Supplementary Online Content

Samadder NJ, Riegert-Johnson D, Boardman L, et al. Comparison of universal genetic testing vs guideline-directed targeted testing for patients with hereditary cancer syndrome. *JAMA Oncol.* Published online October 30, 2020. doi:10.1001/jamaoncol.2020.6252

This supplementary material has been provided by the authors to give readers additional information about their work.

Appendix 1: Invitae Multi-gene panel list by penetrance category

High Penetrance	Moderate Penetrance
ALK, APC, AXIN2, BAP1, BMPR1A, BRCA1, BRCA2, CDH1, CDK4, CDKN1C, CDKN2A, DICER1, EPCAM, FH (biallelic), FLCN, GATA2, HRAS, MEN1, MET, MLH1, MSH2, MSH6, MUTYH (biallelic), NF2, PALB2, PHOX2B, PMS2, PTCH1, PTEN, RB1, RET, RUNX1, SDHA, SDHB, SDHD, SDHC, SMAD4, SMARCA4, SMARCB1, SMARCE1, STK11, SUFU, TERC, TERT, TP53, TSC1, TSC2, VHL	*AIP, ATM, BLM (biallelic), BRIP1, CASR, CDC73, CDKN1B, CEBPA, CHEK2, GREM1, HOXB13, MAX, MITF, NBN, NF1, POLD1, POLE, POT1, PRKAR1A, RAD51C, RAD51D, RECQL4 (biallelic), SDHAF2, TMEM127, WT1*
Low Penetrance	**Recessive Conditions**
APC (I1307K), BARD1, CTNNA1, EGFR, KIT, MUTYH (monoallelic), PDGFRA, RAD50	*BLM (monoallelic), DIS3L2, FH, MSH3, NTHL1, RECQL4, WRN* **X-linked Conditions** *GPC3*

Appendix 2: Methods for Sequencing and determination of pathogenicity of variants

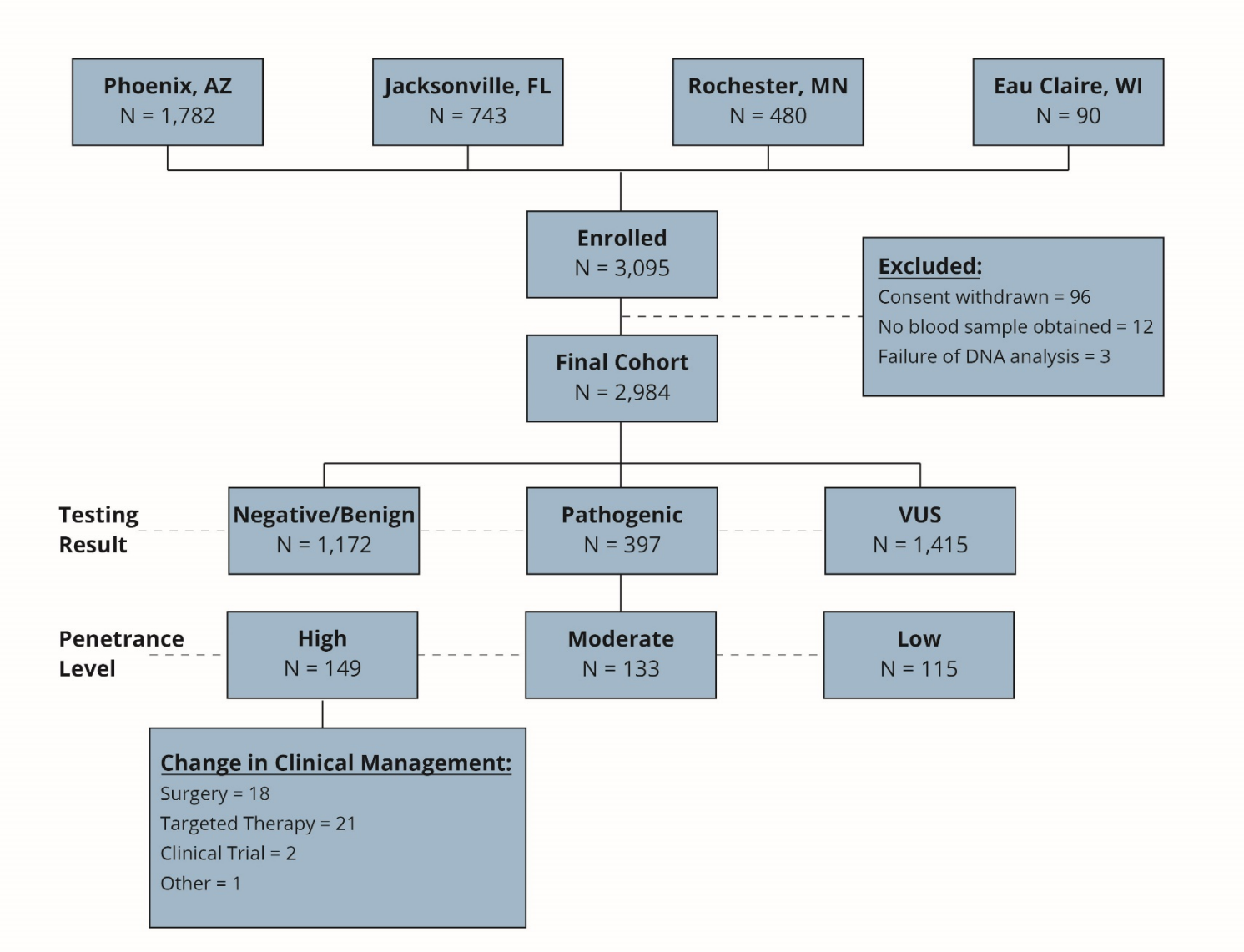
Patients in the study underwent clinical genetic testing at Invitae, a CLIA-certified, NYS-certified and CAP-accredited genetic diagnostics laboratory. Cancer genetic testing at Invitae comprehensively analyzes patients via full-gene sequencing (including coding exons, 10-20 base pairs of adjacent intronic sequencing on either side of the coding exons and select non-coding variants) and deletion-duplication analysis for SNVs, indels, CNVs, and splice site variants. Based on validation study results, this assay achieves >99% analytical sensitivity and specificity for single nucleotide variants, insertions and deletions <15bp in length, and exon-level deletions and duplications. Invitae's methods also detect insertions and deletions larger than 15bp but smaller than a full exon but sensitivity for these may be marginally reduced. Invitae's deletion/duplication analysis determines copy number at a single exon resolution at virtually all targeted exons.

Genomic DNA was extracted from whole blood using a QiaSymphony (Qiagen, Hilden, Germany). Next-generation sequencing and quality control were performed on an Illumina platform as previously described.[1, 2] Reads were aligned to the reference human genome sequence GRCh37 using Novoalign (Novocraft Technologies, Selangor, Malaysia). Small indels and single-nucleotide variants (SNVs) were analyzed using the Genome Analysis Toolkit (GATK). Copy number variant calls were performed using CNVitae.[3] Large structural variants were detected using split-read analysis as described previously.[4]

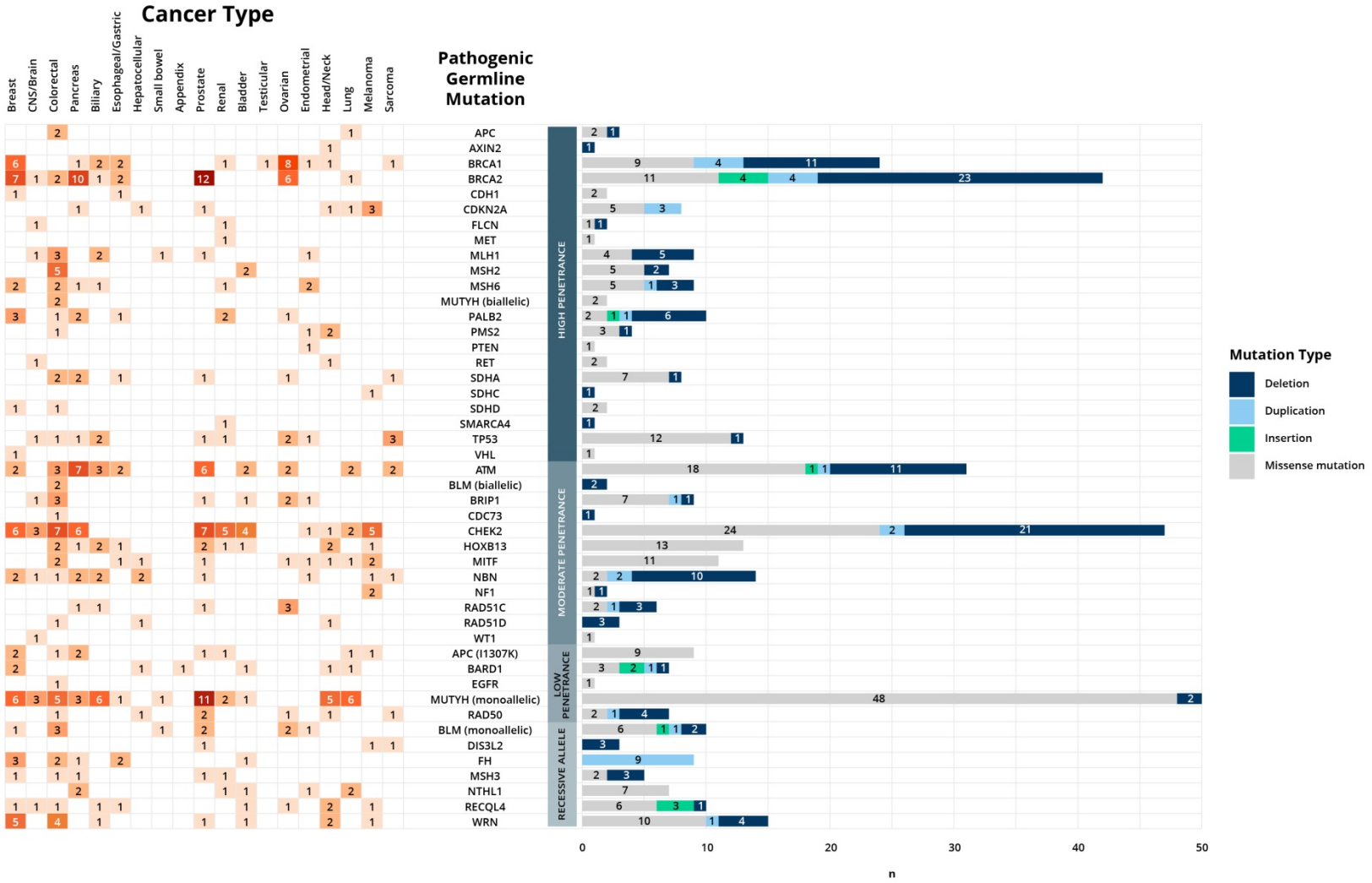
Pathogenicity of candidate variants, including structural variants, was established and they were classified as pathogenic or likely pathogenic if they involved large genomic events or conferred a truncating, initiation codon or splice donor/acceptor effect, if functional data showed an impact on protein function, or if pathogenicity was otherwise reported in published literature.[5] Validation of PGMs observed was performed in accordance with Invitae standard operating practices, wherein orthogonal technology was used to validate pathogenic and likely pathogenic variants via Sanger sequencing or Multiplex Ligation-Dependent Probe Amplification.[6] Confirmed variants were then interrogated using a refinement of American College of Medical Genetics and Genomics (ACMG) criteria (Sherloc).[7]

Genes in which PGMs were identified were organized into high, intermediate and low penetrance categories based on the relative risk (RR) of cancer associated with PGMs in each gene, based on evidence in the literature. Examples include high penetrance genes such as *BRCA1* & *BRCA2*, RR 5-9[8], moderate penetrance genes such as *CHEK2*, RR 3.3[9], low penetrance genes such as *BARD1*, RR <2[10], and recessive genes such as *WRN*.[11] In some cases specific PGMs (e.g. those occurring more frequently in certain populations) were classified individually based on the RR of cancer that has been established for these particular variants, an example of which is *APC* I1307K, RR ~2 in the Ashkenazi Jewish population.[12]

Supplemental Figure 1: Patient flow Diagram



Supplemental Figure 2: Germline mutation type shown by tumor type



eTable 1: Participant characteristics by clinical site

	Arizona	Florida	Rochester	Eau Claire	Total	p value
All	1697	730	467	90	2984	< 0.001[1]
Gender						< 0.001[2]
Male	983 (57.9%)	365 (50.0%)	200 (42.8%)	34 (37.8%)	1582 (53.0%)	
Female	714 (42.1%)	365 (50.0%)	267 (57.2%)	56 (62.2%)	1402 (47.0%)	
Age						< 0.001[3]
Mean (SD)	61.7 (12.6)	61.9 (11.3)	59.1 (12.3)	64.5 (10.3)	61.4 (12.2)	
Median	64.0	63.0	61.0	66.5	64.0	
Range	18.0 - 85.0	20.0 - 81.0	27.0 - 79.0	31.0 - 80.0	18.0 - 85.0	
Ancestry						< 0.001[2]
White	1429 (84.2%)	608 (83.3%)	445 (95.3%)	89 (98.9%)	2571 (86.2%)	
Non-white	268 (15.8%)	122 (16.7%)	22 (4.7%)	1 (1.1%)	413 (13.8%)	
Hispanic/Latino	127 (7.5%)	28 (3.8%)	4 (0.9%)	0 (0.0%)	159 (5.3%)	
Black/African American	35 (2.1%)	69 (9.5%)	6 (1.3%)	0 (0.0%)	110 (3.7%)	
Asian	39 (2.3%)	10 (1.4%)	4 (0.9%)	0 (0.0%)	53 (1.8%)	
American Indian/ Alaskan Native	27 (1.6%)	1 (0.1%)	1 (0.2%)	0 (0.0%)	29 (1.0%)	
Native Hawaiian/Pacific Islander	5 (0.3%)	1 (0.1%)	0 (0.0%)	0 (0.0%)	6 (0.2%)	
Other	35 (2.1%)	13 (1.8%)	7 (1.5%)	1 (1.1%)	56 (1.9%)	
Stage[4]						< 0.001[1]
0/1	273 (16.8%)	147 (20.6%)	92 (21.0%)	23 (25.5%)	535 (18.6%)	
2	248 (15.3%)	152 (21.4%)	68 (15.5%)	9 (10.0%)	477 (16.7%)	
3	310 (19.1%)	153 (21.5%)	116 (26.5%)	14 (15.6%)	593 (20.7%)	
4	792 (48.8%)	259 (36.4%)	162 (37.0%)	44 (48.9%)	1257 (43.9%)	
Missing	3	0	0	0	3	
WHO Grade[5]						0.325[1]
1	1 (1.4%)	1 (5.3%)	0 (0.0%)	2 (1.7%)		

2	17 (23.9%)	3 (15.8%)	4 (13.8%)	24 (20.2%)		
3	13 (18.3%)	4 (21.1%)	11 (37.9%)	28 (23.5%)		
4	40 (56.3%)	11 (57.9%)	14 (48.3%)	65 (54.6%)		
Germline Result						0.026[2]
Pathogenic	210 (12.4%)	123 (16.8%)	57 (12.2%)	7 (7.8%)	397 (13.3%)	
Benign	669 (39.4%)	268 (36.7%)	199 (42.6%)	36 (40.0%)	1172 (39.3%)	
VUS	818 (48.2%)	339 (46.4%)	211 (45.2%)	47 (52.2%)	1415 (47.4%)	
Family History of cancer in a First Degree Relative						< 0.001[2]
Yes	596 (35.1%)	262 (35.9%)	131 (28.1%)	30 (33.3%)	1019 (34.1%)	
No	261 (15.4%)	141 (19.3%)	68 (14.6%)	9 (10.0%)	479 (16.1%)	
Pedigree unavailable	840 (49.5%)	327 (44.8%)	268 (57.4%)	51 (56.7%)	1486 (49.8%)	
Primary Cancer						< 0.001[2]
BREAST	145 (8.5%)	123 (16.8%)	89 (19.1%)	33 (36.7%)	390 (13.1%)	
CNS/BRAIN	71 (4.2%)	19 (2.6%)	29 (6.2%)	0 (0.0%)	119 (4.0%)	
GASTRO-INTESTINAL	542 (31.9%)	208 (28.5%)	183 (39.2%)	23 (25.6%)	956 (32.0%)	
Colorectal	217 (12.8%)	64 (8.8%)	82 (17.6%)	9 (10.0%)	372 (12.5%)	
Pancreas	132 (7.8%)	67 (9.2%)	50 (10.7%)	9 (10.0%)	258 (8.6%)	
Biliary	105 (6.2%)	30 (4.1%)	21 (4.5%)	3 (3.3%)	159 (5.3%)	
Esophageal/Gastric	51 (3.0%)	30 (4.1%)	18 (3.9%)	2 (2.2%)	101 (3.4%)	
Hepatocellular	21 (1.2%)	13 (1.8%)	9 (1.9%)	0 (0.0%)	43 (1.4%)	
Small bowel	7 (0.4%)	3 (0.4%)	2 (0.4%)	0 (0.0%)	12 (0.4%)	
Appendix	7 (0.4%)	1 (0.1%)	1 (0.2%)	0 (0.0%)	9 (0.3%)	
Anal	2 (0.1%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	2 (0.1%)	
GENITO-URINARY	425 (25.0%)	152 (20.8%)	19 (4.1%)	12 (13.3%)	608 (20.4%)	
Prostate	264 (15.6%)	77 (10.5%)	7 (1.5%)	10 (11.1%)	358 (12.0%)	

Renal	87 (5.1%)	43 (5.9%)	5 (1.1%)	2 (2.2%)	137 (4.6%)	
Bladder	68 (4.0%)	32 (4.4%)	6 (1.3%)	0 (0.0%)	106 (3.6%)	
Testicular	5 (0.3%)	0 (0.0%)	1 (0.2%)	0 (0.0%)	6 (0.2%)	
Penis	1 (0.1%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	1 (0.0%)	
GYNECOLOGICAL	107 (6.3%)	73 (10.0%)	38 (8.1%)	8 (8.9%)	226 (7.6%)	
Ovarian	60 (3.5%)	33 (4.5%)	29 (6.2%)	4 (4.4%)	126 (4.2%)	
Endometrial	45 (2.7%)	40 (5.5%)	9 (1.9%)	4 (4.4%)	98 (3.3%)	
Cervical	2 (0.1%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	2 (0.1%)	
HEAD/NECK	119 (7.0%)	59 (8.1%)	16 (3.4%)	6 (6.7%)	200 (6.7%)	
THORAX	77 (4.5%)	31 (4.2%)	7 (1.5%)	5 (5.6%)	120 (4.0%)	
Thymus	1 (0.1%)	0 (0.0%)	1 (0.2%)	0 (0.0%)	2 (0.1%)	
Lung	75 (4.4%)	30 (4.1%)	6 (1.3%)	5 (5.6%)	116 (3.9%)	
Mesothelioma	1 (0.1%)	1 (0.1%)	0 (0.0%)	0 (0.0%)	2 (0.1%)	
SKIN	154 (9.1%)	36 (4.9%)	54 (11.6%)	3 (3.3%)	247 (8.3%)	
Melanoma	153 (9.0%)	35 (4.8%)	54 (11.6%)	3 (3.3%)	245 (8.2%)	
Sebaceous carcinoma	1 (0.1%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	1 (0.0%)	
Squamous cell carcinoma	0 (0.0%)	1 (0.1%)	0 (0.0%)	0 (0.0%)	1 (0.0%)	
NEUROENDOCRINE (NET)	3 (0.2%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	3 (0.1%)	
SARCOMA	54 (3.2%)	29 (4.0%)	32 (6.9%)	0 (0.0%)	115 .9%)	

1. Chi-squared test for given probabilities
2. Pearson's Chi-squared test
3. Kruskal-Wallis rank sum test
4. Stage – all cancer types except CNS/Brain tumors
5. WHO Grade – used for CNS/Brain tumors only

eTable 2: Participant and tumor characteristics by genetic test result

	Pathogenic	Benign	VUS	Total	p value
All	397 (13.3%)	1172 (39.3%)	1415 (47.4%)	2984	< 0.001[2]
Enrollment Site					0.026[1]
Phoenix, Arizona	210 (12.4%)	669 (39.4%)	818 (48.2%)	1697	
Jacksonville, Florida	123 (16.8%)	268 (36.7%)	339 (46.4%)	730	
Rochester, Minnesota	57 (12.2%)	199 (42.6%)	211 (45.2%)	467	
Eau Claire, Wisconsin	7 (7.8%)	36 (40.0%)	47 (52.2%)	90	
Gender					0.669[1]
Male	213 (13.5%)	631 (39.9%)	738 (46.6%)	1582	
Female	184 (13.1%)	541 (38.6%)	677 (48.3%)	1402	
Age					0.086[3]
Mean (SD)	60.0 (13.0)	61.6 (12.0)	61.7 (12.1)	61.4 (12.2)	
Median	62.0	64.0	64.0	64.0	
Range	18.0 - 81.0	20.0 - 82.0	19.0 - 85.0	18.0 - 85.0	
Ancestry					0.008[1]
White	352 (13.7%)	1033 (40.2%)	1186 (46.1%)	2571	
Hispanic/Latino	15 (9.4%)	64 (40.3%)	80 (50.3%)	159	
Black/African American	14 (12.7%)	24 (21.8%)	72 (65.5%)	110	
Asian	8 (15.1%)	19 (35.8%)	26 (49.1%)	53	
American Indian / Alaskan Native	3 (10.3%)	8 (27.6%)	18 (62.1%)	29	
Native Hawaiian / Pacific Islander	0 (0.0%)	1 (16.7%)	5 (83.3%)	6	
Other	5 (8.9%)	23 (41.1%)	28 (50.0%)	56	
Family History of cancer in a First Degree Relatives[5]					0.288[1]
Yes	146 (14.3%)	409 (40.1%)	464 (45.5%)	1019	
No	70 (14.6%)	188 (39.2%)	221 (46.1%)	479	
Stage[4]					0.245[1]
0/1	62 (11.6%)	206 (38.5%)	267 (49.9%)	535	
2	77 (16.1%)	171 (35.8%)	229 (48.0%)	477	
3	84 (14.2%)	234 (39.5%)	275 (46.4%)	593	
4	159 (12.6%)	511 (40.7%)	587 (46.7%)	1257	
Missing	0	0	3	3	
Primary Cancer					0.155[1]
BREAST	47 (12.1%)	160 (41.0%)	183 (46.9%)	390	
CNS/BRAIN	15 (12.6%)	50 (42.0%)	54 (45.4%)	119	
GASTRO-INTESTINAL	147 (15.4%)	365 (38.2%)	444 (46.4%)	956	

Colorectal	57 (15.3%)	144 (38.7%)	171 (46.0%)	372	
Pancreas	41 (15.9%)	103 (39.9%)	114 (44.2%)	258	
Biliary	23 (14.5%)	66 (41.5%)	70 (44.0%)	159	
Esophageal/Gastric	15 (14.9%)	30 (29.7%)	56 (55.4%)	101	
Hepatocellular	7 (16.3%)	14 (32.6%)	22 (51.2%)	43	
Small bowel	3 (25.0%)	3 (25.0%)	6 (50.0%)	12	
Appendix	1 (11.1%)	3 (33.3%)	5 (55.6%)	9	
Anal	0 (0.0%)	2 (100.0%)	0 (0.0%)	2	
GENITO-URINARY	83 (13.7%)	230 (37.8%)	295 (48.5%)	608	
Prostate	49 (13.7%)	147 (41.1%)	162 (45.3%)	358	
Renal	18 (13.1%)	39 (28.5%)	80 (58.4%)	137	
Bladder	15 (14.2%)	44 (41.5%)	47 (44.3%)	106	
Testicular	1 (16.7%)	0 (0.0%)	5 (83.3%)	6	
Penis	0 (0.0%)	0 (0.0%)	1 (100.0%)	1	
GYNECOLOGICAL	39 (17.3%)	81 (35.8%)	106 (46.9%)	226	
Ovarian	26 (20.6%)	49 (38.9%)	51 (40.5%)	126	
Endometrial	13 (13.3%)	31 (31.6%)	54 (55.1%)	98	
Cervical	0 (0.0%)	1 (50.0%)	1 (50.0%)	2	
HEAD/NECK	21 (10.5%)	81 (40.5%)	98 (49.0%)	200	
THORAX	17 (14.2%)	39 (32.5%)	64 (53.3%)	120	
Thymus	0 (0.0%)	2 (100.0%)	0 (0.0%)	2	
Lung	17 (14.7%)	36 (31.0%)	63 (54.3%)	116	
Mesothelioma	0 (0.0%)	1 (50.0%)	1 (50.0%)	2	
SKIN	18 (7.3%)	115 (46.6%)	114 (46.2%)	247	
Melanoma	18 (7.3%)	115 (46.9%)	112 (45.7%)	245	
Sebaceous carcinoma	0 (0.0%)	0 (0.0%)	1 (100.0%)	1	
Squamous cell carcinoma skin	0 (0.0%)	0 (0.0%)	1 (100.0%)	1	
NEUROENDOCRINE (NET)	0 (0.0%)	2 (66.7%)	1 (33.3%)	3	
SARCOMA	10 (8.7%)	49 (42.6%)	56 (48.7%)	115	

1. Pearson's Chi-squared test
2. Chi-squared test for given probabilities
3. Kruskal-Wallis rank sum test
4. Stage – all cancer types except CNS/Brain tumors
5. Limited to patients with a pedigree available (N=1498)

eTable 3: Incidence of findings not predicted by clinical guidelines (incremental findings)

	2018 (N=397)	2020 (N=397)	p value
Did they meet NCCN/NSGC/ACMG testing guidelines?			0.101
Yes	205 (51.6%)	228 (57.4%)	
NCCN	186 (46.8%)	212 (53.4%)	
NSGC/ACMG	147 (37.0%)	148 (37.3%)	
No	192 (48.4%)	169 (42.6%)	

eTable 4: Participant and tumor characteristics by incremental vs non-incremental PGM

	Non-Incremental PGM (N=228)	Incremental PGM (N=169)	Total (N=397)	p value
Enrollment Site				0.530[1]
ARZ	117 (51.3%)	93 (55.0%)	210 (52.9%)	
FLA	69 (30.3%)	54 (32.0%)	123 (31.0%)	
RST	37 (16.2%)	20 (11.8%)	57 (14.4%)	
EU	5 (2.2%)	2 (1.2%)	7 (1.8%)	
Gender				0.012[1]
Male	110 (48.2%)	103 (60.9%)	213 (53.7%)	
Female	118 (51.8%)	66 (39.1%)	184 (46.3%)	
Age				< 0.001[1]
Median (SD)	57.6 (14.0)	63.1 (10.9)	60.0 (13.0)	
Age < 50 Years	68 (29.8%)	17 (10.1%)	85 (21.4%)	
Age ≥ 50 Years	160 (70.2%)	152 (89.9%)	312 (78.6%)	
Ancestry				0.049[1]
White	196 (86.0%)	156 (92.3%)	352 (88.7%)	
Non-white	32 (14.0%)	13 (7.7%)	45 (11.3%)	
Family History of cancer in First Degree Relatives[2]				0.362[1]
Yes	88 (38.6%)	58 (34.3%)	146 (36.8%)	
No	43 (18.9%)	27 (16.0%)	70 (17.6%)	
Lynch MMR Genes				0.004[1]
Yes	24 (10.5%)	5 (3.0%)	29 (7.3%)	
No	204 (89.5%)	164 (97.0%)	368 (92.7%)	
BRCA 1 and 2 Genes				< 0.001[1]
Yes	56 (24.6%)	10 (5.9%)	66 (16.6%)	
No	172 (75.4%)	159 (94.1%)	331 (83.4%)	
Primary Cancer				< 0.001[1]
Breast	38 (16.7%)	9 (5.3%)	47 (11.8%)	
CNS/Brain	3 (1.3%)	12 (7.1%)	15 (3.8%)	
GI	95 (41.7%)	52 (30.8%)	147 (37.0%)	
GU	49 (21.5%)	34 (20.1%)	83 (20.9%)	
GYN	37 (16.2%)	2 (1.2%)	39 (9.8%)	
Head/neck	1 (0.4%)	20 (11.8%)	21 (5.3%)	
Sarcoma	3 (1.3%)	7 (4.1%)	10 (2.5%)	
Skin	2 (0.9%)	16 (9.5%)	18 (4.5%)	
Thorax	0 (0.0%)	17 (10.1%)	17 (4.3%)	
Cancer Stage				0.403[1]

Early Stage (0-2)	78 (34.7%)	61 (38.9%)	139 (36.4%)	
Late Stage (3-4)	147 (65.3%)	96 (61.1%)	243 (63.6%)	
Missing	3	12	15	

1. Pearson's Chi-squared test
2. Limited to patients with a pedigree available (N=1498)

eTable 5: Clinical Implications of those with high penetrance PGM

Patient	Age	Gender	Cancer Type	PGM	Clinical Action	Description
1	36	Female	Breast	*BRCA1*	Surgery	Bilateral nipple-sparing mastectomy plus Bilateral salpingo-oophorectomy
2	54	Male	Prostate	*BRCA2*	Targeted Therapy	Olaparib (PARPi)
3	57	Male	Prostate	*BRCA2*	Clinical Trial	Rucaparib (PARPi) NCT02975934
4	44	Male	Colorectal	*MSH2*	Targeted Therapy	Pembrolizumab
5	67	Female	Ovarian	*BRCA1*	Surgery	Bilateral prophylactic mastectomy
6	32	Female	Breast	*BRCA1*	Surgery	Bilateral mastectomy
7	78	Male	Breast	*BRCA2*	Surgery	Bilateral total mastectomy
8	58	Female	Breast	*BRCA2*	Surgery	Bilateral mastectomy
9	70	Female	Ovarian	*BRCA1*	Targeted Therapy	Olaparib (PARPi)
10	62	Female	Ovarian	*BRCA2, SDHA*	Targeted Therapy	Olaparib (PARPi)
11	44	Female	Ovarian	*BRCA1*	Targeted Therapy	Olaparib
12	54	Female	Ovarian	*BRCA1*	Targeted Therapy	Rucaparib (PARPi)
13	33	Female	Colorectal	*APC*	Surgery	Total colectomy
14	70	Female	Pancreas	*PALB2*	Targeted Therapy	Platinum-based therapy
15	73	Female	Biliary	*BRCA2*	Surgery	Bilateral mastectomy and Bilateral salpingo-oophorectomy plus total hysterectomy
16	66	Female	Biliary	*BRCA1*	Surgery	Bilateral mastectomy
17	44	Female	Endometrial	*BRCA1*	Targeted Therapy	Olaparib (PARPi)
18	40	Female	Colorectal	*MSH6*	Targeted Therapy	Nivolumab + Ipilimumab
19	65	Male	Pancreas	*MSH6*	Targeted Therapy	Pembrolizumab
20	72	Female	Ovarian	*BRCA2*	Surgery	Bilateral mastectomy + Bilateral salpingo-oophorectomy
21	52	Female	Ovarian	*BRCA2*	Targeted Therapy	Olaparib (PARPi)
22	48	Female	Breast	*BRCA2*	Surgery	Bilateral mastectomy + Bilateral salpigo-oophorectomy
23	31	Female	Breast	*BRCA1*	Surgery	Bilateral mastectomy + Bilateral salpingo-oophorectomy
24	52	Female	Breast	*CDH1*	Surgery	Bilateral mastectomy
25	52	Female	Breast	*BRCA2*	Surgery	Bilateral mastectomy + Bilateral salpingo-oophorectomy
26	38	Female	Breast	*BRCA1*	Surgery	Bilateral mastectomy
27	37	Female	Breast	*BRCA2, SDHD*	Surgery	Bilateral mastectomy
28	54	Female	Breast	*BRCA1*	Surgery	Bilateral mastectomy
29	32	Male	Head/ Neck	*RET*	Clinical Trial	RET inhibitor NCT03037385
30	50	Female	Ovarian	*BRCA1*	Targeted Therapy	Olaparib (PARPi)
31	48	Male	Colorectal	*PMS2*	Targeted Therapy	Nivolumab + Ipilimumab
32	66	Male	Prostate	*BRCA2*	Targeted Therapy	Olaparib (PARPi)
33	67	Female	Biliary	*MLH1*	Targeted Therapy	Pembrolizumab

34	44	Female	Breast	*BRCA1*	Surgery	Bilateral mastectomy
35	68	Male	Pancreas	*BRCA2*	Targeted Therapy	Olaparib (PARPi)
36	52	Male	Colorectal	*MSH2*	Targeted Therapy	Pembrolizumab, Nivolumab, Ipilimumab
37	57	Female	Ovarian	*BRCA1*	Surgery	Bilateral mastectomy + Bilateral salpingo-oophorectomy
38	55	Female	Pancreas	*BRCA2*	Targeted Therapy	Rucaparib (PARPi)
39	54	Female	Ovarian	*BRCA2*	Targeted Therapy	Niraparib, Olaparib (PARPi)
40	18	Male	Esophageal/ Gastric	*SDHA*	Other	No adjuvant treatment required
41	53	Female	Ovarian	*BRCA1*	Targeted Therapy	Olaparib (PARPi)
42	72	Female	Ovarian	*PALB2*	Targeted Therapy	Olaparib (PARPi)

eTable 6: Characteristics of participants whose families did or did not undergo cascade family variant testing

	Did not undergo FVT (N=327)	Underwent FVT (N=70)	Total (N=397)	p value
Gender				0.907[1]
Male	175 (53.5%)	38 (54.3%)	213 (53.7%)	
Female	152 (46.5%)	32 (45.7%)	184 (46.3%)	
Age				0.294[2]
Mean (SD)	60.2 (13.1)	58.8 (12.8)	60.0 (13.0)	
Median	62.0	61.0	62.0	
Range	24.0 - 81.0	18.0 - 78.0	18.0 - 81.0	
Ancestry				0.462[1]
White	285 (87.2%)	67 (95.7%)	352 (88.7%)	
Hispanic/Latino	14 (4.3%)	1 (1.4%)	15 (3.8%)	
Black/African American	13 (4.0%)	1 (1.4%)	14 (3.5%)	
Asian	7 (2.1%)	1 (1.4%)	8 (2.0%)	
American Indian / Alaskan Native	3 (0.9%)	0 (0.0%)	3 (0.8%)	
Other	5 (1.5%)	0 (0.0%)	5 (1.3%)	
Cancer Stage				0.102[1]
0/1	46 (14.6%)	16 (23.9%)	62 (16.2%)	
2	60 (19.0%)	17 (25.4%)	77 (20.2%)	
3	73 (23.2%)	11 (16.4%)	84 (22.0%)	
4	136 (43.2%)	23 (34.3%)	159 (41.6%)	
Number of family members tested				
Mean (SD)	NA	2.6 (2.2)	2.6 (2.2)	
Median	NA	2.0	2.0	
Range	NA	1.0 - 14.0	1.0 - 14.0	
BRCA 1 and 2 genes				0.234[1]
Yes	51 (15.6%)	15 (21.4%)	66 (16.6%)	
No	276 (84.4%)	55 (78.6%)	331 (83.4%)	
Lynch MMR genes				0.654[1]
Yes	23 (7.0%)	6 (8.6%)	29 (7.3%)	
No	304 (93.0%)	64 (91.4%)	368 (92.7%)	

1. Pearson's Chi-squared test
2. Kruskal-Wallis rank sum test

eTable 7: Logistic regression ORs and 95% CI of patient and tumor predictors of pathogenic/likely pathogenic mutation

Characteristic	OR	95% CI	P
Age group, y			
≥50	1.0 (ref)		
<50	1.38	1.06-1.78	0.016
Sex			
Male	1.0 (ref)		
Female	0.97	0.79-1.20	0.785
Cancer stage			
Stage 0/1	1.0 (ref)		
Stage 2	1.44	1.00-2.07	0.050
Stage 3	1.23	0.87-1.76	0.244
Stage 4	1.08	0.79-1.49	0.624
Dichotomize Cancer Stage			
Early (stage 0-2)	1.0 (ref)		
Advanced (stage 3-4)	0.95	0.76-1.19	0.652
Family history of cancer in first degree relative			
No	1.0 (ref)		
Yes	0.98	0.72-1.34	0.88
Cancer Type			
Breast	1.0 (ref)	--	--
Colorectal	1.32	0.88-2.0	0.189
Pancreas-biliary	1.38	0.88-2.17	0.164
Prostate	1.16	0.75-1.78	0.504
Melanoma	0.58	0.32-1.00	0.049

eReferences:

1. Beitsch PD, Whitworth PW, Hughes K, et al. Underdiagnosis of Hereditary Breast Cancer: Are Genetic Testing Guidelines a Tool or an Obstacle? J Clin Oncol 2019;37:453-460.
2. Lincoln SE, Kobayashi Y, Anderson MJ, et al. A Systematic Comparison of Traditional and Multigene Panel Testing for Hereditary Breast and Ovarian Cancer Genes in More Than 1000 Patients. J Mol Diagn 2015;17:533-44.
3. Truty R, Paul J, Kennemer M, et al. Prevalence and properties of intragenic copy-number variation in Mendelian disease genes. Genet Med 2019;21:114-123.
4. Nord AS, Lee M, King MC, et al. Accurate and exact CNV identification from targeted high-throughput sequence data. BMC Genomics 2011;12:184.
5. Kurian AW, Hare EE, Mills MA, et al. Clinical evaluation of a multiple-gene sequencing panel for hereditary cancer risk assessment. J Clin Oncol 2014;32:2001-9.
6. Hogervorst FB, Nederlof PM, Gille JJ, et al. Large genomic deletions and duplications in the BRCA1 gene identified by a novel quantitative method. Cancer Res 2003;63:1449-53.
7. Nykamp K, Anderson M, Powers M, et al. Sherloc: a comprehensive refinement of the ACMG-AMP variant classification criteria. Genet Med 2017;19:1105-1117.
8. Ford D, Easton DF, Stratton M, et al. Genetic heterogeneity and penetrance analysis of the BRCA1 and BRCA2 genes in breast cancer families. The Breast Cancer Linkage Consortium. Am J Hum Genet 1998;62:676-89.
9. Cybulski C, Wokolorczyk D, Jakubowska A, et al. Risk of breast cancer in women with a CHEK2 mutation with and without a family history of breast cancer. J Clin Oncol 2011;29:3747-52.
10. Wood ME, McKinnon W, Garber J. Risk for breast cancer and management of unaffected individuals with non-BRCA hereditary breast cancer. Breast J 2020.
11. Lauper JM, Krause A, Vaughan TL, et al. Spectrum and risk of neoplasia in Werner syndrome: a systematic review. PLoS One 2013;8:e59709.
12. Leshno A, Shapira S, Liberman E, et al. The APC I1307K allele conveys a significant increased risk for cancer. Int J Cancer 2016;138:1361-7.