The Synergy of Diverse Treatment Strategies: A Focus of Research and a Promising Path for Osteosarcoma Management

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ABSTRACT

Osteosarcoma begins in the long bones that occurs mainly in children and young adults but it is also seen in an older population with high mortality rate. Zinc beryllium silicate and beryllium oxide, radioactive agents like radium and some C-type viruses have a harsh impact on osteosarcoma, Genetic factors like changes in the p53 tumor suppressor gene, mutation in RECQL4 gene, WRN gene can contribute in the osteosarcoma. Osteosarcoma tumor can be categorized as high grade, intermediate grade, and low grade depending on the degree and duration of tumor growth. Based on the nature of the pathogenesis and location of tumor high grade osteosarcoma can be Osteoblastic, Chondroblastic, Fibroblastic, Telangiectatic, Desmoplastic fibroma, Extraskeletal osteosarcoma; Periosteal Osteosarcoma are of intermediate grade. Parosteal osteosarcoma is a variant of osteosarcoma which is a type of low grade. Paget disease can become malignant and form osteosarcoma. It is associated with some characteristic symptoms like bone pain, formation of lump, walking with difficulties that leading pathologic fractures. Osteosarcoma can be diagnosed by Biopsy, X-ray, MRI, and CT scan and from some biochemical markers like Lactate Dehydrogenase and Alkaline Phosphatase. Tumor staging is based on the location of tumor, whether it is within the bone or outside of a bone, status of metastasis, regional lymph node involvement. Treatment include surgery which has wide safety margin, chemotherapy with Cisplatin and Doxorubicin after that Methotrexate, Adiramycin, Cisplatin. Pre-operative chemotherapy is given 2–6 cycles for 6–18 weeks and radiotherapy. Some Phytochemicals like curcumin, genistein, berberine has a positive impact on osteosarcoma tissue. After completion of treatment patient should be followed up to 6 year with timely surveillance.

Keywords: Osteosarcoma; RECQL4 gene; WRN gene; Phytochemicals; Tumor

Introduction

Osteosarcoma is a very rare type of bone cancer, begins in the osteoblast cells of the long bones which occurs mainly in children and young adults. All OS contain varying amounts of osteoid, most of which are constituted of cartilage and fibrous tissues [1]. Depending on its type, if a particular cell type constitutes 50% of a malignant tumor, that tumor is considered to be an osteoblast, chondroblast, or fibroblast. It is reported higher in male child than in female; it is the eighth most common childhood malignancy but mortality is higher in older individuals. The incidence rate of osteosarcoma within 14 year age range is four cases per year per million people; In the 0 to 19 year age range is five cases per year per million people; in the age group of older than 65, when the appearance of osteosarcoma is more likely to represent secondary cancer resulting from malignant degeneration of Paget disease, sites of bone infarction, etc[2].

Established Etiology of Osteosarcoma

Pathophysiological background of osteosarcoma though unpredictable, but it may has some contributing factors, like genetics, epidemiology, certain viruses. Chemical compound named zinc beryllium silicate and beryllium oxide has shown to produce atrophy in the spleen followed by osteosarcoma in rabbit model or previously surgically removed spleen in rabbit [3]. Another radioactive chemical like Plutonium-Thorotrast injection has shown to develop osteosarcoma practically upto 4 years of radiation [4]. Certain viruses can also contribute in osteosarcoma These viruses are classified based on their genetic substances RNA and DNA. The C type viruses are associated with leukaemia, sarcoma, lymphoma in animal species. In some animals, there are two C-type of viruses that are ecotropic and xenotropic are found; ecotropic virus can infect same species but xenotropic virus infect different species. FBJ virus was isolated from a spontaneously developing osteosarcoma in a mouse and it can induce osteosarcoma in new born mice even if it diluted to thousand folds [5]. Hereditary factors including genetic aberrations in the case of primary osteosarcoma Retinoblastoma start from a condition called leukocoria; symptoms are cloudy iris, poor vision and also it causes, devoid of the "red reflex" in the eye of the patient, this disorder is caused by genetic mutation. Li-Fraumeni Syndrome is a dominant autosomal disorder due to radiation therapy changes in the p53 tumor suppressor gene. This has been found to causes children with osteosarcoma [6]. Patients having this disorder are at a high risk of developing other types of cancer at a very young age. Rothmund-Thompson Syndrome because of a mutation in the RECQL4 gene, symptoms like early onset of poikiloderma, with characteristic rash, changes in the skin colouring, alopecia, hypogonadism, cataracts, ultimately leads to osteosarcoma [7]. Bloom Syndrome is characterized by photosensitivity, short stature. If this syndrome is predisposed with or without other cancers, these allows better surveillance of genetic testing family members. Werner syndrome or adult progeria, is characterized by untimely aging, cataracts, osteoporosis, short stature, skin changes, and prevalence of osteosarcoma caused by faulty WRN gene [8]. Patients with Paget disease, electrical burns, alkylating agents, orthopedic prosthetics, osteochondromatosis, enchondromatosis, as well as bone infection. Osteosarcoma is shown to be developed from a trauma related injury, there are some cases of osteosarcoma development at the site of bullet shot. Diamond Blackfan anaemia can also turn into osteosarcoma characterized by blood cell aplasia, congenital abnormalities in the ribosomal protein formation due to genetic mutation upto nine genes. Mostly mutation occurs in the

RPS19 gene in 19q13.2 zone. Sometimes there is mutation in the RPL5 site along with blocking the action of the FLVCR1 causes apoptosis of erythroid progenitor and mutation in the GATA1 maturation factor. Mutated GATA1 maturation factor and activated P53 gene may ultimately causes Diamond Blackfan anaemia. Paget disease which is another suspected cause of osteosarcoma, occurs in elderly male and in more than 40 years of age. Abnormal osteoclastogenesis and osteoblastogenesis may occur due to defected and mutated growth factors. It has been found that the gene name SQSTM1 it the culprit here to cause Interferon-induced RANK signaling and osteoporosis thus the neoplasia formed. By studying the genomic sequencing from the osteosarcoma germline it was seen that there are single nucleotide variation or structural variation of the TP53, RB1, ATRX, DLG2 gene locus. Transcriptome and genome analysis shows that rearrangements of TP53, RB1, MDM2 and CDKN2A as well as PMP22-ELOVL5 gene fusions and the most frequent TP53 rearrangements (e.g., TP53-VAV1, TP53-EMR1, TP53-PPRAD and TP53-KPNA3) resulted in the inactivation of p53 in osteosarcoma as well as the cessation of cell death in osteosarcoma.

Epidemiology

Osteosarcoma is a neoplasia in long bones, and usually reported with 4.7 per million cases per year and 8.9% of lethality rate. As it is a childhood and pubertal disorder, the chances of healing increases with near about 65 percent. It has been seen from the genomic study of osteosarcoma that African American and Latino populations are mainly affected from the disease. Sometimes the osteosarcoma can occur for the radiation applied to treat other different types of cancers. Mainly long bones are affected like humerus, radioulna and tibiofibula affects with maximum occurrence in femur with 42 percent. A very few 1.25% of osteosarcoma are found in the ribs and fingers.

Classification

The disease can be classified based on their degree and duration of tumor growth, it can be categorized as high grade, inter-mediate grade, and low grade. Based on the nature of the pathogenesis and location of tumor it can be Osteoblastic (tumor cells may be osseous), Chondroblastic (tumor cells may be cartilaginous) and Fibroblastic (fibrous tissues may affect) [1,9]. Tumor, which is of large blood-filled mass and separated by thin bone it is called Telangiectatic [10]. If fibrous stroma is embedded with osseous matrix then they are generally termed as Desmoplastic fibroma; it can be seen in jaw bone, pelvic bone. It has slow growth rate but with the aggressive characteristics. Extraskeletal osteosarcoma grows as a painless tumor in the soft tissue and then produce osteoid. The above mentioned types of osteosarcoma are of high grade osteosarcoma that metastasize fast to the other parts of body. Periosteal Osteosarcoma are of intermediate grade, arises from inner layer of periosteum. Parosteal Osteosarcoma is a variant of osteosarcoma which is a type of low grade occurs on the outer protecting membrane of long bone and it remains localized. Paget disease can become malignant and form Osteosarcoma [11].

Symptoms

Symptoms of osteosarcoma are bone pain, sprain, lump formation; pain increases during physical activity, fever, and paralysis. It is reported that pain increases in night with walking

difficulties that leads to pathologic fractures, sometimes associated with telangiectasia. In Osteosarcoma, there is a chance of rapid growth of bone. In low grade, tumor develop at its place and reside locally; in lung metastasis respiratory symptoms like cough, excess mucous formation, wheezing, and chest pain are found. Osteosarcoma has low background frequency with some other disease. Fibrosarcoma can be diagnosed histologically by presence of fibroblast tissue that invade long bones Osteoblastoma is a large osteoid with vascular leisons. Lymphoma, that increase lymphocyte formation uncontrollably that causes Hodgkin disease for middle aged population and non-hodgkin disease for older population. There are some sarcomas like Ewing sarcoma in which the reason is still unknown [12].

Pathogenesis of Osteosarcoma

Diagnosis

Physical examination shows that patient is having stiff joint, localized pain and warmth, lump, palpable mass, pathological fractures of some patients; and some characteristic symptom of cancer like unwanted weight loss, fever, malaise also appear.

Laboratory diagnosis of Lactate Dehydrogenase (LDH) and Alkaline Phosphatase (ALP) are two Biochemical markers are assessed in the initial diagnosis. ALP levels become elevated by around 40 percent, because of the increased osteoblastic activity which are often associated with osteosarcoma; levels may lower with treatment or rise with residual disease or recurrence. X-ray views of the suspected lump area of whole bone and nearby joints show contusion at the metaphysis of the long bone; Diagnosis with X-ray followed by biopsy can confirm the osteosarcoma diagnosis, X-Ray of chest should also be done to assess the lung metastasis

Magnetic resonance imaging (MRI) is used to assess in details of the contusion and invasion and its extension into the bones and soft tissue and vascular and neural structures of it; skip metastasis which are fragments of bone tumor present in some distance from the primary tumor. MRI with contrast revel those tumor which are situated within intra-articularly.

Computed tomography (CT) scans are useful in biopsy planning and revealing in abnormal irregularities, fracture and also mineralization in cortical bones. The extant and stage of tumor invasion is accurately detectable by MRI. Positron emission tomography scans can detect the primary metabolic sites and also detect metastatic, and has obvious advantages in showing osteosarcoma. Periosteal reactions are not well visible through CT [13].

Biomarker

There is only one biomarker that is helpful to diagnose the phenotype of osteosarcoma called SATB2 but is only helps to identify the osteoblastic phenotype but is incapable to determine the neoplasia is categorized under malignant or benign [14].

Biopsy is an essential procedure after diagnosis, laboratory tests to confirm the presence of a lesion of osteosarcoma .This procedure helps to avoid timely detection and treatment of patients. The surgical procedure must have abscission of the biopsy tract, for identification. Analysis of Biopsy sample gives more accurate knowledge of the pattern and development of lesions. The common biopsy method is incisional biopsy and trucut biopsy. Puncture biopsy is usually performed for 2–10 min to cease the bleeding. Biopsy should be carefully approached

to suppress the chances for the tumor cells to spread in the biopsy tract and nearby other tissues [15].

Stages of Tumor

Orthopedic surgeons are concerned with the anatomical position of the tumor; whether the tumor is within the bone or outside of a bone. they also concerned with the size of the tumor with its treatment response and overall survival. Tumor in Stage IA is low grade, intra compartmental tumor location with no involvement in regional lymph nodes and no metastasis. in Stage IB it is also low grade, extra compartmental tumor location with no involvement in regional lymph nodes and no metastasis. Stage IIA consist of High grade, intra compartmental tumor location, regional lymph nodes are free from tumor spreading and no metastasis. Stage IIB is High grade, extra compartmental tumor location,no regional lymph node involvement no metastasis. Stage III is any grade, any location, metastasis present. Stage IVA: Tumor grade and size could be anything but no lymph node involvement, lung metastasis; Stage IVB: Metastasis in regional lymph node, lung or extra pulmonary region [16].

Treatment

Surgical method

Tissue of Osteosarcoma can be completely excised surgically with a wide safety margin; there are two approaches, limb salvage and amputation for removing the lesion and also its trace to prevent residual disease. Limb salvage is a surgical procedure that provide a safe way for the Osteosarcoma treatment. Limb salvage is consist of resection and reconstruction. The intention of the resection is to eliminate the primary biopsy site. Diagnostic imaging technology, such as bone scan, should be used to determine the quantity of bone to be resected to prevent residual disease; the recurrence of disease is due to poor response of disease. Resection followed by examination of that part to see the effect of chemotherapy on that tumor. The process of reconstruction is mainly utilized for those bones which can heavy carry weight. Reconstruction surgery is two type endoprosthetic replacement and biological reconstruction. The first one is a form of limb salvage reconstruction with a good functional outcomes [15,16]. There are some challenges for the surgical procedure like excision followed by physeal resection lead to create interruption in the child's growth and maturity. In the knee osteosarcoma surgery, the mass around the joint create obstacle for the surgeon; resection followed by tissue regeneration have overcome this challenge. Metallic prosthetics brought new light in surgical reconstruction; it provide large bone and joint replacement. The incidence rate of recurrences in local area after amputations and limb salvage are more or less similar, still limb salvage patients have a higher 5-year survival rate. Limb salvage surgery protects the structural and functional integrity of patient. The surgery should completely eliminate the lesion to prevent local recurrence as well as distant metastasis. If not the recurrence rate can increase upto 25%

There are few complications in the Prostheses like local or systemic infections which are frequent. the prevention for it is antibiotic therapy during lengthy surgery systemic antibiotics are used and after surgery local antibiotics are used [17].

When reconstructing the joint surface it is recommended to use the allograft prosthetic combination can be used to reconstruct the more stable periarticular soft tissue by the allograft and the prosthetic side create the more stable and joint articulation.

Chemotherapy

In Osteosarcoma treatment chemotherapy are used in adjuvant setting after surgery to eliminate the formation of lesions and metastases that could not be completely removed by surgery alone. Preoperative chemotherapy was successfully applied in hospital; this approach is known as neoadjuvant chemotherapy. It is a landmark in the treatment of Osteosarcoma, The chemotherapeutic drugs for OS are adriamycin, methotrexate, cisplatin, and ifosfamide; all of these drugs has significant efficacy. Combining those drugs with the different doses and frequency of administration, that can produce an effective chemotherapy regimen; e.g. Combination of Cisplatin and Doxorubicin (first line) Adjuvant MAP therapy is composed of Methotrexate, Adiramycin, Cisplatin. Pre-operative chemotherapy is given 2–6 cycles for 6–18 weeks[14] If the chemotherapy is responded well less than 10% tumor content, those patient would continue their regimen; and Prescriber must change the combination of drug of patient with poor response The adverse effect of that chemotherapy include suppression of bone marrow, neurotoxicity, nephrotoxitcity, hepatotoxicity, gastrointestinal disturbances. Side effect of chemotherapy treatment related process are included nausea, vomiting, pain at the site of chemotherapy, alopecia etc [12,18,19].

Radiotherapy

In non- metastatic Osteosarcoma, local radiotherapy may create a positive impact. Studies found that external beam radiation therapy with the systemic therapy or induction chemotherapy may relief local symptom. It is used for those patient who cannot undergone resection or post- operative patient whose cancerous tissue is not incompletely removed. In Metastatic Disease Samarium 153-EDTMP Stereotactic radiosurgery (SRS) can be used. Side effect of radiation include dryness of skin and inside of the vagina (in case of brachytherapy), dry mouth, itchiness which can be ameliorate by hydration, menstrual cycle changes etc [20].

Immunotherapy

Body's immune system consist of innate immunity that is the natural immunity of body and adaptive immunity which is the slow but long established immunity. In adaptive immunity T-cells plays the major role. In cancer therapy T- cells are utilized in the cancer cell vaccine. Dendritic cell is an antigen found on T-cell, collected from patient's blood used as cancer vaccine that has ability to slow down cancer progression [20].

Chimeric Antigen receptor T cells (CAR T-cells) are genetically modified T cells are injected in HER2 positive patient in a trial, after 12 to 15 weeks of treatment most of the patient shows stable response which means tumor size is not increasing by 20% nor decreasing by 20%

Immunosuppressive protein PD1 which is a Programmed death protein reside on the T cells and B cells. It induce treatment resistance. Programmed cell death ligand 1 (PDL1) is its ligand. PD1/PDL1 overexpressed in the tumor micro environment and it is an immune check point that cause treatment resistance. Inhibiting PD1/PDL1 checkpoint pathway can be a promising therapy for cancer treatment. Recent Clinical trial of PD-1 inhibitor camrelizumab shows positive response in lung metastasized osteosarcoma treatment, without showing a serious toxicities [20,21].

Gene therapy

Genetic mutation is the foremost reason of Osteosarcoma. Gene therapy attempt to replace the defective or mutated genes with normal gene through a viral or non-viral vector that produce beneficiary effect. In case of cancer, It is the result of a series of genetic alteration like rearrangements of one base, or deletions ,So the target is the pivotal gene example p53 or pRb in the series of gene alteration in cance.P53 gene it can cause increase chemtherapy sensitivity, and it is a biomarker of the overall survival of the patient .The study of thymidine kinase along with propoxyguanosine approach in the cell line of Osteosarcoma can induce apoptosis and hinder the growth of the tumor cell line . This synergistic gene therapy improve the condition significantly. Combining other treatment methods with this gene therapy can be a good strategy in the treating of Osteosarcoma patients [21].

Phytochemicals

Some plant derived compounds has anti-tumor properties, they have potential to undergo clinical trial process as a supplementary of standard treatment of chemotherapy example Curcumin is a natural polyphenol derived from curcuma longa. It has potential to induce apoptosis by activating various gene like MG63, U2OS and HOS. Genistein is an isoflavonoid, isolated from *Genista tinctoria*. Song and colleagues investigated that Genistein affect the PPARγ signalling, which has negative impact on Osteosarcoma proliferation. It also inhibit MG-63 genes, which is associated with Osteosarcoma. Growth of Osteosarcoma is reported to be suppressed by another phytochemical called berberine; it cause down regulation of caspase-1 and IL-1β in the tumor of Osteosarcoma. Berberine also reduce expression of E-cadherin, which is high in Osteosarcoma [14].

Follow-up & Monitoring

Post-operation, follow up visit for patient would be in every three months for 1 to 2 years; after that in every four months for 3 years, then in every six months for 4 and 5 years; from the post-operative sixth year, follow-up visit would be once in a year. The follow up visit should include the physical assessment and Quality of Life (QOL) questionnaire. Imaging the site of operation and thorax through PET CT or CT scan or bone scan to detect any lesions in lungs and pathological tests should be done that include Complete Blood Count (CBC), Liver Function test, Alkaline Phosphatase test. If any lesions or relapse is detected, the physician should start Chemotherapy or resection or palliative radiation depending on patient's condition [15].

Prognosis

The young adult patient population have highest survival rates than middle-age patient population (over 40 years old), patients on the middle age group are more vulnerable to get metastatic lesions in axial skeleton. Patients in the age group of 60 years or beyond, their body show refusal of surgery or chemotherapy. Osteosarcoma patient with pathological fractures cause increase in mortality. Chondroid tumor indicate unfavourable in histology; whereas fibroblastic tumor shows good response. Different studies found that Men show higher chances of recurrence than women because of their less sensitivity for chemo therapy that increase the morbidity. Biomarker like Alkaline phosphatase (ALP) and Lactate dehydrogenase (LDH) are two important biomarker for the Osteosarcoma patient. ALP promotes bone mineralization and associated with the bone resorption, this biomarker may be or may not be in normal level in

the time of diagnosis but it is elevated later on in Osteosarcoma patient. Serum LDH become higher in level in metastatic patients. Rate of survival of Osteosarcoma patients are also depend on the location and volume of tumor. If tumors are diagnosed in the bones of skull, neck, vertebrae, sacrum and tailbone region; that means axial skeleton shows worse prognosis than to those in the bones of shoulder, pelvis, arms, leg region that means appendicular skeleleton. Tumor with larger volume cause lack of response in chemotherapy that leads to recurrence [11].

Discussion

Osteosarcoma, a rare form of bone cancer, has seen promising progress in treatment approaches over time. Scientists and medical researchers are working diligently to develop novel therapeutic strategies to combat this aggressive disease. In recent years, a combination of traditional methods, such as surgical intervention and neo adjuvant chemotherapy, with innovative approaches like immunotherapy and gene therapy, has shown great potential in improving patient outcomes.

Immunotherapy and gene therapy are emerging as groundbreaking methods for managing osteosarcoma. By leveraging the body's immune system to identify and attack cancer cells, immunotherapy aims to enhance the body's natural defenses against the tumor. Gene therapy, on the other hand, involves modifying the patient's genetic material to target and inhibit cancerous cells more effectively. The integration of these modern treatments with established protocols like surgical methods and neo adjuvant chemotherapy can help overcome chemotherapy resistance, ultimately leading to better treatment responses.

The combined treatment regimen has demonstrated promising results, leading to increased progression-free survival rates and an overall improvement in the quality of life for osteosarcoma patients. The synergy of different therapeutic modalities offers a more comprehensive approach to tackling the disease, resulting in more favorable long-term outcomes.

In addition to exploring innovative treatment methods, there have been intriguing proposals for new reconstructive techniques. Among them is the use of liquid nitrogen to destroy tumor cells, followed by the introduction of the patient's own cells to replace the affected tissue. This method, known as frozen autograft, involves a surgical procedure where the neoplast is frozen with liquefied nitrogen and then reconstructed with plates in a process termed arthroplasty. Not only can the frozen autograft approach potentially be a cost-effective method, but its efficacy also warrants further investigation and study to evaluate its long-term benefits.

However, while progress is evident, challenges still remain, particularly in tackling lung metastasis, which remains a concerning aspect of osteosarcoma treatment. Timely and precise diagnosis is of paramount importance to ensure that patients receive the most appropriate and effective treatment promptly.

As research and medical advancements continue, the collaborative efforts of healthcare professionals, scientists, and patients in the fight against osteosarcoma are slowly but steadily making strides. By combining cutting-edge therapies with established treatments and focusing on early detection, there is hope that the management of osteosarcoma will continue to improve, leading to better outcomes and an improved quality of life for affected individuals.

Conclusion

The synergistic combination of various treatment strategies has captured the attention of researchers, as it holds the potential to yield optimal outcomes within the patient's body. We firmly believe that moving forward, osteosarcoma can be treated more holistically, leading to a substantial improvement in the quality of life for affected individuals.

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References

1. Lindsey BA, Markel JE, Kleinerman ES (2017). Osteosarcoma Overview. Rheumatol Ther. 4(1):25-43.
2. Widhe B, Widhe T (2000). Initial symptoms and clinical features in osteosarcoma and Ewing sarcoma. J Bone Joint Surg Am. 82(5):667-74.
3. Wadhwa N (2014). Osteosarcoma: Diagnostic dilemmas in histopathology and prognostic factors. Indian J Orthop. 48(3):247-54.
4. Gorlick R (2009). Current concepts on the molecular biology of osteosarcoma. Cancer Treat Res. 152:467–78.
5. Carrle D, Bielack SS (2006). Current strategies of chemotherapy in osteosarcoma. Int Orthop. 30:445–51.
6. Hameed M, Mandelker D (2008). Tumor Syndromes Predisposing to Osteosarcoma. Adv Anat Pathol. 25(4):217-222.
7. Merlin R, Hamrea,b, Richard K. Seversonb (2002). Osteosarcoma as a second malignant neoplasm, Radiotherapy and Oncology. 65:153–157.
8. Ambroszkiewicz J, Gajewska J, Klepacka T, Chelchowska M, Laskowska-Klita T, Wozniak W (2010). Clinical utility of biochemical bone turnover markers in children and adolescents with osteosarcoma. Adv Med Sci. 55:266–72.
9. Kundu JS (2014). Classification, imaging, biopsy and staging of osteosarcoma. Indian J Orthop. 48(3):238-246.
10. Provisor AJ, Ettinger LJ, Nachman JB, Krailo MD, Makley JT, Yunis EJ, Huvos AG, Betcher DL, Baum ES, Kisker CT, Miser JS (1997). Treatment of nonmetastatic osteosarcoma of the extremity with preoperative and postoperative chemotherapy: a report from the Children's Cancer Group. J Clin Oncol. 15:76–84.
11. Abramson DH, Ellsworth RM, Zimmerman LE (1976). Nonocular cancer in retinoblastoma survivors. Trans Sect Ophthalmol Am Acad Ophthalmol Otolaryngol. 81:454–457.
12. Murphey MD, Wan Jaovisidha S, Temple HT, Gannon FH, Jelinek JS, Malawer MM (2003). Telangiectatic osteosarcoma: radiologic-pathologic comparison. Radiology. 229:545–553.
13. Sim FH, Unni KK, Beabout JW, Dahlin DC (1979). Osteosarcoma with small cells simulating Ewing’s tumor. J Bone Joint Surg Am. 61:207–215.
14. Nakajima H, Sim FH, Bond JR, Unni KK (1997). Small cell osteosarcoma of bone. Review of 72 cases. Cancer. 79:2095–2106.
15. Kumar VS, Barwar N, Khan SA (2014). Surface osteosarcomas: Diagnosis, treatment and outcome Indian J Orthop. 48(3):255–261.

16. Shuman WP, Patton AM, Baron AL, Uddell AM, Conrad EU, Richarson ML (1991). Comparison of STIR and spin-echo MA imaging at 1.5 T in 45 suspected extremity tumors: lesion conspicuity and extent. Radiology. 179:247–252.
17. Hong AM, Millington S, Ahern V, Mccowage G, Boyle R, Tattersall M, Haydu L, Stalley PD (2013). Limb preservation surgery with extracorporeal irradiation in the management of malignant bone tumor: the oncological outcomes of 101 patients. Ann Oncol. 24:2676–2680.
18. Betz M, Dumont CE, Fuchs B, Ulrich Exner G (2012). Physeal distraction for joint preservation in malignant metaphyseal bone tumors in children. Clin Orthop Relat Res. 470:1749–1754.
19. Wong KC, Kumta SM, Chiu KH, Antonio GE, Unwin P, Leung KS (2007). Precision tumour resection and reconstruction using image-guided computer navigation. J Bone Joint Surg Br. 89:943–947.
20. Gosheger G, Goetze C, Hardes J, Joosten U, Winkelmann W, von Eiff C (2008). The influence of the alloy of megaprostheses on infection rate. J Arthroplasty. 23:9–12.
21. Shalaby S, Shalaby H, Bassiony A (2006). Limb salvage for osteosarcoma of the distal tibia with resection arthrodesis, autogenous fibular graft and Ilizarov external fixator. J Bone Joint Surg Br. 88-B:1642–1646.