Supplementary Online Content

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

eMethods. Study Population, Whole-Exome Sequencing, Pathogenicity Scoring and In Silico Analysis, and Statistical Analyses

Study Population

Osteosarcoma patients. 1,244 cases were assembled from participating studies described in **Supplemental Table 1**. 782 osteosarcoma cases were previously reported in a genome-wide association study (GWAS)[1,2], including 48 cases from the Instituto de Oncologia Pediátrica GRAACC/UNIFESP and Universidade Federal de Sao Paulo, Brazil[2], and a total of 462 additional cases were included, drawn from the Childhood Cancer Survivor Study (CCSS)[3], USA, the NCI Bone Disease and Injury Study of Osteosarcoma (BDISO)[4], USA, the Hospital Infantil Manuel De Jesus Rivera, Nicaragua, and from the Unidad Nacional de Oncologia Pediatrica (UNOP), Guatemala[5]. 1,004 osteosarcoma cases with WES performed at the NCI were included as a primary discovery set, and 240 additional (non-overlapping) osteosarcoma cases[6] for replication had WES (N=100) or targeted sequencing (N=140) performed at the University of Minnesota. Neither family history nor tumor sequence data were available. 360 cases in the current study were also included in our prior study of *TP53* targeted sequencing and evaluation of P/LP variants[7]; this is noted in the results.

Population controls. 1,062 in-house, cancer-free adult controls were assembled from participating studies in **Supplemental Table 1**. 994 European DCEG control subjects were adults (mean age at study enrollment: 64.6, SD 7.2) drawn from three large studies: the Prostate, Lung, Colon and Ovarian Cancer Prevention Trial (PLCO)[8], the American Cancer Society Cancer Prevention Study-II (CPSII)[9], and the Environment and Genes in Lung cancer Etiology (EAGLE) study[10]. Additionally, 68 controls from the Instituto de Oncologia Pediátrica GRAACC/UNIFESP and Universidade Federal de Sao Paulo, Brazil drawn from the same Brazilian population as the 48 osteosarcoma cases from Brazil.

The in-house, cancer-free adult controls and the discovery set cases had comparable whole-exome sequencing methods, coverage, variant calling, quality control filtering, and ancestry assessment. SNP GWAS microarray or WES data was used to determine the underlying population substructure of the cases and controls based on STRUCTURE and principal components analyses (PCA) with outliers removed. Individuals with >80% European ancestry were considered European (EUR).

All participating subjects provided informed consent under the auspices of local Institutional Review Boards.

Whole-Exome Sequencing

WES was performed on a discovery set of 1,004 confirmed osteosarcoma cases and 1,062 DCEG controls on germline DNA extracted from either leukocytes or buccal samples. WES detailed methods have been previously described.[11-13] In brief, NimbleGen's SeqCap EZ Human Exome Library, Exome+UTR (Roche NimbleGen, Inc., Madison, WI, USA), capture kit was used for all cases and controls, and sequencing was performed on an Illumina HiSeq2500 (with 125bp paired end reads) with the Bioo Nextflex (Perkin Elmer, Inc., Austin, TX) library prep (all DCEG controls and 721 cases) or on an Illumina HiSeq4000 (with 150bp paired end reads) with Kapa

HyperPlus (Roche Sequencing and Life Science, Kapa Biosystems, Wilmington, MA) library prep (285 cases). For all samples, library fragmentation was performed with parameters optimized to obtain insert sizes of 250bp-350bp. The human reference genome and the "known gene" transcript annotation were downloaded from the UCSC database, version hg19. Reads were trimmed and aligned to the hg19 reference genome using the Novoalign software (v3.00.05). High-quality alignments for each individual were created, local realignments refined, and BAM file level recalibrations were done with modules from the Genome Analysis Toolkit (GATK v3.1)[14].

Variant discovery and genotype calling of multi-allelic substitutions, insertions and deletions were performed on all individuals using the UnifiedGenotyper and HaplotypeCaller modules from Genome Analysis Toolkit (GATK v3.1) as well as the FreeBayes variant caller (v9.9.2). The Ensembl variant calling pipeline (bcbio V0.2.2: https://github.com/chapmanb/bcbio.variation/releases/tag/v0.2.2) was used to integrate variant calling results from the above three callers, and all variants were included in the analyses after the Ensembl calling. Insertions and deletions were left-aligned at both post-alignment (BAM) and post-variant-calling (VCF) levels using GATK's LeftAlignIndels and LeftAlignVariants modules, respectively. Annotation and variant dissemination was performed using an in-house custom software annotation pipeline.

Exome analyses were conducted on samples that passed established in house quality control and variant filters[11,12,15]. Poor quality and contaminated samples were excluded, and variants were excluded if they did not pass our pipeline quality control metrics (*e.g.*, CScorefilter), had read depth of <5, heterozygous allele fraction <0.25, and if the minor allele frequency (MAF) was >1% in our population (cases and controls combined) or in any population within 1,000 Genomes Project[16], NHLBI ESP[17] or ExAC[18].

The average WES coverage was >15 reads in 99.8% of the cases and 99.7% of the controls, with median coverage of 53X and 52X in cases and controls, respectively (**Supplemental Figure 1**).

WES and targeted sequencing of 240 replication set cases. WES of 100 osteosarcoma cases (replication set 1) and targeted sequencing of the 238 cancer-susceptibility genes for 140 additional osteosarcoma cases (replication set 2) was conducted on germline DNA extracted from buccal samples by standard methods at the University of Minnesota. WES Libraries were created using the Agilent SureSelect All Exon V5+UTRs kit and were sequenced by the University of Minnesota Genomics Center (UMGC) using a HiSeq2000 that generated 100bp paired end reads with an average on-target insert size of 188.7bp. We implemented the best practices as delineated in the Genome Analysis Toolkit (GATK) pipeline[19], including using BWA-MEM[20] for alignment, GATK for quality recalibration and indel realignment, and GATK HaplotypeCaller for genotyping. On average, 48.3 million reads were delivered per sample, with 35.0 million on target. The average read depth was 46.7x across the 100 case samples.

Genes for targeted sequencing were selected based on genes identified in the 100 osteosarcoma cases (noted above, replication set 1) with a P/LP variant, plus genes identified as cancer-predisposing genes based on previous reports[21,22] and genes reported in COSMIC[23] with germline effects; the 238 cancer-susceptibility genes assessed in the discovery set were targeted. Targeted sequencing was conducted at UMGC using a HiSeq2500 that generated 125bp paired

end reads with an average on-target insert size of 195.4bp. Best practices were implemented as delineated in the GATK pipeline[19,20]. On average, 6.8 million reads were delivered per sample, with 4.4 million on target. The average read depth was 206.6x across 140 case samples.

Pathogenicity Scoring and In Silico Analysis

Rare variants passing quality control and variant filters were evaluated for pathogenicity. A step-wise pipeline was constructed to evaluate each rare variant identified above in the genes of interest. Variants were classified as "Pathogenic" (P), "Likely Pathogenic" (LP), "Variant of Uncertain Significance" (VUS), "Likely Benign" (LB), or "Benign". The classification of variants was based on previous reports[22,24] and on the guidelines recommended by the American College of Medical Genetics and Genomics and the Association for Molecular Pathology[25] and adapted as follows: Step 1: variants matching a known variant annotated by a badged laboratory in ClinVar[26] were categorized as the designated pathogenicity category given by the badged ClinVar laboratory (P, LP, VUS, LB or B). The ClinVar badged laboratory classification was based on clinical laboratories meeting minimum requirements for data sharing to support quality assurance by ClinGen (https://www.clinicalgenome.org/lablist/); the variant scores from laboratories that were not-badged were disregarded using the archive database downloaded on 5/20/2018[26]. At least one score was needed to classify variants, and a majority rule was applied. Step 2: variants not included in ClinVar were evaluated using InterVar version 2.1.2 (default settings)[27], and categorized by its designated pathogenicity category (P, LP, VUS, LB or B). Step 3: variants of uncertain significance, as determined by either ClinVar or InterVar, that were a high impact variant (frameshift indels, stop gain/loss, or known splice sites) and classified as a disease-causing mutation (DM) by HGMD[28] (2018.1) were categorized as LP. Variants classified as a high impact variant or HGMD DM in established cancer predisposition syndrome genes[29] were manually reviewed based on the published literature to determine/confirm pathogenicity and categorized as LP or VUS. Step 4: all P and LP variants were further filtered by population frequency (public database populations[16-18] and our total case/control frequency), for AR genes the variant allele frequency had to be ≤ 0.005 and ≤ 0.001 for non-AR genes otherwise the variants were down-graded to VUS. The VUS category was further divided into *in silico* predicted damaging (VUS_D) or not-damaging (VUS_ND), described below (see *in silico* prediction algorithm). The final step was a manual review of all P and LP designated variants, and review of the high impact and HGMD DM variants for a final designation of P, LP, or VUS_D. Manual review included review of literature for confirmation of pathogenicity, gene specific database review, confirmation of the phenotype within the spectrum of the associated syndrome or cancer, evaluation of the mutation impact on gene function and the mechanism of action associated with that gene. **Supplemental Table 4** summarizes the cancer-susceptibility gene variant classification schema.

Detailed manual review of the P/LP variants, high impact and HGMD DM variants, as described, was performed for the 1,004 discovery set cases and 1,062 DCEG controls. For both the 240 replication set cases and the 27,173 ExAC NFE resource, Steps 1-4 were identical with an equivalent minimal manual review at the final step of the classification schema (**Supplemental Table 4**). It is not possible to determine whether individuals in ExAC carry more than one variant of interest due to the lack of individual level data. This could result in an overestimation of MAFs in this dataset.

An *in silico* prediction algorithm was used to further filter the VUS category for all of the cancer-susceptibility genes, and for the candidate genes, in order to categorize VUS variants as 'damaging' or 'not damaging'. Three programs were used to assign variants as *in silico* deleterious: if MetaSVM[30] (predicted Damaging), REVEL[31] (score ≥0.5) and CADD[32] (score ≥20) predicted a variant deleterious, the variant was categorized as "VUS_D", or otherwise as "VUS_ND". For candidate and somatic genes, variants were further classified as 'VUS_D' if they were a high impact variant (frameshift indels, stop gain/loss, or known splice sites) or pathogenic or likely pathogenic (P/LP) by ClinVar[26]. The variant population frequency for VUS_D variants had to be ≤0.005 in public database populations[16-18] and in our total population (cases and controls), otherwise variants were down-graded to VUS_ND.

Variant calls for all P/LP indels and approximately 50% of SNVs were manually reviewed by an experienced reviewer, and 3.6% of the indels were excluded due to potentially being a false positive based on the following criteria. The sequencing reads (BAM files) in the genomic regions surrounding the variant calls were reviewed using the Integrative Genomics Viewer (IGV) to exclude sequencing and analysis artifacts, and thus false positive findings, following the recommendations/criteria outlined in Robinson et al. 2017[33] and in the IGV user guide (http://www.broadinstitute.org/igv). Briefly, visual inspection for potential errors in indel or SNV calls was performed by manually reviewing the specific indel/variant aligned reads for: (1) highlighted mismatched bases in individual reads; (2) highlighted ambiguously mapped reads (mapping quality = 0); (3) shaded bases with low read base quality (shaded when quality ≤20); (4) forward and reverse strands for strand bias; and (5) the alignment, sequencing, and platform meta-data.

Statistical Analyses

We compared the 1,004 osteosarcoma cases (discovery set) with the 1,062 DCEG controls (994 controls of EUR ancestry and 67 controls drawn from the same Brazilian population as the studied included 48 osteosarcoma cases from Brazil). For replication, we evaluated 240 osteosarcoma cases with germline WES (replication set 1) or targeted sequencing (replication set 2) data from the University of Minnesota, and we compared the total of the cases to the public resource of ExAC NFE[18].

Rare variant burden tests were conducted between the EUR cases (N=732) and EUR DCEG controls (N=994) using the burden and SKAT-O tests[34] . To adjust for multiple comparisons, a Bonferroni significance threshold of 0.0002 was used for 238 cancer-susceptibility gene tests, and *P*-values that remained significant based on this threshold were noted. Burden tests were performed in three ways: (1) comparing the burden of only the P/LP cancer-susceptibility gene variants (termed "pathogenic variant burden"), (2) comparing the burden of all potentially deleterious variants (including P, LP and VUS_D variants and termed "deleterious variant burden"), (3) comparing the burden of all rare variants, applying a MAF threshold of 0.01 as the definition of rare variation (no pathogenicity criteria, termed "rare variant burden"). Comparisons among cases with and without P/LP cancer-susceptibility gene variants were performed using Chi-squared (χ^2) or Fisher's exact tests for categorical variables, and Mann-Whitney U (MWU) tests for continuous variables (i.e., age). A subregion-based burden test (REBET)[35] was used to determine the *TP53* protein/functional domain that was significantly

enriched for P/LP variants in the cases compared to the controls, while adjusting for multiple comparisons. Lollipop plots of P/LP variants by gene with publicly available somatic mutation pediatric data were visualized in the ProteinPaint genome browser[36].

The ExAC NFE case-control comparisons were only performed for the genes that were identified as significantly different between the primary discovery set cases and DCEG controls that had comparable WES at NCI. Carrier frequencies from ExAC NFE were only used as secondary comparisons because the methods are not directly comparable to our cases due to potential differences in both the sequencing methods (e.g., capture kit, sequencing chemistries) and bioinformatic analyses (*e.g.,* variants in ExAC were only called using HaplotypeCaller and utilized different quality control filters). Exact binomial tests were used to compare the frequencies of variants in select cancer-susceptibility genes in the cases versus those in the ExAC NFE resource; burden tests could not be performed for ExAC comparisons because ExAC does not provide individual level data. We used logistic regression to assess associations between case-control status and the presence of P/LP cancer-susceptibility gene variants. All statistical tests were two-sided and performed with R version 3.3.2 and SPSS version 23.

We conducted a time-to-event analysis in 407 osteosarcoma cases from the discovery set with survival data to investigate the effect of carrying P/LP variants on overall survival. The overall survival time was calculated as the time from the date of osteosarcoma diagnosis until the date of death for those deceased or the last date known to be alive; patients were censored at the last date known to be alive or when lost to follow-up. Cause of death was not available for all cases. We compared overall survival for cases carrying P/LP variants to cases without P/LP variants for all cancer-susceptibility genes, and for *TP53*, using Cox proportional hazards regression and estimated hazard ratios (HR) and 95% confidence intervals (CI). Cox models were adjusted for age at diagnosis, gender, and tumor location (i.e., axial vs. extremity location).

We conducted a pathway enrichment analysis for the 101 cancer-susceptibility genes with one or more pathogenic or likely pathogenic variant identified in the discovery set of 1,004 osteosarcoma cases using algorithms from the webtools KOBAS 3.0[37] (http://kobas.cbi.pku.edu.cn) and PathDIP[38] (http://ophid.utoronto.ca/pathDIP/). Both algorithms used a hypergeometric approach to test pathways that are over represented given a gene set. For this analysis we used: (1) as input, all genes that have at least one P/LP variant in the cases; (2) as background, *Homo sapien* genes provided by each algorithm; and, (3) as pathway databases, KEGG and Reactome. For the network analysis, protein-protein interactions were retrieved from the Integrated Interactions Database (IID)[39] and visualized in Network Analysis, Visualization, & Graphing TORonto (Navigator)[40].

eTable 1. Description of Participating Studies

Study Name	Country of Origin	EUR†	AFR†	ADM†	ASN†	HIS†	Brazil	Total	Age at dx, Mean (SD)	N Male, %	
Cases, discovery set											
NCI Retrospective Study of Genetic Risk Factors for Osteosarcoma, Children's Oncology Group (COG AOST08B1)	USA	182	50	33	0	40	0	305	13.2 (3.6)	166	54.4%
Clínica Universidad de Navarra, Pamplona	Spain	73	0	1	0	0	0	74	14.8 (5.5)	40	54.1%
Royal National Orthopaedic Hospital NHS Trust and University College London Cancer Institute	UK	46	1	5	1	0	0	53	252 (17.8)	34	64.2%
Istituto Ortopedico Rizzoli	Italy	40	0	0	0	0	0	40	23.2 (14.6)	27	67.5%
Ankara Oncology and Education Research Hospital	Turkey	0	0	22	0	0	0	22	11.2 (4.1)	11	50.0%
Instituto de Oncologia Pediátrica GRAACC/UNIFESP and Universidade Federal de Sao Paulo	Brazil	0	0	0	0	0	48	48	18.1 (5.3)	31	64.6%
Childhood Cancer Survivor Study (CCSS)	USA	284	0	0	0	0	0	284	14.5 (3.4)	145	51.4%
NCI Bone Disease and Injury Study of Osteosarcoma (BDISO)	USA	107	3	12	2	0	0	124	25.8 (16.0)	70	56.5%
Unidad Nacional de Oncologia Pediatrica (UNOP)	Guatemala	0	0	0	0	54	0	54	12.9 (3.8)	16	29.6%

Total		732	54	73	3	94	48	1004	16.5 (9.5)	540	53.9%
Cases, replication 1: WES data from University of Minnesota											
Genetic Epidemiology of Osteosarcoma study, Children's Oncology Group (COG AEPI05N2)	USA	87	3	2	1	7	0	100	13.2 (3.3)	57	57.0%
Cases, replication 2: Targeted sequencing from University of Minnesota											
Genetic Epidemiology of Osteosarcoma study, Children's Oncology Group (COG AEPI05N2)	USA	124	5	3	0	8	0	140	14.1 (2.9)	79	56.4%
Total Cases		**943**	**62**	**78**	**4**	**109**	**48**	**1244**	**16 (8.9)**	**684**	**55.0%**
Controls											
Prostate, Lung, Colon and Ovarian Cancer Prevention Trial (PLCO)	USA	375	0	0	0	0	0	375	63.8 (5.7)	218	58%
American Cancer Society Cancer Prevention Study-II (CPSII)	USA	223	0	0	0	0	0	223	62.8 (6.1)	108	48%
Environment And Genes in Lung cancer Etiology (EAGLE)	Italy	396	0	0	0	0	0	396	66.3 (8.5)	317	80%
Instituto de Oncologia Pediátrica GRAACC/UNIFESP and Universidade Federal de Sao Paulo	Brazil	0	0	0	0	0	68	68	—	37	54%
Total DCEG Controls		**994**	**0**	**0**	**0**	**0**	**68**	**1062**	**64.6 (7.2)**	**680**	**64%**

†Ancestry based on GWAS data; EUR, European ancestry; AFR, African ancestry; ADM, admixed ancestry; ASN, Asian ancestry; HIS, Hispanic ancestry; Brazil, cases/controls from Brazil.
Age data was not available for the cancer-free controls from Brazil.

eTable 2. Description of 238 Cancer-Susceptibility Genes Evaluated

Syndromes known to be associated with the occurrence of osteosarcoma are shaded.

Gene	Cancer syndrome(s)	Major associated tumor type(s)	Mode of inheritance	Mechanism of action
ABCB11	Progressive familial intrahepatic cholestasis	Hepatocellular carcinoma, Cholangiocarcinoma	AR	loss-of-function
ACD	Dyskeratosis congenita	MDS, AML, head/neck squamous cell cancer, anogenital adenocarcinoma	AD/AR	loss-of-function
AGL	Glycogen storage disease type III	hepatocellular carcinoma	AR	loss-of-function
AIP	Familial isolated pituitary adenoma	Pituitary adenoma	AD	loss-of-function
ALK		Neuroblastoma	AD	gain-of-function
ANKRD26	Thrombocytopenia 2	MDS	AD	
APC	Familial adenomatous polyposis (FAP)	Colorectal cancer, Hepatoblastoma, Desmoid tumor	AD	loss-of-function
APOBEC3B		Breast Cancer	AR	
AR	Reifenstein syndrome; Androgen insensitivity syndrome	prostate	X-linked, recessive	gain-of-function
ASXL1	Bohring-Opiz Syndrome (germline)	blood-forming cells (leukemias), such as acute myeloid leukemia, chronic myelomonocytic leukemia, and myelodysplastic syndrome	unk or de novo	gain-of-function
ATG2B		MDS	AD	gain-of-function
ATM	Ataxia-Telangiectasia (biallelic mutations)	Biallelic mutations: Lymphoid hematological malignancy (leukemia, lymphoma). Monoallelic mutations: Breast cancer.	AD/AR	loss-of-function
ATR	Cutaneous telangiectasia and cancer syndrome, familial	oropharyngeal cancer	AD	loss-of-function
AXIN2	oligodentia-colorectal cancer syndrome	Colorectal cancer	AD	loss-of-function

BAP1		Melanoma (cutaneous, uveal), Mesothelioma, Meningioma, Lung cancer (adenocarcinoma)	AD	loss-of-function
BARD1		Breast cancer	AD	loss-of-function
BLM	Bloom syndrome	Lymphoma and ALL hematological malignancy, Myeloid hematological malignancy, Squamous cell carcinoma, Gastric, Colorectal cancers, Osteosarcoma	AR	loss-of-function
BMPR1A	Juvenile polyposis syndrome	Colorectal cancer, gastric cancer, hamartoma	AD	loss-of-function
BRAF	Noonan syndrome	Cardiofaciocutaneous syndrome	AD	gain-of-function
BRCA1	Hereditary breast-ovarian cancer	Breast cancer, Ovarian cancer	AD	loss-of-function
BRCA2	Hereditary breast-ovarian cancer Fanconi anemia (D1) (biallelic mutations)	Biallelic mutations: Myeloid hematological malignancy (Medulloblastoma, Wilms tumor). Monoallelic mutations: Breast cancer, Ovarian cancer, Prostate cancer, Pancreas cancer.	AD/AR	loss-of-function
BRIP1	Fanconi anemia (J) (biallelic mutations)	Biallelic mutations: Myeloid hematological malignancy, Squamous cell carcinoma. Monoallelic mutations: Breast cancer, Ovarian cancer.	AD/AR	loss-of-function
BUB1B	Mosaic variegated aneuploidy Syndrome	Wilms Tumor, Rhabdomyosarcoma, Myeloid hematological malignancy	AR	loss-of-function
CBL	Noonan syndrome	JMML	AD	loss-of-function
CDC73	Hyperparathyroidism-jaw tumor syndrome	Parathyroid cancer, Ossifying fibroma (bone)	AD	loss-of-function
CDH1	Hereditary diffuse gastric cancer	Breast cancer (lobular), Gastric cancer (diffuse)	AD	loss-of-function
CDK4		Melanoma	AD	gain-of-function
CDKN1B		Thyroid cancer, Pituitary adenoma	AD/AR	loss-of-function
CDKN1C	Beckwith-Wiedemann Syndrome	Embryonal tumors	AD	loss-of-function

CDKN2A		Melanoma [p16 and p14ARF], Pancreas cancer [p16], Astrocytoma [p14ARF]	AD	loss-of-function
CDKN2B		renal cell carcinoma	AD	loss-of-function
CEBPA		Myeloid hematological malignancy	AD	loss-of-function
CEP57	Mosaic Variegated Aneuploidy Syndrome		AR	loss-of-function
CHEK2		Breast cancer	AD	loss-of-function
COL7A1	Epidermolysis bullosa	Squamous cell carcinoma (skin)	AD/AR	loss-of-function
CREBBP	Rubinstein-Taybi Syndrome	AML, ovarian cancer, MBL	AD	
CTC1	dyskeratosis congenita	acute myeloid leukemia, Squamous cell carcinoma (head + neck, anorectal)	AR	loss-of-function
CTR9			AD	
CXCR4			AD	
CYLD	Brooke-Spiegler syndrome	Cylindroma, spiroadenocarcinoma, Basal cell carcinoma	AD	loss-of-function
DDB2	Xeroderma Pigmentosum (E)	Basal cell carcinoma, Squamous cell carcinoma, Melanoma	AD	loss-of-function
DDX41		MDS/AML	AD	loss-of-function
DHCR7	Smith-Lemli-Opitz Syndrome		AR	
DICER1	DICER1 syndrome	Pleuropulmonary blastoma, Cystic nephroma, Ovarian sex cord tumor	AD	loss-of-function
DIS3L2	Perlman syndrome	Wilms tumor	AR	loss-of-function
DKC1	Dyskeratosis congenita	acute myeloid leukemia, Squamous cell carcinoma (head + neck, anorectal)	X-linked, recessive	loss-of-function
DOCK8	HyperIgE syndrome	Squamous cell carcinoma, Lymphoma	AR	loss-of-function
EGFR		Non-small cell lung cancer	AD	gain-of-function
ELANE	Severe congenital neutropenia	Leukemia	AD	loss-of-function
EP300	Rubstein-taybi syndrome	Medulloblastoma	AD	Case Report
EPCAM	Lynch Syndrome; Hereditary Non-Polyposis Colon Cancer (HNPCC); Muir-Torre Syndrome	Colorectal cancer, Endometrial cancer, Ovarian cancer	AD	partial gene deletion

ERBB4		melanoma, gastric, NSCLC	unk or de novo	
ERCC1	Xeroderma Pigmentosa		AR	
ERCC2	Xeroderma pigmentosum (D)	Basal cell carcinoma, Squamous cell carcinoma, Melanoma	AR	loss-of-function
ERCC3	Xeroderma pigmentosum (B)	Basal cell carcinoma, Squamous cell carcinoma, Melanoma	AR	loss-of-function
ERCC4	Xeroderma pigmentosum (F) Fanconi anemia (Q)	Basal cell carcinoma, Squamous cell carcinoma, Melanoma	AR	loss-of-function
ERCC5	Xeroderma pigmentosum (G)	Basal cell carcinoma, Squamous cell carcinoma, Melanoma	AR	loss-of-function
ETV6		ALL	AD	
EXT1		Chondrosarcoma	AD	loss-of-function
EXT2		Chondrosarcoma	AD	loss-of-function
EZH2	Weaver Syndrome	Lymphoid hematological malignancies (lymphoma, lymphoblastic leukemia), neuroblastoma	AD	loss-of-function
FAH	Tyrosinemia	Hepatocellular carcinoma	AR	loss-of-function
FANCA	Fanconi anemia (A)	Myeloid hematological malignancy, Squamous cell carcinoma (head and neck, esophagus, genital tract)	AR	loss-of-function
FANCB	Fanconi Anemia (B)	Myeloid hematological malignancy (leukemia, myelodysplastic syndrome); Squamous cell carcinoma (head and neck, esophagus, genital tract)	X-linked	loss-of-function
FANCC	Fanconi anemia (C)	Myeloid hematological malignancy, Squamous cell carcinoma (head and neck, esophagus, genital tract)	AR	loss-of-function

FANCD2	Fanconi Anemia (D2)	Myeloid hematological malignancy, Squamous cell carcinoma (head and neck, esophagus, genital tract)	AR	loss-of-function
FANCE	Fanconi Anemia (E)	Myeloid hematological malignancy, Squamous cell carcinoma (head and neck, esophagus, genital tract)	AR	loss-of-function
FANCF	Fanconi Anemia (F)	Myeloid hematological malignancy, Squamous cell carcinoma (head and neck, esophagus, genital tract)	AR	loss-of-function
FANCG	Fanconi anemia (G)	Myeloid hematological malignancy, Squamous cell carcinoma (head and neck, esophagus, genital tract)	AR	loss-of-function
FANCI	Fanconi Anemia (I)	Myeloid hematological malignancy, Squamous cell carcinoma (head and neck, esophagus, genital tract)	AR	loss-of-function
FANCL	Fanconi Anemia (L)	Myeloid hematological malignancy, Squamous cell carcinoma (head and neck, esophagus, genital tract)	AR	loss-of-function
FANCM	Fanconi Anemia (M)	monoallelic: breast cancer	AD/AR	loss-of-function
FAS (aka TNFRSF6)	Autoimmune lymphoproliferative syndrome	Lymphoma	AD	loss-of-function
FAT1		oral squamous cell, chemorefractory CLL, head and neck, pancreatic acinar cell carcinoma	unk or de novo	Limited Data
FGFR2	albert, crouzon, pfeiffer syndromes (craniosynostosis)	somatic mutations	AD	loss-of-function
FGFR3	albert, crouzon, pfeiffer syndromes (craniosynostosis)	somatic mutations	AD	loss-of-function
FH	Hereditary leiomyomatosis and renal cell cancer (HLRCC)	Renal cell cancer, Leiomyosarcoma (uterus)	AD/AR	loss-of-function
FLCN	Birt-Hogg-Dube syndrome	Renal cell cancer, Oncocytoma	AD	loss-of-function

FMR1			unk or de novo	
FOXE1		Thyroid Carcinoma	AD	Case Report
G6PC	Glycogen Storage Disease type Ia (GSDIa)	Hepatocellular carcinoma	AR	
GALNT12		Colon	AD	loss-of-function
GALNT14		Neuroblastoma	AD	
GATA1	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	X-linked, recessive	loss-of-function
GATA2	Emberger MonoMAC syndrome; DBA	Myeloid hematological malignancy, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
GBA	Gauchers type 1	Myeloma, Lymphoma, Hepatocellular carcinoma	AR	loss-of-function
GJB2	Keratosis-icthyosis-deafness syndrome (KID)	Squamous cell carcinoma	AD	loss-of-function
GLI3	Pallister-Hall Syndrome	hypothalamic hamartoma	AD	loss-of-function
GNAS			unk or de novo	
GPC3	Simpson-Golabi-Behmel syndrome	Wilms tumor, Hepatoblastoma, hepatocellular carcinoma, Neuroblastoma, Gonadoblastoma	X-linked, recessive	loss-of-function
GPC4	Simpson-Golabi-Behmel syndrome	Wilms tumor, Hepatoblastoma, hepatocellular carcinoma, Neuroblastoma, Gonadoblastoma	X-linked, recessive	
GREM1		Colon	AD	gain-of-function
GSKIP		AML	unk or de novo	Duplication
HABP2		thyroid cancer, nonmedullary	AD	
HFE	Haemochromatosis	Hepatocellular carcinoma, Cholangiocarcinoma	AR	loss-of-function
HMBS	Porphyria (AI)	hepatocellular carcinoma	AD	loss-of-function

HNF1A	Maturity-Onset Diabetes of The Young, Type 3 (MODY3)	Hepatic adenoma	AD	loss-of-function
HOXB13		Prostate cancer	AD	loss-of-function/gain-off-function
HRAS	Costello syndrome	Rhabdomyosarcoma, Neuroblastoma, Transitional cell carcinoma (bladder)	AD	gain-of-function
IDH1	Ollier Disease	AML, glioblastoma	unk or de novo	
IDH2	Ollier Disease	AML, glioblastoma	unk or de novo	
IKZF1		ALL	unk or de novo	
IPMK	inherited neuroendocrine tumor of small intestine	neuroendocrine tumor small intestine	AD	
ITK	Lymphoproliferative syndrome 1	Hodgkin's lymphoma	AR	loss-of-function
KIT		Gastro-Intestinal Stromal Tumor	AD	gain-of-function
KMT2D	Kabuki syndrome	hepatoblastoma	AD	
KRAS	Noonan syndrome	JMML, neuroblastoma	AD	gain-of-function
L2HGDH	L-2-hydroxyglutaric aciduria	brain tumor (medulloblastoma, high grade glioma)	AR	loss-of-function
LMO1		neuroblastoma	AD	gain-of-function
LZTR1		schwannoma	AD	
MAP2K1	mitogen-activated protein kinase kinase 1	Cardiofaciocutaneous syndrome	AD	
MAP2K2	mitogen-activated protein kinase kinase 2	Cardiofaciocutaneous syndrome	AD	
MAX	Familial paraganglioma-pheochromocytoma syndrome	Paraganglioma, Pheochromocytoma	AD	loss-of-function
MC1R		melanoma	AD	
MEN1	Multiple endocrine neoplasia Type 1	Parathyroid, pituitary adenoma, Neuroendocrine tumor, Carcinoid tumor, Adrenocortical carcinoma	AD	loss-of-function
MET		Renal cell cancer (papillary carcinoma)	AD	gain-of-function
MITF		melanoma, renal cell carcinoma	AD	gain-of-function

MLH1	MMR deficiency syndrome (biallelic mutations); Lynch syndrome / Hereditary Non-Polyposis Colon Cancer (monoallelic mutations)	Biallelic mutations: Brain tumors, Hematological malignancy, Embryonal tumors. Monoallelic mutations: Colorectal cancer, Endometrial cancer, Ovarian cancer.	AD/AR	loss-of-function
MPL	Congenital Amegakaryocytic Thrombocytopenia	AML, MDS	AD/AR	
MRE11A			unk or de novo	
MSH2	MMR deficiency syndrome (biallelic mutations) Lynch syndrome / Hereditary Non-Polyposis Colon Cancer (monoallelic mutations)	Biallelic mutations: Brain tumors, Hematological malignancy, Embryonal tumors. Monoallelic mutations: Colorectal cancer, Endometrial cancer, Ovarian cancer, Sebaceous adenoma, carcinoma, epithelioma.	AD/AR	loss-of-function
MSH3		colorectal carcinoma	AR	
MSH6	MMR deficiency syndrome (biallelic mutations) Lynch syndrome / Hereditary Non-Polyposis Colon Cancer (monoallelic mutations).	Biallelic mutations: Brain tumors, Hematological malignancy, Embryonal tumors. Monoallelic mutations: Colorectal cancer, Endometrial cancer, Ovarian cancer.	AD/AR	loss-of-function
MTAP	Diaphyseal medullary stenosis with malignant fibrous histiocytoma (DMS-MFH)	malignant fibrous histiocytoma (sarcoma)	AD	loss-of-function
MUTYH		Colorectal cancer	AR	loss-of-function
NBN	Nijmegen breakage syndrome	Lymphoma, Medulloblastoma, Glioma, Rhabdomyosarcoma	AR	loss-of-function
NF1	Neurofibromatosis type 1	Glioma, Malignant peripheral nerve sheath tumor	AD	loss-of-function
NF2	Neurofibromatosis type 2	Vestibular schwannoma, Meningioma, Ependymoma	AD	loss-of-function

NFIX			unk or de novo	
NHP2	Dyskeratosis Congenita	acute myeloid leukemia, Squamous cell carcinoma (head + neck, anorectal), Melanoma	AD/AR	
NKX2-1		Thyroid carcinoma	AD	
NOTCH3	fibroblastic tumors involving skin, striated muscles, bones, and viscera	Infantile myofibromatosis	AD	
NRAS	Noonan syndrome	leukemia - JMML	AD	
NSD1	Sotos Syndrome	Neuroblastoma, Presacral ganglioma, Sacrococcygeal teratoma, AML	AD	loss-of-function
NTHL1	Familial adenomatous polyposis 3	Colorectal	AD	
NTRK1		spitzoid tumor, papillary thyroid	AR	
PALB2	Fanconi anemia (N) (biallelic mutations)	Biallelic mutations: Myeloid hematological malignancy, Medulloblastoma, Neuroblastoma, Wilms tumor.Monoallelic mutations: Breast cancer, Pancreas cancer.	AD/AR	loss-of-function
PARN	Dyskeratosis congenita	acute myeloid leukemia, Squamous cell carcinoma (head + neck, anorectal), Melanoma	AD/AR	
PAX5	Familial Clustering of Acute Lymphoblastic Leukemia	ALL	AD	
PAX6	WAGR	Wilms	AD	loss-of-function
PBRM1		renal cell carcinoma	AD	
PDGFRA		Gastro-Intestinal Stromal Tumor	AD	gain-of-function
PDGFRB		Infantile myofibromatosis	AD	
PHF6	Borjeson-Forssman-Lehmann syndrome	t-cell ALL	X-linked, recessive	
PHOX2B		Neuroblastoma	AD	loss-of-function
PMS1	lynch syndrome	colon cancer	AD	

PMS2	MMR deficiency syndrome (biallelic mutations) Lynch syndrome / Hereditary Non-Polyposis Colon Cancer (monoallelic mutations)	Biallelic mutations: Brain tumors, Hematological malignancy, Supratentorial primitive, neuroectodermal tumors. Monoallelic mutations: Colorectal cancer, Endometrial cancer, Ovarian cancer.	AD/AR	loss-of-function
POLD1	PPAP (polymerase proofreading associated polyposis)	Colorectal cancer, Endometrial cancer	AD	loss-of-function
POLE	PPAP (polymerase proofreading associated polyposis)	Colorectal cancer	AD/AR	loss-of-function
POLH	Xeroderma pigmentosa V	Squamous cell cancer (skin)	AR	loss-of-function
POT1	Familial cutaneous malignant melanoma	melanoma	AD/AR	
PPOX	porphyria	hepatocellular carcinoma	AD	loss-of-function
PRF1	Familial Hemophagocytic Lymphohistiocytosis	Lymphoid hematological malignancy (lymphoma)	AR	loss-of-function
PRKAR1A	Carney complex	Myxoma (cardiac/cutaneous/breast), Thyroid cancer, Sex cord-stromal tumor	AD	loss-of-function
PRSS1		Pancreatic cancer	AD	loss-of-function
PTCH1	Nevoid basal cell carcinoma syndrome Gorlin Syndrome	Basal cell carcinoma, Medulloblastoma	AD	loss-of-function
PTCH2	Nevoid basal cell carcinoma syndromeGorlin Syndrome	Basal cell carcinoma, Medulloblastoma	AD	loss-of-function
PTEN	Cowden Syndrome PTEN hamartoma tumor syndrome	Breast cancer, Thyroid cancer, Endometrial cancer	AD	loss-of-function
PTPN11	Noonan syndrome	JMML, neuroblastoma	AD	gain-of-function
PTPN13		colon cancer	unk or de novo	
RAD51			unk or de novo	
RAD51C	Fanconi anemia (O) (biallelic mutations)	Monoallelic mutations: Ovarian cancer	AD/AR	loss-of-function
RAD51D		Ovarian cancer	AD	loss-of-function
RAF1	Noonan syndrome	JMML, neuroblastoma	AD	gain-of-function
RB1		Retinoblastoma, Pinealoma, Sarcoma, Melanoma	AD	loss-of-function

RECQL4	Rothmund-Thompson syndrome	Osteosarcoma, Basal cell carcinoma, Squamous cell carcinoma	AR	loss-of-function
REST		Wilms Tumor	AD	
RET	Multiple endocrine neoplasia 2A/2B Familial medullary thyroid carcinoma	Medullary thyroid cancer, Pheochromocytoma	AD	gain-of-function
RHBDF2		Esophageal cancer	AD	gain-of-function
RPL11	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
RPL15	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
RPL26	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
RPL27	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
RPL31	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
RPL35A	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
RPL5	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
RPS10	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function

RPS19	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
RPS24	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
RPS26	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
RPS27	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
RPS29	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
RPS7	Diamond-Blackfan Anemia (DBA)	Myelodysplastic syndrome, Osteosarcoma, Acute myeloid leukemia, Colon adenocarcinoma	AD	loss-of-function
RTEL1	Dyskeratosis congenita	acute myeloid leukemia, Squamous cell carcinoma (head + neck, anorectal), Melanoma	AD/AR	
RUNX1		Myeloid hematological malignancy (leukemia)	AD	loss-of-function
SAMD9		MDS	unk or de novo	gain-of-function
SAMD9L		MDS	unk or de novo	gain-of-function
SBDS	Schwachman-Diamond syndrome	Myeloid hematological malignancy	AR	loss-of-function
SDHA	Carney-Stratakis syndrome	Paraganglioma, Pheochromocytoma, Gastrointestinal stromal tumor (GIST)	AD/AR	loss-of-function
SDHAF2	Familial paraganglioma-pheochromocytoma syndrome	Paraganglioma, Pheochromocytoma	AD	loss-of-function

SDHB	Familial paraganglioma-pheochromocytoma syndrome	Paraganglioma, Pheochromocytoma, Renal cell cancer	AD	loss-of-function
SDHC	Familial paraganglioma-pheochromocytoma syndrome	Paraganglioma, Pheochromocytoma, Gastrointestinal stromal tumor (GIST)	AD	loss-of-function
SDHD	Familial paraganglioma-pheochromocytoma syndrome	Paraganglioma, Pheochromocytoma, Gastrointestinal stromal tumor (GIST)	AD	loss-of-function
SERPINA1	Alpha1 antitrypsin deficiency	Hepatocellular carcinoma	AR	loss-of-function
SETBP1	Schinzel–Giedion Syndrome	germ cell tumor, CML, AML	unk or de novo	gain-of-function
SH2B3	Susceptibility to ALL	Acute Lymphoblastic Leukemia (per CCSS)	AD	
SH2D1A	Lymphoproliferative disease	Lymphoma	X-linked, recessive	loss-of-function
SHOC2	Noonan syndrome		AD	
SLC25A13	Citrullinemia	Hepatocellular carcinoma	AR	loss-of-function
SLX4	Fanconi Anemia (P)	Squamous cell carcinoma (head and neck)	AR	loss-of-function
SMAD4	Juvenile polyposis syndrome	Colorectal cancer	AD	loss-of-function
SMARCA2			unk or de novo	
SMARCA4	Rhabdoid predisposition syndrome	Rhabdoid tumor	AD	loss-of-function
SMARCB1	Rhabdoid predisposition syndrome	Rhabdoid tumor (renal, extra-renal), Central primitive neuroectodermal tumor	AD	loss-of-function
SMARCE1		Meningioma	AD	loss-of-function
SMO		basal cell carcinoma, medulloblastoma	AD	gain-of-function
SOS1	Noonan syndrome	Rhabdomyosarcoma	AD	gain-of-function
SOS2	Noonan syndrome	Rhabdomyosarcoma	AD	gain-of-function
SPOP		prostate cancer	unk or de novo	
SPRTN		Hepatocellular carcinoma	unk or de novo	
SRP72		Acute myeloid Leukemia	AD	gain-of-function
SRY		Gonadoblastoma	Y-linked	loss-of-function
STAT3	Hyper-immunoglobulin E syndrome	Lymphoma	AD	loss-of-function
STK11	Peutz-Jeghers syndrome	Colorectal cancer, Gastric cancer, Breast cancer Sex cord-stromal tumor	AD	loss-of-function

SUFU		Medulloblastoma, meningioma	AD	loss-of-function
T		chordoma	unk or de novo	brachyury homolog
TERT	Dyskeratosis congenita	acute myeloid leukemia, Squamous cell carcinoma (head + neck, anorectal), Melanoma	AD/AR	loss-of-function
TFAP2A			unk or de novo	
TGFBR1	Multiple self-healing squamous epithelioma (MSSE); Ferguson-Smith syndrome	Squamous cell carcinoma (skin)	AD	loss-of-function
TGFBR2	Hereditary non-polyposis colorectal cancer 6	Colon Cancer; also uterus, ovary, breast, stomach, small intestine, skin, and larynx	AD	
TINF2	Dyskeratosis congenitia	acute myeloid leukemia, Squamous cell carcinoma (head + neck, anorectal), Melanoma	AD/AR	
TMEM127		Pheochromocytoma	AD	loss-of-function
TP53	Li-Fraumeni syndrome	Breast cancer, Sarcoma, Adrenocortical carcinoma, Astrocytoma	AD	loss-of-function
TP63	Ectrodactyly-ectodermal dysplasia-cleft syndrome		AD	
TRIM37	Mulibrey-nanism	Wilms tumor	AR	loss-of-function
TSC1	Tuberous sclerosis 1	Renal cell cancer, angiomyolipoma, Subependymal giant cell astrocytoma, Rhabdomyoma (cardiac)	AD	loss-of-function
TSC2	Tuberous sclerosis 2	Renal cell cancer, angiomyolipoma, Subependymal giant cell astrocytoma, Rhabdomyoma (cardiac)	AD	loss-of-function
TSHR	Nonautoimmune autosomal dominant hyperthyroidism	thyroid neoplasms (papillary and follicular cancers).	AD	
TSR2	Diamond Blackfan Anemia	Myelodysplastic syndrome, osteosarcoma, acute myeloid leukemia, colon adenocarcinoma	X-linked, recessive	

UROD	Porphyria (cutanea tarda)	hepatocellular carcinoma	AD/AR	loss-of-function
VHL	Von Hippel-Lindau syndrome	Renal cell cancer, Pheochromocytoma, Neuroendocrine tumor (pancreas), Hemangioblastoma (central nervous system, retina)	AD	loss-of-function
WAS	Wiskott-Aldrich syndromeWAS-related syndrome	Lymphoma	X-linked, recessive	loss-of-function
WRAP53	Dyskeratosis Congenita	MDS, AML, head/neck squamous cell cancer, anogenital adenocarcinoma	AR	
WRN	Werner syndrome	Sarcoma, melanoma, thyroid cancer	AR	loss-of-function
WT1	WAGR syndrome; Denys-Drash syndrome; Frasier syndrome	Wilms tumor, gonadoblastoma	AD	loss-of-function
XPA	Xeroderma pigmentosum (A)	Basal cell carcinoma, squamous cell carcinoma, melanoma	AR	loss-of-function
XPC	Xeroderma pigmentosum (C)	Basal cell carcinoma, squamous cell carcinoma, melanoma	AR	loss-of-function
XRCC3		Cutaneous malignant melanoma, breast cancer	unk or de novo	

AD, autosomal dominant; AR, autosomal recessive; unk, unknown inheritance; de novo, de novo mutation.

eTable 3. Osteosarcoma Candidate Genes and Known Pediatric/Osteosarcoma Somatically Altered Genes Evaluated

140 Candidate genes†		**596 Somatic genes**
ABCB1		*ABCA13*
ABCC2		*ABCA4*
ABCC3		*ABCD1*
ABCC6		*ABI1*
ABCG2		*ABL1*
ACSL1		*ABL2*
ACYP2		*ACKR3*
ADAMTS6		*ACSL3*
ADRA2A		*ACSL6*
AKT1		*AFF1*
APTX		*AFF3*
ARHGAP35		*AFF4*
ATRX		*AKAP9*
BAZ2B		*AKT2*
BCL2L11		*ALDH2*
BMP2		*AMBRA1*
CA1		*AMER1*
CAMK2D		*AMMECR1*
CCNA1		*ANKHD1*
CCNA2		*ANKRD50*
CCNB1		*APP*
CCNB2		*ARAP3*
CCNE2		*ARHGAP26*
CCNH		*ARHGEF12*
CD3EAP		*ARID1A*
CD86		*ARID1B*
CDC6		*ARID2*
CHEK1		*ARID5B*
CKS1B		*ARIH1*
CNOT1		*ARNT*

CNOT4		*ASCL3*
COL18A1		*ASPSCR1*
COL1A1		*ATAD2B*
COL1A2		*ATF1*
COL2A1		*ATIC*
COX8A		*ATXN2*
CTGF		*BAX*
CTLA4		*BCL10*
CTSK		*BCL11A*
CXCL13		*BCL11B*
CYP2C8		*BCL2*
CYP27A1		*BCL3*
DHFR		*BCL6*
DPYD		*BCL7A*
DROSHA		*BCL9*
EDN1		*BCOR*
EGR1		*BCR*
ERCC6		*BICC1*
ESR1		*BIRC3*
FGF2		*BIRC5*
FOS		*BMP4*
GEMIN4		*BRD3*
GGN		*BRD4*
GH1		*BTG1*
GLDC		*BTK*
GNAS		*BUB1*
GNRH2		*C2orf44*
GRM4		*CACNA1E*
GSTK1		*CAMTA1*
GSTM1		*CANT1*
GSTM3		*CARD11*
GSTP1		*CARS*
GSTT1		*CASC5*

IGF1		*CASP10*
IGF2R		*CASP8*
IGFALS		*CBFA2T3*
IL10		*CBFB*
IL12A		*CBLB*
IL12B		*CBLC*
IL16		*CCDC6*
IL1B		*CCK*
IL23A		*CCNB1IP1*
IL23B (aka IL12B)		*CCND1*
IL27		*CCND2*
IL33		*CCND3*
IL6		*CCNE1*
IL8RA (aka CXCR2)		*CD274*
IL8RB (aka CXCR1)		*CD2AP*
ITGA3		*CD74*
JUN		*CD79A*
KEAP1		*CD79B*
LOX		*CD99*
LTA		*CDC25A*
LY75		*CDH11*
MCM5		*CDH2*
MDM1		*CDH9*
MDM2		*CDK12*
MIR34B		*CDK2*
MIR34C		*CDK6*
MLF1IP (aka CENPU)		*CDKN1A*
MMS19		*CDKN2C*
MPG		*CDX2*
MPO		*CHCHD7*
MTHFD1		*CHD3*
MTHFR		*CHD9*
MTR		*CHIC2*

NAF1		*CHN1*
NDUFS4		*CIC*
NFE2L2		*CIITA*
NFIB		*CITED2*
NR1H2		*CLP1*
NUBP2		*CLTC*
OBFC1		*CLTCL1*
OR2T5		*CNBP*
OR7A1P		*CNTN5*
PARP2		*CNTRL*
PCNA		*COL10A1*
PECAM1		*COL5A3*
PIK3CA		*COLCA2*
POMC		*COPS3*
PON1		*COX6C*
PPP1R13L		*CREB1*
PPP1R13L		*CREB3L1*
PRKCG		*CREB3L2*
RECQL5		*CRLF2*
RFC1		*CRTC1*
RNR1		*CRTC3*
SGPL1		*CSDE1*
SHMT1		*CSMD1*
SLCO1B1		*CTDNEP1*
SND1		*CTNNB1*
SQSTM1		*CUL3*
SSSCA1		*CUX1*
TERF1		*DAB1*
TGFB1		*DAPK1*
TNFa		*DAXX*
TNFRSF11A		*DCC*
TNFRSF11B		*DCDC2*
TP53BP2		*DDIT3*

TRNS1		*DDX1*
TXN2		*DDX10*
TYMS		*DDX5*
VCP		*DDX6*
VDR		*DEK*
VEGFA		*DIP2B*
VIP		*DLC1*
VIPR2		*DLEU2*
XRCC1		*DLG2*
XRCC5		*DNAH17*
ZNF208		*DNAH5*
		DNAJA2
		DNM2
		DNMT3A
		DOCK5
		DUSP10
		DUX4L1
		E2F1
		E2F3
		EBF1
		ECT2L
		EGR2
		EIF3H
		EIF4A1
		EIF4A2
		ELF4
		ELK4
		ELL
		ELN
		EML4
		EPB41
		EPS15
		ERAS

		ERBB2
		ERBB3
		ERC1
		ERG
		ETS1
		ETV1
		ETV4
		ETV5
		EWSR1
		EXD2
		EZR
		FAM175A
		FAM46C
		FAP
		FASLG
		FBXL12
		FBXO11
		FBXW7
		FCGR2B
		FCRL4
		FEV
		FGF9
		FGFR1
		FGFR1OP
		FHIT
		FIP1L1
		FLI1
		FLNA
		FLT3
		FMNL3
		FNBP1
		FOXL2
		FOXO1

		FOXO3
		FOXO4
		FOXP1
		FRG1
		FRG2
		FSTL3
		FUBP1
		FUS
		G6PC3
		GAA
		GAB1
		GABRG2
		GAR1
		GAS7
		GATA3
		GGNBP2
		GIGYF2
		GLDN
		GLI1
		GLI2
		GMPS
		GNA11
		GNAQ
		GOLGA5
		GOPC
		GPHN
		GRB10
		GRIN3A
		GSK3B
		H3F3A
		HAS3
		HAX1
		HCN1

		HECTD4
		HECW1
		HERPUD1
		HEY1
		HIP1
		HIST1H4I
		HLF
		HMGA1
		HMGA2
		HMGN2P46
		HNRNPA2B1
		HOOK3
		HOXA11
		HOXA13
		HOXA9
		HOXC11
		HOXC13
		HOXD11
		HOXD13
		HS3ST4
		HSP90AA1
		HSP90AB1
		HUWE1
		IBSP
		IFNA1
		IFNG
		IGF1R
		IGFBP5
		IL2
		IL21R
		IL6ST
		IL7R
		INO80

		IRF4
		ITCH
		ITGA5
		JAK1
		JAK2
		JAK3
		JAZF1
		KAT6A
		KAT6B
		KCND1
		KDM5A
		KDM5C
		KDM6A
		KDR
		KDSR
		KIAA1549
		KIF1B
		KIF5B
		KIRREL
		KLF6
		KLHL28
		KLK2
		KMT2A
		KMT2C
		KMT2D
		KPNA1
		KPNB1
		KRT15
		KRT36
		KTN1
		LAMA5
		LASP1
		LCK

		LCP1
		LHFP
		LIFR
		LMO2
		LMO7
		LOR
		LPP
		LRCH1
		LRIG3
		LRP1B
		LRP2
		LRRFIP2
		LSAMP
		LYL1
		MAF
		MAFB
		MALAT1
		MALT1
		MAML2
		MAP2K4
		MAPT
		MCL1
		MDC1
		MDM4
		MDN1
		MDS2
		MECOM
		MED12
		MEF2A
		MEF2C
		MFN2
		MGMT
		MKL1

		MLF1
		MLLT1
		MLLT10
		MLLT11
		MLLT3
		MLLT4
		MLLT6
		MN1
		MNX1
		MORF4L1
		MPRIP
		MROH2B
		MSI2
		MSN
		MSRB3
		MTCP1
		MTPN
		MUC1
		MUC16
		MYB
		MYC
		MYC-C
		MYCL
		MYCN
		MYD88
		MYH10
		MYNN
		NACA
		NANOS1
		NBAS
		NCAPD2
		NCKIPSD
		NCOA1

		NCOA2
		NCOA4
		NDRG1
		NEFM
		NEK2
		NFKB1
		NFKB2
		NIN
		NKTR
		NONO
		NOTCH1
		NOTCH2
		NPAT
		NPM1
		NR0B1
		NR4A3
		NRG1
		NTRK3
		NUMA1
		NUP214
		NUP98
		NUTM1
		NUTM2A
		NUTM2B
		OLIG2
		OMD
		OR1B1
		OTX2
		P2RY8
		PALLD
		PAPOLA
		PATZ1
		PAX3

		PAX7
		PAX8
		PBX1
		PCDH10
		PCM1
		PCSK7
		PDCD1LG2
		PDE4DIP
		PDGFB
		PER1
		PICALM
		PIK3R1
		PIM1
		PINK1
		PKHD1
		PKHD1L1
		PLAG1
		PLG
		PMEL
		PML
		POLD3
		POU2AF1
		POU5F1
		PPARG
		PPARGC1A
		PPM1D
		PPP1R1A
		PPP2R1A
		PPP3CA
		PPP6R3
		PRCC
		PRDM1
		PRDM16

		PRDM5
		PRRX1
		PSIP1
		PSME4
		PTPRS
		QKI
		RAB27A
		RABEP1
		RAD50
		RAD51B
		RAG1
		RAG2
		RALGDS
		RANBP17
		RAP1GDS1
		RARA
		RASSF1
		RBL2
		RBM15
		RBM8A
		REL
		RFWD2
		RHOH
		RHPN2
		RICTOR
		RMI2
		RNF213
		ROS1
		RPL22
		RPN1
		RUNX1T1
		RUNX2
		RYR3

		SACS
		SAMD4A
		SCAF8
		SCG5
		SCN7A
		SCN9A
		SDC4
		SEMA4D
		SEMA6D
		SEPT5
		SEPT9
		SERINC3
		SERPINA6
		SET
		SETD2
		SF3B1
		SFPQ
		SGSM1
		SH3GL1
		SH3PXD2A
		SHROOM2
		SIK3
		SLC26A10
		SLC34A2
		SLC37A4
		SLC45A3
		SLITRK1
		SMAD7
		SMARCA5
		SMC1A
		SMEK1
		SNF607
		SNX29

		SOCS1
		SOX2
		SPAG17
		SPATA16
		SPECC1
		SPP1
		SPRED1
		SRCAP
		SRGAP2
		SRGAP3
		SRSF2
		SRSF3
		SS18
		SS18L1
		SSX1
		SSX2
		SSX4
		STAG2
		STX11
		STXBP2
		SUZ12
		SYK
		TAF15
		TAL1
		TAL2
		TANC1
		TARDBP
		TCEA1
		TCF12
		TCF3
		TCF7L1
		TCF7L2
		TCL1A

		TCL6
		TET1
		TET2
		TFE3
		TFEB
		TFG
		TFPT
		TFRC
		TH
		THADA
		THBS1
		THRAP3
		THSD4
		TIE1
		TIMP3
		TLX1
		TLX3
		TMEM52
		TMPRSS2
		TNC
		TNFAIP3
		TNFRSF10D
		TNFRSF14
		TNFRSF17
		TNFSF10
		TNRC6A
		TNS3
		TOP1
		TPM3
		TPM4
		TPR
		TRIM24
		TRIM27

		TRIM33
		TRIP11
		TTL
		TTN
		TYK2
		U2AF1
		UBE2D3
		UBE3A
		UNC13D
		USH2A
		USP6
		USP9X
		VAPA
		VNN3
		VTI1A
		WAC
		WDR33
		WHSC1
		WIF1
		WNK1
		WWTR1
		XPO1
		XRCC2
		YIPF3
		YWHAE
		ZBTB16
		ZBTB42
		ZFAND4
		ZFHX3
		ZHX3
		ZMIZ1
		ZMYM2
		ZMYM3

		ZNF331
		ZNF384
		ZNF521
		ZNF638
		ZRSR2

† Genes lists do not overlap; candidate genes that were also known somatically altered genes were included as a candidate gene.

eTable 4. Details of Criteria for Classification of Pathogenicity Categories

ClinVar	InterVar	Classic High Impact	HGMD DM	In silico[¥]	PopMax and TotalCount[†]	Automated Classification	Manual review[₮]
P					Pass	P	Final Bin: P, LP, VUS_D
					Fail	VUS_ND	
LP					Pass	LP	Final Bin: P, LP, VUS_D
					Fail	VUS_ND	
VUS		Yes	Yes		Pass	LP	Final Bin: LP, VUS_D
					Fail	VUS_ND	
		Yes			Pass	VUS_D	Final Bin: LP, VUS_D
					Fail	VUS_ND	
			Yes		Pass	VUS_D	Final Bin: LP, VUS_D
					Fail	VUS_ND	
				Damaging	Pass	VUS_D	
					Fail	VUS_ND	
						VUS_ND	
LB						LB	
B						B	
NA	P				Pass	P	Final Bin: P, LP, VUS_D
					Fail	VUS_ND	
	LP				Pass	LP	Final Bin: LP, VUS_D
					Fail	VUS_ND	
	VUS/NA	Yes	Yes		Pass	LP	Final Bin: LP, VUS_D
					Fail	VUS_ND	
		Yes			Pass	VUS_D	Final Bin: LP, VUS_D
					Fail	VUS_ND	
			Yes		Pass	VUS_D	Final Bin: LP, VUS_D
					Fail	VUS_ND	
				Damaging	Pass	VUS_D	

					Fail	VUS_ND	
						VUS_ND	
	LB					LB	
	B					B	

P = pathogenic, LP = likely pathogenic, VUS_D = variant of uncertain significance (VUS) *in silico* predicted damaging, VUS_ND = VUS *in silico* predicted not damaging, LB = likely benign, B = benign.

¥ Damaging *in silico* is REVEL ≥ 0.5 and CADD ≥ 20 and MetaSVM = D.

† PopMax ≤ 0.005 for AR genes and ≤ 0.001 for non-AR genes; TotalCount ≤ 10.

Ŧ Based on review of literature confirming pathogenicity, gene specific database review, phenotype within the spectrum of associated syndrome or cancer, mutation impact on gene function and mechanism of action.

eTable 5. Details of Pathogenic and Likely Pathogenic Variants in 1004 Patients With Osteosarcoma in Discovery Set

Path. Score	No. cases	Chr	Position	REF	ALT	Gene	Gene Inher.	HGVS.c	HGVS.p	CytoBand	Effect	Impact	Pop Max Freq
LP	1	chr1	100316614	CAG	C	*AGL*	AR	c.18_19delGA	p.Gln6fs	1p21.2	frameshift_variant	HIGH	0.01%
LP	1	chr1	100366273	C	A	*AGL*	AR	c.3444C>A;c.3396C>A	p.Tyr1148*;p.Tyr1132*;p.Tyr1132*;p.Tyr1131*	1p21.2	stop_gained	HIGH	0.00%
P	1	chr1	241675301	G	C	*FH*	AD/AR	c.521C>G		1q43	structural_interaction_variant	HIGH	0.05%
P	1	chr1	241671943	C	T	*FH*	AD/AR	c.698G>A		1q43	structural_interaction_variant	HIGH	0.02%
LP	1	chr1	155208361	C	G	*GBA*	AR	c.535G>Cc.388G>C;c.196G>C;c.274G>C	p.Asp179His;p.Asp179His;p.Asp130His;p.Asp66His;p.Asp92His	1q22	missense_variant	MODERATE	0.02%
P	1	chr1	155210420	C	T	*GBA*	AR	c.115+1G>An.234+1G>A;n.453+1G>A;n.436+1G>A;n.420+1G>A;n.246+1G>A	1q22	splice_donor_variant&intron_variant	HIGH	0.10%	
P	1	chr1	43804234	CCT	C	*MPL*	AD/AR	c.235_236delCT	p.Leu79fs	1p34.2	frameshift_variant	HIGH	0.01%
LP	1	chr1	43814627	G	A	*MPL*	AD/AR	c.1422G>A	p.Trp474*	1p34.2	stop_gained	HIGH	0.00%
P	2	chr1	43804305	G	C	*MPL*	AD/AR	c.305G>C	p.Arg102Pro	1p34.2	missense_variant	MODERATE	0.10%
P	1	chr1	43804396	G	C	*MPL*	AD/AR	c.391+5G>C		1p34.2	splice_region_variant	LOW	0.03%

											&intron_variant		
P	1	chr1	45798117	C	T	*MUTYH*	AR	c.734G>A;c.650G>A;c.683G>A;c.650G>A;c.653G>A;c.650G>A;c.725G>A;c.695G>A;c.692G>A;c.683G>A;c.44G>A;c.692G>A;c.266G>A;c.683G>A	p.Arg245His;p.Arg217His;p.Arg228His;p.Arg217His;p.Arg218His;p.Arg217His;p.Arg242His;p.Arg232His;p.Arg231His;p.Arg228His;p.Arg15His;p.Arg231His;p.Arg89His;p.Arg228His	1p34.1	missense_variant	MODERATE	0.15%
P	1	chr1	45797371	AG	A	*MUTYH*	AR	c.1147delC;c.1063delC;c.1096delC;c.1063delC;c.1066delC;c.1063delC;c.1138delC;c.1108delC;c.1105delC;c.1096delC;c.480delC;c.1105delC	p.Ala385fs;p.Ala357fs;p.Ala368fs;p.Ala357fs;p.Ala358fs;p.Ala357fs;p.Ala382fs;p.Ala372fs;p.Ala371fs;p.Ala368fs;p.Leu161fs;p.Ala371fs	1p34.1	frameshift_variant	HIGH	0.12%
P	1	chr1	45798627	C	T	*MUTYH*	AR	c.467G>A;c.383G>A;c.416G>A;c.383G>A;c.386G>A;c.383G>A;c.458G>A;c.428G>A;c.425G>A;c.416G>A;c.	p.Trp156*;p.Trp128*;p.Trp139*;p.Trp128*;p.Trp129*;p.Trp128*;p.Trp153*;p.Trp143*;p.Trp142*;p.Trp139*;p.Trp142*;p.Trp139*	1p34.1	stop_gained	HIGH	0.06%

								425G>A;c.416G>A					
P	1	chr1	45796890	TTCC	T	*MUTYH*	AR	c.1437_1439delGGA;c.480_482delGGA;c.438_440delGGA;c.48_50delGGA;c.1353_1355delGGA;c.1386_1388delGGA;c.1353_1355delGGA;c.1356_1358delGGA;c.1353_1355delGGA;c.1428_1430delGGA;c.1398_1400delGGA;c.1395_1397delGGA;c.438_440delGGA;c.1386_1388delGGA;c.426_428delGGA;c.1395_1397delGGA	p.Glu480del;p.Glu161del;p.Glu147del;p.Glu17del;p.Glu452del;p.Glu463del;p.Glu452del;p.Glu453del;p.Glu452del;p.Glu477del;p.Glu467del;p.Glu466del;p.Glu147del;p.Glu463del;p.Glu143del;p.Glu466del	1p34.1	disruptive_inframe_deletion	MODERATE	0.02%
LP	1	chr1	156838007	AG	A	*NTRK1*	AR	c.543delG;c.453delG;c.453delG;c.543delG;c.543delG	p.Leu183fs;p.Leu153fs;p.Leu153fs;p.Leu183fs;p.Leu183fs	1q23.1	frameshift_variant	HIGH	0.00%
LP	1	chr1	93301840	G	A	*RPL5*	AD	c.418G>A;c.268G>A	p.Gly140Ser;p.Gly90Ser	1p22.1	missense_variant	MODERATE	0.03%

LP	2	chr2	169828535	C	T	*ABCB11*	AR	c.1460G>A;c.8G>A	p.Arg487His;p.Arg3His	2q31.1	missense_variant	MODERATE	0.14%
P	1	chr2	169825008	A	C	*ABCB11*	AR	c.2012-8T>G;c.*482-8T>G		2q31.1	splice_region_variant&intron_variant	LOW	0.01%
LP	1	chr2	233208190	C	T	*DIS3L2*	AR	c.1717C>T	p.Arg573*	2q37.1	stop_gained	HIGH	0.00%
LP	1	chr2	233208193	CTG	C	*DIS3L2*	AR	c.1722_1723delGT	p.Phe576fs	2q37.1	frameshift_variant	HIGH	0.00%
LP	1	chr2	233200919	AAC	A	*DIS3L2*	AR	c.161_162delCA;c.*2837-3_*2837-2delCA	p.Thr54fs;	2q37.1	frameshift_variant;splice_acceptor_variant&splice_region_variant&intron_variant	HIGH	0.00%
LP	1	chr2	233201091	AG	A	*DIS3L2*	AR	c.253delG	p.Val85fs	2q37.1	frameshift_variant	HIGH	0.08%
LP	1	chr2	128030505	T	TC	*ERCC3*	AR	c.1762dupG;c.1570dupG	p.Glu588fs;p.Glu524fs	2q14.3	frameshift_variant	HIGH	0.01%
LP	1	chr2	128050332	G	A	*ERCC3*	AR	c.325C>T;c.133C>T	p.Arg109*;p.Arg45*	2q14.3	stop_gained	HIGH	0.08%
LP	1	chr2	58388667	CATA	C	*FANCL*	AR	c.1022_1024delTAT;c.923_925delTAT;c.1007_1009delTAT;c.656_658delTAT;c.830_832delTAT	p.Ile341_Cys342delinsSer;p.Ile308_Cys309delinsSer;p.Ile336_Cys337delinsSer;p.Ile219_Cys220delinsSer;p.Ile277_Cys278delinsSer	2p16.1	disruptive_inframe_deletion	MODERATE	0.07%
LP	1	chr2	209108190	T	C	*IDH1*	UNK	c.659A>G		2q34	structural_interaction_variant	HIGH	0.08%

LP	1	chr2	209116175	T	C	*IDH1*	UNK	c.101A>G		2q34	structural_interaction_variant	HIGH	0.00%
LP	2	chr2	47637301	T	G	*MSH2*	AD/AR	c.435T>G		2p21	structural_interaction_variant	HIGH	0.10%
LP	1	chr2	47643513	C	G	*MSH2*	AD/AR	c.1021C>G; c.823C>G	p.Leu341Val;p.Leu275Val;p.Leu341Val	2p21	missense_variant	MODERATE	0.00%
LP	1	chr2	47643537	C	G	*MSH2*	AD/AR	c.1045C>G		2p21	structural_interaction_variant	HIGH	0.04%
LP	1	chr2	48026818	G	A	*MSH6*	AD/AR	c.1696G>A; c.1306G>A; c.790G>A	p.Gly566Arg;p.Gly436Arg;p.Gly264Arg	2p16.3	missense_variant	MODERATE	0.24%
LP	1	chr2	190728500	C	T	*PMS1*	AD	c.1888C>T; c.1360C>T; c.1771C>T; c.1360C>T; c.1705C>T; c.52C>T	p.Arg630*;p.Arg454*;p.Arg591*;p.Arg591*;p.Arg454*;p.Arg569*;p.Arg18*	2q32.2	stop_gained	HIGH	0.02%
LP	1	chr2	3627860	C	T	*RPS7*	AD	c.517C>T	p.Gln173*	2p25.3	stop_gained	HIGH	0.00%
LP	1	chr2	39216456	C	T	*SOS1*	AD	c.3347-1G>A		2p22.1	splice_acceptor_variant&intron_variant	HIGH	0.14%
LP	1	chr2	39239306	A	G	*SOS1*	AD	c.2351T>C	p.Ile784Thr;p.Ile784Thr;p.Ile784Thr	2p22.1	missense_variant	MODERATE	0.00%
P	1	chr3	48611297	A	AG	*COL7A1*	AD/AR	c.6527dupC; c.6431dupC	p.Gly2177fs;p.Gly2145fs	3p21.31	frameshift_variant	HIGH	0.04%
LP	1	chr3	48622467	A	T	*COL7A1*	AD/AR	c.3975+2T>A		3p21.31	splice_donor_variant&intron_variant	HIGH	0.01%
LP	1	chr3	48613168	C	T	*COL7A1*	AD/AR	c.5870G>A; c.5774G>A	p.Arg1957Gln; p.Arg1925Gln	3p21.31	missense_variant	MODERATE	0.00%

LP	1	chr3	48631927	G	A	*COL7A1*	AD/AR	c.140C>T	p.Ser47Leu	3p21.31	missense_variant	MODERATE	0.00%
LP	1	chr3	48601943	GAGGCTACAAC	G	*COL7A1*	AD/AR	n.211-7_213delGTTGTAGCCT	3p21.31	splice_acceptor_variant&splice_region_variant&intron_variant&non_coding_transcript_exon_variant	HIGH	0.00%	
P	1	chr3	10135009	T	G	*FANCD2*	AR	c.3888+2T>G;c.*44+2T>G	3p25.3	splice_donor_variant&intron_variant	HIGH	0.00%	
P	2	chr3	10108951	G	A	*FANCD2*	AR	c.2444G>A;c.941G>A	p.Arg815Gln;p.Arg815Gln;p.Arg815Gln;p.Arg815Gln;p.Arg314Gln	3p25.3	missense_variant	MODERATE	0.11%
P	1	chr3	37038201	G	A	*MLH1*	AD/AR	c.207+1G>A;c.*85+1G>A;c.*287+1G>A;c.*85+1G>A;c.180+1G>A;c.-662+1G>A;n.329+1G>A;c.-425+1G>A;c.-517+1G>A;c.-425+1G>A;c.-83+1G>A;c.-	3p22.2	splice_donor_variant&intron_variant	HIGH	0.00%	

								83+1G>A;n.230+1G>A;n.134+1G>A;c.-517+1G>A;n.211+1G>A					
LP	1	chr3	37059011	T	A	*MLH1*	AD/AR	c.805T>A;c.778T>A;c.82T>A;c.511T>A;c.82T>A;c.145T>A	p.Ser269Thr;p.Ser260Thr;p.Ser28Thr;p.Ser28Thr;p.Ser28Thr;p.Ser28Thr;p.Ser171Thr;p.Ser28Thr;p.Ser49Thr	3p22.2	missense_variant	MODERATE	0.00%
LP	1	chr3	12632325	T	C	*RAF1*	AD	c.1402A>G;c.1342A>G;c.697A>G;c.979A>G;c.1099A>G	p.Ile468Val;p.Ile448Val;p.Ile233Val;p.Ile327Val;p.Ile367Val	3p25.2	missense_variant	MODERATE	0.04%
LP	1	chr3	197680532	G	A	*RPL35A*	AD	c.*163+1G>A		3q29	splice_donor_variant&intron_variant	HIGH	0.00%
LP	1	chr3	189584569	C	T	*TP63*	AD	c.865C>T;c.610C>T;c.583C>T;c.328C>T	p.Pro289Ser;p.Pro204Ser;p.Pro289Ser;p.Pro289Ser;p.Pro289Ser;p.Pro289Ser;p.Pro195Ser;p.Pro195Ser;p.Pro195Ser;p.Pro195Ser;p.Pro110Ser;p.Pro195Ser	3q28	missense_variant	MODERATE	0.00%
LP	1	chr3	189585649	T	C	*TP63*	AD	c.910T>C;c.655T>C;c.6	p.Tyr304His;p.Tyr219His;p.Tyr304His;p.Ty	3q28	missense_variant	MODERATE	0.00%

								28T>C;c.373T>C	r304His;p.Tyr304His;p.Tyr304His;p.Tyr210His;p.Tyr210His;p.Tyr210His;p.Tyr210His;p.Tyr125His;p.Tyr210His				
LP	2	chr3	189612062	G	A	*TP63*	AD	c.1814G>A		3q28	structural_interaction_variant	HIGH	0.07%
LP	2	chr3	189455575	C	T	*TP63*	AD	c.109C>T	p.Arg37*	3q28	stop_gained	HIGH	0.02%
P	1	chr3	10191605	C	T	*VHL*	AD	c.598C>T		3p25.3	structural_interaction_variant	HIGH	0.06%
LP	2	chr3	10183722	G	T	*VHL*	AD	c.191G>T		3p25.3	structural_interaction_variant	HIGH	0.00%
LP	1	chr3	10183534	G	T	*VHL*	AD	c.3G>T	p.Met1?	3p25.3	start_lost	HIGH	0.00%
P	1	chr3	14199738	GCA	G	*XPC*	AR	c.1643_1644delTG;c.1532_1533delTG	p.Val548fs;p.Val511fs	3p25.1	frameshift_variant	HIGH	0.00%
LP	1	chr5	112154970	G	A	*APC*	AD	c.1241G>A		5q22.2	structural_interaction_variant	HIGH	0.02%
LP	1	chr5	112162920	G	C	*APC*	AD	c.1524G>C		5q22.2	structural_interaction_variant	HIGH	0.00%
LP	1	chr5	112175750	A	G	*APC*	AD	c.4459A>G		5q22.2	protein_protein_contact	HIGH	0.00%
LP	1	chr5	79952308	C	T	*MSH3*	AR	c.316C>T	p.Gln106*	5q14.1	stop_gained	HIGH	0.00%
LP	1	chr5	177580563	T	A	*NHP2*	AD/AR	c.161-2A>T;n.228-2A>T	5q35.3	splice_acceptor_varian	HIGH	0.00%	

										t&intron_variant			
P	1	chr5	240511	G	T	*SDHA*	AD/AR	c.1471G>T; c.1327G>T; c.124G>T	p.Glu491*;p.Glu491*;p.Glu443*;p.Glu42*	5p15.33	stop_gained	HIGH	0.00%
LP	1	chr5	1279527	G	A	*TERT*	AD/AR	c.2009C>T	p.Ala670Val	5p15.33	missense_variant	MODERATE	0.00%
LP	1	chr5	1280427	C	T	*TERT*	AD/AR	c.1796G>A	p.Arg599Gln	5p15.33	missense_variant	MODERATE	0.01%
LP	1	chr5	1294106	C	T	*TERT*	AD/AR	c.895G>A	p.Val299Met	5p15.33	missense_variant	MODERATE	0.14%
P	1	chr6	35423696	C	T	*FANCE*	AR	c.421C>T	p.Arg141*	6p21.31	stop_gained	HIGH	0.00%
P	1	chr6	35423630	C	T	*FANCE*	AR	c.355C>T	p.Gln119*	6p21.31	stop_gained	HIGH	0.00%
LP	1	chr6	26092870	G	A	*HFE*	AR	c.649+1G>A		6p22.2	splice_donor_variant&intron_variant	HIGH	0.00%
LP	1	chr7	55220349	G	T	*EGFR*	AD	c.739G>T;c.604G>T;c.580G>T	p.Asp247Tyr;p.Asp202Tyr;p.Asp247Tyr;p.Asp247Tyr;p.Asp247Tyr;p.Asp194Tyr	7p11.2	missense_variant	MODERATE	0.00%
LP	1	chr7	55266472	G	A	*EGFR*	AD	c.2764G>A; c.2629G>A; c.2605G>A	p.Glu922Lys;p.Glu877Lys;p.Glu869Lys	7p11.2	missense_variant	MODERATE	0.00%
LP	1	chr7	55260490	T	C	*EGFR*	AD	c.2657T>C; c.2522T>C; c.2498T>C	p.Ile886Thr;p.Ile841Thr;p.Ile833Thr	7p11.2	missense_variant	MODERATE	0.00%
LP	1	chr7	55266550	G	A	*EGFR*	AD	c.2842G>A; c.2707G>A; c.2683G>A	p.Val948Ile;p.Val903Ile;p.Val895Ile	7p11.2	missense_variant	MODERATE	0.08%
LP	1	chr7	55268045	G	A	*EGFR*	AD	c.2885G>A		7p11.2	structural_interaction_variant	HIGH	0.20%

LP	1	chr7	148511116	C	T	*EZH2*	AD	c.1786G>A; c.1618G>A; c.1771G>A; c.1654G>A; c.1744G>A	p.Ala596Thr;p.Ala540Thr;p.Ala591Thr;p.Ala552Thr;p.Ala540Thr;p.Ala540Thr;p.Ala582Thr	7q36.1	missense_variant	MODERATE	0.05%
LP	1	chr7	148508755	C	T	*EZH2*	AD	c.1894G>A		7q36.1	structural_interaction_variant	HIGH	0.00%
LP	1	chr7	116398601	C	T	*MET*	AD	c.2191C>T; c.31C>T	p.Arg731*;p.Arg731*;p.Arg731*;p.Arg11*	7q31.2	stop_gained	HIGH	0.00%
LP	1	chr7	6022534	C	G	*PMS2*	AD/AR	c.2095G>C; c.1777G>C; c.892G>C	p.Asp699His;p.Asp593His;p.Asp298His	7p22.1	missense_variant	MODERATE	0.00%
P	1	chr7	6022511	CT	C	*PMS2*	AD/AR	c.2117delA; c.1799delA; c.914delA	p.Lys706fs;p.Lys600fs;p.Lys305fs	7p22.1	frameshift_variant	HIGH	0.00%
LP	1	chr7	124532320	C	A	*POT1*	AD/AR	c.124G>T	p.Asp42Tyr	7q31.33	missense_variant&splice_region_variant	MODERATE	0.00%
LP	1	chr7	124482897	T	C	*POT1*	AD/AR	c.1127A>G; c.734A>G	p.Gln376Arg;p.Gln245Arg;p.Gln376Arg;p.Gln376Arg	7q31.33	missense_variant	MODERATE	0.10%
LP	1	chr7	124511015	G	A	*POT1*	AD/AR	c.205C>T	p.Leu69Phe	7q31.33	missense_variant	MODERATE	0.00%
LP	2	chr7	124499043	C	T	*POT1*	AD/AR	c.670G>A		7q31.33	structural_interaction_variant	HIGH	0.01%
P	1	chr7	124464068	TTA	T	*POT1*	AD/AR	c.1851_1852delTA;c.201_202delTA; c.1458_1459delTA;c.345_346delTA	p.Asp617fs;p.Asp67fs;p.Asp486fs;p.Asp115fs	7q31.33	frameshift_variant	HIGH	0.02%

LP	1	chr7	92735355	A	T	*SAMD9*	AD	c.56T>A	p.Val19Glu;p.Val19Glu	7q21.2	missense_variant	MODERATE	0.00%
P	2	chr7	66459273	T	A	*SBDS*	AR	c.184A>T	p.Lys62*;p.Lys62*	7q11.21	stop_gained	HIGH	0.14%
P	1	chr7	95813688	G	A	*SLC25A13*	AR	c.1081C>T;c.1078C>T;c.754C>T	p.Arg361*;p.Arg360*;p.Arg252*	7q21.3	stop_gained	HIGH	0.01%
LP	1	chr7	128845209	G	T	*SMO*	AD	c.703G>T	p.Ala235Ser	7q32.1	missense_variant	MODERATE	0.00%
LP	1	chr7	128846049	G	A	*SMO*	AD	c.979G>A;c.61G>A	p.Ala327Thr;p.Ala21Thr	7q32.1	missense_variant	MODERATE	0.01%
LP	1	chr7	128843230	C	T	*SMO*	AD	c.337C>T	p.Arg113Trp	7q32.1	missense_variant	MODERATE	0.00%
LP	2	chr7	128843386	G	C	*SMO*	AD	c.493G>C	p.Asp165His	7q32.1	missense_variant	MODERATE	0.00%
LP	1	chr7	128846127	C	G	*SMO*	AD	c.1057C>G;c.139C>G	p.Leu353Val;p.Leu47Val	7q32.1	missense_variant	MODERATE	0.00%
P	1	chr8	90955582	C	A	*NBN*	AR	c.2083G>T;c.1837G>T	p.Gly695*;p.Gly613*	8q21.3	stop_gained	HIGH	0.00%
P	1	chr8	90967765	TG	T	*NBN*	AR	c.1142delC;c.896delC	p.Pro381fs;p.Pro299fs	8q21.3	frameshift_variant	HIGH	0.00%
LP	1	chr8	90955480	C	A	*NBN*	AR	c.2184+1G>T;c.*2057+1G>T;c.1938+1G>T	8q21.3	splice_donor_variant&intron_variant	HIGH	0.00%	
LP	1	chr8	145738323	G	A	*RECQL4*	AR	c.2662C>T;c.832C>T	p.Gln888*;p.Gln278*	8q24.3	stop_gained	HIGH	0.00%
P	1	chr8	145738509	G	A	*RECQL4*	AR	c.2476C>T;c.646C>T	p.Arg826*;p.Arg216*	8q24.3	stop_gained	HIGH	0.02%
P	2	chr8	145740366	CA	C	*RECQL4*	AR	c.1573delT;c.427delT	p.Cys525fs;p.Cys143fs	8q24.3	frameshift_variant	HIGH	0.04%
LP	1	chr8	145739833	A	G	*RECQL4*	AR	c.1697T>C;c.65T>C;c.551T>C	p.Leu566Pro;p.Leu22Pro;p.Leu184Pro	8q24.3	missense_variant	MODERATE	0.10%
LP	3	chr8	145742480	G	A	*RECQL4*	AR	c.308C>T;c.179C>T	p.Pro103Leu;p.Pro60Leu	8q24.3	missense_variant	MODERATE	0.10%

LP	2	chr8	30954294	C	T	*WRN*	AR	c.1909C>T	p.Arg637Trp	8p12	missense_variant	MODERATE	0.14%
LP	1	chr8	31024680	TC	T	*WRN*	AR	c.4128delC	p.Gly1377fs	8p12	frameshift_variant	HIGH	0.00%
LP	1	chr8	30977902	TCA	T	*WRN*	AR	c.2594_2595delAC	p.His865fs	8p12	frameshift_variant	HIGH	0
LP	1	chr8	30922443	G	A	*WRN*	AR	c.368G>A		8p12	structural_interaction_variant	HIGH	0.00%
LP	3	chr9	21970985	C	G	*CDKN2A*	AD	c.373G>C;c.373G>C;c.220G>C;c.373G>C;c.220G>C;c.220G>C;c.373G>C;c.220G>C;c.220G>C	p.Asp125His;p.Asp125His;p.Asp74His;p.Asp125His;p.Asp74His;p.Asp74His;p.Asp125His;p.Asp74His;p.Asp74His	9p21.3	missense_variant	MODERATE	0.10%
LP	5	chr9	21974681	A	G	*CDKN2A*	AD	c.146T>C	p.Ile49Thr	9p21.3	missense_variant	MODERATE	0.46%
P	1	chr9	21971057	C	A	*CDKN2A*	AD	c.467G>T;c.301G>T;c.344G>T;c.301G>T;c.148G>T;c.344G>T;c.301G>T;c.148G>T;c.148G>T;c.301G>T;c.148G>T;c.148G>T	p.Arg156Leu;p.Gly101Trp;p.Arg115Leu;p.Gly101Trp;p.Gly50Trp;p.Arg115Leu;p.Gly101Trp;p.Gly50Trp;p.Gly50Trp;p.Gly101Trp;p.Gly50Trp;p.Gly50Trp	9p21.3	missense_variant	MODERATE	0.00%
LP	1	chr9	21970994	C	T	*CDKN2A*	AD	c.364G>A;c.364G>A;c.211G>A;c.364G>A;c.211G>A;c.211G>A;c.364G>A;c.211G>A;c.211G>A	p.Gly122Ser;p.Gly122Ser;p.Gly71Ser;p.Gly122Ser;p.Gly71Ser;p.Gly71Ser;p.Gly122Ser;p.Gly71Ser;p.Gly71Ser	9p21.3	missense_variant	MODERATE	0

LP	1	chr9	21974681	A	C	*CDKN2A*	AD	c.146T>G	p.Ile49Ser	9p21.3	missense_variant	MODERATE	0.00%
LP	1	chr9	21974794	A	AGGCTCCATGCTGCTCCCCGCCGCC	*CDKN2A*	AD	c.9_32dupGGCGGCGGGGAGCAGCATGGAGCC	p.Pro11_Ser12insAlaAlaGlySerSerMetGluPro	9p21.3	disruptive_inframe_insertion	MODERATE	0.00%
P	1	chr9	271626	G	T	*DOCK8*	AR	c.54-1G>T;c.54-1G>T;c.*17-1G>T;c.-151-1G>T;n.163-1G>T;c.-151-1G>T	9p24.3	splice_acceptor_variant&intron_variant	HIGH	0.04%	
P	1	chr9	97912338	G	A	*FANCC*	AR	c.553C>T	p.Arg185*	9q22.32	stop_gained	HIGH	0.03%
LP	1	chr9	97888864	C	G	*FANCC*	AR	c.844-1G>C;c.844-1G>C;n.171-1G>C;n.29-1G>C;n.199-1G>C	9q22.32	splice_acceptor_variant&intron_variant	HIGH	0.00%	
LP	1	chr9	35075080	C	G	*FANCG*	AR	c.1481-1G>C;c.*957-1G>C	9p13.3	splice_acceptor_variant&intron_variant	HIGH	0.00%	
LP	1	chr9	101599363	G	A	*GALNT12*	AD	c.1145G>A	p.Arg382His	9q22.33	missense_variant	MODERATE	0.00%
LP	1	chr9	98240450	C	A	*PTCH1*	AD	c.1234G>T;c.781G>T;c.781G>T;c.1036G>T;c.8	p.Ala412Ser;p.Ala261Ser;p.Ala261Ser;p.Ala346Ser;p.Ala2	9q22.32	missense_variant	MODERATE	0.02%

								71G>T;c.1036G>T;c.781G>T;c.1231G>T;c.385G>T	91Ser;p.Ala346Ser;p.Ala261Ser;p.Ala411Ser;p.Ala129Ser				
LP	1	chr9	98279098	TC	T	*PTCH1*	AD	c.4delG	p.Glu2fs	9q22.32	frameshift_variant	HIGH	0.01%
LP	1	chr9	2039717	C	T	*SMARCA2*	UNK	c.607C>T	p.Gln203*	9p24.3	stop_gained	HIGH	0.00%
LP	1	chr10	123239526	C	T	*FGFR2*	AD	c.981G>A	p.Trp327*	10q26.13	stop_gained	HIGH	0.00%
P	1	chr10	72360543	G	T	*PRF1*	AR	c.116C>A	p.Pro39His	10q22.1	missense_variant	MODERATE	0.00%
LP	1	chr10	72360526	C	T	*PRF1*	AR	c.133G>A	p.Gly45Arg	10q22.1	missense_variant	MODERATE	0.10%
LP	2	chr10	72358173	G	A	*PRF1*	AR	c.1304C>T	p.Thr435Met	10q22.1	missense_variant	MODERATE	0.01%
LP	1	chr10	89692831	T	A	*PTEN*	AD	c.315T>A	p.Cys105*	10q23.31	stop_gained	HIGH	0.00%
P	1	chr11	108181032	C	T	*ATM*	AD/AR	c.5908C>T	p.Gln1970*	11q22.3	stop_gained	HIGH	0.00%
P	1	chr11	108121752	CAG	C	*ATM*	AD/AR	c.1564_1565delGA	p.Glu522fs	11q22.3	frameshift_variant	HIGH	0.05%
LP	1	chr11	108202286	T	C	*ATM*	AD/AR	c.7629+2T>C;c.7629+2T>C;n.3844+2T>C;n.3033+2T>C	11q22.3	splice_donor_variant&intron_variant	HIGH	0.00%	
P	1	chr11	108202611	CTCTAGAATT	C	*ATM*	AD/AR	c.7638_7646delTAGAATTTC	p.Arg2547_Ser2549del	11q22.3	disruptive_inframe_deletion	MODERATE	0.09%
P	1	chr11	108236142	C	CA	*ATM*	AD/AR	c.9079dupA	p.Ser3027fs	11q22.3	frameshift_variant	HIGH	0.00%
P	1	chr11	71146512	C	T	*DHCR7*	AR	c.1337G>A;c.587G>A	p.Arg446Gln;p.Arg196Gln	11q13.4	missense_variant	MODERATE	0.01%

LP	1	chr11	71148910	C	T	*DHCR7*	AR	c.911G>A;c.266G>A;c.911G>A;c.161G>A;c.278G>A	p.Trp304*;p.Trp89*;p.Trp304*;p.Trp54*;p.Trp93*	11q13.4	stop_gained	HIGH	0.00%
LP	1	chr11	71158655	C	T	*DHCR7*	AR	c.-7+1G>A		11q13.4	splice_donor_variant&intron_variant	HIGH	0.00%
LP	1	chr11	71155910	C	G	*DHCR7*	AR	c.89G>C	p.Gly30Ala	11q13.4	missense_variant	MODERATE	0.10%
LP	2	chr11	71146861	C	T	*DHCR7*	AR	c.988G>A;c.238G>A	p.Val330Met;p.Val80Met	11q13.4	missense_variant	MODERATE	0.14%
LP	1	chr11	44254000	C	T	*EXT2*	AD	c.1859C>T;c.1760C>T	p.Thr620Met;p.Thr587Met	11p11.2	missense_variant	MODERATE	0.13%
LP	1	chr11	118963136	G	A	*HMBS*	AD	c.674G>A		11q23.3	structural_interaction_variant	HIGH	0.05%
LP	3	chr11	64572021	G	A	*MEN1*	AD	c.1633C>T;c.1453C>T;c.1513C>T	p.Pro545Ser;p.Pro485Ser;p.Pro505Ser;p.Pro540Ser	11q13.1	missense_variant	MODERATE	0.05%
LP	1	chr11	64575365	G	A	*MEN1*	AD	c.652C>T		11q13.1	structural_interaction_variant	HIGH	0.00%
P	1	chr11	64572548	C	A	*MEN1*	AD	c.1308G>T		11q13.1	structural_interaction_variant	HIGH	0.00%
LP	1	chr11	94204810	G	A	*MRE11A*	AD/AR	c.784C>T	p.Gln262*;p.Gln259*	11q21	stop_gained	HIGH	0.00%
LP	1	chr12	121437161	G	C	*HNF1A*	AD	c.1592G>C	p.Ser531Thr	12q24.31	missense_variant	MODERATE	0.01%
LP	1	chr12	25368455	G	A	*KRAS*	AD	c.490C>T	p.Arg164*	12p12.1	structural_interaction_variant;stop_gained	HIGH	0.03%
LP	2	chr12	133249340	T	C	*POLE*	AD/AR	c.1559A>G;c.1478A>G;	p.Gln520Arg;p.Gln493Arg;p.	12q24.33	missense_variant	MODERATE	0.00%

								c.899A>G;c.413A>G	Gln300Arg;p.Gln138Arg				
LP	1	chr12	133249420	CA	C	*POLE*	AD/AR	c.1478delT;c.1397delT;c.818delT;c.332delT	p.Leu493fs;p.Leu466fs;p.Leu273fs;p.Leu111fs	12q24.33	frameshift_variant	HIGH	0.03%
LP	1	chr12	133245023	AG	A	*POLE*	AD/AR	c.2091delC;c.2010delC;c.1431delC	p.Leu698fs;p.Leu671fs;p.Leu478fs	12q24.33	frameshift_variant	HIGH	0.05%
LP	1	chr13	32936732	G	C	*BRCA2*	AD/AR	c.7878G>C;c.7878G>C	p.Trp2626Cys;p.Trp2626Cys	13q13.1	missense_variant	MODERATE	0.00%
P	1	chr13	32912623	C	CTGAGGA	*BRCA2*	AD/AR	c.4131_4132insTGAGGA	p.Asn1377_Thr1378insTerGly;p.Asn1377_Thr1378insTerGly	13q13.1	stop_gained&conservative_inframe_insertion	HIGH	0.00%
P	1	chr13	32953577	C	T	*BRCA2*	AD/AR	c.8878C>T	p.Gln2960*	13q13.1	stop_gained	HIGH	0.00%
P	1	chr13	32954222	C	T	*BRCA2*	AD/AR	c.9196C>T;c.9196C>T;c.151C>T	p.Gln3066*;p.Gln3066*;p.Gln51*	13q13.1	stop_gained	HIGH	0.00%
P	1	chr13	32913125	CT	C	*BRCA2*	AD/AR	c.4638delT	p.Phe1546fs	13q13.1	frameshift_variant	HIGH	0.00%
P	1	chr13	32914437	GT	G	*BRCA2*	AD/AR	c.5946delT	p.Ser1982fs	13q13.1	frameshift_variant	HIGH	0.04%
P	2	chr13	32913558	C	CA	*BRCA2*	AD/AR	c.5073dupA	p.Trp1692fs	13q13.1	frameshift_variant	HIGH	0.00%
P	1	chr13	32954260	CG	C	*BRCA2*	AD/AR	c.9235delG;c.9235delG;c.190delG	p.Val3079fs;p.Val3079fs;p.Val64fs	13q13.1	frameshift_variant	HIGH	0.01%
LP	1	chr13	103527976	AATCATCTGAT	A	*ERCC5*	AR	c.3285_3294delATCATCTGAT;c.984_993delATCATCTGAT	p.Ser1096fs;p.Ser329fs	13q33.1	frameshift_variant	HIGH	0.00%

P	1	chr13	49033844	C	T	*RB1*	AD	c.1981C>T		13q14.2	structural_interaction_variant	HIGH	0.08%
P	1	chr13	48936995	C	T	*RB1*	AD	c.763C>T	p.Arg255*	13q14.2	stop_gained	HIGH	0.00%
LP	1	chr13	49039375	G	A	*RB1*	AD	c.2360G>A	p.Arg787Gln	13q14.2	missense_variant	MODERATE	0.02%
LP	1	chr13	49039504	G	A	*RB1*	AD	c.2489G>A		13q14.2	protein_protein_contact	HIGH	0.00%
LP	1	chr14	95562349	G	GA	*DICER1*	AD	c.4907_4908insT;c.941_942insT	p.Ser1637fs;p.Ser315fs	14q32.13	frameshift_variant	HIGH	0
LP	1	chr14	45618181	C	T	*FANCM*	AD/AR	c.901C>T;c.901C>T;c.823C>T	p.Gln301*;p.Gln301*;p.Gln275*	14q21.2	stop_gained	HIGH	0.00%
LP	1	chr14	45645406	C	G	*FANCM*	AD/AR	c.3449C>G;c.3371C>G;c.1997C>G;c.245C>G	p.Ser1150*;p.Ser1124*;p.Ser666*;p.Ser82*	14q21.2	stop_gained	HIGH	0.00%
LP	2	chr14	45667921	C	T	*FANCM*	AD/AR	c.5791C>T;c.5713C>T;c.4339C>T;c.2692C>T;c.169C>T;c.5791C>T;c.5791C>T	p.Arg1931*;p.Arg1905*;p.Arg1447*;p.Arg898*;p.Arg57*;;	14q21.2	structural_interaction_variant;stop_gained	HIGH	0.45%
P	1	chr14	45636336	C	T	*FANCM*	AD/AR	c.1972C>T;c.1972C>T;c.1894C>T;c.520C>T	p.Arg658*;p.Arg658*;p.Arg632*;p.Arg174*	14q21.2	stop_gained	HIGH	0.10%
LP	1	chr14	94847286	T	A	*SERPINA1*	AR	c.839A>T	p.Asp280Val	14q32.13	missense_variant	MODERATE	0.09%
LP	2	chr14	94849325	C	T	*SERPINA1*	AR	c.250G>A	p.Ala84Thr	14q32.13	missense_variant	MODERATE	0.07%
LP	1	chr14	94849249	G	A	*SERPINA1*	AR	c.326C>T	p.Thr109Met	14q32.13	missense_variant	MODERATE	0.10%

P	1	chr14	94844912	T	TA	*SERPINA1*	AR	c.1130dupT	p.Leu377fs	14q32.13	frameshift_variant	HIGH	0.01%
LP	1	chr14	24709617	CCACT	C	*TINF2*	AD/AR	c.1065_*3delAGTG;c.822_*3delAGTG;c.423_*3delAGTG	p.Ter355fs;p.Ter274fs;p.Ter141fs	14q12	frameshift_variant&stop_lost	HIGH	0.05%
LP	1	chr15	40501902	T	G	*BUB1B*	AR	c.2252T>G;c.2210T>G	p.Leu751*;p.Leu737*	15q15.1	stop_gained	HIGH	0.01%
P	1	chr15	80460605	G	T	*FAH*	AR	c.554-1G>T;c.554-1G>T;c.554-1G>T;c.344-1G>T;n.482-1G>T;n.100-1G>T	15q25.1	splice_acceptor_variant&intron_variant	HIGH	0.03%	
LP	1	chr15	80464583	C	G	*FAH*	AR	c.699C>G;c.699C>G;c.699C>G;c.489C>G	p.Asp233Glu;p.Asp233Glu;p.Asp233Glu;p.Asp163Glu	15q25.1	missense_variant	MODERATE	0.00%
P	1	chr15	80472572	G	A	*FAH*	AR	c.164G>A	p.Ser55Asn	15q25.1	missense_variant	MODERATE	0.16%
LP	1	chr16	67693137	T	C	*ACD*	AD/AR	c.746A>G;c.737A>G;c.29A>G;c.497A>G;c.317A>G	p.Asn249Ser;p.Asn246Ser;p.Asn10Ser;p.Asn166Ser;p.Asn106Ser	16q22.1	missense_variant	MODERATE	0.05%
LP	1	chr16	3807934	T	C	*CREBBP*	AD	c.3485A>G		16p13.3	structural_interaction_variant	HIGH	0.01%
LP	1	chr16	50828116	A	AT	*CYLD*	AD	c.2470-3dupT;c.2461-3dupT;c.2470-3dupT;c.2461-	16q12.1	splice_acceptor_variant&intron_variant	HIGH	0.00%	

								3dupT;c.2461-3dupT;c.2461-3dupT;c.2470-3dupT;c.1915-3dupT;c.49-3dupT					
LP	1	chr16	14020608	G	A	*ERCC4*	AR	c.579G>A;c.579G>A;c.198G>A	p.Trp193*;p.Trp193*;p.Trp66*	16p13.12	stop_gained	HIGH	0.02%
LP	1	chr16	14031642	G	A	*ERCC4*	AR	c.1831G>A;c.22G>A	p.Gly611Arg;p.Gly8Arg	16p13.12	missense_variant	MODERATE	0.00%
LP	1	chr16	89805462	G	A	*FANCA*	AR	c.397C>T	p.Gln133*	16q24.3	stop_gained	HIGH	0.00%
LP	1	chr16	89809219	C	A	*FANCA*	AR	c.3754G>T;c.3754G>T;c.232G>T;c.82G>T;c.16G>T	p.Glu1252*;p.Glu1252*;p.Glu78*;p.Glu28*;p.Glu6*	16q24.3	stop_gained	HIGH	0.00%
LP	1	chr16	89882683	G	C	*FANCA*	AR	c.341C>G	p.Ser114*	16q24.3	stop_gained	HIGH	0.08%
LP	1	chr16	89986531	T	C	*MC1R*	AD	c.865T>C;c.865T>C	p.Cys289Arg;p.Cys289Arg	16q24.3	missense_variant	MODERATE	0.02%
P	1	chr16	23641191	G	GAGCAAGTTGGGGTGTGC	*PALB2*	AD/AR	c.2267_2283dupGCACACCCCAACTTGCT	p.His762fs	16p12.2	frameshift_variant	HIGH	0.00%
P	1	chr16	23646296	G	C	*PALB2*	AD/AR	c.1571C>G	p.Ser524*	16p12.2	stop_gained	HIGH	0.00%
P	1	chr16	23614911	GAAGT	G	*PALB2*	AD/AR	c.3426_3429delACTT;c.	p.Leu1142fs;;	16p12.2	frameshift_variant;stru	HIGH	0.00%

								3426_3429delACTT;c.3426_3429delACTT			ctural_interaction_variant		
LP	2	chr16	2138446	G	C	*TSC2*	AD	c.5260-1G>C;c.4915-1G>C;c.5059-1G>C;c.5131-1G>C;c.4951-1G>C;c.5191-1G>C;c.*4427-1G>C;c.5092-1G>C;n.2983-1G>C;n.2375-1G>C;c.1441-1G>C	16p13.3	splice_acceptor_variant&intron_variant	HIGH	0.00%	
LP	1	chr16	2129652	C	T	*TSC2*	AD	c.3379C>T;c.3103C>T;c.3247C>T;c.3250C>T;c.3139C>T;c.3379C>T;c.3280C>T	p.Arg1127Trp;p.Arg1035Trp;p.Arg1083Trp;p.Arg1084Trp;p.Arg1047Trp;p.Arg1127Trp;p.Arg1094Trp	16p13.3	missense_variant	MODERATE	0.09%
LP	1	chr17	63526149	GTC	G	*AXIN2*	AD	c.2475_2476delGA;c.2280_2281delGA	p.Glu825fs;p.Glu760fs	17q24.1	frameshift_variant	HIGH	0.00%
P	1	chr17	41246748	GAA	G	*BRCA1*	AD	c.798_799delTT;c.798_7	p.Ser267fs;p.Ser267fs;p.Ser2	17q21.31	frameshift_variant	HIGH	0.00%

								99delTT;c.798_799delTT;c.798_799delTT;c.657_658delTT;c.147_148delTT;c.798_799delTT;c.720_721delTT;c.798_799delTT;c.393_394delTT	67fs;p.Ser267fs;p.Ser220fs;p.Ser50fs;p.Ser267fs;p.Ser241fs;p.Ser267fs;p.Ser132fs				
P	1	chr17	41245293	A	T	*BRCA1*	AD	c.2255T>A;c.2255T>A;c.1367T>A;c.2255T>A;c.2255T>A;c.2114T>A	p.Leu752*;p.Leu752*;p.Leu456*;p.Leu752*;p.Leu752*;p.Leu705*	17q21.31	stop_gained	HIGH	0.00%
P	1	chr17	41215934	A	C	*BRCA1*	AD	c.5172T>G;c.5109T>G;c.1560T>G;c.4221T>G;c.4392T>G;c.1683T>G;c.1797T>G;c.39T>G;c.582T>G;c.4968T>G;c.1797T>G;c.1422T>G;c.1797T>G;c.1659T>G;c.5109T>G	p.Tyr1724*;p.Tyr1703*;p.Tyr520*;p.Tyr1407*;p.Tyr1464*;p.Tyr561*;p.Tyr599*;p.Tyr13*;p.Tyr194*;p.Tyr1656*;p.Tyr599*;p.Tyr474*;p.Tyr599*;p.Tyr553*	17q21.31	structural_interaction_variant;stop_gained	HIGH	0.00%
P	1	chr17	41256137	A	T	*BRCA1*	AD	c.441+2T>A;c.441+2T>A;c.441+2T>A;c.-448+2T>A;c	17q21.31	splice_donor_variant&intron_variant	HIGH	0.00%	

								.441+2T>A;c.441+2T>A;c.441+2T>A;c.441+2T>A;c.300+2T>A;c.*227+2T>A;c.441+2T>A;c.189+2T>A;c.441+2T>A;c.300+2T>A;c.189+2T>A;n.505+2T>A;c.441+2T>A;c.363+2T>A;c.*377+2T>A;c.441+2T>A;c.162+2T>A;c.441+2T>A;c.*227+2T>A					
P	1	chr17	59761413	CTTTG	C	*BRIP1*	AD/AR	c.2990_2993delCAAA	p.Thr997fs	17q23.2	frameshift_variant	HIGH	0.00%
P	1	chr17	8140757	GCTTT	G	*CTC1*	AR	c.724_727delAAAG	p.Lys242fs	17p13.1	frameshift_variant	HIGH	0.03%
LP	1	chr17	41061435	G	C	*G6PC*	AR	c.562G>C;c.485G>C	p.Gly188Arg;p.Arg162Thr	17q21.31	missense_variant&splice_region_variant	MODERATE	0.01%
LP	1	chr17	41061435	G	A	*G6PC*	AR	c.562G>A;c.485G>A	p.Gly188Ser;p.Arg162Lys	17q21.31	missense_variant&splice_region_variant	MODERATE	0.00%
P	1	chr17	41055947	G	A	*G6PC*	AR	c.231-1G>A;c.231-1G>A;c.231-	17q21.31	splice_acceptor_variant&intron_variant	HIGH	0.00%	

								1G>A;n.296-1G>A					
LP	1	chr17	41063408	C	T	*G6PC*	AR	c.1039C>T	p.Gln347*	17q21.31	stop_gained	HIGH	0.10%
LP	1	chr17	29533306	G	GTAGT	*NF1*	AD	c.1310_1311insAGTT;c.1310_1311insAGTT;c.1310_1311insAGTT;c.1412_1413insAGTT;c.308_309insAGTT	p.Glu438fs;p.Glu438fs;p.Glu438fs;p.Glu472fs;p.Glu104fs	17q11.2	frameshift_variant	HIGH	0.01%
LP	1	chr17	66521095	C	T	*PRKAR1A*	AD	c.545C>T	p.Thr182Met	17q24.2	missense_variant;missense_variant&splice_region_variant	MODERATE	0.10%
LP	1	chr17	66518939	C	T	*PRKAR1A*	AD	c.220C>T	p.Arg74Cys	17q24.2	missense_variant	MODERATE	0.01%
P	1	chr17	33433425	G	A	*RAD51D*	AD	c.616C>T;c.556C>T;c.556C>T;c.199C>T;c.421C>T;c.220C>T;c.199C>T;c.199C>T;c.562C>T;c.25C>T	p.Arg206*;p.Arg186*;p.Arg186*;p.Arg67*;p.Arg141*;p.Arg74*;p.Arg67*;p.Arg67*;p.Arg188*;p.Arg9*	17q12	stop_gained	HIGH	0.01%
P	1	chr17	33446581	G	A	*RAD51D*	AD	c.52C>T	p.Gln18*	17q12	stop_gained	HIGH	0.00%
LP	1	chr17	7578469	13bpdel		*TP53*	AD			17p13.1	frameshift_variant	HIGH	0.00%
LP	2	chr17	7579368	A	G	*TP53*	AD	c.319T>C		17p13.1	structural_interaction_variant	HIGH	0.08%

P	1	chr17	7578479	G	C	*TP53*	AD	c.451C>G;c.55C>G;c.172C>G;c.451C>G	p.Pro151Ala;p.Pro19Ala;p.Pro58Ala;p.Pro151Ala	17p13.1	missense_variant	MODERATE	0.00%
P	1	chr17	7577121	G	A	*TP53*	AD	c.817C>T		17p13.1	structural_interaction_variant	HIGH	0.00%
P	1	chr17	7578190	T	C	*TP53*	AD	c.659A>G		17p13.1	structural_interaction_variant	HIGH	0.00%
P	1	chr17	7577106	G	C	*TP53*	AD	c.832C>G		17p13.1	structural_interaction_variant	HIGH	0.00%
P	1	chr17	7579329	T	C	*TP53*	AD	c.358A>G		17p13.1	structural_interaction_variant	HIGH	0.00%
P	3	chr17	7577538	C	T	*TP53*	AD	c.743G>A		17p13.1	protein_protein_contact	HIGH	0.03%
P	1	chr17	7577082	C	T	*TP53*	AD	c.856G>A		17p13.1	structural_interaction_variant	HIGH	0.00%
P	1	chr17	7577121	G	T	*TP53*	AD	c.817C>A		17p13.1	structural_interaction_variant	HIGH	0.01%
P	1	chr17	7577120	C	T	*TP53*	AD	c.818G>A		17p13.1	structural_interaction_variant	HIGH	0.10%
P	1	chr17	7578406	C	T	*TP53*	AD	c.524G>A		17p13.1	structural_interaction_variant	HIGH	0.00%
LP	1	chr17	7577057	TC	T	*TP53*	AD	c.880delG;c.880delG;c.880delG;c.880delG;c.880delG;c.484delG	p.Glu294fs;p.Glu294fs;p.Glu294fs;p.Glu294fs;p.Glu294fs;p.Glu162fs	17p13.1	frameshift_variant	HIGH	0.00%

P	1	chr17	7574021	C	A	*TP53*	AD	c.1006G>T;c.1006G>T	p.Glu336*;p.Glu336*	17p13.1	stop_gained	HIGH	0.00%
LP	1	chr17	7577035	T	TG	*TP53*	AD	c.902dupC;c.902dupC;c.902dupC;c.902dupC;c.902dupC;c.506dupC	p.Gly302fs;p.Gly302fs;p.Gly302fs;p.Gly302fs;p.Gly302fs;p.Gly170fs	17p13.1	frameshift_variant	HIGH	0.00%
P	1	chr17	7579340	G	GAATGCAA	*TP53*	AD	c.340_346dupTTGCATT	p.Ser116fs	17p13.1	frameshift_variant;structural_interaction_variant	HIGH	0.00%
LP	1	chr17	7578490	AC	A	*TP53*	AD	c.439delG;c.439delG;c.439delG;c.439delG;c.439delG;c.439delG;c.43delG;c.160delG;c.439delG;c.418delG;c.439	p.Val147fs;p.Val147fs;p.Val147fs;p.Val147fs;p.Val147fs;p.Val147fs;p.Val15fs;p.Val54fs;p.Val147fs;p.Val140fs;;;;;;;;;;;;;;;;;;;;	17p13.1	frameshift_variant;structural_interaction_variant	HIGH	0.00%

								delG;c.439delG;c.439delG;c.439delG					
P	2	chr17	7578263	G	A	*TP53*	AD	c.586C>T	p.Arg196*	17p13.1	structural_interaction_variant;stop_gained	HIGH	0.00%
P	1	chr17	7578463	CGGGTGCCGGGCGG	C	*TP53*	AD	c.454_466delCCGCCCGGCACCC	p.Pro152fs	17p13.1	frameshift_variant;structural_interaction_variant	HIGH	0.00%
P	2	chr17	7577035	TG	T	*TP53*	AD	c.902delC;c.902delC;c.902delC;c.902delC;c.902delC;c.506delC	p.Pro301fs;p.Pro301fs;p.Pro301fs;p.Pro301fs;p.Pro301fs;p.Pro169fs	17p13.1	frameshift_variant	HIGH	0.00%
P	1	chr17	7590694	C	A	*TP53*	AD	c.-29+1G>T;c.-29+1G>T;c.-29+1G>T;c.-26+1G>T;c.-22+1G>T;c.-29+1G>T;n.111+1G>T;c.-29+1G>T;c.-132+1G>T	17p13.1	splice_donor_variant&intron_variant	HIGH	0.00%	
LP	1	chr17	7577497	A	C	*TP53*	AD	c.782+2T>G;c.782+2T>G;c.782+2T>G;n.664+2	17p13.1	splice_donor_variant&intron_variant	HIGH	0.00%	

								T>G;n.664+2T>G;n.664+2T>G;c.782+2T>G;c.782+2T>G;c.782+2T>G;c.386+2T>G					
P	1	chr17	7590694	C	G	*TP53*	AD	c.-29+1G>C;c.-29+1G>C;c.-29+1G>C;c.-26+1G>C;c.-22+1G>C;c.-29+1G>C;n.111+1G>C;c.-29+1G>C;c.-132+1G>C	17p13.1	splice_donor_variant&intron_variant	HIGH	0.00%	
P	1	chr17	7577498	C	T	*TP53*	AD	c.782+1G>A;c.782+1G>A;c.782+1G>A;n.664+1G>A;n.664+1G>A;n.664+1G>A;c.782+1G>A;c.782+1G>A;c.782+1G>A;c.386+1G>A	17p13.1	splice_donor_variant&intron_variant	HIGH	0.00%	
P	1	chr17	7577018	C	T	*TP53*	AD	c.919+1G>A;c.919+1G>A;n.801+1	17p13.1	splice_donor_variant	HIGH	0.00%	

								G>A;n.801+1G>A;n.801+1G>A;c.919+1G>A;c.919+1G>A;c.919+1G>A;c.523+1G>A		&intron_variant			
LP	1	chr17	7578372	A	T	*TP53*	AD	c.558T>A;c.558T>A;c.558T>A;c.558T>A;c.558T>A;c.558T>A;c.162T>A;c.279T>A	p.Asp186Glu;p.Asp186Glu;p.Asp186Glu;p.Asp186Glu;p.Asp186Glu;p.Asp186Glu;p.Asp54Glu;p.Asp93Glu	17p13.1	missense_variant&splice_region_variant	MODERATE	0.01%
P	1	chr17	7577557	A	G	*TP53*	AD	c.724T>C;c.724T>C;c.724T>C;c.724T>C;c.724T>C;c.724T>C;c.328T>C;c.445T>C	p.Cys242Arg;p.Cys242Arg;p.Cys242Arg;p.Cys242Arg;p.Cys242Arg;p.Cys242Arg;p.Cys110Arg;p.Cys149Arg	17p13.1	missense_variant	MODERATE	0.00%
LP	1	chr17	7576562	C	T	*TP53*	AD	c.1016G>A	p.Cys339Tyr	17p13.1	missense_variant	MODERATE	0.00%
LP	1	chr17	7576897	G	T	*TP53*	AD	c.949C>A;c.949C>A;c.949C>A;c.949C>A;c.949C>A;c.7C>A;c.553C>A	p.Gln317Lys;p.Gln317Lys;p.Gln317Lys;p.Gln317Lys;p.Gln317Lys;p.Gln3Lys;p.Gln185Lys	17p13.1	missense_variant	MODERATE	0.00%
LP	1	chr17	7576872	C	A	*TP53*	AD	c.974G>T;c.974G>T;c.974G>T;c.974G>T;c.974G>T;c.974	p.Gly325Val;p.Gly325Val;p.Gly325Val;p.Gly325Val;p.Gly325Val;p.	17p13.1	missense_variant	MODERATE	0.00%

								G>T;c.32G>T;c.578G>T	Gly11Val;p.Gly193Val				
LP	2	chr17	7569531	T	C	*TP53*	AD	c.1025A>G	p.His342Arg	17p13.1	missense_variant	MODERATE	0.00%
LP	1	chr17	7574032	A	C	*TP53*	AD	c.995T>G;	p.Ile332Ser	17p13.1	missense_variant&splice_region_variant	MODERATE	0.00%
P	1	chr17	7578286	A	G	*TP53*	AD	c.563T>C;c.563T>C;c.563T>C;c.563T>C;c.563T>C;c.563T>C;c.167T>C;c.284T>C	p.Leu188Pro;p.Leu188Pro;p.Leu188Pro;p.Leu188Pro;p.Leu188Pro;p.Leu188Pro;p.Leu56Pro;p.Leu95Pro	17p13.1	missense_variant	MODERATE	0.00%
LP	1	chr17	7578376	C	T	*TP53*	AD	c.554G>A;c.554G>A;c.554G>A;c.554G>A;c.554G>A;c.554G>A;c.158G>A;c.275G>A	p.Ser185Asn;p.Ser185Asn;p.Ser185Asn;p.Ser185Asn;p.Ser185Asn;p.Ser185Asn;p.Ser53Asn;p.Ser92Asn	17p13.1	missense_variant	MODERATE	0.02%
LP	1	chr17	7577541	T	C	*TP53*	AD	c.740A>G		17p13.1	protein_protein_contact	HIGH	0.00%
LP	1	chr17	7577577	T	C	*TP53*	AD	c.704A>G		17p13.1	structural_interaction_variant	HIGH	0.09%
LP	1	chr17	7578484	GAATCAA	G	*TP53*	AD	c.440_445delTTGATT		17p13.1	structural_interaction_variant	HIGH	0.00%
LP	1	chr17	7578531	C	T	*TP53*	AD	c.3G>A;c.399G>A	p.Met1?	17p13.1	structural_interaction_variant;start_lost	HIGH	0.00%

LP	1	chr17	57181695	AC	A	*TRIM37*	AR	c.81delG;c.81delG;c.81delG;c.81delG;c.78delG	p.Cys28fs;p.Cys28fs;p.Cys28fs;p.Cys28fs;p.Cys27fs;p.Cys28fs	17q22	frameshift_variant;structural_interaction_variant	HIGH	0.00%
LP	1	chr19	852396	G	C	*ELANE*	AD	c.67+1G>C		19p13.3	splice_donor_variant&intron_variant	HIGH	0.00%
LP	3	chr19	45855906	G	A	*ERCC2*	AR	c.1904C>T;c.1670C>T;c.1832C>T	p.Ala635Val;p.Ala557Val;p.Ala611Val	19q13.32	missense_variant&splice_region_variant	MODERATE	0.40%
LP	1	chr19	45855828	G	C	*ERCC2*	AR	c.1982C>G;c.1748C>G;c.1910C>G	p.Ala661Gly;p.Ala583Gly;p.Ala637Gly	19q13.32	missense_variant	MODERATE	0.00%
LP	1	chr19	45868349	C	T	*ERCC2*	AR	c.428G>A;c.356G>A;c.356G>A;c.356G>A;c.356G>A;c.278G>A	p.Arg143Gln;p.Arg119Gln;p.Arg119Gln;p.Arg119Gln;p.Arg119Gln;p.Arg93Gln	19q13.32	missense_variant	MODERATE	0.05%
LP	1	chr19	45867721	G	A	*ERCC2*	AR	c.679C>T;c.445C>T;c.607C>T	p.Arg227Cys;p.Arg149Cys;p.Arg203Cys	19q13.32	missense_variant	MODERATE	0.11%
LP	1	chr19	45860761	G	A	*ERCC2*	AR	c.1348C>T;c.1114C>T;c.1276C>T;c.469C>T	p.Arg450Cys;p.Arg372Cys;p.Arg426Cys;p.Arg157Cys	19q13.32	missense_variant	MODERATE	0.00%
LP	1	chr19	45860760	C	T	*ERCC2*	AR	c.1349G>A;c.1115G>A;c.1277G>A;c.470G>A	p.Arg450His;p.Arg372His;p.Arg426His;p.Arg157His	19q13.32	missense_variant	MODERATE	0.04%
LP	1	chr19	45856397	C	T	*ERCC2*	AR	c.1775G>A;c.1541G>A;c.1703G>A	p.Arg592His;p.Arg514His;p.Arg568His	19q13.32	missense_variant	MODERATE	0.09%

LP	1	chr19	45856059	C	G	*ERCC2*	AR	c.1847G>C;c.1613G>C;c.1775G>C	p.Arg616Pro;p.Arg538Pro;p.Arg592Pro	19q13.32	missense_variant	MODERATE	0.02%
LP	2	chr19	45867291	G	A	*ERCC2*	AR	c.902C>T;c.668C>T;c.830C>T;c.23C>T;c.830C>T;c.830C>T;c.830C>T	p.Thr301Met;p.Thr223Met;p.Thr277Met;p.Thr8Met;p.Thr277Met;p.Thr277Met;p.Thr277Met	19q13.32	missense_variant	MODERATE	0.00%
LP	1	chr19	45873449	T	C	*ERCC2*	AR	c.47A>G	p.Tyr16Cys	19q13.32	missense_variant	MODERATE	0.43%
LP	1	chr19	45858047	C	T	*ERCC2*	AR	c.1606G>A;c.1372G>A;c.1534G>A	p.Val536Met;p.Val458Met;p.Val512Met	19q13.32	missense_variant	MODERATE	0.05%
LP	1	chr19	45855803	CCT	C	*ERCC2*	AR	c.2005_2006delAG;c.1771_1772delAG;c.1933_1934delAG	p.Arg669fs;p.Arg591fs;p.Arg645fs	19q13.32	frameshift_variant	HIGH	0.01%
LP	1	chr19	45860730	A	G	*ERCC2*	AR	c.1377+2T>C;n.1465+2T>C;n.548+2T>C;c.1143+2T>C;c.1305+2T>C;c.498+2T>C	19q13.32	splice_donor_variant&intron_variant	HIGH	0.00%	
LP	1	chr19	45873795	TCATGGCGCC	T	*ERCC2*	AR	c.-6_3delGGCGCCATG	p.Met1del	19q13.32	start_lost&splice_region_variant&conservative_inframe_deletion	HIGH	0.00%
LP	1	chr19	50921180	AC	A	*POLD1*	AD	c.3383delC;c.3305delC;c.3305delC	p.Pro1128fs;p.Pro1102fs;p.Pro1102fs	19q13.33	frameshift_variant	HIGH	0.02%
LP	1	chr19	42364900	C	T	*RPS19*	AD	c.56C>T	p.Ala19Val	19q13.2	missense_variant	MODERATE	0.00%

LP	1	chr20	62293748	G	C	*RTEL1*	AD/AR	c.396-1G>C		20q13.33	splice_acceptor_variant&intron_variant	HIGH	0.00%
LP	1	chr20	62327208	G	T	*RTEL1*	AD/AR	c.3899+1G>T		20q13.33	splice_donor_variant&intron_variant	HIGH	0.12%
LP	1	chr22	29130600	C	CT	*CHEK2*	AD	c.109_110insA;c.139_140insA	p.Gly37fs;p.Gly47fs	22q12.1	frameshift_variant	HIGH	0.00%
P	1	chr22	29107974	C	A	*CHEK2*	AD	c.844G>T;c.715G>T;c.715G>T;c.442G>T;c.52G>T;c.715G>T;c.715G>T;c.715G>T;c.715G>T;c.715G>T;c.715G>T;c.442G>T;c.514G>T;c.52G>T;c.808G>T	p.Glu282*;p.Glu239*;p.Glu239*;p.Glu148*;p.Glu18*;p.Glu239*;p.Glu239*;p.Glu239*;p.Glu239*;p.Glu239*;p.Glu148*;p.Glu172*;p.Glu18*;p.Glu270*	22q12.1	stop_gained	HIGH	0.00%
P	1	chr22	29121230	C	T	*CHEK2*	AD	c.573+1G>A;c.444+1G>A;c.444+1G>A;c.444+1G>A;c.-334+1G>A;c.444+1G>A;c.444+1G>A;c.444+1G>A;c.444+1G>A;c.444+1G>A;c.444+1G>A;c.444+1G>A;c.444+1G>A;c.444+1G>A;	22q12.1	splice_donor_variant&intron_variant	HIGH	0.03%	

								c.444+1G>A;c.-220+1G>A;c.6+1G>A;c.537+1G>A;c.*424+1G>A;c.474+1G>A					
LP	2	chr22	29107974	C	T	*CHEK2*	AD	c.844G>A;c.715G>A;c.715G>A;c.442G>A;c.52G>A;c.715G>A;c.715G>A;c.715G>A;c.715G>A;c.715G>A;c.442G>A;c.514G>A;c.52G>A;c.808G>A	p.Glu282Lys;p.Glu239Lys;p.Glu239Lys;p.Glu148Lys;p.Glu18Lys;p.Glu239Lys;p.Glu239Lys;p.Glu239Lys;p.Glu239Lys;p.Glu148Lys;p.Glu172Lys;p.Glu18Lys;p.Glu270Lys	22q12.1	missense_variant	MODERATE	0.03%
LP	1	chr22	21346078	TC	T	*LZTR1*	AD	c.955delC;c.898delC	p.Gln319fs;p.Gln300fs	22q11.21	frameshift_variant	HIGH	0.00%
LP	1	chr22	21349014	AAG	A	*LZTR1*	AD	c.1784_1785delAG;c.1727_1728delAG;c.96_97delAG;c.65_66delAG	p.Lys595fs;p.Lys576fs;p.Gly33fs;p.Lys22fs	22q11.21	frameshift_variant&splice_region_variant	HIGH	0.00%
P	1	chr22	30035077	A	C	*NF2*	AD	c.241-2A>C		22q12.2	splice_acceptor_variant&intron_variant	HIGH	0.00%
LP	1	chr22	30067870	C	T	*NF2*	AD	c.1055C>T;c.932C>T;c.1055C>T;c.806C>T;c.8	p.Thr352Met;p.Thr311Met;p.Thr352Met;p.Thr269Met;p.	22q12.2	missense_variant	MODERATE	0.00%

								06C>T;c.1055C>T;c.929C>T;c.1055C>T	Thr269Met;p.Thr352Met;p.Thr310Met;p.Thr352Met				
P	1	chrX	154002944	C	T	*DKC1*	XLR	c.1223C>T;c.584C>T	p.Thr408Ile;p.Thr195Ile	Xq28	missense_variant	MODERATE	0.00%
LP	1	chrX	132730460	T	C	*GPC3*	XLR	c.768A>G	p.Ter256Trpext*?	Xq26.2	stop_lost	HIGH	0.00%
LP	2	chrX	48547812	T	C	*WAS*	XLR	c.1442T>C	p.Ile481Thr	Xp11.23	missense_variant	MODERATE	0.00%

P = pathogenic, LP = likely pathogenic, VUS = variant of uncertain significance, LB = likely benign.
AD, autosomal dominant; AR, autosomal recessive; XLR, X-linked recessive; UNK, unknown inheritance. DM, disease-causing mutation.
DM, disease-causing mutation.
Gene inher., gene inheritance.

eTable 6. Prevalence of Potentially Pathogenic Variants in 238 Cancer-Susceptibility Genes in 1244 Patients With Osteosarcoma Compared With 28 235 Individuals Without Cancer

238 genes: inheritance*	**1,244 Osteosarcoma cases**						**28,235 Cancer-free controls**			
	1,004 cases, combined discovery: NCI WES		100 cases, replication 1: WES data		140 cases, replication 2: targeted sequencing		1,062 in-house cancer-free controls: NCI WES		27,173 ExAC NFE: WES data	
	N	% of cases	N	% of cases	N	% of cases	N	% of controls	N	% of controls
AD, AD/AR	185	18.4%	16	16.0%	24	17.1%	56	5.3%	1491	5.5%
AR	92	9.2%	10	10.0%	14	10.0%	72	6.8%	1041	3.8%
XLR	4	0.4%	2	2.0%	0	0.0%	0	0.0%	7	0.03%
Total prevalence		**28.0%**		**28.0%**		**27.1%**		**12.1%**		**9.3%**
95% CI		22.7-33.2		11.4-44.6		13.0-41.3		6.4-17.7		8.2-10.5

Abbreviations: AD, autosomal dominant; AR, autosomal recessive; XLR, X-linked recessive; WES, whole exome sequencing; P, pathogenic; LP, likely pathogenic; NFE, non-Finnish European.

*, unknown inheritance P/LP variants (N=3) are included with AD, AD/AR.

eTable 7. Top Statistically Significant Cancer-Susceptibility Genes With a Higher Burden of Variants

Higher burden of variance in European (EUR) patients with osteosarcoma (n = 732) and European in-house individuals in the control group (n = 994) for the specified burden test, subsequently compared with individuals of non-Finnish European ancestry in the ExAC group (n = 27 173).

Genes evaluated, Burden test	Top genes	Inheritance	P_{burden}	N, EUR cases	% of cases	N, EUR controls	% of controls	N, ExAC NFE	% ExAC NFE	$P^{\ddagger}$	OR*	95% CI
Cancer-susceptibility genes												
Pathogenic variants (P, LP)												
	TP53†	AD	**3.2E-8**	30	4.1%	3	0.3%	17	0.1%	**9.0E-44**	79.5	46.3 to 136.6
	CDKN2A	AD	3.1E-3	8	1.1%	0	0.0%	0	0.0%	**2.2E-13**	637.6	36.8 to 11057.9
	MEN1	AD	0.020	4	0.5%	0	0.0%	0	0.0%	**4.7E-7**	335.7	18.1 to 6241.3
	VHL	AD	0.020	4	0.5%	0	0.0%	2	0.0%	**3.3E-7**	74.6	13.6 to 408.1
	POT1	AD/AR	0.020	4	0.5%	0	0.0%	7	0.0%	**4.5E-5**	21.3	6.2 to 73.0
	RECQL4	AR	0.021	7	1.0%	1	0.1%	50	0.2%	4.9E-4	5.2	2.4 to 11.6
	FAH	AR	0.043	3	0.4%	0	0.0%	33	0.1%	0.061	3.4	1.0 to 11.1
	APC	AD	0.043	3	0.4%	0	0.0%	3	0.0%	**8.2E-5**	37.3	7.5 to 184.9
	MSH2	AD/AR	0.043	3	0.4%	0	0.0%	6	0.0%	6.2E-4	18.6	4.6 to 74.6
Damaging variants (P, LP, VUS_D)												
	RB1	AD	0.008	9	1.2%	2	0.2%	43	0.2%	**3.5E-6**	7.8	3.8 to 16.2
	MRE11A	AD/AR	0.009	7	1.0%	1	0.1%	110	0.4%	0.031	2.4	1.1 to 5.1
	RECQL4	AR	0.010	8	1.1%	1	0.1%	71	0.3%	8.1E-4	4.2	2.0 to 8.8
	VHL	AD	0.020	6	0.8%	1	0.1%	24	0.1%	**5.8E-5**	9.3	3.8 to 22.9
	MEN1	AD	0.042	5	0.7%	1	0.1%	57	0.2%	0.020	3.3	1.3 to 8.2
	CEP57	AR	0.043	3	0.4%	0	0.0%	5	0.0%	3.7E-4	22.4	5.3 to 93.7
	FGFR2	AD	0.043	3	0.4%	0	0.0%	54	0.2%	0.180	2.1	0.6 to 6.6
All rare variants												
	ERCC4	AR	5.0E-4	57	7.8%	37	3.7%	819	3.0%	**1.7E-10**	2.7	2.0 to 3.6

	GLI3	AD	0.008	39	5.3%	28	2.8%	1124	4.1%	0.114	1.3	0.9 to 1.8
	PHOX2B	AD	0.014	23	3.1%	12	1.2%	64	0.2%	**1.5E-18**	13.7	8.5 to 22.3
	WT1	AD	0.035	21	2.9%	14	1.4%	196	0.7%	**1.7E-7**	4.1	2.6 to 6.4
	BRAF	AD	0.045	15	2.0%	6	0.6%	84	0.3%	**1.7E-8**	6.7	3.9 to 11.7

Abbreviations: AD, autosomal dominant; AR, autosomal recessive; P, pathogenic; LP, likely pathogenic; VUS_D = variant of uncertain significance (VUS) *in silico* predicted damaging; NFE, non-Finnish European; N, number of individuals with the specified rare variants.

†, *TP53* was the top most significant gene in all burden analyses, it is shown only once.

‡, exact binomial test P value, tests if the proportion of cases with variants is the same as observed in the ExAC NFE controls.

Bolded values remains significant at 0.0002 if a Bonferroni correction for multiple tests is used.

*, EUR cases vs. ExAC NFE, odds of cases carrying a mutation in the specified gene.

eTable 8. Characteristics of Individuals With *TP53* Pathogenic/Likely Pathogenic Variants and All Pathogenic/Likely Pathogenic Cancer-Susceptibility Gene Variants in 1004 Patients With Osteosarcoma in the Discovery Set

Variable[†]		N Individuals Evaluable	No *TP53* P/LP variant		*TP53* P/LP variant		*P*	No P/LP variant		Any P/LP variant[€]		*P*	2 or more P/LP variants		*P*
			N	N %	N	N %		N	N %	N	N %		N	N %	
Age at dx, mean (SD)		974	16.5 (10)		14.7 (6)		0.155	16.9 (10)		15.3 (7)		0.015	15.6 (7)		0.327
Age group (years)	0-10	151	142	94.0%	9	6.0%		107	70.9%	44	29.1%		6	4.0%	
	11-20	698	672	96.3%	26	3.7%		521	74.6%	177	25.4%		25	3.6%	
	21-30	67	62	92.5%	5	7.5%		48	71.6%	19	28.4%		5	7.5%	
	31-40	26	25	96.2%	1	3.8%		19	73.1%	7	26.9%		1	3.8%	
	41+	32	32	100%	0	0.0%	0.316	30	93.8%	2	6.3%	0.107	0	0.0%	0.396
Gender	Male	540	519	96.1%	21	3.9%		397	73.5%	143	26.5%		25	4.6%	
	Female	462	440	95.2%	22	4.8%	0.490	346	74.9%	116	25.1%	0.620	13	2.8%	0.165
Ancestry [¥]	EUR	732	702	95.9%	30	4.1%		544	74.3%	188	25.7%		24	3.3%	
	AFR	54	49	90.7%	5	9.3%		40	74.1%	14	25.9%		3	5.6%	
	ADM	73	71	97.3%	2	2.7%		60	82.2%	13	17.8%		2	2.7%	
	ASN	3	3	100%	0	0.0%		3	100%	0	0.0%		0	0.0%	
	His	142	136	95.8%	6	4.2%	0.422	97	68.3%	45	31.7%	0.196	9	6.3%	0.480
AFR vs. non-AFR ancestry	non-AFR	950	912	96.0%	38	4.0%		704	74.1%	246	25.9%		35	3.7%	
	AFR	54	49	90.7%	5	9.3%	0.060	40	74.1%	14	25.9%	0.990	3	5.6%	0.387
Osteosarcoma location	Lower long bones	746	719	96.4%	27	3.6%		562	75.3%	184	24.7%		28	3.8%	
	Lower short bones	7	7	100.0%	0	0.0%		4	57.1%	3	42.9%		0	0.0%	
	Upper long bones	91	86	94.5%	5	5.5%		66	72.5%	25	27.5%		2	2.2%	
	Upper short bones	2	2	100%	0	0.0%		2	100%	0	0.0%		0	0.0%	
	Face or skull	2	2	100%	0	0.0%		2	100%	0	0.0%		0	0.0%	
	Mandible	2	2	100%	0	0.0%		1	50.0%	1	50.0%		0	0.0%	
	Chest region	10	8	80.0%	2	20.0%		6	60.0%	4	40.0%		0	0.0%	
	Pelvic region	31	27	87.1%	4	12.9%		21	67.7%	10	32.3%		4	12.9%	
	Soft tissue	5	4	80.0%	1	20.0%		3	60.0%	2	40.0%		0	0.0%	
	Vertebral column	1	1	100%	0	0.0%	0.086	1	100%	0	0.0%	0.740	0	0.0%	0.346
	Extremity	846	814	96.2%	32	3.8%		634	74.9%	212	25.1%		30	3.5%	

Axial vs. Extremity location	Axial	51	44	86.3%	7	13.7%	0.001	34	66.7%	17	33.3%	0.188	4	7.8%	0.309
Metastases at dx	No	382	365	95.5%	17	4.5%		277	72.5%	105	27.5%		11	2.9%	
	Yes	138	126	91.3%	12	8.7%	0.060	95	68.8%	43	31.2%	0.413	9	6.5%	0.089
Relapse	No	231	218	94.4%	13	5.6%		170	73.6%	61	26.4%		10	4.3%	
	Yes	136	126	92.6%	10	7.4%		97	71.3%	39	28.7%		5	3.7%	
	Progression	4	4	100%	0	0.0%	0.703	3	75.0%	1	25.0%	0.890	0	0.0%	0.817
Percent necrosis at surgery	Poor, <90%	140	133	95.0%	7	5.0%		99	70.7%	41	29.3%		5	3.6%	
	Good, >90%	134	130	97.0%	4	3.0%	0.396	92	68.7%	42	31.3%	0.711	6	4.5%	0.814
Conventional vs. Surface subtype	Conv.	342	327	95.6%	15	4.4%		238	69.6%	104	30.4%		14	4.1%	
	Surface	22	21	95.5%	1	4.5%	0.970	19	86.4%	3	13.6%	0.094	0	0.0%	0.491

dx, diagnosis; EUR, European ancestry; AFR, African ancestry; ADM, admixed; HIS, Hispanic; ASN, Asian; P, pathogenic; LP, likely pathogenic.
P values for the difference between cases with the specified P/LP variants and cases without these variants using a Chi-Square test.
[†]Not all cases had all variable data, counts (% of total) are given for the cases with these data.
[€]Includes cases with all pathogenic/likely pathogenic variants, and both AD and AR inheritance gene variants.
[¥]Ancestry based on GWAS data.
[§]Based on the presence of a death date or last known alive date.

eTable 9. Pathways Significantly Enriched for 101 Cancer-Susceptibility Genes With 1 or More Pathogenic/Likely Pathogenic Variants in 1004 Patients With Osteosarcoma

Pathway Source	Pathway Name	P-value*	q-value†	N genes
Reactome	DNA Repair	3.38E-28	2.77E-25	32
KEGG	Fanconi anemia pathway	1.30E-18	1.07E-15	14
Reactome	Resolution of D-loop Structures through Synthesis-Dependent Strand Annealing (SDSA)	1.72E-15	1.41E-12	10
KEGG	Pathways in cancer	1.21E-14	9.89E-12	23
Reactome	Resolution of D-Loop Structures	4.04E-14	3.32E-11	10
Reactome	HDR through Homologous Recombination (HRR)	6.88E-14	5.64E-11	12
Reactome	Transcriptional Regulation by TP53	2.00E-13	1.64E-10	21
KEGG	MicroRNAs in cancer	2.89E-13	2.37E-10	15
Reactome	TP53 Regulates Transcription of DNA Repair Genes	1.08E-12	8.84E-10	11
Reactome	DNA Double-Strand Break Repair	1.30E-12	1.07E-09	15
Reactome	Resolution of D-loop Structures through Holliday Junction Intermediates	1.74E-12	1.43E-09	9
KEGG	Homologous recombination	9.24E-12	7.58E-09	9
Reactome	HDR through Homologous Recombination (HR) or Single Strand Annealing (SSA)	1.50E-11	1.23E-08	13
Reactome	Homologous DNA Pairing and Strand Exchange	1.91E-11	1.56E-08	9
Reactome	Diseases of Mismatch Repair (MMR)	2.17E-11	1.78E-08	5
Reactome	Homology Directed Repair	2.66E-11	2.18E-08	13
KEGG	Endometrial cancer	1.48E-10	1.21E-07	9
Reactome	Fanconi Anemia Pathway	3.87E-10	3.17E-07	14
Reactome	Presynaptic phase of homologous DNA pairing and strand exchange	3.87E-10	3.17E-07	8
KEGG	Colorectal cancer	7.68E-10	6.30E-07	9
Reactome	Mismatch Repair	7.33E-10	6.01E-07	6

KEGG	Central carbon metabolism in cancer	1.57E-09	1.29E-06	9
Reactome	Cell Cycle	6.36E-09	5.21E-06	21
Reactome	HDR through Single Strand Annealing (SSA)	9.35E-09	7.66E-06	7
KEGG	Breast cancer	1.15E-08	9.44E-06	11
KEGG	Mismatch repair	1.41E-08	1.15E-05	6
KEGG	Prostate cancer	2.04E-08	1.67E-05	9
KEGG	Glioma	3.06E-08	2.51E-05	8
Reactome	Generic Transcription Pathway	4.20E-08	3.44E-05	24
Reactome	Disease	4.64E-08	3.81E-05	23
Reactome	Mismatch repair (MMR) directed by MSH2:MSH3 (MutSbeta)	4.11E-08	3.37E-05	5
Reactome	Mismatch repair (MMR) directed by MSH2:MSH6 (MutSalpha)	4.11E-08	3.37E-05	5
KEGG	Nucleotide excision repair	4.60E-08	3.77E-05	7
KEGG	Melanoma	5.49E-08	4.50E-05	8
Reactome	DNA Damage/Telomere Stress Induced Senescence	1.28E-07	1.05E-04	8
Reactome	Telomere Maintenance	1.56E-07	1.28E-04	8
KEGG	Non-small cell lung cancer	1.87E-07	1.54E-04	7
Reactome	Meiosis	2.40E-07	1.97E-04	9

*Fisher exact test.

†Shown for pathways significant at 0.0002 after Bonferroni correction for multiple tests.

eTable 10. Number of Rare Variants by Pathogenicity Score in 1004 Patients With Osteosarcoma and 1062 Individuals Without Cancer for 238 Cancer-Susceptibility Genes and Pathogenic/Likely Pathogenic Variants in 240 Patients in the Replication Set

Gene	Inheritance	P		LP		VUS_D[†]		VUS_ND		LB		B		Summary of all P + LP Variants					
		N Cases	N Ctrls	N Cases	N Ctrls	N Cases	N Ctrls	N Cases	N Ctrls	N Cases	N Ctrls	N Cases	N Ctrls	N Disc. Cases	% Disc. Cases	N Rep. Cases	% Rep. Cases	N Ctrls	% Ctrls
Autosomal Dominant (AD)																			
AIP	AD							10	19	1	1	0	1						
ALK	AD					0	5	38	30	6	4	4	2						
ANKRD26	AD					2	1	41	34	17	14	10	3						
APC	AD			3	0	11	11	34	41	22	22	16	16	3	0.3				
ATG2B	AD					3	1	47	51										
ATR	AD					1	3	36	36	8	14								
AXIN2	AD			1	0	2	0	19	18	18	19	0	1	1	0.1				
BAP1	AD					0	1	9	5	1	1								
BARD1	AD			0	1	2	2	6	15	10	10	5	6					1	0.1
BMPR1A	AD					0	1	3	7	8	5								
BRAF	AD							12	5	0	1								
BRCA1	AD	4	0			3	2	9	16	22	10	30	21	4	0.4	1	0.4		
CBL	AD					4	3	5	9	8	11								
CDC73	AD							1	1										
CDH1	AD					4	1	10	9	11	11	17	16						
CDK4	AD					1	1	10	11	1	2								

CDKN1C	AD							4	0			11	0						
CDKN2A	AD	1	0	11	0	1	4	2	6	12	7			12	1.2				
CDKN2B	AD					0	1	5	9										
CEBPA	AD							7	1										
CHEK2	AD	2	0	3	1	2	4	16	17	4	5	2	1	5	0.5			1	0.1
CREBBP	AD			1	0	4	3	30	24	7	15	9	20	1	0.1				
CTR9	AD							11	19										
CXCR4	AD							2	2	1	0								
CYLD	AD			1	0	0	2	6	4	2	3			1	0.1				
DDB2	AD							3	7	12	4								
DDX41	AD					3	4	9	11	2	2								
DICER1	AD			1	0	1	0	18	10	20	18	6	1	1	0.1				
EGFR	AD			5	0	0	3	18	24	7	3	2	1	5	0.5	1	0.4		
ELANE	AD			1	0	1	0	5	2	1	2			1	0.1				
EP300	AD					5	4	34	38	22	28	13	7						
EPCAM	AD					2	2	10	3	2	4	10	19			1	0.4		
ETV6	AD							5	5										
EXT1	AD			0	1	1	3	7	0	1	3	1	0					1	0.1
EXT2	AD			1	1	7	7	5	8	11	10			1	0.1			1	0.1
EZH2	AD			2	1	1	1	8	7	3	1			2	0.2			1	0.1
FAS	AD							5	12										

FGFR2	AD			1	0	2	0	7	7	3	6	1	0	1	0.1				
FGFR3	AD					5	6	6	12	6	6	13	14						
FLCN	AD					4	4	10	7	14	16					1	0.4		
FOXE1	AD							27	24										
GALNT12	AD			1	0	3	2	5	8	6	5			1	0.1	1	0.4		
GALNT14	AD					2	0	16	20	9	9								
GATA2	AD					8	4	2	1	3	2								
GJB2	AD					11	8	8	23	10	6								
GLI3	AD					3	1	21	13	32	21								
GREM1	AD							2	1	7	5								
GSKIP	AD					0	2	0	2										
HABP2	AD					6	7	20	28	1	0	3	1						
HMBS	AD			1	0	6	2	9	11	9	3			1	0.1				
HNF1A	AD			1	1	8	8	24	17	1	5			1	0.1			1	0.1
HOXB13	AD					1	1	8	6										
HRAS	AD					0	3	0	2	0	2	10	11						
IPMK	AD					0	2	9	11										
KIT	AD					0	2	5	11	11	12	5	2						
KMT2D	AD			0	1	9	6	67	60	124	96	4	8					1	0.1
KRAS	AD			1	0	1	1	1	4	3	2	0	2	1	0.1				
LMO1	AD					2	2	1	2										

LZTR1	AD			2	0	5	3	17	13	1	1			2	0.2				
MAP2K1	AD							4	0	1	1					1	0.4		
MAP2K2	AD					3	3	2	2	2	6	4	1						
MAX	AD							2	7	3	3								
MC1R	AD			1	0	4	5	32	24	11	8	1	0	1	0.1				
MEN1	AD	1	0	4	0	1	1	4	1	3	0	7	7	5	0.5	1	0.4		
MET	AD			1	0	0	7	18	26	20	19	1	2	1	0.1				
MITF	AD					2	1	23	12							1	0.4		
MTAP	AD			0	1	5	2	7	6	3	3							1	0.1
NF1	AD			1	0	1	3	29	24	13	18	10	14	1	0.1	2	0.8		
NF2	AD	1	0	1	0	5	3	5	8	3	0	2	3	2	0.2				
NFIX	AD							1	4	1	1								
NKX2-1	AD					1	0	9	5	1	0								
NOTCH3	AD					12	18	43	38	11	7								
NRAS	AD							3	2	2	4								
NSD1	AD					2	2	45	25	19	9	6	8						
NTHL1	AD					3	2	17	21	3	0								
PAX5	AD					2	2	6	5	2	4								
PAX6	AD					0	1			1	5								
PBRM1	AD					4	1	20	27	1	0								
PDGFRA	AD					0	4	24	29	14	15	1	1						

PDGFRB	AD			0	1	2	1	25	20	13	5	7	10					1	0.1
PHOX2B	AD					0	1	7	6	17	6	5	1						
PMS1	AD			1	1	5	5	37	37					1	0.1			1	0.1
POLD1	AD			1	0	0	1	22	30	12	10	9	13	1	0.1				
PPOX	AD					2	1	6	9	1	1								
PRKAR1A	AD			2	0	1	0	4	13	2	4			2	0.2				
PRSS1	AD					2	7	18	27			0	1						
PTCH1	AD			2	0	7	5	19	13	12	18	7	6	2	0.2				
PTCH2	AD					8	8	13	6	5	8	8	7						
PTEN	AD			1	0	1	1	2	2	0	2			1	0.1				
PTPN11	AD	0	1			1	2	1	7	5	2							1	0.1
RAD51D	AD	2	0			0	1	9	12	8	9	5	1	2	0.2				
RAF1	AD			1	0	3	1	10	22	2	3	2	0	1	0.1				
RB1	AD	2	0	2	0	7	2	9	6	10	9	1	1	4	0.4				
REST	AD			0	1	0	1	15	32	4	4	6	12					1	0.1
RET	AD					9	5	15	12	13	10	1	0						
RHBDF2	AD					0	0	24	17	6	6								
RPL11	AD					0	1	2	1	1	0								
RPL15	AD					1	0	1	5										
RPL26	AD					0	1	1	0										
RPL27	AD							3	1										

RPL31	AD							2	6										
RPL35A	AD			1	0			0	1	2	0			1	0.1				
RPL5	AD			1	1			6	1	4	5			1	0.1	1	0.4	1	0.1
RPS10	AD					0	2	2	1	0	1								
RPS19	AD			1	0			14	11					1	0.1				
RPS24	AD							6	6	1	4								
RPS26	AD					0	1			1	0								
RPS27	AD							2	1										
RPS29	AD					1	2	1	0										
RPS7	AD			1	0	1	1	1	1					1	0.1				
RUNX1	AD					0	2	4	2	3	2					1	0.4		
SAMD9	AD			1	2	5	5	35	38					1	0.1			2	0.2
SAMD9L	AD					3	4	23	25										
SDHAF2	AD					0	1	0	1	2	1								
SDHB	AD					2	4	0	1	1	0								
SDHC	AD					1	0	6	12	1	1								
SDHD	AD					0	2	5	3										
SH2B3	AD					12	4	24	18										
SHOC2	AD							4	2			2	0						
SMAD4	AD							3	2	8	1								
SMARCA4	AD					2	3	17	12	7	1								

SMARCB1	AD							1	1	2	4	5	0						
SMARCE1	AD							18	17										
SMO	AD			6	3	0	3	38	20					6	0.6			3	0.3
SOS1	AD			2	0	0	1	7	12	5	10	7	10	2	0.2				
SOS2	AD					2	0	11	11	3	2	3	2						
SRP72	AD							6	10	12	6								
STAT3	AD					0	1	12	14	2	0								
STK11	AD					0	1	35	34	5	7	1	2						
SUFU	AD			0	1	1	0	8	7	7	9	1	0					1	0.1
T	AD					1	0	5	7	0	2								
TGFBR1	AD					2	4	1	4	10	8	5	7						
TGFBR2	AD					3	2	0	4	12	10	3	5						
TMEM127	AD					3	1	1	0	1	0								
TP53	AD	25	0	19	3	1	1	0	3	3	1			44	4.4	13	5.4	3	0.3
TP63	AD			6	2	1	2	7	3					6	0.6	2	0.8	2	0.2
TSC1	AD					2	0	7	7	10	16	16	18						
TSC2	AD			3	0	10	6	21	16	41	50	15	26	3	0.3	1	0.4		
TSHR	AD			0	1	1	7	22	20	0	1					1	0.4	1	0.1
VHL	AD	1	0	3	0	2	1	1	3			0	3	4	0.4				
WT1	AD							9	8	15	7								
ACD	AD/AR			1	0			11	22					1	0.1				

ATM	AD/AR	4	3	1	0	11	11	42	46	48	46	34	49	5	0.5	1	0.4	3	0.3
BRCA2	AD/AR	8	5	1	1	4	7	27	16	59	47	52	52	9	0.9	2	0.8	6	0.6
BRIP1	AD/AR	1	2	0	1	0	6	12	8	22	23	3	4	1	0.1			3	0.3
CDKN1B	AD/AR					2	3	6	13	2	3								
COL7A1	AD/AR	1	3	4	1	16	13	88	106	17	21			5	0.5			4	0.4
FANCM	AD/AR	1	0	4	2	1	3	44	38	15	22	1	0	5	0.5			2	0.2
FH	AD/AR	2	0			0	4	4	3	0	1			2	0.2				
MLH1	AD/AR	1	0	1	0	10	7	7	4	22	24	11	10	2	0.2				
MPL	AD/AR	4	1	1	2	0	3	7	8	2	2	0	1	5	0.5	3	1.3	3	0.3
MRE11A	AD/AR			1	0	8	1	5	13	7	3			1	0.1				
MSH2	AD/AR			4	0	3	9	5	3	7	11	20	18	4	0.4	1	0.4		
MSH6	AD/AR			1	1	5	8	23	11	9	10	7	7	1	0.1	2	0.8	1	0.1
NHP2	AD/AR			1	1	0	1	9	10	1	2			1	0.1			1	0.1
PALB2	AD/AR	3	1					20	13	12	11	8	7	3	0.3			1	0.1
PARN	AD/AR							33	55	2	1								
PMS2	AD/AR	1	1	1	0	6	7	18	17	12	14	7	7	2	0.2			1	0.1
POLE	AD/AR			4	2	1	1	53	58	12	12	5	1	4	0.4			2	0.2
POT1	AD/AR	1	0	5	0	0	2	7	3	8	12	2	0	6	0.6				
RAD51C	AD/AR					1	0	2	4	2	0	5	7						
RTEL1	AD/AR	0	3	2	0	3	1	72	52	11	16			2	0.2	1	0.4	3	0.3
SDHA	AD/AR	1	0	0	1	4	8	24	32	10	13			1	0.1			1	0.1

TERT	AD/AR			3	0	3	3	11	7	6	9			3	0.3				
TINF2	AD/AR			1	0	2	2	5	3	12	14			1	0.1				
UROD	AD/AR			0	1	1	1	3	4									1	0.1
AD/AR Total		67	20	135	37	368	395	2205	2237	1161	1093	468	477	202	20.1	39	16.3	57	5.4
Autosomal Recessive (AR)																			
ABCB11	AR	1	0	2	3	6	7	27	22	5	6	0	1	3	0.3	2	0.8	3	0.3
AGL	AR			2	1	11	6	24	24	4	7	8	13	2	0.2			1	0.1
APOBEC3B	AR					1	0	20	13	2	1								
BLM	AR	0	3			1	2	24	25	31	21	15	21					3	0.3
BUB1B	AR			1	1	2	3	38	35	1	0			1	0.1			1	0.1
CEP57	AR					4	0	10	12	6	0								
CTC1	AR	1	2	0	2	6	3	33	21	7	3	3	2	1	0.1			4	0.4
DHCR7	AR	1	1	5	2	5	1	33	28	4	5	1	1	6	0.6			3	0.3
DIS3L2	AR			4	1	0	2	35	41	4	5	3	3	4	0.4			1	0.1
DOCK8	AR	1	1			0	1	63	96	22	18	3	0	1	0.1			1	0.1
ERCC1	AR					3	7	3	7										
ERCC2	AR			17	6	0	1	14	19					17	1.7	3	1.3	6	0.6
ERCC3	AR	0	1	2	3	4	1	19	18					2	0.2			4	0.4
ERCC4	AR			2	0	7	6	47	30	4	2	6	4	2	0.2	1	0.4		
ERCC5	AR			1	4	0	1	62	54			2	8	1	0.1	2	0.8	4	0.4
FAH	AR	2	0	1	0	8	8	4	1					3	0.3	4	1.7		
FANCA	AR	0	4	3	2	11	10	48	71	6	14	4	0	3	0.3	5	2.1	6	0.6

FANCC	AR	1	1	1	0			5	8	5	7	1	3	2	0.2			1	0.1
FANCD2	AR	3	0	0	1			46	40	13	14			3	0.3			1	0.1
FANCE	AR	2	0	0	1			10	11	0	2	2	2	2	0.2			1	0.1
FANCF	AR							9	5	2	0	0	1						
FANCG	AR	0	1	1	1			12	7	3	5			1	0.1			2	0.2
FANCI	AR	0	1	0	2	5	8	20	19	7	10	14	9					3	0.3
FANCL	AR			1	4	1	0	18	12	6	13	1	0	1	0.1			4	0.4
G6PC	AR	1	0	3	5	0	1	9	9	0	1			4	0.4	1	0.4	5	0.5
GBA	AR	1	0	1	1	6	10	3	2	1	4			2	0.2	1	0.4	1	0.1
HFE	AR			1	0	7	5	5	2	2	0			1	0.1				
ITK	AR					5	3	10	12	1	1								
L2HGDH	AR					3	1	9	12	0	1								
MSH3	AR			1	0	7	8	15	19	21	13			1	0.1				
MUTYH	AR	4	1	0	1	7	16	18	11	12	7	0	4	4	0.4	1	0.4	2	0.2
NBN	AR	2	0	1	1	5	7	18	17	9	5	2	5	3	0.3			1	0.1
NTRK1	AR			1	0	9	7	20	23	6	15			1	0.1				
POLH	AR					1	2	11	18	12	15					1	0.4		
PRF1	AR	1	1	3	1	5	4	23	20	2	2	1	1	4	0.4			2	0.2
RECQL4	AR	3	1	5	0	2	0	49	65	22	17	16	12	8	0.8	2	0.8	1	0.1
SBDS	AR	2	1	0	1	10	14	1	1	3	7			2	0.2			2	0.2
SERPINA1	AR	1	0	4	7	10	12	13	12	3	1	3	6	5	0.5	1	0.4	7	0.7

SLC25A13	AR	1	0			3	4	10	11	1	0			1	0.1				
SLX4	AR					2	5	55	34	36	34	3	3						
TRIM37	AR	0	1	1	0	0	1	13	15	0	1			1	0.1			1	0.1
WRAP53	AR					1	2	12	11	1	0								
WRN	AR			5	4	4	1	25	29	13	18	34	47	5	0.5	3	1.3	4	0.4
XPA	AR					0	1	4	1	2	0								
XPC	AR	1	0	0	1			17	10	10	6			1	0.1			1	0.1
AR Total		29	20	69	56	162	171	964	953	289	281	122	146	98	9.8	27	11.3	76	7.2
X-linked/Y-linked																			
AR	X-linked					2	6	28	15	3	2	3	1						
DKC1	X-linked	1	0					23	12	0	1			1	0.1	1	0.4		
FANCB	X-linked							4	7	6	7	2	1						
FMR1	X-linked							3	1	0	1	4	6						
GATA1	X-linked							2	6	0	2	3	2						
GPC3	X-linked			1	0			17	10	1	2			1	0.1				
GPC4	X-linked							4	6										
PHF6	X-linked							2	1										
SH2D1A	X-linked							0	1	5	4								
SRY	Y-linked							1	0										
TSR2	X-linked							1	0										
WAS	X-linked			2	0	1	1	4	4	2	4			2	0.2	1	0.4		

X/Y-linked Total		1	0	3	0	3	7	89	63	17	23	12	10	4	0.4	2	0.8		
De Novo or Unknown																			
ASXL1	de novo or unk					0	4	24	16	24	25								
ERBB4	de novo or unk					2	2	15	20										
FAT1	de novo or unk					18	10	162	183	6	5	1	0						
GNAS	de novo or unk					3	3	42	45										
IDH1	de novo or unk			2	2	1	2	4	14	3	3			2	0.2	1	0.4	2	0.2
IDH2	de novo or unk					4	0	6	4	12	6								
IKZF1	de novo or unk					0	1	5	9	2	2	0	1						
PTPN13	de novo or unk					2	7	108	90	6	9	19	10						
RAD51	de novo or unk					2	1	4	3										
SETBP1	de novo or unk					1	1	32	35	4	15								
SMARCA2	de novo or unk			1	0	2	3	9	23	18	15			1	0.1				
SPOP	de novo or unk					1	0	3	0	0	2					1	0.4		
SPRTN	de novo or unk					0	1	5	6	0	1								

TFAP2A	de novo or unk			0	1	1	0	1	2	1	2					1	0.4	1	0.1
XRCC3	de novo or unk							7	5	1	0								
Unk Total		0	0	3	3	37	35	427	455	77	85	20	11	3	0.3	3	1.3	3	0.3
Grand Total*		**97**	**40**	**210**	**96**	**570**	**608**	**3685**	**3708**	**1544**	**1482**	**622**	**644**	**307**	**30.6**	**71**	**29.6**	**136**	**12.8**

Abbreviations: P, pathogenic; LP, likely pathogenic

†Rare variants of uncertain significance but predicted damaging based on in silico predictions (MetaSVM, REVEL, and CADD scores).

Disc, 1,004 discovery set cases; Rep., 240 total independent replication set cases.

*Overall pathogenic variant prevalence appears a little higher here due to some people carrying more than one pathogenic gene variant and counted more than once here.

eTable 11. Predicted Pathogenic and Likely Pathogenic Variants in 240 Patients in the Replication Set

Path. Score	No. Cases	Chr	Position	REF	ALT	Gene	Gene Inher.	HGVS.c	Effect	Impact	Classsic High Impact	Pop Max Freq	Sequencing
P	1	chr1	45798475	T		*MUTYH*	AR	c.536A>G;c.452A>G;c.485A>G;c.452A>G;c.455A>G;c.452A>G;c.527A>G; c.497A>G;c.494A>G;c.485A>G;c.494A>G;c.68A>G;c.485A>G	Missense variant	ODERATE		0.30%	rgeted
P	1	chr1	43803600	T	A	*MPL*	AD/AR	c.79+2T>A;c.79+2T>A	HIGH		%	Targeted	
P	1	chr1	43804396	G	C	*MPL*	AD/AR	c.391+5G>C;c.391+5G>C	LOW		%	Targeted	
LP	1	chr1	93299353	T	G	*RPL5*	AD	c.190T>G	Stop lost & splice region variant	IGH		0.00%	rgeted
P	1	chr1	155205634	T	C	*GBA*	AR	c.1226A>G;c.1226A>G;c.1079A>G;c.887A>G;c.965A>G	Missense variant & splice region variant	ODERATE		0.36%	ES
P	1	chr1	43804305	G	C	*MPL*	AD/AR	c.305G>C;c.305G>C	Missense variant	ODERATE		0.10%	ES
LP	1	chr2	169853219	C	T	*ABCB11*	AR	c.403G>A	Missense variant	ODERATE		0.01%	rgeted
P	1	chr2	48026250	AAAGAG	A	*MSH6*	AD/AR	c.1135_1139delAGAGA;c.745_749delAGAGA;c.229_233delAGAGA	Frameshift variant	IGH		0.00%	rgeted
LP	1	chr2	169783792	CA	C	*ABCB11*	AR	c.3491delT	Frameshift variant	IGH		0.00%	rgeted
P	1	chr2	209108190	T	C	*IDH1*	UNK	c.659A>G	Structural interaction variant; missense variant	IGH		0.08%	rgeted
P	1	chr2	47637389	CTG	C	*MSH2*	AD/AR	c.528_529delTG;c.330_331delTG;c.330_331delTG;c.528_529delTG; c.528_529delTG;c.528_529delTG	Frameshift variant structural interaction variant	IGH		0.00%	ES
P	1	chr2	48023116	G	T	*MSH6*	AD/AR	c.541G>T;c.244G>T;c.244G>T;c.244G>T	Stop gained	IGH		0.00%	ES
LP	1	chr2	47596772	G	GCGGCCCCCGGCCCT	*EPCAM*	AD	c.138_151dupGCCCTCGGCCCCCG	Frameshift variant	IGH		0.00%	ES
LP	1	chr3	70008546	A	G	*MITF*	AD	c.1154A>G;c.1136A>G;c.980A>G;c.1133A>G;c.1088A>G;c.1061A>G; c.815A>G;c.833A>G;c.647A>G	Missense variant	ODERATE		0.01%	ES
LP	1	chr3	189587125	A	G	*TP63*	AD	c.1142A>G;c.887A>G;c.1142A>G;c.1142A>G;c.1142A>G;c.1130A>G; c.860A>G;c.860A>G;c.860A>G;c.860A>G;c.605A>G;c.848A>G	Missense variant	ODERATE		0.00%	ES
LP	1	chr3	189607139	G	A	*TP63*	AD	c.1518G>A;c.1263G>A;c.1518G>A;c.1506G>A;c.1236G>A;c.1236G>A; c.981G>A;c.1224G>A	Missense variant	ODERATE		0.00%	ES
LP	1	chr6	10411828	ATCTGCGAAGAG	A	*TFAP2A*	UNK	c.-10_1delCTCTTCGCAGA;c.-10_1delCTCTTCGCAGA	Frameshift variant & start lost	IGH		0.00%	ES
LP	1	chr6	43550081	G	T	*POLH*	AR	c.25G>T	HIGH		%	WES	
LP	1	chr7	55270399	C	T	*EGFR*	AD	c.3217C>T	Stop gained	IGH		0.00%	ES
P	1	chr8	30938648	C	T	*WRN*	AR	c.1105C>T	Stop gained	IGH		0.03%	rgeted
P	1	chr8	31004567	A	G	*WRN*	AR	c.3384-2A>G;n.2017-2A>G	HIGH		%	Targeted	
P	1	chr8	30946482	G	A	*WRN*	AR	c.1652+1G>A;n.353+1G>A	HIGH		%	Targeted	
P	1	chr8	145738796	G	A	*RECQL4*	AR	c.2269C>T	Stop gained	IGH		0.04%	ES
P	1	chr8	145741453	CCT	C	*RECQL4*	AR	c.1048_1049delAG;c.568_569delAG	Frameshift variant	IGH		0.02%	ES
LP	1	chr9	101602374	G	T	*GALNT12*	AD	c.1303G>T	Stop gained	IGH		0.00%	ES
P	1	chr11	108121531	C	T	*ATM*	AD/AR	c.1339C>T;c.1339C>T;c.1339C>T	Stop gained	IGH		0.01%	rgeted
P	1	chr11	64572613	G	A	*MEN1*	AD	c.1258C>T;c.1138C>T;c.1243C>T	Structural interaction variant stop gained	IGH		0.00%	ES
P	1	chr13	32914437	GT	G	*BRCA2*	AD/AR	c.5946delT;c.5946delT	Frameshift variant	IGH		0.04%	rgeted
LP	1	chr13	103520471	C	T	*ERCC5*	AR	c.2542C>T;c.241C>T	Missense variant	ODERATE		0.03%	rgeted
LP	1	chr13	32944697	A	G	*BRCA2*	AD/AR	c.8487+3A>G;c.8487+3A>G	LOW		%	WES	
LP	1	chr13	103519117	C	T	*ERCC5*	AR	c.2455C>T;c.154C>T	Missense variant	ODERATE		0.00%	ES
LP	1	chr14	94849388	G	A	*SERPINA1*	AR	c.187C>T	Missense variant	ODERATE		0.30%	rgeted
P	1	chr14	81558873	A	G	*TSHR*	AD	c.468-2A>G;c.468-2A>G;c.468-2A>G;c.468-2A>G;c.468-2A>G	HIGH		%	Targeted	
P	3	chr15	80460605	G	T	*FAH*	AR	c.554-1G>T;c.554-1G>T;c.554-1G>T;c.344-1G>T;n.482-1G>T;n.100-1G>T	HIGH		%	Targeted	
LP	1	chr15	80472673	G	A	*FAH*	AR	c.*84+1G>A	Splice donor variant	IGH		0.14%	rgeted
LP	1	chr15	66781553	C	T	*MAP2K1*	AD	c.961C>T;c.433C>T	Missense variant & splice region variant	ODERATE		0.02%	ES
P	1	chr16	2134966	A	C	*TSC2*	AD	c.4508A>C;c.4163A>C;c.4307A>C;c.4379A>C;c.4199A>C;c.4439A>C; c.4340A>C;c.689A>C	Missense variant	ODERATE		0.00%	rgeted
P	1	chr16	89805118	T	C	*FANCA*	AR	c.4261-2A>G;c.4265-2A>G;c.481-2A>G;n.532-2A>G;c.635-2A>G	HIGH		%	Targeted	
LP	1	chr16	89882683	G	C	*FANCA*	AR	c.341C>G	Stop gained	IGH		0.08%	rgeted
LP	1	chr16	89865516	T	A	*FANCA*	AR	c.123A>T;c.951A>T	Stop lost	IGH		0.29%	rgeted

LP	1	chr16	89805692	AG	A	*FANCA*	AR	c.4015delC;c.4015delC;c.166delC;c.343delC;c.214delC	Frameshift variant	IGH		0.01%	ES
LP	1	chr16	14022000	A	C	*ERCC4*	AR	c.225A>C	Stop lost	IGH		0.01%	ES
LP	1	chr16	89829189	T	C	*FANCA*	AR	c.153A>G	Stop lost	IGH		0.00%	ES
LP	1	chr17	7574017	C	A	*TP53*	AD	c.1010G>T;c.1010G>T	Missense variant	IODERATE		0.00%	rgeted
LP	2	chr17	7577568	C	T	*TP53*	AD	c.713G>A;c.713G>A;c.713G>A;c.713G>A;c.713G>A;c.713G>A; c.713G>A;c.713G>A;c.713G>A;c.713G>A;c.713G>A;c.713G>A	Structural interaction variant	IGH		0.00%	ES
P	1	chr17	41219665	ATTAG	A	*BRCA1*	AD	c.5093_5096delCTAA;c.5030_5033delCTAA;c.1481_1484delCTAA; c.4142_4145delCTAA;c.1604_1607delCTAA;c.1718_1721delCTAA; c.503_506delCTAA;c.4889_4892delCTAA;c.1718_1721delCTAA; c.1343_1346delCTAA;c.1718_1721delCTAA;c.1580_1583delCTAA; c.5030_5033delCTAA	Frameshift variant	IGH		0.00%	rgeted
P	1	chr17	7577539	G	A	*TP53**	AD	c.742C>T;c.742C>T;c.742C>T;c.742C>T	Protein protein contact missense variant	IGH		0.00%	rgeted
P	1	chr17	7574003	G	A	*TP53**	AD	c.1024C>T;c.1024C>T;c.1024C>T;c.1024C>T	Stop gained	IGH		0.00%	rgeted
P	1	chr17	7577022	G	A	*TP53**	AD	c.916C>T;c.916C>T;c.916C>T;c.916C>T;c.916C>T;c.520C>T	Stop gained	IGH		0.00%	rgeted
P	1	chr17	7577094	G	A	*TP53**	AD	c.844C>T	HIGH	I	%	Targeted	
P	1	chr17	7577120	C	T	*TP53**	AD	c.818G>A	HIGH	I	%	Targeted	
P	1	chr17	7577548	C	T	*TP53**	AD	c.733G>A;c.733G>A	Structural interaction variant; missense variant	IGH		0.01%	rgeted
P	1	chr17	7578406	C	T	*TP53**	AD	c.524G>A	HIGH	I	%	Targeted	
LP	1	chr17	7578400	G	C	*TP53*	AD	c.530C>G;c.530C>G;c.530C>G;c.530C>G;c.530C>G;c.530C>G; c.134C>G;c.251C>G	Missense variant	IODERATE		0.01%	rgeted
LP	1	chr17	7579368	A	G	*TP53*†	AD	c.319T>C	HIGH	I	%	Targeted	
P	1	chr17	47684623	C	G	*SPOP*	UNK	c.826G>C	Structural interaction variant; missense variant	IGH		0.00%	rgeted
LP	1	chr17	7577102	C	T	*TP53*	AD	c.836G>A	HIGH	I	%	WES	
P	1	chr17	41061435	G	C	*G6PC*	AR	c.562G>C;c.485G>C	Missense variant & splice region variant	IODERATE		0.01%	ES
LP	1	chr17	17124819	C	T	*FLCN*	AD	c.903G>A	Stop gained	IGH		0.08%	ES
LP	2	chr17	29706042	G	A	*NF1*	AD	HIGH	Splice donor variant & intron variant	IGH		0.30%	ES
P	1	chr19	45855804	CT	C	*ERCC2*	AR	c.2005delA;c.1771delA;c.1933delA	Frameshift variant	IGH		0.00%	rgeted
LP	1	chr19	45855795	G	A	*ERCC2*	AR	c.2015C>T;c.1781C>T;c.1943C>T	Missense variant	IODERATE		0.02%	rgeted
LP	1	chr19	45860760	C	T	*ERCC2*	AR	c.1349G>A;c.1115G>A;c.1277G>A;c.470G>A	Missense variant	IODERATE		0.04%	rgeted
LP	1	chr20	62293801	C	T	*RTEL1*	AD/AR	c.448C>T	Stop gained	IGH		0.01%	rgeted
P	1	chr21	36252995	C	G	*RUNX1*	AD	c.286G>C;c.286G>C	Structural interaction variant; missense variant	IGH		0.01%	rgeted
LP	1	chrX	153991099	C	G	*DKC1*	XLR	c.-142C>G	5 prime UTR premature start codon gain variant	OW		0.13%	ES
LP	1	chrX	48547322	C	CACGAT	*WAS*	XLR	c.1205_1206insACGAT	Frameshift variant	IGH		0.00%	ES

P = pathogenic, LP = likely pathogenic, VUS = variant of uncertain significance, LB = likely benign.
AD, autosomal dominant; AR, autosomal recessive; XLR, X-linked recessive; UNK, unknown inheritance; DM, disease-causing mutation; WES, whole exome sequencing.
*TP53** variant at this position present in the IARC germline mutation TP53 database (R20) in Li-Fraumeni syndrome, Li-Fraumeni-like syndrome, or *TP53* Chompret classified families/individuals.
TP53† variant present in an osteosarcoma case in the discovery and replication set and also in an individual in the IARC germline mutation TP53 database (R20) with osteosarcoma.

eTable 12. Top Statistically Significant Candidate Genes With a Higher Burden of Variants

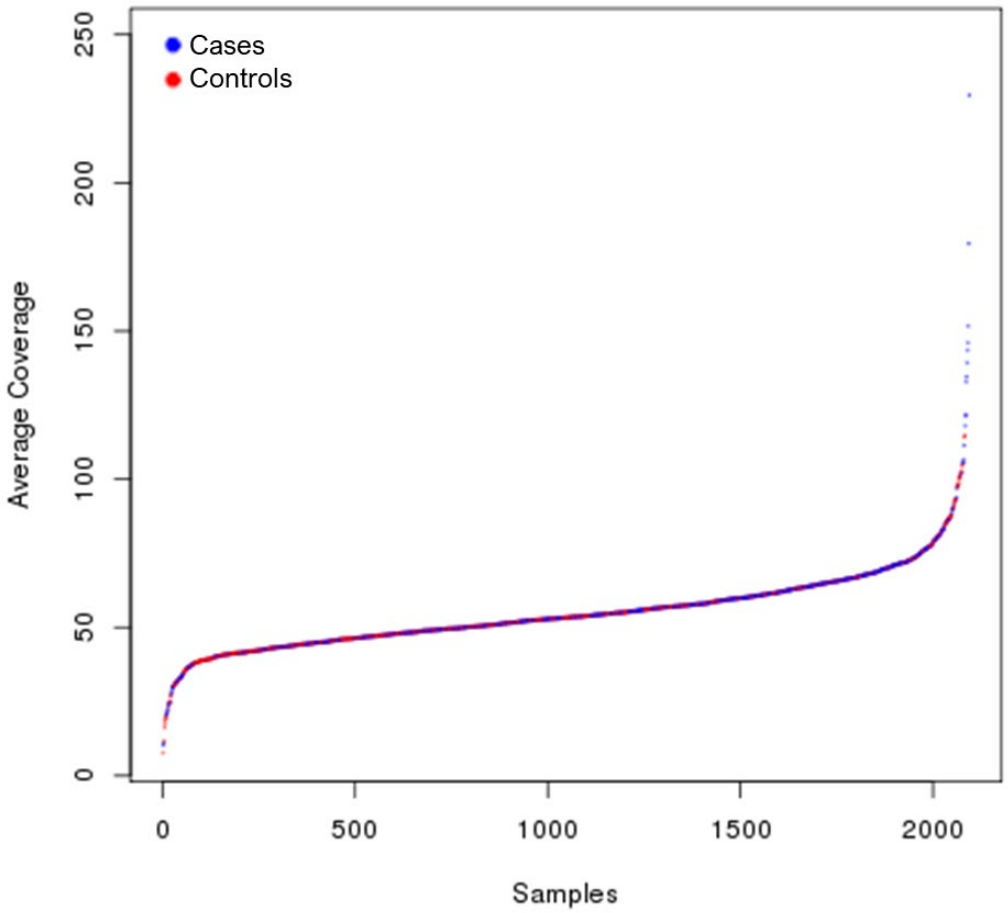
European patients (n = 732) compared with European individuals in the control group (n = 994) for the specified burden test.

Genes evaluated, Burden test	Top genes[†]	P_{burden}	N, EUR cases	% of cases	N, EUR controls	% of controls
Somatically altered genes						
Damaging variants						
	SUZ12	0.002	9	1.2%	1	0.1%
	MLLT4	0.004	10	1.4%	2	0.2%
	ALDH2	0.020	4	0.5%	0	0.0%
	THSD4	0.020	6	0.8%	1	0.1%
All rare variants						
	NACA	0.0006	99	13.5%	73	7.3%
	MNX1	0.0008	33	4.5%	10	1.0%
	HOXA13	0.0028	24	3.3%	9	0.9%
Candidate genes from previous studies						
Damaging variants						
	PRKCG	0.043	3	0.4%	0	0.0%
All rare variants						
	ZNF208	0.002	100	13.7%	73	7.3%
	NFE2L2	0.007	27	3.7%	16	1.6%
	CAMK2D	0.028	8	1.1%	2	0.2%
	ATRX	0.041	28	3.8%	18	1.8%
	CYP2C8	0.045	15	2.0%	9	0.9%

N, number of individuals with the specified rare variants.

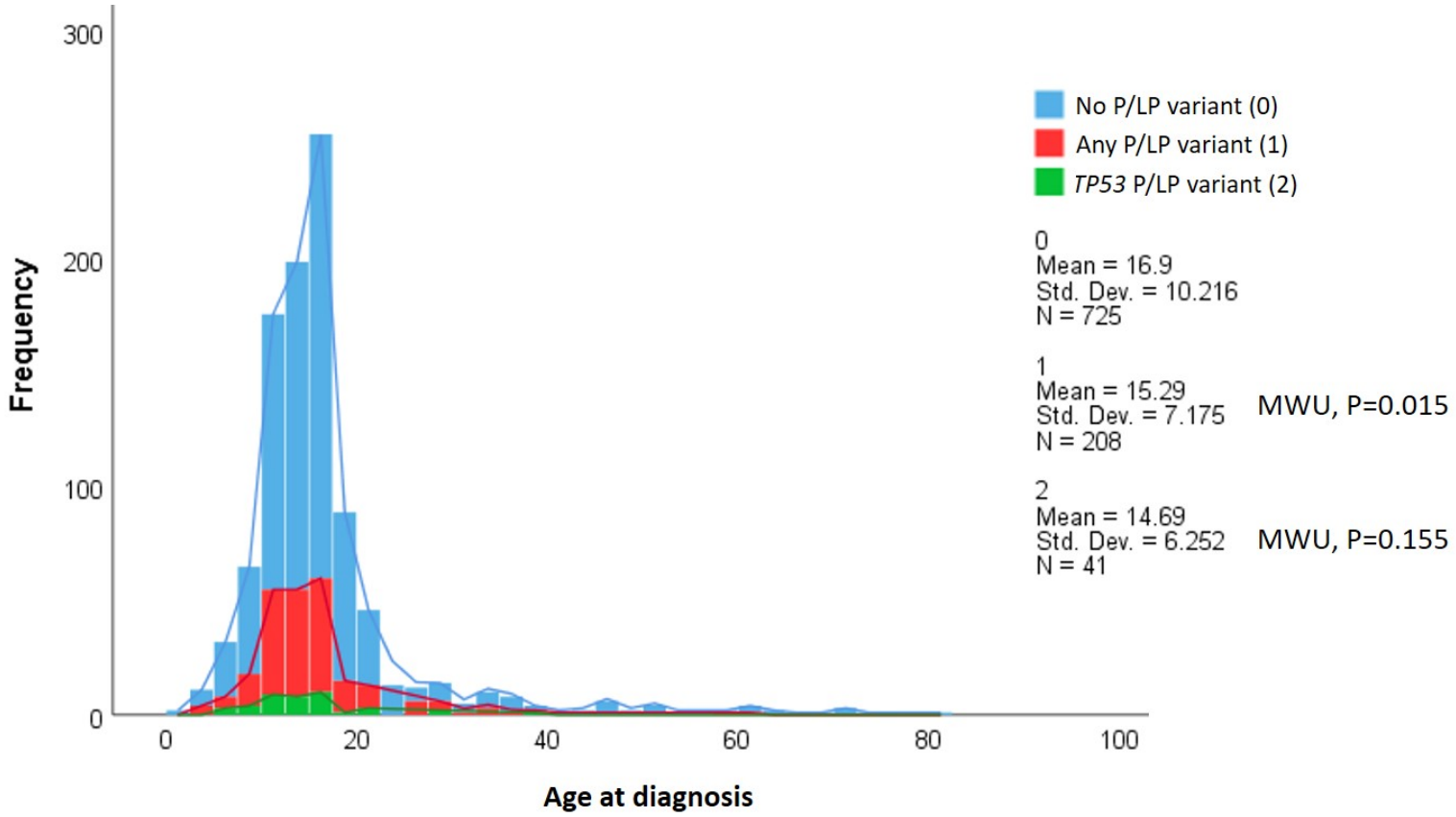
¥ only the top significant genes are shown.

eFigure 1. Mean Whole-Exome Sequencing Coverage per Sample



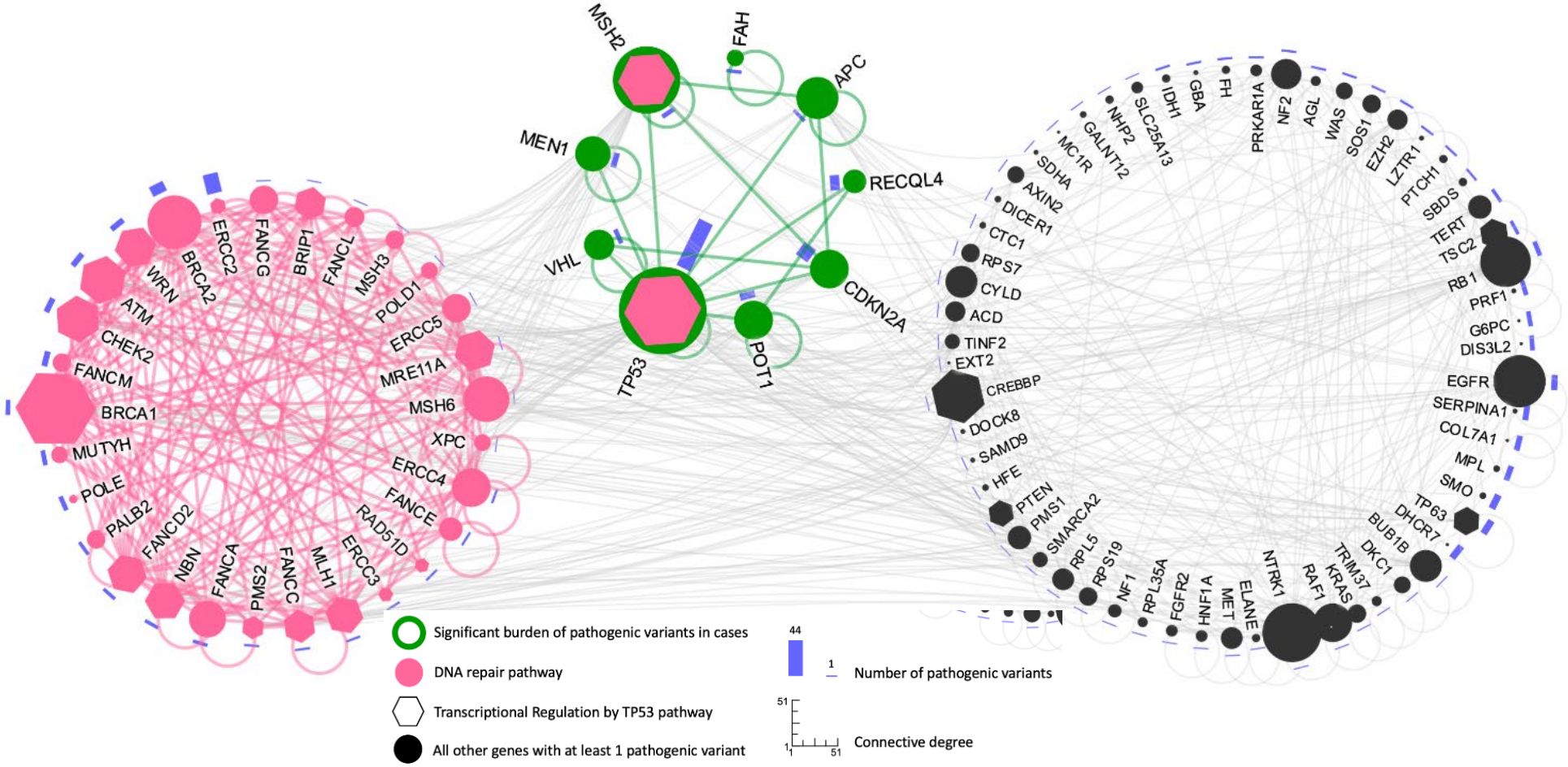
Coverage across the 238 cancer-susceptibility genes for the 1004 patients in the primary discovery set and 1062 individuals the control group. Depth of coverage was estimated after excluding the duplicate reads generated during sequencing. The mean coverage for all participant samples was 57.13 (standard deviation 16.16) and 53.18 for all control group samples (standard deviation 12.94).

eFigure 2. Frequency of Rare Pathogenic/Likely Pathogenic Variants in the *TP53* Gene and the Other Cancer-Susceptibility Genes



Frequency compared with individuals without a pathogenic/likely pathogenic variant by age at diagnosis. Mann-Whitney U (MWU) test *P* value for the distribution of age at osteosarcoma diagnosis for individuals with and without pathogenic/likely pathogenic variants for all cancer-susceptibility genes (1) and for *TP53* (2). Not all patients had age at diagnosis data. The all pathogenic/likely pathogenic variant (1) category in red excludes the individuals with *TP53* P/LP variants (2) shown in green.

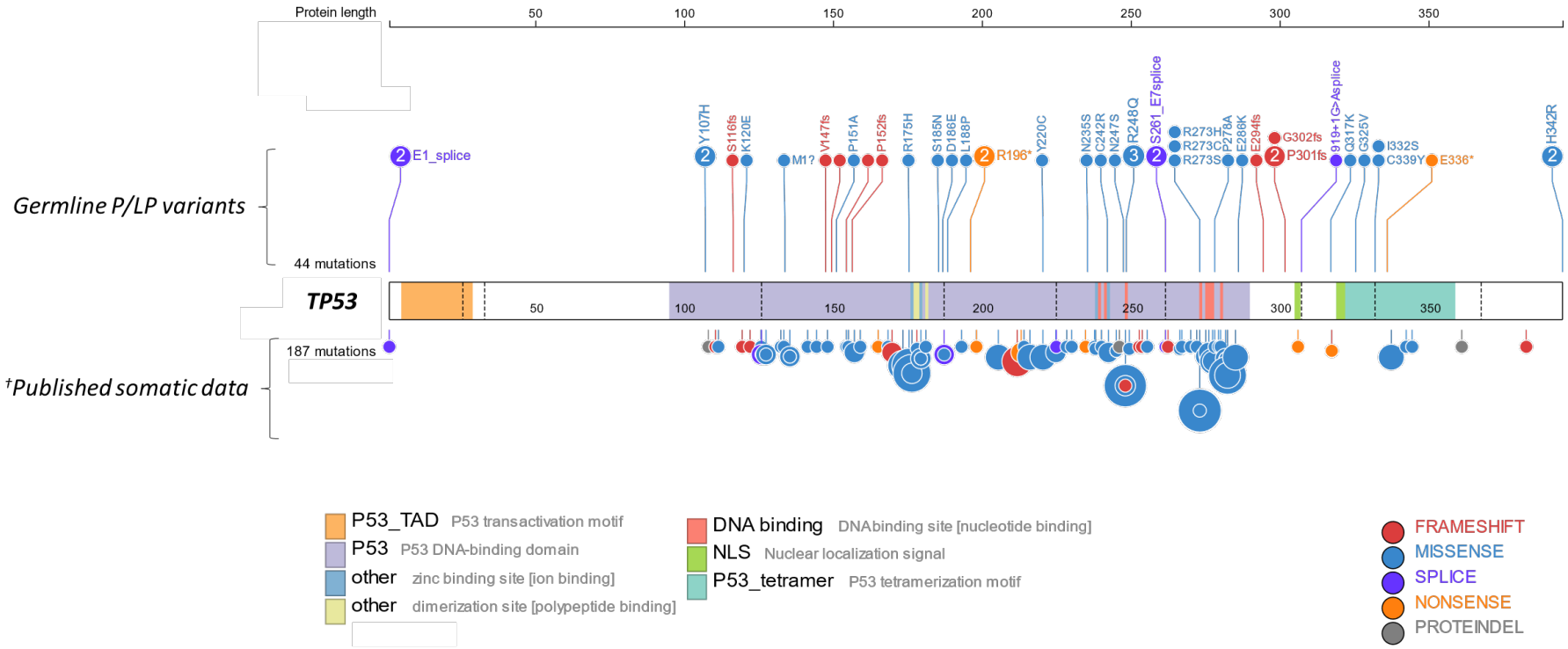
eFigure 3. Pathway Enrichment Illustrated With a Network Analysis and Protein-Protein Interactions for 101 Cancer-Susceptibility Genes With 1 or More Pathogenic/Likely Pathogenic Variant Identified in Discovery Set of 1004 Patients With Osteosarcoma



Node size represents the number of protein-protein interactions within the network, with a maximum of 51 and minimum of 1. Pathway enrichment analyses were performed using algorithms from the web tools, KOBAS 3.0 and PathDIP. Both algorithms used a hypergeometric approach to test pathways that are overrepresented given a gene set.

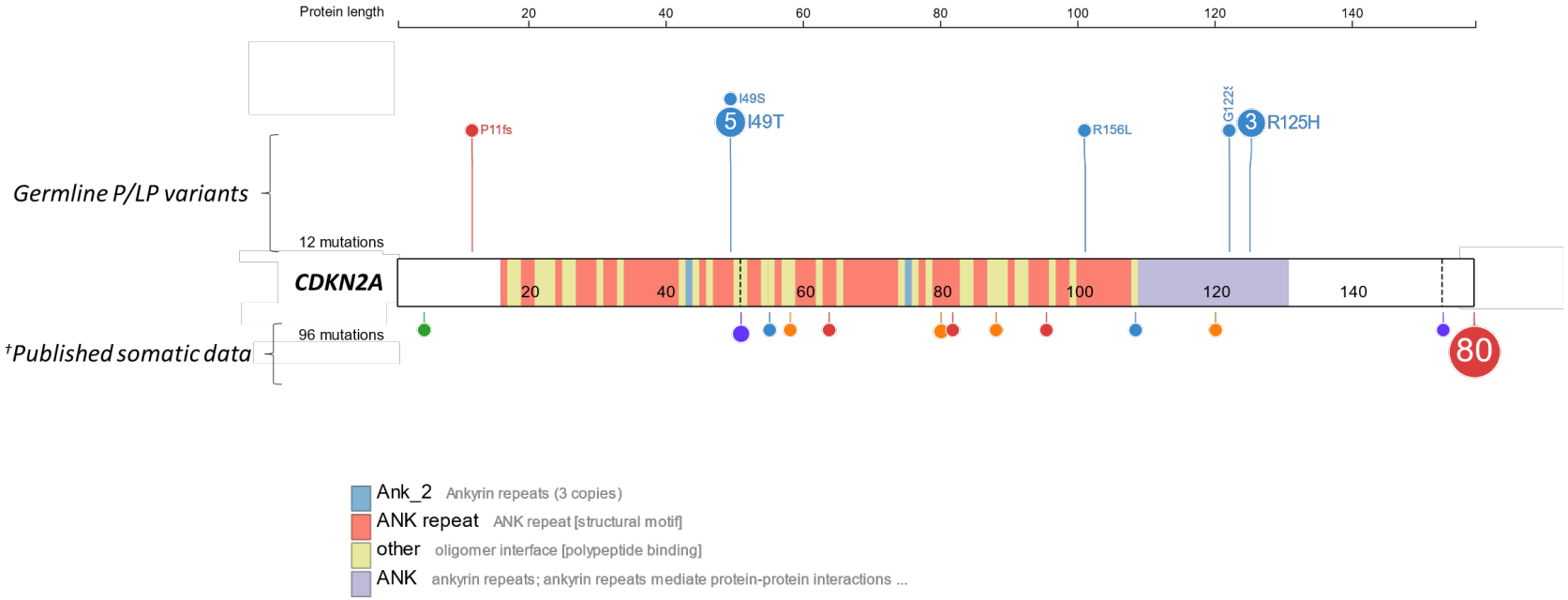
eFigure 4. Lollipop Plots Illustrating Location of Variants Within the Specified Genes That Contain a Significantly Increased Burden (Number) of Rare Pathogenic/Likely Pathogenic Variants in Patients Compared With the Control Group by Protein/Functional Domain

A. *TP53*



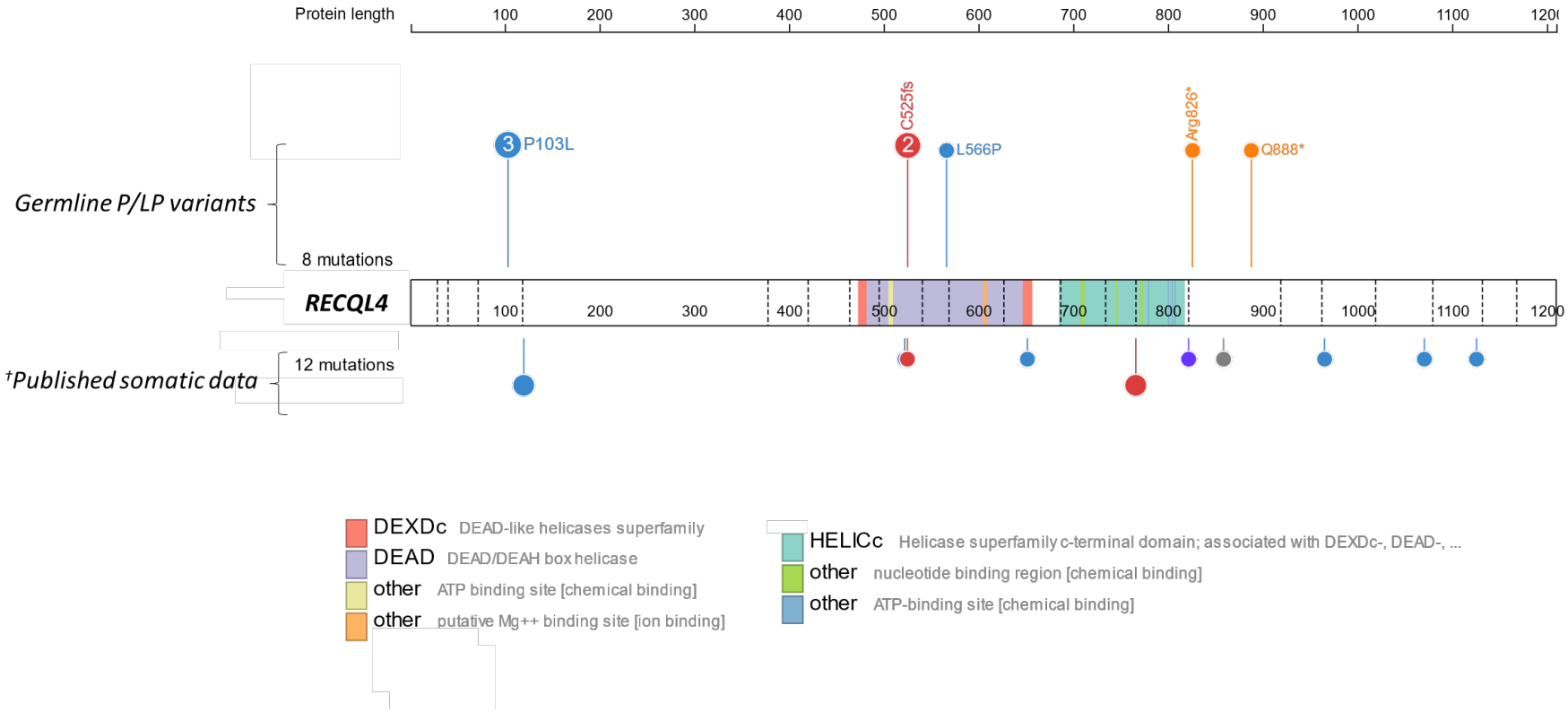
Pathogenic/likely pathogenic variants observed in the 1004 patients with osteosarcoma are shown above each plot. † Published somatic variants from the Therapeutically Applicable Research To Generate Effective Treatments (TARGET) osteosarcoma study and the Pediatric Cancer Genome Project (PCGP) osteosarcoma study and from COSMIC bone cancers visualized in the ProteinPaint genome browser. Published somatic variants from all the pediatric cancer data in TARGET and PCGP and from COSMIC bone cancers were retrieved with the ProteinPaint genome browser. The number of patients with a specific variant are shown within each circle if more than 1.

B. *CDKN2A*



Pathogenic/likely pathogenic variants observed in the 1004 patients with osteosarcoma are shown above each plot. † Published somatic variants from the Therapeutically Applicable Research To Generate Effective Treatments (TARGET) osteosarcoma study and the Pediatric Cancer Genome Project (PCGP) osteosarcoma study and from COSMIC bone cancers visualized in the ProteinPaint genome browser. Published somatic variants from all the pediatric cancer data in TARGET and PCGP and from COSMIC bone cancers were retrieved with the ProteinPaint genome browser. The number of patients with a specific variant are shown within each circle if more than 1.

C. *RECQL4*



Pathogenic/likely pathogenic variants observed in the 1004 patients with osteosarcoma are shown above each plot. † Published somatic variants from the Therapeutically Applicable Research To Generate Effective Treatments (TARGET) osteosarcoma study and the Pediatric Cancer Genome Project (PCGP) osteosarcoma study and from COSMIC bone cancers visualized in the ProteinPaint genome browser. Published somatic variants from all the pediatric cancer data in TARGET and PCGP and from COSMIC bone cancers were retrieved with the ProteinPaint genome browser. The number of patients with a specific variant are shown within each circle if more than 1.

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