Atherosclerosis Supplements 42 (2020) e35–e40

Contents lists available at ScienceDirect

Atherosclerosis Supplements

journal homepage: www.elsevier.com/locate/atherosclerosis

How registers could enhance knowledge and characterization of genetic dyslipidaemias: The experience of the LIPIGEN in Italy and of other networks for familial hypercholesterolemia

Marta Gazzotti [a, *, 1], Manuela Casula [a, b, 1], Elena Olmastroni [a], Maurizio Averna [c], Marcello Arca [d], Alberico L. Catapano [a, b]

[a] Epidemiology and Preventive Pharmacology Service (SEFAP), Department of Pharmacological and Biomolecular Sciences, University of Milan, Milan, Italy
[b] IRCCS MultiMedica, Sesto San Giovanni (MI), Italy
[c] Department ProMISE (Department of Health Promotion, Mother and Child Care, Internal Medicine and Medical Specialties), University of Palermo, Palermo, Italy
[d] Department of Translational and Precision Medicine, Sapienza University of Rome, Italy

ARTICLE INFO

Article history:

Keywords:
Familial hypercholesterolemia
Genetic dyslipidaemias
Pathology registers

ABSTRACT

Familial hypercholesterolemia (FH) is a common genetic disorder of lipid metabolism, still underdiagnosed and undertreated in the general population. Pathology registers could play a crucial role in the creation of a comprehensive and integrated global approach to cover all aspects of this disease. Systematic data collection of patients affected by FH has increased dramatically worldwide in the past few years. Moreover, results from registers already established for the longest time showed their potentialities in the implementation of the knowledge of FH, comparing country-specific approaches and providing real-world data about identification, management and treatment of FH individuals in the clinical practice.

The potential fields of research through registers are related to the deepening of the genetic basis of disease, the study of genotype-phenotype correlation, the local adaption and implementation of diagnostic algorithms, the comparison of pharmacological approaches and treatment gaps in real-life clinical practice, the evaluation of specific subpopulations, and the identification of factors modifying cardiovascular disease risk. Registers could become also a valid resource for other rare dyslipidaemias, contributing towards the evidence-based enhancement in the worldwide care of uncommon diseases.

Familial hypercholesterolemia (FH) is a common genetic disorder of lipid metabolism, characterized by raised total cholesterol (TC) and low-density lipoprotein-cholesterol (LDL-C) concentrations since birth. Affected patients cannot clear LDL particles from the circulation, and that predisposes to early atherosclerotic lesions and premature coronary heart disease (CHD).

In the last decade, the prevalence of FH has been consistently shown to be more common than what traditionally estimated, reaching 1:200/250 [1] and 1:160,000/320,000 [2,3] for heterozygous FH (HeFH) and homozygous FH (HoFH), respectively. This suggests that the number of subjects affected by this disease could potentially reach 4.5 million of HeFH individuals in Europe and possibly 35 million worldwide. Although prompt identification and treatment are crucial to prevent CHD and improve clinical outcomes, FH still remains underdiagnosed and undertreated in the general population [4].

As remarked in 2015 with the “global call to arms” prompted by the European Atherosclerosis Society FH Studies Collaboration (FHSC), huge efforts are needed to deal with the FH burden worldwide. Vallejo-Vaz et al. underlined how a comprehensive and integrated global approach is necessary to cover all aspects of this disease (e.g. early detection, a cost-effective screening, continuous research activities, raise of awareness and education, improved health policies, adequate therapy), identifying a crucial role for pathology registers [5]. Following this statement, the creation and dissemination of systems for the systematic collection of

* Corresponding author. Epidemiology and Preventive Pharmacology Service (SEFAP), Department of Pharmacological and Biomolecular Sciences, University of Milan, via Balzaretti 9, 20133, Milan, Italy.
E-mail address: marta.gazzotti@unimi.it (M. Gazzotti).
[1] Contributed equally.

https://doi.org/10.1016/j.atherosclerosissup.2021.01.007

information on FH patients have increased dramatically, as evinced also by the exponential growth of related publications over the last 5 years: published articles describing the design of the FH registers or data results increased tenfold, with more than 20 new national registers activated in recent years [6]. Moreover, also national programs were implemented in order to investigate for the prevalence of FH and the genetics of lipid disorders [7,8]. A consortium of major FH registers was then created, including data from more than 70 countries. Pooling together information of country-specific pathology registers allow to obtain large-scale real-world data necessary to fill the gaps in knowledge, evaluate the impact of FH in general population, improve the collective awareness of this pathology, and ensure a continuous care program for patients.

The potential of this type of initiatives are well illustrated by results published by registers established for the longest time: among them, the CASCADE FH Registry in United States, the SAFEHEART registry in Spain, the FH Canada in Canada, and the LIPIGEN study in Italy.

CASCADE FH (CAscade SCreening for Awareness and DEtection of Familial Hypercholesterolemia) registry was created in 2013 by the FH foundation [9], a patient-led, non-profit, charitable organization in the United States. The enrolment is characterized by a hybrid recruitment using a variety of mechanisms: clinical enrolment in specialized lipid clinics, self-enrolment using an online screening mechanism available to general public, and the case identification through electronic health records (EHRs). Inclusion criteria are based on the presence of clinical or genetic diagnosis of FH or on untreated or treated LDL-cholesterol and total cholesterol levels (untreated LDL-C > 190 mg/dL or TC > 300 mg/dL; treated LDL-C > 124 mg/dL or TC > 195 mg/dL). At the end of 2018, the study accounted for more than 5000 subjects followed by 40 sites (of which 5 were paediatric) in 23 US states [10].

SAFEHEART registry (The SpAnish Familial HypErcHolEsterolaemiA CohoRt STudy), an open, multicentre, long-term prospective cohort study, began in 2004 in 19 outpatient lipid clinics in Spain [11]. Based on the inclusion criteria, all the index cases with a genetic diagnosis of FH and their relatives older than 15 years with a genetic diagnosis of FH, and relatives over 15 years old without a genetic diagnosis of FH as control group, are included in the registry. At October 2015 [12], more than 4000 subjects (19% index cases) from 25 lipid clinics were enrolled in the study. About the 90% of them were adults (2746 FH and about 1000 unaffected relatives).

Canadian FH registry was established in 2013 in the province of British Columbia, and then expanded to include 19 academic centres across Canada, as well as numerous peripheral sites. At the end of December 2017, data of more than 3000 subjects (14 with HoFH) were entered in the database. To be enrolled, patients should receive a diagnosis of HeFH according to the Dutch Lipid Clinic Network (DLCN) score criteria (possible, probable, definite) or to the Canadian definition (probable, definite), or a physician diagnosis of HoFH [13]. Genetic testing, which is not routinely available in most of Canada, was not considered to be necessary for the enrolment into the register.

LIPIGEN-FH (LIpid TransPort Disorders Italian GEnetic Network) study is an observational, multicentre, retrospective and prospective study [14], sponsored by the Foundation of Italian Atherosclerosis Society (SISA) and established in Italy since 2009 to promote the diagnosis of genetic dyslipidaemias, with a primary focus on FH. At July 2020, more than 50 Italian clinical centres are involved in the project, accounting for more than 8500 FH patients (about 15.0% of whom under 18 years), with a clinical (based on DLCN score criteria) and/or genetic diagnosis of FH.

These registers allow to implement the knowledge on FH from various points of view, through comparing different strategies and country-specific approaches, and providing real-world data about how FH subjects are identified, managed, and treated in clinical practice (Fig. 1).

1. Deepening of the genetic basis of disease

When genetic testing is performed in individuals with FH, causal *LDLR* mutations are found in about 60–80% of cases [15]. The identification of families with phenotypic FH without *LDLR* mutations led to the search for other causal genes: the gene encoding apolipoprotein B (apoB), which is now thought to account for 4–5% of familial hypercholesterolemia cases (more in certain populations), or the proprotein convertase subtilisin kexin 9 (PCSK9) gene, so far accounting for less than 1% of FH overall [15].

Although genetic testing has notably improved the identification of patients suffering from FH [16,17], a relevant proportion [18,19] of individuals clinically diagnosed with FH does not present a causative mutation in any of the conventionally tested genes. Importantly, a negative genetic test does not rule out an underlying genetic cause [16]. Recently, the failure in detection of mutation in patients with a clinically diagnosis of FH raises the question of the presence of a polygenic aetiology due to co-inheritance of common LDL-C raising variants [20] or the existence of other still unknown variants/genes. The continuous progress in the knowledge of the genetic basis of this disease is illustrated, for example, by the identification of new variants associated with the disease, or by the characterization of pathogenic variants among those previously defined as “of uncertain clinical significance” (VUS) [21]. The genetic results of 2938 FH subjects enrolled in the SAFEHEART study [22] identified 194 variants (88% classified as pathogenic or likely pathogenic), 24 (21 in *LDLR* and 3 in *APOB*) of them have not been reported before. Likewise, among 213 variants detected in 1076 subjects enrolled in the LIPIGEN study, 36 mutations had not been described before [23]. *In silico* analysis showed that among 32 new variants on *LDLR* gene, 27 could potentially be defined as pathogenic or likely pathogenic while only one of the four not previously reported variants on *LDLRAP1* gene could be presumed to be pathogenic. The increase of number of variants associated with FH discovered for *LDLR*, the gene implicated in the greatest number of cases, is emblematic: it has gone from about 300 at the end of the nineties [24,25] to 2600 (ClinVar database, https://clinvarminer.genetics.utah.edu) nowadays. Finally, registers may also offer the opportunity to identify new FH-causing genes, due to possibility to detect several individuals (or families) with a FH-like phenotype, but without sequence variations in the canonical genes.

2. Genotype-phenotype correlation

For any genetic disease, one of the most important questions is whether there is a relationship between any disease-causative mutation and the risk of developing a specific outcome. In this context, registers allow to study the relationship between genotype, LDL-C levels and outcomes. In 2000, Bertolini et al. [26] started to evaluate the impact of *LDLR* mutation on clinical expression of FH in an Italian FH heterozygous cohort, stratifying by LDLR residual activity. They showed that receptor-negative subjects presented a more severe phenotype compared to receptor-defective group. In the same way, Bourbon et al. [22] classified the SAFEHEART cohort with a *LDLR* mutation according to the LDLR residual activity: carriers of a null allele variant, or carriers of a defective allele variant. Also in this case, individuals who carry *LDLR* mutations that completely abolish LDLR activity have even higher LDL-C levels (264.1 mg/dL vs 253.9 mg/dL, $p < 0.001$) and a worse clinical status (presence of xanthomas, 16.6%, vs 10.7%, $p < 0.001$; history of premature atherosclerotic cardiovascular disease, 10.2%

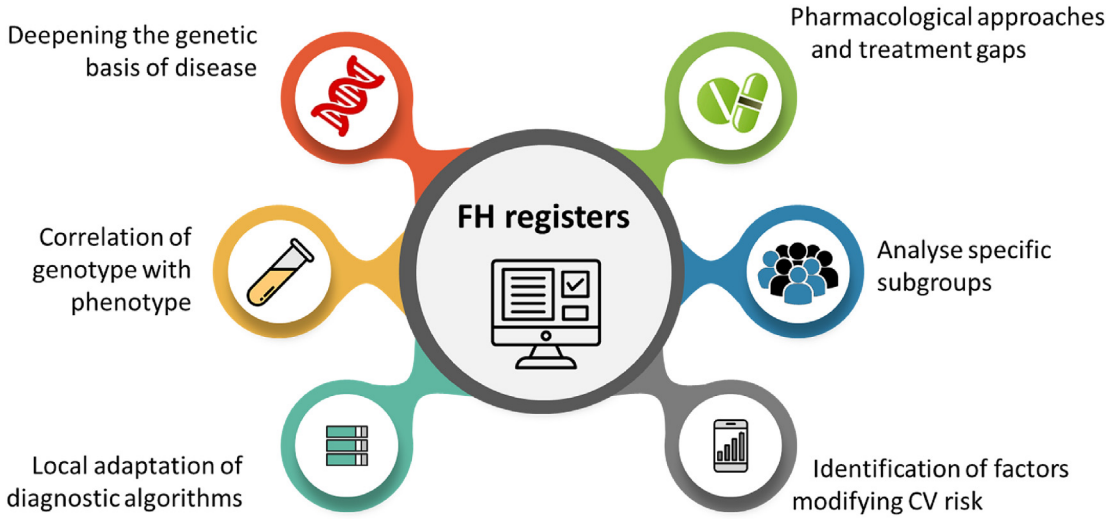

Fig. 1. Fields of research potentially investigable through the registers.

vs 9.1%, p = 0.114) than patients with defective allele variants. The same pattern was observed in Chinese [27], Greek [28], and German [29] FH patients. Moreover, recent genetic studies highlighted the great variability of the clinical phenotypes in homozygous FH and heterozygous FH and the large clinical overlap between them, as a consequence of the variations in the number of genes involved and the specific pathogenic mutations. Analysing HoFH patients from LIPIGEN study, Bertolini et al. [30] showed that true homozygotes (carrying two identical pathogenic variants) showed a more severe lipid phenotype (LDL-C 607 mg/dL vs 520 mg/dL, p = 0.005) and higher prevalence of atherosclerotic cardiovascular disease (66.6% vs 43.1%, p = 0.009) as compared with compound heterozygotes (two different pathogenic variants). A less severe phenotype was also observed in the few double heterozygotes (carrying a pathogenic variant in *LDLR* in combination with a variant in either *APOB* or *PCSK9* gene). Similar results were obtained in an analysis of HoFH Spanish patients [31].

Registers allow also the comparisons between index cases and affected (or unaffected) family members [32,33]. For instance, it is possible to find people (and families) with classic FH mutations but without a previous history of CHD. Similarly, there are families with 'mild' mutations that have a much higher prevalence of CHD than might be expected. Finally, it is possible to study to what extent environmental factors, such as diet, and other classical risk factors play a significant role in modulating the FH phenotype [34].

3. Local adaption of diagnostic algorithms

Conventionally, clinical scores such as Simon Broome (from UK) [35], DLCN (from the Netherlands) [36], and Make Early Diagnosis to Prevent Early Death (MEDPED) (from the USA) [37], are used to diagnose FH. These scores consider patients' lipid levels, cutaneous stigmata, and personal and family history of CHD to some extent. Nevertheless, their ability of efficiently diagnose FH subjects could be affected due to the fact that they have been created and validated long ago, and could result to be less efficient in an era in which lifestyle modifications, the widespread therapy with statins and more recent new lipid-lowering drugs have modified the epidemiology of some conditions classically associated with FH [3,8]. Moreover, they have not been adapted by country and by age groups, and some data, required for the calculation of scores, can be difficult to be found [38,39].

In the last decade, taking advantage from information retrieved within registers, efforts have been made to overcome these limitations. In Canada, a new simplified diagnostic criteria has been proposed through modification and adaptation of DLCN and Simon-Broome Registry criteria to Canadian population [40]. Authors identified major criteria (DNA mutation, tendon xanthomas or LDL-C $\geq$ 8.5 mmol/L) and minor criteria (family history of LDL-C >95th percentile, as evaluated in Canadian population, according to age and sex; or a history of ASCVD in the index patient or in a first-degree relative younger than 55 years for men or < 65 years for women). A suspect of FH should be considered in case of LDL-C level $\geq$ 5 mmol/L (or LDL-C $\geq$4.0 mmol/L for age <18 years; LDL-C $\geq$ 4.5 mmol/L for age $\geq$18 years and <40 years). A definite diagnosis of FH is also performed based upon the presence of one or more major criteria. When no major criteria are present, a probable diagnosis of FH could be performed if at least one of minor criteria is present. This simplified definition of FH showed excellent agreement with the most widely used FH criteria, and was well adapted to the Canadian population.

In the LIPIGEN cohort, the performance of the DLCN score was evaluated in 1337 adult patients with a genetic diagnosis of FH, addressing the question whether missing information may affect the identification of FH subjects [38]. Compared with subjects without missing data, the lack of information related to the family clinical history did not modify the rate of patient identification; in contrast, the lack of information concerning the physical signs, typical of FH, or the personal history of cardio/cerebrovascular events strongly reduced the percentage of subjects with a probable/definite FH diagnosis. Recently, this evaluation was deepened carrying out a stepwise logistic regression analysis on the LIPIGEN cohort to identifying which factors among conventional DLCN criteria and other additional covariates (i.e. non-HDL-C, lipoprotein (a) [Lp(a)], LDL-C>180 mg/dL since age<18) had a greater impact in predicting the probability of having monogenic FH. We found that, among the nine items of the DLCN score calculation, the presence of tendinous xanthomata and very high LDL-C levels (either in the patient or in first degree relatives) showed the strongest association with the outcome. Regarding the additional factors, the only one included in the final model was the detection of LDL-C>180 mg/dL before 18 years old.

In Wales, Haralambos et al. [41] implemented a scoring system based on a modification of the DLCN score criteria, extending the

detection of family history criteria to 2nd degree relative, increasing the points for tendon xanthoma in 1st and 2nd degree relatives to 6 points, identifying a negative value for raised triglycerides, and providing different points according to the age of premature CHD in the index patient. The new score system was shown to efficiently guide selection of patients for DNA testing when applied by health professionals, targeting resources to those individuals and families that are most likely to benefit from FH genetic testing.

4. Pharmacological approaches and treatment gaps

The nature of registers makes them a useful tool to obtain information from real-life clinical practice, and to evaluate the effectiveness of the available lipid-lowering treatment to achieve LDL-C goals in FH patients.

A cross-sectional analysis of HeFH adults enrolled in the CASCADE-FH Registry [42], demonstrated limited use of high-intensity statin therapy (42%) and combination of LDL-lowering therapy (45%); among the 25% of patients not receiving statin treatment, reported reasons for the lack of statin use were mainly intolerance or allergy (60%), patient preference (11%), and physician preference (11%). Another evaluation from the same registry suggested that sex and racial/ethnic factors may contribute to undertreatment [43]. In a longitudinal analysis [10], during a mean follow-up of 20 months, only 48% of patients achieved LDL-C < 100 mg/dL and 22% achieved LDL-C < 70 mg/dL.

Taking advantages by longitudinal data collected in the SAFEHEART study, authors investigated the achievement of LDL-C therapy goals in 2170 FH subjects [44], detecting a failure in the majority of subjects in reaching LDL-C goals: over a mean follow-up of 5 years, despite a significant increase in the use of high-intensity statins and combined lipid-lowering therapies, and a reduction in mean LDL-C values, less than 10% of FH subjects reached the LDL-C goals. Similar low rates were reported in the analysis of PLANET registry from Czech Republic and Slovakia [45] and in an evaluation of the German CaReHigh Registry [46]. Currently the most effective way of reducing LDL-C levels in HoFH patients is lipoprotein apheresis. An evaluation from a Turkish registry [47] showed that, in these subjects, there is a long delay between FH diagnosis and the initiation of this treatment. Although apheresis is a lifesaving therapy for patients with HoFH, in real clinical practice, most patients experience it as ineffective, and fail to reach LDL goals.

Overall, this observation emphasizes that FH patients should receive more intensive lipid-lowering treatment, but also highlight a medical need for new therapies to help patients to reach lower LDL-C levels and prevent development of premature ASCVD. The analyses from registers could determine the factors leading to late referral, undertreatment, and treatment failure on country level, supporting the development of structured approaches.

5. Evaluation of specific subgroups

Local registers could also provide useful information on specific subpopulations, such as children and adolescents [48]. They could be studied with *ad hoc* registers, or data could be extrapolated from ongoing national registers including also adults. The UK National Paediatric Familial Hypercholesterolaemia Register, established in 2012 to collect baseline and long-term follow-up data on all children with HeFH in the United Kingdom [49], the Czech MedPed registry, a paediatric care network well established in the Czech Republic since 1998 [50], or the Greek Paediatric FH Register, started in 1993 [51], are valuable examples. International collaborations of country-specific databases, such as the International Paediatric FH register [52], offer the opportunity to compare the impact of different approaches, policies and care-pathways on identification and treatment of FH. Pooling together this information was identified, for example, a variability between countries related to the mean age at diagnosis, age at which therapy is started, type of lipid-lowering treatment prescribed, and proportion of subjects above 10 years not treated but with LDL-C levels higher than recommended levels. In Italy, as the LIPIGEN-FH cohort included both adults and children/adolescents, in 2018 a paediatric group was established. This sub-study is aimed at improving detection, diagnosis, and management of FH patients <18 years throughout Italy. At July 2020, 30 lipid clinics were involved, and more than 1300 subjects were entered in the database. This network, established between lipid clinics and paediatric centres, allowed to implement the baseline data collection, integrating follow-up data and additional specific information (i.e., presence of premature CHD in second-degree family members, lipid levels of both parents, puberty status, and waist circumference), with the aim of refining diagnostic approach in paediatric patients.

6. Identification of factors modifying cardiovascular risk

In FH subjects, the exposure to elevated level of LDL-C since birth confers high risk of cardiovascular disease (CVD). The phenotypic variability that characterizes FH offers the opportunity to stratify affected patients, identifying those at the highest CV risk, and therefore deserving of a closer monitoring and a more aggressive treatment. Part of the heterogeneity in risk may be attributable to non-LDL-C traditional risk factors. Since genotype has a major impact on these subjects, well-established risk calculators such as the Framingham algorithm [53] or the European SCORE [54] are not applicable to FH patients, which made accurate CV risk calculation challenging [55]. Consequently, specific algorithms were developed in the recent years, to predict the CV risk of FH subject.

The presence of follow-up data in a large cohort of FH patients enrolled into the SAFEHEART registry allowed to build a risk equation (SAFEHEART-RE) [56] to predict incident ASCVD events in genetically-confirmed HeFH adults, based on age, sex, history of ASCVD, blood pressure, body mass index, smoking, and plasma LDL-C, and Lp(a) levels.

Another combination of cardiovascular risk factors greatly increasing CVD in FH was identified in Canadian subjects with a FH causative mutation in the *LDLR* gene. This developed novel tool incorporated several independent risk factors (age, HDL-C, gender, hypertension and smoking) in the Montreal-FH-SCORE [57]. This model was derived from a cohort of 670 adult French Canadian FH individuals carrying a mutation in the *LDLR* gene; it has been further validated in another French Canadian cohort of FH patients, and additional validation in the FH Canada national registry is currently underway.

Conclusions and future perspective

Despite recent progress, FH is still underestimated, underdiagnosed, and undertreated, and it represents a significant problem as a common risk factor for CV morbidity and mortality. Pathology registers have proven to be a valuable resource for increasing our knowledge and improving patient management. In the future, the efforts around the world will have to focus on increasing the number of patients included in national databases, promoting the awareness in the general population and among medical experts, supporting patient organizations, and promoting the standardization of registries to facilitate the comparison of the results and to allow the construction of large multinational databases pooling several local experiences [58].

As registers could significantly contribute towards the evidence-based enhancement in the worldwide care of uncommon diseases, they can become a resource for other rare dyslipidaemias besides FH. As well depicted by the recent Consensus Statement by EAS [59], although each rare disease affects a small number of people, collectively these conditions pose a considerable health burden. The development of registers has the potential to improve awareness, management, and access to effective therapy for these conditions.

Financial support

The work of M Casula is supported by Ministry of Health-IRCCS MultiMedica GR-2016-02361198 and Fondazione SISA. The work of M Arca is supported by Telethon GGP14066; Fondazione Roma Grant 2018-H2217; National Institute of Health (USA), NIH R01-HL 131961; Progetti Ricerca Ateneo, RM11715C683968E; Grandi Progetti Università C26H15ZWC9, Fondazione SISA. The work of AL Catapano is supported by Ministry of Health - Ricerca Corrente - IRCCS MultiMedica, PRIN 2017H5F943, ERANET ER-2017-2364981, and Fondazione SISA.

Author contributions

M Gazzotti and M Casula designed the review. M Gazzotti, M Casula, and E Olmastroni wrote and M Averna, M Arca, and AL Catapano provided critical revision of the article. All authors commented the article and accepted the final version.

Declaration of competing interest

M Casula, M Gazzotti, and E Olmastroni report no disclosures. M Averna received research funding and/or honoraria for advisory boards, consultancy or speaker bureau from Aegerion, Akcea, Amgen, Merck or MSD, Pfizer, Sanofi-Regeneron. M Arca has received research funding and/or honoraria for advisory boards or speaker bureau from Alfasigma, Amryt, Amgen, Akcea/Ionis, Boheringer, Daichi-Sankio, Novartis, Pfizer, Regeneron, Sanofi. AL Catapano reports grants from Amgen, Sanofi, Regeneron and personal fees from Merck, Sanofi, Regeneron, AstraZeneca, Amgen, Novartis, outside the submitted work.

Acknowledgments

This article is part of a Supplement entitled “Plasma lipids and cardiovascular risk: Nutritional and therapeutic approaches” published with support from Società Italiana di Terapia Clinica e Sperimentale (SITeCS).

References

[1] Akioyamen LE, Genest J, Shan SD, Reel RL, Albaum JM, Chu A, et al. Estimating the prevalence of heterozygous familial hypercholesterolaemia: a systematic review and meta-analysis. BMJ open 2017 Sep 1;7(9):e016461. PubMed PMID: 28864697. Pubmed Central PMCID: 5588988.

[2] Mach F, Baigent C, Catapano AL, Koskinas KC, Casula M, Badimon L, et al. ESC/EAS Guidelines for the management of dyslipidaemias: lipid modification to reduce cardiovascular risk. 2020 Jan 1 Eur Heart J 2019;41(1):111–88. PubMed PMID: 31504418.

[3] Beheshti SO, Madsen CM, Varbo A, Nordestgaard BG. Worldwide prevalence of familial hypercholesterolemia: meta-analyses of 11 million subjects. J Am Coll Cardiol 2020 May 26;75(20):2553–66. PubMed PMID: 32439005.

[4] Nordestgaard BG, Chapman MJ, Humphries SE, Ginsberg HN, Masana L, Descamps OS, et al. Familial hypercholesterolaemia is underdiagnosed and undertreated in the general population: guidance for clinicians to prevent coronary heart disease. Eur Heart J 2013 Dec;34(45):3478. PubMed PMID: WOS:000328393500009. English.

[5] Vallejo-Vaz AJ, Kondapally Seshasai SR, Cole D, Hovingh GK, Kastelein JJ, Mata P, et al. Familial hypercholesterolaemia: a global call to arms. Atherosclerosis 2015 Nov;243(1):257–9. PubMed PMID: 26408930.

[6] Collaboration Easfhs, Vallejo-Vaz AJ, De Marco M, Stevens CAT, Akram A, Freiberger T, et al. Overview of the current status of familial hypercholesterolaemia care in over 60 countries - the EAS Familial Hypercholesterolaemia Studies Collaboration (FHSC). Atherosclerosis 2018 Oct;277:234–55. PubMed PMID: 30270054.

[7] Jóźwiak J, Kasperczyk S, Tomasik T, Osadnik T, Windak A, Studziński K, et al. Design and rationale of a nationwide screening analysis from the LIPIDOGRAM2015 and LIPIDOGEN2015 studies. Arch Med Sci 2020 05/24:16.

[8] de Ferranti SD, Rodday AM, Mendelson MM, Wong JB, Leslie LK, Sheldrick RC. Prevalence of familial hypercholesterolemia in the 1999 to 2012 United States national health and nutrition examination surveys (NHANES). Circulation 2016 Mar 15;133(11):1067–72. PubMed PMID: 26976914.

[9] O'Brien EC, Roe MT, Fraulo ES, Peterson ED, Ballantyne CM, Genest J, et al. Rationale and design of the familial hypercholesterolemia foundation CAscade SCreening for Awareness and DEtection of Familial Hypercholesterolemia registry. Am Heart J 2014 Mar;167(3):342–349 e17. PubMed PMID: 24576518.

[10] Duell PB, Gidding SS, Andersen RL, Knickelbine T, Anderson L, Gianos E, et al. Longitudinal low density lipoprotein cholesterol goal achievement and cardiovascular outcomes among adult patients with familial hypercholesterolemia: the CASCADE FH registry. Atherosclerosis 2019 Oct;289:85–93. PubMed PMID: WOS:000489281400012. English.

[11] Mata N, Alonso R, Badimon L, Padro T, Fuentes F, Muniz O, et al. Clinical characteristics and evaluation of LDL-cholesterol treatment of the Spanish familial hypercholesterolemia longitudinal cohort study (SAFEHEART). Lipids Health Dis 2011 Jun 10:10. PubMed PMID: WOS:000292875100001. English.

[12] de Isla LP, Alonso R, Mata N, Fernandez-Perez C, Muniz O, Diaz-Diaz JL, et al. Predicting cardiovascular events in familial hypercholesterolemia the SAFEHEART registry (Spanish familial hypercholesterolemia cohort study). Circulation 2017 May 30;135(22):2133. PubMed PMID: WOS:000402290600016. English.

[13] Brunham LR, Ruel I, Khoury E, Hegele RA, Couture P, Bergeron J, et al. Familial hypercholesterolemia in Canada: initial results from the FH Canada national registry. Atherosclerosis 2018 Oct;277:419–24. PubMed PMID: WOS:000445908000063. English.

[14] Averna M, Cefalu AB, Casula M, Noto D, Arca M, Bertolini S, et al. Familial hypercholesterolemia: the Italian Atherosclerosis society network (LIPIGEN). Atherosclerosis Suppl 2017 Oct;29:11–6. PubMed PMID: WOS:000415623500002. English.

[15] Berberich AJ, Hegele RA. The complex molecular genetics of familial hypercholesterolaemia. Nat Rev Cardiol 2019 Jan;16(1):9–20. PubMed PMID: 29973710.

[16] Berberich AJ, Hegele RA. The role of genetic testing in dyslipidaemia. Pathology 2019 Feb;51(2):184–92. PubMed PMID: WOS:000456806400008. English.

[17] Banach M, Penson PE. Genetic testing in familial hypercholesterolaemia: what does it add? Eur J Prevent Cardiol 2020 Jan;27(1):105–6. PubMed PMID: 31408367.

[18] Talmud PJ, Futema M, Humphries SE. The genetic architecture of the familial hyperlipidaemia syndromes: rare mutations and common variants in multiple genes. Curr Opin Lipidol 2014 Aug;25(4):274–81. PubMed PMID: WOS:000339244900005. English.

[19] Hughes DP, Viljoen A, Wierzbicki AS. Familial hypercholesterolaemia in the era of genetic testing. Curr Cardiol Rep 2016 May;18(5). PubMed PMID: WOS:000374590900008. English.

[20] Talmud PJ, Shah S, Whittall R, Futema M, Howard P, Cooper JA, et al. Use of low-density lipoprotein cholesterol gene score to distinguish patients with polygenic and monogenic familial hypercholesterolaemia: a case-control study. 1293-301 Lancet 2013 Apr 13;381(9874). PubMed PMID: WOS:000317351000032. English.

[21] Kindt I, Mata P, Knowles JW. The role of registries and genetic databases in familial hypercholesterolemia. Curr Opin Lipidol 2017 Apr;28(2):152–60. PubMed PMID: WOS:000396026100009. English.

[22] Bourbon M, Alves AC, Alonso R, Mata N, Aguiar P, Padro T, et al. Mutational analysis and genotype-phenotype relation in familial hypercholesterolemia: the SAFEHEART registry. Atherosclerosis 2017 Jul;262:8–13. PubMed PMID: WOS:000405311200002. English.

[23] Pirillo A, Garlaschelli K, Arca M, Averna M, Bertolini S, Calandra S, et al. Spectrum of mutations in Italian patients with familial hypercholesterolemia: new results from the LIPIGEN study. Atherosclerosis Suppl 2017 Oct;29:17–24. PubMed PMID: WOS:000415623500003. English.

[24] Hobbs HH, Brown MS, Goldstein JL. Molecular genetics of the LDL receptor gene in familial hypercholesterolemia. Hum Mutat 1992;1(6):445–66.

[25] Day I, Whittall R, O'Dell S, Haddad L, Bolla M, Gudnason V, et al. Spectrum of LDL receptor gene mutations in heterozygous familial hypercholesterolemia. Hum Mutat 1997;10(2):116–27.

[26] Bertolini S, Cantafora A, Averna M, Cortese C, Motti C, Martini S, et al. Clinical expression of familial hypercholesterolemia in clusters of mutations of the LDL receptor gene that cause a receptor-defective or receptor-negative phenotype. Arterioscler Thromb Vasc Biol 2000 Sep;20(9):E41–52. PubMed PMID: 10978268.

[27] Sun D, Zhou BY, Li S, Sun NL, Hua Q, Wu SL, et al. Genetic basis of index patients with familial hypercholesterolemia in Chinese population: mutation spectrum and genotype-phenotype correlation. Lipids Health Dis 2018 Nov

6;17(1):252. PubMed PMID: 30400955. Pubmed Central PMCID: 6220500.
[28] Mollaki V, Progias P, Drogari E. Familial Hypercholesterolemia in Greek children and their families: genotype-to-phenotype correlations and a reconsideration of LDLR mutation spectrum. Atherosclerosis 2014 Dec;237(2):798–804. PubMed PMID: WOS:000346066600092. English.
[29] Grenkowitz T, Kassner U, Wuhle-Demuth M, Salewsky B, Rosada A, Zemojtel T, et al. Clinical characterization and mutation spectrum of German patients with familial hypercholesterolemia. Atherosclerosis 2016 Oct;253:88–93. PubMed PMID: WOS:000389401200014. English.
[30] Bertolini S, Calandra S, Arca M, Averna M, Catapano AL, Tarugi P, et al. Homozygous familial hypercholesterolemia in Italy: clinical and molecular features. Atherosclerosis 2020 Sep 9;312:72–8. PubMed PMID: 32977124.
[31] Sanchez-Hernandez RM, Civeira F, Stef M, Perez-Calahorra S, Almagro F, Plana N, et al. Homozygous familial hypercholesterolemia in Spain: prevalence and phenotype-genotype relationship. Circul Cardiovasc Genet 2016 Dec;9(6):504–10. PubMed PMID: 27784735.
[32] Bourbon M, Alves AC, Medeiros AM, Silva S, Soutar AK. Investigators of Portuguese FHS. Familial hypercholesterolaemia in Portugal. Atherosclerosis 2008 Feb;196(2):633–42. PubMed PMID: 17765246.
[33] Bertolini S, Pisciotta L, Fasano T, Rabacchi C, Calandra S. The study of familial hypercholesterolemia in Italy: a narrative review. Atherosclerosis Suppl 2017 Oct;29:1–10. PubMed PMID: 28965614.
[34] Pimstone SN, Sun XM, du Souich C, Frohlich JJ, Hayden MR, Soutar AK. Phenotypic variation in heterozygous familial hypercholesterolemia - a comparison of Chinese patients with the same or similar mutations in the LDL receptor gene in China or Canada. Arterioscl Throm Vas 1998 Feb;18(2):309–15. PubMed PMID: WOS:000072127600022. English.
[35] Risk of fatal coronary heart disease in familial hypercholesterolaemia. Scientific Steering Committee on behalf of the Simon Broome Register Group. BMJ 1991 Oct 12;303(6807):893–6. PubMed PMID: 1933004. Pubmed Central PMCID: 1671226.
[36] Defesche JC, Lansberg PJ, Umans-Eckenhausen MA, Kastelein JJ. Advanced method for the identification of patients with inherited hypercholesterolemia. Semin Vasc Med 2004 Feb;4(1):59–65. PubMed PMID: 15199434.
[37] Programme Whohg. Familial hypercholesterolaemia (FH) : report of a second WHO consultation, Geneva, 4 September 1998. Geneva: World Health Organization; 1999.
[38] Casula M, Olmastroni E, Pirillo A, Catapano AL. Members of the Lipigen Steering C, center PIC, et al. Evaluation of the performance of Dutch Lipid Clinic Network score in an Italian FH population: the LIPIGEN study. Atherosclerosis 2018 Oct;277:413–8. PubMed PMID: 30270079.
[39] Damgaard D, Larsen ML, Nissen PH, Jensen JM, Jensen HK, Soerensen VR, et al. The relationship of molecular genetic to clinical diagnosis of familial hypercholesterolemia in a Danish population. Atherosclerosis 2005 May;180(1):155–60. PubMed PMID: 15823288.
[40] Ruel I, Brisson D, Aljenedil S, Awan Z, Baass A, Belanger A, et al. Simplified Canadian definition for familial hypercholesterolemia. Can J Cardiol 2018 Sep;34(9):1210–4. PubMed PMID: 30093300.
[41] Haralambos K, Whatley SD, Edwards R, Gingell R, Townsend D, Ashfield-Watt P, et al. Clinical experience of scoring criteria for Familial Hypercholesterolaemia (FH) genetic testing in Wales. Atherosclerosis 2015 May;240(1):190–6. PubMed PMID: 25797312.
[42] deGoma EM, Ahmad ZS, O'Brien EC, Kindt I, Shrader P, Newman CB, et al. Treatment gaps in adults with heterozygous familial hypercholesterolemia in the United States: data from the CASCADE-FH registry. Circul Cardiovasc Genet 2016 Jun;9(3):240–9. PubMed PMID: 27013694. Pubmed Central PMCID: 5315030.
[43] Amrock SM, Duell PB, Knickelbine T, Martin SS, O'Brien EC, Watson KE, et al. Health disparities among adult patients with a phenotypic diagnosis of familial hypercholesterolemia in the CASCADE-FH (TM) patient registry. Atherosclerosis 2017 Dec;267:19–26. PubMed PMID: WOS:000416032800003. English.
[44] de Isla LP, Alonso R, Watts GF, Mata N, Cerezo AS, Muniz O, et al. Attainment of LDL-cholesterol treatment goals in patients with familial hypercholesterolemia 5-year SAFEHEART registry follow-up. J Am Coll Cardiol 2016 Mar 22;67(11):1278–85. PubMed PMID: WOS:000371881400003. English.
[45] Vrablik M, Raslova K, Vohnout B, Blaha V, Satny M, Kyselak O, et al. Real-life LDL-C treatment goals achievement in patients with heterozygous familial hypercholesterolemia in the Czech Republic and Slovakia: results of the PLANET registry. Atherosclerosis 2018 Oct;277:355–61. PubMed PMID: 30270071.
[46] Schmidt N, Dressel A, Grammer TB, Gouni-Berthold I, Julius U, Kassner U, et al. Lipid-modifying therapy and low-density lipoprotein cholesterol goal attainment in patients with familial hypercholesterolemia in Germany: the CaReHigh Registry. Atherosclerosis 2018 Oct;277:314–22. PubMed PMID: WOS:000445908000048. English.
[47] Kayikcioglu M, Tokgozoglu L, Yilmaz M, Kaynar L, Aktan M, Durmus RB, et al. A nation-wide survey of patients with homozygous familial hypercholesterolemia phenotype undergoing LDL-apheresis in Turkey (A-HIT 1 registry). Atherosclerosis 2018 Mar;270:42–8. PubMed PMID: WOS:000427639800007. English.
[48] Martin AC, Gidding SS, Wiegman A, Watts GF. Knowns and unknowns in the care of pediatric familial hypercholesterolemia. J Lipid Res 2017 Sep;58(9):1765–76. PubMed PMID: 28701353. Pubmed Central PMCID: 5580907.
[49] Ramaswami U, Cooper J, Humphries SE, Group FHPRS. The UK paediatric familial hypercholesterolaemia register: preliminary data. Arch Dis Child 2017 Mar;102(3):255–60. PubMed PMID: 26948823. Pubmed Central PMCID: 5339547.
[50] Vrablik M, Vaclova M, Tichy L, Soska V, Blaha V, Fajkusova L, et al. Familial hypercholesterolemia in the Czech Republic: more than 17 years of systematic screening within the MedPed project. Physiol Res 2017 Apr 5;66(Suppl 1):S1–9. PubMed PMID: 28379025.
[51] Mollaki V, Progias P, Drogari E. Novel LDLR variants in patients with familial hypercholesterolemia: in silico analysis as a tool to predict pathogenic variants in children and their families. Ann Hum Genet 2013 Sep;77(5):426–34. PubMed PMID: 23815734.
[52] Ramaswami U, Futema M, Bogsrud MP, Holven KB, van Lennep JR, Wiegman A, et al. Comparison of the characteristics at diagnosis and treatment of children with heterozygous familial hypercholesterolaemia (FH) from eight European countries. Atherosclerosis 2020 Jan;292:178–87. PubMed PMID: WOS:000503400300024. English.
[53] D'Agostino RB, Vasan RS, Pencina MJ, Wolf PA, Cobain M, Massaro JM, et al. General cardiovascular risk profile for use in primary care - the Framingham Heart Study. Circulation 2008 Feb 12;117(6):743–53. PubMed PMID: WOS:000253090700005. English.
[54] Conroy RM, Pyorala K, Fitzgerald AP, Sans S, Menotti A, De Backer G, et al. Estimation of ten-year risk of fatal cardiovascular disease in Europe: the SCORE project. Eur Heart J 2003 Jun;24(11):987–1003. PubMed PMID: WOS:000183577800002. English.
[55] Villa G, Wong B, Kutikova L, Ray KK, Mata P, Bruckert E. Prediction of cardiovascular risk in patients with familial hypercholesterolaemia. Eur Heart J Qual Care Clin Outcomes 2017 Oct 1;3(4):274–80. PubMed PMID: 29044394.
[56] Perez de Isla L, Alonso R, Mata N, Fernandez-Perez C, Muniz O, Diaz-Diaz JL, et al. Predicting cardiovascular events in familial hypercholesterolemia: the SAFEHEART registry (Spanish familial hypercholesterolemia cohort study). Circulation 2017 May 30;135(22):2133–44. PubMed PMID: 28275165.
[57] Paquette M, Dufour R, Baass A. The Montreal-FH-SCORE: a new score to predict cardiovascular events in familial hypercholesterolemia. J Clin Lipidol 2017 Jan - Feb;11(1):80–6. PubMed PMID: 28391914.
[58] Collaboration EASFHS, Vallejo-Vaz AJ, Akram A, Kondapally Seshasai SR, Cole D, Watts GF, 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 2016 Dec;22:1–32. PubMed PMID: 27939304.
[59] Hegele RA, Boren J, Ginsberg HN, Arca M, Averna M, Binder CJ, et al. Rare dyslipidaemias, from phenotype to genotype to management: a European Atherosclerosis Society task force consensus statement. Lancet Diabetes Endo 2020 Jan;8(1):50–67. PubMed PMID: WOS:000503224800013. English.