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ARTICLE

An spanish study of secondary findings in families affected with mendelian disorders: choices, prevalence and family history

Marta Codina-Solà[1,2,3 ✉], Laura Trujillano[1,2,4], Anna Abulí[1,2,3], Eulàlia Rovira-Moreno[1,2,3], Patricia Muñoz-Cabello[1,2], Berta Campos[1,2], Paula Fernández-Álvarez[1,2,3], Dolors Palau[1], Estela Carrasco[5,6], Irene Valenzuela[1,2,3], Anna Maria. Cueto-González[1,2], Amaia Lasa-Aranzasti[1,2,3], Javier Limeres[7,8,9], Jordi Leno-Colorado[1,2], Mar Costa-Roger[1,2], Alejandro Moles-Fernández[1,2], Judith Balmaña[5,6], Orland Díez[2,5], Ivon Cuscó[1,2,3,4,11], Elena Garcia-Arumí[1,2,3,4,10] and Eduardo Fidel Tizzano[1,2,3]

Clinical exome sequencing has the potential to identify pathogenic variants unrelated to the purpose of the study (secondary findings, SFs). Data describing actual choices of SFs in participants in a clinical setting and factors influencing their decision are virtually non-existant in Europe. In this work, we report the acceptance rate of SFs, calculate their prevalence and study factors associated with the decision in a cohort of patients affected with a rare genetic disorder in a Spanish Hospital. Finally, we re-examine the presence of previously non reported family history in positive cases. We retrospectively reviewed informed consent choices and SF results from 824 unrelated probands affected with rare genetic disorders who underwent whole-genome or exome sequencing. Ninety percent of families (740/824) affected with rare disorders wished to be informed of SFs. Declining SFs was associated with a prenatal setting (30% vs. 8.7%, $p = 0.025$), consanguinity (19% vs. 8.7%, $p = 0.013$), male gender (10.6% vs. 1.5%, $p = 0.00865$) and the proband being a minor (10.6% vs. 1.5%, $p = 0.014$). Overall, 27 pathogenic or likely pathogenic variants were identified in 27 individuals, with an SF prevalence of 3.6%. Disclosure of SFs increased the percentage of positive family histories and resulted in early diagnosis or changes in the management of 10 individuals from five families. We show that the acceptance of SFs in Spain is high and the disclosure of SFs leads to a clinically meaningful change in the medical management of individuals.

INTRODUCTION

Exome sequencing has become an established clinical test for the diagnosis of rare genetic disorders, with a variable diagnostic yield depending on the clinical indication. Besides identifying the genetic variant associated with the symptoms of a particular disease, it allows the screening of additional variants unrelated to the purpose of the study. In 2013, the American College of Medical Genetics (ACMG) coined the term "incidental findings" and defined a list of 56 genes for which it recommended reporting pathogenic variants, regardless of the patient's preferences [1]. Later, in 2017, it revised the terminology to "secondary findings" (SFs), updated the list to 59 genes and included the option to opt out (v2) [2]. The latest update (v3.1) published in 2022, includes 78 genes and encourages the continued nomination of genes to further expand the list [3, 4]. This position differs from that of the European Society of Human Genetics (ESHG), which advocates for a cautious approach to opportunistic genome screening and

recommends a targeted approach in which unsolicited findings are minimized [5, 6]. In the context of European countries and their mostly publicly funded healthcare systems, the ESHG argues that derived healthcare expenditures must be taken into account. In addition, there is an ethical debate surrounding the benefits and limitations of studying adult-onset conditions in minors, among other questions. However, both societies acknowledge that further studies are needed to understand the implications of reporting SFs and resolve ongoing debates, such as ethical issues, clinical utility and penetrance in the absence of family history.

Despite the ongoing debate of scientific societies, several studies have shown a high acceptance of SFs among participants of exome sequencing studies, ranging from 76% to 93.5%. Although published studies vary greatly according to participants included (adults, children or prenatal) setting (clinical or research), type of findings offered (closed vs. opened list), situation (hypothetical vs. real) and age of the proband, the high

[1]Medicine Genetics Group, Vall d'Hebron Institut de Recerca (VHIR), Vall d'Hebron Barcelona Hospital Campus, Vall d'Hebron Hospital Universitari, Barcelona, Spain. [2]Department of Clinical and Molecular Genetics, Vall d'Hebron Barcelona Hospital Campus, Vall d'Hebron Hospital Universitari, Barcelona, Spain. [3]European Reference Network on Rare Congenital Malformations and Rare Intellectual Disability ERN-ITHACA, Barcelona, Spain. [4]Instituto de Salud Carlos III, Centro de Investigación Biomédica en Red de Enfermedades Raras (CIBERER), Barcelona, Spain. [5]Hereditary Cancer Genetics Group, Vall d'Hebron Institute of Oncology (VHIO), Vall d'Hebron Barcelona Hospital Campus, Hospital Universitari Vall d'Hebron, Barcelona, Spain. [6]Medical Oncology Department, Vall d'Hebron Barcelona Hospital Campus, Vall d'Hebron Hospital Universitari, Barcelona, Spain. [7]European Reference Networks for rare, low prevalence and complex diseases of the heart (ERN GUARD-Heart), Barcelona, Spain. [8]Unidad de Cardiopatías Familiares, Servicio de Cardiología, Vall d'Hebron Institut de Recerca (VHIR), Vall d'Hebron Barcelona Hospital Campus, Vall d'Hebron Hospital Universitari, Barcelona, Spain. [9]Centre for Biomedical Network Research on Cardiovascular Diseases (CIBERCV), Madrid, Spain. [10]Research Group on Neuromuscular and Mitochondrial Disorders, Vall d'Hebron Institut de Recerca (VHIR), Vall d'Hebron Barcelona Hospital Campus, Vall d'Hebron Hospital Universitari, Barcelona, Spain. [11]Present address: Department of Genetics, Hospital Sant Pau, Barcelona, Spain. ✉email: marta.codina@vallhebron.cat

Table 1. Summary of previous studies exploring participant's preferences for SFs.

Authors, ref.	Participants (n)	Site	Age of participants	Categories of SF	Setting	Situation	Rate of acceptance
Shahmirzadi et al. [7]	200	USA/Canada	Adults, children	Four categories of SFs defined accordingly to age of onset of the disease and reproductive actionability	Clinical	Real world	94% (for any category)
Regier et al. [8]	1200	Canada	Adults	Discrete choice questionnaire evaluating 5 attributes (penetrance, treatability, severity of the disease, carrier status and cost of receiving the results	Research	Hypothethical	66% (for high-penetrance, medically treatable disorders)
Fiallos et al. [9]	790	USA	Adults, children	ACMG v1	Research	Real world	83%
Wynn et al. [22]	219	USA	Adults	11 types of genetic results with different degree of risk; availability and effectiveness of screening, prevention and treatment and acceptability of screening, prevention and treatment	Research	Hypothethical	73% (all results)
Rini et al. [11]	152	USA	Adults	Six categories of SFs with low medical actionability	Research	Hypothethical	78%
Similuk et al. [12]	66	USA	Adults, children	ACMG v2	Research	Real world	98%
Swanson et al. [35]	685	USA	Children, prenatal	ACMG v2	Clinical	Real world	84%
Horiuchi et al. [14]	2480	Japan	Adults, children	ACMG v2	Research	Real world	69%
Rego et al. [15]	150	USA	Children, prenatal	Hypothetical categories of SFs dfined according to severity of the disease, availability of treatment, reproductive utility and age of onset.	Research	Hypothethical	79% (at least one category)
This study	824	Spain	Children, adult, prenatal	ACMG v2	Clinical	Real world	90%

acceptability of SFs is a consisting finding [7–15] (see Table 1 for a summary of the studies published until now). Some studies exploring factors that could influence the decision found that non-European ancestry, professional providing consent and anticipatory regret could modulate the participant's decision [9, 11]. However, to our knowledge, none of these studies have been carried out in Europe and most evaluated decisions in a research setting or in a hypothetical situation in which SFs were not actually returned to participants. Since participants enrolling in a research setting may show differences in healthcare-decision making and the hypothetical scenario may not reflect real situations, actual acceptance rates may vary in clinical situations.

Regarding their prevalence, previous studies have shown a consistent SF frequency, defined according to the v1 or v2 ACMG list, of 1.1 to 2.8% [8, 12, 16–23]. Few studies have evaluated their penetrance or assessed clinical and family data as a proxy. Data from Van Hout et al. indicated a lower penetrance for *BRCA1/2* pathogenic variants identified through the UK Biobank compared with previous estimates obtained from high-risk families, suggesting that SFs may be associated with lower risk in the absence of family history [20]. This poses important challenges for counseling and clinical management and questions the clinical utility of including genes for which low penetrance is already known.

The aim of this work was to establish the acceptance rate of being informed regarding SFs among 824 Spanish patients affected with a rare genetic disorder, analyze factors that influence this decision, and calculate the prevalence of SFs. Finally, we reviewed the presence of family history in positive cases as a proxy for penetrance.

SUBJECTS AND METHODS
Study population and data collection
The study was conducted at the Department of Clinical and Molecular Genetics at the Vall d'Hebron Hospital (Barcelona, Spain). The population included 824 families who were offered singleton exome or genome sequencing as part of consultation and follow-up studies for a rare disorder between September 2016 and March 2021. Exome or whole-genome sequencing was offered to all families in which a monogenic condition was suspected but previous genetic testing had failed to detect a cause. All families received extensive genetic counseling by a qualified professional (medical geneticist or genetic counselor). All professionals belonged to the same Department, ensuring consistency in the approach and content of the information provided in the pre-test session and informed consent process. A detailed family history, including at least three generations, was collected for all patients prior to exome sequencing.

All participants were offered to receive SFs as defined in the ACMG recommendations v2, which includes 59 genes [2]. The consenting process and consent form included discussion of the study purpose, option to receive only primary findings or primary and SFs, method for sample collection, data storage and future usage, reanalysis and authorization for future re-contacting. Consent was provided by either both parents or a legal representative if the patient was a minor under 16 years or if they were over 16 years but incapable of providing consent for themselves. Capable patients (16 years or older) provided consent, according to Spanish law (21/2000), which regulates the patient's autonomy and rights to clinical information. The consent form provided space for participants to opt in or opt out to receive SFs. Informed consent forms that were left blank were considered invalid and were not included in the study. Consent forms were manually reviewed by two investigators. The choice to receive SFs, as well as the person who provided the consent and relationship to the study participant were recorded. All participants signed the informed consent approved by the local IRB.

Demographic and clinical data were obtained from the clinical history and clinical database of the clinical unit, including age of the proband when the informed consent was signed, sex of the proband and presence of consanguinity in the family. Only postnatal cases were included in the analysis of factors influencing the decision (n = 749). For the prevalence analysis, only cases who did consent to SF analysis and whose final report was completed were included (n = 740).

Table 2. Factors influencing choice of receiving SFs.

Variable		# (%)	Choice of SF		P value
			No (%)	Yes (%)	(FET)
Setting	Prenatal	20 (2%)	6 (30%)	14 (70%)	0.03
	Postnatal	749 (91%)	72 (10%)	677 (90%	
	Pregnancy termination	55 (7%)	6 (11%)	49 (89%)	
Person providing consent[a]	Parent or legal representative	621 (83%)	60 (10%)	561 (90%)	0.68
	Self	128 (17%)	12 (9%)	116 (91%)	
Consanguinity[a]	Yes	63 (8%)	12 (19%)	51 (81%)	0.01
	No	686 (92%)	60 (9%)	626 (91%)	
Sex of the consenter[b]	Female	81 (63%)	3 (4%)	78 (96%)	0.01
	Male	47 (37%)	9 (19%)	38 (81%)	
Age of the proband when another is providing consent[c]	Minor (<=16)	556 (90%)	59 (11%)	497 (89%)	0.01
	Adult (>16)	65 (10%)	1 (2%)	64 (98%)	

P values were calculated using Fisher's Exact Test (FET).
[a]This analysis includes only postnatal cases.
[b]This analysis includes only postnatal cases and participants providing consent for one-self.
[c]This analysis includes only postnatal cases and cases for which a parent or legal representative is providing consent. See Fig. 1 for a general scheme of the analysis performed.

Definition of SF and variant classification
An SF was defined according to the ACMG v2 list (59 genes) [2]; variants were classified according to ACMG guidelines [24]. All variants were independently reviewed by two clinical molecular geneticists to ensure the classification process. Variants that were classified discordantly were further discussed by a multidisciplinary team that included medical geneticists, genetic counselors, molecular geneticists, cardiologists and oncologists and only those considered pathogenic or likely pathogenic were included in the final analysis.

Statistical analysis
Categorical variables were reported as frequency (%) and compared between groups by Fisher's exact test (FET, two-sided). A P value < 0.05 was considered significant.

Personal and family history review
In each case, a three-generation pedigree was collected by a trained genetic professional (either a genetic counselor or a clinical geneticist) in the pre-test session. The family history taken during the pre-test session was reviewed and compared to the updated family history obtained at the time of SF disclosure. All at-risk relatives of index cases were offered genetic counseling and direct testing at the Hospital. Carriers were referred for clinical evaluation and the clinical history was reviewed thereafter. Personal history was considered positive if the individual was clinically diagnosed with the genetic condition or showed signs compatible with the disorder.

RESULTS
Preferences for receiving SFs
We retrospectively examined demographic and clinical factors related to declining SFs. Overall, 90% of patients wished to receive SFs (Table 2 and Fig. 1). We first examined if the setting in which exome sequencing was offered (during an ongoing pregnancy, after pregnancy termination or in a postnatal context) influenced the SF acceptance. Declining SFs was positively associated with genetic testing being offered in a prenatal setting with 30% of participants not wanting to receive SFs, compared with 8.7% and 11% after pregnancy termination or in a postnatal case, respectively (FET, $p = 0.025$).

In order to avoid confounding factors, only postnatal cases were included for further analysis (Fig. 1). There were no differences in SF acceptance rates according to being the patient or their parent/legal representative who provided the consent (FET, $p = 0.68$). Regarding consanguinity, 19% of cases with self-reported consanguinity declined SFs compared with 8% for non-consanguineous families (FET, $p = 0.013$).
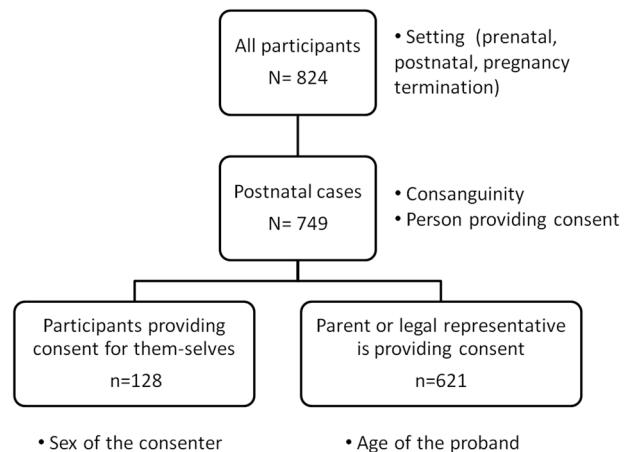

Fig. 1 Scheme showing different subgroups used in each statistical analysis. For the whole cohort, preferences were analysed according to the setting (prenatal, postnatal, pregnancy termination). We evaluated whether consanguinity and the person providing consent influenced choice only in the cohort of postnatal cases. Finally we evaluated how the sex of the consenter influenced acceptance for participants providing consent for themselves and if the age of the proband influenced choices when a parent or legal representative was providing consent.

We then separately examined factors influencing the acceptance rate in patients who provided consent for themselves and in patients for whom a parent or a legal representative provided consent. When patients provided consent for themselves, being female was associated with a higher acceptance rate (96%) than being male (81%) (FET, $p = 0.00865$). In addition, the proband's age was a factor that influenced choice in patients for whom a parent or a legal representative provided consent. Adult age of the proband was associated with a higher SF acceptance rate, compared with acceptance for probands who were under 16 (98% vs. 89%).

Prevalence of SFs and characteristics
Estimates of prevalence were based on the 740 probands who underwent genome ($n = 4$) or exome sequencing ($n = 736$) and
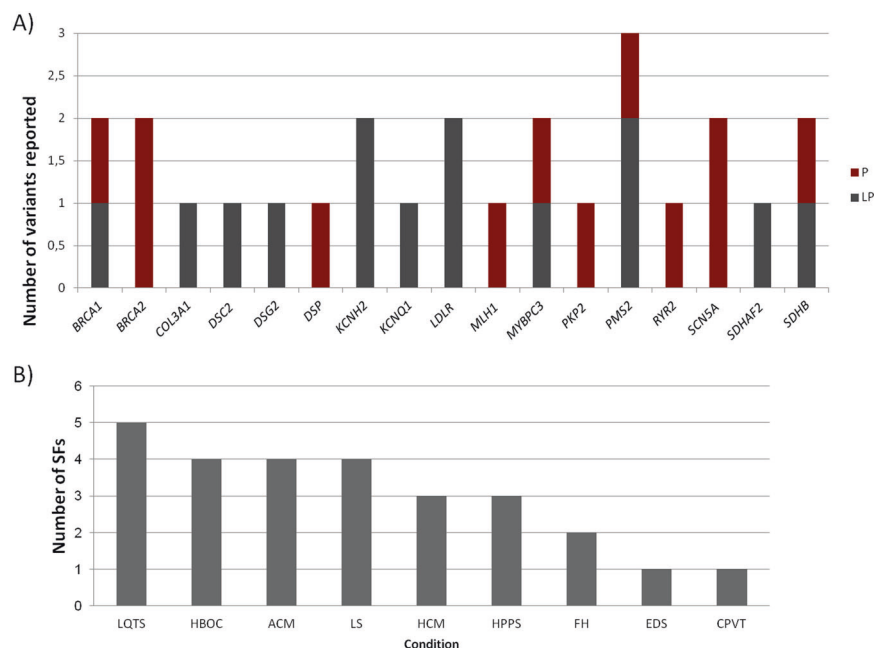
Fig. 2 Genes and conditions identified as SFs. A Number of pathogenic and likely pathogenic variants identified per gene in the whole cohort analysed (27/740). LP likely pathogenic, P pathogenic. **B** Conditions associated to the SFs identified in the cohort. LQTS Long QT syndrome, HBOC Hereditary Breast and Ovarian Cancer, ACM Arrhythmogenic Cardiomyopathy, LS Lynch syndrome, HCM Hypertrophic Cardiomyopahty, HPPS Hereditary Paraganglioma-Pheochromocytoma Syndromes, FH Familial Hypercholesterolemia, EDS Ehlers-Danlos syndrome and CPVT Catecholaminergic Polymorphic Ventricular Tachycardia.

wished to receive SFs. A total of 27 pathogenic or likely pathogenic variants were identified in 27 individuals, with an SF prevalence of 3.6%. Six variants were downgraded to variants of unknown significance due to insufficient evidence and, therefore, excluded from further analysis (see Supplementary Table 1 for details). All variants classified as pathogenic or likely pathogenic were unique and were found in 18 different genes, with eight genes (*BRCA1, BRCA2, KCNH2, LDLR, MYBPC3, PKP2, PMS2, SCN5A, SDHB*) harboring more than one variant (Fig. 2A). The most common disorders were long QT syndrome (LQTS), Arrythmogenic cardiomyopathy (ACM), Hereditary Breast and Ovarian Cancer (HBOC) and Lynch syndrome (LS) (Fig. 2B).

Family history known prior to result disclosure and following SF disclosure

Family history was available in only 25 of the 27 families as one of the probands was adopted and another born via egg donation and their father was not a carrier of the SF identified (Fig. 3). Before testing family history was positive in two cases (8%). In the first case where a pathogenic variant in *LDLR* was found (Family 17), the proband was diagnosed with hypercholesterolemia, but the cholesterol levels or the family history did not raise any suspicion of familial hypercholesterolemia nor did it meet Dutch criteria for genetic testing [25]. In the second case (Family 18), a pathogenic *BRCA2* variant was identified. The family referred a paternal aunt who died of breast cancer before the age of 40, and a paternal uncle with prostate cancer diagnosed at 71 years of age but no affected individual was available for genetic testing (see Supplementary Table 2 for detailed family history before and after SF disclosure).

Of the 23 cases with negative family history before testing, two participants had a *de novo* variant (*BRCA1, RYR2*), explaining the lack of family history (Fig. 3 and Supplementary Table 2). Reinterrogation after SF disclosure revealed additional family history in 4/23 cases (Fig. 3 and Supplementary Table 2). In one case, in which an SF in *BRCA1* was identified (Family 25) the genetic result was previously known in the family but the father of

the proband had not undergone genetic testing and it was not reported to the clinical geneticist during the pre-test session.

Clinical utility and results of clinical screening after SF disclosure

SF disclosure resulted in a mean of 2.7 direct studies per family, with a total of 73 genetic studies being performed. Genetic testing was offered to at-risk relatives, according to age and genetic testing recommendations in asymptomatic minors. After familial genetic testing, 28 relatives were found to carry the same SF identified in the proband, with a total of 55 individuals being carriers of an SF (including probands).

All probands and carrier relatives were offered a referral for a complete clinical evaluation if indicated according to the condition associated with the SF and age of the individual. During these evaluations, 10 individuals from five families showed signs compatible with the condition (Fig. 3, Supplementary Table 2). Therefore, after clinical evaluation and specific family history reinterrogation, 11/27 (41%) families had at least one individual presenting signs compatible with the SF identified.

Regarding changes in clinical management, in three of the families with an SF in a gene related to long QT syndrome 1 or 2 (*KCNQ1* and *KCNH2*), all relatives showing the respective variants started beta blocker medication (Families 10, 12 and 13), although a prolonged QT interval was only observed in individuals from Family 12. Also, the proband harboring a pathogenic variant in *COL3A1* (Family 19) had a minimal aortic dilation seen by echocardiography (Supplementary Table 2). In contrast, none of the individuals carrying a pathogenic or likely pathogenic variant in a gene related to arrhythmogenic cardiopathy (11 individuals from four families) showed any sign compatible with the disease after the first cardiological evaluation. Actionability of SFs in cancer susceptibility genes has been previously reported [26, 27]. As previously shown, the clinical evaluation in the two families with pathogenic variants in *SHDB* (Families 1 and 9) resulted in the diagnoses of two paragangliomas at ages 55 and 35 in family 1 and the diagnosis of one paraganglioma at age 10 in family 9.
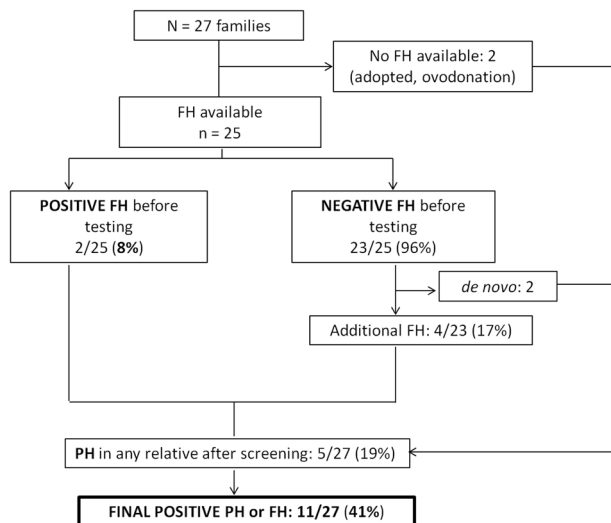
Fig. 3 Family and personal history before and after the disclosure of the SF. From 27 families, in two, family history was not available. In the remaining 25 cases, only two had a positive family history before testing. In the 23 cases with a negative family history before testing, two cases were de novo. Disclosure of the SF revealed a positive family history in 4 additional families. When probands and carrier relatives were evaluated for the condition related to the SF, 5 individuals from 27 families showed signs compatible with the condition. Considering both personal and family history, 11/27 families showed the disease or a previous history related with the condition. PH personal history, FH family history.

Finally, in one of the families in which a pathogenic variant in *PMS2* was found (Family 16), one of the relatives was found to have two tubular adenomas with low-grade dysplasia.

DISCUSSION

This study describes the acceptance of SFs, demographic factors associated with disclosure decision-making, prevalence and family history in a cohort of patients with rare disorders followed up in a clinical setting in a tertiary Hospital in Spain. To our knowledge, this is the first study conducted in Europe exploring patients' preferences for SFs in a routine clinical scenario. In our study, the majority of patients (90%) opted to receive SFs. This is in agreement with previous studies reporting that 76–97% of patients wished to know SFs [7, 9, 10, 28, 29], although differences among studies made direct comparisons difficult. Interestingly, acceptance rates of SFs do not seem to differ among studies conducted in real situations or hypothetical scenarios. It is known that family history and risk perception influence genetic testing uptake across different conditions and situations [30–32]. In the context of offering SFs in which patients lack a previous experience with the conditions screened and where there is a low probability of identifying a positive result, the situation may be very similar to a hypothetic scenario. This highlights the difficulty of conveying the implications of receiving SFs and the difficulty of engaging the patient in the discussion and obtaining a truly informed consent in the context of genomic sequencing [33]. Given these nuances, it is recommended that pre-test genetic counseling sessions are provided by qualified professionals, such as genetic counselors or clinical geneticists.

In our study, we found significant differences in the SF acceptance rate between families who were offered genetic testing during an ongoing pregnancy, after pregnancy termination or in a postnatal context. Being offered SFs during an ongoing pregnancy was associated with a lower acceptance of SF. Although SFs in a prenatal context could facilitate information

to couples of reproductive utility, this difference could be due to participants experiencing a greater decisional burden or information overload during the genetic counseling process, making them more prone to decline SFs if a decision regarding the pregnancy had not been reached. An alternative two-step model for informed consent, where discussion of SFs is carried out after the delivery of primary findings may help families reduce the decisional burden in a prenatal context [34]. However, further studies are needed to assess the feasibility and acceptance of a two-step model in a prenatal context. There is limited information regarding the preferences for SFs during an ongoing pregnancy, but a previous study by Swanson et al. carried out in the USA, reported an acceptance rate of 86.2% in fetal exome sequencing [35], which is higher than our results (70%). The same study did not report any differences among families where exome sequencing had been offered in a postnatal setting or after pregnancy termination or loss [35]. The difference between our results and those of Swanson et al. could be due to cultural differences between our subjects or sociodemographic differences between the prenatal and pediatric subgroups. Further studies regarding SF acceptance in the prenatal context are required to confirm our findings.

We also found significant differences in the return of SFs among consanguineous and non-consanguineous families, with a higher frequency of consanguineous families declining SFs. Since the frequency of consanguineous unions varies according to geographical origin and ethnicity, we hypothesiz that this difference is driven by a difference in ancestry among consanguineous and non-consanguineous families. Therefore, our results could corroborate previous studies in which families of non-European ancestry were more prone to decline SFs [9]. As discussed by Fiallos et al. this could be due to differences in attitudes towards genetic testing in individuals of non-European background, which usually represent underserved communities.

As previously described by Fiallos et al. we found no significant differences according to the person who provided consent (patient vs. parent or legal representative). When considering only the group of patients that provided consent themselves, we observed that women requested SFs more frequently. This could be explained by a bias in the examples used when discussing SF. Genetic professionals usually use practical examples of conditions included in the ACMG list, with ovarian and breast cancer risk associated with *BRCA1/2* pathogenic variants being one of the most known by the general public [36]. Therefore, using examples of conditions most commonly affecting women could be responsible for a higher acceptance rate among females and a lower acceptance among men. Finally, when considering only cases in which parents or legal representatives provided consent, we observed that SF acceptance was lower when the proband was a minor. This difference could be due to a reticence from parents and legal guardians to know the risk of adult-onset conditions in children in order to respect their child's right not to know. Although the principle of autonomy may be compromised when SFs are disclosed in children before the age of consent, it is also true that the disclosure of SFs results in a familial benefit. Children belong to a larger system (the family) and therefore, information about SFs of adult-onset may be beneficial to other family members [37]. Genetic counseling should be provided to families to allow them to carefully balance the benefits and limitations of disclosing SFs in a child.

Family history data showed that only 8% (2/25) of family histories were positive in the pre-test compilation of family information (2/23, 8.7% when excluding de novo cases). After SF disclosure, this percentage increased to 26% (6/23). A similar trend was observed by Hart et al. in which the frequency of a positive family history increased from 34% to 48% after SF disclosure [17]. As hypothesized by the authors, this could be due to the passage of time, targeted family history questions or the triggering of specific recollections.

The long follow-up time of our study (at least 1 year after the SF disclosure) allowed us to assess the results of clinical evaluations performed in the proband and carrier relatives after the disclosure of an SF. Overall, we detected after clinical evaluation and specific reinterrogation of family history, 11/27 families showed signs compatible with the SF identified. This resulted in the diagnosis of the SF-related condition in 11 individuals from five families. This allowed the start of beta blocker medication in three families with long QT syndrome. The clinical utility of disclosing SF of genes related to inherited cardiac conditions has been previously shown by Ormondroyd et al. [38]. In addition, early cancer detection was possible in two families with a pathogenic variant in *SDHB*. Our results are remarkable considering the relatively low penetrance of pathogenic variants in *SDHB* (between 8% and 37% across several studies) [39].

In contrast, despite being the third most common condition in our cohort, none of the 11 individuals from 4 families with a pathogenic variant in a gene related to arrhythmogenic cardiopathy (*DSC2, DSG2, DSP, PKP2*) showed any sign compatible with the disease after the first evaluation. This is in agreement with a recent study showing that 1 in 435 individuals of European descent carries a pathogenic variant in arrhythmogenic cardiopathy, with an estimated penetrance of 6% in the general population [40]. This could explain the results observed in our cohort and suggests, as previously hypothesized, that penetrance may be higher in individuals with a strong family background compared to those without it [41]. This fact may prompt to rethink the clinical utility of including low penetrance genes as SFs given the potential consequences of their return, such as overdiagnosis, emotional and psychological impact and healthcare expenses. Further studies assessing the prevalence and penetrance of SFs in large general population cohorts are needed to refine penetrance estimates and assess its clinical utility in the absence of family history.

Besides this matter, the definition of a closed list of SFs, such as the one elaborated by the ACMG, has other relevant potential issues. First, it does not take into account patient's preferences. For example, some families may find knowing their reproductive risk more useful than their risk of developing a late-onset condition. Second, a closed list of SFs does not take into account that the penetrance or frequency of a disease may vary across different countries. In this sense, the last update of the ACMG SFs list (v3.1) includes hereditary *TTR* (transthyretin) amyloidosis as a new condition to screen for. It is widely known that the prevalence of *TTR* pathogenic variants varies widely according to ethnicity, since approximately 3.0–3.9% of African Americans are heterozygous for the pathogenic variant p.(Val142Ile) and that penetrance of other pathogenic variants, such as p.(Val50Met), varies widely according to geographic region and it is higher in endemic foci [42, 43]. Therefore, a closed list of genes may need to be modified depending on the country where it is applied in terms of inclusion of genes and clinical management of carrier individuals. In the same direction, availability of treatments may vary among different countries due to differences in healthcare system and drug regulation. As an example, *RPE65*-related retinopathy was included in the ACMG v3.0 due to the availability of gene therapy treatment, but is not yet available in many countries [44]. Despite the limitations of the ACMG guidelines, they are currently the only guidelines that suggests which findings should be reported across diverse conditions and are regularly updated. Indeed, the results of our study following the ACMG guidelines show the favorable opinion of Spanish patients towards reporting additional genetic findings.

LIMITATIONS
This study has some limitations. First, this was a retrospective, cross-sectional analysis and we had limitations regarding the data

available for the preference analysis, such as self-reported ethnic origin. For instance, we lacked information regarding ancestry, since it is not usually collected in our database. Second, our sample size is relatively low to estimate the penetrance of SFs. Third, some of the families refused follow-up actions and, therefore, our estimates of positive history after the disclosure of SFs may be underestimated.

CONCLUSIONS
To our knowledge, this study is the first to quantify choices for the return of SFs in a clinical context in Europe. In our study, the vast majority of families affected with rare disorders desired to receive information about SFs. We also identified several factors associated with SF preferences, such as the postnatal setting, no consanguinity, female consenter and adult age of the proband. These findings provide suggestions for future research, especially on how to adapt genetic counseling to ethnically diverse populations and provide unbiased information and examples of SFs. The high acceptance of SFs in our population suggests that return of results in the clinical context may be favorably received, particularly when information is offered by expert professionals (genetic counselor, clinical geneticist) and provides data for future guideline updates in the European context. Our results also showed that the disclosure of SFs led to a change in management and early diagnosis of related conditions in probands and carrier relatives, indicating that SF disclosure is of clinical utility. However, further issues should be addressed before implementing opportunistic genome screening, such as which type of genetic findings to return, how to best provide genetic counseling and what is the emotional and psychological impact of receiving SFs. The results of this study represent a suitable addition to the growing information available regarding SFs preference and familial implications, contributing towards the improvement of clinical practice and the development of future policies and research planning.

DATA AVAILABILITY
All data and methods used in the analysis are described or included in this article and the electronic Supplementary Information. Raw data is available upon request.

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AUTHOR CONTRIBUTIONS

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COMPETING INTERESTS
The authors declare no competing interests.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE
All individuals participating in the study or their parents/legal representatives signed the informed consent approved by the institutional review board (v1-2016, v2-2021).

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Correspondence and requests for materials should be addressed to Marta Codina-Solà.

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