MEDICAL SCIENCE MONITOR

LAB/IN VITRO RESEARCH

e-ISSN 1643-3750
© Med Sci Monit, 2017; 23: 5892-5898
DOI: 10.12659/MSM.908004

Received: 2017.11.11
Accepted: 2017.11.27
Published: 2017.12.12

Genetic Polymorphisms of Insulin-Like Growth Factor 1 Are Associated with Osteosarcoma Risk and Prognosis

Authors' Contribution:
Study Design A
Data Collection B
Statistical Analysis C
Data Interpretation D
Manuscript Preparation E
Literature Search F
Funds Collection G

ABCEG 1 Jianshui Mao
BCEF 1 Genying Zhuang
CDEF 2 Zhikang Chen

1 Department of Orthopedics, The Fourth Affiliated Hospital of Zhejiang University School of Medicine, Yiwu, Zhejiang, P.R. China
2 School of Public Health, Fudan University, Shanghai, P.R. China

Corresponding Author: Jianshui Mao, e-mail: zhoujuansh@sina.com
Source of support: Departmental sources

Background: Insulin-like growth factor 1 (IGF-1) gene plays an important role in bone and soft tumors. IGF-1 gene polymorphisms have been revealed to be correlated with the carcinogenesis and progression of solid malignancies. We therefore hypothesized that IGF-1 genetic polymorphisms might be associated with the risks and outcomes of osteosarcomas in Chinese individuals.

Material/Methods: This study included 173 conventional osteosarcoma individuals and 175 tumor-free controls. Five single nucleotide polymorphisms (SNPs) of IGF-1 (rs6214, rs6218, rs35767, rs5742612, and rs5742714) were genotyped. DNA was extracted from peripheral blood and analyzed for SNP genotyping using PCR.

Results: We found that rs6218 had a predictive role for the susceptibility and progression of osteosarcoma. The presence of TC and CC genotypes of rs6218 indicated higher risk of osteosarcoma. In addition, rs6218 TC and CC genotypes were discovered to be associated with later stage and elevated risk of osteosarcoma metastasis.

Conclusions: IGF-1 polymorphisms are potential prognostic predictors of osteosarcoma susceptibility and outcomes.

MeSH Keywords: Osteosarcoma • Polymorphism, Single Nucleotide •Prognosis

Full-text PDF: https://www.medscimonit.com/abstract/index/idArt/908004

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS] [Index Copernicus]

Background

Osteosarcoma is the most common kind of malignancy in musculoskeletal tumors and has the characteristic of forming early lung metastases and unfavorable survival [1]. Although in the past decades, surgery in combination with neoadjuvant and postoperative chemotherapy has extended the overall survival time for osteosarcoma patients, there are still controversies regarding the appropriate dose of chemotherapy agents [2]. In addition, once osteosarcoma becomes chemo-resistant, there are few therapeutic strategies available. In high-grade cases, the puzzling question arises as to whether patients should receive more aggressive chemotherapy regimens or radiotherapy. Also, numerous efforts have been made to assess second-line chemotherapy agents for refractory cases; however, there is limited methodology available to evaluate the response rate of conventional chemotherapy except for the evaluation of necrosis [3,4]. If there were reliable biomarkers that could predict osteosarcoma susceptibility or prognosis, clinicians could apply aggressive regimens or radiotherapy for high-risk patients.

Insulin-like growth factor 1 (IGF-1) is a member of the IGF family, a family of insulin-related proteins or peptides [5]. IGF-1 is involved in the regulation of cell survival, proliferation, and differentiation. The downstream signaling pathways include the MAPK pathway, PI3K-Akt pathway, and NF-κB pathway [6,7]. The gene locus that encodes IGF-1 is located on chromosome 12. Given the fact that IGF-1 is involved in regulating mitogenesis and anti-apoptosis, the role of the IGF family members in osteosarcoma tumorigenesis or progression is still poorly understood. The receptor of IGF-1 (IGF-1R) has been found to be overexpressed in multiple musculoskeletal sarcomas, including osteosarcoma [8]. Increasing evidence indicates that IGF-1 related pathways are involved in osteosarcoma progression [9,10]. However, there is still controversy regarding the prognostic value of IGF-1R or IGF-1 expression in osteosarcomas. For instance, Wang et al. reported that higher expression of IGF-1R was associated with poorer survival [10], while van de Luijtgaarden et al. showed that IGF-1R seemed to be correlated with better overall survival [11]. Larger cohort studies are needed to reach a more accurate conclusion; however, the low incidence of osteosarcoma severely limits large studies. Thus, studies utilizing blood samples or other easy-to-obtain data are of great value.

Single nucleotide polymorphisms (SNPs) have been validated to be correlated with the risk and prognosis of solid malignancies. However, SNPs role in osteosarcomas are still elusive because of the rarity of the disease. As emerging studies revealed the prognostic role played by SNPs in tumors, it will be important to evaluate genetic polymorphisms of given genes in osteosarcoma [3]. SNPs of IGF-1 have been proven to be associated with susceptibility and outcomes of some cancers, including prostate cancer, colorectal cancer, and breast cancer. Hence, we conjectured that IGF-1 polymorphisms might be associated with osteosarcoma tumorigenesis.

We collected peripheral blood samples from 173 osteosarcoma cases and 175 healthy controls to analyze IGF-1 polymorphisms and to reveal the relationship between IGF-1 SNPs and osteosarcomas. In this case-control study, we analyzed five IGF-1 SNPs (rs6214, rs6218, rs35767, rs5742612, and rs5742714) and tried to validate the hypothesis that IGF-1 polymorphisms might be associated with susceptibility and progression of osteosarcoma.

Material and Methods

Ethics approval

This case-control study involving patients was approved by the ethical committee board of the two institutions. Signed informed consents were obtained from all participants or their legal guardians.

Osteosarcoma cases and controls

There were 173 primary osteosarcoma individuals and 175 tumor-free controls involved in this case-control study. All the study patients were Chinese Han people. All osteosarcoma cases were pathologically diagnosed from October 2009 to March 2012. Before performing neoadjuvant chemotherapy, blood samples from individuals were collected and preserved in liquid nitrogen. All osteosarcoma cases received established therapeutic strategies including essential neoadjuvant chemotherapy, surgical excision, and postoperative chemotherapy. Surgeries were performed by experienced orthopedic surgeons and all osteosarcoma individuals were followed for at least 60 months. Tumor-free controls were trauma cases and were matched to osteosarcoma individuals by age and sex. All the clinical materials were reviewed from saved clinical records.

DNA extraction

Total DNA was isolated from peripheral blood by proteinase K digestion and phenol-chloroform method. A Blood DNA Extraction Kit from Qiagen was utilized for DNA isolation. In brief, blood samples were mixed with proteinase K and cultured for 10 minutes at 55°C, and then centrifuged to remove drops from the inside of the lid. Then 200 µL ethanol was added and mixed by pulse-vortex, and then centrifuged. This mixture was applied to the QIAamp Mini spin column from the kit. After centrifuging at 6,000 g for one minute, 500 µL Buffer AW2 was added and then centrifuged at 20,000 g for three minutes. Extracted DNA was used for further genotyping.

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS] [Index Copernicus]

Table 1. General characteristics.

Variables		Osteosarcoma cases [n (%)]	Tumor-free control n (%)]	*P*
Age	Mean ±SD (year)	15.35±2.91	15.76±3.40	0.352
Gender	Male	92 (53.18)	95 (54.28)	0.521
	Female	81 (46.82)	80 (45.72)	
Location	Trunk	18 (10.40)		
	Limbs	155 (89.60)		
Enneking stages	IA or IB	21 (12.13)		
	IIA or IIB or III	152 (87.87)		
Operation	Amputation	24 (13.87)		
	Limb salvage	149 (86.13)		
Metastasis	Yes	83 (47.98)		
	No	90 (52.02)		

SNP genotyping

We reviewed the HapMap data and picked out frequent SNPs (frequency >5% in the Chinese Han people). Five SNPs (rs6214, rs6218, rs35767, rs5742612, and rs5742714) were genotyped and evaluated. Sequence Detection Systems software on an ABI StepOnePlus system (Thermo Fisher Scientific) was used for data collection and analyses. Primers and probes for TaqMan assays were designed online on the ABI Assay-by-Design services. We used 384-well plates from ABI, and all samples were run in triplicate. Two technicians independently performed SNP genotyping in a blinded manner. The amplification conditions were two minutes at 50°C and 10 minutes at 95°C, followed by 40 cycles at 95°C for 15 seconds and at 60°C for 60 seconds.

Haplotype analysis

Computational haplotyping method was used for haplotype analysis. The five tagging SNPs were analyzed on SHEsis system (*http://analysis.bio-x.cn/myAnalysis.php*) to find haplotypes with frequencies over 3%.

Statistical analysis

The Pearson's χ^2 test was used to evaluate the differences in frequency distributions of characteristics including age, sex, and Enneking stage. Selected variables and genotypes of *IGF-1* changes were compared between the osteosarcoma individuals and the tumor-free controls. Odds ratios (ORs) and 95% confidential intervals (95% CIs) were calculated to evaluate the association between the five selected tagging SNPs and the susceptibility or prognosis of osteosarcoma. Unconditioned logistic regression analysis was preformed to evaluate crude ORs, and subsequently adjusted for age and sex. The Hardy-Weinberg equilibrium was also evaluated using Pearson's χ^2 test. A value of $p<0.05$ was considered statistically significant. All statistical analyses were two-sided and performed by SPSS 21.0.

Results

Clinical characteristics

Clinical information of all included osteosarcoma individuals and tumor-free controls are displayed in Table 1. There were 92 male osteosarcoma cases and 81 female osteosarcoma cases. The medium age and range of osteosarcoma cases and control cases was 15.35±2.91 years and 15.76±3.40 years, respectively, with no statistical difference for age (p=0.352) or sex (p=0.521). The Enneking GTM System was used for clinical staging [12]. In the osteosarcoma group, there were 21 early-stage cases (stage I) and 152 late-stage cases (stage II or III). Among the 173 osteosarcoma cases, 24 individuals underwent amputation and 149 cases underwent limb salvage. During the five-year-follow-up, 83 cases developed lung metastases.

IGF-1 SNP rs6218 was correlated with the risk of osteosarcoma

The pooled data of the five evaluated SNPs (rs6214, rs6218, rs35767, rs5742612, and rs5742714) are shown in Table 2. In the tumor-free control individuals, the genotype distributions of the evaluated five tagging SNPs were all within Hardy-Weinberg equilibrium (p=0.552, 0.710, 0.225, 0.189, and

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS] [Index Copernicus]

Table 2. Logistic regression analyses of associations between *IGF-1* rs6214, rs6218, rs35767, rs5742612, and rs5742714 polymorphisms and risk of osteosarcoma.

IGF-1 genotype	Cases (n=173)		Controls (n=175)		Crude OR (95%CI)	*P*	Adjusted OR (95%CI)	*P*
	n	%	n	%				
rs6214 G/A								
GG	49	28.32	51	29.14	1.00		1.00	
GA	84	48.55	86	49.14	1.22 (0.73–1.59)	0.287	1.21 (0.72–1.58)	0.293
AA	40	23.13	38	21.72	1.29 (0.83–1.42)	0.311	1.30 (0.82–1.44)	0.301
GA+AA	124	71.68	124	70.86	1.13 (0.79–1.35)	0.233	1.13 (0.79–1.36)	0.225
GG+GA	133	76.87	137	78.28	1.00		1.00	
AA	40	23.13	38	21.72	1.10 (0.57–1.25)	0.310	1.12 (0.58–1.29)	0.320
rs6218 T/C								
TT	90	52.02	110	62.86	1.00		1.00	
TC	69	39.88	60	34.29	1.51 (1.15–1.82)	0.002*	1.50 (1.13–1.82)	0.002*
CC	14	8.10	5	2.85	1.42 (1.10–1.77)	0.009*	1.43 (1.09–1.88)	0.008*
TC+CC	83	47.98	249	87.06	1.39 (1.11–1.76)	0.003*	1.38 (1.08–1.78)	0.004*
TT+TC	159	91.90	165	57.69	1.00		1.00	
CC	14	8.10	121	42.31	1.07 (1.03–1.39)	0.017*	1.10 (1.05–1.40)	0.015*
rs35767 C/T								
CC	63	36.41	66	37.71	1.00		1.00	
CT	86	49.71	83	47.43	0.85 (0.48–1.22)	0.331	0.84 (0.47–1.21)	0.341
TT	24	13.88	26	14.86	1.12 (0.77–1.53)	0.561	1.10 (0.72–1.50)	0.560
CT+TT	110	63.59	109	62.29	0.91 (0.60–1.38)	0.620	0.92 (0.63–1.40)	0.610
CC+CT	149	86.12	149	85.14	1.00		1.00	
TT	24	13.88	26	14.86	1.23 (0.89–1.91)	0.320	1.22 (0.86–1.94)	0.317
rs5742612 T/C								
TT	76	43.93	83	47.43	1.00		1.00	
TC	79	45.66	76	43.43	0.73 (0.48–1.09)	0.220	0.75 (0.49–1.12)	0.222
CC	18	10.41	16	9.14	1.10 (0.77–1.83)	0.441	1.10 (0.76–1.83)	0.440
TC+CC	97	56.07	92	52.57	0.96 (0.73–1.11)	0.125	0.92 (0.71–1.07)	0.121
TT+TC	155	89.59	159	90.86	1.00		1.00	
CC	18	10.41	16	9.14	1.25 (0.80–1.90)	0.109	1.24 (0.76–1.88)	0.108
rs5742714 G/C								
GG	112	64.74	115	65.71	1.00		1.00	
GC	53	30.64	50	28.57	0.60 (0.41–1.25)	0.520	0.60 (0.40–1.22)	0.518
CC	8	4.62	10	5.72	0.89 (0.50–1.52)	0.771	0.91 (0.51–1.53)	0.750
GC+CC	61	35.26	60	34.29	0.82 (0.71–1.39)	0.472	0.85 (0.75–1.39)	0.463
GG+GC	165	95.38	165	94.28	1.00		1.00	
CC	8	4.62	10	5.72	1.21 (0.87–1.60)	0.229	1.20 (0.86–1.62)	0.219

* Statistically significant (P<0.05).

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS] [Index Copernicus]

Table 3. Correlations between genotype frequencies of *IGF-1* rs6218T/C and clinical features in osteosarcoma individuals.

Variables	n	TT n (%)	TC n (%)	CC n (%)	*P*
Location					
Trunk	18	10 (55.56)	7 (38.89)	1 (5.55)	0.106
Limbs	155	83 (53.55)	55 (35.48)	17 (10.97)	
Enneking stages					
IA or IB	21	11 (52.38)	8 (38.09)	2 (9.53)	0.007*
IIA or IIB or III	152	68 (44.74)	79 (51.97)	5 (3.29)	
Operation					
Amputation	24	12 (50.00)	10 (41.67)	2 (8.33)	0.082
Limb salvage	149	72 (48.32)	70 (49.98)	7 (4.70)	
Metastasis					
Yes	83	38 (45.78)	44 (53.01)	1 (1.21)	0.002*
No	90	29 (32.22)	56 (62.22)	5 (5.56)	

Statistically significant ($P<0.05$).

Table 4. Confounding variables (Enneking stages).

Confounding variables		IA or IB cases [n (%)]	IIA or IIB or III cases [n (%)]	*P*
Age	Mean ±SD (year)	14.57±3.16	15.02±2.86	0.772
Gender	Male	11 (52.38)	80 (53.69)	0.102
	Female	10 (47.62)	79 (46.31)	

Table 5. Confounding variables (metastasis).

Confounding variables		Metastasis cases [n (%)]	Non-metastasis cases [n (%)]	*P*
Age	Mean ±SD (year)	14.66±3.02	15.03±3.12	0.581
Gender	Male	44 (53.01)	49 (54.44)	0.312
	Female	39 (46.99)	41 (45.56)	

0.312, respectively). In IGF-1 SNP rs6218, when the TT homozygote genotype was set as the reference group, the TC genotype was found to be statistically significantly elevated for the risk of osteosarcoma (TC versus TT: crude OR=1.51, 95% CI=1.15–1.82, p=0.002; adjusted OR=1.50, 95% CI=1.13–1.82, p=0.002). Furthermore, the TT homozygote genotype was also found to be associated with the risk of osteosarcoma (CC versus TT: crude OR=1.42, 95% CI=1.10–1.77, p=0.009; adjusted OR=1.43, 95% CI=1.09–1.88, p=0.008). In addition, in the dominant model (TC+CC), a statistical difference was also revealed (TC+CC versus TT: crude OR=1.39, 95% CI=1.11–1.76, p=0.003; adjusted OR=1.38, 95% CI=1.08–1.78, p=0.004). In the recessive model (TT+TC), when compared with the CC homozygote genotype, a statistical difference was also found (TT+TC versus CC: crude OR=1.07, 95% CI=1.03–1.39, p=0.017; adjusted OR=1.10, 95% CI=1.05–1.40, p=0.015).

The other four tagging SNPs (rs6214, rs35767, rs5742612, and rs5742714) did not show potential correlations with osteosarcoma susceptibility in Chinese Han individuals.

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS] [Index Copernicus]

Table 6. Haplotype analyses.

Haplotype	Cases (n=173) n (%)	Controls (n=175) n (%)	*P*	OR (95% CI)
GCGGT	5.71 (0.033)	4.91 (0.028)	0.133	1.125 (0.610–1.733)
CGCGA	7.27 (0.042)	21.35 (0.122)	0.008*	1.787 (1.535–2.208)
CCCTT	15.40 (0.089)	16.10 (0.092)	0.525	0.611 (0.375–1.196)
CAACA	17.65 (0.102)	19.78 (0.113)	0.093	1.190 (0.733–1.574)
CAGGG	6.06 (0.035)	7.18 (0.041)	0.109	0.688 (0.472–1.337)
CCGGG	7.27 (0.042)	8.58 (0.049)	0.331	0.835 (0.577–1.395)
CCGGT	12.11 (0.070)	13.83 (0.079)	0.115	0.815 (0.606–1.070)
GCGGC	21.63 (0.125)	23.63 (0.135)	0.227	1.182 (0.785–1.545)

* Statistically significant (P<0.05).

rs6218 was associated with the Enneking stage and metastatic risk of osteosarcoma

Pooled data on tumor primary location, Enneking stage, operation technique, and metastasis of osteosarcoma was used to study the correlations between the five selected SNPs and osteosarcomas (Table 3). For rs6218, the frequency of the genotype TC and CC at late Enneking stages (51.97% and 3.29%, respectively) were significantly higher when compared with early-stage cases (38.09% and 9.53%, respectively), and a statistical difference in distribution was revealed (p=0.007). Furthermore, the evaluation of tumor metastasis also showed statistically significant results. The genotype TC displayed significantly higher frequency (62.22%) in metastasized cases when compared with metastasis-free cases (53.01%), and the difference in distributions was statistically significant (p=0.002). This data indicated that rs6218 polymorphisms were associated with late Enneking stages and higher risk of lung metastasizing.

The possible confounding variables (Enneking stages and metastasis) are shown in Tables 4 and 5. No statistical differences were found.

Haplotype analyses showed statistical differences between osteosarcoma cases and control cases

The evaluation of the five SNPs revealed eight frequent (frequency >3%) haplotypes: GCGGT, CGCGA, CCCTT, CAACA, CAGGG, CCGGG, CCGGT, and GCGGC (Table 6). The haplotype CGCGA showed a statistically significantly difference between the osteosarcoma cases and tumor-free control cases (p=0.008, OR 1.787, 95% CI=1.535–2.208). The other seven haplotypes did not reveal any differences between osteosarcoma cases and cancer-free control cases.

Discussion

Osteosarcoma is a rare malignancy, with morbidity about five per million population annually [13]. The low morbidity rate severely restricts clinical sample-based studies. Although accumulating studies have reported some potential biological biomarkers for osteosarcoma, further studies are needed to improve the accuracy of those markers. In addition, attempts to find alternative therapeutic strategies, especially non-surgical strategies, have been refractory for osteosarcomas [14,15]. Until now, there have been no satisfactory strategies that include first-line regimens in the clinical care setting of osteosarcomas. Thus, finding new prognostic markers that can predict osteosarcoma risks or outcomes for conventional therapy would be of great value.

Emerging studies have revealed genetic factors that are potentially associated with osteosarcoma carcinogenesis and/or progression. However, one of the most important factors for improving the clinical value of biomarkers is improving the method used to evaluate the markers. Genetic polymorphisms, which can be studied based on blood samples and evaluated by PCR, are good biomarker candidates based on available methodology. In osteosarcoma, some studies have demonstrated promising prognostic roles for biomarkers including *NAT2* [16], *WWOX* [17], and *TCF21* [18]. However, in most circumstances, using only one genetic polymorphism factor is not considered reliable due to the limited number of cohorts available that restricts accuracy. IGF-1 is produced by the liver and by some malignant cells, and is believed to be involved in osteosarcoma tumorigenesis. Recently, epidemiologic and experimental studies have revealed the value of IGF-1 genetic polymorphisms in predicting tumor risk and prognosis. In a study involving a large cohort of prostate cancer cases (5,887 cases), the IGF signaling pathway was found to be associated

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS] [Index Copernicus]

with prostate cancer mortality [19]. Other studies have also emphasized the prognostic role of IGF-1. In a study recruiting 110 colorectal cancer cases and 137 breast cancer cases, the genetic polymorphisms of IGF-1 were evaluated. The authors reported that IGF-1 SNPs were correlated with cancer risk and prognosis [20]. In addition, another recent study provided some evidence that targeting IGF-1R might suppress osteosarcoma progression, highlighting the role that the IGF-1 family plays in osteosarcoma [21].

Our study was based on a large number of cases to evaluate whether genetic polymorphisms in IGF-1 was associated with osteosarcoma risk and progression. Five candidate SNPs were genotyped and analyzed; and rs6218 was found to be associated with significantly higher osteosarcoma susceptibility. Moreover, rs6218 was discovered to be correlated with metastasis potential of osteosarcoma, indicating that alterations in this SNP were associated with poorer prognosis of osteosarcoma. According to our results, if a patient suffering from osteosarcoma has worse phenotypes of rs6218, which indicates higher risk of metastasis potential, clinicians should consider using more aggressive therapeutic strategies and perform more frequent follow-ups. However, although the other four study SNPs were not related to osteosarcoma risk or progression in our study, they have been found to be associated with some other malignancies. Thus, we could not reach a conclusion that the other four SNPs were irrelevant to osteosarcoma.

In addition, we also performed haplotype analyses. The frequency of haplotype CGCGA was statistically significantly different between the osteosarcoma cases and the tumor-free control cases.

There were limitations to our study. We could not avoid inherent bias as all the blood samples were preserved and obtained from one institution. Furthermore, the sample size weakened the power of statistical analyses, especially for the results of homozygotic cases. Some homozygotic types only had a very small number of cases. Expanded studies that recruit more osteosarcoma cases would help improve the efficacy of the prognostic effect of IGF-1 genetic polymorphisms.

Conclusion

In conclusion, to the best of our knowledge, this is the first study to report that IGF-1 genetic polymorphisms were associated with osteosarcoma susceptibility and progression, especially metastasis. Thus, *IGF-1* rs6218 SNP polymorphisms can be considered valuable prognostic biomarkers for osteosarcoma.

Conflicts of interest

None.

References:

1. Isakoff MS, Bielack SS, Meltzer P et al: Osteosarcoma: Current treatment and a collaborative pathway to success. J Clin Oncol, 2015; 33(27): 3029–35
2. Ferrari S, Serra M: An update on chemotherapy for osteosarcoma. Expert Opin Pharmacother, 2015; 16(18): 2727–36
3. Savage SA, Mirabello L, Wang Z et al: Genome-wide association study identifies two susceptibility *loci* for osteosarcoma. Nat Genet, 2013; 45(7): 799–803
4. Vanneman M, Dranoff G: Combining immunotherapy and targeted therapies in cancer treatment. Nat Rev Cancer, 2012; 12(4): 237–51
5. Chan JM, Stampfer MJ, Giovannucci E et al: Plasma insulin-like growth factor-I and prostate cancer risk: A prospective study. Science, 1998; 279(5350): 563–66
6. Yu H, Spitz MR, Mistry J et al: Plasma levels of insulin-like growth factor-I and lung cancer risk: A case-control analysis. J Natl Cancer Inst, 1999; 91(2): 151–56
7. Vivanco I, Sawyers CL: The phosphatidylinositol 3-Kinase AKT pathway in human cancer. Nat Rev Cancer, 2002; 2(7): 489–501
8. Xie Y, Skytting B, Nilsson G et al: Expression of insulin-like growth factor-1 receptor in synovial sarcoma: association with an aggressive phenotype. Cancer Res, 1999; 59(15): 3588–91
9. Rikhof B, de Jong S, Suurmeijer A J et al: The insulin-like growth factor system and sarcomas. J Pathol, 2009; 217(4): 469–82
10. Wang Y H, Han X D, Qiu Y et al: Increased expression of insulin-like growth factor-1 receptor is correlated with tumor metastasis and prognosis in patients with osteosarcoma. J Surg Oncol, 2012; 105(3): 235–43
11. van de Luijtgaarden AC, Roeffen MH, Leus MA et al: IGF signaling pathway analysis of osteosarcomas reveals the prognostic value of pAKT localization. Future Oncol, 2013; 9(11): 1733–40
12. Enneking WF: A system of staging musculoskeletal neoplasms. Clin Orthop Relat Res, 1986; (204): 9–24
13. Biermann JS, Adkins DR, Agulnik M et al: Bone cancer. J Natl Compr Canc Netw, 2013; 11(6): 688–723
14. Omer N, Le Deley MC, Piperno-Neumann S et al: Phase-II trials in osteosarcoma recurrences: A systematic review of past experience. Eur J Cancer, 2017; 75: 98–108
15. Kansara M, Teng MW, Smyth MJ et al: Translational biology of osteosarcoma. Nat Rev Cancer, 2014; 14(11): 722–35
16. Huang Z, Yuan L, Jiang Z et al: Associations of polymorphisms in NAT2 gene with risk and metastasis of osteosarcoma in young Chinese population. Onco Targets Ther, 2015; 8: 2675–80
17. Zhang N, Jiang Z, Ren W et al: Association of polymorphisms in WWOX gene with risk and outcome of osteosarcoma in a sample of the young Chinese population. Onco Targets Ther, 2016; 9: 807–13
18. Jiang Z, Zhang W, Chen Z et al: Transcription Factor 21 (TCF21) rs12190287 polymorphism is associated with osteosarcoma risk and outcomes in East Chinese population. Med Sci Monit, 2017; 23: 3185–91
19. Cao Y, Lindstrom S, Schumacher F et al: Insulin-like growth factor pathway genetic polymorphisms, circulating IGF1 and IGFBP3, and prostate cancer survival. J Natl Cancer Inst, 2014; 106(6): u85
20. Costa-Silva DR, da Conceição Barros-Oliveira M, Borges RS et al. Insulin-like growth factor 1 gene polymorphism in women with breast cancer. Med Oncol, 2017; 34(4): 59
21. Zhang SW, Wan WG, Miao HX et al: Leptocarpin suppresses proliferation, migration, and invasion of human osteosarcoma by targeting Type-1 insulin-like growth factor receptor (IGF-1R). Med Sci Monit, 2017; 23: 4132–40

Indexed in: [Current Contents/Clinical Medicine] [SCI Expanded] [ISI Alerting System] [ISI Journals Master List] [Index Medicus/MEDLINE] [EMBASE/Excerpta Medica] [Chemical Abstracts/CAS] [Index Copernicus]