Osteosarcoma, the most common malignant bone tumor in children and adolescents, occurs in a high number of cancer predisposition syndromes that are defined by highly penetrant germline mutations. The germline genetic susceptibility to osteosarcoma outside of familial cancer syndromes remains unclear.

OBJECTIVE To investigate the germline genetic architecture of 1244 patients with osteosarcoma.

DESIGN, SETTING, AND PARTICIPANTS Whole-exome sequencing (n = 1104) or targeted sequencing (n = 140) of the DNA of 1244 patients with osteosarcoma from 10 participating international centers or studies was conducted from April 21, 2014, to September 1, 2017. The results were compared with the DNA of 1062 individuals without cancer assembled internally from 4 participating studies who underwent comparable whole-exome sequencing and 27,173 individuals of non-Finnish European ancestry who were identified through the Exome Aggregation Consortium (ExAC) database. In the analysis, 238 high-interest cancer-susceptibility genes were assessed followed by testing of the mutational burden across 736 additional candidate genes. Principal component analyses were used to identify 732 European patients with osteosarcoma and 994 European individuals without cancer, with outliers removed for patient-control group comparisons. Patients were subsequently compared with individuals in the ExAC group. All data were analyzed from June 1, 2017, to July 1, 2019.

MAIN OUTCOMES AND MEASURES The frequency of rare pathogenic or likely pathogenic genetic variants.

RESULTS Among 1244 patients with osteosarcoma (mean [SD] age at diagnosis, 16 [8.9] years [range, 2-80 years]; 684 patients [55.0%] were male), an analysis restricted to individuals with European ancestry indicated a significantly higher pathogenic or likely pathogenic variant burden in 238 high-interest cancer-susceptibility genes among patients with osteosarcoma compared with the control group (732 vs 994, respectively; P = 1.3×10−18). A pathogenic or likely pathogenic cancer-susceptibility gene variant was identified in 281 of 1004 patients with osteosarcoma (28.0%), of which nearly three-quarters had a variant that mapped to an autosomal-dominant gene or a known osteosarcoma-associated cancer predisposition syndrome gene. The frequency of a pathogenic or likely pathogenic cancer-susceptibility gene variant was 128 of 1062 individuals (12.1%) in the control group and 2527 of 27,173 individuals (9.3%) in the ExAC group. A higher than expected frequency of pathogenic or likely pathogenic variants was observed in genes not previously linked to osteosarcoma (eg, CDKN2A, MEN1, VHL, POT1, APC, MSH2, and ATRX) and in the Li-Fraumeni syndrome-associated gene, TP53.

CONCLUSIONS AND RELEVANCE In this study, approximately one-fourth of patients with osteosarcoma unselected for family history had a highly penetrant germline mutation requiring additional follow-up analysis and possible genetic counseling with cascade testing.
The peak incidence of osteosarcoma (OMIM 259500) occurs during the pubertal growth spurt. Osteosarcoma risk factors include tall height, previous radiotherapy, and at least 8 established cancer predisposition syndromes, including autosomal-dominant disorders (Li-Fraumeni syndrome [OMIM 161431], hereditary retinoblastoma [OMIM 180200], and Bloom syndrome [OMIM 210900]). Candidate genes and genome-wide association studies suggest that common single-nucleotide polymorphisms are also associated with osteosarcoma, affirming a complex underlying architecture for its genetic etiology but one that appears to be weighted disproportionately toward rare variants.

An earlier study reported that 4% of patients with osteosarcoma younger than 30 years with an unknown family history of cancer carried a pathogenic germline variant of TP53 (OMIM 191170) that was known to be or highly likely to be associated with Li-Fraumeni syndrome; in addition, 6% of those patients carried rare likely pathogenic TP53 variants. A survey of 72 candidate genes across 1162 sarcomas, including 124 osteosarcomas, observed that 217 individuals (18.7%) had a pathogenic or likely pathogenic germline variant in autosomal-recessive or autosomal-dominant disorders (primarily DNA helicase disorders, such as Rothmund-Thomson syndrome [OMIM 268400], RAPADILINO syndrome [OMIM 266280], Werner syndrome [OMIM 277700], and Bloom syndrome [OMIM 210900]). Candidate genes and genome-wide association studies suggest that common single-nucleotide polymorphisms are also associated with osteosarcoma, confirming a complex underlying architecture for its genetic etiology but one that appears to be weighted disproportionately toward rare variants.

We used a 2-phase approach to evaluate rare germline variants in 1244 patients with osteosarcoma, beginning with assessment of 238 cancer-susceptibility genes followed by burden testing for an additional 736 candidate genes. We compared the frequency of pathogenic/likely pathogenic variants in patients with those of 1062 individuals without cancer (the control group), and for significant findings, with 27173 individuals of non-Finnish European ancestry who were identified through the Exome Aggregation Consortium (ExAC) database (the ExAC group; Figure 1).

### Methods

The NCI Retrospective Study of Genetic Risk Factors for Osteosarcoma was approved by the institutional review board.
Pathogenic Germline Variants in Cancer-Susceptibility Genes in Patients With Osteosarcoma

Paulo, Brazil; the Childhood Cancer Survivor Study, the Instituto de Oncologia Pediatrica, Grupode Apoio ao Adolescente e a Crianca com Cancer/Universidade Federal de Sao Paulo, Brazil; the Istituto Ortopedico Rizzoli, Bologna, Italy; and the Ankara Oncology Training and Research Hospital, Ankara, Turkey. eMethods and eTable 1 in the Supplement). Of those, 782 patients were previously reported in a genome-wide association study, which included 48 patients from the Instituto de Oncologia Pediatrica, Grupo de Apoio ao Adolescente e a Crianca com Cancer /Universidade Federal de Sao Paulo. A total of 462 additional patients were included, drawn from the Childhood Cancer Survivor Study, the Hospital Infantil Manuel De Jesus Rivera (Managua, Nicaragua), and from the Unidad Nacional de Oncologia Pediatrica, Guatemala City, Guatemala; the Royal National Orthopaedic Hospital NHS Trust and University College London Cancer Institute (Middlesex, United Kingdom); the Instituto Ortopedico Rizzoli, Bologna, Italy; and the Ankara Oncology Training and Research Hospital. The population frequency of pathogenic/likely pathogenic germline variants was estimated for 238 cancer-susceptibility genes using publicly available noncancer whole-exome sequencing data from the ExAC database. All analyses evaluated variants with minor allele frequencies of less than 0.01 that passed quality-control filters. For the patient replication sets, we used buccal sample DNA to conduct whole-exome sequencing on 100 patients with osteosarcoma and targeted sequencing of 238 cancer-susceptibility genes on an additional 140 patients at the University of Minnesota from August 1, 2017, to September 1, 2018 (eMethods in the Supplement).

**Genes and Variants**
We assembled a set of 238 cancer-susceptibility genes, including 114 cancer-predisposing genes, 38 14 genes associated with Diamond-Blackfan anemia, and 110 cancer-associated genes previously described or reported to have germline associations in the Catalogue of Somatic Mutations in Cancer (eTable 2 in the Supplement). These genes were grouped by mode of inheritance: 141 genes were autosomal-dominant, 45 were autosomal-recessive, 25 were autosomal-dominant and autosomal-recessive, 11 were X-linked, 1 was Y-linked, and 15 had de novo or unknown inheritance patterns (eTable 2 in the Supplement). An additional 736 candidate genes were evaluated, including 140 genes associated with osteosarcoma that were identified through the Human Genome Epidemiology (HuGE) phenopedia and manual curation of published reports and 596 genes somatically altered in pediatric bone cancers or recurrent in any pediatric cancer that were identified through the Catalogue of Somatic Mutations in Cancer and annotation of published osteosarcoma somatic data (eTable 3 in the Supplement).

A stepwise pipeline was constructed to evaluate each rare variant that passed quality-control filters in the genes of interest. Variants were classified as pathogenic, likely patho-
genic, of uncertain significance, likely benign, or benign based on previous reports and recommendations from the American College of Medical Genetics and Genomics and the Association for Molecular Pathology (eMethods and eTable 4 in the Supplement). An in silico prediction algorithm was also used to further filter the variant of uncertain significance category as damaging or not damaging (eMethods in the Supplement). All pathogenic/likely pathogenic variants are summarized in eTable 5 in the Supplement.

Statistical Analyses
We analyzed the 1004 patients with osteosarcoma in the discovery set, which included 732 patients of European ancestry, with the 1062 individuals in the control group, which included 994 patients of European ancestry (Figure 1; eMethods in the Supplement). The replication set consisted of 240 patients with osteosarcoma who had germline whole-exome sequencing or targeted sequencing data available, and we performed secondary patient comparisons with individuals in the ExAC group.

Rare-variant burden tests were conducted on the 732 European patients in the discovery set and the 994 European individuals in the control group using burden and optimal sequence kernel association tests. The comparisons between the patient group and the ExAC group were restricted to genes identified as substantially different between the primary discovery set of patients and the control group. Comparisons among individuals with and without pathogenic/likely pathogenic variants were performed using 2-sided χ² or Fisher exact tests for categorical variables and Mann-Whitney U tests for continuous variables (eg, age). We used 2-sided exact binomial tests and logistic regression models to compare the frequencies of pathogenic/likely pathogenic variants between patients and individuals in the ExAC group only for the selected genes identified as substantially different between the primary discovery set of patients and individuals in the control group who had comparable whole-exome sequencing performed at the National Cancer Institute. We compared overall survival between patients carrying pathogenic/likely pathogenic variants and individuals without pathogenic/likely pathogenic variants for all cancer-susceptibility genes and the TP53 gene using adjusted Cox proportional hazards regression models and estimated hazard ratios (HRs) and 95% CIs. All data were analyzed from June 1, 2017, to July 1, 2019.

Results
Among 1244 patients with osteosarcoma, the mean (SD) age at diagnosis was 16 (8.9) years (age range, 2-80 years), and 684 patients (55.0%) were male (eTable 1 in the Supplement). Our primary analyses were based on patients and individuals in the control group with whole-exome sequencing data jointly called that yielded comparable quality-control measures and coverage (Figure 1; eFigure 1 in the Supplement).

We assessed the frequency of pathogenic/likely pathogenic variants in 238 cancer-susceptibility genes in the discovery set of patients and the control group. Overall, 281 of 1004 patients with osteosarcoma (28.0%; 95% CI, 22.7%-33.2%) had a pathogenic/likely pathogenic variant in a gene of interest, which was significantly higher than the frequency observed in the control group (128 of 1062 individuals [12.1%]; 95% CI, 6.4%-17.7%; Fisher exact P = 1.3 × 10⁻¹⁸; Figure 2A, Figure 2B, and Figure 3; eTable 6 in the Supplement). The pathogenic/likely pathogenic frequency among European patients with osteosarcoma was also higher compared with the frequency among individuals in the ExAC group (2527 individuals [9.3%]; 95% CI, 8.2%-10.5%; Fisher exact P = 2.3 × 10⁻⁵⁵; Figure 2A and Figure 2B; eTable 6 in the Supplement). Patients with pathogenic/likely pathogenic variants were significantly younger (mean [SD] age, 15.3 [7.2] years; age range, 2-61 years) than patients without pathogenic/likely pathogenic variants (mean [SD] age, 16.9 [10.2] years; age range, 2-80 years; Mann-Whitney U P = .02; Figure 4; eFigure 2 in the Supplement).

Among 364 patients with osteosarcoma subtype information, cancer-susceptibility genes with pathogenic/likely pathogenic variants were less common in those with surface subtypes (3 of 22 patients [13.6%]) vs conventional subtypes (104 of 342 patients [30.4%]; eTable 8 in the Supplement). A pathway enrichment analysis of the 101 cancer-susceptibility genes with pathogenic/likely pathogenic variants indicated enrichment in DNA repair pathway genes (Fisher exact P = 3.4 × 10⁻⁶⁵; eFigure 3 and eTable 9 in the Supplement).

Autosomal-Dominant Genes
Overall, 185 of 1004 patients (18.4%; 95% CI, 12.8%-24.0%) with osteosarcoma had a pathogenic/likely pathogenic variant in an autosomal-dominant or an autosomal-dominant and autosomal-recessive cancer-susceptibility gene, whereas the variant frequency was 56 individuals (5.3%; 95% CI, 0%-11.1%) in the control group and 1494 individuals (5.5%; 95% CI, 4.3%-6.6%) in the ExAC group (Figure 2A and Figure 2B; eTable 6 in the Supplement). The highest frequency of pathogenic/likely pathogenic autosomal-dominant cancer-susceptibility gene variants was found in patients aged 0 to 10 years (37 of 151 patients [24.5%]; Mann-Whitney U P = .006; Figure 4). The 732 European patients with cancer had a higher burden of pathogenic/likely pathogenic autosomal-dominant variants than the 994 European individuals in the control group (burden P = 1.9 × 10⁻¹⁰). This higher burden translated to a nearly 4-fold greater risk of carrying a pathogenic/likely pathogenic variant compared with the ExAC group (odds ratio [OR], 3.9; 95% CI, 3.3-4.6).

Eighteen patients (1.8%) had more than 1 pathogenic/likely pathogenic autosomal-dominant variant compared with 4 individuals (0.4%) in the control group (Fisher exact P = .002). No significant difference was observed in overall patient survival for those carrying any pathogenic/likely pathogenic variant or an autosomal-dominant pathogenic/likely pathogenic variant compared with patients without these variants (Cox [adjusted for age, sex, and tumor location] P = .55 for all genes and P = .34 for autosomal-dominant genes) in the subset of 407 patients for whom outcome data was available.
Pathogenic/likely pathogenic variants in the TP53 gene were the most frequent of all autosomal-dominant genes (44 of 1004 total patients [4.4%]; 30 of 732 European patients [4.1%]) and substantially higher than those observed in the control group (3 of 1062 total individuals [0.3%]; 3 of 994 European individuals [0.3%]; burden $P = 3.2 \times 10^{-8}$) and the ExAC group (27 individuals [0.1%]; Fisher exact $P = 9.0 \times 10^{-44}$; Figure 2B and Figure 3; eTable 7 in the Supplement). This finding is consistent with a previous study, which included 360 patients who were also in the current study. Analyses restricted to European patients who did not participate in the previous study found that 32 of 644 patients (5.0%) had a pathogenic TP53 variant.

All pathogenic/likely pathogenic TP53 variants were observed in patients younger than 30 years at diagnosis, with the exception of 1 patient, who was aged 39 years at diagnosis (Mann-Whitney $U = .05$; Figure 2 and eTable 8 in the Supplement). Patients aged 0 to 10 years (n = 151) had the highest estimated likelihood of carrying a TP53 pathogenic/likely pathogenic variant (OR, 108; 95% CI, 47-247; Figure 2B and Figure 4). Patients with a pathogenic/likely pathogenic TP53 variant were more likely to have osteosarcoma of the axial skeleton ($\chi^2 P = .001$), and the data suggested that patients with TP53 pathogenic/likely pathogenic variants were more likely to have metastases at diagnosis ($\chi^2 P = .06$; eTable 8 in the Supplement). In the subset of patients with outcome data, an adjusted Cox proportional hazards model indicated that patients carrying a TP53 pathogenic/likely pathogenic variant had significantly worse overall survival compared with patients without these variants (HR, 2.2; 95% CI, 1.2-4.0; Cox $P = .009$). These variants occurred in several functional domains, including the DNA-binding domain (subregion-based burden $P = 1.5 \times 10^{-44}$; eFigure 4A in the Supplement), which is consistent with previous studies.

The gene CDKN2A (OMIM 600160) had the second highest frequency of pathogenic/likely pathogenic variants in the patients with osteosarcoma (12 of 1004 total patients [1.2%]; 8 of 732 European patients [1.1%]) compared with no pathogenic/likely pathogenic variants among individuals in the control group (burden $P = 3.1 \times 10^{-3}$) and the ExAC group (Fisher exact $P = 2.2 \times 10^{-13}$; Figure 2B and Figure 3; eTable 7 in the Supplement). Individuals with a CDKN2A pathogenic/likely pathogenic variant were younger (mean [SD] age, 12.9 [4.4] years) than patients without pathogenic/likely pathogenic variants (mean [SD] age, 6.9 [10.2] years; Mann-Whitney $U = .03$). Notably, the youngest patients (aged 0-10 years) had the highest frequency of these variants (3 of 151 patients [2.0%]; Figure 4). The CDKN2A variants mapped to sites that were somatically mutated in bone cancers (eFigure 4B in the Supplement). Five additional autosomal-dominant genes (MEN1 [OMIM 613733], VHL [OMIM 608537], POT1 [OMIM 606478], APC [OMIM 611731], and MSH2 [OMIM 609309]) had a significantly higher pathogenic/likely pathogenic burden in European patients compared with European individuals in the control group (Figure 3; eTable 7 in the Supplement).

We compared the frequency of pathogenic/likely pathogenic variants among individuals in the ExAC group and observed that the risk of carrying a pathogenic/likely pathogenic variant in genes MENI, VHL, POT1, and APC was elevated in European patients with osteosarcoma after a Bonferroni adjustment (Figure 2B; eTable 7 in the Supplement). Fifty-five additional autosomal-dominant genes had pathogenic/likely pathogenic variants in 1 or more patients (each were present...
Most of the specific variants observed in patients were absent in individuals in both the control group and the ExAC group as well as other public databases (the 1000 Genomes Project and the Exome Sequencing Project).

In addition, 316 patients (25.4%) with osteosarcoma had a rare variant of uncertain significance that was predicted to be damaging in silico in an autosomal-dominant gene, in the absence of another pathogenic/likely pathogenic autosomal-dominant variant. Altogether, 545 patients (43.8%) had at least 1 pathogenic variant, likely pathogenic variant, or variant of uncertain significance that was predicted to be damaging in silico in an autosomal-dominant gene. The European patients had more variants of uncertain significance that were predicted to be damaging in silico in the genes *RB1* (OMIM 614041) and *VHL* (OMIM 608537) compared with individuals in the control group after adjustment for multiple testing (eTable 7 in the Supplement).

**Autosomal-Recessive Genes**

A total of 92 of 1004 patients (9.2%; 95% CI, 3.3%-15.1%) had a pathogenic/likely pathogenic variant in 33 autosomal-recessive genes, which is higher than that of the control group (72 of 1062 individuals [6.8%]; burden \( P = .03 \)) and the ExAC group (1041 individuals [3.8%]; Fisher exact \( P = 2.6 \times 10^{-13} \); Figure 2A and Figure 2B; eTable 6 in the Supplement). All autosomal-recessive gene variants were present as single heterozygotes, with the exception of 1 patient aged 13 years who...
had osteosarcoma with 2 RECQL4 (OMIM 603780) pathogenic/likely pathogenic variants; we were unable to phase the variants. The gene RECQL4 had the highest frequency of pathogenic/likely pathogenic variants in European patients with osteosarcoma (7 of 732 patients [1.0%]) compared with European individuals in the control group (1 of 994 individuals [0.1%]; burden \( P = .02 \); Figure 2B and Figure 3; eFigure 4C and eTable 7 in the Supplement). One RECQL4 variant was previously reported in a patient with Rothmund-Thomson syndrome and osteosarcoma (c.2476C>T, p.Arg826*).\(^6^0\) Several other autosomal-recessive genes had more pathogenic/likely pathogenic variants in patients than in the control group but were not significantly associated (Figure 2B and Figure 3; eTable 7 and eTable 10 in the Supplement).

We observed a preponderance of male patients (4 of 1004 patients [0.4% of the total patients and 0.7% of the 540 male patients in the discovery set)) who carried a pathogenic/likely pathogenic variant in an X-linked cancer-susceptibility gene (DKC1 [OMIM 300126], GPC3 [OMIM 300037], or WAS [OMIM 300392]) compared with no individuals in the control group (1 of 994 individuals [0.1%]; burden \( P = .02 \); Figure 2B and Figure 3; eFigure 4C and eTable 7 in the Supplement). One variant was pathogenic/likely pathogenic variant in a syndrome with osteosarcoma 22,49 with double the sample. These data suggest that germline genetic association with osteosarcoma existed, we evaluated rare variants in 736 candidate genes, which included 140 genes previously associated with osteosarcoma and 596 somatically altered genes (eTable 3 in the Supplement).

Candidate Gene Rare-Variant Burden

To explore whether unidentified germline genetic associations with osteosarcoma existed, we evaluated rare variants in 736 candidate genes, which included 140 genes previously associated with osteosarcoma and 596 somatically altered genes (eTable 3 in the Supplement). Burden tests of in silico–predicted damaging variants (minor allele frequency \( \leq 0.005 \)) and all rare variants (minor allele frequency \( \leq 0.01 \)) did not identify an association with the evaluated genes (eTable 12 in the Supplement). One exception was observed; the gene ATRX (OMIM 300032) had a higher rare-variant burden in European patients (28 of 732 patients [3.8%]) compared with European individuals in the control group (18 of 994 individuals [1.8%]). One variant was pathogenic (c.6532C>T, p.Arg2178Trp, in 1 male patient; absent in the control group) and was previously reported to be pathogenic for alpha-thalassemia X-linked (ATR-X) intellectual disability syndrome in a patient with ATR-X syndrome who also developed osteosarcoma.\(^4^1,4^2\)

Discussion

We report that 28.0% of patients with osteosarcoma had a pathogenic/likely pathogenic variant in a cancer-susceptibility gene, with 18.4% of those variants in an autosomal-dominant gene; to our knowledge, this frequency is higher than previously reported for any other pediatric cancer.\(^2^3-2^5,6^3\) The highest carrier frequency was observed in the youngest patients, with 24.5% of patients aged 0 to 10 years carrying a pathogenic/likely pathogenic variant in an autosomal-dominant gene. An additional 25.4% of the total patients had an in silico–predicted damaging variant of uncertain significance in an autosomal-dominant cancer-susceptibility gene. We confirmed previous observations of a high frequency of germline TP53 pathogenic/likely pathogenic variants in patients with osteosarcoma 22,49 with double the sample. These data suggest that germline TP53 pathogenic/likely patho-
Pathogenic Germline Variants in Cancer-Susceptibility Genes in Patients With Osteosarcoma

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OBJECTIVE To report on the discovery and replication of rare cancer-susceptibility variants in patients with osteosarcoma.

METHODS We analyzed whole-exome sequencing data from patients with osteosarcoma identified through the Childhood Cancer Survivor Study, which derived its data from multiple sources. We performed bioinformatics analysis of the data to identify rare cancer-susceptibility variants. The variants were classified as pathogenic/likely pathogenic if they met at least one of the following criteria: (1) they were pathogenic/likely pathogenic in at least one of the databases (ACG, ClinVar, ExAC), (2) they had a frequency less than or equal to 0.005 in the ExAC database, and (3) they had a deleterious stop, frameshift, or splice-site variant. A total of 1244 patients who were newly diagnosed with osteosarcoma were included in the study.

RESULTS We identified 15 pathogenic/likely pathogenic variants in 5 genes: CDKN2A, MEN1, VHL, POT1, and ATRX. Two of these variants were new: p.Asp125His and p.Gly101Trp. A total of 41 individuals were identified as having a pathogenic/likely pathogenic variant. The most frequent variants were p.Asp125His and p.Gly101Trp. The p.Asp125His variant has been reported previously to be associated with a cancer-susceptibility syndrome and has been associated with melanoma, and the p.Gly101Trp variant has been reported to be associated with osteosarcoma.

CONCLUSIONS Our data underscore the high frequency of potentially actionable cancer risk variants in patients with osteosarcoma. The p.Asp125His variant has been reported previously to be associated with a cancer-susceptibility syndrome and has been associated with melanoma, and the p.Gly101Trp variant has been reported to be associated with osteosarcoma. Further studies are needed to refine our observations and identify optimal approaches to genetic testing and counseling for patients with osteosarcoma.
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REFERENCES
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