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Original Article

What has changed in the last 25 years in osteosarcoma treatment? A single center experience

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ABSTRACT

Background. Osteosarcoma is the most common type of primary malignant bone tumor in the extremities. The main purpose of this study was to determine clinical features, prognostic factors, and treatment results of patients with osteosarcoma at our center.

Methods. We retrospectively analyzed the medical records of children with osteosarcoma between the years 1994-2020.

Results. 79 patients were identified (54.4% male, 45.6% female). The most common primary site was the femur (62%). Twenty-six of them (32.9%) had lung metastasis at diagnosis. The patients were treated between 1995-2013 according to the Mayo Pilot II Study protocol, while the others were treated with the EURAMOS protocol between the years 2013-2020. Sixty-nine patients underwent limb salvage surgery as a local treatment, whereas seven underwent amputation. The median follow-up time was 53 months (2.5-265 months). The event-free survival (EFS) and overall survival (OS) rates at 5 years were 52.1% and 61.5%. The 5-year EFS and OS rates were 69.4% and 80% in females; 37.1% and 45.5% in males (p=0.008/p=0.001). The 5-year EFS and OS rates of the patients without metastasis were 63.2% and 66.3%; with metastasis 28.8% and 51.8% (p=0.002/p=0.05). For good-responders, the 5-year EFS and OS rates were 80.2% and 89.1%; while for poor-responders, 35% and 46.7% (p=0.001). Mifamurtide was used in addition to chemotherapy as of the year 2016 (n=16). The 5-year EFS and OS rates were 78.8% and 91.7%, respectively for the mifamurtide group; 55.1% and 45.9%, respectively for the non-mifamurtide group (p=0.015, p=0.027).

Conclusions. Metastasis at diagnosis and poor response to preoperative chemotherapy were the most important predictors of survival. Females had a better outcome than males. In our study group, the mifamurtide group's survival rates were significantly higher. Further large studies are needed to validate the efficacy of mifamurtide.

Key words: osteosarcoma, childhood, treatment, surgery, chemotherapy.

Osteosarcoma is the most common type of primary malignant bone tumor in children. Osteosarcoma accounts for approximately 3% of all pediatric malignancies. The incidence rises with age and reaches a peak incidence during puberty.[1] The tumor usually arises from the extremities and especially from the long bones' metaphyseal region. The most common site is the distal femur, followed by the proximal tibia. However, axial bones (pelvis, vertebra, head bones) can also be involved. Osteosarcomas are high-grade malignancies, and 15-17% of patients usually have lung metastasis at diagnosis.[2]

Current treatment for this aggressive tumor is neoadjuvant multiagent chemotherapy followed

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by surgery and adjuvant chemotherapy. With this multimodal treatment, 5-year event-free survival (EFS) rate is about 60-70%. Cisplatin, doxorubicin, methotrexate, and in some regimens ifosfamide are the main drugs of this combination chemotherapy.[3-7]

In surgery, resection of the tumor with wide margins is important. With the introduction of neoadjuvant multiagent chemotherapy, limb-sparing surgery rather than amputation is the treatment of choice in most extremity tumors.[8] After resection, patients usually undergo external prosthesis replacement. However, in some cases, extracorporeal irradiation (ECI) and reimplantation have been preferred in recent years.

Adjuvant chemotherapy after surgery usually depends on the histologic response of the patient. Treatment protocols include similar drugs for good responders, but for poor responders, it is controversial. Which drug is effective for the latter group is not well-established. Current prospective trials evaluate whether altering postoperative chemotherapy in poor responders improves outcomes.

There has been little improvement in the survival rates of osteosarcoma patients in more than three decades. Therefore, novel strategies are needed to improve survival. Mifamurtide is a synthetic lipophilic analog of muramyl dipeptide. This molecule acts as an immunostimulant with an anti-tumor effect. In recent years, the addition of immunostimulant mifamurtide after surgery to postoperative chemotherapy has been reported to have a significant effect on the overall survival of non-metastatic patients, however, it is yet to be answered for metastatic patients.[9,10]

The main purposes of this study were to share our treatment experience, and to document demographic characteristics, clinical features, and prognostic factors of non-metastatic and metastatic patients with osteosarcoma of the extremities treated at our center.

Material and Methods

We retrospectively analyzed 79 children with extremity osteosarcoma treated at Ege University Hospital between the years 1994 and 2020. All patients underwent an initial tru-cut biopsy for definitive diagnosis at the Department of Orthopedics. The extent of the disease was evaluated by magnetic resonance imaging of the lesion, computerized tomography of the chest, and a radionucleotide bone scan.

All patients were treated according to the Mayo Pilot II protocol between the years 1995 and 2013 or EURAMOS protocol between 2013 and 2020. As per the Mayo Pilot II study, patients received cisplatin (120 mg/m^2/day; week 10) and doxorubicin (25 mg/m^2/day x 3, week 0, 5), ifosfamide (1.8 g/m^2/day x 5, week 0, 5, 10), and high-dose methotrexate (12 g/m^2 week 3, 4, 8, 9, 13, 14) with leucovorin rescue. Surgery was carried out at around week 15 or earlier if tumor progression was seen based on clinical and radiological findings. The surgery aimed to remove the tumor and achieve wide margins. Limb-sparing surgery was the treatment of choice. Amputation was restricted to those for whom limb-sparing surgery could not yield wide margins or adequate function. The Huvos necrosis grading system was used in histopathological evaluation to assess chemotherapy response.[11] Based on the percentage of tumor necrosis after chemotherapy, patients can be classified as poor or good responders. The patients who achieved at least 90% of tumor necrosis in the resected specimen were categorized as good responders. They continued to receive similar postoperative chemotherapy to complete 42 weeks. Poor responders (less than 90% tumor necrosis) received the same regimen before 1996, but high-dose ifosfamide alone (14 g/m^2/day over 3.5 days, in 21-day intervals) after this year.

In the EURAMOS study protocol, all patients were planned for the same pre-operative therapy for 10 weeks consisting of 120 mg/m^2 of cisplatin and 75 mg/m^2 of doxorubicin (weeks 1 and 6)

followed by 12 g/m^2 of high-dose methotrexate (weeks 4, 5, 9 and 10). Surgery was carried out at around weeks 11-12. Patients with a good histological response (≥ 90% of tumor necrosis) were continued with postoperative therapy for 29 weeks consisting of 120 mg/m^2 of cisplatin (weeks 12 and 17), 75 mg/m^2 of doxorubicin (weeks 12, 17, 22, 26) followed by 12 g/m^2 of high-dose methotrexate (weeks 15, 16, 20, 21, 24, 25, 28 and 29). The patients with a poor histological response (<90% of tumor necrosis) were continued with postoperative therapy for 40 weeks with cisplatin, doxorubicin, and high-dose methotrexate with additional ifosfamide and etoposide.

As an adjuvant therapy, only mifamurtide was used in addition to postoperative chemotherapy treatment after 2016. Mifamurtide was given 2 mg/m^2 twice weekly at least 3 days apart for 12 weeks, followed by once-weekly treatments for an additional 24 weeks for a total of 48 doses in 36 weeks.

The ethical committee of our institution approved the study (Ege University Faculty of Medicine, report number: 22-4T/55).

Statistical analysis

Data were analyzed using SPSS software (version 21 for Windows). Continuous variables are presented as means (ranges) and categorical variables as numbers (percentages). A p-value ≤ 0.05 was considered to indicate statistical significance. Kaplan-Meier survival analysis followed by log-rank tests were used to identify significant relationships among EFS, categorical variables, and overall survival (OS).

Results

Patients

There were 43 male and 36 female (M:F=1.2) patients in the study with a mean age of 13.1 ± 2.8 years (4.5-18 years). Demographic features of the patients are given in Table I. The most common primary tumor site was the femur (62%). According to WHO histologic classification, 49 patients had conventional osteoblastic osteosarcoma. Twenty-six out of 79 patients (32%) had metastasis at diagnosis, of whom 24 (92.3%) had pulmonary metastases, 2 (7.7%) had both pulmonary and bone metastases. Between 1995 and 2013, the patients were treated according to the Mayo Pilot II Study Protocol (n=59), and the EURAMOS treatment protocol between 2013 and 2020 (n=20) (Table II).

Table I. The demographic and clinical characteristics of patients (n=79).

Sex	
Male	43 (54.4%)
Female	36 (45.6%)
Age, years	
Mean ± SD	13.1± 2.8 years
< 10 years	10 (12.7%)
≥ 10 years	69 (87.3%)
Tumor site	
Femur	49 (62%)
Tibia	18 (22.8%)
Humerus	10 (12.7%)
Fibula	2 (2.5%)
WHO histological classification	
Osteoblastic	49 (62%)
Chondroblastic	15 (19%)
Fibroblastic	4 (5.1%)
Telangiectatic	4 (5.1%)
Others	7 (8.9%)
Metastasis	
Lung	24 (30.4%)
Lung + bone	2 (2.5%)
Treatment protocol	
Mayo Pilot II study	59 (74.7%)
EURAMOS study	20 (25.3%)
Histologic response	
Good (≥90%)	32 (40.5%)
Poor (<90%)	42 (53.2%)

Surgical Outcomes

Local control using surgical resection was planned after pre-operative chemotherapy. Upfront surgical resection was performed in

Table II. Demographic and clinical characteristics of patients according to the treatment groups (n=79).

	Mayo Pilot II Study Protocol (n=59)	EURAMOS (n=20)
Sex		
Male	33 (55.9%)	10 (50%)
Female	26 (44.1%)	10 (50%)
Age, years		
< 10 years	7 (11.9%)	3 (15%)
≥ 10 years	52 (88.1%)	17 (85%)
Metastasis		
Lung	19 (32.2%)	5 (25%)
Multifocal metastasis	2 (3.4%)	0
Surgery		
Limb salvage	49 (83.1%)	20 (100%)
Amputation	7 (11.9%)	0
Histologic response		
Good	21 (35.6%)	11 (55%)
Poor	33 (55.9%)	9 (45%)

two patients, and three patients (3.8%) were not operated on because of the tumor progression. The median time for surgery was 19.5 weeks in the patients treated with the Mayo Pilot II study and 14.5 weeks in patients treated with the EURAMOS protocol. Sixty-nine (87.3%) patients underwent limb salvage surgery, whereas seven patients (12.7%) underwent amputation. Out of these 69 patients, 63 patients (79.7%) experienced a prosthesis replacement following tumor resection. Six patients (7.6%) received extracorporeal irradiation and reimplantation of the bone. The number of patients undergoing limb-salvage surgery (n=20) increased in the EURAMOS group (83.1% vs. 100%). By contrast, the number of patients undergoing amputation (n=0) decreased (11.9% vs. 0%), but this was not statistically significant (p=0.18).

Seventy-four out of 79 patients were evaluated for histologic response. Of these patients, 32 (40.5%) had a good response, and 42 (53.2%) had a poor response. Comparison of the treatment responses of the two treatment groups revealed that the number of patients with good responses increased in the EURAMOS group (n=11) (55% in EURAMOS vs. 35.6% in Mayo Pilot II). Consequently, the poor responders (n=9) seemed to be less in the EURAMOS treatment group (45% in EURAMOS vs. 55.9% in Mayo Pilot II), but this was not significant (p=0.21).

Twenty-six patients with poor histologic response were given high-dose ifosfamide alone as a postoperative regimen after 1996. The other patients with a poor response and good responders were treated according to the postoperative chemotherapy regimen as mentioned in the protocols. Among the poor responders, there was no difference in EFS and OS rates with the addition of high-dose ifosfamide (p=0.33).

Survival

Median follow-up time was 53 months (2.5-265 months). The EFS and OS rates for all patients were 52.1% and 61.5% at 5 years, respectively (Fig. 1). The estimated 5-year EFS and OS rates were 49% and 57.3% for the Mayo Pilot II study group. The estimated 5-year EFS and OS rates were 60.9% and 71.8%, respectively for the EURAMOS treatment group (Fig. 2).

The females had significantly better outcomes than the males. The 5-year EFS rate was 69.4% in females versus 37.1% in males (p=0.008). The 5-year OS was 80% in females versus 45.5%

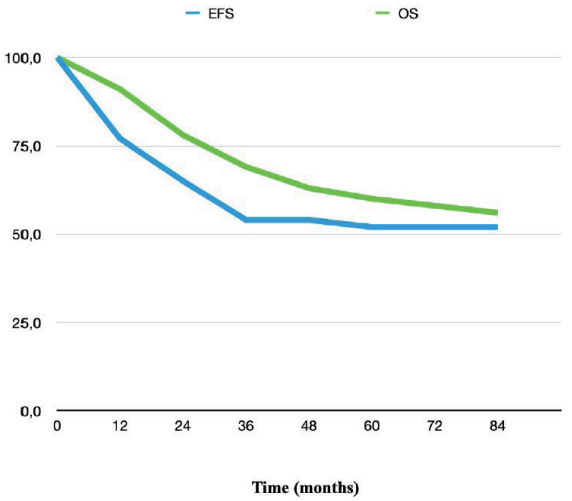

Fig. 1. Survival analysis of all patients.

in males (p=0.001). There was no significant relationship between age groups and survivals. The estimated 5-year OS rate was 42% in patients <10 years of age (n=10), and the estimated 5-year OS rate was 63.8% in patients ≥ 10 years of age (n=69) (p=0.28).

For the non-metastatic group the 5-year EFS rate was 63.2% while for the metastatic disease it was 28.8% (p=0.002). The 5-year OS rates was 66.3% for the non-metastatic group and 51.8% for metastatic patients (p=0.05) (Fig. 3).

Among the non-metastatic patients (n=53), 21 patients (39.6%) were good-responders and 29 patients (60.3%) poor-responders. Three patients did not have a pathological evaluation. Kaplan–Meier analysis showed that good responders have higher EFS at 5 years than poor responders (80.2% vs. 35%, p=0.001). The 5-year OS rates were 94.4% in the non-metastatic good-responders and 50.8% in the non-metastatic poor-responders (p=0.001).

Among the metastatic patients (n=26), 11 patients (42.3%) were good-responders and 13 patients (50%) were poor-responders. Two patients did not have a pathological evaluation. The 5-year EFS rates were higher at good-responders than those for poor-responders (51.1% vs. 15.4%, p=0.008). The 5-year OS rates were 77.9% in metastatic good responders and 38.5% in metastatic poor responders (p=0.04).

Among all patients, the estimated 5-year EFS and OS rates for good-responders were 80.2% and 89.1%, while for poor-responders the same rates were 35% and 46.7 % (p=0.001 vs. p=0.001) (Fig. 4.).

Disease progression occurred in 12 patients and relapse in 25 patients after treatment cessation. The median time to progression or relapse was 13 months (2.5-55 months). Among these 25 patients, 19 patients relapsed only with pulmonary metastases, three had a local plus pulmonary relapse, and three had distant bone plus pulmonary metastasis. Thirty-two patients (40.5%) died from the disease during follow-up. In the poor-response group (n=42), 18 patients relapsed, and 9 patients had progressive disease. In contrast, 6 patients relapsed in the good response group (n=32). We treated most of the poor responders (n=26) with high-dose ifosfamide in our study group. There was

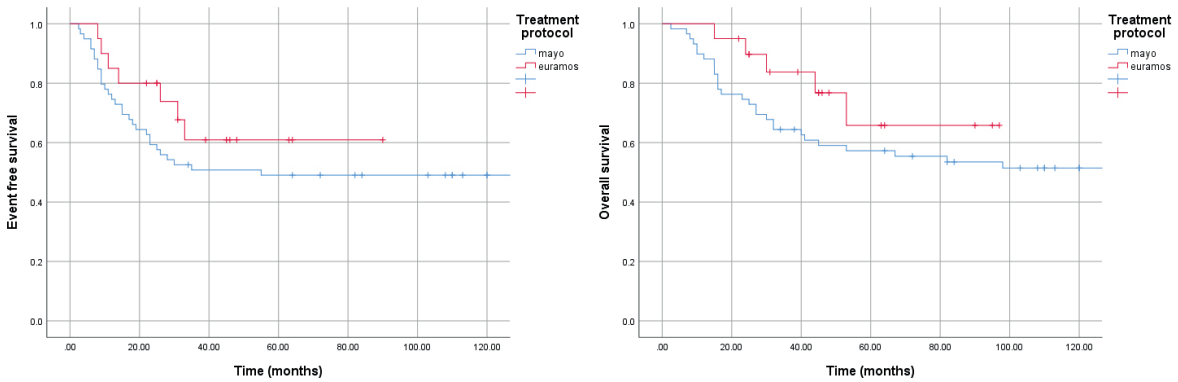

Fig. 2. Survival analysis of patients according to the treatment groups.

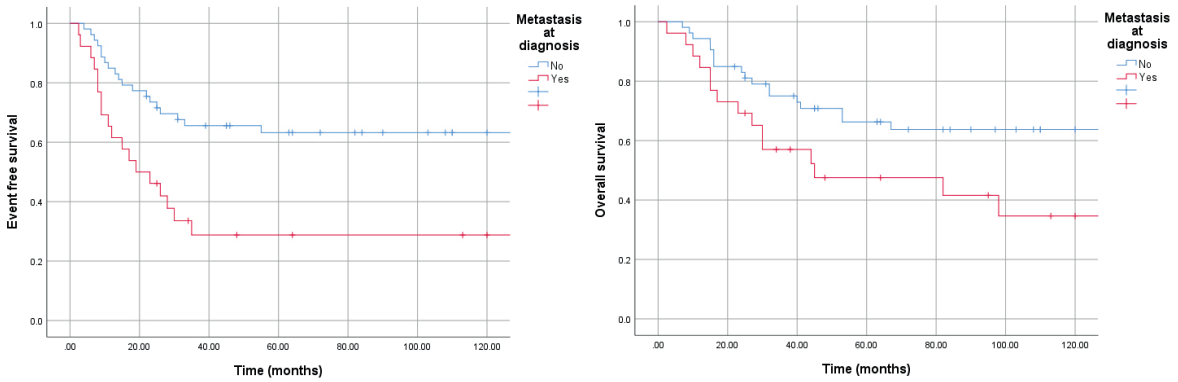

Fig. 3. Survival analysis of all patients according to the metastasis diagnosis.

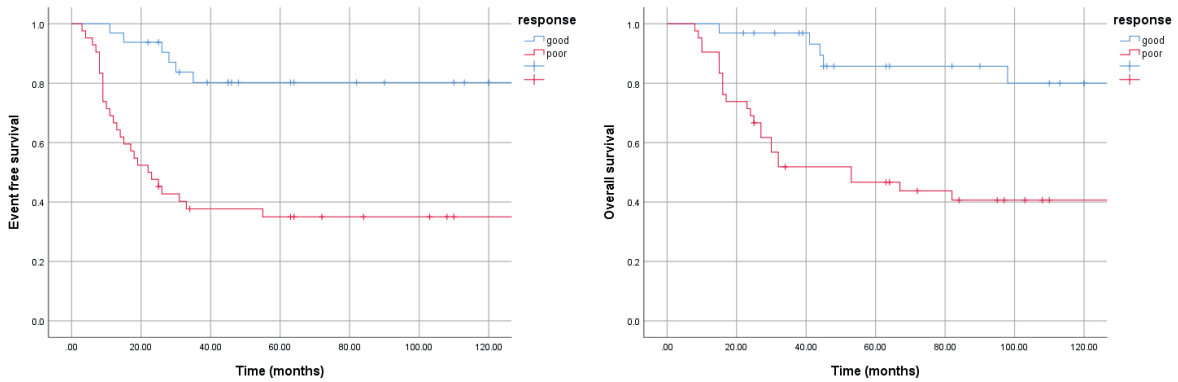

Fig. 4. Survival analysis of all patients according to the histologic response.

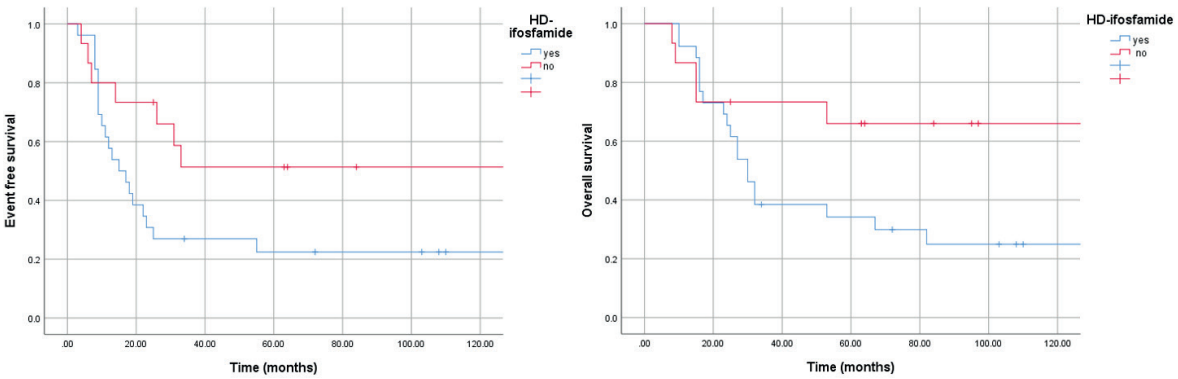

Fig. 5. Survival analysis of patients treated with HD-ifosfamide.

no difference in EFS and OS rates of patients treated with high-dose ifosfamide (Fig. 5).

Mifamurtide was given to 16 patients (10 female, 6 male). Of these, 13 had non-metastatic (8 good response, 5 poor response), and 3 had metastatic disease (all good response). The 5-year EFS and OS rates were 78.8% and 91.7% for the mifamurtide group, and 55.1% and 45.9%, for the non-mifamurtide group (p=0.015 vs. p=0.027), respectively. To evaluate the efficacy of mifamurtide treatment, we evaluated the patients in the EURAMOS treatment protocol because, in the Mayo Pilot II study protocol, only 2/59 patients received mifamurtide. In the EURAMOS treatment group (n=20), 5-year EFS and OS in the mifamurtide group (n=14) were

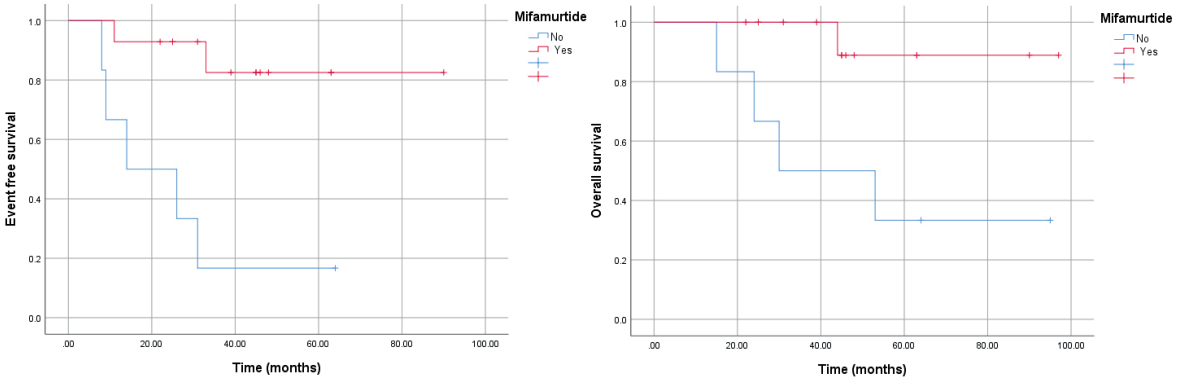

Fig. 6. Effect of mifamurtide in survival analysis of patients in EURAMOS treatment group.

82.5% and 100%, whereas 5-year EFS and OS in the non-mifamurtide group (n=6) were 16.7% and 33.3%, respectively (p=0.001 vs. p=0.003) (Fig. 6.). There were five patients (83.3%) with tumor relapse in the non-mifamurtide group and two patients (14.3%) with tumor relapse in the mifamurtide group (p=0.007). From these patients in the poor response group, 5-year EFS and OS in the mifamurtide group were 50% and 100%, while 5-year EFS and OS in the non-mifamurtide group were 16.7% and 33.3%, respectively. From 16 patients with mifamurtide treatment, 15 patients were alive (3 metastatic at diagnosis), and only one patient died from the poor response group.

Discussion

This study retrospectively reviewed 79 patients diagnosed and treated with osteosarcoma at Ege University Hospital over 25 years. One of the important outcomes of our study was that females had a significantly better outcomes than the males. Most of the studies in the literature reported no significant effect of gender on survival. However, following our results, the EURAMOS-1 study protocol reported a more favorable outcome for females.[12] Two other studies, one of them being a systematic review of 40 studies, reported that females experienced significantly higher overall survival rates than males.[13,14] On the other hand, these two series reported more favorable prognoses for younger patients. Our study did not detect any significant relationship between age groups and survival (<10 yrs vs. ≥10 yrs). The poorer prognosis of male patients might be related to several factors. Firstly, osteosarcoma is more common in males. Furthermore, since females generally reach puberty earlier than males, the peak incidence of osteosarcoma is seen in females at younger ages. As reported in previous studies, the prognosis is better in younger patients. All of these bring to mind the effect of hormonal activity and its effect on skeletal growth. However, the interaction of gender and age has never been formally studied in osteosarcoma patients.

One of the differences in our study was the high rate of lung metastases at diagnosis. According to the literature, 15-17% of the patients were considered to have metastases at diagnosis.[15] In our study population, it was high (32%). This might be due to the late presentation of our patients. However, our patient's event-free and overall survival rates were compatible with the literature.

Between the years 1995 and 2013, we used the Mayo Pilot II Study Protocol. The backbone of chemotherapy was ifosfamide, cisplatin, and high-dose methotrexate. In 2013 we started to use the EURAMOS protocol and added mifamurtide to postoperative chemotherapy after 2016. During this period (between these two protocols), the estimated 5-year EFS and OS rates increased, but this was not statistically significant. Our results were compatible with

previous studies. The EURAMOS-1 trial was a risk-stratified randomized controlled trial investigating treatment based on histological response to preoperative chemotherapy. They reported that 5-year EFS was 54% and 5-year OS 71%.[12] In our patient group with the same treatment protocol after 2013, 5-year EFS was 60.9% and 5-year OS 71.8%. We could not compare our results with the Mayo Pilot II Study Protocol as there was no published data (personal communication with Dr. Carola Arndt).

Increasing the doses of preoperative chemotherapy did not improve good histologic response and survival rates in osteosarcoma of the extremity in the study by Bacci et al.[16], including children and adults. They recommend that preoperative treatment's degree of tumor necrosis reflects an innate sensitivity to chemotherapy, which is not altered by increasing drug doses. On the other hand, intensifying chemotherapy with increased dose intensity resulted in a statistically significant increase in favorable histologic response rate, but not in increased progression-free or overall survival.[5]

In our study population, the histological response is one of the strongest predictors of survival. Patients with a poor response to preoperative chemotherapy have a worse survival rate than those with a good response. The chemotherapy regimen in the postoperative period is controversial, especially for poor responders. Several studies suggest that altering postoperative chemotherapy might improve the outcome for patients with a poor histological response. We treated most of the poor responders (n=26) with high-dose ifosfamide in our study group. Our results showed no difference in EFS and OS rates of patients treated with high-dose ifosfamide.

Similarly, the EURAMOS-1 study results showed that event-free survival did not differ with the addition of ifosfamide-etoposide to postoperative chemotherapy in patients with poorly responding osteosarcoma.[17] Another randomized controlled trial from the Italian Sarcoma Study Group evaluated the addition of ifosfamide to postoperative chemotherapy for poor responders. There was no significant difference in survival rates with the addition of ifosfamide.[18] Also, postoperative intensification with high-dose cyclophosphamide and melphalan in patients with localized osteosarcoma with poor histological response did not improve survival.[19] As a result, intensification of postoperative chemotherapy did not impact survival rates.

Other study groups using different 3- or 4-drug schedules from these same active drugs have reported similar results. Therefore ifosfamide was recommended for patients with a poor histologic response to methotrexate, cisplatin, and doxorubicin.[18-20] In the EURAMOS treatment protocol, preoperative chemotherapy consisted of three drugs (MAP), and the time of surgery was earlier. In our study, the patients in the EURAMOS group had better EFS and OS rates than the Mayo Pilot II Study group; preoperative chemotherapy consisted of four drugs (MAP plus ifosfamide).

Similar conclusions might be drawn from the INT- 0133 study.[9] In this Children's Oncology Group study, patients treated with MAP had a better 5-year EFS of 64% than patients treated with the four-drug combination (MAP plus ifosfamide), who had a 6-year EFS of 58%. They reported that the addition of ifosfamide to cisplatin, doxorubicin, and methotrexate did not enhance EFS or OS for patients with osteosarcoma.

In recent years, the addition of mifamurtide to postoperative chemotherapy after surgery was reported with a statistically significant effect on the overall survival of non-metastatic patients. Meyers et al.[9] reported that the addition of MTP to chemotherapy resulted in a statistically significant improvement in overall survival and a better EFS. Múdry et al.[21] analyzed the treatment results of patients with localized osteosarcoma treated with or without mifamurtide. They reported significantly

better EFS and PFS for mifamurtide group. According to these, mifamurtide addition could be a promising treatment option for localized osteosarcoma. However, it is not clear as to whether it should be added to metastatic patients. Chou et al.[10] reported that the 5-year EFS in the metastatic patients who had received mifamurtide with the chemotherapy regimen was 42% compared to 26% for patients who had received chemotherapy alone; however, this was not statistically significant. Since 2016, we have used mifamurtide with chemotherapy in the postoperative period. As our study was a retrospective study, and we started mifamurtide treatment at the same period as the EURAMOS treatment protocol, we have a limited number of patients to evaluate the effect of mifamurtide.

Nevertheless, survival rates were significantly higher in the mifamurtide group. Again relapse rate was significantly lower in the mifamurtide group with a median 45-month follow-up time. In the EURAMOS treatment group, only three patients with a poor response group had mifamurtide treatment. The number of patients was insufficient to evaluate mifamurtide's effect on EFS and OS rates. Our results were similar to another study from our country. Taçyıldız et al.[22] reported that adding mifamurtide in the high-risk group decreased the probability of distant metastasis, increased the median time to distant metastasis, and improved event-free survival.

In conclusion, we had a broad experience with 79 patients with osteosarcoma. Treatment seems to be more successful in females. Limb salvage surgery rate is greater than amputation in our series. International treatment protocols such as Mayo Pilot II Study Protocol and EURAMOS give similar results for non-metastatic and metastatic patients in collaboration with experienced orthopedic surgeons and pediatric oncologists. The addition of ifosfamide to the postoperative period does not influence the outcome. Our limited experience with mifamurtide addition seems promising, but it should be tested in large randomized studies.

Ethical approval

Ege University Ethics Committee approved the study (report number: 22-4T/55). Informed consent form was obtained from the parents.

Author contribution

The authors confirm contribution to the paper as follows: study conception and design: EA, MK; data collection: ŞÖG, HK, İT, BK, MA, BD, DS, ZB; analysis and interpretation of results: EA, ŞÖG, MK; draft manuscript preparation: EA. All authors reviewed the results and approved the final version of the manuscript.

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Conflict of interest

The authors declare that there is no conflict of interest.

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