Genetic testing for the Tier 1 genomics conditions in a population-level cohort

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Introduction

An estimated 2 million people in the United States are at increased risk for the CDC Tier 1 genomic conditions hereditary breast and ovarian cancer (HBOC), Lynch syndrome (LS), and familial hypercholesterolemia (FH).[1] Recently, there has been momentum in the field towards offering genetic testing for these conditions at the population-level because such screening leads to early detection and intervention.[2–4] Color Genomics has been partnering with organizations to offer genetic testing for these conditions as an employer-sponsored, voluntary, confidential employee health benefit. Here, we report the frequency and spectrum of pathogenic variants in HBOC (*BRCA1* and *BRCA2*), LS (*MLH1, MSH2, MSH6, PMS2*, and *EPCAM*), and FH (*APOB, LDLR*, and *PCSK9*) in employees who represent an average-risk population-level cohort. Importantly, as these individuals were referred for genetic testing independent of testing guidelines, we also evaluated the results with respect to the Genetic/Familial High-Risk Assessments provided by the National Comprehensive Cancer Network (NCCN).

Methods

8918 employees (hereafter referred to as individuals) were ordered a Color test by a healthcare provider that analyzes genes in which variants have been associated with elevated risk for HBOC (*BRCA1* and *BRCA2*) and LS (*MLH1, MSH2, MSH6, PMS2*, and *EPCAM*). 1223 individuals were ordered a Color test by a healthcare provider that analyzes genes in which pathogenic variants have been associated with FH (*APOB, LDLR*, and *PCSK9*). Analysis, variant calling and reporting focus on the complete coding sequence and adjacent intronic sequence of the primary transcript(s), unless otherwise indicated: in *PMS2*, exons 12-15 were not analyzed; in *EPCAM*, only large deletions and duplications that include the 3' end of the gene and affect the expression of neighboring *MSH2* were analyzed; and in *APOB*, exon 1 is not analyzed. For the *LDLR* promoter region, the detection of deletions, duplications, and complex structural rearrangements may have been limited.

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 instrument. 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, 1 bp), 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,[5] and all variant classifications were signed out by a board certified medical geneticist or pathologist. 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 a benign, likely benign, or variant of uncertain significance was detected at the time of data collection.

All individuals consented to have their de-identified information and sample used in anonymized studies. All information was reported by the individual; information not provided was noted as such.

References

1. Tier1 | Tier 1 Genomic Applications Toolkit for Public Health Departments | Genomics | CDC. Accessed August 10, 2018.
2. Buchanan AH, Manickam K, Meyer MN, et al. *Genet Med.* 2018.
3. Pearlman R, Frankel WL, Swanson B, et al. *JAMA Oncol*. 2017.
4. Stoffel EM, Koeppe E, Everett J, et al. *Gastroenterology.* 2018.
5. Richards S, Aziz N, Bale S, et al. *Genet Med*. 2015.
6. Dewey FE, Murray MF, Overton JD, et al. *Science.* 2016.

Results

Table 1. Cohort demographic details for HBOC and LS.

The cohort that received genetic testing for HBOC and LS was almost equal parts men and women, the majority of which were under age 40. Nearly half of individuals were non-Caucasian. Only three individuals reported a personal history of cancer. PV, pathogenic variant.

		Individuals (n)	Population	Individuals w/ PV (n)	Pathogenic Frequency
Total		8918	100.0%	88	1.0%
Gender	Female	4743	53.2%	53	1.1%
	Male	4175	46.8%	35	0.8%
Age (Years)	18-30	2294	25.7%	14	0.6%
	31-40	2645	29.7%	33	1.2%
	41-50	2182	24.5%	25	1.1%
	51-65	1689	18.9%	16	0.9%
	65+	108	1.2%	0	0.0%
Ethnicity	African	219	2.5%	1	0.5%
	Ashkenazi Jewish	426	4.8%	13	3.1%
	Asian	1981	22.2%	22	1.1%
	Caucasian	5093	57.1%	41	0.8%
	Hispanic	373	4.2%	3	0.8%
	Multiple Ethnicity	553	6.2%	6	1.1%
	Native American	11	0.1%	0	0.0%
	Unknown	262	2.9%	2	0.8%
Personal Cancer History	Breast	63	0.7%	2	3.2%
	Ovarian	4	0.0%	0	0.0%
	Uterine	7	0.1%	0	0.0%
	Colorectal	7	0.1%	0	0.0%
	Melanoma	65	0.7%	1	1.5%
	Pancreatic	1	0.0%	0	0.0%
	Prostate	15	0.2%	0	0.0%
	Stomach	1	0.0%	0	0.0%
	Other cancer	137	1.5%	1	0.7%
	No cancer	7103	79.6%	69	1.0%
	Information not provided	1488	16.7%	12	0.8%

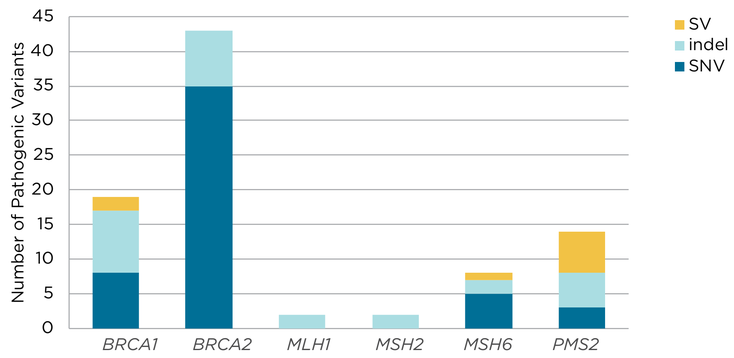
Table 2. Cohort demographic details for FH.

The majority of individuals who received genetic testing for FH were females and under age 40. Nearly half of individuals were non-Caucasian. Only four individuals reported a personal history of high cholesterol. An LDL-C level > 190 mg/dL was considered high. PV, pathogenic variant.

		Individuals (n)	Population	Individuals w/ PV (n)	Pathogenic Frequency
Total		1223	100.0%	4	0.3%
Gender	Female	753	61.6%	2	0.3%
	Male	470	38.4%	2	0.4%
Age (Years)	18-30	325	26.6%	0	0.0%
	31-40	373	30.5%	3	0.8%
	41-50	289	23.6%	0	0.0%
	51-65	225	18.4%	1	0.4%
	65+	11	0.9%	0	0.0%
Ethnicity	African	103	8.4%	1	1.0%
	Ashkenazi Jewish	49	4.0%	0	0.0%
	Asian	158	12.9%	1	0.6%
	Caucasian	710	58.1%	2	0.3%
	Hispanic	54	4.4%	0	0.0%
	Multiple Ethnicity	101	8.3%	0	0.0%
	Native American	1	0.1%	0	0.0%
	Unknown	47	3.8%	0	0.0%
Cholesterol	High	270	22.1%	4	1.5%
	Low/moderate	822	67.2%	0	0.0%
	Unsure	92	7.5%	0	0.0%
	Information not provided	39	3.2%	0	0.0%

Table 2. Genes with pathogenic variants, stratified by type in the HBOC and LS cohort.

(A) In the cohort that received genetic testing for HBOC and LS, pathogenic variants were most frequently found in *BRCA2* (43) and *PMS2* (14), respectively. No pathogenic variants were found in *EPCAM*. Types include single nucleotide variant (SNV), small insertion/deletion, (indel), and large structural variant (SV).

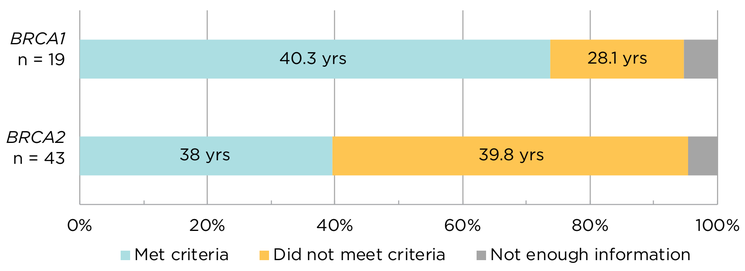


(B) In the cohort that received genetic testing for FH, four pathogenic variants were found in *LDLR*. No pathogenic variants were found in *APOB* or *PCSK9*.

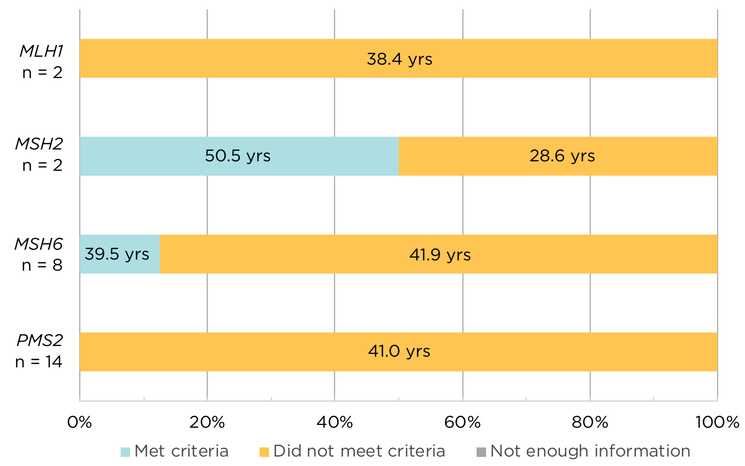
Variant	Gender	Age, range (Years)	LDL-C Level, range (mg/dL)	FH Clinical Diagnosis
LDLR c.718G>A	Female	31-40	250-329	No
LDLR c.1019G>A	Male	31-40	250-329	No
LDLR c.1061A>G	Male	31-40	Information not provided	No
LDLR c.2096C>T	Female	51-65	250-329	No

Figure 3. Genetic testing recommendations for HBOC and LS by NCCN guidelines.

(A) Of the 62 individuals with a pathogenic variant in *BRCA1* or *BRCA2*, 31 (50.0%) would have met criteria for genetic testing for HBOC, 28 (45.2%) would not, and 3 (4.8%) did not provide enough information to determine. The median age (years) of those who met criteria and did not meet criteria is shown.



(B) Of the 26 individuals with a pathogenic variant in *MLH1, MSH2, MSH6*, or *PMS2*, 2 (7.7%) would have met criteria for genetic testing for LS, 24 (92.3%) would not, and 0 (0.0%) did not provide enough information to determine. The median age (years) of those who met criteria and did not meet criteria is shown.



Conclusions

- The demographics of this cohort are in stark contrast to typically studied multi-gene panel cohorts of women, over age 50, with high clinical risk.
- The pathogenic frequency in this cohort (1:75, 1.33%) is double the estimated prevalence from previous studies but closely matches that from the recent Geisinger MyCode cohort (1:78, 1.28%).[6]
- Current guidelines for HBOC and LS were developed in high-risk populations and are therefore not comprehensive enough to identify all carriers in an unselected population.
- Population screening, such as through an optional employee benefit, can identify individuals at an age when screening and preventative measures can be undertaken.