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# Lipoprotein apheresis and PCSK9 inhibitors for severe familial hypercholesterolaemia: Experience from Australia and New Zealand

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**Short title**: LDL-apheresis and PCSK9 inhibitors

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**Abstract**

**Introduction**: Severe familial hypercholesterolaemia (FH) causes premature disability and death due to atherosclerotic cardiovascular disease and is refractory to standard lipid-lowering therapies. Lipoprotein apheresis (LA) has long been a standard of care for patients with severe FH, but is invasive, expensive and time-consuming for patients and their caregivers. Newer drug therapies, including the proprotein convertase subtilisin/kexin type 9 (PCSK9) inhibitors, may reduce the need for LA.

**Materials and methods**: We audited the records of 16 patients (eight homozygous, eight heterozygous) treated with LA in Australia and New Zealand, 14 of whom subsequently commenced PCSK9 inhibitor therapy. LA was performed by cascade filtration in all centres.

**Results**: LDL-cholesterol was acutely lowered by 69 ± 7% in patients with homozygous FH and by 72 ± 9% in those with heterozygous FH, representing time-averaged reductions of 36 ± 12% and 34 ± 5%, respectively. LA was well-tolerated, and patients reported comparable quality of life to population and disease-related norms. After commencement of PCSK9 inhibitors, four of seven patients with homozygous FH had meaningful biochemical responses, with a reduction in the frequency of LA permitted in one patient and complete cessation in another. Four of seven patients with heterozygous FH were able to be managed without LA after commencing PCSK9 inhibitors.

**Conclusion**: While PCSK9 inhibitors have reduced the need for LA, some patients with severe FH continue to require LA, and will require it for the foreseeable future. However,

emerging therapies, including angiopoetin-like 3 inhibitors, may further reduce the need for LA.

**Key words**: Lipoprotein apheresis, familial hypercholesterolaemia, PCSK9 inhibitors, health-related quality of life.

## Introduction

Familial hypercholesterolaemia (FH) is an autosomal co-dominant disorder of lipoprotein metabolism characterised by increased plasma concentrations of low-density lipoprotein (LDL)-cholesterol. It is most commonly caused by loss-of-function variants in *LDLR*, which encodes the LDL receptor. Other causes include loss-of-function variants in *APOB*, encoding the ligand for the LDL receptor, apolipoprotein (apo)-B100 (apoB); and rarely gain-of-function variants in *PCSK9*, which encodes proprotein convertase subtilisin/kexin type 9 (PCSK9), a protein that inhibits LDL receptor recycling. Each of these negatively impacts LDL receptor function, leading to decreased LDL particle catabolism.[1,2]

A pathogenic gene variant in a single allele causes heterozygous FH, which results in plasma LDL-cholesterol concentrations of 5 - 10 mmol/L and if untreated symptomatic atherosclerotic cardiovascular disease (ASCVD) by middle age.[1] There is considerable heterogeneity in the clinical presentation of heterozygous FH, which is influenced by the severity of variant, tolerability and response to treatment, and the presence of additional cardiovascular risk factors.

Homozygous FH is caused by the presence of pathogenic variants in both alleles. If the variants are non-identical, as seen in a majority of cases in many populations, the condition may otherwise be referred to as compound heterozygous FH. This is clinically and metabolically indistinguishable from true homozygous FH. LDL-cholesterol concentrations

are usually > 10 mmol/L, and if untreated, death from ASCVD can occur in childhood or adolescence.

The mainstay of the medical treatment of FH is diet and lifestyle modification, attention to co-morbid risk factors contributing to ASCVD, and pharmacotherapy with statins, ezetimibe, PCSK9 inhibitors, and sometimes other agents such as bile acid sequestrants.[3,4] Patients with heterozygous FH often respond satisfactorily to this approach, having at least one normal copy of *LDLR* that can be upregulated by statin therapy, with the resultant protein's function enhanced by PCSK9 inhibitor therapy. PCSK9, secreted by the liver, binds to LDL receptors and in doing so prevents their recycling. PCSK9 inhibitors disrupt this process, thereby increasing LDL particle catabolism.[5] As these agents depend on at least some residual LDL receptor function, they are generally ineffective in patients homozygous for null variants in *LDLR*, as are statins.[6] Patients with homozygous FH may therefore require advanced therapies including lipoprotein apheresis (LA) or, rarely, liver transplantation.[1,7-10]

LA is a therapeutic modality involving the regular extracorporeal removal of LDL and usually other apoB-containing lipoproteins including lipoprotein(a) [Lp(a)], an LDL-like particle with atherogenic properties whose plasma concentrations may be increased independently of LDL-cholesterol, but for which few effective treatments are available.[11,12] LA has been used in Australia and New Zealand since 2003, predating the availability of PCSK9 inhibitor therapy. We present the outcome of our first 16 patients to undergo LA for the treatment of severe FH, and include data relating to the use of PCSK9 inhibitors.

## Materials and methods

*Data collection*

We obtained data from hospital and clinic records including pathology and medical imaging for all patients who underwent treatment with LA at our centres between December 2003 and December 2019. For each patient we recorded demographics; past medical history; examination findings; genetic characterisation of FH; current and previous lipid-lowering therapies including biochemical and clinical treatment response and adverse effects; details of the number and frequency of LA procedures; and adverse effects associated with LA.

Adverse effects recorded were those known to occur with LA including headache, nausea, hypotension, chest pain and anaphylactoid reactions; deficiency states requiring supplementation of iron, calcium or cryoprecipitate; and any others observed and documented by clinical staff. Anaphylactoid reactions were classified as minor, which were transient reactions occurring during the LA procedure and requiring a pause or change in flow rate, but not requiring termination of the procedure; or major, which were those reactions requiring termination of the procedure to ensure patient comfort or safety.

*Eligibility*

Patients are generally considered for LA where there has been an unsatisfactory biochemical response, or evidence of progressive ASCVD, on standard therapies.[13-15] Local criteria for

consideration of LA are: homozygous FH and on-treatment LDL-cholesterol > 7 mmol/L; or heterozygous FH with documented ASCVD and on-treatment LDL-cholesterol > 5 mmol/L.[4]

*LA technique*

LA in all centres was performed by cascade filtration with centrifugal cell separation (Perth, Christchurch, Hobart: COBE Spectra Apheresis System; CaridianBCT Inc., Colorado, USA. Melbourne: COM.TEC; Fresenius Kabi AG, Bad Homburg, Germany). This system involves an initial centrifugal separation of plasma from cells, with the latter returned to the patient. The plasma passes through a column containing a filtration membrane (Evaflux 5A; Kuraray Medical Inc., Tokyo, Japan) which by size exclusion removes larger particles, including LDL and Lp(a), relatively preserving smaller components such as high-density lipoprotein (HDL) and albumin. Calcium is removed by the procedure and is routinely supplemented.

*Laboratory methods*

Total cholesterol, HDL-cholesterol and triglyceride were measured by routine clinical chemistry methods (Perth, Christchurch: Architect chemistry series analysers (Abbott, Chicago, IL, USA); Hobart, Melbourne: Cobas chemistry series analysers (Roche, Basel, Switzerland)). LDL-cholesterol was calculated by the Friedewald formula.[16] Lp(a) was measured by turbidimetry (Perth, Christchurch: QUANTIA assay on Architect immunoassay series analysers); or rate nephelometry (Melbourne, Hobart: LPAX assay on IMMAGE analyser (Beckman Coulter, Brea, CA, USA)).

*Quality of life survey*

All patients were asked to complete the RAND 36-item Health Survey, developed from a cohort of patients with chronic disease in the Medical Outcomes Study, giving scores for eight domains of health and wellbeing.[17]

*Cost of treatment*

We estimated the price of each LA procedure, including drugs and other consumables such as filtration columns and lines, staffing and an infrastructure loading of 20%, adjusted to 2020 Australian and New Zealand dollars.[18,19] We multiplied this by the mean number of treatments per annum to calculate the annual cost of LA per patient. The cost of evolocumab was determined from the reimbursement amount provided by the Australian Pharmaceutical Benefits Scheme, and the same cost for equivalent doses was assumed for alirocumab which, while not yet listed for public reimbursement in Australia, some of our patients continued to receive after previous enrolment in clinical trials. Neither evolocumab nor alirocumab currently attracts government subsidy in New Zealand.

*Calculations and statistical analyses*

We calculated the acute reduction in plasma cholesterol and lipoprotein concentrations attributable to each LA procedure including total cholesterol (TC), LDL-cholesterol, HDL-cholesterol, triglycerides (TG), and Lp(a). We calculated the mean change in each parameter for all procedures for each patient and calculated the mean and standard deviation for all patients combined.

Owing to the "sawtooth" pattern of rebound in lipoproteins that occurs between LA procedures, time-averaged concentrations were used to assess the response to treatment. We determined the time-averaged concentrations of TC, LDL-cholesterol and Lp(a) using the method described by Kroon et al., whereby the time-averaged concentration, $C_{AVG} = C_{MIN} + k(C_{MAX}-C_{MIN})$.[20] For the first-order rate-constant *k*, we used Kroon's value of 0.73 for patients with heterozygous FH and a *k* value of 0.65 for patients with homozygous FH, based on a study of lipoprotein rebound kinetics in homozygous FH.[21]

A Student's t-test was used to determine the significance of the acute and time-averaged reduction in lipids and lipoprotein concentrations.

*Ethical considerations*

This study was approved by the Royal Perth Hospital and Austin Health Human Research Ethics Committees and was exempted from ethical review by the New Zealand Health and Disability Ethics Committees. All adult patients gave informed written consent, and patients aged < 18 years at the time of data collection gave assent with at least one parent providing consent.

**Results**

*Patient characteristics*

Patient demographics, relevant clinical information, and lipid profile on maximum lipid-lowering therapy prior to commencing LA are shown in **Table 1** for the 14 adult and two paediatric patients enrolled in the study. Indications for LA were homozygous FH (eight patients, including four with compound heterozygous FH) and heterozygous FH (eight patients, including one with familial defective apo-B100). Patient 10 had both heterozygous FH and hyperLp(a)aemia with progressive CAD. With maximal lipid-modifying therapy before the commencement of LA, LDL-cholesterol was 8.2 ± 1.1 mmol/L in patients with homozygous FH and 6.7 ± 1.9 mmol/L in patients with heterozygous FH. Prior to the availability of PCSK9 inhibitors, patient 3 underwent liver transplantation, ceased treatment with LA and has been reported elsewhere.[7] Of the ten patients remaining on LA, the mean duration of follow-up since commencement is 9.6 years (range 5 - 17 years).

*Biochemical effects of LA*

The effect of LA on the plasma concentrations of lipids and lipoproteins in patients with FH, prior to the commencement of PCSK9 inhibitors or other novel lipid lowering therapies such as mipomersen or lomitapide, is shown in **Table 2**. LDL-cholesterol was acutely lowered by a mean of 69 ± 7% in patients with homozygous FH and by 72 ± 9% in those with heterozygous FH, with a time-averaged reduction of 36 ± 12% and 34 ± 5% respectively.

*Effect of PCSK9 inhibitor therapy on LDL-cholesterol concentrations and LA frequency*

Seven patients with homozygous FH were commenced on PCSK9 inhibitors. In four of these treated patients, the pre-apheresis LDL-cholesterol concentrations were lower than time-averaged LDL-cholesterol concentrations prior to commencement of PCSK9 inhibitors. In two cases, LDL cholesterol-lowering was deemed sufficient to reduce the frequency of LA, and in one, to cease LA altogether (**Table 3**).

Of seven patients with heterozygous FH commenced on PCSK9 inhibitors, four were able to cease LA. Patient 16, severely intolerant to all previously attempted lipid lowering drugs and receiving four-weekly LA prior to commencement of a low monthly dose of alirocumab, increased frequency of LA from monthly to fortnightly in an attempt to achieve better control of LDL-cholesterol concentrations. There had not been evidence of progressive CAD in this case.

*Safety and tolerability of LA*

Adverse events occurred during 5.5% of LA procedures (**Table 4**). Anaphylactoid reactions, as described in **Table 4**, were mainly attributed to a particular batch of LA columns used in two centres across a particular period of time. Symptoms and signs were chiefly hypotension and flushing, and the reactions were managed by slowing the rate of LA. This reaction has not occurred since 2011; therefore, the current adverse event rate is lower.

One patient developed prolonged bleeding from her venepuncture sites and was found to have low fibrinogen levels, which was remedied by administering cryoprecipitate at the end

of each procedure. Four patients developed iron deficiency anaemia, requiring iron supplementation by the oral or parenteral route.

*Stability of ASCVD*

No patients reported symptoms suggestive of progressive ASCVD whilst on LA. As this was a retrospective review of routine clinical care without an *a priori* study protocol, there were insufficient imaging and other empirical data to meaningfully report.

*Quality of life*

All patients had been established on regular lipoprotein apheresis prior to completion of the SF-36 survey. The results are presented in **Figure 1**. Overall, they suggested that our patients with severe hypercholesterolaemia undergoing treatment with LA experienced quality of life that was not inferior to the Australian population with ASCVD, and in several domains to that of a healthy population.

*Cost*

In Australia, the mean cost of each LA procedure was AUD $2,178 (USD $1,568) and in New Zealand it was NZD $2,689 (USD $1,776). The mean annual cost of providing LA across both countries was AUD $50,256 (range $32,907 - $66,355), NZD $54,779 (range $35,868 - $72,327) or USD $36,184 ($23,693 - $47,775) prior to the commencement of PCSK9 inhibitors. After the commencement of PCSK9 inhibitors, the total cost of combined therapy was estimated to decrease by a mean of AUD $3,491, NZD $3,805 or USD $2,513

per annum per patient with homozygous FH (4% cost reduction); and by AUD $19,287, NZD $21,023 or USD $13,887 per annum per patient with heterozygous FH (32% cost reduction).

## Discussion

We present the first comprehensive description of the use of LA in Australia and New Zealand, with a contemporary perspective on the real-world efficacy of PCSK9 inhibitors in reducing the need for LA in patients with severe FH. LA was generally well-tolerated, and no patient ceased LA because of effects on quality of life (QOL). After the commencement of PCSK9 inhibitor therapy, LA was able to be ceased in only one of seven patients with homozygous FH, but in a majority of those with heterozygous FH. The introduction of PCSK9 inhibitors was associated with an estimated overall cost-saving to the health system.

### *PCSK9 inhibitors and the need for LA*

Two major studies have examined the ability of PCSK9 inhibitors to reduce the need for LA: the open-label, single-arm TAUSSIG study of evolocumab, and the randomised, double-blind, placebo-controlled ODYSSEY ESCAPE study of alirocumab. These studies included phenotypically similar cohorts to ours, and found that a small minority of patients with homozygous FH, but around half of those with heterozygous FH, could be withdrawn from LA following the use of PCSK9 inhibitors.[2,22]

Several major factors might have influenced the ability of PCSK9 inhibitors to reduce the need for LA in the aforementioned studies and in our cohort. First, the stringency of local eligibility criteria for LA dictates the average phenotypic severity of disease. Patients with lower baseline concentrations of LDL-cholesterol receiving LA are more likely to achieve therapeutic targets when transitioned to PCSK9 therapy, and therefore not require ongoing

LA. This was illustrated in a German study of 110 hypercholesterolaemic patients, most of whom did not have FH and who had lower baseline LDL-cholesterol concentrations than our cohort. The addition of PCSK9 inhibitor therapy enabled an extra 41.8% of patients to achieve LDL-cholesterol < 1.8 mmol/L.[23] Only one of our 16 patients achieved this target with combined LA and PCSK9 inhibitor therapy.

Second, the treatment target may vary according to local clinical protocols and practices, and patient preferences. For example, in one multinational randomised, controlled, open-label study of the initiation of evolocumab in patients who mostly had a background of heterozygous FH and ASCVD, 84% of patients receiving evolocumab were able to cease LA after six weeks, compared with 10% of those who did not, using an LDL-cholesterol treatment target of <2.6 mmol/L.[24] By contrast, around 40-60% of patients were able to cease LA after commencement of a PCSK9 inhibitor in studies using a target of <1.8 mmol/L.[2,22,24]

Third, ongoing increased plasma concentrations of Lp(a) may be more broadly accepted as a separate indication to continue LA, even where the on-treatment LDL-cholesterol concentration is satisfactory without LA, in some jurisdictions.[25]

*Response to PCSK9 inhibitors in homozygous FH*

Whilst it is predictable that patients with homozygous FH due to LDL receptor-null variants respond poorly to PCSK9 inhibitor therapy, some patients with receptor-defective variants may also poorly respond.[6] Furthermore, individual patients with the same receptor-defective

variants may respond differently from one another, for unclear reasons.[26] Patient 1 responded well to PCSK9 inhibitor therapy, with a reduction in LA frequency and a reduction in pre-apheresis LDL-cholesterol of > 50%. By contrast, patient 2 showed a complete absence of response, despite homozygosity for the same pathogenic variant in *LDLR*. Variability in response to PCSK9 inhibitor therapy with this receptor-defective variant, c.681C>G (also known as Afrikaaner-1), has been shown both *in vivo* and *in vitro*.[26] Therefore, a satisfactory response to PCSK9 inhibitor therapy cannot yet be predicted by receptor-defective *LDLR* status alone. Phenotypic predictors that might augment the use of genotype to predict treatment response are also unknown and warrant further investigation.

*Health-related quality of life*

Although LA might be expected to be a burdensome treatment, our patients generally reported similar or better health-related quality of life (HRQOL) than the broader population with ischaemic heart disease. Our data are consistent with the findings of studies designed to determine the effects of LA on measures of HRQOL, which have not found a significant adverse impact. [27,28] None of our patients elected to cease LA because of detrimental effects on their QOL. Nonetheless, QOL and mental health remain important considerations in patient selection and monitoring.

*Economic implications*

The introduction of PCSK9 inhibitors was associated with substantial cost savings in patients with both homozygous and heterozygous FH. Very few comprehensive analyses of the health

economic implications of LA have been published, and none since the introduction of PCSK9 inhibitors. Based on an estimate of 778 patients with FH in Ontario, Canada, a 2006 analysis estimated the cost of fortnightly LA per CAD death avoided at Can$18.8 M.[29] A number of assumptions used in the modelling may have affected the validity of this conclusion, including the use of Framingham-based models to estimate cardiovascular risk (not validated in the FH population); the patient characteristics assumed for the purposes of risk estimation; and a lack of robust data illuminating the cardiovascular risk reduction attributable to LA. Furthermore, CAD deaths avoided is not the only outcome by which lipid-lowering therapy is usually judged: avoidance of nonfatal major adverse cardiovascular events and the associated quality life years gained are also important.

*Limitations*

Most patients in our study, including those who ceased LA after commencement of PCSK9 inhibitors, did not achieve the guideline recommended target of LDL-cholesterol < 1.8 mmol/L.[30,31] However, clinical decisions to cease LA took into account patient preferences in the context of improved lipid profiles and the presumed commensurate decrease in ASCVD risk after the commencement of PCSK9 inhibitors. We cannot comment on the stability of ASCVD based on imaging in our patients, but none experienced major adverse cardiovascular events while on LA. Collection of formal quality of life data prior to commencement of LA is not routine, so we lacked baseline SF-36 scores with which to compare the on-LA scores, and an assessment of untreated matched controls was not possible. A comprehensive health economic evaluation of the treatment of patients with

severe FH was beyond the scope of the present report. A comprehensive, contemporary health economic evaluation would be of value, but would require accurate risk modelling and risk reduction estimates attributable to LA and contemporary pharmacological therapies in patients with homozygous and heterozygous FH.

**Conclusion**

PCSK9 inhibitors have helped to mitigate some of the problems associated with LA, including its cost and geographical accessibility. Advances in drug therapy, particularly the development of agents that enhance the non-LDL receptor-mediated clearance of atherogenic lipoproteins, [32-34] may further reduce the need for LA. However, LA maintains its place in the management of FH as the only currently available therapeutic modality able to enhance the clearance of lipoproteins independently of the LDL receptor. This is particularly important in homozygous FH, in which there is a demonstrated need for multimodal therapy to reduce LDL-cholesterol and improve survival.[35 36]

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**Figure 1 Legend:**

Health-related quality of life, as determined by SF-36 Questionnaire, in Australian and New Zealand patients undergoing lipoprotein apheresis, compared with Australian reference values for patients with ischaemic heart disease,[37] and Australian population norms[38]

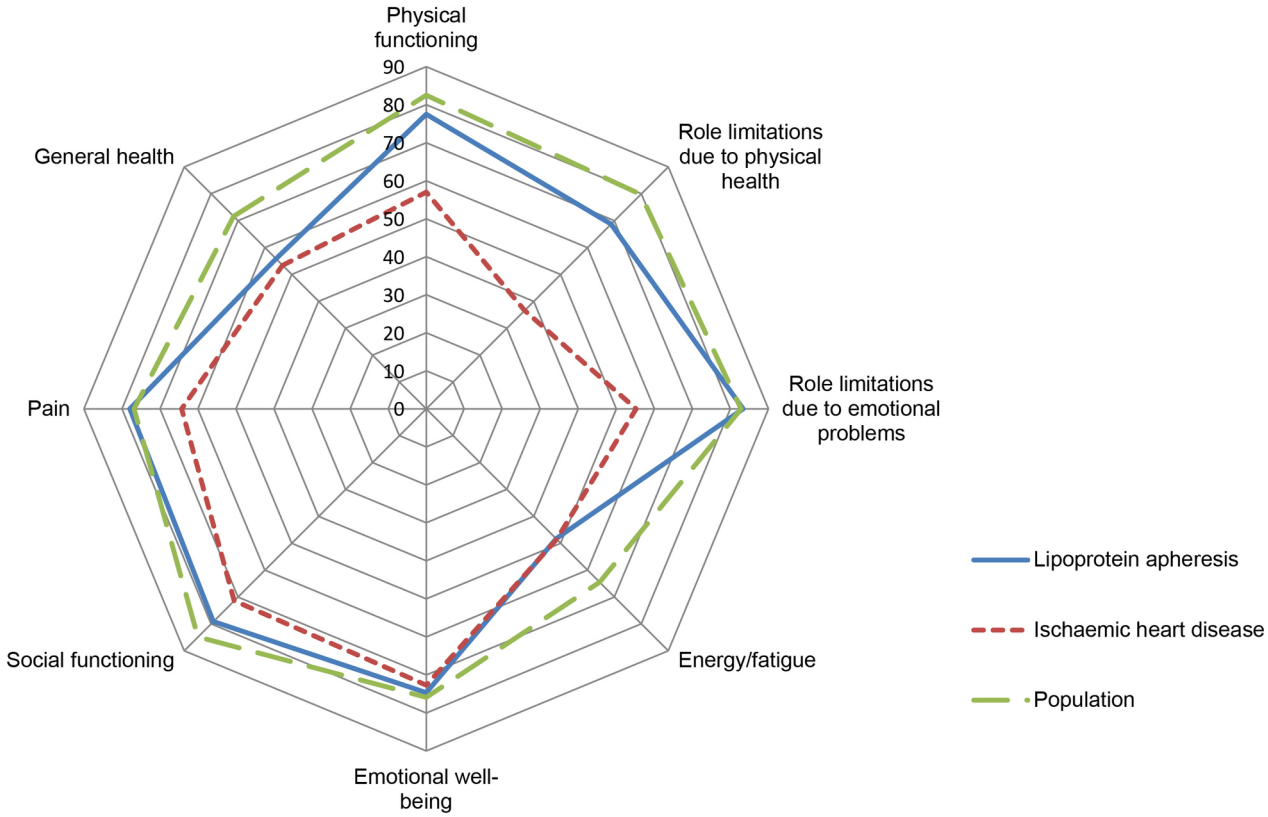


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**Table 1:** Patient demographics

| Patient | Age | Sex | Diagnosis | DLCNS | Gene Variant | Cardiovascular history | Lipid-modifying therapies | TC (mmol/L) | LDL-C (mmol/L) | HDL-C (mmol/L) | TG (mmol/L) | Lp(a) (g/L) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 1 | 39 | F | HoFH | 22 | c.681C>G, c.681C>G | Severe AS, AVR | Rosuvastatin 40 mg, ezetimibe 10 mg | 12.5 | 10.8 | 1.3 | 0.8 | NA |
| 2 | 14 | F | HoFH | 25 | c.681C>G, c.681C>G | No CV history | Rosuvastatin 40 mg, ezetimibe | 12.7 | 10.1 | 1.0 | 0.9 | 1.2 |
| 3 | 24 | M | HoFH | 19 | c.686A>T, c.686A>T | CABG age 14 | Rosuvastatin 40 mg, ezetimibe 10 mg | 14.9 | 13.8 | 0.7 | 0.9 | NA |
| 4 | 39 | M | HoFH | 17 | Not done | CABG age 39 | Rosuvastatin 40mg, ezetimibe 10 mg | 9.6 | 7.7 | 1.4 | 1.1 | 1.86 |
| 5 | 33 | F | HoFH | 25 | c.681C>G, c.523G>A | No CV history | Rosuvastatin 40 mg, ezetimibe 10 mg | 8.2 | 6.9 | 0.9 | 1.0 | < 0.03 |
| 6 | 40 | F | HoFH | 26 | c.661G>A, c.681C>G | Asymptomatic CAD | Atorvastatin 80 mg, ezetimibe | 10.4 | 7.6 | 1.3 | 5.1 | 0.10 |
| 7 | 44 | F | HoFH | 26 | c.660delC, c.326G>A | Asymptomatic CAD | Atorvastatin 80 mg, ezetimibe 10 mg | 16.1 | 14.5 | 0.7 | 1.5 | 0.50 |

| | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 8 | 14 | F | HoFH | 24 | c.417C>G, c.693C>A | No CV history | Simvastatin 40 mg, ezetimibe 10 mg | 12.7 | 10.4 | 1.6 | 1.5 | 0.50 |
| 9 | 52 | M | HeFH | 25 | c.301G>A | PTCI age 26, CABG age 45, statin intolerance | Ezetimibe 10 mg | 11.2 | 9.2 | 1.0 | 2.2 | 0.75 |
| 10 | 49 | F | HeFH | 26 | c.681C>G | Unstable angina, PTCI | Rosuvastatin 40 mg, ezetimibe 10 mg, fenofibrate 145 mg | 5.8 | 3.9 | 1.3 | 1.2 | 1.86 |
| 11 | 50 | F | HeFH | 8 | Not found | Stable angina | Atorvastatin 80 mg, ezetimibe 10 mg | 11.3 | 7.9 | 1.8 | 3.4 | NA |
| 12 | 59 | M | HeFH | 20 | c.191-2A>G | Mixed multivessel CAD | Atorvastatin 80 mg, ezetimibe 10 mg, fenofibrate 145 mg | 7.1 | 5.4 | 1.1 | 1.3 | 0.37 |
| 13 | 68 | F | HeFH | 6 | Not found | Angina, PTCI | Rosuvastatin 40 mg, ezetimibe 10 mg, fenofibrate 145 mg | 8.4 | 5.8 | 1.5 | 2.4 | 0.08 |
| 14 | 33 | F | HeFH | 19 | Not done | No CV history | Rosuvastatin 40mg, ezetimibe 10 mg | 7.3 | 5.6 | 0.9 | 1.9 | 0.38 |

| | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 15 | 40 | F | HeFH | 26 | c.1813C>T | PTCI age 32 | Nil due to multiple intolerances | 11.6 | 9.0 | 1.2 | 2.9 | 2.00 |
| 16 | 71 | F | HeFDB | 26 | *APOB* c.10580G>A | CABG age 56; statin intolerance | Nil due to multiple intolerances | 9.3 | 7.1 | 1.8 | 1.1 | 0.40 |

Age is in years at the time of commencement of lipoprotein apheresis. All lipid and lipoprotein concentrations are on maximal lipid lowering therapy, as specified in the Table, prior to lipoprotein apheresis or PCSK9 inhibitors. Gene variants named according to reference sequences *LDLR:* NM_000527.5 (Patients 1-15) and *APOB:* NM_000384.3 (Patient 16).

Abbreviations: AS, aortic stenosis; AVR, aortic valve replacement; CABG, coronary artery bypass graft; CAD, coronary artery disease; DLCNS, Dutch Lipid Clinic Network Score; HDL-C, high-density lipoprotein-cholesterol; HeFDB, heterozygous familial defective apolipoprotein-B100; HeFH, heterozygous familial hypercholesterolaemia; HoFH, homozygous familial hypercholesterolaemia; LDL-C, low-density lipoprotein-cholesterol; Lp(a), lipoprotein(a); NA, not available or not applicable; PTCI, percutaneous transluminal coronary intervention; TC, total cholesterol; TG, triglycerides

**Table 2:** Plasma lipid and lipoprotein concentrations, and response to LA in patients with homozygous and heterozygous FH on standard therapies, not including PCSK9 inhibitors; mean ± SD shown

| Plasma level or change | TC | LDL-C | HDL-C | TG | Lp(a) |
|---|---|---|---|---|---|
| **(A) Homozygous FH (n=8)** | | | | | |
| Baseline, off all treatment | 17.2 ± 4.7 mmol/L | | | | |
| On maximal drug therapy, prior to starting lipoprotein apheresis | 12.1 ± 2.6 mmol/L | 10.2 ± 2.8 mmol/L | 1.1 ± 0.3 mmol/L | 1.7 ± 1.3 mmol/L | 0.83 ± 0.70 g/L |
| Immediately before each apheresis treatment | 9.9 ± 1.2 mmol/L | 8.2 ± 1.1 mmol/L | 1.1 ± 0.3 mmol/L | 1.5 ± 1.0 mmol/L | 1.08 ± 0.57 g/L |
| Immediately after each apheresis treatment | 3.6 ± 0.7 mmol/L | 2.6 ± 0.8 mmol/L | 0.7 ± 0.2 mmol/L | 0.6 ± 0.4 mmol/L | 0.32 ± 0.23 g/L |
| Acute change attributable to apheresis | - 64 ± 7 % | - 69 ± 7 % | - 35 ± 9 % | - 58 ± 6 % | - 74 ± 5 % |
| Time-averaged concentration | 7.7 ± 0.9 mmol/L | 6.2 ± 1.0 mmol/L | | | |
| Change in time-averaged concentration attributable to apheresis, from levels on maximal drug therapy | - 34 ± 12 % | - 36 ± 12 % | | | |
| **(B) Heterozygous FH (n=8)** | | | | | |
| Baseline, off all treatment | 11.1 ± 2.4 mmol/L | | | | |
| On maximal drug therapy, prior to starting lipoprotein apheresis | 9.0 ± 2.2 mmol/L | 6.7 ± 1.9 mmol/L | 1.3 ± 0.3 mmol/L | 2.1 ± 0.8 mmol/L | 0.83 ± 0.77 g/L |
| Immediately before each apheresis treatment | 8.0 ± 1.5 mmol/L | 5.8 ± 1.4 mmol/L | 1.2 ± 0.3 mmol/L | 2.4 ± 1.1 mmol/L | 0.83 ± 0.56 g/L |

| | | | | | |
|---|---|---|---|---|---|
| Immediately after each apheresis treatment | 2.8 ± 0.7 mmol/L | 1.6 ± 0.7 mmol/L | 0.8 ± 0.2 mmol/L | 1.2 ± 0.5 mmol/L | 0.16 ± 0.07 g/L |
| Acute change attributable to apheresis | - 65 ± 7 % | - 72 ± 9 % | - 33 ± 10 % | - 49 ± 14 % | - 76 ± 12 % * |
| Time-averaged levels | 6.4 ± 1.2 mmol/L | 4.6 ± 1.1 mmol/L | | | |
| Change in time-averaged levels attributable to apheresis, from levels on maximal drug therapy | - 26 ± 5 % | - 34 ± 5 % | | | |

* $p = 0.08$; $p < 0.05$ for all other acute and time-averaged changes in lipid and lipoprotein concentrations.

**Table 3:** Effect of PCSK9 inhibitor therapy on frequency of apheresis and LDL-cholesterol concentrations

| Indication | Patient | LDL-cholesterol before PCSK9 inhibitor therapy | | | | LDL-cholesterol with PCSK9 inhibitor therapy | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | Apheresis frequency | Pre-apheresis | Post-apheresis | Time-averaged | PCSK9 inhibitor regimen | Apheresis frequency | Pre-apheresis or off apheresis | Post-apheresis |
| HoFH | 1 | Fortnightly | 8.7 mmol/L | 2.5 mmol/L | 6.5 mmol/L | Evolocumab 420 mg fortnightly | Every eight weeks | 3.7 mmol/L | 1.2 mmol/L |
| | 2 | Fortnightly | 7.9 mmol/L | 2.9 mmol/L | 6.1 mmol/L | Evolocumab 420 mg fortnightly | Fortnightly | 8.1 mmol/L | 2.8 mmol/L |
| | 4 | Fortnightly | 8.1 mmol/L | 3.2 mmol/L | 6.4 mmol/L | Evolocumab 420 mg fortnightly | Fortnightly | 6.8 mmol/L | 2.4 mmol/L |
| | 5 | Fortnightly | 6.5 mmol/L | 2.3 mmol/L | 5.0 mmol/L | Evolocumab 420 mg fortnightly | Fortnightly | 3.6 mmol/L | 1.5 mmol/L |
| | 6 | Fortnightly | 7.7 mmol/L | 1.5 mmol/L | 5.5 mmol/L | Evolocumab 420 mg fortnightly | Stopped | 2.4 mmol/L | NA |
| | 7 | Weekly | 8.3 mmol/L | 2.4 mmol/L | 6.2 mmol/L | Alirocumab 450 mg monthly | Weekly | 8.1 mmol/L | 1.1 mmol/L |
| | 8 | Fortnightly | 7.7 mmol/L | 2.0 mmol/L | 5.7 mmol/L | Alirocumab 450 mg fortnightly | Every four weeks | 5.0 mmol/L | 1.0 mmol/L |
| HeFH | 9 | Fortnightly | 7.9 mmol/L | 1.2 mmol/L | 6.0 mmol/L | Alirocumab 150 mg fortnightly | Stopped | 4.1 mmol/L | NA |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| 10 | Fortnightly | 3.2 mmol/L | 0.5 mmol/L | 2.4 mmol/L | Evolocumab 140 mg fortnightly | Stopped | 1.1 mmol/L | NA |
| 12 | Fortnightly | 5.0 mmol/L | 1.6 mmol/L | 4.0 mmol/L | Evolocumab 140mg fortnightly | Stopped | 2.0 mmol/L | NA |
| 13 | Fortnightly | 5.1 mmol/L | 1.6 mmol/L | 4.1 mmol/L | Evolocumab 140 mg fortnightly | Stopped* | 2.1 mmol/L | |
| 14 | Fortnightly | 6.2 mmol/L | 2.2 mmol/L | 4.8 mmol/L | Evolocumab 140 mg fortnightly | Fortnightly | 6.4 mmol/L | 2.0 mmol/L |
| 15 | Fortnightly | 7.1 mmol/L | 3.0 mmol/L | 5.9 mmol/L | Evolocumab 420 mg fortnightly | Fortnightly | 3.9 mmol/L | 1.4 mmol/L |
| 16 | Every four weeks | 6.3 mmol/L | 1.4 mmol/L | 4.9 mmol/L | Alirocumab 75 mg every four weeks | Every four weeks | 5.4 mmol/L | 1.0 mmol/L |

Time-averaged mean LDL-cholesterol concentrations were not calculated for patients remaining on lipoprotein apheresis after commencement of PCSK9 inhibitor therapy, because of uncertain validity of the equation in this setting.

* Ceased apheresis due to problems with vascular access; commenced evolocumab three months later

**Table 4:** Adverse effects occurring during, or within two hours after, lipoprotein apheresis (n = 1,971 procedures)

| Type of reaction | Incidence |
|---|---|
| Anaphylactoid (major) | 4 (0.2%) |
| Anaphylactoid (minor) | 23 (1.2%) |
| Chest pain | 18 (0.9%) |
| Hypotension | 24 (1.2%) |
| Blurred vision | 16 (0.8%) |
| Nausea | 6 (0.3%) |
| Headache | 6 (0.3%) |
| Dizziness | 6 (0.3%) |
| Paraesthesia | 6 (0.3%) |
| **Total** | **109 (5.5%)** |