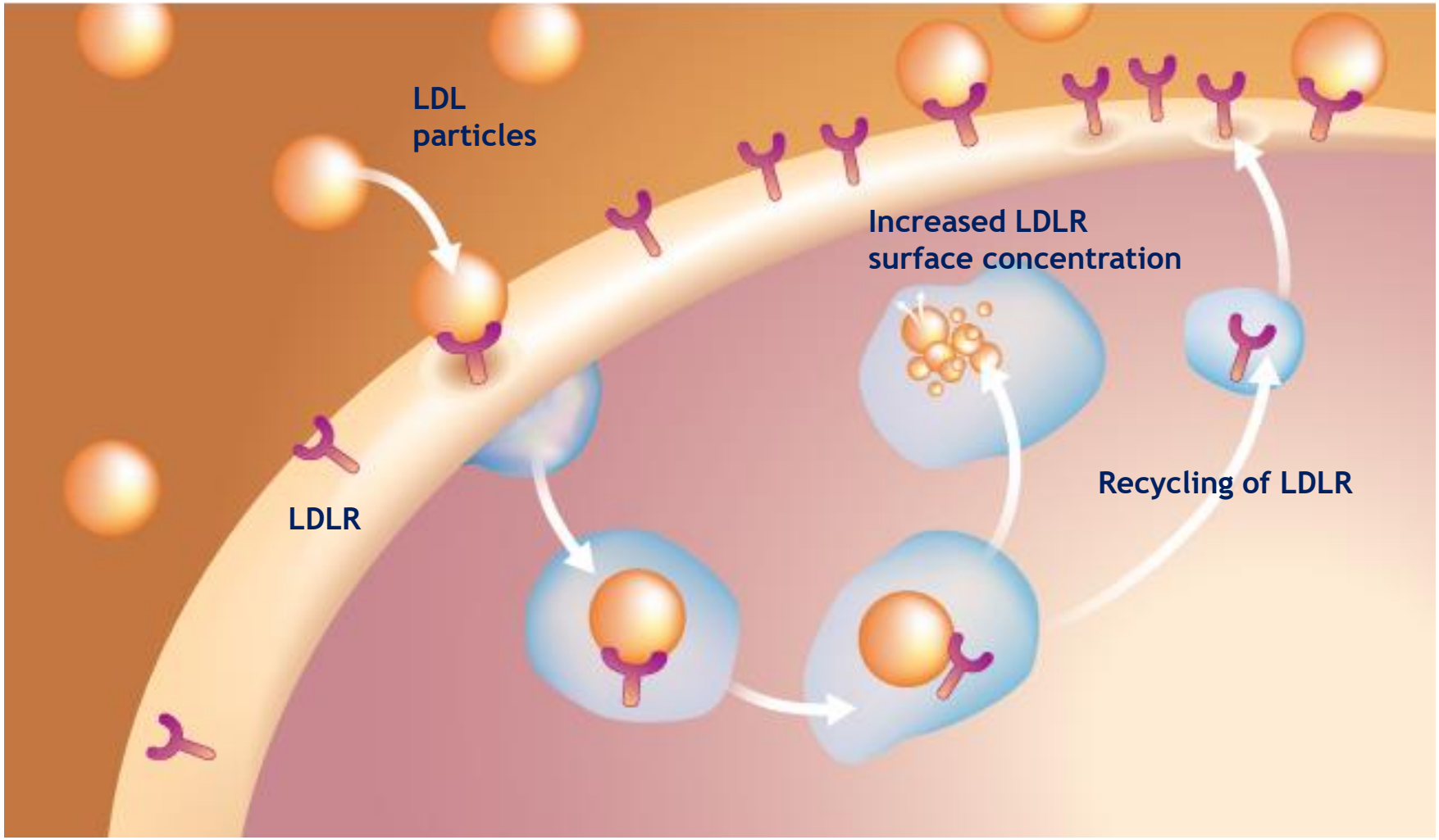


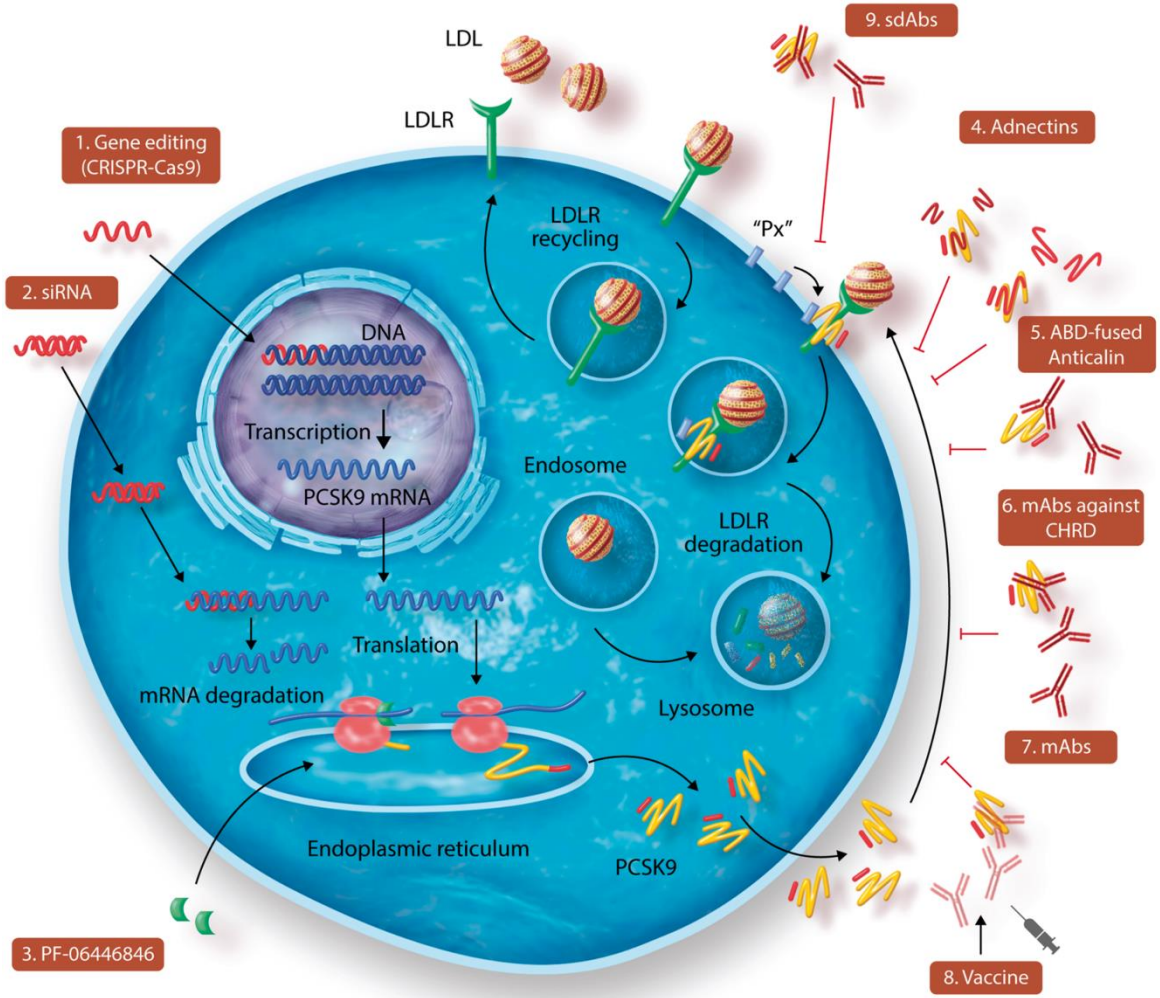
Plasma LDL-C levels are controlled by hepatic low-density lipoprotein receptor (LDLR)



Brown et al. Proc Natl Acad Sci USA 1979;76:3330–3337.

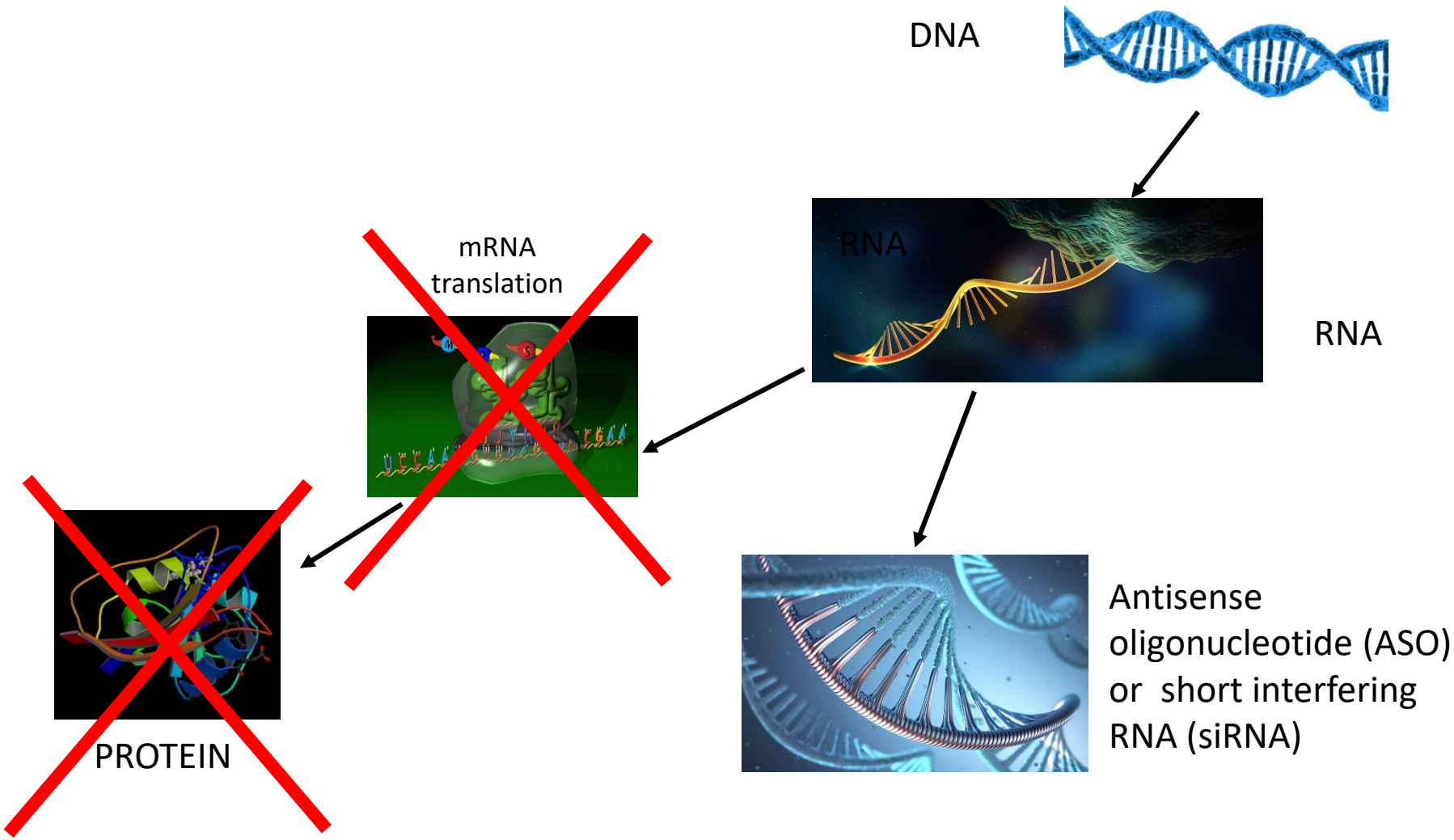
Modulators of LDLR half-life: PCSK9 inhibitors

Targeting PCSK9 beyond monoclonal antibodies

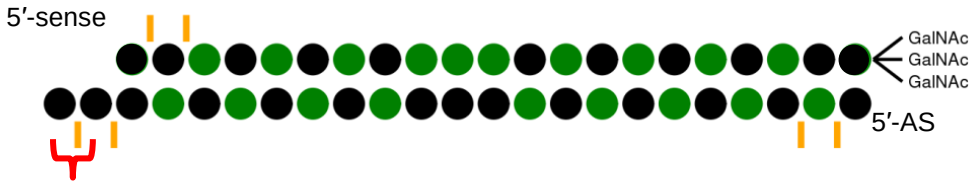


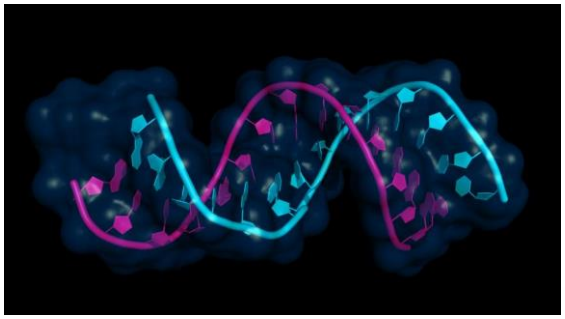
Seidah N, Norata GD. Cardiovasc Res 2019; 115:510-518.

Targeting RNA



PCSK9 gene silencing (Inclisiran)





Inclisiran binds to ASGPR

ASGPR

Hepatocyte membrane

Hepatocyte cytoplasm

ASGPR recycles

Endosome

Inclisiran enters cytoplasm

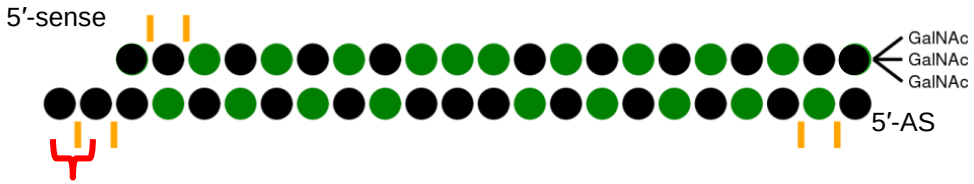
$GalNAc_3$

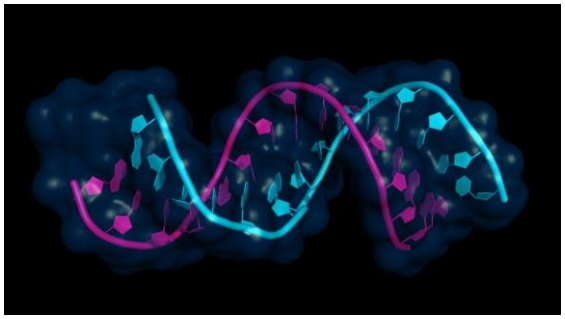
Inclisiran

Golgi Apparatus

Nucleus

PCSK9 gene silencing (Inclisiran)



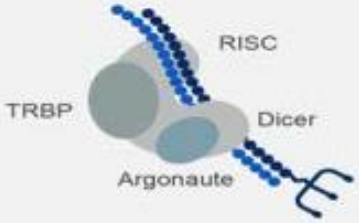


Hepatocyte membrane

Hepatocyte cytoplasm

Inclisiran

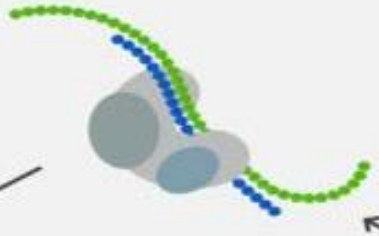
Inclisiran incorporates into RISC



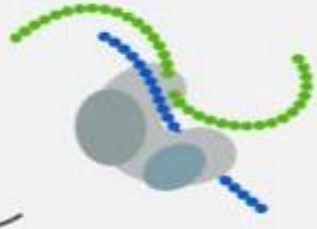
Strand separation

PCSK9 mRNA

PCSK9 mRNA pairing



PCSK9 mRNA is cleaved



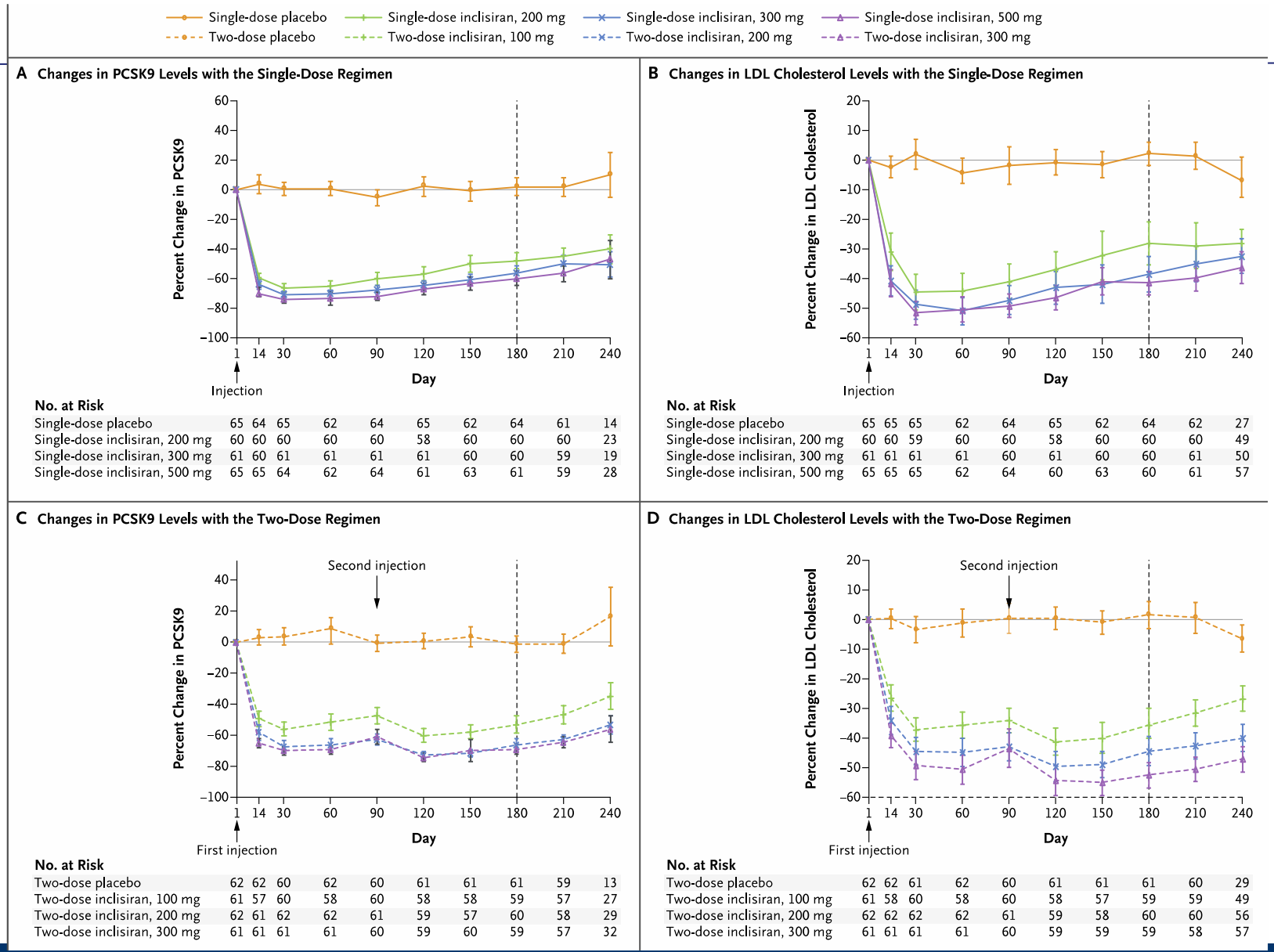
Catalytic degradation of PCSK9 mRNA

Inhibition of PCSK9 Synthesis

Golgi Apparatus

Nucleus

PCSK9 gene silencing: ORION-1 (Inclisiran)



Ray K et al, NEJM 2017

Inclisiran on going development

ASCVD or ASCVD risk equivalents
Total 18,500

ORION-4
(15,000 high risk ASCVD)
Phase III

ORION-11
(1500 ASCVD/RE)
Phase III

ORION-1
(500 ASCVD)
Phase II

ORION-10
(1500 ASCVD)
Phase III

ORION-3
Phase II extension

ORION-8
Phase III extension

HeFH & HoFH

ORION-2 & -5 (60 HoFH)
Phase II & III

ORION-13
(12 HoFH peds)
Phase III

ORION-9
(400 HeFH)
Phase III

ORION-16
(40 HeFH peds)
Phase III

ORION-19 HoFH (8-11)

ORION-20 HeFH (8-11)

Special populations

ORION-6 & -7
renal & hepatic
impaired) PK

ORION-14 & -15
PK/PD Japan &
China

ORION-12 TQT

Inclisiran: efficacy in ASCVD patients (Phase III: ORION 10, 11)

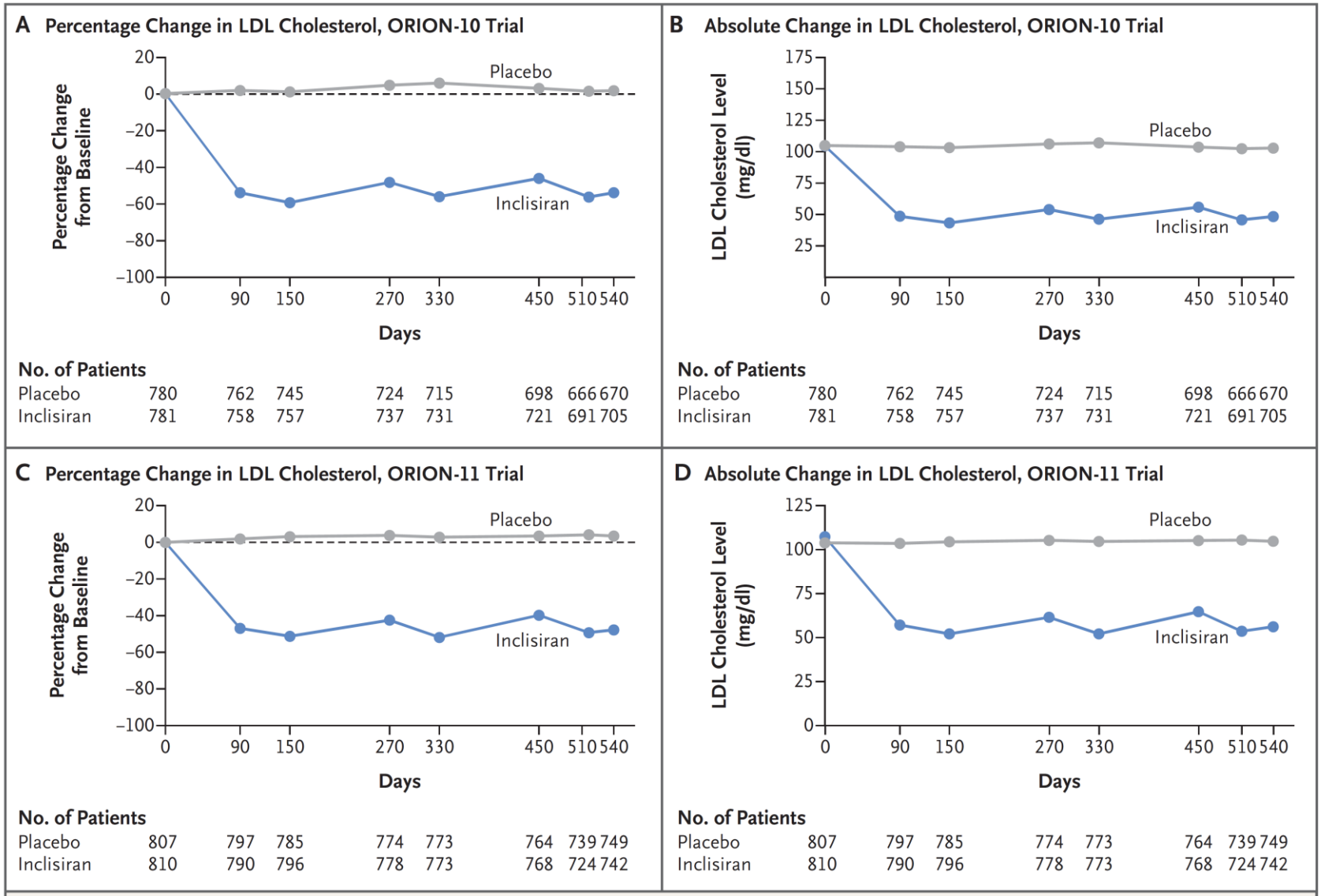


Figure 1. Efficacy of Inclisiran or Placebo in Lowering LDL Cholesterol over the 540-Day Trial Period (Intention-to-Treat Population).

Ray K et al, NEJM 2020

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Inclisiran: efficacy in ASCVD patients (Phase III: ORION 10, 11)

Subgroup	Inclisiran	Placebo	Least-Squares Mean Percentage Difference in LDL Cholesterol (95% CI)
	no.		
All patients	781	780	−57.0 (−60.1 to −53.8)
Sex			
Male	535	548	−55.5 (−59.1 to −51.9)
Female	246	232	−61.0 (−67.1 to −55.0)
Age			
<65 yr	297	333	−57.3 (−62.2 to −52.3)
≥65 yr	484	447	−56.9 (−61.0 to −52.9)
<75 yr	638	649	−57.2 (−60.7 to −53.7)
≥75 yr	143	131	−56.1 (−63.2 to −49.1)
Body-mass index			
≤Median	394	385	−54.1 (−58.3 to −49.9)
>Median	387	394	−59.8 (−64.4 to −55.2)
Race			
White	653	685	−58.2 (−61.4 to −55.0)
Black	110	87	−51.6 (−62.1 to −41.1)
Other	18	8	−44.8 (−92.2 to 2.5)
Baseline statin treatment			
Yes	701	692	−57.3 (−60.7 to −54.0)
No	80	88	−54.8 (−62.0 to −47.6)
Intensity of statin treatment			
High	538	546	−58.2 (−62.1 to −54.2)
Not high	243	234	−54.9 (−59.7 to −50.1)
Metabolic disease			
Diabetes	371	331	−55.2 (−60.6 to −49.9)
Metabolic syndrome	195	207	−60.4 (−65.8 to −54.9)
Neither	215	242	−56.8 (−61.6 to −52.0)
Renal function			
Normal	395	410	−56.9 (−61.0 to −52.8)
Mild impairment	269	260	−55.2 (−60.4 to −49.9)
Moderate impairment	113	107	−61.6 (−71.7 to −51.5)

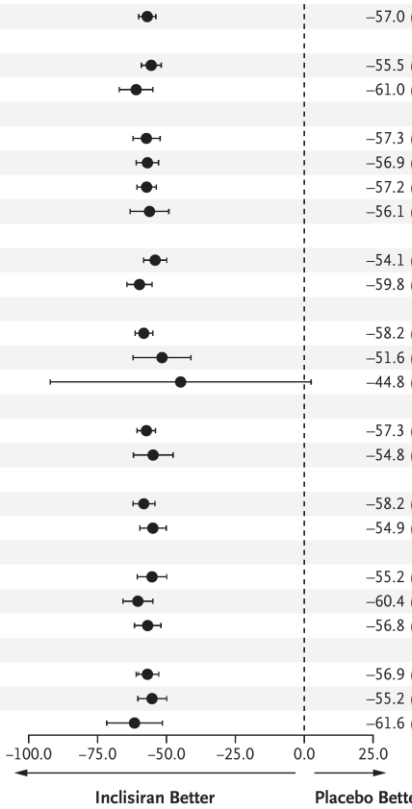


Figure 3. Subgroup Analysis of Placebo-Corrected Percentage Change in LDL Cholesterol from Baseline to Day 510 with Inclisiran in the ORION-10 Trial (Intention-to-Treat Population).

Subgroup	Inclisiran	Placebo	Least-Squares Mean Percentage Difference in LDL Cholesterol (95% CI)
	no.		
All patients	810	807	−52.7 (−55.8 to −49.7)
Sex			
Male	579	581	−52.5 (−56.1 to −48.9)
Female	231	226	−53.0 (−58.8 to −47.2)
Age			
<65 yr	367	366	−54.0 (−59.1 to −48.9)
≥65 yr	443	441	−51.7 (−55.4 to −48.1)
<75 yr	721	693	−52.8 (−56.1 to −49.4)
≥75 yr	89	114	−53.1 (−59.6 to −46.5)
Body-mass index			
≤Median	433	375	−51.5 (−55.7 to −47.3)
>Median	377	431	−54.5 (−59.0 to −50.1)
Race			
White	791	796	−52.6 (−55.7 to −49.5)
Black	12	8	−60.5 (−91.1 to −29.9)
Baseline statin treatment			
Yes	766	766	−53.3 (−56.5 to −50.1)
No	44	41	−41.6 (−51.1 to −32.1)
Intensity of statin treatment			
High	734	729	−53.4 (−56.6 to −50.2)
Not high	76	78	−45.4 (−55.0 to −35.8)
Metabolic disease			
Diabetes	296	272	−56.3 (−61.1 to −51.5)
Metabolic syndrome	212	236	−52.4 (−57.8 to −47.0)
Neither	302	299	−49.2 (−54.9 to −43.6)
Risk category			
ASCVD	712	702	−53.3 (−56.6 to −50.1)
ASCVD equivalent	98	105	−47.2 (−56.1 to −38.3)
Renal function			
Normal	428	444	−53.6 (−58.0 to −49.2)
Mild impairment	315	280	−51.8 (−56.8 to −46.9)
Moderate impairment	67	81	−51.8 (−59.7 to −43.9)
Geographic region			
Europe	750	746	−51.7 (−54.9 to −48.5)
South Africa	60	61	−66.3 (−74.9 to −57.7)

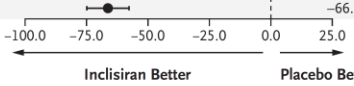


Figure 4. Subgroup Analysis of Placebo-Corrected Percentage Change in LDL Cholesterol from Baseline to Day 510 with Inclisiran in the ORION-11 Trial (Intention-to-Treat Population).

Ray K et al, NEJM 2020

Inclisiran: efficacy in HeFH patients (Phase III: ORION 9)

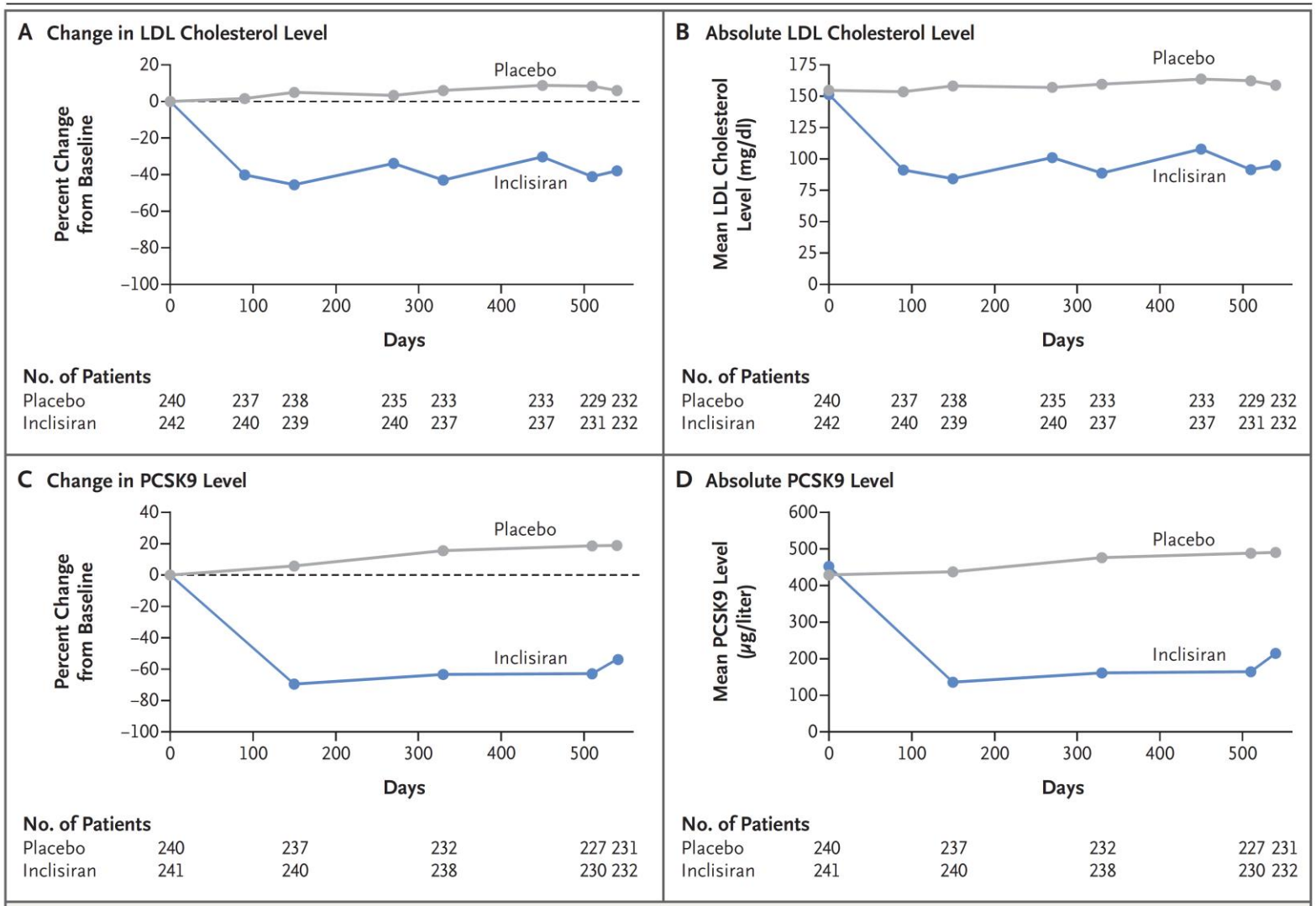
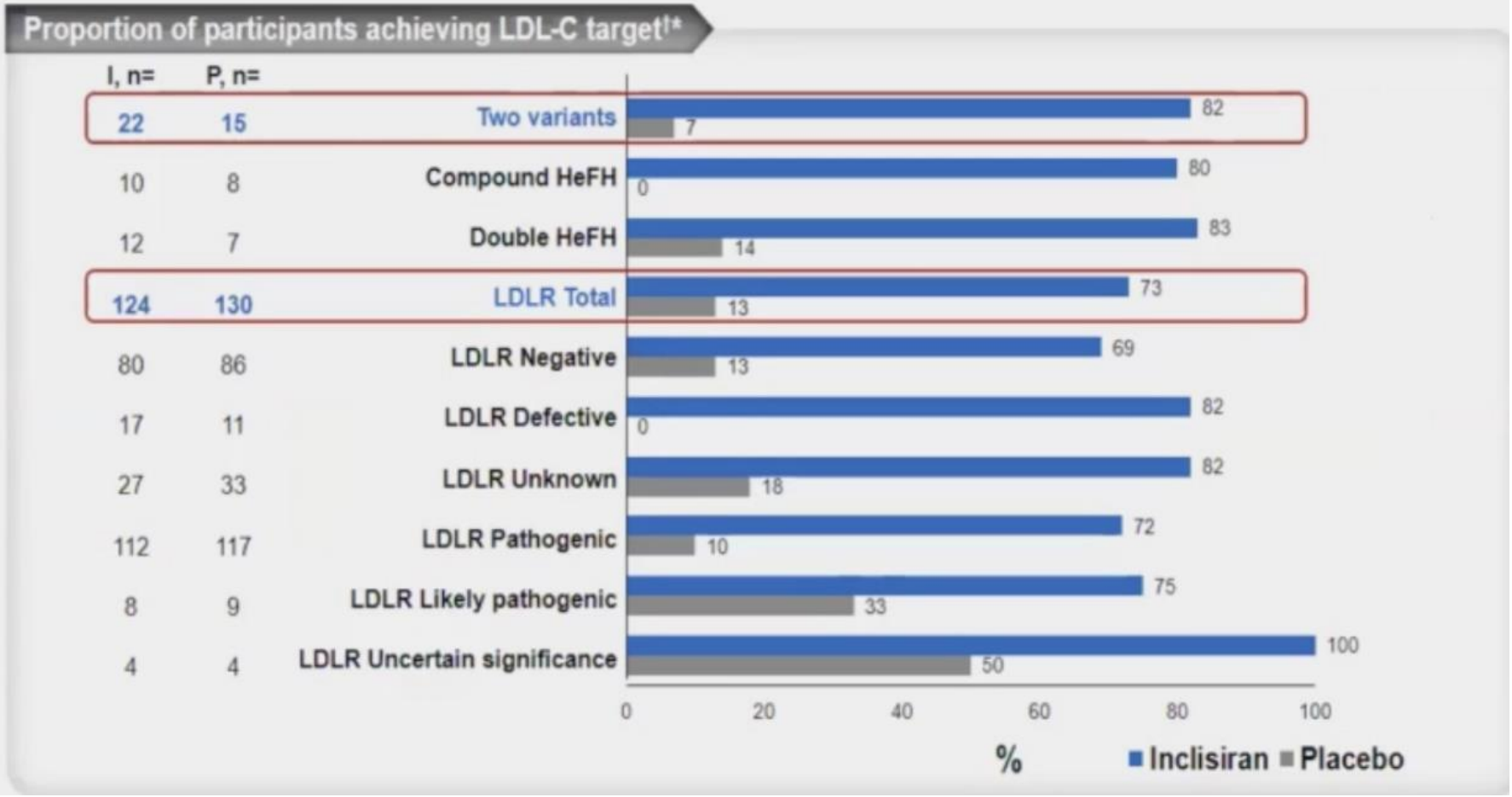


Figure 1. Percent and Absolute Changes in Low-Density Lipoprotein (LDL) Cholesterol and PCSK9 Levels during the 540-Day Trial Period (Intention-to-Treat Population).

Raal FJ et al, NEJM 2020

ORION-9: Efficacy

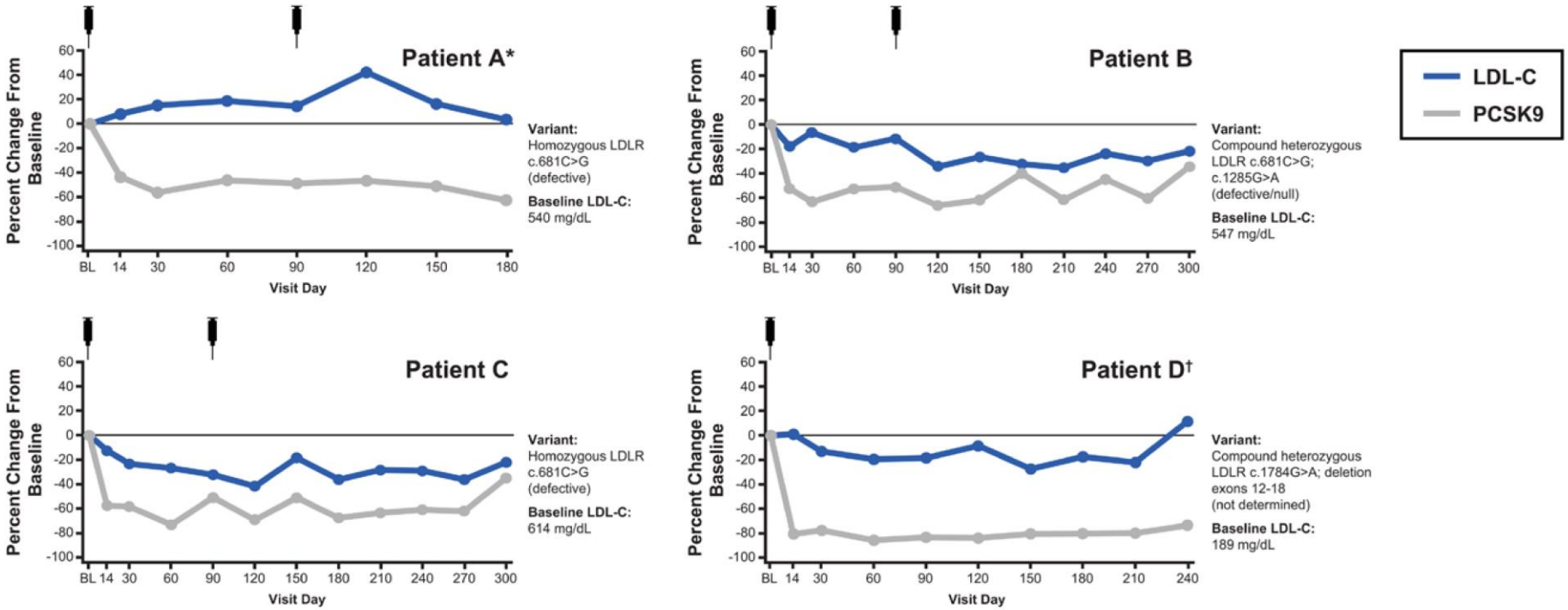
A greater proportion of participants achieved LDL-C targets with inclisiran *vs* placebo



RESEARCH LETTER

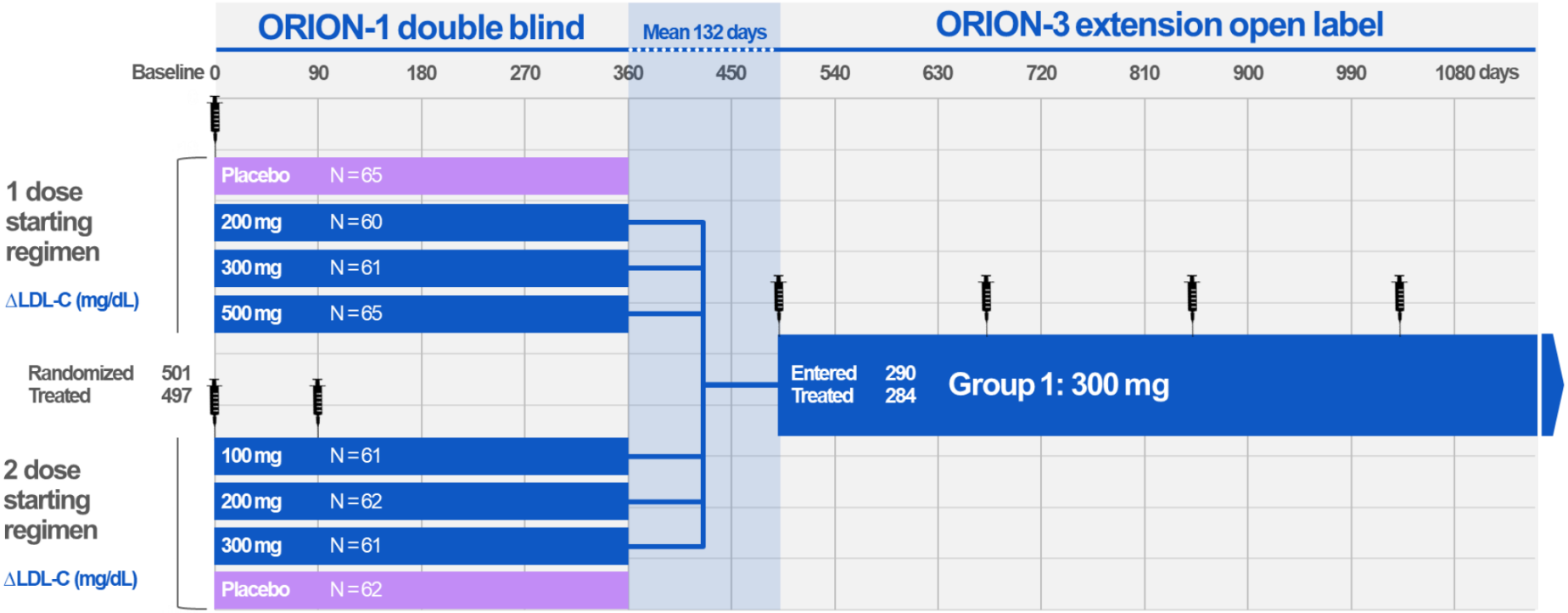
Inclisiran Durably Lowers Low-Density Lipoprotein Cholesterol and Proprotein Convertase Subtilisin/Kexin Type 9 Expression in Homozygous Familial Hypercholesterolemia

The ORION-2 Pilot Study



ORION-3: OLE of ORION 1

Total ~3 years treatment & observation

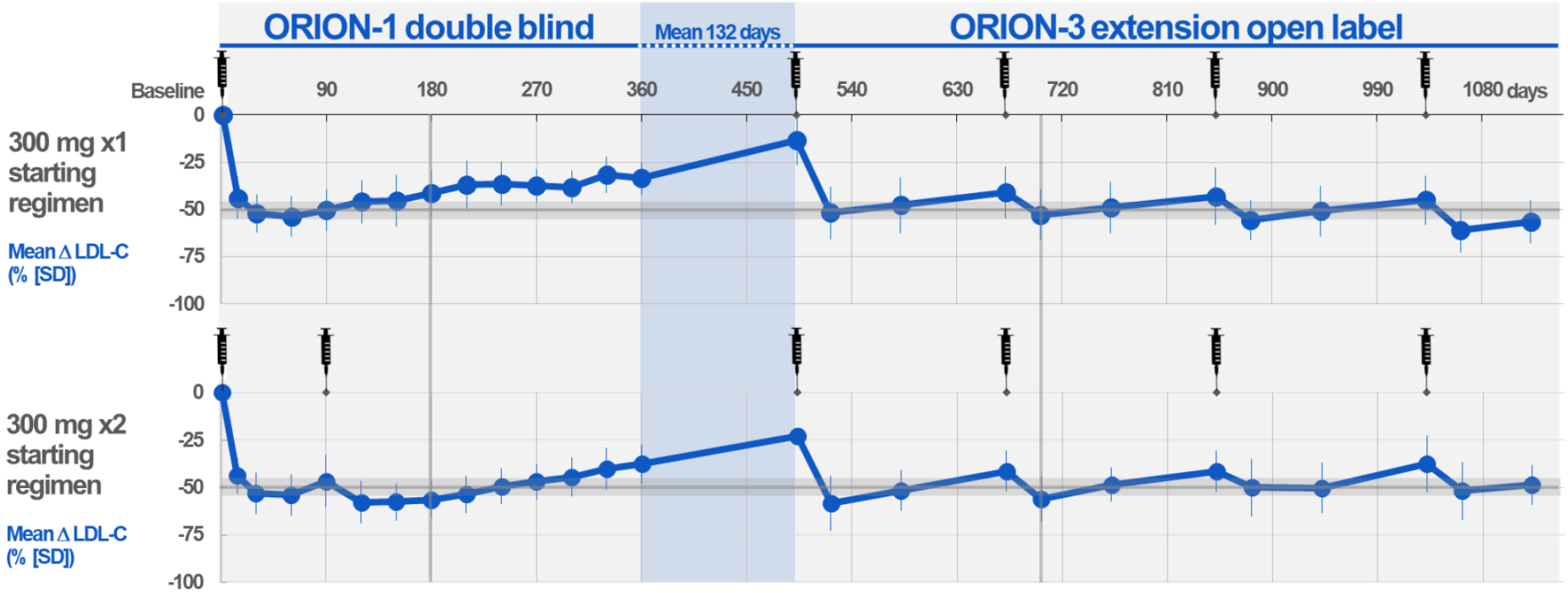


Data Sources: ORION-1 Table 5.10.1.1 final and ORION-3 Table 5.1.1.ORN3.calc ORION-3 09 May 2019

RESULTS

Long-term effect of 300 mg inclisiran on LDL-C

Consistent lowering of LDL-C >50% with no loss of effect over ~3 years



Data Sources: ORION-1 Table 5.10.1.1 final and ORION-3 Table 5.1.1.ORN3.calc ORION-3 09 May 2019

Inclisiran: Safety (Phase III: ORION 9, 10 and 11)

TABLE 3 Treatment-Emergent Adverse Events and Key Safety Findings*

	Inclisiran (n = 1,833)	Placebo (n = 1,822)	Risk Ratio (95% CI)
TEAE			
≥1 TEAE	1,430 (78.0)	1,409 (77.3)	1.01 (0.97–1.04)
≥1 TEAE leading to drug discontinuation	45 (2.5)	35 (1.9)	1.28 (0.83–1.98)
Serious TEAE			
≥1 serious TEAE	374 (20.4)	419 (23.0)	0.89 (0.78–1.00)
Death	27 (1.5)	27 (1.5)	0.99 (0.59–1.69)
New, worsening, or recurrent cancer	44 (2.4)	49 (2.7)	0.89 (0.60–1.33)
Clinically relevant TEAE at the injection site†			
Any reaction	91 (5.0)	12 (0.7)	7.54 (4.14–13.71)
Mild	67 (3.7)	11 (0.6)	6.05 (3.21–11.42)
Moderate	24 (1.3)	1 (0.1)	23.86 (3.23–176.15)
Severe	0 (0.0)	0 (0.0)	–
Persistent	0 (0.0)	0 (0.0)	–
Liver function			
Alanine aminotransferase >3× ULN	9 (0.5)	7 (0.4)	1.28 (0.48–3.42)
Aspartate aminotransferase >3× ULN	8 (0.4)	10 (0.5)	0.80 (0.31–2.01)
Alkaline phosphatase >2× ULN	8 (0.4)	5 (0.3)	1.59 (0.52–4.85)
Bilirubin >2× ULN	14 (0.8)	14 (0.8)	0.99 (0.48–2.08)
Kidney function: creatinine >2 mg/dl	36 (2.0)	42 (2.3)	0.85 (0.55–1.32)
Muscle: creatine kinase >5× ULN	24 (1.3)	22 (1.2)	1.08 (0.61–1.93)
Hematology: platelet count <75 × 10^9/l	1 (0.1)	2 (0.1)	0.50 (0.05–5.48)

Wright RS et al, JACC 2021

ESC European Society of Cardiology
Cardiovascular Research (2021) **117**, 24–26
doi:10.1093/cvr/cvaa139

Insights from ORION studies: focus on inclisiran safety

Alberico Luigi Catapano[1,2]*, Angela Pirillo[2,3], and Giuseppe Danilo Norata[1,3]*

Table 1 **Summary of safety parameters in Orion clinical trials**

	ORION-1			ORION-9		ORION-10		ORION-11	
Platelet count	Platelet count <75 000/μL			Platelet count <75 000/μL		Platelet count <75 000/μL		Platelet count <75 000/μL	
	Single dose								
	Placebo	0/65 (0%)		Placebo	1/240 (0.4%)	Placebo	0/778 (0%)	Placebo	1/804 (0.1%)
	Inclisiran	0/186 (0%)		Inclisiran	0/241 (0%)	Inclisiran	1/781 (0.1%)	Inclisiran	0/811 (0%)
	Double dose								
	Placebo	0/62 (0%)							
	Inclisiran	0/184 (0%)							
ADA positive samples	0.03% (2 samples from 1 patient, total samples 6068)			2.6% (25 samples from 18 patients)		2.0%		2.5%	
Inflammation	Change median, % (interquartile range)								
		TNF-α	IL-6						
	Single dose								
	Placebo	-4.6 (-14.1 to 7.2)	-1.7 (-16.7 to 38.8)						
	Incl. 200 mg	-0.2 (-7.8 to 13.5)	6.0 (-7.5 to 22.1)						
	Incl. 300 mg	-1.9 (-8.7 to 9.1)	3.1 (-15.9 to 32.7)						
	Incl. 500 mg	1.1 (-8.7 to 11.7)	4.2 (-35.5 to 21.8)						
	Double dose								
	Placebo	-0.9 (-6.9 to 14.7)	-21.3 (-30.8 to 23.8)						
	Incl. 100 mg	-0.6 (-11.9 to 13.7)	-12.3 (-28.0 to 16.7)						
	Incl. 200 mg	1.3 (-6.3 to 12.1)	14.8 (-12.8 to 46.6)						
	Incl. 300 mg	2.7 (-8.3 to 15.5)	15.9 (0.3 to 46.1)	NA		NA		NA	

NA, not available.

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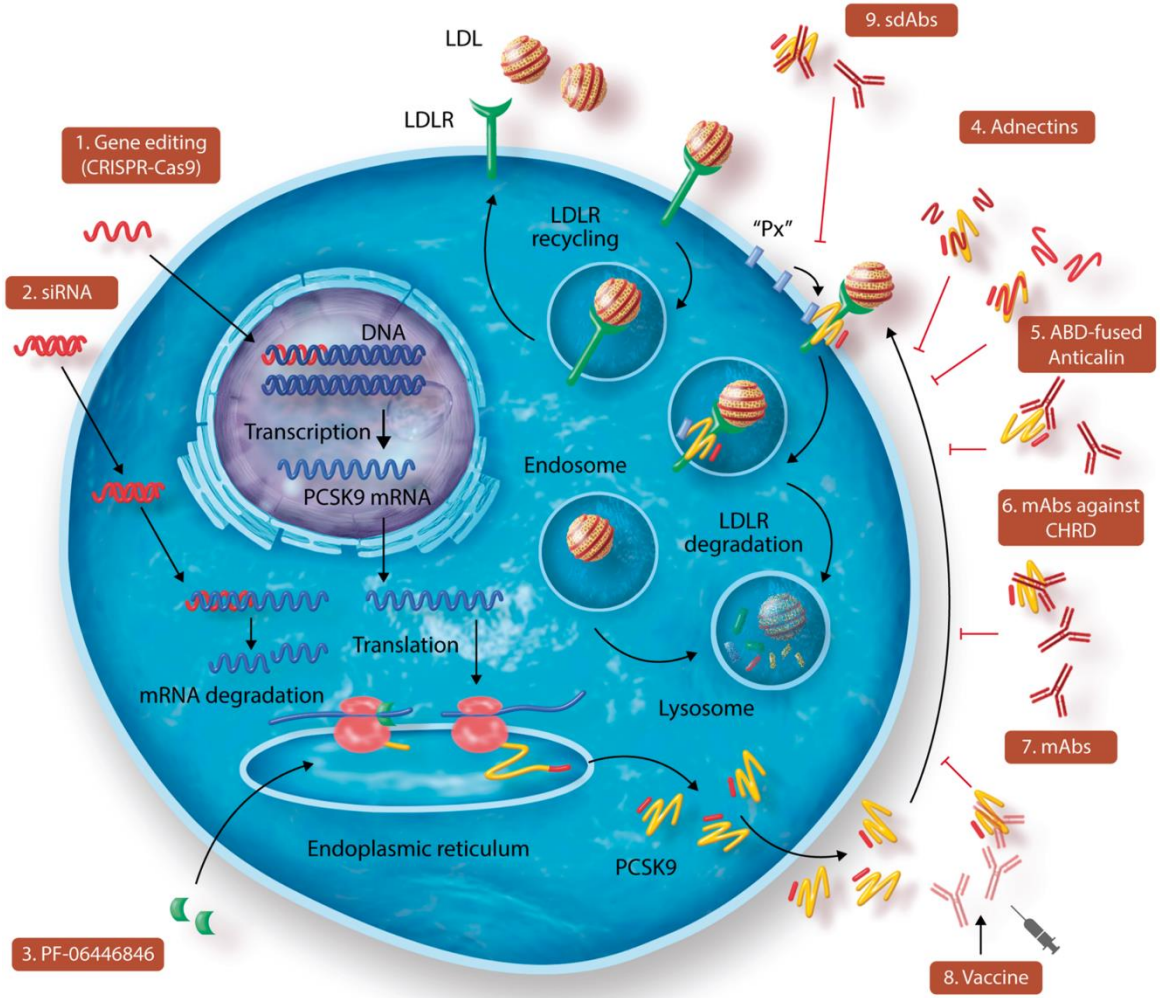
Modulators of LDLR half-life: PCSK9 inhibitors

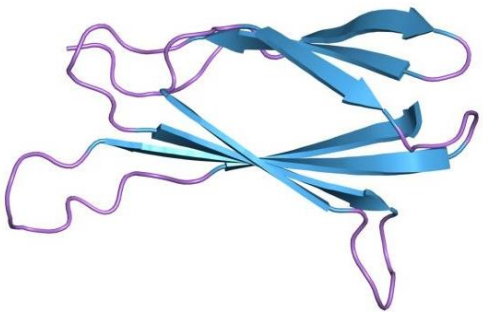
Targeting PCSK9 beyond monoclonal antibodies

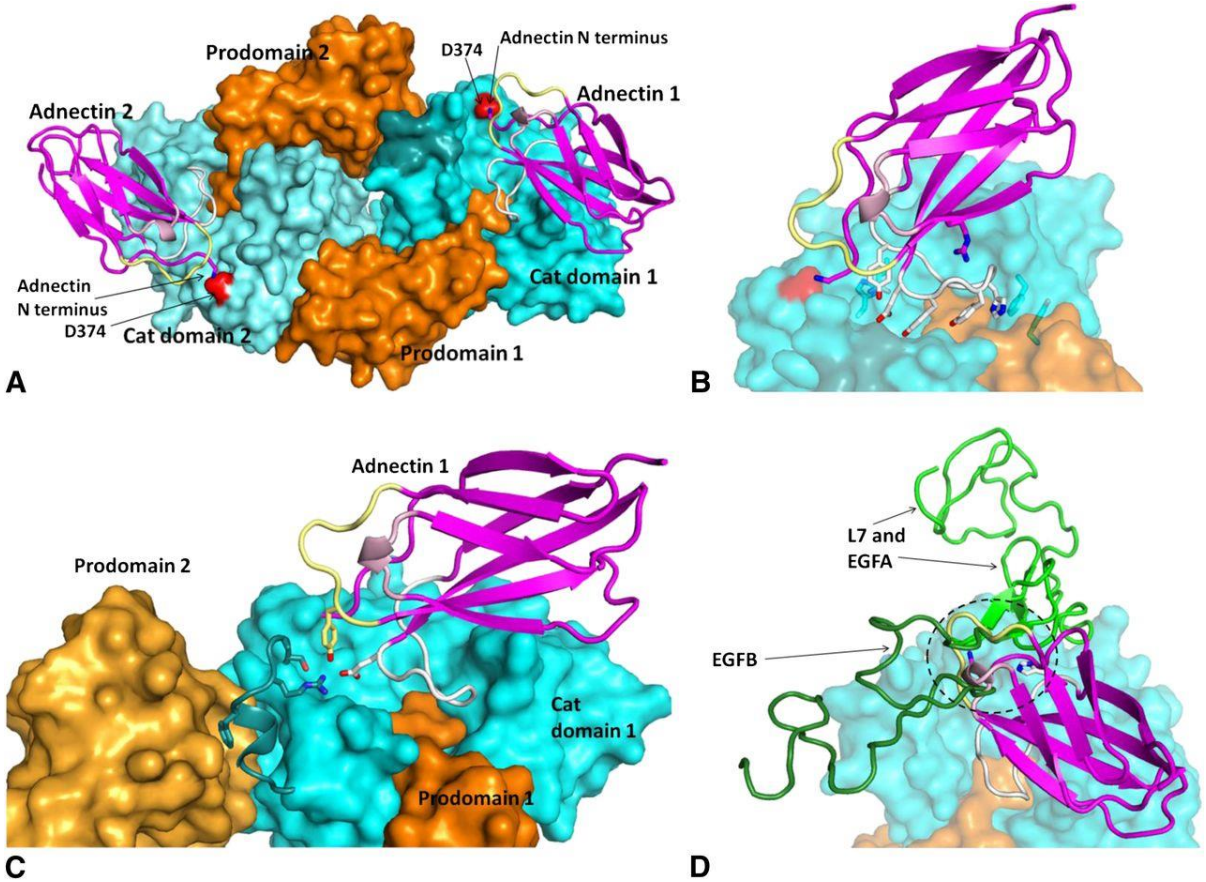


Seidah N, Norata GD. Cardiovasc Res 2019; 115:510-518.

Adnectins anti PCSK9:LIB003 Phase II

Scaffold proteins





Pharmacokinetics aspects of scaffold proteins compared to mAbs

ADVANTAGES

Small MW (10-20Kd) vs

Increased solubility

Increased stability

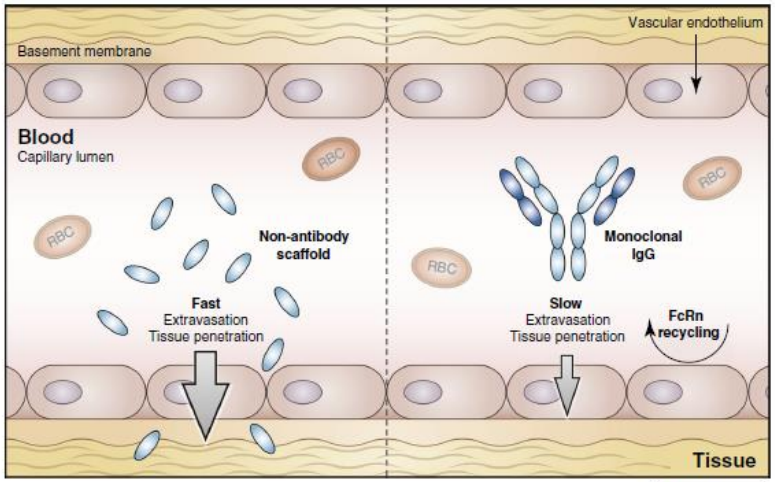
- **Elevated tissue distribution**
- **Simple to produce (Escherichia Coli)**
- **Low cost**

DISADVANTAGES

Short half life (PEG or albumin BD conjugation)

Immunogenicity

MoA similar to mAb

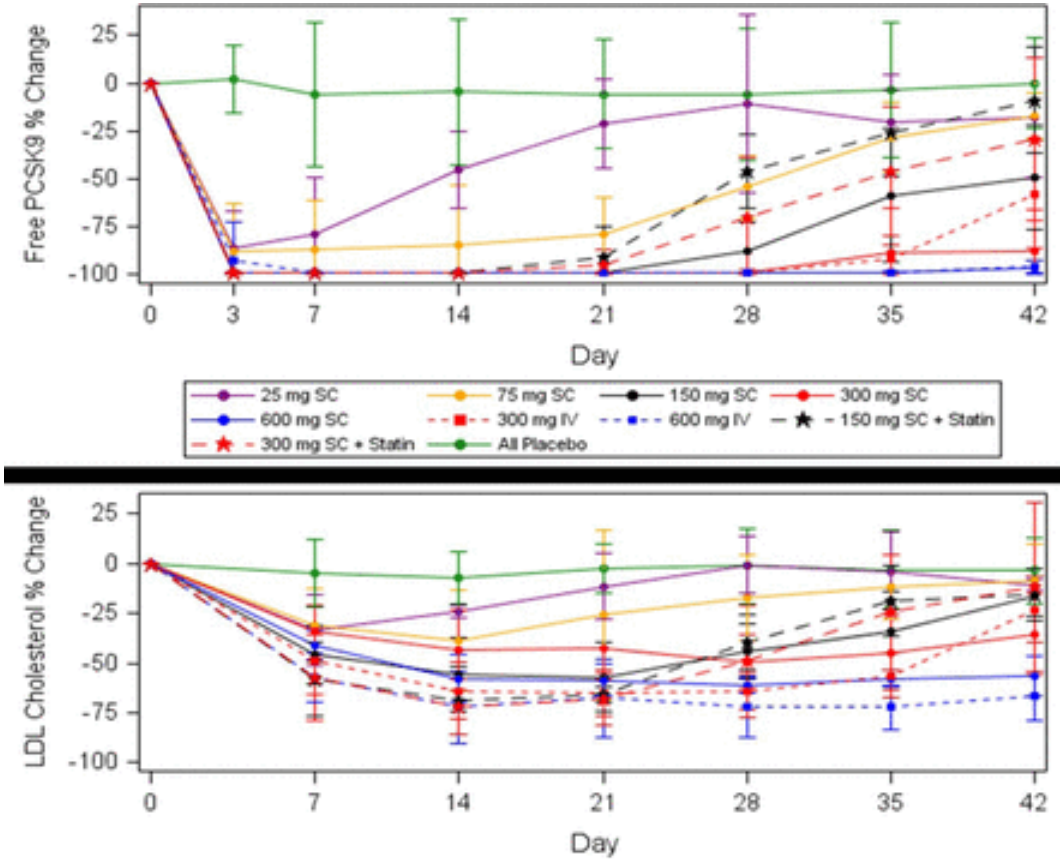


Drug Discovery Today

Adnectins anti PCSK9:LIB003

LIB003 (LIB) is a recombinant fusion protein of a PCSK9-binding domain (adnectin) and human serum albumin bind domain which offers small injection volumes for monthly (Q4W) dosing.

Placebo (PBO)-controlled, double-blind, single ascending dose study of 9 cohorts with 7 subjects per cohort (5 LIB003:2 PBO). Cohorts 1-7 were on diet alone and cohorts 8-9 on a statin. Subjects were in-patients for 72 hours post dose and followed as out-patients on days 5, 8, 11, 15, 18, 22, 29, 36 and 43. Randomization to next cohort dose commenced after safety review at day 8 of ≥5 subjects in a cohort.



Adnectins anti PCSK9:LIB003

Conclusion: LIB 300 mg dosed QM in 1.2 mL was well tolerated, safe and provided sustained PCSK9 with LDL-C reductions >60%. In 1 patient ADAs exceeded the level of assay sensitivity recommended by FDA with no impact on efficacy

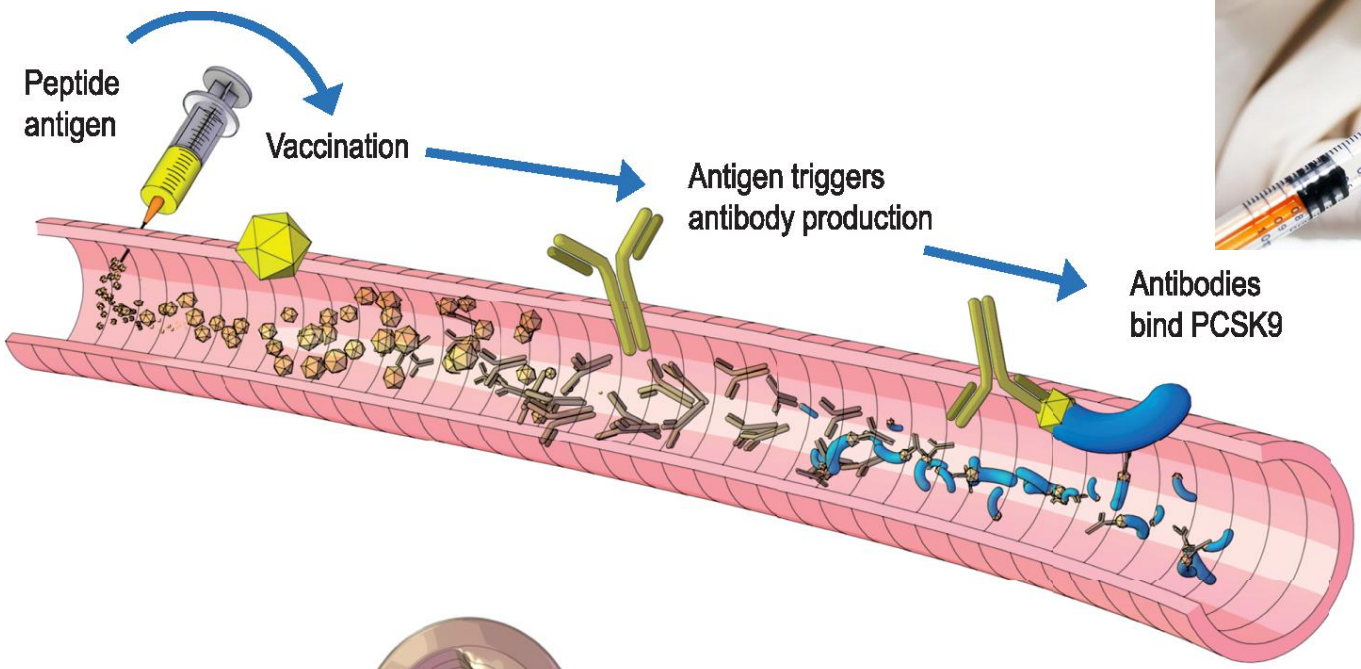
Phase II

Patients; mean (range) 62.5 (31-80) years, 62.5% female, 78% white, CVD risk (28% ASCVD; 47% diabetic, 66% HTN, 31% FH)
Lipid Therapy 84% on statin and 25% ezetimibe

	Baseline	week 4	week 8	week 12	week 16	week 20	week 24	week 28	week 32
LDL-C mg/dL ¥	124.3	54.8	53.9	52.1	55.4	53.4	56.0	51.3	48.9
ITT analysis n	32	32	31	31	30	30	27	29	29
% change - mean	n/a	-57.2	-59.2	-59.0	-57.2	-58.9	-57.5	-61.9	-62.1
SD	n/a	16.65	17.47	18.55	18.57	22.92	22.17	20.86	20.21
PP analysis n	32	32	30	31	28	27	24	24	28
% change - mean	n/a	-57.2	-60.0	-59.0	-58.3	-61.7	-61.1	-65.7	-63.4
SD	n/a	16.65	17.23	18.55	18.75	20.25	20.84	17.95	19.30
Free PCSK9 ng/mL¥	260.9	45.5	50.0	45.5	42.5	40.9	71.0		
ITT analysis n	32	32	31	31	29	30	28		
% change - mean	n/a	-83.0	-81.4	-84.2	-85.2	-83.7	-75.8		
SD	n/a	24.80	26.56	23.38	21.57	25.20	32.69		
PP analysis n *	32	32	30	31	28	27	25		
% change - mean	n/a	-83.0	-83.0	-84.2	-85.0	-86.1	-82.6		
SD	n/a	24.80	25.36	23.38	21.96	25.29	25.36		
Lp(a) nmol/L ¥	131.8			94.2			90.1		
ITT analysis n	32			31			28		
% change - mean	n/a			-26.2			-29.1		
SD	n/a			22.54			18.00		
Apo B mg/dL ¥	99.8			54.2			57.7		
ITT analysis n	32			31			28		
% change - mean	n/a			-46.1			-44.2		
SD	n/a			14.59			18.52		

ITT= intent to treat; PP=per protocol dosing 28±3 days; ¥mean

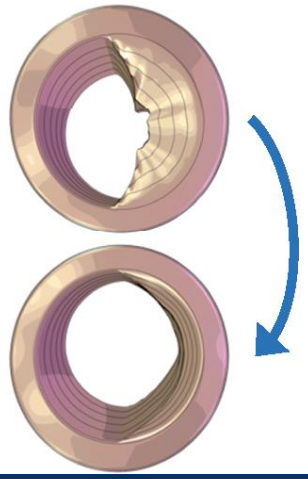
SAIT (specific active immunotherapy anti-PCSK9)



PCSK9 ↓

LDL-R ↑

Serum LDL-C ↓



Atherosclerotic plaque ↓

-New frontiers in hypercholesterolaemia

PCSK9 (Beyond Monoclonal antibodies)

ACLY (Bempedoic acid)

ANGPTL3 (Evinacumab)

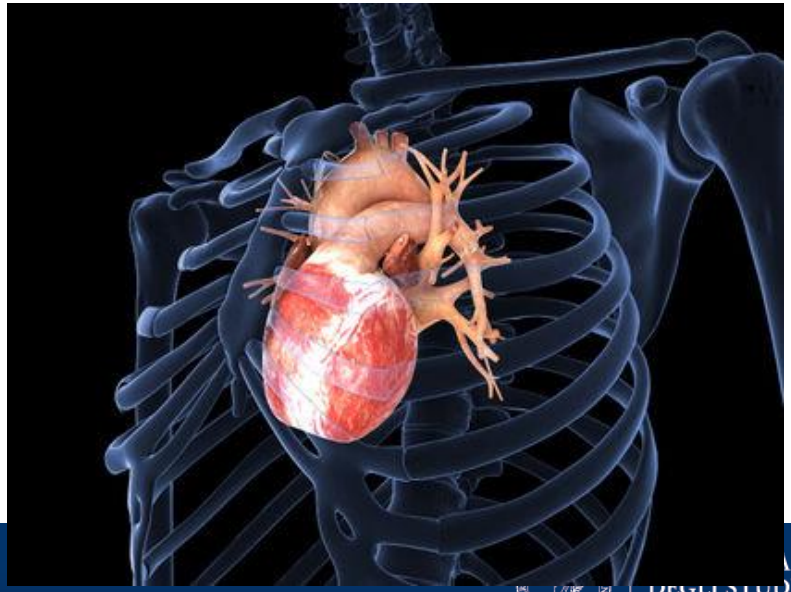
-Targeting tryglicerides rich lipoproteins

Icopentanyl EPA

APO CIII (Volanesorsen)

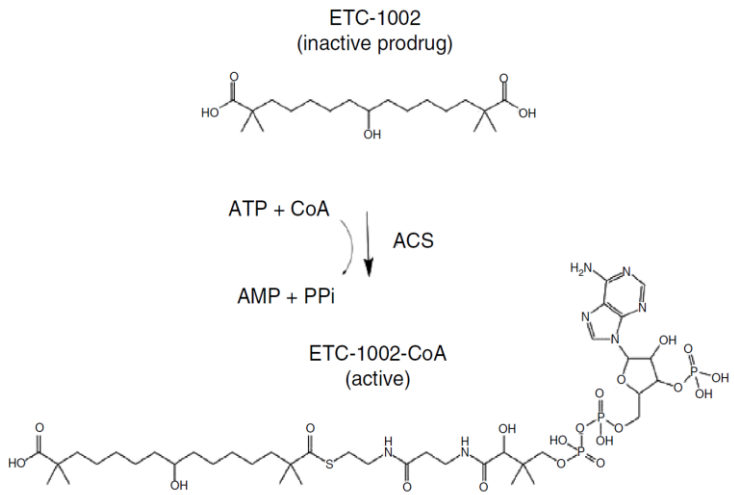
-Targeting Lp(a)

Apo(a) Gene silencing

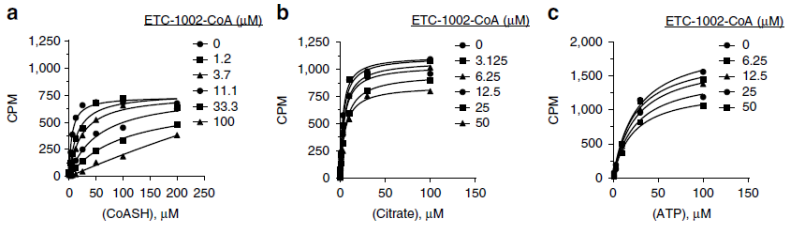


Bempedoic Acid: mechanism of action

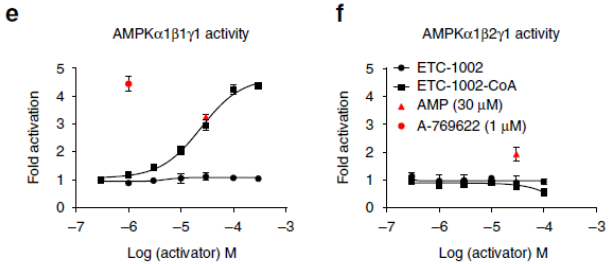
Prodrug



ATP citrate lyase (ACLy) inhibitor



AMPK beta1 activator

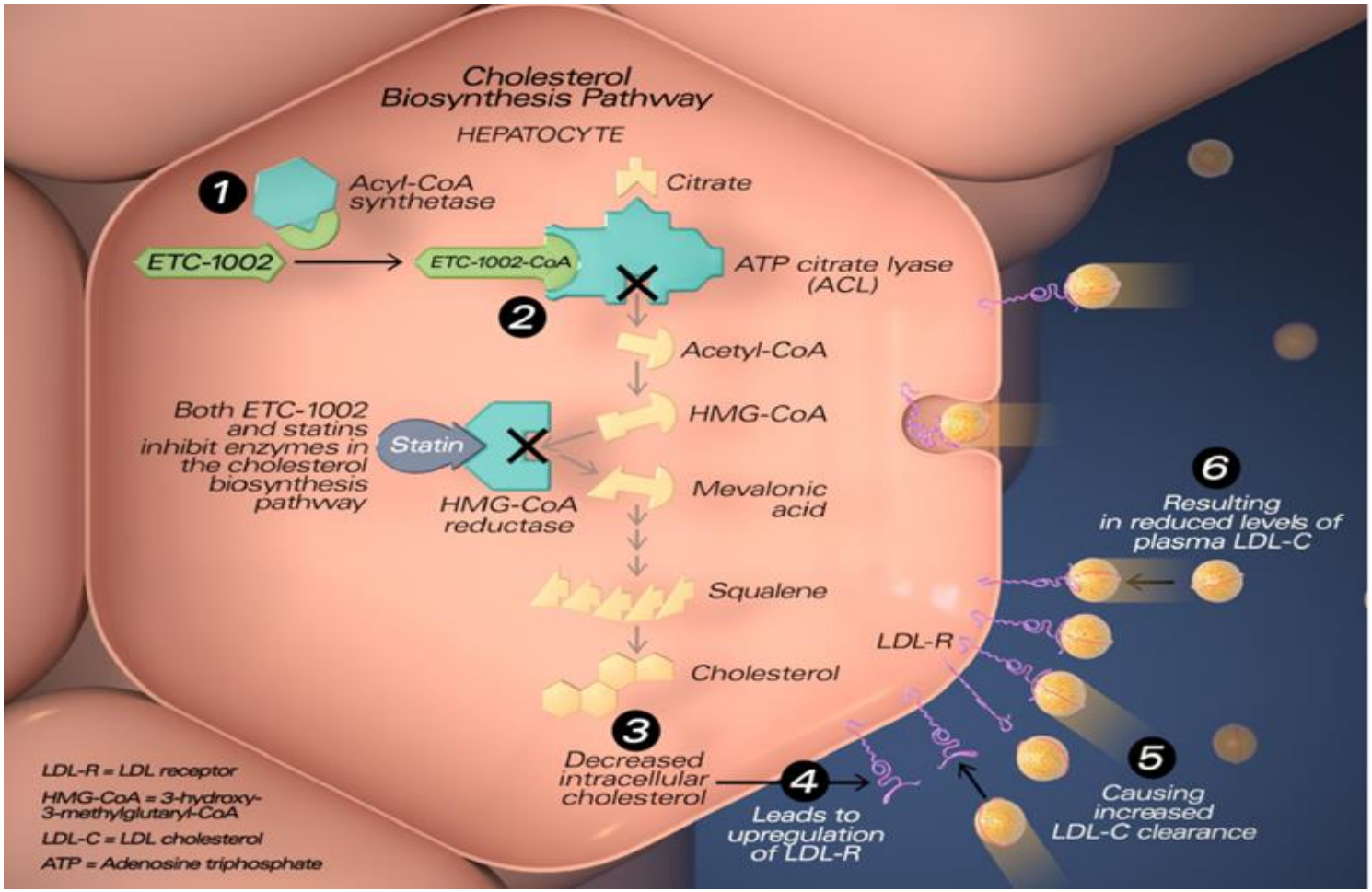


ACSVL1:Very long chain acyl-coA synthase

Pinkosky SL, et al. Nat Commun. 2016 Nov 28;7:13457.

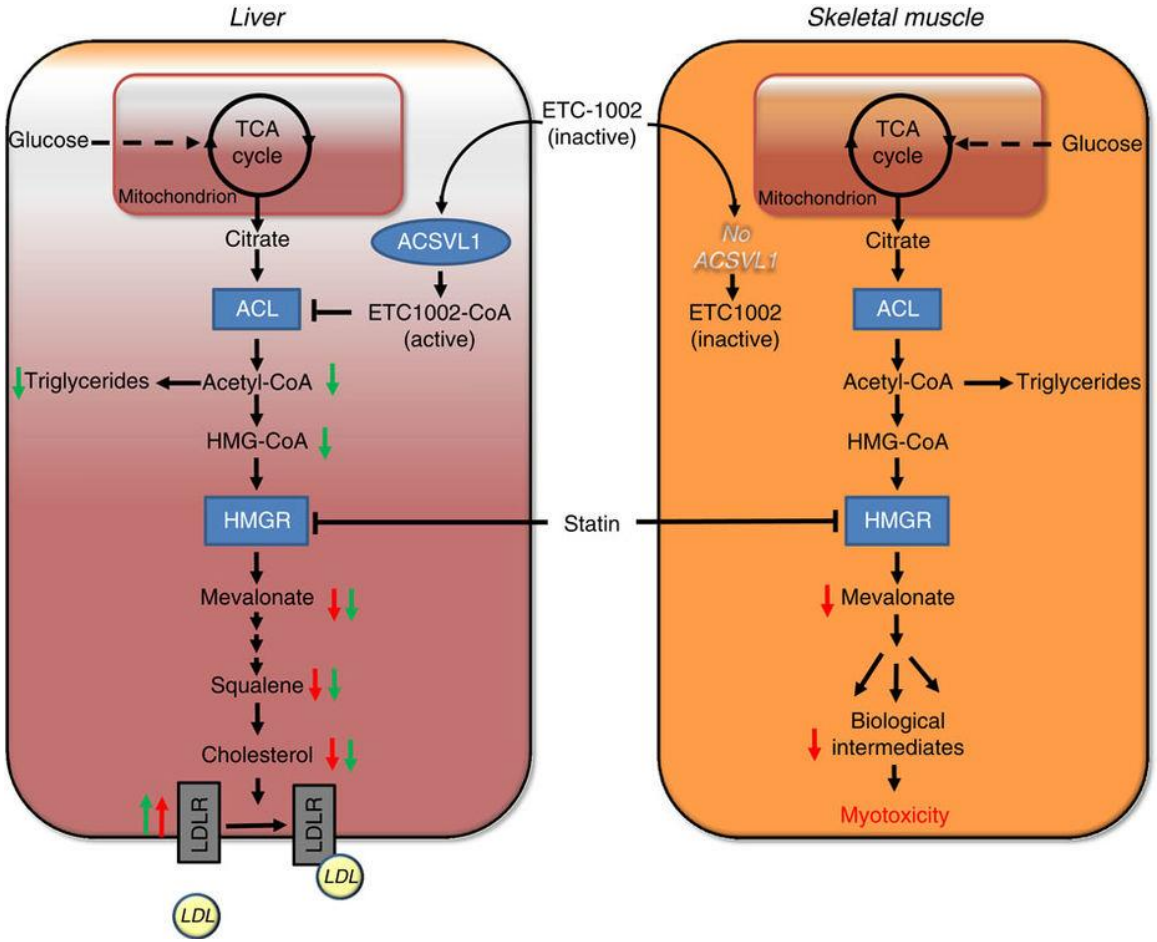
Bempedoic Acid: mechanism of action

ATP citrate lyase (ACLy) inhibition



Liver selectivity of Bempedoic Acid

Prodrug activation requires the activity of very long chain fatty acids synthase



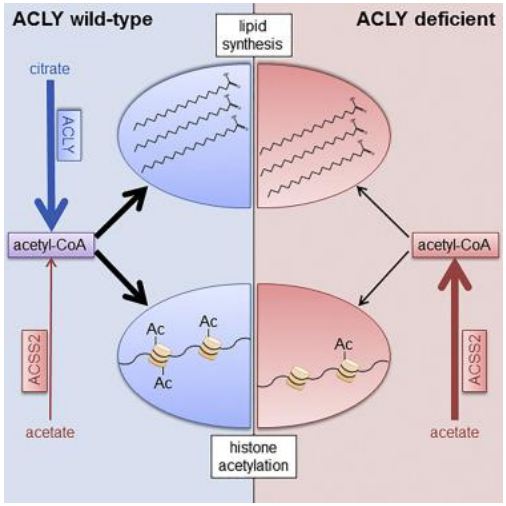
Pinkosky SL, et al. Nat Commun. 2016 Nov 28;7:13457.

Pharmacology of Bempedoic Acid

-Potent inhibition of ATP citrate lyase in the liver resulting in decreased LDL-C levels

-Activation of AMPK (not involved in lipid lowering in animal models).

-No active drug produced in smooth muscle cells.

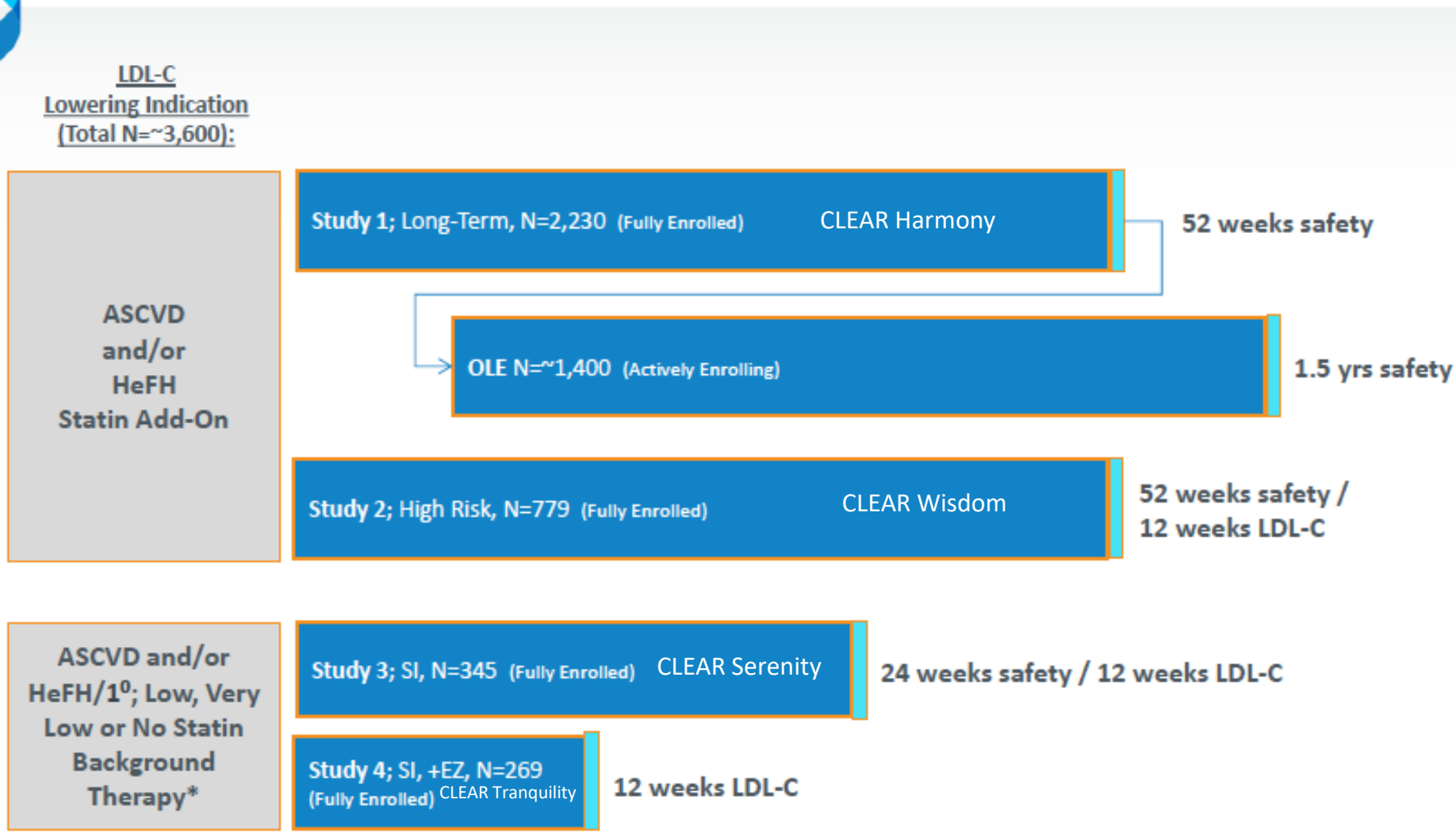


LDL-C reduction in clinical trials

Table 1. Published phase 2 and 3 clinical studies of bempedoic acid

Reference	Patient population	Participants (treated/placebo)	Duration (weeks)	Dose (mg/day)	LDL-C reduction (%)[a]
Phase 2					
[40]	Hypercholesterolemia	133/44	12	40, 80, 120	Up to 27%
[41]	Hypercholesterolemia and type 2 diabetes	30/30	4	80, 2 weeks; 120, 2 weeks	43%
[14]	Hypercholesterolemia and hypertension	72/71	6	180	21%
[42]	Hypercholesterolemia and on stable statin therapy	88/46	12	120, 180	Up to 24%
[43]	Hypercholesterolemia and history of statin-intolerance	37/19	8	60, 120, 180, 240	32%
[44]	Hypercholesterolemia with or without statin-intolerance, compared to ezetimibe	249/100	12	120, 180 (ezetimibe 10)	BemA Up to 30% BemA + ezetimibe Up to 48%
Phase 3					
[45▪▪]	Hypercholesterolemia and history of statin-intolerance, BemA + ezetimibe + low-dose statin compared to ezetimibe + low-dose statin	181/88	12	BemA 180, ezetimibe 10	29%
[46]	ASCVD and/or Het FH on maximally tolerated statin therapy	1487/742	24	BemA 180	20%

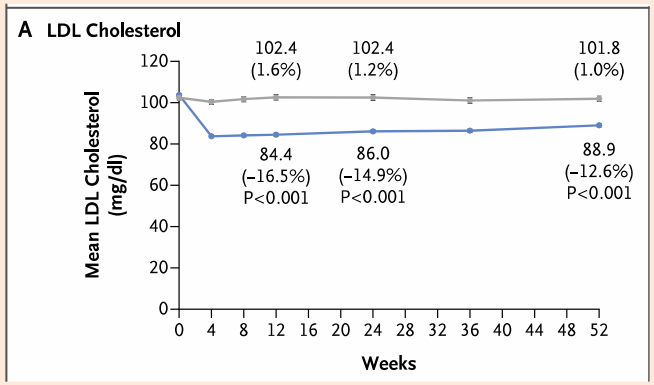
Bempedoic Acid Phase 3 Program for LDL-C Lowering

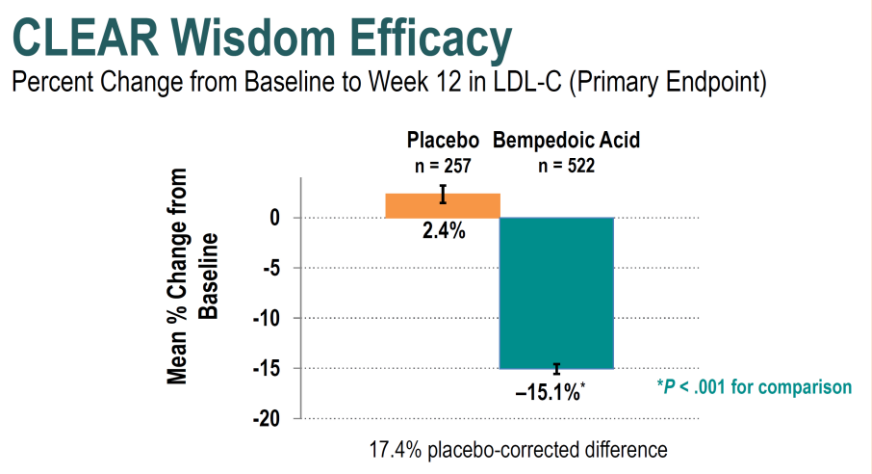


*Studies are being conducted to obtain an indication for use in patients on no background statin therapy in Europe

Summary of bempedoic acid phase III studies

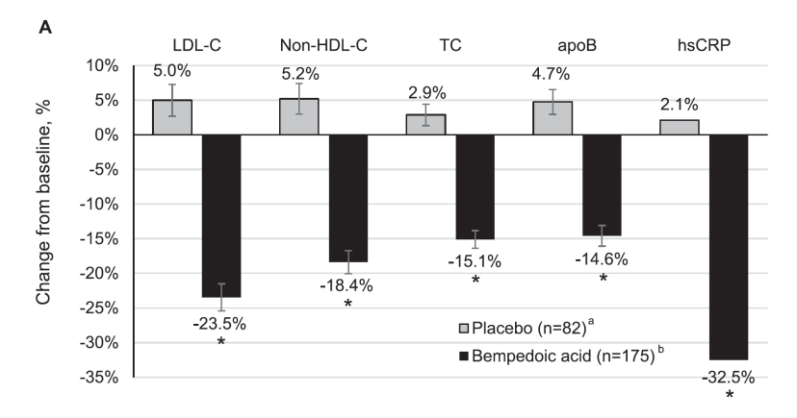
CLEAR Harmony





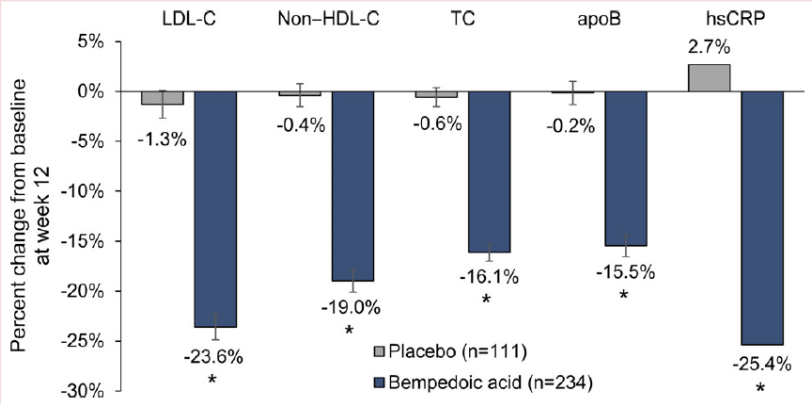
On top of max tolerated dose of statins

CLEAR Tranquillity



+ ezetimibe

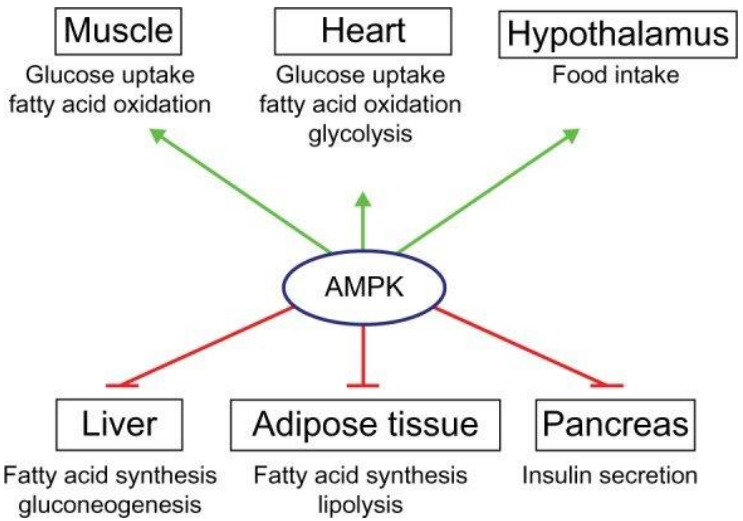
CLEAR Serenity

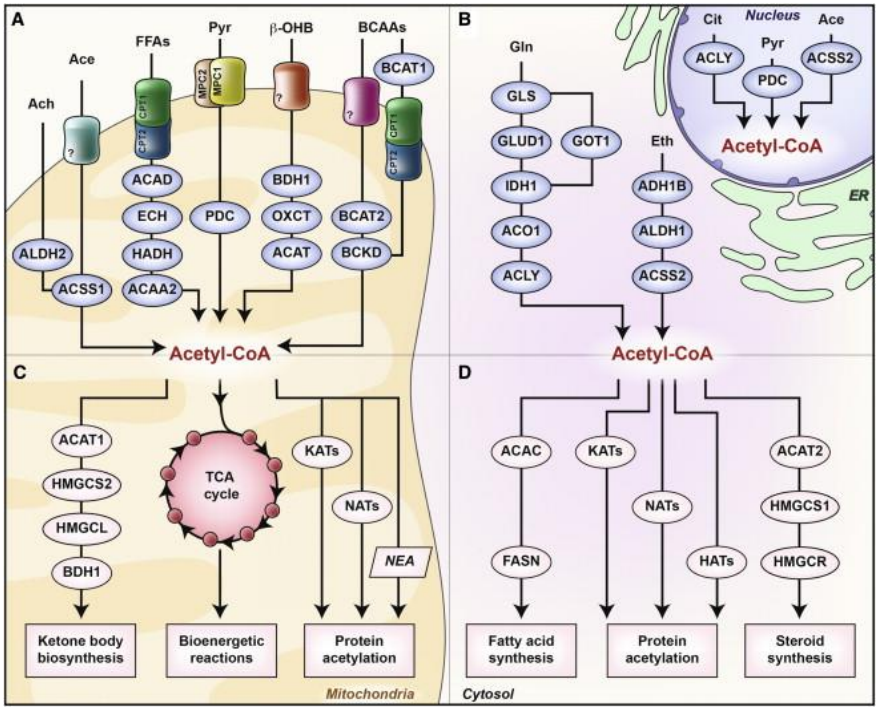


Statin intollerant

Open questions:

Clinical impact of AMPK activation?

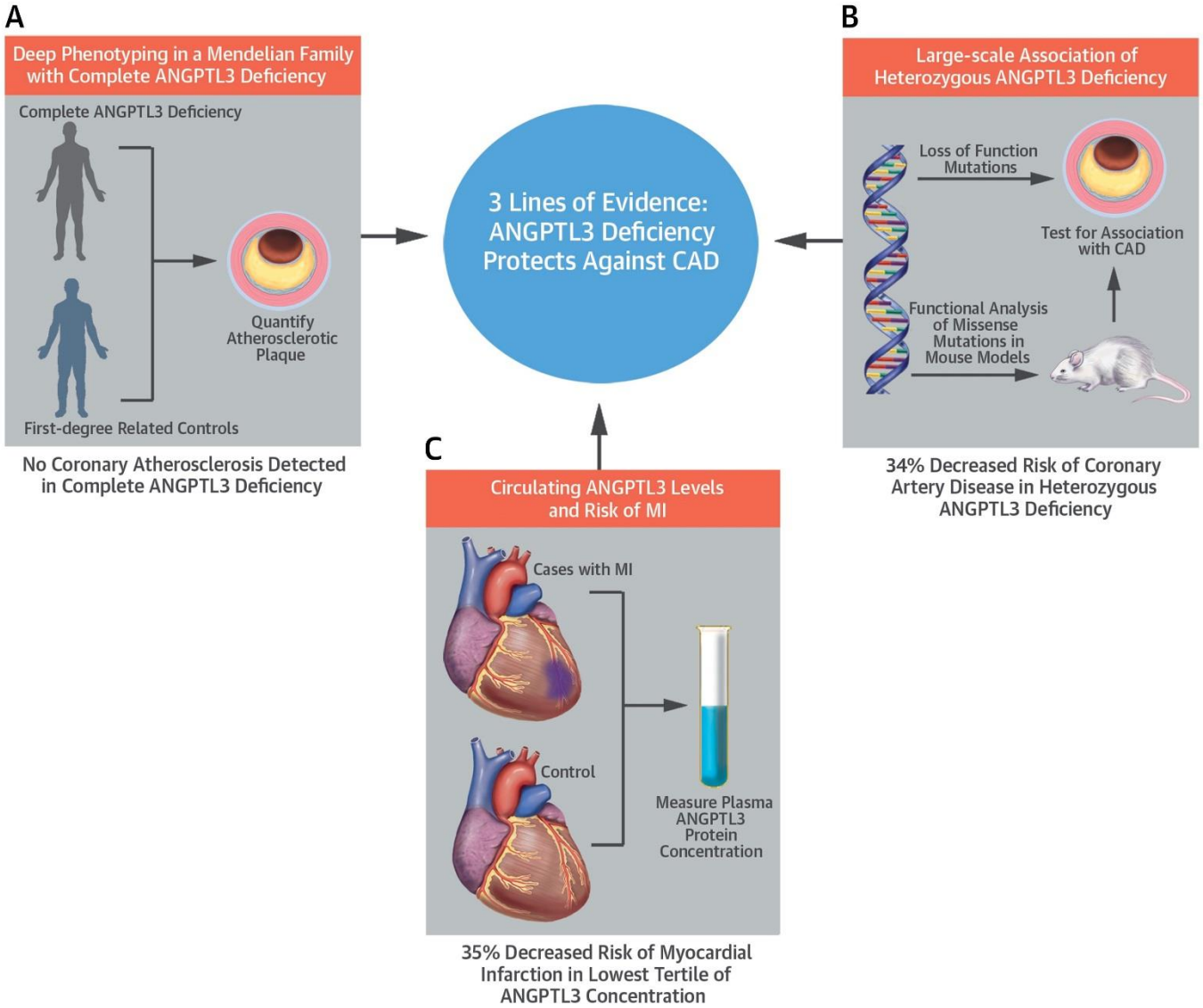




Clinical implication of acetyl-CoA production inhibition beyond the mevalonate pathway?

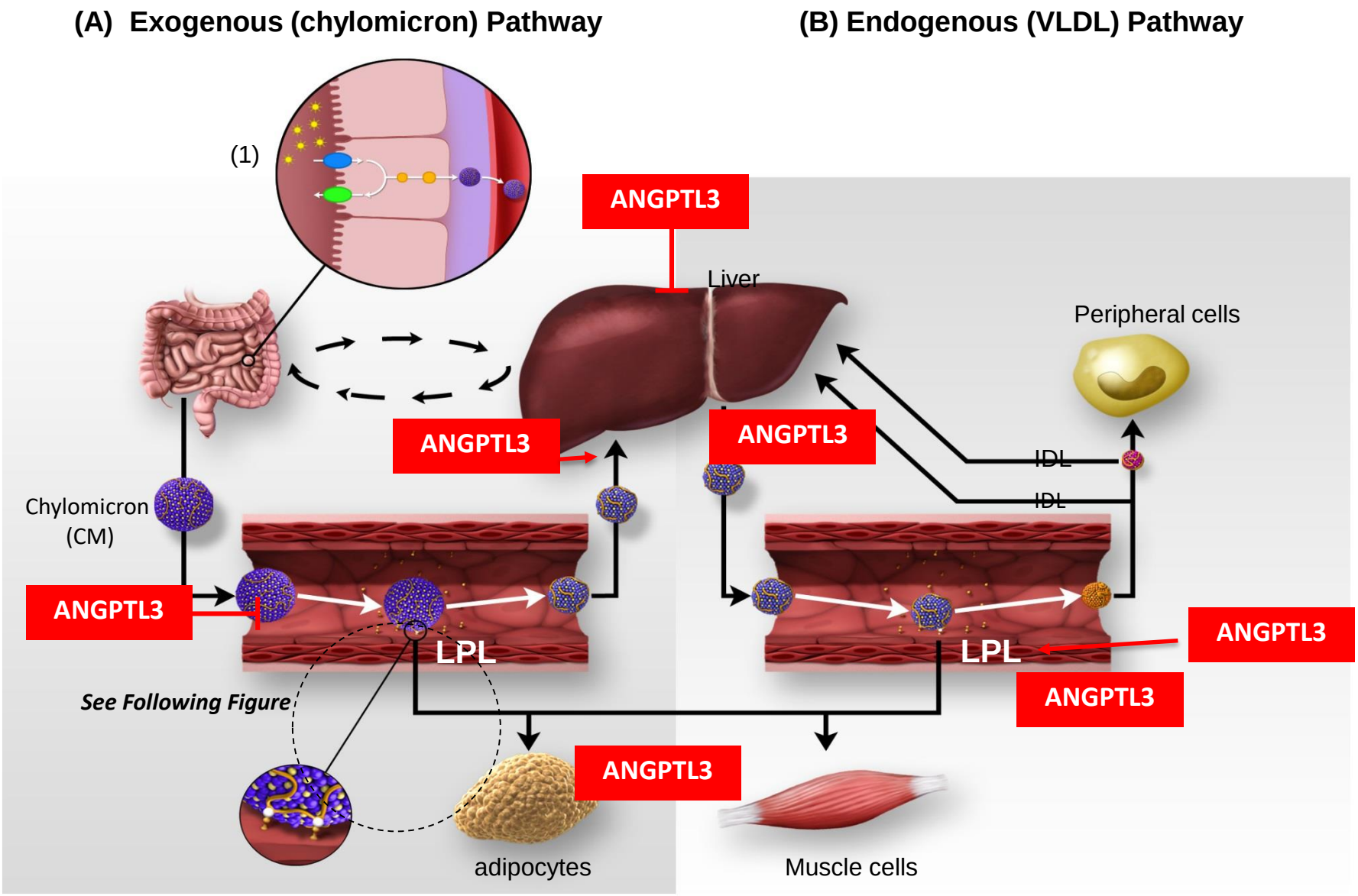
ANGPTL3
(angiopoietin-like 3 protein)

CENTRAL ILLUSTRATION: ANGPTL3 Deficiency and Protection From Coronary Artery Disease

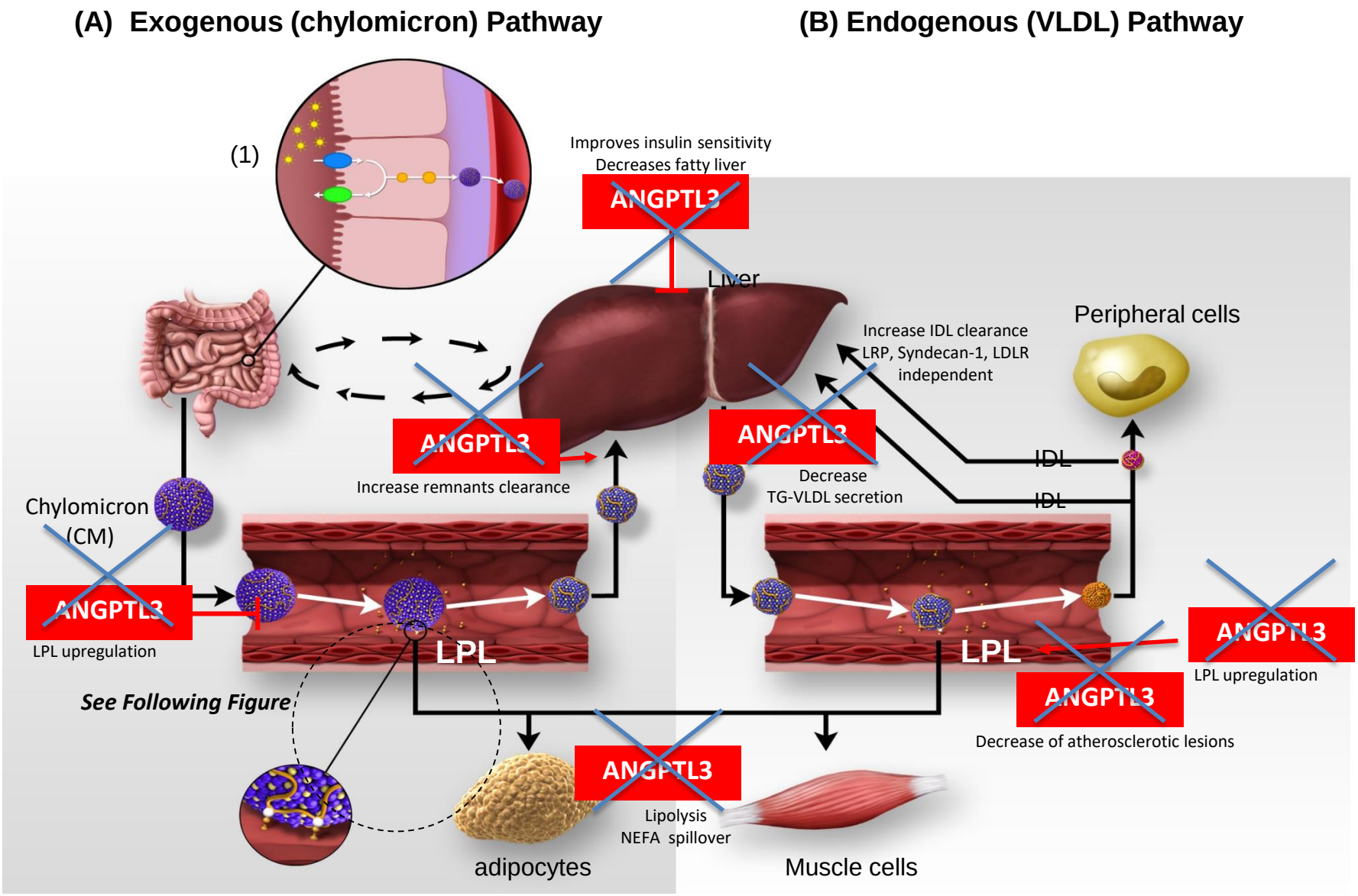


Stitziel, N.O. et al. J Am Coll Cardiol. 2017;69(16):2054–63.

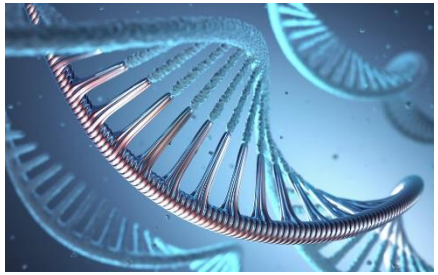
Pre-Clinical and Human Early Phase Studies Suggest that Several Mechanisms are Involved in the Lipid Lowering Effect of ANGPTL3 Blockade



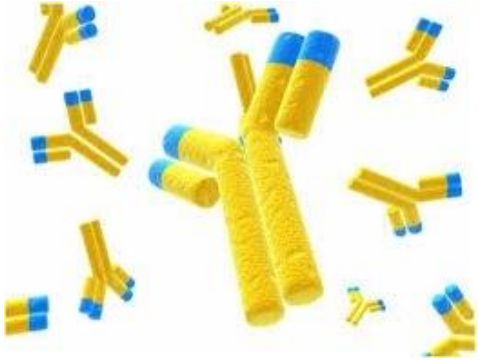
Pre-Clinical and Human Early Phase Studies Suggest that Several Mechanisms are Involved in the Lipid Lowering Effect of ANGPTL3 Blockade



Two approaches to target ANGPTL3 in clinical development



Vupanorsen
Antisense oligonucleotide
(ASO)



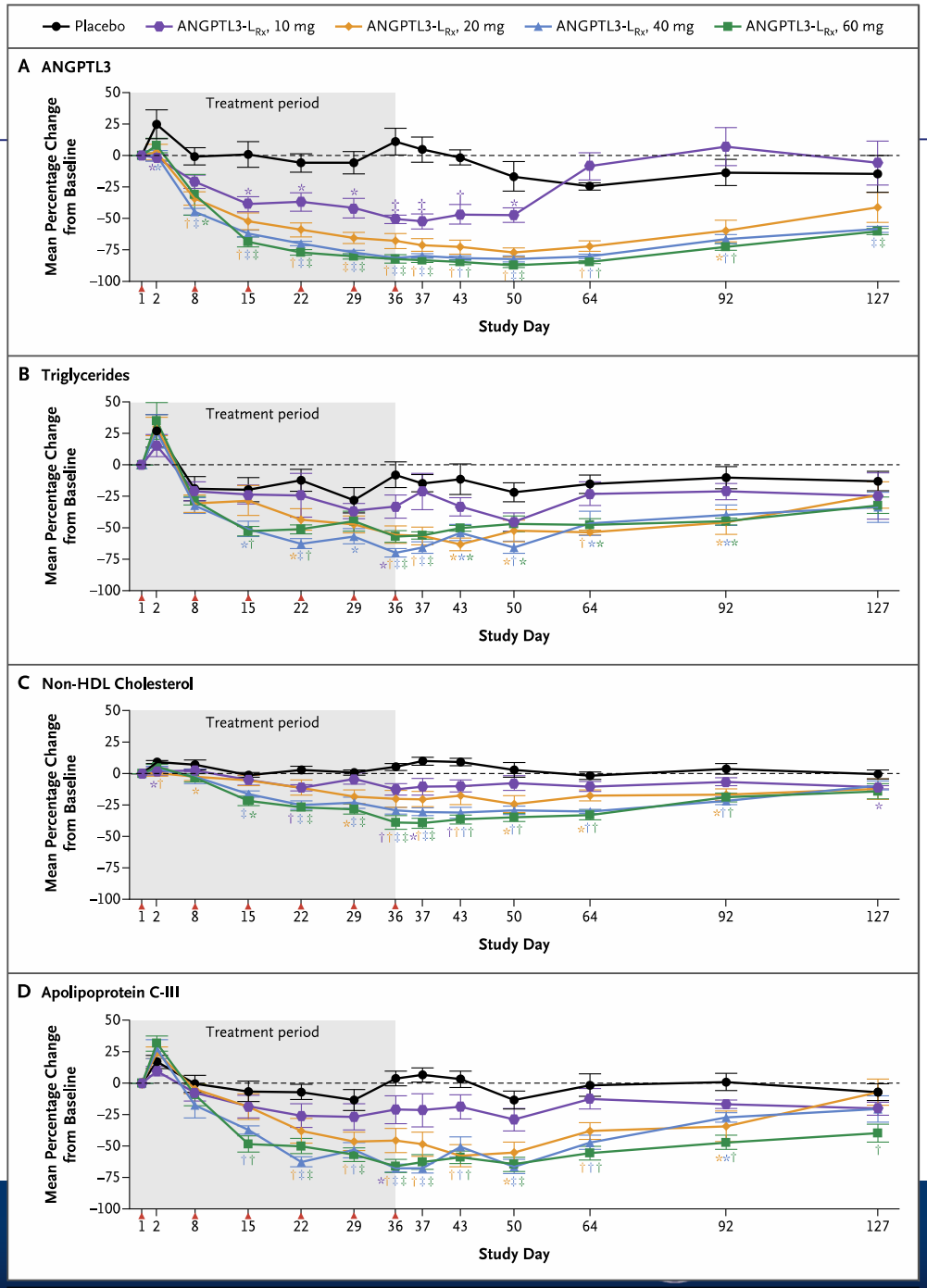
Evinacumab

Figure 2 (facing page). Effect of ANGPTL3-L_{Rx} on Levels of ANGPTL3 Protein, Apolipoprotein C-III, Triglycerides, and Non-HDL Cholesterol in the Multiple-Dose Groups.

The data are the mean percentage changes in levels from baseline; I bars denote standard errors. Red arrowheads indicate the timing of dosing weekly for 6 weeks; the shaded area denotes the dosing duration, and the white area the time of follow-up. Day 43 was used as the therapeutic end point for ANGPTL3 protein and lipid variables that reflects the maximal predicted reduction in levels after the final dose. P values are for the differences in efficacy at day 43 as determined by the exact Wilcoxon rank-sum test comparing ANGPTL3-L_{Rx} and placebo. An asterisk denotes P<0.05, a dagger P<0.01, and a double dagger P<0.001. HDL denotes high-density lipoprotein.

Table 3. Percentage Changes in ANGPTL3, Lipid, and Lipoprotein Levels at Day 43 after Initiation of ANGPTL3-L_{Rx} Treatment in the Multiple-Dose Groups.*

Measure	Placebo (N=8)	ANGPTL3-L_{Rx}			
		10 mg (N=6)	20 mg (N=6)	40 mg (N=6)	60 mg (N=6)
	percentage change from baseline				
ANGPTL3	−1.6±15.4	−46.6±18.5†	−72.5±12.4†	−81.3±4.0†	−84.5±5.1†
Triglycerides	−11.4±31.9	−33.2±17.8	−63.1±10.9‡	−53.8±15.6‡	−50.4±5.9‡
LDL cholesterol	13.6±12.1	−1.3±23.6	−4.3±18.5	−25.4±16.5†	−32.9±10.4†
VLDL cholesterol	−4.0±30.8	−27.9±18.3	−60.0±15.5†	−48.5±14.3†	−48.7±8.5‡
Apolipoprotein B	11.0±7.7	−3.4±17.0	−13.3±15.0†	−25.7±10.8†	−22.2±12.7†
Non-HDL cholesterol	9.1±8.2	−10.0±11.6†	−17.6±16.2†	−31.1±11.0†	−36.6±8.9†
Apolipoprotein C-III	3.1±17.0	−18.9±23.4	−57.8±19.4†	−50.7±19.3†	−58.8±12.3†



Graham MJ *et al. NEJM 2017*

Vupanorsen Produced Significant Reductions of Plasma Triglycerides and Total Cholesterol Levels in Healthy Subjects

ANGPTL3

Up to -93%
(mean 84%)

Triglycerides

Up to -63%
(mean 49%)

Total Cholesterol

Up to -46%
(mean 28%)

Summary Vupanorsen

- Administration of an ANGPTL3 ASO produced dose-dependent reductions in hepatic target mRNA, plasma TG, and cholesterol in preclinical models of hypercholesterolemia and metabolic dyslipidemia.
  - Reductions in plasma TG were independent of apoC-III expression
  - Plasma lipids were reduced by inhibiting hepatic TG secretion and increasing plasma TG clearance
- In diet induced obese mice, ANGPTL3 ASO administration reduced plasma triglyceride and LDL-C, reduced hepatic TG, and improved insulin and leptin sensitivity
- Antisense inhibition of ANGPTL3 may provide a unique approach to treatment of patients with elevated cholesterol, metabolic dyslipidemia, and fatty liver

Monoclonal antibodies anti ANGPTL3

ORIGINAL ARTICLE

Genetic and Pharmacologic Inactivation of ANGPTL3 and Cardiovascular Disease

F.E. Dewey, V. Gusarova, R.L. Dunbar, C. O'Dushlaine, C. Schurmann, O. Gottesman, S. McCarthy, C.V. Van Hout, S. Bruse, H.M. Dansky, J.B. Leader, M.F. Murray, M.D. Ritchie, H.L. Kirchner, L. Habegger, A. Lopez, J. Penn, A. Zhao, W. Shao, N. Stahl, A.J. Murphy, S. Hamon, A. Bouzelmat, R. Zhang, B. Shumel, R. Pordy, D. Gipe, G.A. Herman, W.H.H. Sheu, I-T. Lee, K.-W. Liang, X. Guo, J.I. Rotter, Y.-D.I. Chen,* W.E. Kraus, S.H. Shah, S. Damrauer, A. Small, D.J. Rader, A.B. Wulff, B.G. Nordestgaard, A. Tybjærg-Hansen, A.M. van den Hoek, H.M.G. Princen, D.H. Ledbetter, D.J. Carey,* J.D. Overton, J.G. Reid, W.J. Sasiela, P. Banerjee, A.R. Shuldiner, I.B. Borecki, T.M. Teslovich, G.D. Yancopoulos, S.J. Mellis, J. Gromada, and A. Baras

TG: -76%

LDL-C: 23%

BACKGROUND

Loss-of-function variants in the angiopoietin-like 3 gene (*ANGPTL3*) have been associated with decreased plasma levels of triglycerides, low-density lipoprotein (LDL) cholesterol, and high-density lipoprotein (HDL) cholesterol. It is not known whether such variants or therapeutic antagonism of ANGPTL3 are associated with a reduced risk of atherosclerotic cardiovascular disease.

METHODS

We sequenced the exons of *ANGPTL3* in 58,335 participants in the DiscovEHR human genetics study. We performed tests of association for loss-of-function variants in *ANGPTL3* with lipid levels and with coronary artery disease in 13,102 case patients and 40,430 controls from the DiscovEHR study, with follow-up studies involving 23,317 case patients and 107,166 controls from four population studies. We also tested the effects of a human monoclonal antibody, evinacumab, against Angptl3 in dyslipidemic mice and against ANGPTL3 in healthy human volunteers with elevated levels of triglycerides or LDL cholesterol.

RESULTS

In the DiscovEHR study, participants with heterozygous loss-of-function variants in *ANGPTL3* had significantly lower serum levels of triglycerides, HDL cholesterol, and LDL cholesterol than participants without these variants. Loss-of-function variants were found in 0.33% of case patients with coronary artery disease and in 0.45% of controls (adjusted odds ratio, 0.59; 95% confidence interval, 0.41 to 0.85; $P=0.004$). These results were confirmed in the follow-up studies. In dyslipidemic mice, inhibition of Angptl3 with evinacumab resulted in a greater decrease in atherosclerotic lesion area and necrotic content than a control antibody. In humans, evinacumab caused a dose-dependent placebo-adjusted reduction in fasting triglyceride levels of up to 76% and LDL cholesterol levels of up to 23%.

CONCLUSIONS

Genetic and therapeutic antagonism of ANGPTL3 in humans and of Angptl3 in mice was associated with decreased levels of all three major lipid fractions and decreased odds of atherosclerotic cardiovascular disease. (Funded by Regeneron Pharmaceuticals and others; ClinicalTrials.gov number, NCT01749878.)

Evinabumab in HoFH: LDL-C and lipids/Lipoproteins at Week 4 (N = 9)

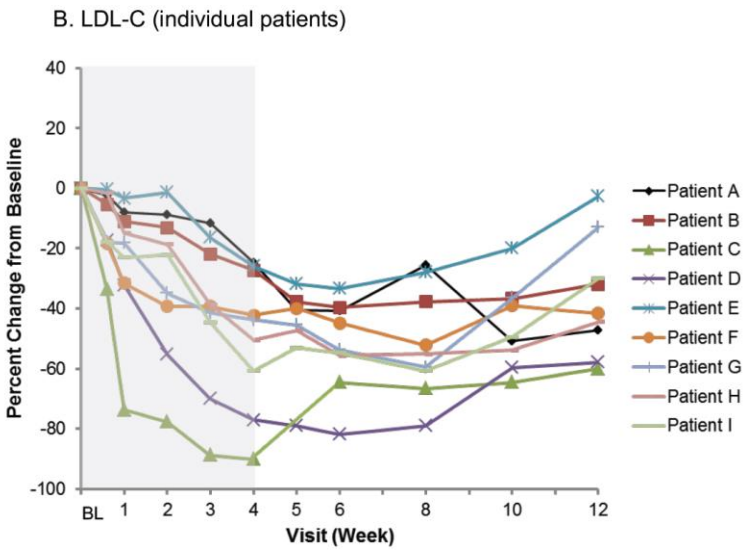
> 45% mean decrease in LDL-C, apo B and TG at W 4

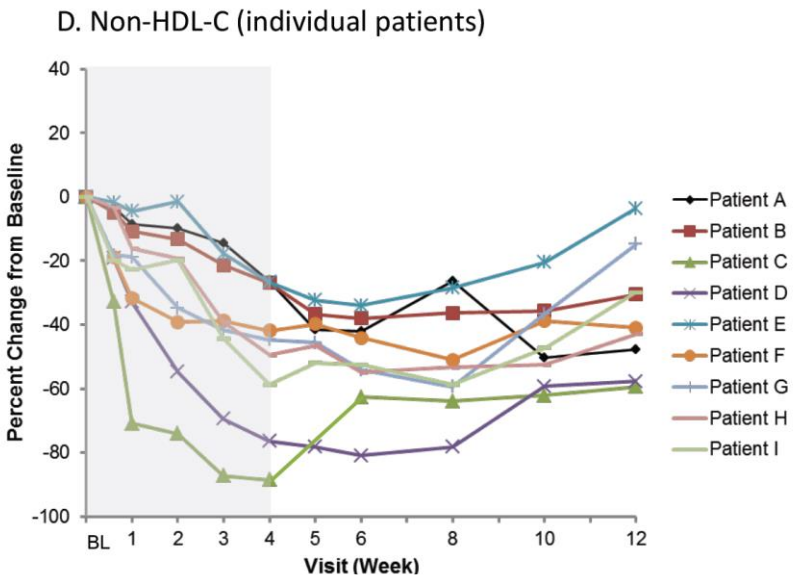
Variable	Mean±SD % change	Mean±SD or median (Q1, Q3) absolute change
LDL-C (mmol/L)	–49 ± 23	–4.1 ± 2.3
Non-HDL cholesterol (mmol/L)	–49 ± 22	–4.3 ± 2.4
Apolipoprotein A1 (g/L)	–39 ± 9	–0.4 ± 0.2
Apolipoprotein B (g/L)	–46 ± 18	–1.0 ± 0.6
Total cholesterol (mmol/L)	–47 ± 19	–4.7 ± 2.3
Lipoprotein(a) (nmol/L)	−19 (−27, 1)	−27 (−29, 1)
HDL-cholesterol (mmol/L)	−36 ± 16	−0.4 ± 0.3
Triglycerides (mmol/L)	−47 (−57, −38)	−0.3 (−0.2, −0.6)

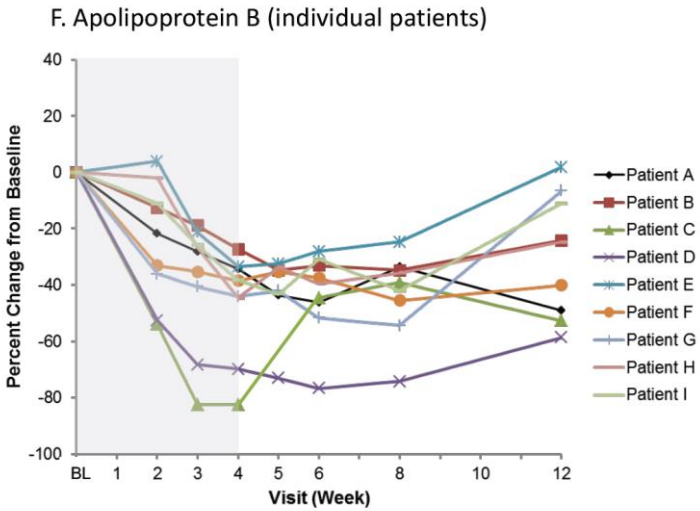
Data given as mean ± SD or median (Q1, Q3)

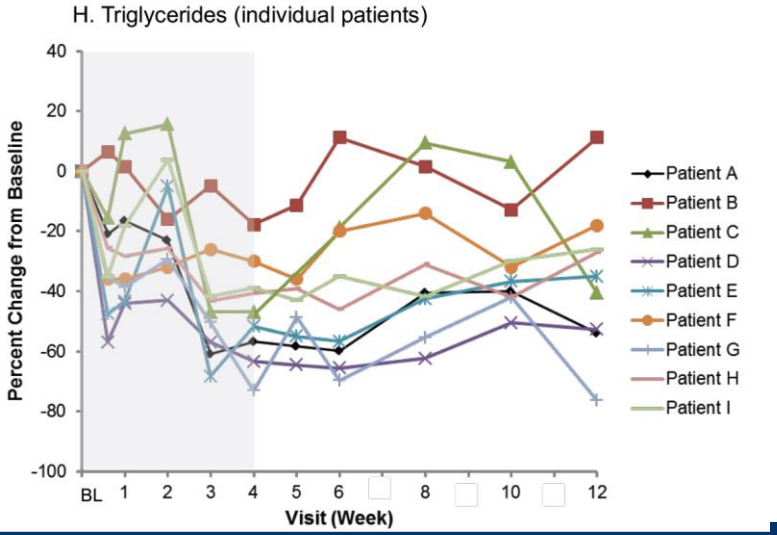
Gaudet D et al. NEJM 2017

Evinabumab in HoFH: LDL-C and lipids/Lipoproteins at Week 4 (N = 9)









Gaudet D et al. NEJM 2017

The NEW ENGLAND JOURNAL of MEDICINE

ESTABLISHED IN 1812 AUGUST 20, 2020 VOL. 383 NO. 8

Evinacumab for Homozygous Familial Hypercholesterolemia

Frederick J. Raal, M.D., Ph.D., Robert S. Rosenson, M.D., Laurens F. Reeskamp, M.D., G. Kees Hovingh, M.D., Ph.D., John J.P. Kastelein, M.D., Ph.D., Paolo Rubba, M.D., Shazia Ali, Pharm.D., Poulabi Banerjee, Ph.D., Kuo-Chen Chan, Ph.D., Daniel A. Gipe, M.D., Nagwa Khilla, M.S., Robert Pordy, M.D., David M. Weinreich, M.D., George D. Yancopoulos, M.D., Ph.D., Yi Zhang, Ph.D., and Daniel Gaudet, M.D., Ph.D., for the ELIPSE HoFH Investigators*

METHODS

In this double-blind, placebo-controlled, phase 3 trial, we randomly assigned in a 2:1 ratio 65 patients with homozygous familial hypercholesterolemia who were receiving stable lipid-lowering therapy to receive an intravenous infusion of evinacumab (at a dose of 15 mg per kilogram of body weight) every 4 weeks or placebo. The primary outcome was the percent change from baseline in the LDL cholesterol level at week 24.

UNIVERSITÀ DEGLI STUDI DI MILANO

Evinacumab: ECLIPSE Study

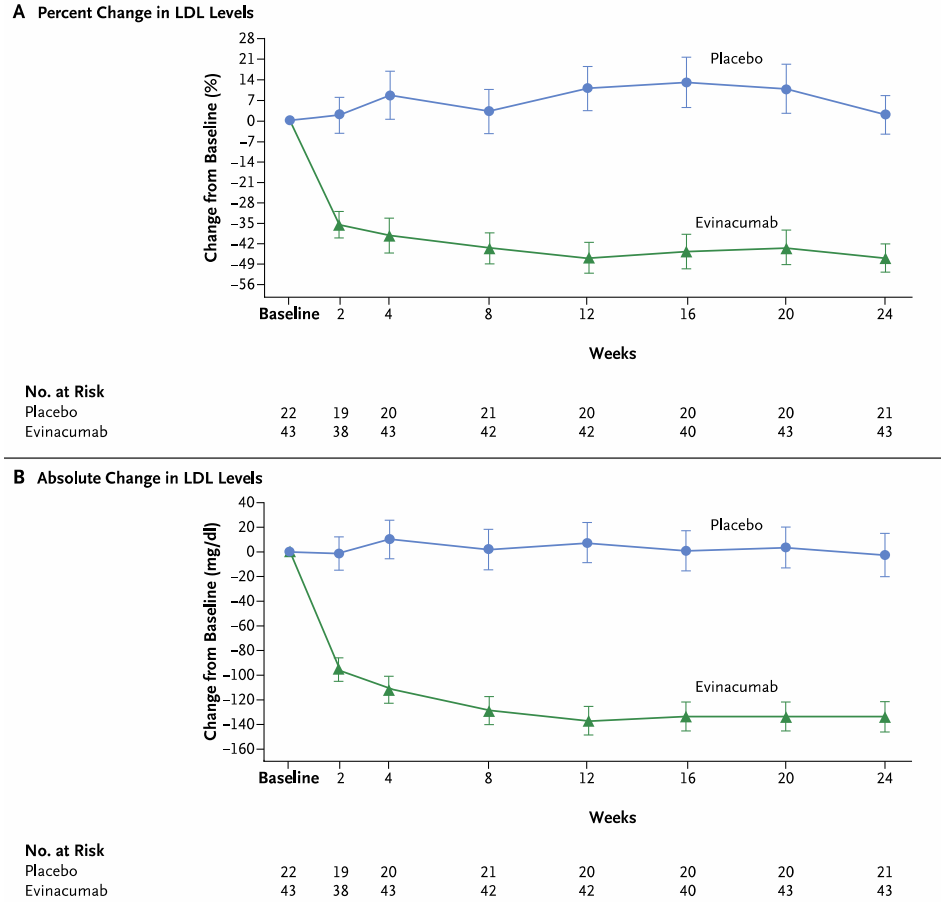
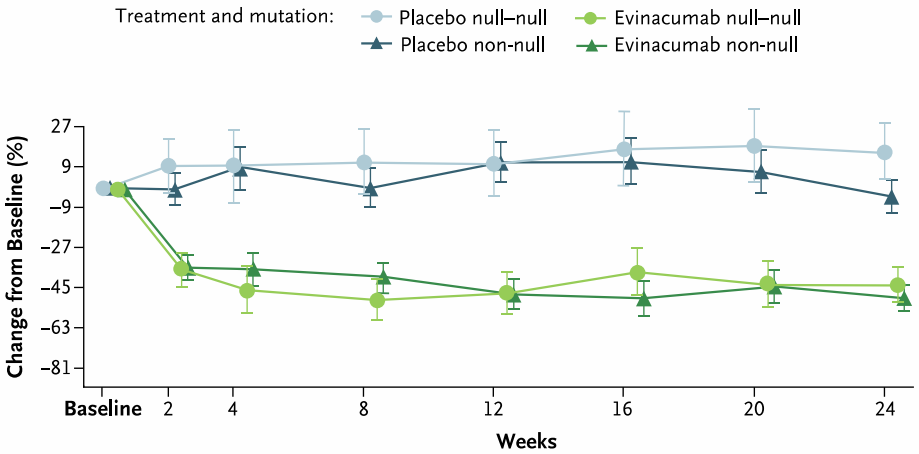


Figure 1. Changes from Baseline in Low-Density Lipoprotein (LDL) Cholesterol Levels at 24 Weeks.
Shown is the least-squares mean percent change (Panel A) and absolute change (Panel B) in calculated LDL cholesterol levels from baseline to week 24 in the evinacumab group and the placebo group. The I bars indicate standard errors.

Outcome	Evinacumab (N=43)	Placebo (N=22)	LS Mean (±SE) Difference 95% CI
Primary outcome			
Percent change from baseline in LDL cholesterol	−47.1±4.6	1.9±6.5	−49.0±8.0 (−65.0 to −33.1)
Key secondary outcomes			
Percent change from baseline in apolipoprotein B	−41.4±3.3	−4.5±4.8	−36.9±5.9 (−48.6 to −25.2)
Percent change from baseline in non-HD lipoprotein cholesterol	−49.7±3.8	2.0±5.4	−51.7±6.6 (−64.8 to −38.5)
Percent change from baseline in total cholesterol	−47.4±3.0	1.0±4.2	−48.4±5.1 (−58.7 to −38.1)

Evinacumab: ECLIPSE Study



No. at Risk

Placebo null–null	6	4	6	6	6	6	6	6
Placebo non-null	16	15	14	15	14	14	14	15
Evinacumab null–null	15	14	15	15	14	15	15	15
Evinacumab non-null	28	24	28	27	28	25	28	28

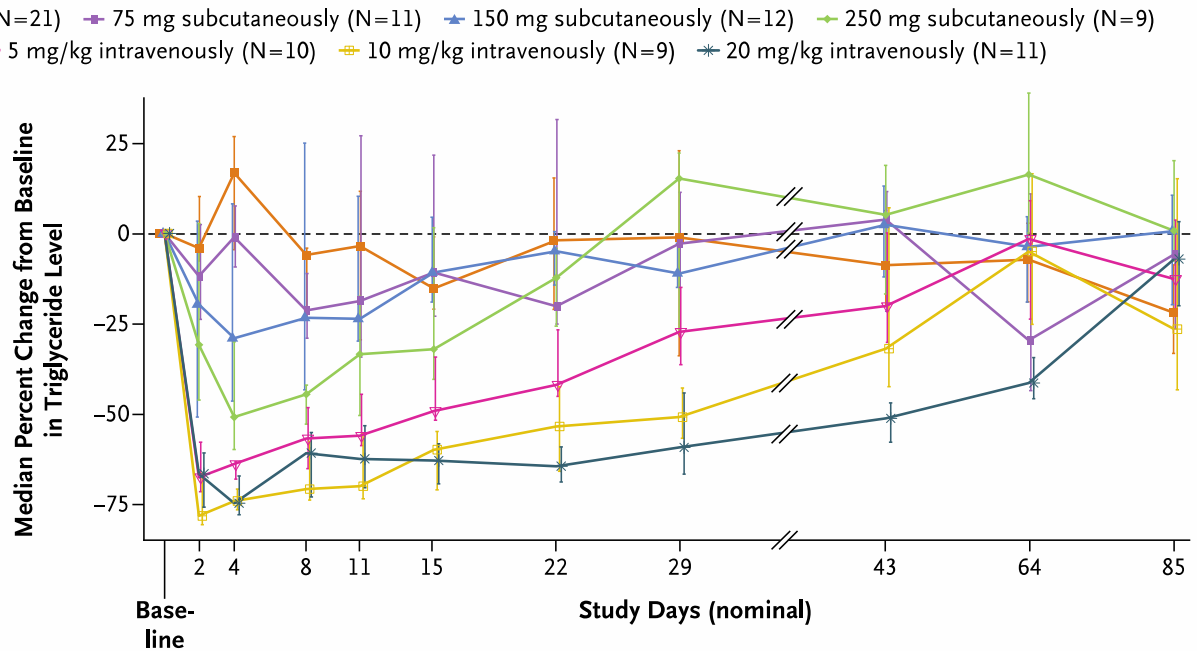
Figure 2. Percent Change in LDL Cholesterol Levels, According to Type of LDL-Receptor Variants.

Shown is the least-squares mean percent change from baseline in LDL cholesterol levels in the evinacumab group and the placebo group, according to the type of loss-of-function variant in the LDL receptor. Genetic variants that cause a virtually complete absence of LDL-receptor expression (null–null variants) result in higher LDL cholesterol levels than variants that partially reduce LDL-receptor activity (non-null variants). Patients with null–null variants have LDL-receptor activity of less than 15%.

Table 3. Adverse Events during the Treatment Period.*

Adverse Events	Evinacumab (N=44)	Placebo (N=21)
	no. (%)	
Any adverse event	29 (66)	17 (81)
Nasopharyngitis	7 (16)	5 (24)
Influenza-like illness	5 (11)	0
Headache	4 (9)	5 (24)
Rhinorrhea	3 (7)	0
Gastroenteritis	2 (5)	0
Infusion-site pruritus	2 (5)	0
Pyrexia	2 (5)	1 (5)
Cough	2 (5)	0
Dental caries	2 (5)	0
Diarrhea	2 (5)	1 (5)
Dyspepsia	2 (5)	0
Toothache	2 (5)	2 (10)
Dizziness	2 (5)	0
Urinary tract infection	0	2 (10)
Increased aspartate aminotransferase	0	2 (10)
Myalgia	0	2 (10)
Any serious adverse event	2 (5)	0
Urosepsis	1 (2)	0
Suicide attempt	1 (2)	0

Evinacumab: TG reduction in healthy volunteers



No. at Risk											
Placebo	21	21	21	21	20	18	19	19	19	18	19
75 mg subcutaneously	11	11	11	9	10	10	9	8	8	9	9
150 mg subcutaneously	12	12	12	12	12	12	11	11	10	8	8
250 mg subcutaneously	9	9	9	7	7	7	7	7	7	7	6
5 mg/kg intravenously	9	10	10	10	10	9	10	10	10	9	9
10 mg/kg intravenously	9	9	9	9	8	9	8	8	8	7	6
20 mg/kg intravenously	11	11	11	11	11	11	11	10	10	10	10

Figure 4. Effects of Inhibition of ANGPTL3 with a Monoclonal Antibody on Triglyceride Levels in Human Volunteers.
The median percent change from baseline in the triglyceride level (measured in milligrams per deciliter) is shown for the placebo group and for the groups receiving evinacumab at various doses. Individual data points are median values, with I bars showing interquartile ranges.

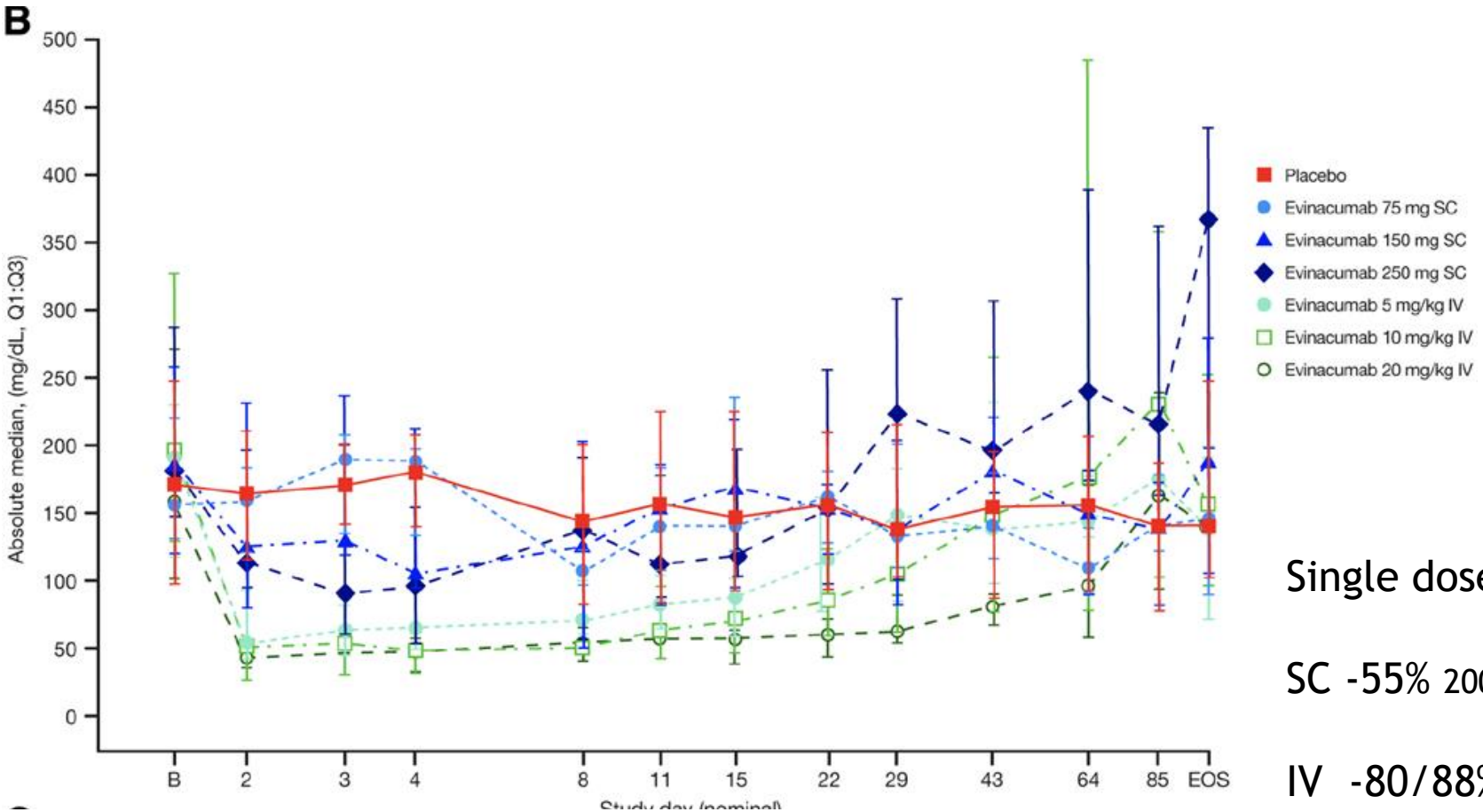
Circulation

ORIGINAL RESEARCH ARTICLE

Inhibition of Angiopoietin-Like Protein 3 With a Monoclonal Antibody Reduces Triglycerides in Hypertriglyceridemia

Methods: Subjects with triglycerides >150 but ≤450 mg/dL and LDL-C ≥100 mg/dL (n=83 for single ascending dose study [SAD]; n=56 for multiple ascending dose study [MAD]) were randomized 3:1 to evinacumab:placebo. SAD subjects received evinacumab subcutaneously (SC) at 75/150/250 mg, or intravenously (IV) at 5/10/20 mg/kg, monitored up to day (d) 126. MAD

Evinacumab and TGs



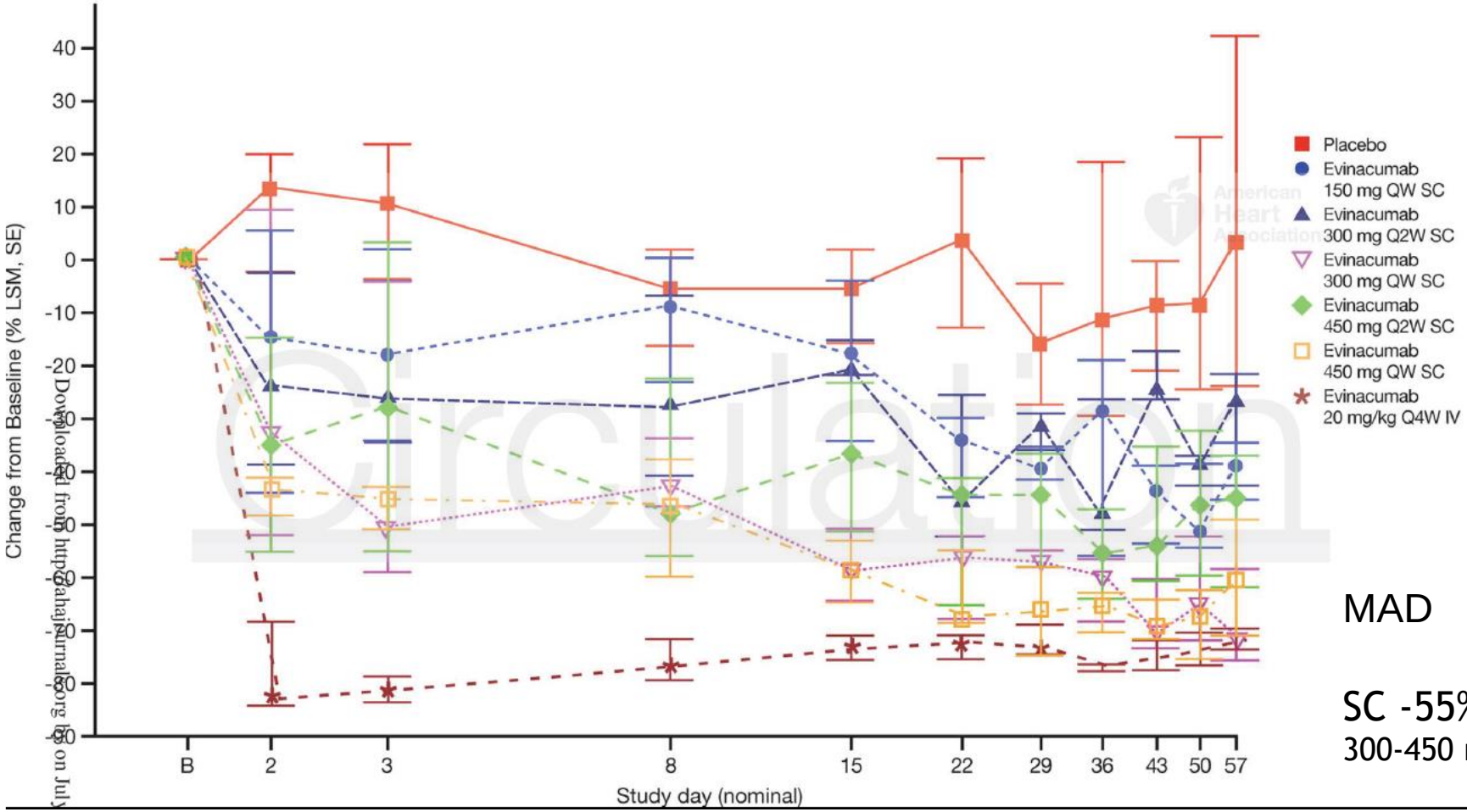
Single dose day4

SC -55% 200 mg

IV -80/88%
5 to 20 mg/kg

Evinacumab and TGs

Plasma TG



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MAD

SC -55%
300-450 mg

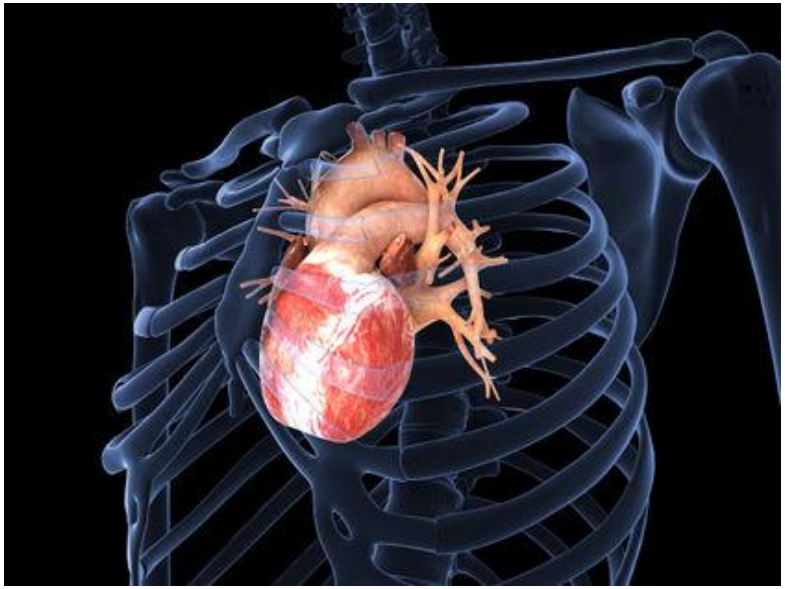
IV -88%
20 mg/kg

-New frontiers in hypercholesterolaemia
PCSK9 (Beyond Monoclonal antibodies)
ACLY (Bempedoic acid)
ANGPTL3 (Evinacumab, Vupanorsen, ARO-ANG3)

-Targeting trygliceridesrich lipoproteins
Icopentanyl EPA
APO CIII (Volanesorsen)
Pemafibrato
ANGPTL3

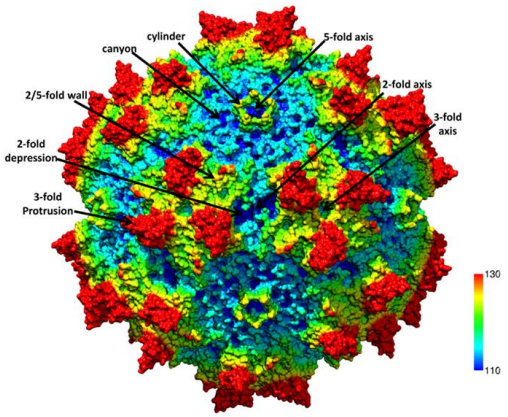
-Targeting Lp(a)
Apo(a) Gene silencing

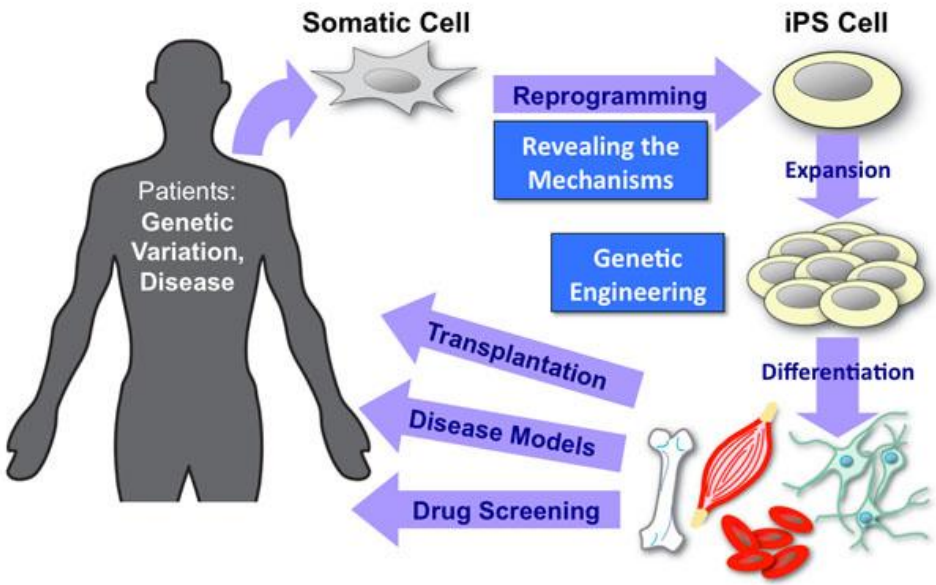
-Targeting Inflammation
IL1beta (Canakinumab)
Colchicine

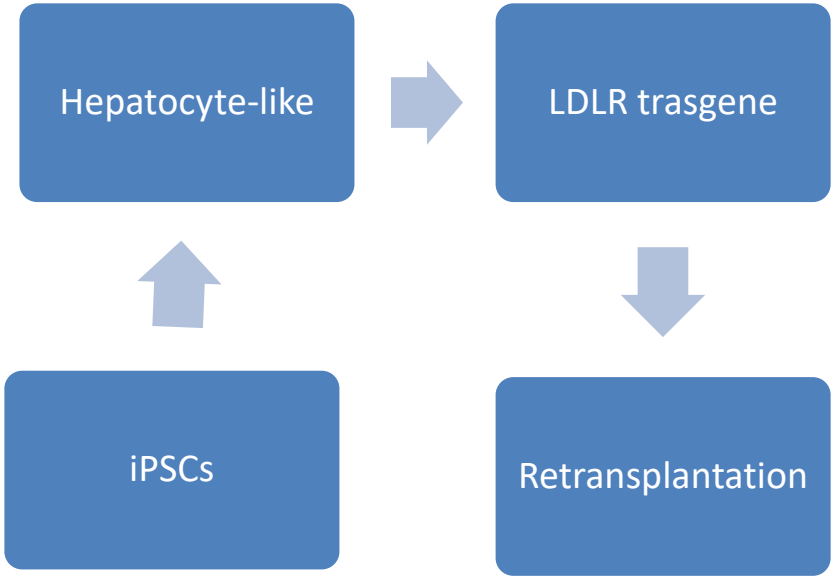


Long term vision? : AAV8.TBG.hLDLR

Agent	Stage of development
Recombinant *LDLR*-expressing AAV8 (AAV8.TBG.h*LDLR*)	Phase I/2a first-in-human study (NCT02651675). 12 patients. Primary safety endpoint at 1 year. Total follow-up of 5 years.







Thanks