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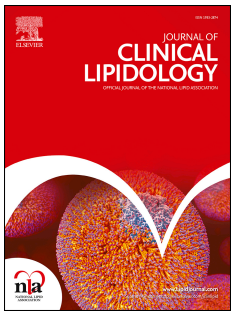

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Alirocumab efficacy in patients with double heterozygous, compound heterozygous, or homozygous familial hypercholesterolemia

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Running title: Alirocumab efficacy in pts with >1 FH mutations

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Abstract

BACKGROUND: Mutations in the genes for the low-density lipoprotein receptor (*LDLR*), apolipoprotein B (*APOB*), and proprotein convertase subtilisin/kexin type 9 (*PCSK9*) have been reported to cause heterozygous and homozygous familial hypercholesterolemia (FH).

OBJECTIVE: To examine the influence of double heterozygous, compound heterozygous, or homozygous mutations underlying FH on the efficacy of alirocumab.

METHODS: Patients from six alirocumab trials with elevated low-density lipoprotein cholesterol (LDL-C) and FH diagnosis were sequenced for mutations in the *LDLR*, *APOB*, *PCSK9*, *LDLR* adaptor protein 1 (*LDLRAP1*), and signal-transducing adaptor protein 1 (*STAP1*) genes. The efficacy of alirocumab was examined in patients who had double heterozygous, compound heterozygous, or homozygous mutations.

RESULTS: Of 1191 patients sequenced, 20 patients were double heterozygotes (n=7), compound heterozygotes (n=10), or homozygotes (n=3). Mean baseline LDL-C levels were similar between patients treated with alirocumab (n=11; 198 mg/dL) versus placebo (n=9; 189 mg/dL). All patients treated with alirocumab 75/150 or 150 mg every 2 weeks had an LDL-C reduction of ≥15% at either Week 12 or Week 24. At Week 12, one patient had an increase of 7.1% in LDL-C, while in others, LDL-C was reduced by 21.7–63.9% (corresponding to 39–114 mg/dL absolute reduction from baseline). At Week 24, LDL-C was reduced in all patients by 8.8–65.1% (10–165 mg/dL absolute reduction from baseline). Alirocumab was generally well tolerated in the six trials.

CONCLUSION: Clinically meaningful LDL-C-lowering activity was observed in patients receiving alirocumab who were double heterozygous, compound heterozygous, or homozygous for genes which are causative for FH.

KEYWORDS: alirocumab, hypercholesterolemia, *LDLR, APOB, LDLRAP1*

Introduction

Mutations in the genes for the low-density lipoprotein receptor (*LDLR*), apolipoprotein B (*APOB)*, and proprotein convertase subtilisin/kexin type 9 (*PCSK9)* have been reported to cause heterozygous familial hypercholesterolemia (HeFH) and homozygous familial hypercholesterolemia (HoFH), conditions which are characterized by high levels of low-density lipoprotein cholesterol (LDL-C) and increased risk of coronary heart disease.[1-3] Mutation in LDLR adaptor protein 1 (*LDLRAP1)* gene is recessive and causes HoFH.[1] LDL-C levels can vary markedly due to the phenotypic variability of mutations in the LDL-C pathway. Residual LDLR pathway activity correlates with disease severity and response to some lipid-lowering agents.[4,5] For example, patients who are *LDLR* negative have higher LDL-C levels and poorer clinical prognosis compared with patients who are *LDLR* defective.[6,7]

In general, patients with homozygous (identical mutations in both alleles) *LDLR* negative mutations or with compound heterozygous (different mutations in both alleles of the same gene) *LDLR* negative mutations have overall the highest mean LDL-C levels.[5] This is followed by those with compound heterozygous *LDLR* defective plus *LDLR* negative mutations, those with homozygous *LDLRAP1* or *LDLR* defective mutations, homozygous *APOB* or *PCSK9* gain-of-function (GOF) mutations, those with double heterozygous (mutations in two different genes) mutations, and then those with HeFH.[5,8] However, LDL-C level is the main determinant of cardiovascular disease risk and not the genetic defect *per se*.[7,9]

We have previously reported the effect of single mutations in genes causative for familial hypercholesterolemia (FH) in 1191 patients enrolled in one Phase 2 and five Phase 3 studies of the PCSK9 antibody alirocumab.[10] Here, we focus on the

treatment effect of alirocumab in patients with FH who were double heterozygotes, compound heterozygotes, or homozygotes.

Methods

DNA samples from patients with a diagnosis of FH who were enrolled and provided written consent for participation in six clinical trials, and also provided written consent for the present genotyping analysis, were sequenced for mutations in genes causative for FH (*LDLR*, *APOB*, *PCSK9*, *LDLRAP1*, and signal-transducing adaptor protein 1 [*STAP1*]). The trials included one Phase 2 trial (NCT01375764)[11] and five Phase 3 clinical trials from the ODYSSEY program (LONG TERM [NCT01507831],[12] HIGH FH [NCT01617655],[13] FH I [NCT01623115], FH II [NCT01709500],[14] and ALTERNATIVE [NCT01709513][15]). The original diagnosis of FH was performed either by previous genotyping or on clinical presentation. Clinical diagnosis was based on the Simon Broome criteria for definite FH or the World Health Organization/Dutch Lipid Network criteria (score >8 points).[16-18] The original genotyping results were not recorded in the trials, hence patients were sequenced regardless of how they were originally diagnosed. Full details of the genotyping analysis for the present study have been described previously.[10]

The present analysis focuses on those patients who had more than one mutation in one or more of the sequenced genes. No patients from the ALTERNATIVE trial were found to have more than one mutation. Study designs of the other trials were as follows. In the 12-week Phase 2 study, patients received one of four alirocumab doses (150 mg every 2 weeks [Q2W], 150 mg every 4 weeks [Q4W], 200 mg Q4W, 300 mg Q4W) or placebo.[11] In the 78-week Phase 3 trials, patients received either alirocumab 150 mg Q2W (LONG TERM and HIGH FH) or an initial alirocumab dose of 75 mg Q2W, which was increased to 150 mg Q2W at Week 12 if LDL-C was ≥70 mg/dL at Week 8 (FH I and FH II); control was placebo in each trial.[12-14] The

primary efficacy endpoint in the Phase 3 trials was the percentage reduction in LDL-C from baseline to Week 24. Safety assessments included treatment-emergent adverse events (TEAEs), which were events occurring from first to last dose and up to 70 days after the last dose (follow-up).

LDL-C levels were calculated using the Friedewald equation[19] except when triglyceride levels exceeded 400 mg/dL, in which case LDL-C was determined by direct measurement using beta quantification.[20] In this *post hoc* analysis, a clinically meaningful response to alirocumab was defined as a reduction in LDL-C of ≥15% at Week 12 or Week 24 (the available timepoints), as described previously.[10] Analysis of lipid and lipoprotein parameters was performed at a central laboratory. Lipoprotein (a) [Lp(a)] levels were analyzed using a validated immunoturbidimetric assay as previously described.[21]

Results

Patients

Of 1191 patients sequenced, 20 patients were double heterozygous (n=7), compound heterozygous (n=10), or homozygous (n=3) for genes causative of FH, and included in the present analysis (Table 1). Six patients were double heterozygotes with mutations in both *APOB* and *LDLR*, of whom three patients were *APOB* defective/*LDLR* negative and the remaining three *APOB* defective/*LDLR* defective. One patient was double heterozygote with *LDLR* negative and *PCSK9* GOF mutations. Of those who were compound heterozygotes, three were *LDLR* defective/*LDLR* negative and seven were *LDLR* defective/*LDLR* defective. Of the three patients who were homozygotes, one had *LDLR* defective mutations (further details on this patient are presented in the Supplementary) and two were homozygous for mutations in *LDLRAP1*.

In this analysis, 11 of 20 patients received alirocumab and the remaining nine received placebo (Table 1). The mean age at baseline was 49.2 years and 50% were males. Baseline characteristics of individual patients are presented in Supplementary Table 1. The mean baseline LDL-C level was 198 mg/dL for those treated with alirocumab and 189 mg/dL for those treated with placebo. All patients were receiving concomitant statin and the majority were receiving additional lipid-lowering therapies (LLTs) at baseline (Supplementary Table 1). Most patients were at very high cardiovascular risk at baseline. The cardiovascular history of individual patients at baseline is presented in Supplementary Table 2.

Influence of double heterozygous, compound heterozygous, or homozygous mutations on the efficacy of alirocumab

Percentage changes from baseline in LDL-C at Week 12 and 24 for individual patients with available data are shown in Figure 1A and Figure 1B, respectively; absolute changes are shown in Supplementary Table 3. In this analysis, an LDL-C reduction of ≥15% at Week 12 or 24 was observed in patients who had received alirocumab 75/150 or 150 mg Q2W (Figure 1). At Week 12, an LDL-C reduction of 21.7–63.9% (corresponding to 39–114 mg/dL absolute reduction) with alirocumab treatment was observed in all but one patient (Patient 10, *LDLRAP1* negative, baseline LDL-C of 140 mg/dL, from FH I study) who had an LDL-C increase of 7.1%; however, this patient had an LDL-C reduction of 34.3% (absolute reduction of 48 mg/dL) from baseline to Week 24. LDL-C reduction from baseline to Week 24 in other patients was 8.8–65.1% (absolute reduction of 10–165 mg/dL).

Furthermore, Patient 5 (*LDLR* defective/*LDLR* negative from FH II study) had an LDL-C reduction of 52.6% (absolute reduction of 60 mg/dL from baseline value of 114 mg/dL) at Week 12, compared with a reduction of 8.8% (absolute reduction of 10 mg/dL) at Week 24. Patient 9 (*LDLR* defective homozygous from HIGH FH study) had an LDL-C reduction of 22.9% (absolute reduction of 92 mg/dL from baseline value of 402 mg/dL) at Week 12 compared with a reduction of 11.9% (absolute reduction of 48 mg/dL) at Week 24.

Overall, alirocumab treatment provided LDL-C reductions of 39.3–55.7% and 55.1–62.0% in patients with double heterozygous mutations (*APOB* defective/*LDLR* negative and *APOB* defective/*LDLR* defective) at Week 12 and Week 24, respectively. The corresponding reductions in patients with compound heterozygous

mutations (*LDLR* defective/*LDLR* negative and *LDLR* defective/*LDLR* defective) were 21.7–63.9% and 8.8–65.1% at Week 12 and Week 24, respectively.

At Week 12, two patients (Patients 5 and 6, both *LDLR* defective/*LDLR* negative) achieved an LDL-C level of <70 mg/dL with alirocumab treatment. In addition, two patients (Patient 1 [*APOB* defective/*LDLR* negative] and Patient 4 [*APOB* defective/*LDLR* defective]) achieved LDL-C <100 mg/dL. Overall, the LDL-C levels were maintained in these patients at Week 24 except in Patient 5 who had an LDL-C level of 104 mg/dL, compared with 54 mg/dL at Week 12.

Reductions with alirocumab treatment at Weeks 12 and 24 were also observed across the mutation backgrounds in ApoB, Lp(a), non-high-density lipoprotein cholesterol, and triglycerides (Supplementary Figure 1–4). The patient with the *PCSK9* GOF and *LDLR* negative mutations (Patient 11) received a different administration regimen of alirocumab (150 mg Q4W) in the Phase 2 study and is not included in Figure 1 or Supplementary Figures 1–4; an LDL-C reduction of 44.1% (corresponding to 60 mg/dL absolute reduction in LDL-C from baseline value of 136 mg/dL) was observed at Week 10, 2 weeks after the last alirocumab dose was administered.

Safety

Safety data for all patients sequenced for mutations in genes causative for FH (n=1191) have been reported previously.[10] The rates of TEAEs in the overall sequenced cohort were comparable for alirocumab (82.9%) versus comparator (83.3%; comparator included placebo as well as ezetimibe).[10] The incidence of injection-site reactions (mostly mild and transient) was higher for alirocumab (11.4%)

versus comparator (8.8%).[10] Given the small population (n=20) for the present analysis, no further safety analysis was performed for this specific cohort.

Discussion

In the present analysis, we identified 20 patients with double heterozygous, compound heterozygous, and homozygous FH mutations, from six of the alirocumab clinical trials. All patients who received alirocumab 75/150 or 150 mg Q2W (majority of whom were receiving background statins) in the trials responded to treatment (defined by LDL-C reduction ≥15% on at least Week 12 or Week 24). At Week 12, alirocumab treatment resulted in LDL-C reductions of 21.7–63.9% (absolute reductions of 39–114 mg/dL) in all but one patient (Patient 10; a 39-year-old female with *LDLRAP1* negative mutations) who had an increase of 7.1% in LDL-C (baseline LDL-C was 140 mg/dL); however, a reduction of 34.3% (absolute reduction of 48 mg/dL) from baseline to Week 24 was observed in this patient, following alirocumab dose increase from 75 mg Q2W to 150 mg Q2W at Week 12.

Furthermore, two alirocumab-treated patients showed inconsistent LDL-C reductions at Week 12 vs Week 24. Patient 5 had an LDL-C reduction of 52.6% and 8.8% at Weeks 12 and 24, respectively. The corresponding values for Patient 9 were 22.9% and 11.9%, respectively. Although there is no firm explanation for the differences in response between Week 12 and Week 24 LDL-C reductions in these patients, nonadherence to therapy cannot be excluded.

Reductions of 24–30% in LDL-C, regardless of baseline levels, have been reported to provide clinical benefits, including reduced risks of cardiovascular events and deaths.[22-24] With the range of LDL-C reductions observed in this analysis, patients with more than one FH mutation will be expected to have reduced cardiovascular risks with alirocumab treatment. Although at Week 12 only two and four alirocumab-treated patients achieved risk-specific LDL-C goals of <70 mg/dL or <100 mg/dL,

respectively, those who did not achieve the LDL-C goals had reductions of 21.7–39.3%, equivalent to 39–114 mg/dL absolute reductions in LDL-C (despite high baseline LDL-C level of ≥180 mg/dL). With these high baseline LDL-C levels, achievement of LDL-C <70 mg/dL is unlikely, but these patients will be expected to have reduced risk of cardiovascular events and improved survival with the observed reductions in their LDL-C.

The LDLR mediates uptake of low-density lipoprotein (LDL) particles into the liver cell, via interaction with the APOB component of LDL. PCSK9 binds to the LDLR and prevents the receptor recycling to the cell surface, targeting the LDLR for degradation by endocytosis. Inhibition of PCSK9 with the monoclonal antibody alirocumab reduces LDL-C levels by increasing the level of LDLRs on the liver cell surface, resulting in an increased uptake of LDL particles.[25] Hence, alirocumab's mode of action involves the LDLR, ApoB, and PCSK9 (and likely other proteins such as LDLRAP1, which interacts with LDLR), and mutations in genes encoding these proteins could conceivably impact the treatment effect of alirocumab. For example, complete loss of both copies of *LDLR* may be expected to nullify the effect of a PCSK9 inhibitor. Indeed, another PCSK9 inhibitor showed no effect on LDL-C levels when examined in three patients with *LDLR* negative/negative mutations,[26,27] with similar results seen in a large open-label study.[28] None of the patients examined in our analysis was *LDLR* negative/negative.

In this analysis, alirocumab treatment provided substantial reductions in LDL-C in patients with FH and residual *LDLR* function (including patients with mutations in both copies of the gene). Double heterozygous mutations in *APOB* and *LDLR* appeared not to influence the efficacy of alirocumab, with reductions in the same

range as reported for the overall pooled analysis of FH patients from alirocumab Phase 3 trials (mean reductions from baseline to Week 24 of 48.8% and 55.0% with alirocumab doses of 75 mg Q2W [with possible dose increase to 150 mg Q2W at Week 12] and 150 mg Q2W, respectively).[29]

Published data have shown a mean reduction in LDL-C of 29.6% at Week 12 in 20 HoFH patients with *LDLR* defective mutations in one or both alleles, following biweekly treatment with another PCSK9 inhibitor, supporting the efficacy of PCSK9 inhibitors in patients with defective *LDLR* function.[27] In our study, alirocumab 75/150 or 150 mg Q2W treatment in seven patients with defective *LDLR* function (Patients 3–9) provided LDL-C reductions of 21.7–63.9% at Week 12, a mean reduction of 41.2%. Of note, this includes patients who also have other mutations including defective *APOB* function (Patients 3 and 4) and negative *LDLR* function (Patients 5 and 6).

Alirocumab treatment resulted in LDL-C reduction in the patient with *LDLR* negative and *PCSK9* GOF mutations, lending further support to previously published results suggesting that *PCSK9* GOF mutations in general do not impair the efficacy of alirocumab;[30] similar findings were observed with another PCSK9 inhibitor.[28]

Previous reports have indicated mean reductions in Lp(a) of approximately 20% with alirocumab treatment.[21] Lp(a) is known to be an independent risk factor for cardiovascular disease.[31] In the present analysis, Lp(a) reductions with alirocumab varied between Week 12 and Week 24. At Week 24, reductions in the range 19.8–49.5% were observed across the patients treated with alirocumab, although (for reasons which are unclear) two patients (with *LDLRAP1* and *LDLR* defective

homozygous mutations, respectively) did not have an Lp(a) reduction at Week 24. Baseline Lp(a) levels also varied considerably between patients (25–99 mg/dL).

The alirocumab safety profile in the cohort of sequenced patients was comparable between those who received alirocumab or placebo,[10] consistent with pooled safety data from the overall FH populations of alirocumab Phase 3 trials.[29]

Limitations

Limitations of this *post hoc* analysis include the small number of patients with each mutation type; however, this is inevitable given the rarity of these mutations. Furthermore, patients with a known history of HoFH were excluded in the individual clinical trials, and so very few patients with HoFH were included in the present analysis. However, in general, the data is robust, with low heterogeneity. The analysis was well controlled with a similar group of patients who received placebo during the study. The impact of rare mutation types may be better assessed in specifically designed trials using a placebo-phase approach, whereby each patient acts as their own control, as previously described.[30]

Conclusion

A clinically meaningful LDL-C-lowering activity was observed in patients receiving alirocumab who are double or compound heterozygous, or homozygous for genes which are causative for FH, such as *LDLR*, *APOB*, *PCSK9*, and *LDLRAP1*. LDL-C-lowering activity of alirocumab in these mutations is likely to be attributable to the presence of at least one partially functional allele.

Author contributions

Merel L. Hartgers: Interpretation of data.

Joep C. Defesche: Interpretation of data.

Gisle Langslet: Acquisition of data (trial investigator), interpretation of data.

Paul N. Hopkins: Interpretation of data.

John J. P. Kastelein: Concept/design, acquisition of data (trial investigator), interpretation of data.

Marie T. Baccara-Dinet: Interpretation of data.

Werner Seiz: Concept/design, interpretation of data.

Sara Hamon: Concept/design, interpretation of data.

Poulabi Banerjee: Concept/design, interpretation of data.

Claudia Stefanutti: Interpretation of data.

All authors were involved in critical review of manuscript drafts and approved the final article for submission.

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Table 1. Distribution of mutations and treatment received by each patient (sequenced cohort)

Patient number	Study	Mutation category	Genotype	Treatment
1	FH I	*APOB* defective/*LDLR* negative	p.Arg3527Gln.c.1846-?_2140+?del	Alirocumab 75/150 mg Q2W[†]
2[‡]	HIGH FH	*APOB* defective/*LDLR* negative	p.Arg3527Gln.2390-?_2583+?del	Alirocumab 150 mg Q2W
3	FH I	*APOB* defective/*LDLR* defective	p.Arg3527Gln.p.Asp227Glu	Alirocumab 75/150 mg Q2W[†]
4	FH II	*APOB* defective/*LDLR* defective	p.Arg3527Gln.p.Cys209Tyr	Alirocumab 75/150 mg Q2W[†]
5	FH II	*LDLR* defective/*LDLR* negative	c.(-16)G>C.p.Trp562*	Alirocumab 75/150 mg Q2W[†]
6	FH II	*LDLR* defective/*LDLR* negative	c.313+1G>A.p.Val462Ile	Alirocumab 75/150 mg Q2W[†]
7	R727-CL-1003 Phase 2	*LDLR* defective/*LDLR* defective	p.Arg81Cys.c.(-268)G>T	Alirocumab 150 mg Q2W
8	HIGH FH	*LDLR* defective/*LDLR* defective	p.Asp266Asn.p.Gly592Glu	Alirocumab 150 mg Q2W
9	HIGH FH	*LDLR* defective homozygous	p.Asp227Glu.p.Asp227Glu	Alirocumab 150 mg Q2W
10	FH I	*LDLRAP1* negative	c.344+1G>A.c.344+1G>A	Alirocumab 75/150 mg Q2W[†]
11	R727-CL-1003 Phase 2	*LDLR* negative/*PCSK9* GOF	p.Cys143.p.Leu22_Leu23dup	Alirocumab 150 mg Q4W
12	FH I	*APOB* defective/*LDLR* negative	p.Arg3527Gln.p.Tyr375Trpfs*7	Placebo
13	FH I	*APOB* defective/*LDLR* defective	p.Arg3527Gln.p.Gly478Arg	Placebo

14	FH I	*LDLR* defective/*LDLR* negative	p.Glu600Asp.c.191-?_1060+?del	Placebo
15	FH I	*LDLR* defective/*LDLR* defective	p.Glu408Lys.p.Gln770Arg	Placebo
16	FH I	*LDLR* defective/*LDLR* defective	p.Glu337Lys.p.Asp482Asn	Placebo
17	LONG TERM	*LDLR* defective/*LDLR* defective	p.Asp651Asn.p.Asp221Gly	Placebo
18	LONG TERM	*LDLR* defective/*LDLR* defective	p.Asp700Glu.p.Asp227Glu	Placebo
19	LONG TERM	*LDLR* defective/*LDLR* defective	p.Leu432Val.p.Tyr465Asn.p.Pro685Leu	Placebo
20	LONG TERM	*LDLRAP1* negative	p.Gly24Alafs*32.p.Gly24Alafs*32	Placebo

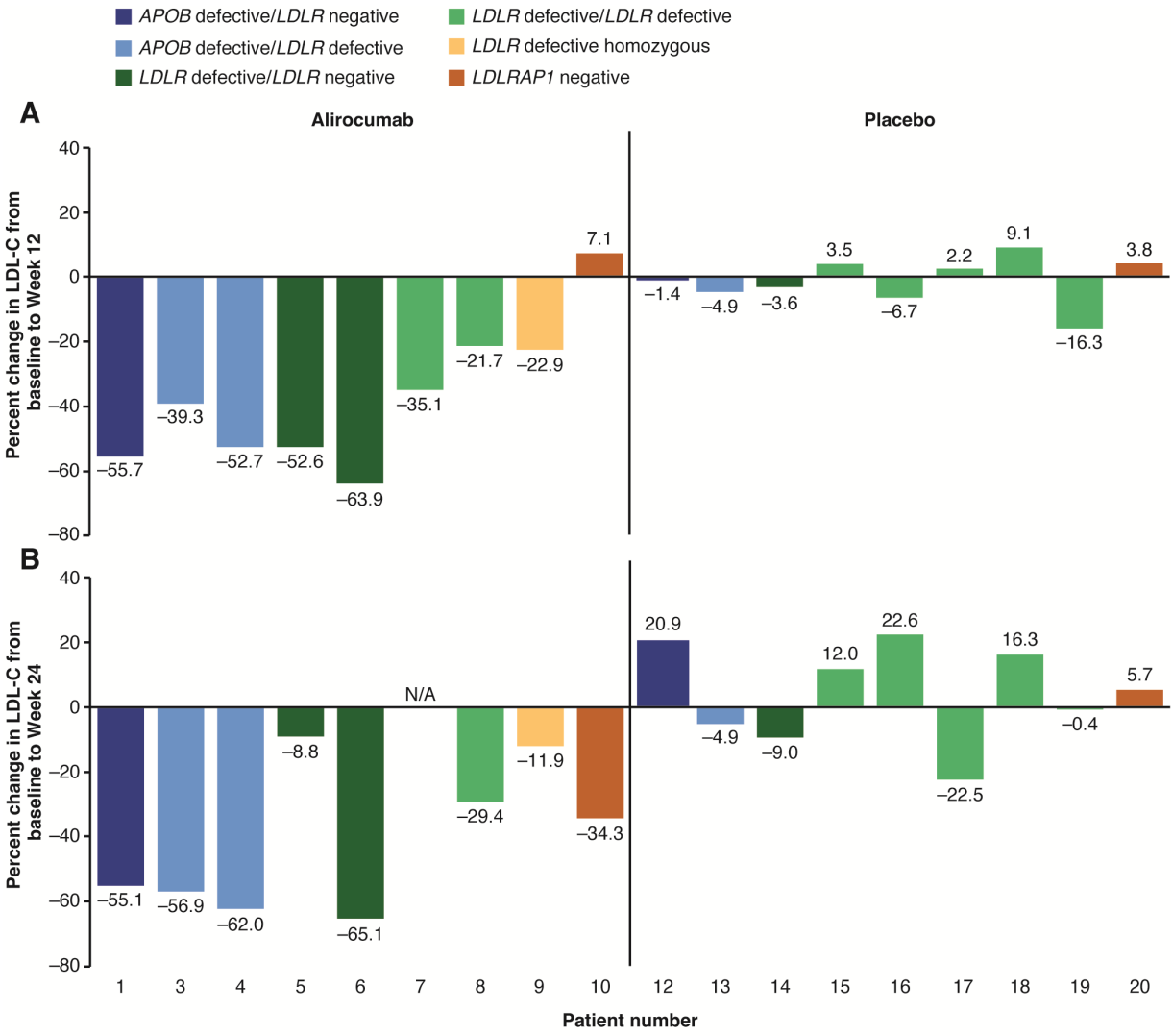
APOB, apolipoprotein B; GOF, gain-of-function; LDL-C, low-density lipoprotein cholesterol; *LDLR*, low-density lipoprotein receptor; *LDLRAP1*, LDLR adaptor protein 1; *PCSK9*, proprotein convertase subtilisin/kexin type 9; Q2W, every 2 weeks; Q4W, every 4 weeks.

[†]Alirocumab 75 mg Q2W was increased to 150 mg Q2W at Week 12 depending on LDL-C at Week 8.

[‡]Data for all lipid endpoints was not available for Patient 2.

Figure 1 Percentage reduction from baseline in LDL-C at (A) Week 12 and (B) Week 24 for individual patients

Data was not available for Patient 2 (*APOB* defective/*LDLR* negative). Patient 7 was from the 12-week Phase 2 study hence no data was available at Week 24. Patient 11 (*PCSK9* GOF and *LDLR* negative) received a different alirocumab administration regimen and is not included in this figure. *APOB*, apolipoprotein B; LDL-C, low-density lipoprotein cholesterol; *LDLR*, low-density lipoprotein receptor; *LDLRAP1*, LDLR adaptor protein 1; NA, not available.


APOB defective/LDLR negative
APOB defective/LDLR defective
LDLR defective/LDLR negative
LDLR defective/LDLR defective
LDLR defective homozygous
LDLRAP1 negative
A
Alirocumab
Placebo
Percent change in LDL-C from baseline to Week 12
40
20
0
−20
−40
−60
−80
−55.7
−39.3
−52.7
−52.6
−63.9
−35.1
−21.7
−22.9
7.1
−1.4
−4.9
−3.6
3.5
−6.7
2.2
9.1
−16.3
3.8
B
Percent change in LDL-C from baseline to Week 24
−55.1
−56.9
−62.0
−8.8
−65.1
N/A
−29.4
−11.9
−34.3
20.9
−4.9
−9.0
12.0
22.6
−22.5
16.3
−0.4
5.7
1
3
4
5
6
7
8
9
10
12
13
14
15
16
17
18
19
20
Patient number

Highlights

- The effect of >1 FH mutation on alirocumab efficacy was assessed in 1191 patients
- Mutations in *LDLR*, *APOB*, *PCSK9*, *STAP1*, and *LDLRAP1* were assessed
- 20 patients had double or compound heterozygous, or homozygous mutations
- Clinically meaningful reductions in LDL-C were observed with alirocumab treatment

Supplementary Material

Further details for patient with homozygous familial hypercholesterolemia

In this analysis, one patient (Patient 9) was genotyped as having homozygous low-density lipoprotein receptor (*LDLR*)-defective mutations. This patient was a 28-year-old male with homozygous familial hypercholesterolemia diagnosed at the age of 5 years. The patient was treated with simvastatin 80 mg and nicotinic acid 1500 mg daily; he had a body mass index of 23 kg/m^2. Xanthomas were present on both knees. Medical history was hypertension, aortic stenosis, and stable angina. Patient was treated with alirocumab 150 mg every 2 weeks with baseline LDL-C 402 mg/dL and achieving an LDL-C reduction of 22.9% at Week 12.

Supplementary Table 1 Patient characteristics at baseline

Patient number	Age (years)	Gender	Mutation category	LDL-C (mg/dL)	ApoB (mg/dL)	Lp(a) (mg/dL)	Non-HDL-C (mg/dL)	Triglycerides (mg/dL)	Statin	Other LLTs
1	59	F	*APOB* defective/*LDLR* negative	167	137	99	191	120	Rosuvastatin 20 mg	Ezetimibe
2	50	F	*APOB* defective/*LDLR* negative	155	113	2	183	138	Rosuvastatin 40 mg	NA
3	69	F	*APOB* defective/*LDLR* defective	290	168	25	317	137	Atorvastatin 80 mg	Ezetimibe
4	47	F	*APOB* defective/*LDLR* defective	150	109	38	165	73	Rosuvastatin 40 mg	Ezetimibe
5	61	F	*LDLR* defective/*LDLR* negative	114	87	86	126	61	Atorvastatin 80 mg	Ezetimibe
6	58	F	*LDLR* defective/*LDLR* negative	166	124	54	187	104	Atorvastatin 80 mg	Ezetimibe, bile acid sequestrants
7	36	M	*LDLR* defective/*LDLR* defective	205	162	38	233	139	Simvastatin 40 mg, rosuvastatin 10 mg	Fish oil, nicotinic acid
8	31	M	*LDLR* defective/*LDLR* defective	180	124	80	194	68	Rosuvastatin 40 mg	NA
9	28	M	*LDLR* defective homozygous	402	202	69	419	83	Simvastatin 80 mg	Nicotinic acid
10	39	F	*LDLRAP1* negative	140	100	71	149	43	Atorvastatin	Ezetimibe,

									80 mg	fenofibrate
11	54	M	*LDLR* negative/*PCSK9* GOF	136	NA	NA	NA	NA	Rosuvastatin 40 mg	Ezetimibe
12	58	F	*APOB* defective/*LDLR* negative	296	203	50	336	198	Atorvastatin 80 mg	Ezetimibe, fish oil
13	54	M	*APOB* defective/*LDLR* defective	163	120	91	175	61	Atorvastatin 80 mg	Ezetimibe, nicotinic acid
14	35	M	*LDLR* defective/*LDLR* negative	167	138	2	201	169	Rosuvastatin 20 mg	Ezetimibe
15	60	M	*LDLR* defective/*LDLR* defective	142	132	185	170	142	Rosuvastatin 40 mg	Ezetimibe, fish oil, bile acid sequestrants
16	59	M	*LDLR* defective/*LDLR* defective	164	128	178	185	103	Rosuvastatin 40 mg	Ezetimibe, nicotinic acid, bile acid sequestrants
17	51	M	*LDLR* defective/*LDLR* defective	232	164	115	252	102	Rosuvastatin 40 mg	Ezetimibe
18	49	F	*LDLR* defective/*LDLR* defective	192	119	2	207	73	Simvastatin 40 mg	N/A
19	41	F	*LDLR* defective/*LDLR* defective	208	143	4	227	93	Rosuvastatin 40 mg	Ezetimibe, bile acid sequestrants
20	45	M	*LDLRAP1* negative	141	110	2	156	74	Rosuvastatin 40 mg	Ezetimibe, nicotinic acid

ApoB, apolipoprotein B; F, female; GOF, gain-of-function; LDL-C, low-density lipoprotein cholesterol; *LDLR*, low-density lipoprotein receptor; *LDLRAP1*, LDLR adaptor protein 1; Lp(a), lipoprotein(a); M, male; NA, not available; non-HDL-C, non-high-density lipoprotein cholesterol; *PCSK9*, proprotein convertase subtilisin/kexin type 9.

Supplementary Table 2 Cardiovascular history at baseline

Patient number	Dictionary-derived term for reported cardiovascular history	Categorization of cardiovascular risk per protocol
1	Coronary revascularization, coronary artery disease, cardiac catheterization, angina pectoris, familial risk factor, mitral valve prolapse, unstable angina, coronary artery bypass, cardiac stress test	Very high cardiovascular risk
2	Hypertension	High cardiovascular risk
3	Hypertension, familial risk factor, type 2 diabetes mellitus	Very high cardiovascular risk
4	Coronary artery disease, cardiac stress test, carotid arteriosclerosis	Very high cardiovascular risk
5	Coronary revascularization, coronary artery disease, familial risk factor, angina pectoris, acute myocardial infarction, unstable angina, percutaneous coronary intervention	Very high cardiovascular risk
6	Hypertension, intermittent claudication, ankle brachial index, cardiac murmur, familial risk factor	Very high cardiovascular risk
7	None reported	Not applicable
8	Coronary revascularization, coronary artery disease, arteriosclerosis, arteriosclerosis coronary artery, angina pectoris, coronary angioplasty, coronary arterial stent insertion	Very high cardiovascular risk
9	Coronary artery disease, familial risk factor, hypertension, aortic stenosis, angina pectoris	Very high cardiovascular risk
10	Sinus bradycardia, abdominal bruit, carotid bruit, cardiac murmur, familial risk factor	High cardiovascular risk
11	None reported	Not applicable
12	None reported	Not applicable
13	Coronary revascularization, coronary artery disease, acute myocardial infarction,	Very high cardiovascular risk

	percutaneous coronary intervention	
14	None reported	Not applicable
15	Coronary revascularization, familial risk factor, acute myocardial infarction, coronary artery disease, coronary artery bypass, coronary angioplasty, unstable angina, coronary arterial stent insertion, myocardial infarction, ventricular extrasystoles	Very high cardiovascular risk
16	Coronary revascularization, familial risk factor, coronary artery disease, acute myocardial infarction, coronary artery bypass, hypertension, type 2 diabetes mellitus, coronary arterial stent insertion, percutaneous coronary intervention, dyslipidemia, abnormal lipoprotein	Very high cardiovascular risk
17	Familial risk factor, coronary revascularization, coronary arterial stent insertion, hypertension, acute myocardial infarction, unstable angina, coronary artery disease	Very high cardiovascular risk
18	None reported	Not applicable
19	Acute myocardial infarction, coronary revascularization, bradycardia	Very high cardiovascular risk
20	Coronary revascularization, coronary artery disease, familial risk factor, acute myocardial infarction	Very high cardiovascular risk

Supplementary Table 3 Change in LDL-C from baseline to Week 12 and Week 24

Patient number	Treatment*	Baseline LDL-C (mg/dL)	LDL-C at Week 12 (mg/dL)	Change in LDL-C from baseline to Week 12 (mg/dL)	LDL-C at Week 24 (mg/dL)	Change in LDL-C from baseline to Week 24 (mg/dL)
1	Alirocumab 75/150 mg Q2W	167	74	–93	75	–92
3	Alirocumab 75/150 mg Q2W	290	176	–114	125	–165
4	Alirocumab 75/150 mg Q2W	150	71	–79	57	–93
5	Alirocumab 75/150 mg Q2W	114	54	–60	104	–10
6	Alirocumab 75/150 mg Q2W	166	60	–106	58	–108
7	Alirocumab 150 mg Q2W	205	133	–72	NA	NA
8	Alirocumab 150 mg Q2W	180	141	–39	127	–53
9	Alirocumab 150 mg Q2W	402	310	–92	354	–48
10	Alirocumab 75/150 mg Q2W	140	150	+10	92	–48
12	Placebo	296	292	–4	358	+62
13	Placebo	163	155	–8	155	–8
14	Placebo	167	161	–6	152	–15
15	Placebo	142	147	+5	159	+17
16	Placebo	164	153	–11	201	+37
17	Placebo	232	237	+5	180	–52

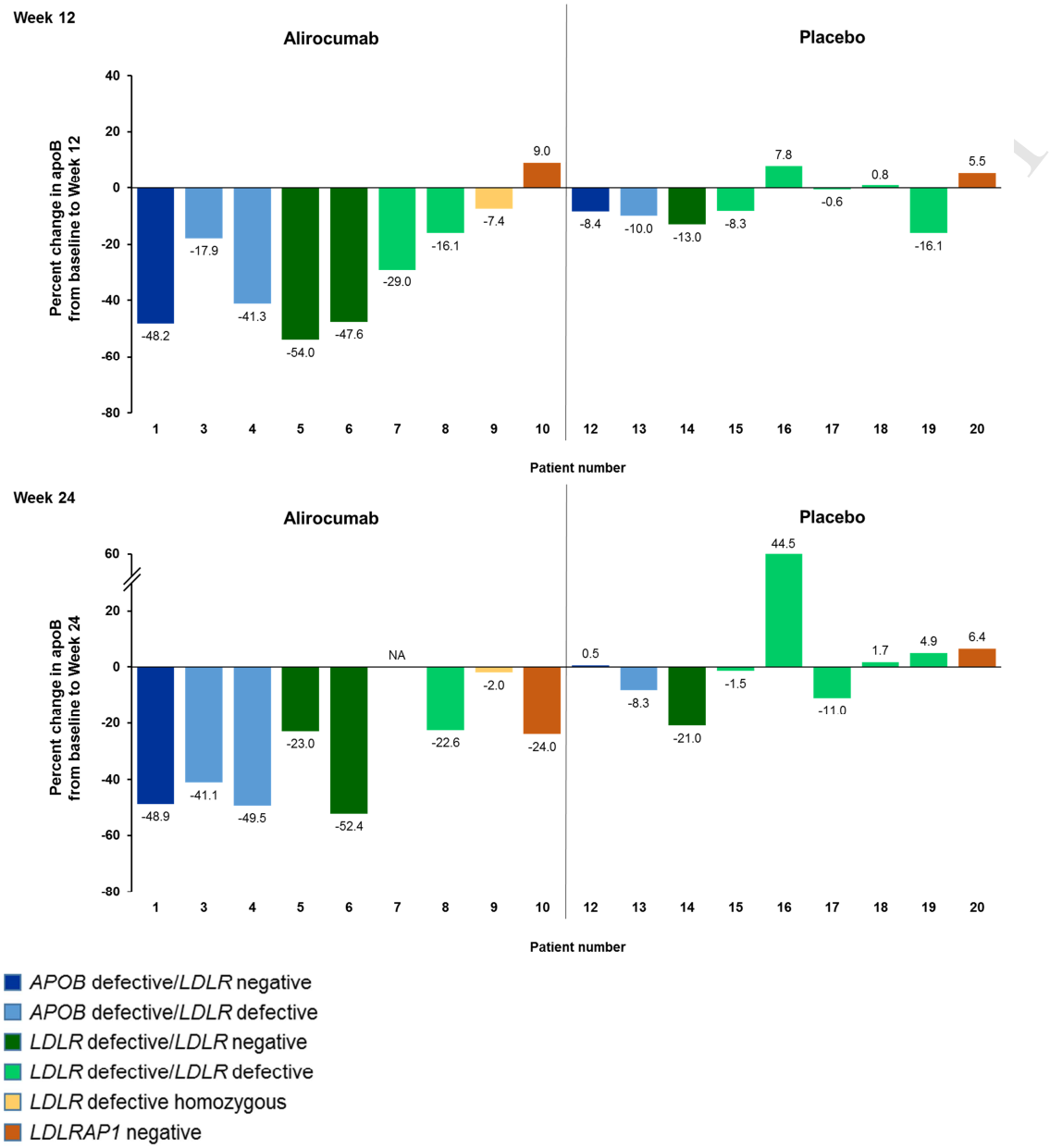
18	Placebo	192	209	+17	223	+31
19	Placebo	208	174	–34	207	–1
20	Placebo	141	147	+5	149	+8

LDL-C, low-density lipoprotein cholesterol; NA, not available; Q2W, every 2 weeks.

Data was not available for Patient 2 (*APOB* defective/*LDLR* negative). Patient 7 was from the 12-week Phase 2 study hence no data was available at Week 24. Patient 11 (*PCSK9* GOF and *LDLR* negative) received a different alirocumab administration regimen and is not included in this table.

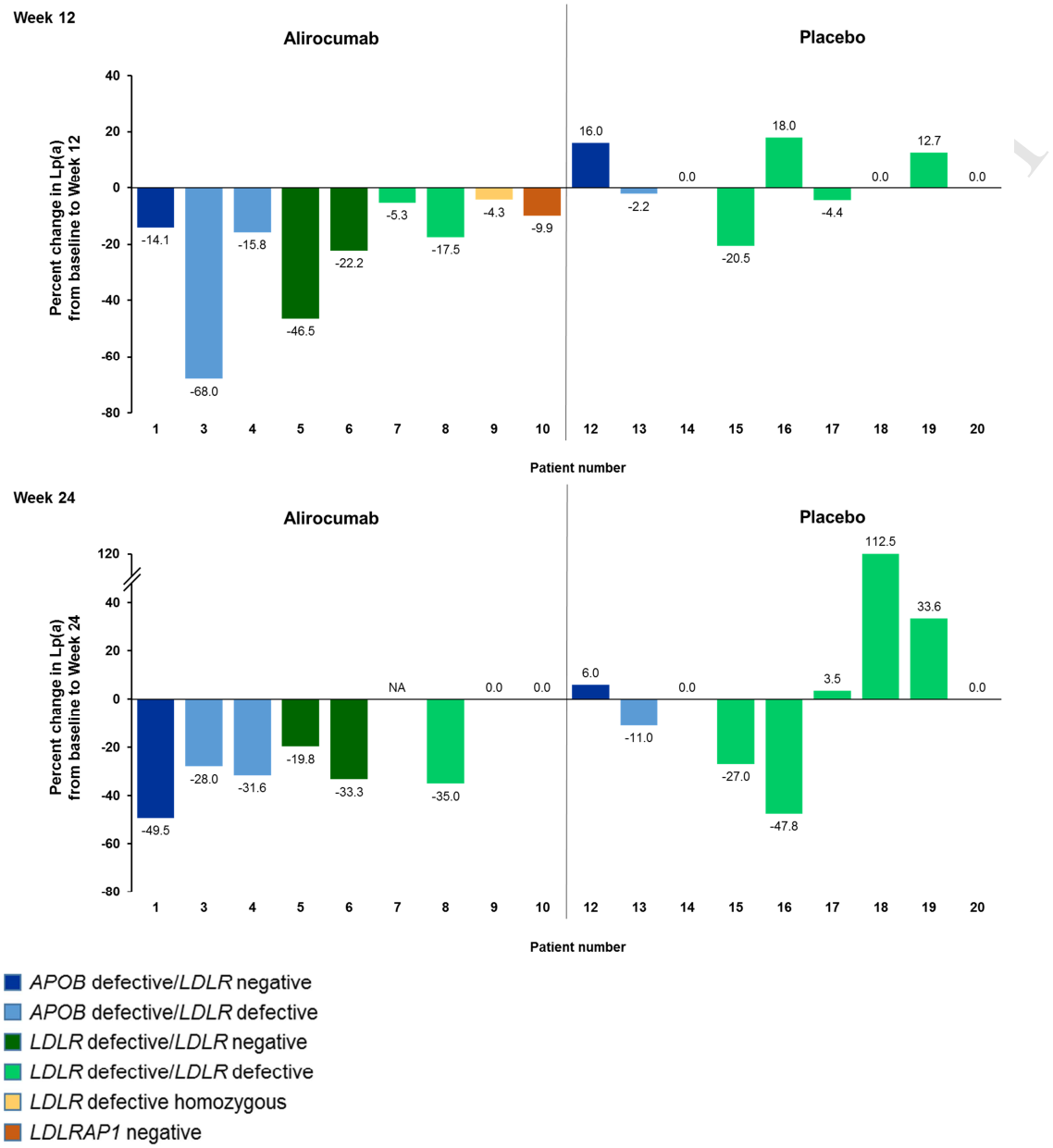
*Alirocumab 75 mg Q2W was increased to 150 mg Q2W at Week 12 depending on LDL-C at Week 8.

Supplementary Figure 1 Percentage reduction from baseline in apoB at Weeks 12 and 24



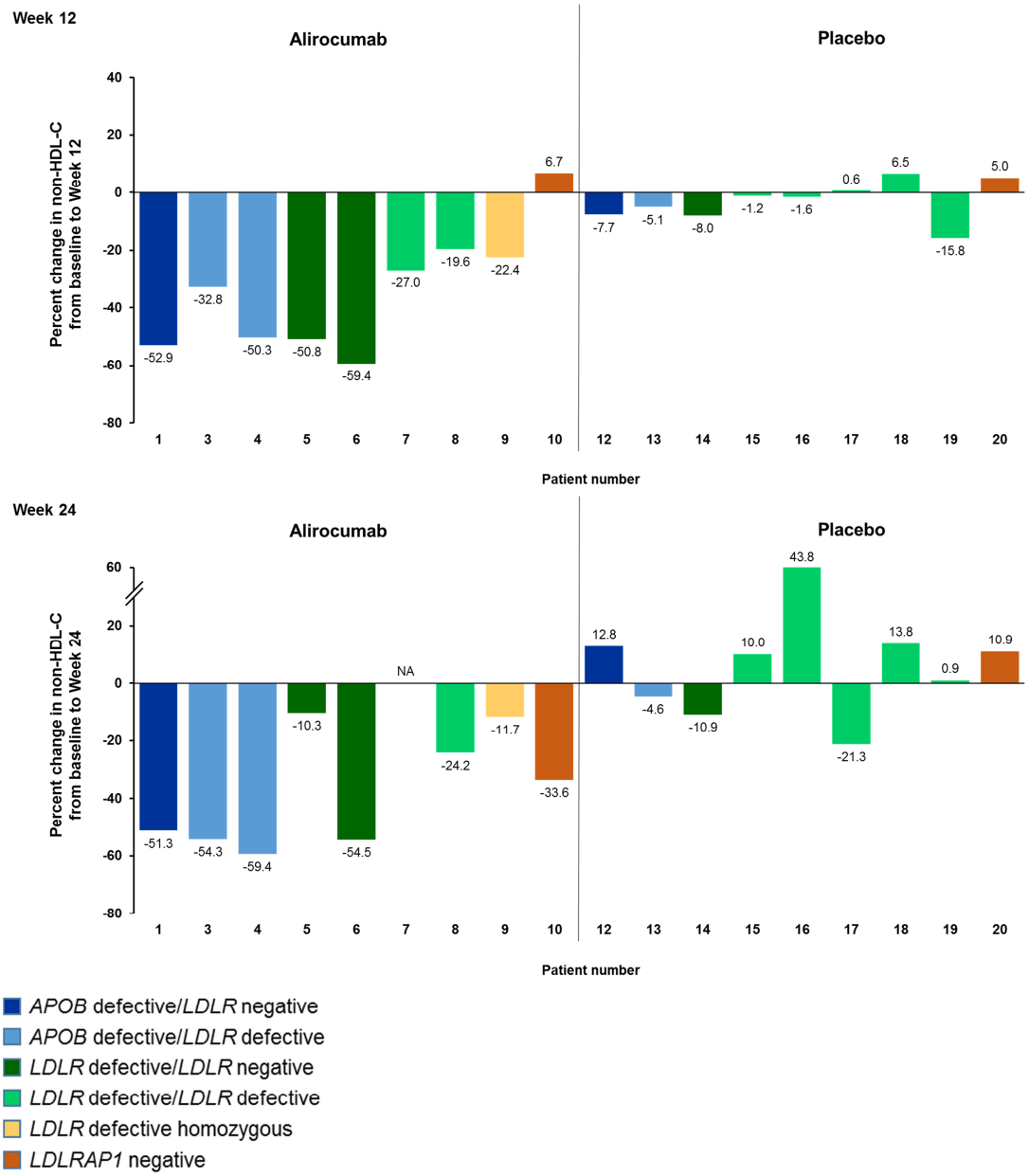
Data was not available for Patient 2 (*APOB* defective/*LDLR* negative). Patient 7 was from the 12-week Phase 2 study hence no data was available at Week 24. Patient 11 (*PCSK9* GOF and *LDLR* negative) received a different alirocumab administration regimen and is not included in this figure. *APOB*, apolipoprotein B; *LDLR*, low-density lipoprotein receptor; *LDLRAP1*, LDLR adaptor protein 1; NA, not available.

Supplementary Figure 2 Percentage reduction from baseline in Lp(a) at Weeks 12 and 24



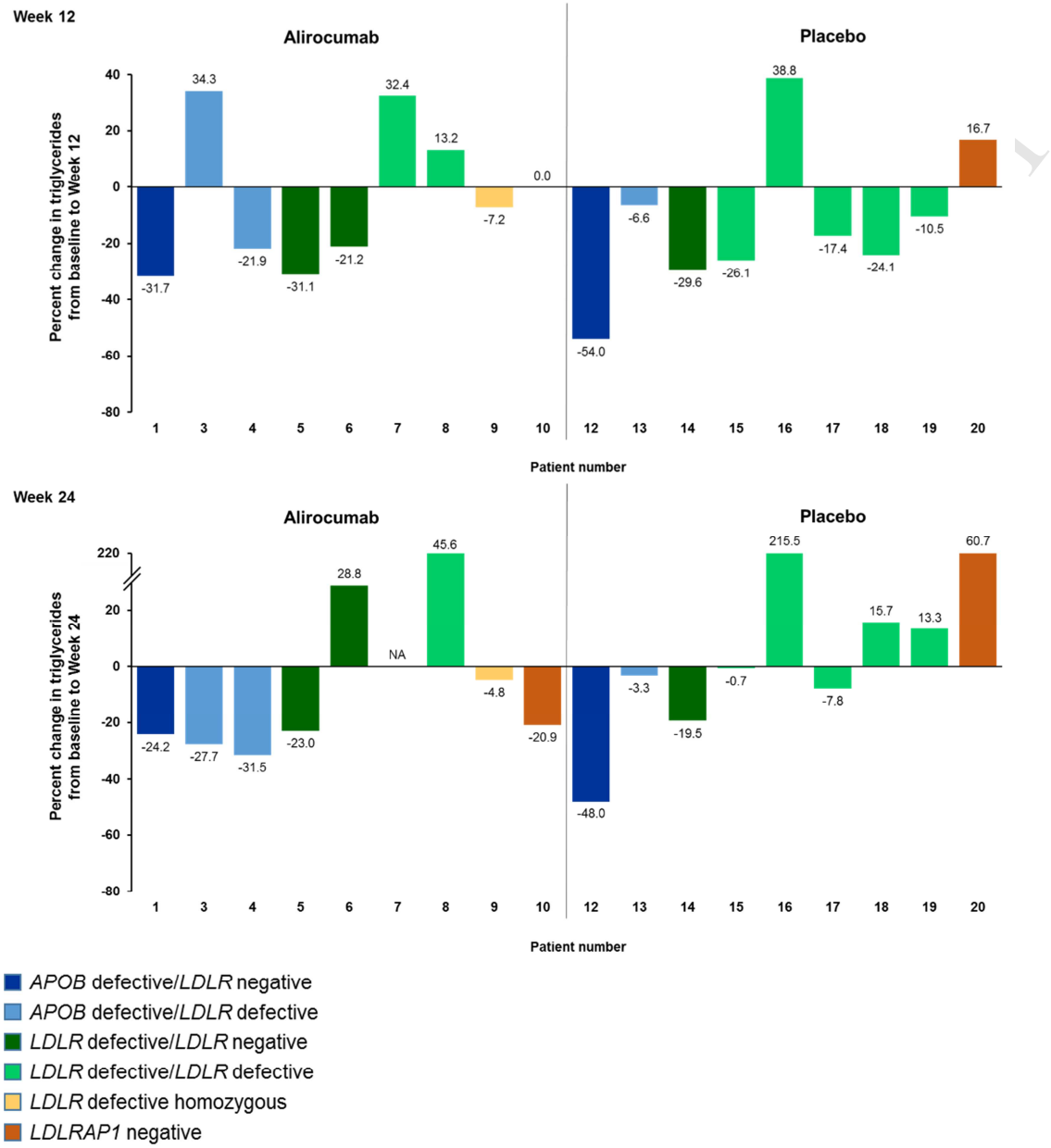
Data was not available for Patient 2 (*APOB* defective/*LDLR* negative). Patient 7 was from the 12-week Phase 2 study hence no data was available at Week 24. Patient 11 (*PCSK9* GOF and *LDLR* negative) received a different alirocumab administration regimen and is not included in this figure. *APOB*, apolipoprotein B; *LDLR*, low-density lipoprotein receptor; *LDLRAP1*, LDLR adaptor protein 1; Lp(a), lipoprotein (a); NA, not available.

Supplementary Figure 3 Percentage reduction from baseline in non-HDL-C at Weeks 12 and 24



Data was not available for Patient 2 (*APOB* defective/*LDLR* negative). Patient 7 was from the 12-week Phase 2 study hence no data was available at Week 24. Patient 11 (*PCSK9* GOF and *LDLR* negative) received a different alirocumab administration regimen and is not included in this figure. *APOB*, apolipoprotein B; *LDLR*, low-density lipoprotein receptor; *LDLRAP1*, LDLR adaptor protein 1; non-HDL-C, non-high-density lipoprotein cholesterol.

Supplementary Figure 4 Percentage reduction from baseline in triglycerides at Weeks 12 and 24



Data was not available for Patient 2 (*APOB* defective/*LDLR* negative). Patient 7 was from the 12-week Phase 2 study hence no data was available at Week 24. Patient 11 (*PCSK9* GOF and *LDLR* negative) received a different alirocumab administration regimen and is not included in this figure. APOB, apolipoprotein B; LDLR, low-density lipoprotein receptor; LDLRAP1, LDLR adaptor protein 1; NA, not available.