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Heterozygous familial hypercholesterolaemia in specialist centres in South Africa, Australia and Brazil: Importance of early detection and lifestyle advice

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ABSTRACT

Background and aims: Familial hypercholesterolaemia (FH) is the commonest monogenic disorder that accelerates atherosclerotic cardiovascular disease. We compared and contrasted the characteristics of patients from three specialist centres in the southern hemisphere.
Methods: Adult index-cases with molecularly diagnosed heterozygous FH attending specialist lipid centres in Cape Town, Perth and São Paulo were studied. Myocardial infarction, revascularisation, hypertension, diabetes, smoking and lipid-lowering treatment were recorded at the time of diagnosis and compared across the three centres.
Results: The spectrum of genetic variants causative of FH was significantly different in patients attending the centres in South Africa compared with Australia and Brazil. Hypertension and diabetes were more prevalent in Brazilian and Australian patients, than in South African patients, but the frequency of smoking was significantly greater in South Africa than the other two centres ($p<0.01$). Age, male sex and smoking were significant independent predictors of coronary artery disease (CAD) in all three countries ($p<0.05$).
Conclusions: Patients with FH in three specialist centres in the southern hemisphere exhibit a high prevalence of non-cholesterol cardiovascular disease risk factors. Older age, male sex and smoking were more common among subjects with CAD. In all three countries, there should be vigorous programmes for the control of risk factors beyond good control of hypercholesterolaemia among patients with FH. Promotion of a healthy lifestyle, especially anti-smoking advice, is of paramount importance.

1. Introduction

Familial hypercholesterolaemia (FH) is characterised by elevated low-density lipoprotein cholesterol (LDL-C) levels owing to mutations in genes involved in the low-density lipoprotein receptor (LDLR) pathway. Heterozygous FH (heFH) is the commonest monogenic lipid disorder that accelerates atherosclerotic cardiovascular disease (CVD). The relatively high population frequency estimates of 1:250 to 1:300 [1–5] implies that FH must be viewed as a public health problem throughout the world, noting also that

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the majority of people with FH are undiagnosed and/or undertreated [6]. Beyond LDL-C, lifestyle and other risk factors also need to be addressed in the care of patients with FH [7].

Most reports in the medical literature on the detection, management and treatment of FH have been focused on the northern hemisphere [8], with fewer reports on the southern hemisphere [9]. International comparisons among patients attending specialist centres remain outstanding. As part of the "Ten Countries Study" [10], we took the opportunity to compare and contrast the clinical characteristics, CVD risk factors, genetic variants, and the corresponding associations with the prevalence of coronary artery disease (CAD) in molecularly confirmed adult index cases with heFH attending three specialist centres in South Africa, Australia and Brazil. We wanted to explore whether apart from differences in FH genetic variants, patients attending the different centres have variable prevalence and severity of non-cholesterol risk factors that could impact on the risk of CAD.

2. Patients and methods

We obtained data from hospital records and clinic databases on all index patients (first to be diagnosed in a family) aged 18 years or more and known to have a pathogenic mutation affecting the LDLR pathway (*LDLR*, *APOB* and *PCSK9* genes). Data from three specialist centres in Cape Town (South Africa) [11], Perth (Australia) [12] and São Paulo (Brazil) [13] were collated for the study between 1990 (post-statin) to 2017. All three centres followed a consistent service delivery model of care over this time period. Details of the services provided, including genetic testing, and the characteristics of patients attending these centres have been published elsewhere [11–13]. Patients with homozygous and compound heFH patients were excluded owing to the inordinately high prevalence of this type of FH in South Africa as a consequence of a founder effect; details of the homozygous FH cohort have been reported elsewhere [14]. Local ethics committee approval was obtained and informed written consent was obtained from all patients regarding the use of de-identified information.

Clinical characteristics such as history of CAD (defined as a myocardial infarction and/or coronary revascularisation), hypertension, diabetes, tendon xanthomata (assessed by clinician), smoking and lipid-lowering treatments were abstracted from hospital records at the time of FH diagnosis. Premature CAD was defined as <55 years for men and <60 years for women. Hypertension and diabetes were diagnosed according to local standard criteria. Smoking was defined as current or previous smoking. The highest pre-treatment LDL-C was also recorded. If an untreated LDL-C was not available, a correction factor was applied to estimate the pre-treatment LDL-C [15–17]. Laboratory methodology for lipid and genetic testing have been previously described [11–13,18].

Data were collected using Microsoft Excel and Access 2013. Databases were aligned, amalgamated and analysed using STATA 13.1 (College Station, TX: StataCorp LP). Continuous variables were described as mean ± standard deviation and categorical variables were expressed as proportions. The characteristics of the patients from Australia and Brazil were compared with those from South Africa, as the reference group. Group differences were investigated using regression analyses. Differences in patients with and without CAD were assessed by paired *t*-tests and chi-square tests. Univariate and multivariate logistic regression analyses were performed to predict the probability of CAD. We restricted the selection of variables in the multivariate model to only those that were significantly associated with CAD in univariate analyses. Significance was defined at 5%.

3. Results

Table 1 summarises the clinical characteristics of the 875 adult index cases with molecularly defined heFH from South Africa (n = 353), Australia (n = 266) and Brazil (n = 256). FH patients from South Africa were younger at presentation to the specialist clinic compared with patients from Australia and Brazil ($p<0.001$). Diagnosis was relatively delayed in Brazil at a mean age of 50.4 ± 14.1 years; this is almost a decade later than patients from South Africa at 41.0 ± 13.3 years and 5 years later than patients from Australia at 45.5 ± 13.8 years.

Modifiable CVD risk factors, such as hypertension and diabetes, were more prevalent in patients from Brazil and Australia compared with those from South Africa ($p<0.01$), however, smoking rates were significantly higher in South African patients ($p<0.01$).

Despite the earlier diagnosis, rates of coronary events were particularly high in South Africa with 21.0% having experienced a myocardial infarction and 18.4% having undergone a revascularisation procedure. Among those with CAD, age of first coronary event was also earlier in South Africa compared with Australia and

Table 1
Clinical characteristics of adult index cases with molecularly defined heterozygous familial hypercholesterolaemia from South Africa, Australia and Brazil.

	All	South Africa	Australia	Brazil
	n = 875	n = 353	n = 266	n = 256
Age (years)	45.1 ± 14.2	41.0 ± 13.3	45.5 ± 13.8[a]	50.4 ± 14.1[a]
Sex (% male)	45.6	51.3	41.4[b]	42.2[b]
Myocardial infarction (%)	16.5	21.0	9.8[a]	17.2
Coronary revascularisation (%)	16.8	18.4	18.1	13.3
CAD (%)	24.6	28.6	20.3[c]	23.1
Premature CAD (%)	22.9	26.4	19.2[c]	21.9
Age at first event (years)	43.3 ± 10.8	39.3 ± 10.5	46.8 ± 9.5[a]	46.7 ± 10.5[a]
History of hypertension (%)	20.5	11.9	22.9[a]	29.7[a]
History of diabetes (%)	5.3	0.6	5.3[b]	11.7[a]
History of smoking (%)	43.2	52.4	41.0[b]	32.8[a]
Tendon xanthomata (%)	43.0	76.2	31.2[a]	10.7[a†]
Pre-treatment LDL-C (mmol/L)	7.5 ± 1.9	7.4 ± 1.9	8.0 ± 1.9[a]	7.0 ± 2.0[b]
HDL-C (mmol/L)	1.2 ± 0.4	1.2 ± 0.4	1.3 ± 0.4[a]	1.2 ± 0.3
Non-HDL-C (mmol/L)	8.1 ± 2.0	8.1 ± 1.9	8.4 ± 1.9	7.8 ± 2.0
Lipid-lowering treatment (%)	59.2	31.7	74.4[a]	81.3[a]

Continuous variables are expressed as mean ± standard deviation and categorical variables are expressed as proportions (%). Significantly different from South Africa (reference group); [a]$p<0.001$, [b]$p<0.01$, [c]$p<0.05$. †16% of the cohort from Brazil had no comment about tendon xanthomata.
CAD: coronary artery disease; LDL-C: low-density lipoprotein cholesterol; HDL-C: high-density lipoprotein cholesterol.

Table 2
Pre-treatment plasma LDL cholesterol according to the type of gene variants causative of familial hypercholesterolaemia in adult index cases attending clinics in South Africa, Australia and Brazil.

Pre-treatment LDL-C	All	South Africa	Australia	Brazil
	n = 875	n = 353	n = 266	n = 256
LDLR gene variants	7.5 ± 1.9 (n = 831)	7.4 ± 1.9 (n = 349)	8.1 ± 1.8[a] (n = 238)	7.0 ± 2.0[c] (n = 244)
APOB gene variants	6.7 ± 1.5 (n = 39)	5.2 ± 0.4 (n = 3)	7.1 ± 1.5[c] (n = 25)	6.0 ± 1.0 (n = 11)
PCSK9 gene variants	9.0 ± 2.8 (n = 5)	11.2 (n = 1)	9.0 ± 3.3 (n = 3)	6.8 (n = 1)

Continuous variables are expressed as mean ± standard deviation. Significantly different from South Africa (reference group); [a]$p<0.001$, [b]$p<0.01$, [c]$p<0.05$.
LDLR: low-density lipoprotein receptor; APOB: apolipoprotein B; PCSK9: proprotein convertase subtilisin/kexin type 9.

Brazil ($p<0.001$). This may relate to the significantly higher proportion of patients from Australia and Brazil treated with lipid-lowering therapies, prior to referral to the specialist centre, compared with South Africa ($p<0.001$).

The spectrum of FH mutation types (*APOB*, *LDLR* and *PCSK9* gene variants) was more diverse in Australia and Brazil compared with South Africa. Australia had the highest frequencies of *APOB* and *PCSK9* mutations detected (Table 2). Seventeen different *LDLR* variants were identified in South Africa; 124 in Australia and 93 in Brazil. The most common mutations, FH Afrikaner-1 (*LDLR* p. Asp227Glu), FH Afrikaner-2 (*LDLR* p. Val429Met) and FH Afrikaner-3 (*LDLR* p. Asp175Asn), accounted for 48%, 23% and 7%, respectively, of the total number of heFH patients from South Africa.

Fig. 1 shows the distribution of LDL-C concentration according to FH mutation types. HeFH patients with *LDLR* and *PCSK9* mutations had higher pre-treatment LDL-C concentrations than those with *APOB* mutations ($p<0.05$). Comparing pre-treatment plasma LDL-C concentrations in each mutation type in the three countries, LDL-C concentrations in those with *LDLR* mutations were significantly higher ($p<0.001$) from Australia and significantly lower ($p = 0.017$) from Brazil compared with South Africa. In those with *APOB* mutations, LDL-C concentrations were higher in Australian heFH patients compared with South Africa ($p = 0.023$). Among the types of sequence variants in the *LDLR* gene, pre-treatment LDL-C concentrations were significantly higher in non-missense variants (nonsense, frameshift, deletion, duplication) compared with missense variants ($p = 0.01$).

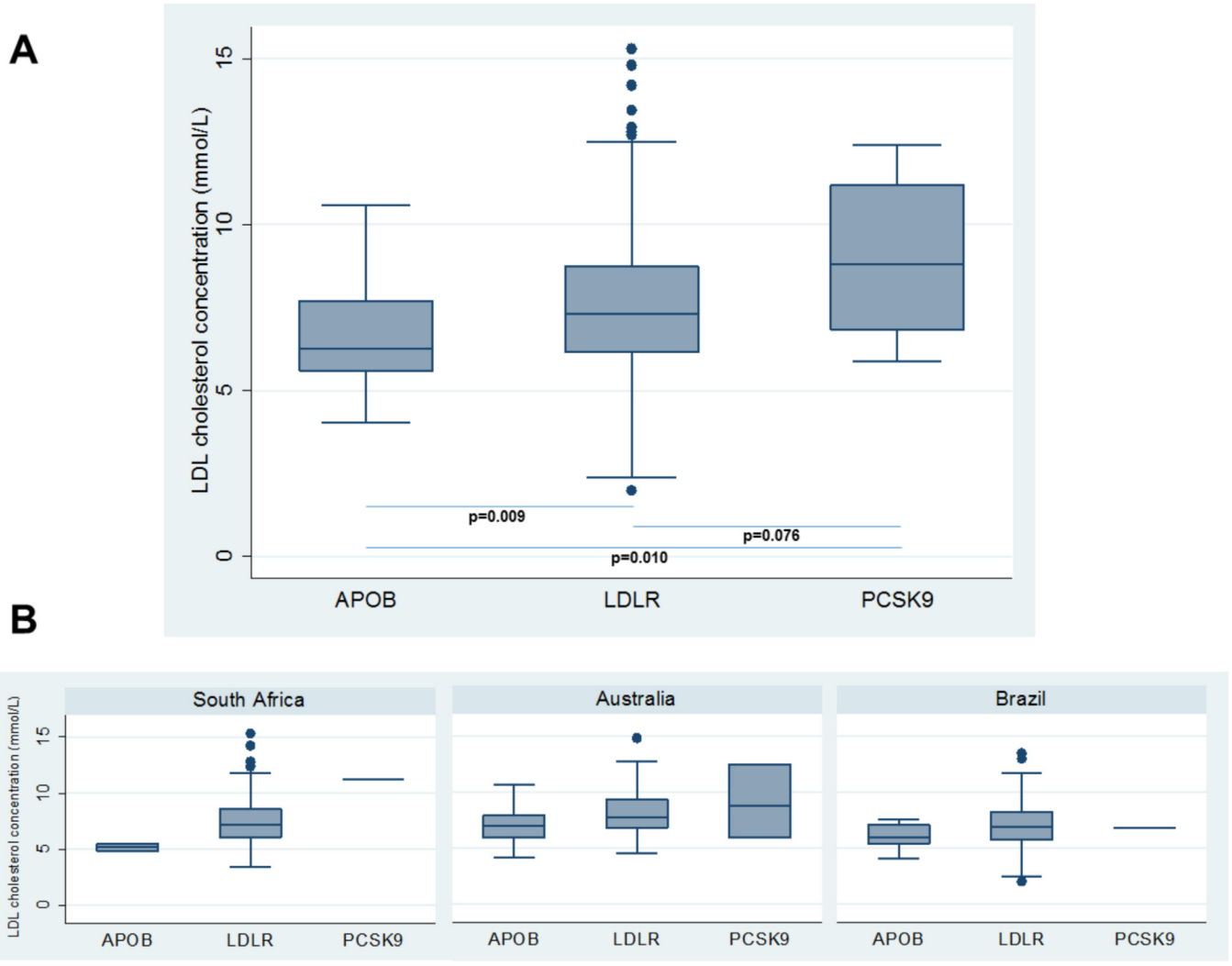

Fig. 1. Distribution of pre-treatment plasma low-density lipoprotein cholesterol concentration according to type of gene variants in (**A**) all three countries and in (**B**) South Africa, Australia and Brazil.

Table 3 compares the clinical and genetic characteristics of the patients with and without CAD. Overall, older age, male sex, hypertensive patients, those who smoked, had tendon xanthomata, on lipid-lowering treatment and had lower high-density lipoprotein cholesterol (HDL-C) were more likely to have CAD. Table 4 shows the odds ratio (OR) of the variables for coronary events in univariate analyses in the heFH patients. Age, male sex, hypertension and smoking were significant predictors of coronary events ($p<0.001$) in a pooled analysis of all three countries. In the multivariate logistic regression models, smoking remained significantly associated with coronary events ($p<0.001$) after adjusting for age and sex; this was observed in all three countries (Table 4). In a gender stratified analysis, age was equally significant in men and women (both $p<0.001$). However, smoking was a more significant predictor of CAD in men than in women (OR 3.04, $p<0.001$ *vs.* OR 1.96, $p = 0.010$).

Use of lipid-lowering treatment and low HDL-C were also associated with the likelihood of CAD in the pooled analysis. The association of lipid-lowering therapy with the likelihood of CAD was primarily driven by South Africa and the association of low HDL-C with the likelihood of CAD was primarily driven by Brazil.

Table 3
Comparison of clinical and genetic characteristics of patients with heterozygous familial hypercholesterolaemia with and without coronary artery disease in clinics from South Africa, Australia and Brazil.

Variables	CAD+	CAD-	*p*-value
All Three Countries	n = 214	n = 661	
Age (years)	50.1 ± 12.3	43.5 ± 14.4	<0.001
Sex (% male)	67.0	38.6	<0.001
Hypertension (%)	31.3	17.1	<0.001
Diabetes (%)	6.5	4.8	0.333
Smoking (%)	62.6	36.9	<0.001
Tendon xanthomata (%)	58.3	40.2	<0.001
Lipid-lowering treatment (%)	73.8	54.5	<0.001
Pre-treatment LDL-C (mmol/L)	7.4 ± 2.2	7.5 ± 1.9	0.716
HDL-C (mmol/L)	1.1 ± 0.4	1.3 ± 0.4	<0.001
Non-HDL-C (mmol/L)	8.0 ± 2.2	8.1 ± 1.9	0.695
LDLR gene variant (%)	94.9	95.0	0.931
APOB gene variant (%)	4.7	4.4	0.860
PCSK9 gene variant (%)	0.5	0.6	0.816
South Africa	n = 101	n = 252	
Age (years)	45.6 ± 11.5	39.2 ± 13.5	<0.001
Sex (% male)	73.3	42.5	<0.001
Hypertension (%)	16.8	9.9	0.070
Diabetes (%)	0	0.8	0.369
Smoking (%)	68.3	46.0	<0.001
Tendon xanthomata (%)	85.2	72.6	0.012
Lipid-lowering treatment (%)	53.5	23.0	<0.001
Pre-treatment LDL-C (mmol/L)	7.3 ± 1.9	7.4 ± 1.8	0.409
HDL-C (mmol/L)	1.1 ± 0.5	1.2 ± 0.4	0.059
Non-HDL-C (mmol/L)	8.0 ± 2.0	8.1 ± 1.9	0.731
Australia	n = 54	n = 212	
Age (years)	53.3 ± 11.0	43.5 ± 13.7	<0.001
Sex (% male)	66.7	34.9	<0.001
Hypertension (%)	44.4	17.9	<0.001
Diabetes (%)	9.3	4.3	0.141
Smoking (%)	64.8	34.9	<0.001
Tendon xanthomata (%)	46.3	27.4	0.007
Lipid-lowering treatment (%)	90.7	70.3	0.002
Pre-treatment LDL-C (mmol/L)	8.4 ± 2.1	7.9 ± 1.8	0.072
HDL-C (mmol/L)	1.2 ± 0.3	1.4 ± 0.4	0.022
Non-HDL-C (mmol/L)	8.6 ± 2.2	8.3 ± 1.8	0.263
Brazil	n = 59	n = 197	
Age (years)	54.9 ± 12.2	49.1 ± 14.4	0.005
Sex (% male)	57.6	37.6	0.006
Hypertension (%)	44.1	25.4	0.006
Diabetes (%)	15.3	10.7	0.336
Smoking (%)	50.9	27.4	0.001
Tendon xanthomata (%)	17.7	8.7	0.069
Lipid-lowering treatment (%)	93.2	77.7	0.007
Pre-treatment LDL-C (mmol/L)	6.7 ± 2.3	7.0 ± 1.9	0.350
HDL-C (mmol/L)	1.1 ± 0.3	1.3 ± 0.3	<0.001
Non-HDL-C (mmol/L)	7.6 ± 2.4	7.9 ± 1.9	0.276

CAD+: with coronary artery disease; CAD-: without coronary artery disease; LDL-C: low-density lipoprotein cholesterol; HDL-C: high-density lipoprotein cholesterol.

4. Discussion

This is the first comparison of the clinical and genetic characteristics of molecularly defined heFH across three lipid specialist centres in the southern hemisphere. We demonstrated significant differences in time of diagnosis, prevalence of non-cholesterol risk factors, spectrum of mutations and treatment of heFH patients attending specialist lipid clinics in Cape Town, Perth and São Paulo. More importantly, we showed that older age, male sex and smoking were consistent predictors of CAD.

The narrower spectrum of mutations from South Africa (mostly *LDLR* missense mutations) was at least in part owing to the limited genetic testing for locally prevalent *LDLR* mutations [11]. Because of a gene founder effect in South Africa [19], index cases, although referred independently, may be related to one another. By contrast, there was a significantly more diverse spectrum of FH mutations in Australia and Brazil. The higher proportion of familial defective apoB-100 mutations (in particular, *APOB* p. Arg3527Gln [18]) from Western Australia, was perhaps owing to the largely European extractions of the population [20].

The tendon xanthomata reflect the cholesterol life-years in FH and are predictive of CAD [21]. Xanthomata, important in the clinical diagnosis of FH and a useful clinical sign, are somewhat subjective and confirmation by imaging techniques is costly and may not always be available. The high prevalence of tendon xanthomata in South African patients indicates a stricter use of criteria for diagnosing FH as well as more sustained exposure to raised plasma cholesterol. The low prevalence of tendon xanthomata in Brazilian patients reflects probably an underestimation of the true frequency, noting that data were not recorded in 16% of that cohort. This provides a mandate for standardising the assessment of physical signs of FH.

The higher frequency of coronary events in South Africa may also reflect poorer healthcare resources, along with less healthy lifestyles and possibly also an ascertainment bias in which those for secondary prevention are referred to the clinic. Increasing the availability of high-intensity statins at all levels of care and authorising community doctors to be able to prescribe statins will be important to improve outcomes in South Africa. Differences in tobacco costs, marketing and policies among the countries could account for higher smoking rates in South Africa, and could also account for the higher frequency of CAD. Although the Brazilian cohort was older by almost a decade, coronary events were significantly less frequent, but the prevalence of hypertension and diabetes was higher than in South African patients. Although, hypertension, diabetes and smoking were similarly high in the Australian heFH patients compared with Brazil, the earlier diagnosis and hence treatment might have contributed to the relatively lower rates of myocardial infarctions in Australia. Contemporary registries around the world have described heterozygous FH adults [22]. However, comparisons of our clinic datasets with other FH populations are constrained by different diagnostic criteria and sampling methods [15,22–29]. Nevertheless, previous studies from countries in the northern hemisphere such as Canada [25], USA [30], the Netherlands [31,32], the UK [33] and Spain [34] have demonstrated that smoking, hypertension, diabetes and obesity are

Table 4
Predictors of coronary artery disease in patients with heterozygous familial hypercholesterolaemia from South Africa, Australia and Brazil.

Variables	OR[a]	95% CI	*p*-value	OR[b]	95% CI	*p*-value
All three countries						
Age	1.03	1.02–1.05	<0.001	1.04	1.02–1.05	<0.001
Sex (male)	3.28	2.36–4.54	<0.001	3.19	2.17–4.70	<0.001
Hypertension	2.21	1.55–3.15	<0.001	1.38	0.90–2.12	0.145
Diabetes	1.38	0.72–2.63	0.334			
Smoking	2.86	2.08–3.94	<0.001	2.25	1.57–3.22	<0.001
Tendon xanthomata	2.08	1.51–2.86	<0.001	1.97	1.36–2.86	<0.001
Lipid-lowering treatment	2.36	1.68–3.32	<0.001	2.05	1.36–3.09	<0.001
Pre-treatment LDL-C	0.99	0.91–1.07	0.715			
HDL-C	0.33	0.21–0.54	<0.001	0.60	0.35–1.02	0.059
Non-HDL-C	0.98	0.91–1.07	0.695			
South Africa						
Age	1.04	1.02–1.06	<0.001	1.04	1.01–1.06	0.001
Sex (male)	3.71	2.24–6.16	<0.001	3.75	2.12–6.63	<0.001
Hypertension	1.84	0.94–3.57	0.073			
Smoking	2.53	1.55–4.11	<0.001	1.90	1.10–3.26	0.021
Tendon xanthomata	2.16	1.17–4.00	0.014	1.45	0.72–2.93	0.298
Lipid-lowering treatment	3.84	2.36–6.27	<0.001	2.98	1.75–5.08	<0.001
Pre-treatment LDL-C	0.95	0.84–1.08	0.408			
HDL-C	0.55	0.29–1.03	0.061			
Non-HDL-C	0.98	0.87–1.10	0.730			
Australia						
Age	1.06	1.03–1.09	<0.001	1.06	1.03–1.10	<0.001
Sex (male)	3.73	1.98–7.02	<0.001	3.95	1.85–8.43	<0.001
Hypertension	3.66	1.93–6.96	<0.001	1.45	0.67–3.13	0.349
Diabetes	2.30	0.74–7.17	0.151			
Smoking	3.44	1.84–6.42	<0.001	2.59	1.27–5.29	0.009
Tendon xanthomata	2.29	1.24–4.23	0.008	1.30	0.64–2.65	0.467
Lipid-lowering treatment	4.14	1.58–10.89	0.004	1.89	0.65–5.49	0.240
Pre-treatment LDL-C	1.16	0.99–1.35	0.074			
HDL-C	0.32	0.12–0.85	0.022	0.60	0.20–1.78	0.355
Non-HDL-C	1.09	0.94–1.28	0.263			
Brazil						
Age	1.03	1.01–1.05	0.006	1.04	1.01–1.06	0.012
Sex (male)	2.26	1.25–4.08	0.007	2.11	1.03–4.33	0.041
Hypertension	2.32	1.26–4.25	0.007	1.78	0.87–3.66	0.116
Diabetes	1.51	0.65–3.50	0.338			
Smoking	2.74	1.51–4.99	0.001	2.09	1.07–4.06	0.031
Tendon xanthomata	2.26	0.92–5.52	0.074			
Lipid-lowering treatment	3.95	1.36–11.52	0.012	3.15	0.88–11.28	0.079
Pre-treatment LDL-C	0.93	0.80–1.08	0.349			
HDL-C	0.14	0.05–0.42	<0.001	0.17	0.05–0.64	0.008
Non-HDL-C	0.92	0.79–1.07	0.275			

LDL-C: low-density lipoprotein cholesterol; HDL-C: high-density lipoprotein cholesterol.
[a] Univariate analyses.
[b] Multivariate analyses adjusting for all other variables significantly associated with coronary artery disease in univariate analyses.

risk factors for CVD in FH. We also demonstrate that smoking and hypertension are associated with coronary events in the present study. However, in each country cohort, smoking consistently remained a significant predictor of coronary events, after adjusting for age and sex.

The generalisability of our findings are constrained by the sampling population being derived from specialist referral centres. FH patients in the primary care setting may be less treated and have a different spectrum of CVD and CVD risk factors [35,36]; we also did not report on homozygous FH and children. A gene founder effect for FH is well-recognised in South Africa [14]; the outcomes of treatment have been conjointly reported with the UK [37]. The impact of different mutations on the phenotype in the different sample populations is worthy of further investigation but could not be fully addressed in the present study. Future studies comparing mutation spectra and phenotypes across countries are warranted.

The limitations of the present study include the lack of follow-up and post-treatment LDL-C data from the Cape Town clinic. Owing to constraints of healthcare resources, patients are often discharged to community care clinics. Additionally, the genetic diagnosis of FH was performed with limited research budgets that did not support the systematic search for all FH-causing variants. Post-treatment data has been communicated previously by Australia [12] and Brazil [13] as part of cascade screening programmes. Although we did not have data on body-mass-index, patients from Brazil had a higher frequency of hypertension and diabetes, and given their older age, this suggested a higher prevalence of central obesity and the metabolic syndrome phenotype additional to the FH status. Data on ethnicity would be interesting given the mixed ancestry and multi-ethnic populations in all three countries. Lipoprotein(a) data was incomplete and was not reported in the present study, but has been previously demonstrated in the Australian cohort as an independent predictor of coronary disease in FH [38]. Information on treatment duration, compliance, safety, intolerances and optimisation was not detailed in the medical records. Future longitudinal studies are merited to

compare cardiovascular outcomes and mortality. Existing and desirable variables will be retrospectively and prospectively measured and collected over time and will be pooled with the FH Studies Collaboration registry [39].

We demonstrate that, beyond hypercholesterolaemia, CVD risk factors were highly prevalent in a large cohort of molecularly defined heFH patients from three specialist centres in the southern hemisphere. Earlier identification and treatment of FH patients are imperative, and particularly counselling to never smoke and smoking cessation in individuals who do smoke. Extensive research, education and awareness programmes can hopefully improve FH diagnosis, treatment and CVD prevention, and mitigate CVD and CVD risk factors in our FH patients. Further studies are required to examine a wider spectrum of environmental and genetic risk factors in the heFH population. The application of a more sophisticated model of care, addressing these risk factors, should be applied to improve the healthcare of FH patients in all three countries, especially in South Africa.

Conflicts of interest

The authors have no conflict of interest to declare in relation to the preparation and submission of this article. ADM has received research funding unrelated to this study from Aegerion, Amgen, Astra Zeneca, Bayer, Bristol Myers Squibb, Isis, Merck Sharpe Dohme, Parke Davis, Pfizer, Schering Plough, Takeda and Unilever for clinical trials. DJB reports that his institution has received research grants from Sanofi-Aventis, Regeneron, Novartis, Eli Lilly & Company, Amgen, Ionis and Aegerion; and reports receiving honoraria unrelated to the study from Aegerion and Gemphire for serving on steering committees; from Sanofi-Aventis, Aegerion, Amgen, AstraZeneca, Ionis, and MSD for serving on advisory boards; from Sanofi-Aventis, Regeneron, Aegerion, Amgen, AstraZeneca, MSD, Pfizer, Servier, and Unilever for lectures; from Amgen, Sanofi Aventis and Aegerion for travel assistance to attend scientific meetings; and from Sanofi-Aventis and Regeneron for non-financial support (editorial assistance and statistical analyses) unrelated to this study. RDS has received honoraria unrelated to this study from Amgen, Astra Zeneca, Akcea, Biolab, Kowa, Merck, Novo-Nordisk, Pfizer and Sanofi/Regeneron. GFW has received honoraria unrelated to the study from Amgen, Kowa and Sanofi/Regeneron.

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Author contributions

JP, ADM and GFW contributed to the conception and design of the research. JP, BCB, PRSS, CEJ, ACP, AJH and RDS contributed to the acquisition of data. JP performed all statistical analyses and with GFW, drafted the manuscript. All authors, JP, ADM, DJB, BCB, PRSS, CEJ, ACP, AJH, KKR, RDS and GFW, contributed to data interpretation and critically revised the manuscript.

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References

[1] G.F. Watts, J.E. Shaw, J. Pang, et al., Prevalence and treatment of familial hypercholesterolemia in Australian communities, Int. J. Cardiol. 185 (2015) 69–71.
[2] S.D. de Ferranti, A.M. Rodday, M.M. Mendelson, et al., Prevalence of familial hypercholesterolemia in the 1999 to 2012 United States national health and nutrition examination surveys (NHANES), Circulation 133 (2016) 1067–1072.
[3] N.S. Abul-Husn, K. Manickam, L.K. Jones, et al., Genetic identification of familial hypercholesterolemia within a single U.S. health care system, Science (2016) 354.
[4] D.S. Wald, J.P. Bestwick, J.K. Morris, et al., Child–parent familial hypercholesterolemia screening in primary care, N. Engl. J. Med. 375 (2016) 1628–1637.
[5] J. Pang, A.C. Martin, T.A. Mori, et al., Prevalence of familial hypercholesterolaemia in adolescents: potential value of universal screening? J. Pediatr. 170 (2016) 315–316.
[6] B.G. Nordestgaard, M.J. Chapman, Familial hypercholesterolaemia is underdiagnosed and undertreated in the general population: guidance for clinicians to prevent coronary heart disease, Eur. Heart J. (2013) 34.
[7] G.F. Watts, S. Gidding, A.S. Wierzbicki, et al., Integrated guidance on the care of familial hypercholesterolaemia from the international FH foundation, Int. J. Cardiol. 171 (2014) 309–325.
[8] J. Pang, P.J. Lansberg, G.F. Watts, International developments in the care of familial hypercholesterolemia: where now and where to next? J. Atherosclerosis Thromb. 23 (2016) 505–519.
[9] B.G. Nordestgaard, M. Benn, Genetic testing for familial hypercholesterolaemia is essential in individuals with high LDL cholesterol: who does it in the world? Eur. Heart J. 38 (2017) 1580–1583.
[10] G.F. Watts, P.Y. Ding, P. George, et al., Translational research for improving the care of familial hypercholesterolemia: the “ten countries study” and beyond, J. Atherosclerosis Thromb. 23 (2016) 891–900.
[11] J.C. Firth, A.D. Marais, Familial hypercholesterolaemia: the Cape Town experience, S. Afr. Med. J. 98 (2008) 99–104.
[12] D.A. Bell, J. Pang, S. Burrows, et al., Effectiveness of genetic cascade screening for familial hypercholesterolaemia using a centrally co-ordinated clinical service: an Australian experience, Atherosclerosis 239 (2015) 93–100.
[13] C.E. Jannes, R.D. Santos, P.R. de Souza Silva, et al., Familial hypercholesterolemia in Brazil: cascade screening program, clinical and genetic aspects, Atherosclerosis 238 (2015) 101–107.
[14] F.J. Raal, G.J. Pilcher, V.R. Panz, et al., Reduction in mortality in subjects with homozygous familial hypercholesterolemia associated with advances in lipid-lowering therapy, Circulation 124 (2011) 2202–2207.
[15] M. Benn, G.F. Watts, A. Tybjaerg-Hansen, et al., Familial hypercholesterolemia in the Danish general population: prevalence, coronary artery disease, and cholesterol-lowering medication, J. Clin. Endocrinol. Metab. 97 (2012) 3956–3964.
[16] K.L. Ellis, J. Pang, D.C. Chan, et al., Familial combined hyperlipidemia and hyperlipoprotein (a) as phenotypic mimics of familial hypercholesterolemia: frequencies, associations and predictions, J. Clin. Lipidol. 10 (2016) 1329–1337.
[17] K. Haralambos, P. Ashfield-Watt, I.F. McDowell, Diagnostic scoring for familial hypercholesterolaemia in practice, Curr. Opin. Lipidol. 27 (2016) 367–374.
[18] A.J. Hooper, L.T. Nguyen, J.R. Burnett, et al., Genetic analysis of familial hypercholesterolaemia in Western Australia, Atherosclerosis 224 (2012) 430–434.
[19] H.C. Seftel, S.G. Baker, M.P. Sandler, et al., A host of hypercholesterolaemic homozygotes in South Africa, Br. Med. J. 281 (1980) 633–636.
[20] A.R. Miserez, P.Y. Muller, Familial defective apolipoprotein B-100: a mutation emerged in the mesolithic ancestors of Celtic peoples? Atherosclerosis 148 (2000) 433–436.
[21] F. Civeira, S. Castillo, R. Alonso, et al., Tendon xanthomas in familial hypercholesterolemia are associated with cardiovascular risk independently of the low-density lipoprotein receptor gene mutation, Arterioscler. Thromb. Vasc. Biol. 25 (2005) 1960–1965.
[22] E.M. deGoma, Z.S. Ahmad, E.C. O'Brien, et al., Treatment gaps in adults with heterozygous familial hypercholesterolemia in the United States: data from the CASCADE-FH registry, Circulation: Cardiovascular Genetics (2016) 240–249.
[23] L. Mundal, M. Sarancic, L. Ose, et al., Mortality among patients with familial hypercholesterolemia: a registry-based study in Norway, 1992–2010, J. Am. Heart Assoc. 3 (2014) e001236.
[24] N. Mata, R. Alonso, L. Badimón, et al., Clinical characteristics and evaluation of LDL-cholesterol treatment of the Spanish familial hypercholesterolemia longitudinal cohort study (SAFEHEART), Lipids Health Dis. 10 (2011) 94.
[25] M.D. Allard, R. Saeedi, M. Yousefi, et al., Risk stratification of patients with familial hypercholesterolemia in a multi-ethnic cohort, Diabetes 13 (2014) 65.
[26] S. Béliard, V. Carreau, A. Carrié, et al., Improvement in LDL-cholesterol levels of patients with familial hypercholesterolemia: can we do better? Analysis of results obtained during the past two decades in 1669 French subjects, Atherosclerosis 234 (2014) 136–141.
[27] M. Harada-Shiba, H. Arai, T. Okamura, et al., Multicenter study to determine the diagnosis criteria of heterozygous familial hypercholesterolemia in Japan, J. Atherosclerosis Thromb. 19 (2012) 1019–1026.

[28] A.H. Pijlman, R. Huijgen, S.N. Verhagen, et al., Evaluation of cholesterol lowering treatment of patients with familial hypercholesterolemia: a large cross-sectional study in The Netherlands, Atherosclerosis 209 (2010) 189–194.
[29] S.G. Hadfield, S. Horara, B.J. Starr, et al., Are patients with familial hypercholesterolaemia well managed in lipid clinics? An audit of eleven clinics from the Department of Health Familial Hypercholesterolaemia Cascade Testing project, Ann. Clin. Biochem. 45 (2008) 199–205.
[30] P.N. Hopkins, S. Stephenson, L.L. Wu, et al., Evaluation of coronary risk factors in patients with heterozygous familial hypercholesterolemia, Am. J. Cardiol. 87 (2001) 547–553.
[31] P.R.W. De Sauvage Nolting, J.C. Defesche, R.J.A. Buirma, et al., Prevalence and significance of cardiovascular risk factors in a large cohort of patients with familial hypercholesterolaemia, J. Intern. Med. 253 (2003) 161–168.
[32] A.C.M. Jansen, E.S. van Aalst-Cohen, M.W. Tanck, et al., The contribution of classical risk factors to cardiovascular disease in familial hypercholesterolaemia: data in 2400 patients, J. Intern. Med. 256 (2004) 482–490.
[33] H.A.W. Neil, V. Seagroatt, D.J. Betteridge, et al., Established and emerging coronary risk factors in patients with heterozygous familial hypercholesterolaemia, Heart 90 (2004) 1431–1437.
[34] R. Alonso, N. Mata, S. Castillo, et al., Cardiovascular disease in familial hypercholesterolaemia: influence of low-density lipoprotein receptor mutation type and classic risk factors, Atherosclerosis 200 (2008) 315–321.
[35] A.W. Vickery, J. Ryan, J. Pang, et al., Increasing the detection of familial hypercholesterolaemia using general practice electronic databases, Heart Lung Circ. 26 (2017) 450–454.
[36] M. Benn, G.F. Watts, A. Tybjærg-Hansen, et al., Mutations causative of familial hypercholesterolaemia: screening of 98 098 individuals from the Copenhagen General Population Study estimated a prevalence of 1 in 217, Eur. Heart J. 37 (2016) 1384–1394, https://doi.org/10.1093/eurheartj/ehw1028.
[37] G.R. Thompson, D.J. Blom, A.D. Marais, et al., Survival in homozygous familial hypercholesterolaemia is determined by the on-treatment level of serum cholesterol, Eur. Heart J. (2017) 1162–1168.
[38] D.C. Chan, J. Pang, A.J. Hooper, et al., Elevated lipoprotein(a), hypertension and renal insufficiency as predictors of coronary artery disease in patients with genetically confirmed heterozygous familial hypercholesterolemia, Int. J. Cardiol. 201 (2015) 633–638.
[39] A.J. Vallejo-Vaz, A. Akram, S.R. Kondapally Seshasai, et al., Pooling and expanding registries of familial hypercholesterolaemia to assess gaps in care and improve disease management and outcomes: rationale and design of the global EAS Familial Hypercholesterolaemia Studies Collaboration, Atherosclerosis Suppl. 22 (2016) 1–32.