Experience Delivering a 30 Gene Panel for Cardiovascular Disorders to Over 23,000 Individuals in an Unselected Population

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Introduction

Cardiovascular disease is the leading cause of death for women and men in the US, accounting for one-third of deaths worldwide. Major risk factors include high blood pressure, elevated LDL cholesterol, and smoking. In addition, genetic factors increase the risk of serious events such as cardiac arrest, heart failure, sudden cardiac death, and stroke. Many individuals with hereditary cardiovascular disorders are asymptomatic, which makes early diagnosis and treatment difficult and puts individuals at risk for adverse cardiovascular events.

Color's Hereditary Heart Health Genetic Test detects pathogenic variants in 30 genes associated with four major categories of cardiovascular disorders: arrhythmias, arteriopathies, cardiomyopathies, and familial hypercholesterolemia (FH). These cardiovascular genes meet expert consensus for having clinical validity and utility and are associated with cardiovascular disorders that have preventive measures and/or treatments available.[1]

Methods

All individuals were ordered a Color test by a healthcare provider that analyzes 30 genes associated with four major categories of cardiovascular disorders: arrhythmias, arteriopathies, cardiomyopathies, and FH. These cardiovascular genes meet expert consensus for having clinical validity and utility and are associated with cardiovascular disorders that have preventive measures and/or treatments available.[1] The 30 genes analyzed are *ACTA2*, *ACTC1*, *APOB*, *COL3A1*, *DSC2*, *DSG2*, *DSP*, *FBN1*, *GLA*, *KCNH2*, *KCNQ1*, *LDLR*, *LMNA*, *MYBPC3*, *MYH7*, *MYH11*, *MYL2*, *MYL3*, *PCSK9*, *PKP2*, *PRKAG2*, *RYR2*, *SCN5A*, *SMAD3*, *TGFBR1*, *TGFBR2*, *TMEM43*, *TNNI3*, *TNNT2*, and *TPM1*. In *APOB* analysis was limited to chr2:g.21229159_21229161 (codon 3527).

Laboratory procedures were performed at the Color laboratory under CLIA and CAP compliance. Briefly, DNA was extracted, enriched for select regions using SureSelect XT probes, and then sequenced using NextSeq 500/550 or NovaSeq 6000 instruments. Sequence reads were aligned against human genome reference GRCh37.p12, and variants were identified using a suite of bioinformatic tools designed to detect single nucleotide variants (SNVs), small insertions and deletions (indels, 2-50 bp), and large structural variants (SVs, > 50 bp).

Variants were classified according to the American College of Medical Genetics and Genomics 2015 guidelines for sequence variant interpretation,2 and all variant classifications were signed out by a board certified medical geneticist. Results were counted as positive if one or more pathogenic or likely pathogenic (hereafter referred to as pathogenic) variant was detected and negative if no variant or only benign, likely benign, or variant of uncertain significance was detected at the time of data collection.

Genetic testing results were returned to individuals and their healthcare providers via a secure online portal. For individuals with a positive result, educational information about the hereditary disorder(s) associated with the affected gene accompanied the genetic results. Due to the spectrum of disorders, uncertain penetrance, and overlapping of phenotype-genotype correlations that can be associated with some of these genes, educational information was curated to be sufficiently broad to apply to all individuals who may or may not have had an established clinical diagnosis.

Genetic counseling via telephone was included with genetic testing. All individuals included in this study consented to have their de-identified information used in anonymized studies. All personal and family health information was reported by the individual.

References

1. Green, R. C. et al. ACMG recommendations for reporting of incidental findings in clinical exome and genome sequencing. *Genet. Med.* 15, 565–574 (2013).
2. Richards, S. et al. Standards and guidelines for the interpretation of sequence variants: a joint consensus recommendation of the American College of Medical Genetics and Genomics and the Association for Molecular Pathology. *Genet. Med.* 17, 405–424 (2015).
3. Austin, M. A., Hutter, C. M., Zimmern, R. L. & Humphries, S. E. Genetic causes of monogenic heterozygous familial hypercholesterolemia: a HuGE prevalence review. *Am. J. Epidemiol.* 160, 407–420 (2004).

Results

Table 1. Cohort demographics

This cohort included 23,775 individuals. Pathogenic variants were identified in 248 individuals, for a pathogenic frequency of 1.04%. 10.3% of this cohort self-reported a personal history of a cardiovascular disorder. Other ethnicity includes Middle Eastern, Native American, unknown and no answer. Personal history of cardiovascular disorder includes heart attack, heart failure, arteriopathy, FH, arrhythmia, cardiomyopathy, and stroke. Family history of cardiovascular disorder includes any relative who died from cardiac or unexplained causes, or had a heart attack, heart failure, arteriopathy, FH, arrhythmia, cardiomyopathy, and stroke.

		Individuals (n)	Fraction of Population	Individuals w/ pathogenic variants (n)	Pathogenic Frequency
Total		23,775	1	248	1.04%
Gender	Female	15,789	0.664	169	1.07%
	Male	7986	0.336	79	0.99%
Age (Years)	18-30	3979	0.167	62	1.56%
	31-40	5288	0.222	54	1.02%
	41-50	5139	0.216	40	0.78%
	51-65	6851	0.288	71	1.04%
	65+	2518	0.106	21	0.83%
Ethnicity	Caucasian	15,303	0.644	175	1.14%
	Ashkenazi Jewish	1987	0.084	7	0.35%
	Multiple Ethnicities	1676	0.070	26	1.55%
	Asian	1294	0.054	14	1.08%
	Hispanic	976	0.041	5	0.51%
	African	583	0.025	5	0.86%
	Other	1956	0.082	16	0.82%
Personal history of cardiovascular disorder	True	2460	0.103	53	2.15%
	False	18,255	0.768	169	0.93%
	No Answer	3060	0.129	26	0.85%
Family history of cardiovascular disorder	True	13,760	0.579	154	1.12%
	False	2058	0.087	13	0.63%
	No Answer	7957	0.335	81	1.02%

Figure 1. Reports with pathogenic variants by gene

Number of samples (reports) with a pathogenic variant identified in each gene, by disorder category. Of the six individuals with pathogenic variants in GLA, four were female carriers and two were male. Of note, one individual in the cohort carried two different pathogenic variants in genes in the panel.

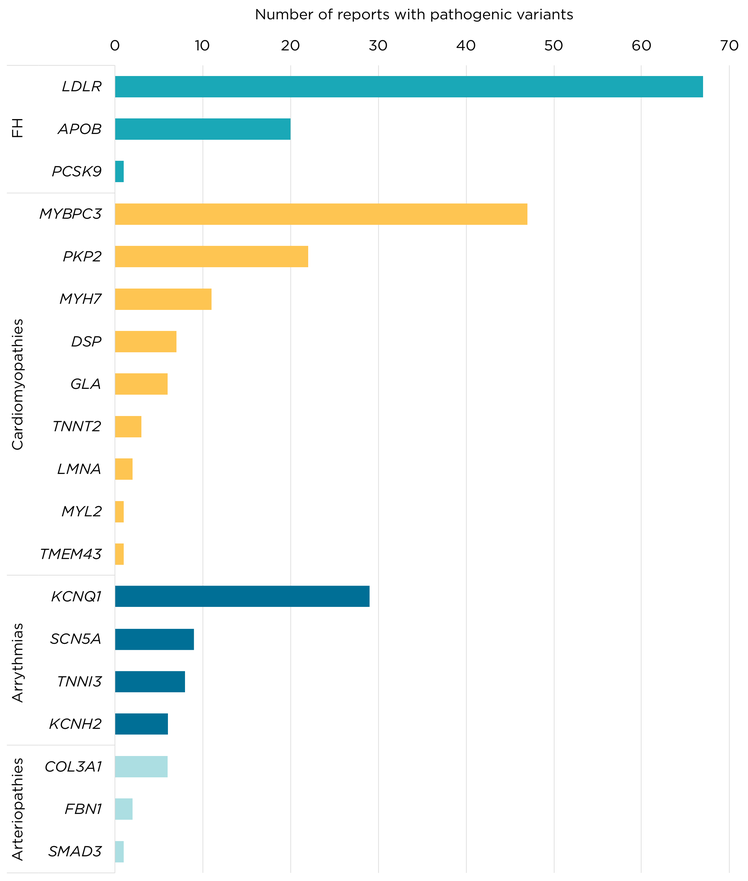


Figure 2. Personal and family history of cardiovascular disease in individuals with FH-related pathogenic variants

A total of 48 individuals with pathogenic variants in FH-related genes (*APOB*, *LDLR*, and *PCSK9*) provided sufficient health history information to calculate a score of clinical factors for FH diagnosis using the Dutch Lipid Clinic Criteria.[3] Only eight individuals (17%) had a score of six or higher using non-genetic criteria, indicative of a clinical diagnosis of FH.

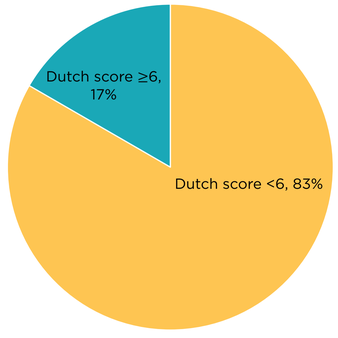
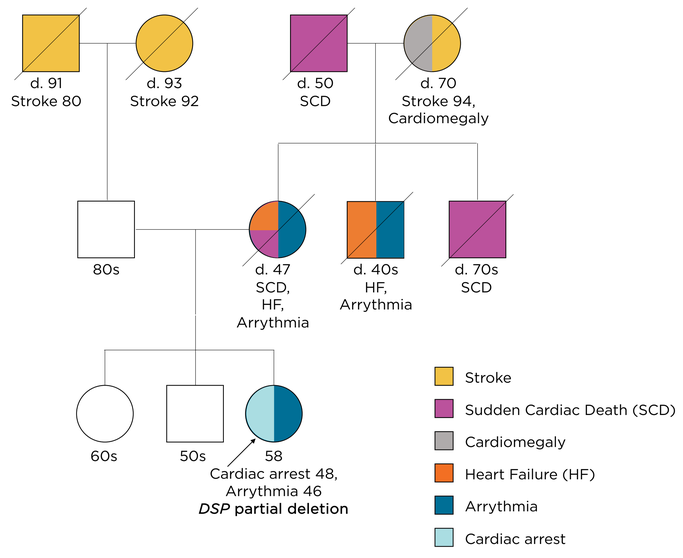


Figure 3. *DSP* case study

An individual with a personal and family history of cardiovascular disorders was self-motivated to seek genetic testing. A *DSP* exon 24 partial deletion was identified in this proband, consistent with multiple phenotypes suggestive of hereditary arrhythmias in this family.



Conclusions

- In a cohort of over 23,000 individuals referred for genetic testing independent of specific clinical characteristics, a pathogenic variant was identified in 1.27% of individuals.
- Pathogenic variants were identified in genes associated with all four disorders included in the panel (arrhythmias, arteriopathies, cardiomyopathies, and FH), most frequently in *LDLR*, *MYBPC3*, and *KCNQ1*.
- Of individuals with a pathogenic variant in an FH-related gene, 83% did not meet clinical criteria for FH diagnosis, and targeted care may have been missed in the absence of genetic diagnosis.
- In one case study, an individual who had not been previously referred for genetic testing used the patient-initiated model to access genetic testing. A pathogenic variant consistent with her personal and family history was identified and indicated a need for individualized follow-up care.