Table 4 Univariate survival analysis in 29 soft tissue sarcoma patients with lung metastasis

Characteristics	*n*	3-year survival (%)	*P* value
Age			
≧65	11	23.9	0.54
<65	18	38.9	
Gender			
Male	17	32.1	0.87
Female	12	36.7	
Local recurrence			
Yes	15	53.6	0.12
No	14	20.0	
No. of lung met.			
>3	11	30.3	0.76
≦3	18	36.1	
Maximum tumor size of lung met.			
>10	7	17.1	0.70
≦10	22	38.6	
Lung met. at presentation			
Yes	8	12.5	0.06
No	21	43.4	
Distribution			
Lateral	14	21.4	0.08
Bilateral	15	47.0	
VDT			
>26 days	15	50.6	0.01
≦26 days	14	15.9	

VDT Volume doubling time

Table 5 Multivariate survival analysis in 29 soft tissue sarcoma patients with lung metastasis

Variables		HR (95% CI)	*P* value
Lung met. at presentation	No	0.36 (0.139–0.93)	0.03
Distribution	Unilateral	1.83 (0.76–4.47)	0.18
VDT	>26 days	0.27 (0.11–0.69)	0.006

95% CI: 95% Confidence interval

HR Hazard Risk, *VDT* volume doubling time

method. The analysis of the growth rate of a tumor expressed as the "tumor doubling time" appears to be an accurate and precise method to compare the biological aggressiveness of tumors in different patients [8–14]. However, there are few reports on the growth rate of lung metastases from soft tissue and skeletal sarcoma [11, 12, 15, 16]. Although these previous analyses used chest radiography, the current study measured the volume doubling time using chest CT images in patients with lung metastases, because lung metastases are detected more accurately and earlier with CT scan than with chest radiography.

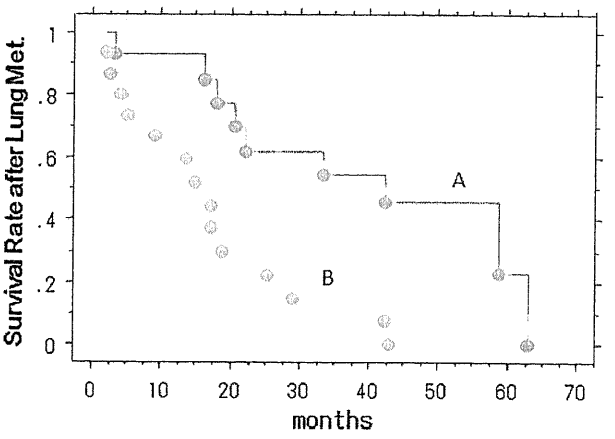

Fig. 3 Kaplan–Meier survival curve shows survival after lung metastasis of the 29 patients with soft tissue sarcomas. (**a** patients with a VDT of more than 26 days **b** patients with a VDT of within 26 days)

The current measurement of lung metastases showed that there was variability in the growth rate both between metastases in different patients and between different histological tumor types. The VDT of patients with high grade tumors was significantly shorter than that of low grade tumors. These data are consistent with several reports of OS, MFH, and LMS (Table 7). Interestingly, the VDTs of the patients with myxoid liposarcoma were relatively short (20 and 26 days). In contrast, the VDTs of synovial sarcoma patients were relatively longer than other high grade sarcomas (27, 65, and 185 days).

Approximately 10–38% of patients with sarcomas have isolated lung metastatic disease [7, 17, 18]. It is very important to determine whether the lung nodule is malignant or benign, because it usually affects the subsequent treatment plan, including surgical excision, radiation therapy and chemotherapy. However, chest radiography usually do not allow detection of small lung nodules (<10 mm) [19]. Improvements in CT scanning have enabled clinicians to detect small lung nodules less than 5 mm in size [20]. A lung nodule of 5 mm in patients with a short VDT (within 30 days) tumor, like OS, LMS, MPNST, would grow to about 10 mm within 3 months based on the Schwaltz equation [9]. A tumor with a VDT of 10, 20 and 30 days would grow to 10 mm in 30, 60, and 90 days, respectively. Therefore, the next chest CT should be taken within 2 months in patients with small lung nodules. Treatment such as RFA or surgical excision of the nodules is recommended if the nodule becomes larger. The nodules normally continue to be benign if they remain 5 mm or smaller than 5 mm in size for more than 6 months [20].

Several large series have defined a number of clinical prognostic factors that are associated with post-metastatic

Table 6 Univariate survival analysis in 11 bone sarcoma patients with lung metastasis

Characteristics	*N*	3-year survival (%)	*P* value
Age			
≧65	3	N.A	0.90
<65	8	25.0	
Gender			
Male	6	33.3	0.97
Female	5	20.0	
Local recurrence			
Yes	4	25.0	0.83
No	7	28.6	
No. of lung met.			
>3	6	16.7	0.27
≦3	5	40.0	
Maximum tumor size of lung met.			
>10	5	60.0	0.16
≦10	6	0	
Lung met. at presentation			
Yes	4	25.0	0.82
No	7	28.6	
Distribution			
Lateral	2	N.A	0.36
Bilateral	9	22.2	
VDT			
>13 days	4	50.0	0.24
≦13 days	7	14.3	

N.A Not applicable, *VDT* volume doubling time

Table 7 Review of the literature describing volume doubling time in sarcoma patients

Authors	Device	Histology	*n*	VDT (day)
Joseph et al. [11]	X-ray	Osteogenic sarcoma	25	11–360
		Fibrosarcoma	12	13–340
		Ewing sarcoma	8	7–18
		Liposarcoma	6	11–100
		Rhabdomyosarcoma	5	10–20
		Synovial sarcoma	4	16–80
Rooser et al. [16]	X-ray	Leiomyosarcoma	3	32–198
		MFH	2	17–42
		Liposarcoma	2	9–38
Band and Kocandrle [15]	X-ray	Osteogenic sarcoma	8	11–79
		Fibrosarcoma	2	11–23
		Ewing sarcoma	2	25–37
Blomqvist et al. [12]	X-ray	Leiomyosarcoma	3	11–276
		Ewing sarcoma	2	16–28
		MFH	2	7–9
		Liposarcoma	2	35–75
Nakamura et al. [Present study]	CT	Osteosarcoma	6	7–13
		MPNST	5	10–37
		Leiomyosarcoma	5	11–38
		MFH	5	12–65
		ESCS	4	64–410
		Synovial sarcoma	3	27–187
		Chondrosarcoma	2	33–255
		Liposarcoma	2	20–26

VDT Volume doubling time, *MFH* malignant fibrous histiocytoma, *ESCS* extra-skeletal chondrosarcoma

survival [1–7]. Present study investigated whether there was any correlation between the VDT and survival after lung metastasis. Joseph et al. [11] reported that patients with a VDT of greater than 40 days had better prognosis than those within 40 days in 113 patients with lung metastatic disease, including 64 sarcoma patients. Roth et al. [10] reported that patients with a VDT less than 20 days had significantly poorer prognosis after metastasectomy than patients with a VDT of 20 days or more in soft tissue sarcoma with lung metastasis, although the histological types were not recorded in this study. The current univariate analysis revealed significantly poorer predictive values for VDT in all 40 sarcoma patients and 29 soft tissue sarcoma patients. A multivariate analysis showed the VDT to be an independent predictor of survival in 29 soft tissue sarcoma patients. However, VDT was not a significant predictive value for bone sarcoma patients because the majority of patients had rapid VDTs and there were only a small number of bone sarcoma patients.

These results suggest that patients with a shorter VDT and consequently a more rapid rate of tumor growth have a significantly lower chance for long term survival especially in soft tissue sarcoma. Furthermore, these results indicate that the VDT appears to enable one to select a group of patients for metastasectomy and/or RFA who will have a more favorable prognosis. The patients with lung metastasis that have a longer VDT should therefore be considered for metastasectomy and/or lung RFA even if the lesions are multiple and/or bilateral.

There are some limitations in the current study. Although our study represents, one of the largest series of CT screening that measured lung metastasis in sarcoma patients, it includes variety histological types of sarcomas which consist of the relatively small number of patients. Another limitation is patient's selection bias. Thirty-two of 72 patients were excluded from this study because of various reasons such as treatment, progression of lung metastases. There may be a bias when evaluating the prognosis of the patients with lung metastases.

Furthermore, it would be useful to further investigate whether the treatment such as RFA, metastasectomy or

chemotherapy influenced VDT of the remaining or new metastatic lesions in larger number of patients.

Conclusion

There was variability in the growth rate both between metastases in different patients and between different histological tumor types in our study.

The next chest CT images should be taken within 2 months in sarcoma patients with lung small nodules. Treatment such as RFA or surgical excision of the nodules is recommended if the nodule becomes larger. However, the patients with a shorter VDT and consequently a more rapid rate of tumor growth have a significantly lower chance for long term survival, especially in soft tissue sarcoma.

Conflict of interest None.

References

1. Casson AG, Putnam JB, Natarajan G et al (1992) Five-year survival after pulmonary metastasectomy for adult soft tissue sarcoma. Cancer 69(3):662–668
2. Billingsley KG, Burt ME, Jara E et al (1999) Pulmonary metastases from soft tissue sarcoma: analysis of patterns of disease and postmetastasis survival. Ann Surg 229(5):602–610
3. Verazin GT, Warneke JA, Driscoll DL et al (1992) Resection of lung metastases from soft-tissue sarcomas. A multivariate analysis. Arch Surg 127(12):1407–1411
4. van Geel AN, Hoekstra HJ, van Coevorden F et al (1994) Repeated resection of recurrent pulmonary metastatic soft tissue sarcoma. Eur J Surg Oncol 20(4):436–440
5. Billingsley KG, Lewis JJ, Leung DH et al (1999) Multifactorial analysis of the survival of patients with distant metastasis arising from primary extremity sarcoma. Cancer 85:389–395
6. Putnam JB Jr, Roth JA, Wesley MN et al (1984) Analysis of prognostic factors in patients undergoing resection of pulmonary metastases from soft tissue sarcomas. J Thorac Cardiovasc Surg 87(2):260–268
7. Harting MT, Blakely ML, Jaffe N et al (2006) Long-term survival after aggressive resection of pulmonary metastases among children and adolescents with osteosarcoma. J Pediatr Surg 41(1): 194–199
8. Collins VP, Loeffler RK, Tivey H (1956) Observations on growth rates of human tumors. Am J Roentgenol 76:988–1000
9. Schwartz M (1961) A biomathematical approach to clinical tumor growth. Cancer 14:1272–1294
10. Roth JA, Putnam JB, Wesley MN (1985) Differing determinations of prognosis following resection of pulmonary metastases from osteogenic and soft tissue sarcoma patients. Cancer 55: 1361–1366
11. Joseph WL, Morton DL, Adkins PC (1971) Prognostic significance of tumor doubling time in evaluating operability in pulmonary metastatic disease. J Thorac Cardiovasc Surg 61:23–32
12. Blomqvist C, Wiklund T, Tarkkanen M et al (1993) Measurement of growth rate of lung metastases in 21 patients with bone or soft-tissue sarcoma. Br J Cancer 68:414–417
13. Nietert PJ, Ravenel JG, Leue WM et al (2009) Inprecision in automated volume measurement of pulmonary nodules and its effect on the level of uncertainly in volume doubling time estimation. Chest 135:1580–1587
14. Lindell RM, Hartman TE, Swensen SJ et al (2007) Five-year lung cancer screening experience: CT appearance, growth rate, location, and histologic features of 61 lung cancers. Radiology 242: 555–562
15. Band PR, Kocandrle C (1975) Growth rate of pulmonary metastases in human sarcomas. Cancer 36:471–474
16. Rooser B, Pettersson H, Alvegard T (1987) Growth rate of pulmonary metastases from soft tissue sarcoma. Acta Oncol 26: 189–192
17. Kane JM, Finley JW, Driscoll D et al (2002) The treatment and outcome of patients with soft tissue sarcomas and synchronous metastases. Sarcoma 6(2):69–73
18. Pollock RE, Karnell LH, Menck HR et al (1996) The national cancer data base report on soft tissue sarcoma. Cancer 78(10): 2247–2257
19. Wang JC, Sone S, Feng L et al (2000) Rapidly growing small peripheral lung cancers detected by screening CT: correlation between radiological appearance and pathological features. Br J Radiol 73:930–937
20. Nakamura T, Matsumine A, Niimi R et al (2009) Management of small pulmonary nodules in patients with sarcoma. Clin Exp Metastasis 26(7):713–718

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Acridine Orange Inhibits Pulmonary Metastasis of Mouse Osteosarcoma

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Abstract. *Although the survival of patients with osteosarcoma has improved following development of chemotherapy and surgery, the presence of pulmonary metastases indicate a poor prognosis. We developed photodynamic and radiodynamic therapies with acridine orange (AO-PDT and AO-RDT) for minimally invasive surgery to treat musculoskeletal sarcomas and reported a good clinical outcome of local control and limb function. We investigated the effect of AO-PDT using flash-wave light (FWL) on pulmonary metastasis of mouse osteosarcoma. In in vitro and in vivo studies, AO alone and AO-PDT significantly inhibited cell invasion and the growth of pulmonary metastases from primary mouse osteosarcoma. AO may have a specific metastasis-inhibitory effect, different from the effect of AO-PDT. The fluorovisualization effect on pulmonary metastases following intravenous AO administration showed that pulmonary metastases localized on the lung surface were recognized as brilliant green lesions. In conclusion, AO-PDT using FWL inhibited cell invasion and pulmonary metastases in mouse osteosarcoma; therefore, this treatment modality might be applicable for treating pulmonary metastasis from malignant musculoskeletal tumors in humans.*

The survival of patients with osteosarcoma has improved due to the development of chemotherapy and surgical techniques (1); however, approximately 15-20% of osteosarcoma patients undergo detectable metastatic disease (2-4). Primary metastases of osteosarcoma affect the lung in 87% of the cases, distant bones in 21% of the cases, and other soft tissues in 9% of the cases, thus result in poor prognosis (5).

We developed photodynamic and radiodynamic therapies with acridine orange (AO-PDT and AO-RDT) as a minimally invasive surgery for treating musculoskeletal sarcomas, making it possible to preserve excellent limb function with a low risk of local tumor recurrence. On the basis of the satisfactory outcome of clinical trials involving more than 100 patients with high-grade malignant bone and soft tissue sarcomas, our previous reports revealed that this modality was clinically applicable (6-11). Nevertheless, in those clinical trials, AO was locally administered by flooding of the surgical field after tumor resection, and prognosis of these patients was better than that of patients treated with conventional wide tumor resection. Therefore, AO-PDT may inhibit metastasis. Furthermore, our previous studies demonstrated that intravenous AO administration had excellent fluorovisualization effect for photodynamic diagnosis (PDD) of mouse osteosarcoma (12) and that AO-PDT with intravenous AO injection significantly inhibited tumor growth of mouse osteosarcoma (13). Moreover, we have previously reported that a high-power flash-wave light (FWL) from a xenon lamp in AO-PDT exerts a stronger cytocidal effect than a continuous-wave light (CWL) on a mouse osteosarcoma cell line (14, 15).

In this study, we investigated whether PDT using FWL with intravenous AO administration exhibits anti-metastatic activity in the development of pulmonary metastasis in mouse osteosarcoma.

Materials and Methods

Tumor cell line and cell culture. The mouse osteosarcoma cell line derived from Dunn's osteosarcoma, LM8, which is highly metastatic, was used in the present study (16). LM8 cells were harvested in Dulbecco's modified Eagle's medium (DMEM, Invitrogen, Carlsbad, CA, USA) containing 10% fetal bovine serum at 37°C in 5% CO_2. All experiments described below were started after 24 h of cell culture.

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Key Words: Acridine orange, osteosarcoma, pulmonary metastasis, photodynamic therapy, flash-wave light, fluorovisualization.

Light sources. A xenon lamp was used as the source of FWL (17). The illumination machine, High Power Strobo Flash XF-1000 (Nissin Electric Co., Kyoto, Japan), was used for FWL irradiation. The light irradiation frequency of the FWL was 30 Hz and the pulse width was less than 1 ms. The energy generated by one shot-irradiation with FWL was 0.1 J cm^2, and the illuminance level was 10^6 lux.

In vitro study. Cell invasion assay: LM8 cells were divided into six groups (n=6): group 1, exposure to AO-free DMEM and no FWL irradiation (control group (C)); group 2, exposure to AO-free DMEM and irradiation with FWL for 10 min (irradiation group (IR)); groups 3-6, exposure to different concentrations (0.1 and 1.0 μg/ml) of AO (Sigma-Aldrich Munich, Germany) for 10 min and no FWL irradiation (group 3, 0.1-AO; group 4, 1.0-AO; group 5, 0.1-AO+IR; group 6, 1.0-AO+IR). At the beginning of the treatment, the medium in each group was exchanged with DMEM containing different concentrations of AO (0.1 and 1.0 μg/ml) or with AO-free DMEM. After exposure to AO for 10 min, the medium was washed out to remove the AO, and AO-free DMEM was added. The cells were isolated from the culture dishes by trypsinization and loaded into the upper chamber in growth medium-containing 6-well BD Matrigel™ Invasion Chamber, pore size, 8 μm (BD Bioscience, CA, USA) at a cell density of 5×10^4 cells per well and were excited with FWL for 10 min. After this treatment, the cells were harvested for 24 h at 37°C in 5% CO_2, following which, duplicate membranes of the chambers were processed and stained with HE. Invading cells were evaluated by counting the number of cells in all fields under high-power light microscopy.

In vivo study. Mouse osteosarcoma model: A suspension containing 1×10^6 cells isolated from culture dishes using trypsinization was inoculated into the soft tissues, including the subcutaneous tissue and muscles of the back, after removal of the hair at the implantation site in C3H mice (5-week-old males) (Japan SLC. Inc., Shizuoka, Japan). Subsequent experiments were conducted on tumors that grew to a macroscopically detectable size (3-6 mm in diameter) within 10 days.

Inhibition of pulmonary metastasis by AO-PDT using FWL: Tumor-bearing mice were divided randomly into four groups of five mice each: group 1, no treatment (C); group 2, irradiation with FWL alone for 10 min (IR); group 3, intravenous administration of AO at 1.0 mg/kg alone (AO); and group 4, intravenous administration of AO at 1.0 mg/kg followed by irradiation with FWL for 10 min (AO+IR). Tumor-bearing mice administered AO *via* the tail vein were exposed to FWL illumination for 10 min at 2 h after AO injection, as previously described (13). Briefly, AO mice administered were placed in a stainless steel bowl under anesthesia induced by intraperitoneally administered pentobarbital sodium and were exposed to FWL irradiation using an illumination machine (XF-1000) for 10 min. AO was used at a concentration of 1.0 mg/kg because our previous studies showed that this concentration yields the strongest cytocidal effect and the lowest toxicity in mice (12, 13). The irradiation time (10 min) was also determined on the basis of the results of previous studies (12, 13, 15, 18, 19). On day 28 after the above treatment, AO was administered intravenously at a concentration of 1.0 mg/kg to tumor-bearing mice *via* the tail vein. After 2 h, mice in each group were sacrificed under anesthesia and the lungs were removed. The lungs were illuminated using a 5,000-luminance blue light selected through an interference filter (450-490 nm) from a 500-W high-power xenon lamp source (SAN-EI Electric MFG Co., Ltd., Tokyo, Japan) and guided through a single fiber tube. The fluorescence emitted from the AO accumulated in the pulmonary metastases was detected using a digital camera system C-5050 equipped with an absorption filter (>520 nm) set at a distance of 10 cm from the tumor surface (photodynamic diagnosis with acridine orange (AO-PDD)). This procedure was conducted in a dark room (12). Image data from the photogram were entered into a personal computer using Adobe Photoshop 7.0 software (Adobe Systems Co., MD, USA). The number of metastatic lesions on the surface of the lungs was counted on the AO-PDD images, regardless of the tumor size (18). The removed lungs were fixed in 10% formalin and embedded in paraffin. The paraffin-embedded lung tissues were cut at their maximum dimensions and were stained with HE. The number of pulmonary metastases was then counted based on histological findings, regardless of the tumor size, under high-powered light microscopy.

Statistical analysis. Statistical analysis was performed using the StatView statistical software version 5.0 (SAS Institute Inc Cary, NC, USA). Significant differences among the groups were evaluated using Student's *t*-test. *p*-Values less than 0.05 were considered statistically significant.

All experiments were performed in accordance with the guidelines in the Declaration of Helsinki and the Interdisciplinary Principles and Guidelines for the Use of Animals in Research, Testing, and Education.

Results

In vitro study. Cell invasion assay: Figure 1 shows the results of the study involving the BD Matrigel™ Invasion Chamber and demonstrates that the average number of LM8 cells that had passed through the chamber membrane after 24 h was 114±14 in group 1 (C) and 117±12 in group 2 (IR). There were no significant differences between these groups. In the AO group alone, the average numbers of invading cells were 61±19 in group 3 (0.1-AO) and 61±6 in group 4 (1.0-AO). The ability of cells to pass through the membrane was significantly inhibited in the AO group than in the control group ($p<0.01$). In the AO-PDT group, the number of invading cells was 34±19 in group 5 (0.1-AO+IR) and 5±1 in group 6 (1.0-AO+IR). In both groups, there was a remarkable decrease of the invading number of cells compared to groups 1 (C) and 2 (IR) ($p<0.01$). In the AO-PDT group, the inhibitory effect of AO on cell invasion was concentration dependent.

In vivo study. Count of pulmonary metastatic lesions by histology and fluorescence imaging after AO administration: Figure 2 shows the brilliant green fluorescence emitted from the *ex vivo* pulmonary metastatic lesions localized on the lung surface in group 1 (C) under blue light excitation of AO selectively bound to tumor cells. Visualization of the pulmonary metastases on fluorescence images (right side of the figure, fluorovisualization) is much easier than macroscopic detection under normal light (left side of the figure). Figure 3 shows the histological findings of the pulmonary metastases

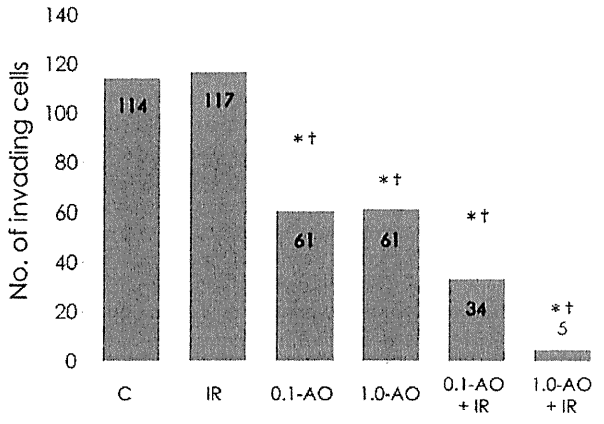

Figure 1. *Number of invading LM8 cells after acridine orange (AO) exposure or followed by illumination with flash-wave light (FWL) in the BD Matrigel™ Invasion Chamber assay. C, control group; IR, FWL irradiation group; 0.1-AO, exposure to 0.1 μg/ml of AO group; 1.0-AO, exposure to 1.0 μg/ml of AO group; 0.1-AO+IR, exposure to 0.1 μg/mL of AO and FWL irradiation group; 1.0-AO+IR, exposure to 1.0 μg/mL of AO and FWL irradiation group. *, $p<0.01$ vs. C; †, $p<0.01$ vs. IR.*

originating from the osteosarcoma in group 1 (C). Not only were surface metastatic lesions seen on fluorescence images but deeply located lesions were detectable as well. The graph in Figure 4 presents both the numbers of the pulmonary metastatic lesions counted on histological sections and the surface metastatic lesions visualized using AO fluorescence images in each group. There was no significant differences in the number of lesions detected with each method in groups 1 (C), 2 (IR), and 4 (AO+IR); however, in group 3 (AO), twice the number of lesions was observed on fluorescence images as compared to the ones detected by histological examination. Numerous pulmonary metastases were detected in groups 1 (C) and 2 (IR), whereas in group 4 (AO+IR), a markedly reduced number of metastases was detected (C *vs.* AO+IR: histological examination, $p<0.04$ and fluorescence imaging, $p<0.04$; IR *vs.* AO+IR: histological examination, $p<0.03$ and fluorescence imaging, $p<0.01$). Group 3 (AO) also exhibited a significant decrease in the number of metastatic lesions as compared to groups 1 (C) and 2 (IR) (C *vs.* AO: histological examination, $p<0.05$ and fluorescence imaging, $p<0.05$; IR *vs.* AO: histological examination, $p<0.05$ and fluorescence imaging, $p<0.05$).

Discussion

Patients with osteosarcoma commonly undergo hematogenous metastasis to the lungs (3, 20). The prognosis of patients with pulmonary metastasis remains poor despite multimodal treatments, including chemotherapy and surgery (3, 5). Recently, the efficacy of percutaneous radiofrequency ablation in treating pulmonary metastases arising from sarcoma (21) was reported; however, there are presently few effective therapies available for treating multiple pulmonary metastases from osteosarcoma.

We developed and established a limb salvage surgery combined with minimal invasive tumor excision and AO-PDT followed by AO-RDT to preserve excellent limb function with a low risk of local tumor recurrence (6-11). In clinical studies, AO was locally administered by flooding of the surgical field after tumor resection, but the prognosis of patients was better than that of patients treated with conventional wide tumor resection surgery. Therefore, we speculated that AO-PDT may inhibit metastasis.

The results of this study revealed that AO-PDT using FWL and AO alone, had a remarkable *in vitro* anti-invasive effect and an *in vivo* anti-metastatic effect on mouse osteosarcoma cells. Since previous studies reported that AO-PDT has a strong cytocidal effect on malignant musculoskeletal sarcomas (14, 15, 19), cell apoptosis by AO-PDT may reduce cellular invasive ability. Our *in vivo* study demonstrated that AO-PDT using FWL as the excitation light after intravenous AO injection, was also highly effective for inhibiting the growth of pulmonary metastases from primary mouse osteosarcomas, without resection of the local lesion, thus suggesting that AO-PDT prevents metastasis to the lung. This effect may be primarily due to growth inhibition of the primary tumor; however, cellular pulmonary metastasis is considered to occur within 10 days after tumor cell inoculation in our osteosarcoma model (22) because mice in which the primary tumor had been widely resected later died of pulmonary metastasis without local recurrence. Therefore, a possible mechanism underlying on the pulmonary metastasis-inhibitory effect of AO-PDT may be the destruction of cellular metastatic lesions by AO or AO-PDT. Since mice treated using AO-PDT received illumination over the entire body, the high-power FWL penetrated the chest wall to exert a cytocidal effect against metastatic lesions incorporating intravenously administered AO.

Interestingly and unexpectedly, cell invasion was significantly suppressed in the group exposed to AO alone *in vitro*. Inhibition of pulmonary metastases was also observed in mice treated with AO alone *in vivo*. It has been established that AO alone, does not have a strong cytocidal effect against mouse osteosarcomas either *in vitro* or *in vivo* (13, 15); therefore, the mechanism of this effect remains unclear. AO may have a specific metastasis-inhibitory effect, which in different from the effect of AO-PDT. High-grade malignant tumors exist in an acidic environment, which is considered to accelerate metastasis (23). AO selectively binds to acidic structures in the tumor cells, such as lysosomes or acidic vesicles containing a large amount of protons (low pH), in order to neutralize the acidic conditions (24). The lysosome contains many enzymes, some of which

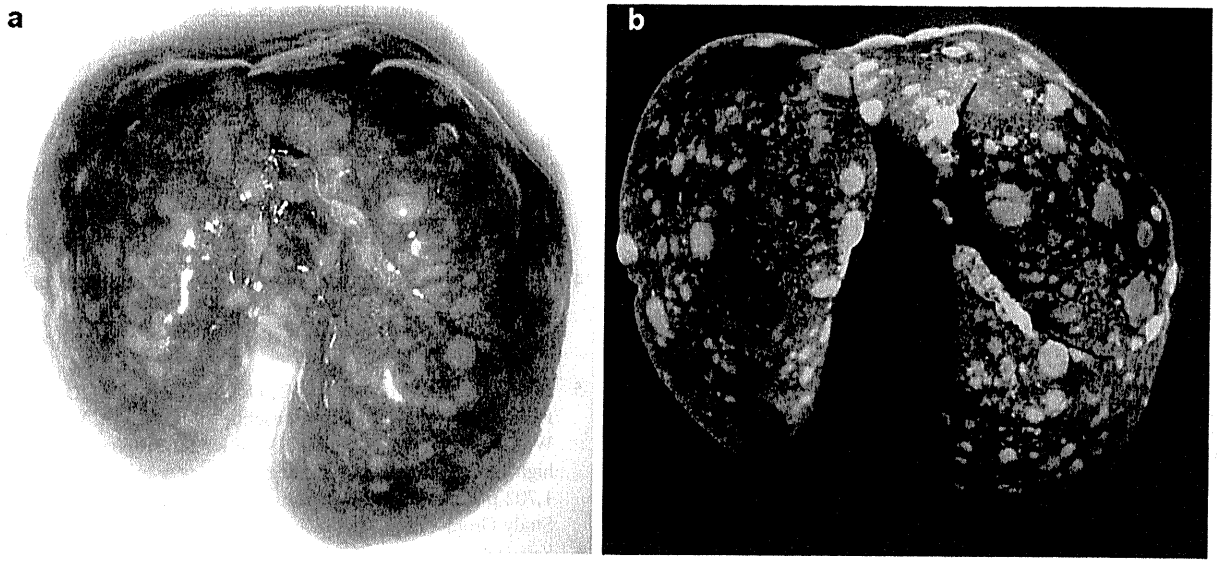

Figure 2. *Stereoscopic findings of pulmonary metastatic lesions localized on the lung surface under ordinary light (a) and under blue light using a yellow absorption filter (b).*

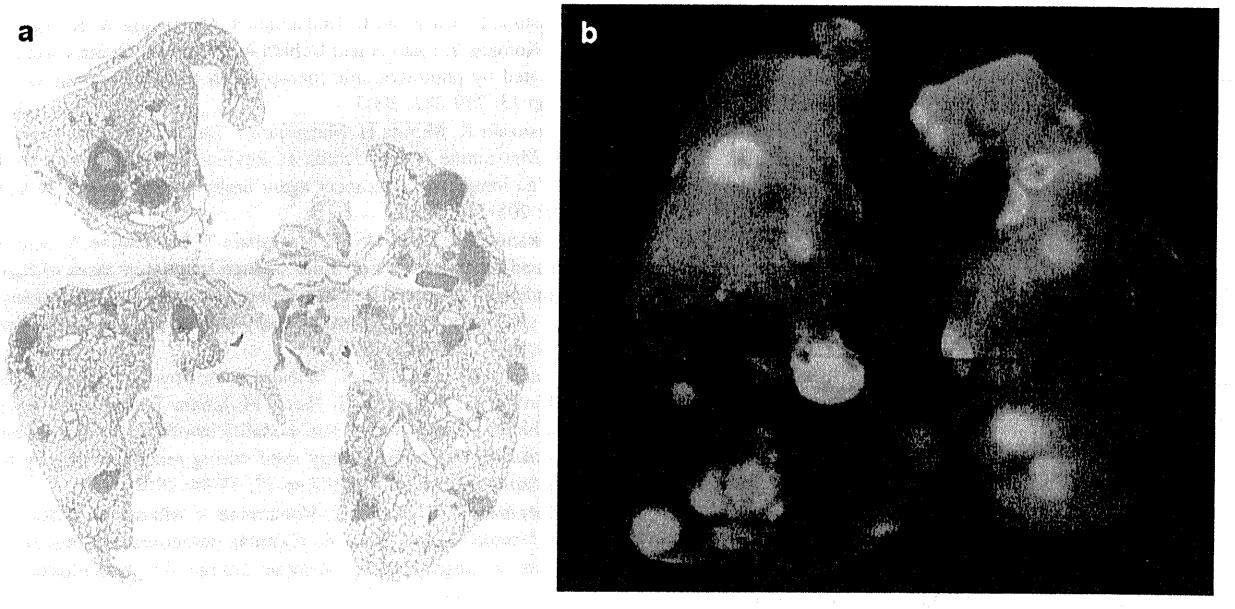

Figure 3. *Histological findings (a) and fluorescence images in photodynamic diagnosis with acridine orange (AO-PDD) (b) of pulmonary metastatic tumors from mouse osteosarcoma.*

are important for tumor invasion and metastasis, such as cathepsin groups, metalloproteases, and heparinases (25-27). AO may induce dysfunction of lysosomal enzymes related to pH. This may be one of the mechanisms underlying the prevention of pulmonary metastasis by AO, although further investigation is needed. Another potential mechanism is that after PDT, antitumor immunity may be enhanced, and tumor antibodies may kill pulmonary metastatic tumor cells (28). Recently, it was reported that exosomes induced by PDT also enhance antitumor immunity after stimulating macrophage antigen expression (29). These tumor immunity-related mechanisms which suppress pulmonary metastasis of osteosarcoma are fascinating in the clinical application of AO-PDT.

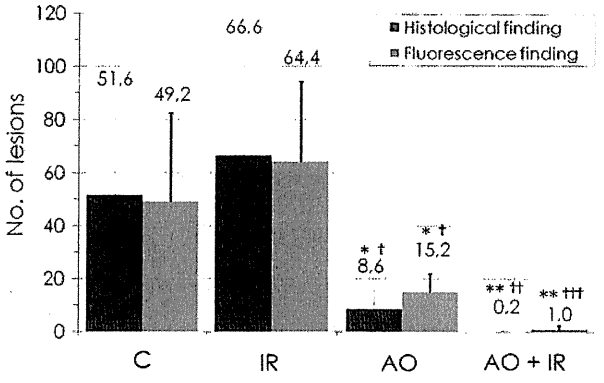

Figure 4. *Number of pulmonary metastases detected using histological and fluorescence imaging methods in each group studied with acridine (AO) alone and photodynamic therapy with acridine orange (AO-PDT). C, control group; IR, flash-wave light (FWL) irradiation group; AO, intravenous administration of 1.0 μg/ml of AO group; AO+IR, intravenous administration of 1.0 μg/ml of AO and FWL irradiation group. *, p<0.05 vs. C; †, p<0.05 vs. IR; **, p<0.04 vs. C; ††, p<0.03 vs. IR; †††, p<0.01 vs. IR.*

An *ex vivo* fluorescence imaging study of the fluorovisualization effect on pulmonary metastases following intravenous AO administration showed that pulmonary metastases localized on the lung surface were recognized as brilliant green lesions. Clinically, this effect may be very useful for determining the surgical margin during endoscopic resections, such as thoracoscopy, bronchoscopy, cystoscopy, and laparoscopy (30-32). Although many experimental methods have been suggested for assessing pulmonary metastases (33, 34), fluorescent detection following intravenous AO administration, referred to as AO-PDD, is useful and easier than other methods. If the fluorescence intensity of AO over the entire lung was measured using a photomultiplier, more accurate estimation of the metastatic tumor volume, as compared with that using the count method, could be possible.

In conclusion, AO-PDT using FWL inhibited cell invasion and pulmonary metastasis in mouse osteosarcoma; therefore, we believe that this treatment modality may be applicable for treating pulmonary metastasis of malignant musculoskeletal tumors in humans, although additional studies are needed to verify this finding.

References

1 Link MP, Goorin AM, Miser AW, Green AA, Pratt CB, Belasco JB, Pritchard J, Malpas JS, Baker AR, Kirkpatrick JA, Ayala AG, Shuster JJ, Abelson HT, Simone JV and Vietti TJ: The effect of adjuvant chemotherapy on relapse-free survival in patients with osteosarcoma of the extremity. N Engl J Med *314*: 1600-1606, 1986.

2 Goorin AM, Delorey MJ, Lack EE, Gelber RD, Price K, Cassady JR, Levey R, Tapper D, Jaffe N, Link M and Abelson HT: Prognostic significance of complete surgical resection of pulmonary metastases in patients with osteogenic sarcoma: analysis of 32 patients. J Clin Oncol 2: 425-431, 1984.

3 Briccoli A, Rocca M, Salone M, Guzzardella GA, Balladelli A and Bacci G: High grade osteosarcoma of the extremities metastatic to the lung: Long-term results in 323 patients treated combining surgery and chemotherapy, 1985-2005. Surg Oncol *19*: 193-199, 2010.

4 Rasalkar DD, Chu WC, Cheng FW, Lee V, Lee KH and Li CK: Pulmonary metastases in children with osteosarcoma: characteristics and impact on patient survival. Pediatr Radiol *41*: 227-236, 2010.

5 Bielack SS, Kempf-Bielack B, Delling G, Exner GU, Flege S, Helmke K, Kotz R, Salzer-Kuntschik M, Werner M, Winkelmann W, Zoubek A, Jürgens H and Winkler K: Prognostic factors in high-grade osteosarcoma of the extremities or trunk: an analysis of 1,702 patients treated on neoadjuvant Cooperative Osteosarcoma Study Group protocols. J Clin Oncol *20*: 776-790, 2002.

6 Kusuzaki K, Murata H, Matsubara T, Miyazaki S, Shintani K, Seto M, Matsumine A, Hosoi H, Sugimoto T and Uchida A: Clinical outcome of a novel photodynamic therapy technique using acridine orange for synovial sarcomas. Photochem Photobiol *81*: 705-709, 2005.

7 Yoshida K, Kusuzaki K, Matsubara T, Matsumine A, Kumamoto T, Komada Y, Naka N and Uchida A: Periosteal Ewing's sarcoma treated by photodynamic therapy with acridine orange. Oncol Rep *13*: 279-282, 2005.

8 Kusuzaki K, Murata H, Matsubara T, Satonaka H, Wakabayashi T, Matsumine A and Uchida A: Review. Acridine orange could be an innovative anticancer agent under photon energy. In Vivo *21*: 205-214, 2007.

9 Nakamura T, Kusuzaki K, Matsubara T, Matsumine A, Murata H and Uchida A: A new limb salvage surgery in cases of high-grade soft tissue sarcoma using photodynamic surgery, followed by photo- and radiodynamic therapy with acridine orange. J Surg Oncol *97*: 523-528, 2008.

10 Matsubara T, Kusuzaki K, Matsumine A, Murata H, Satonaka H, Shintani K, Nakamura T, Hosoi H, Iehara T, Sugimoto T and Uchida A: A new therapeutic modality involving acridine orange excitation by photon energy used during reduction surgery for rhabdmyosarcomas. Oncol Rep *21*: 89-94, 2009.

11 Matsubara T, Kusuzaki K, Matsumine A, Murata H, Nakamura T, Uchida A and Sudo A: Clinical outocomes of minimally invasive surgery using acridine orange for musculoskeletal sarcomas around the forearm, compared with conventional limb salvage surgery after wide resection. J Surg Oncol *102*: 271-275, 2010.

12 Satonaka H, Kusuzaki K, Matsubara T, Shintani K, Wakabayashi T, Matsumine A and Uchida A: Extracorporeal photodynamic image detection of mouse osteosarcoma in soft tissues utilizing fluorovisualization effect of acridine orange. Oncology *70*: 465-473, 2006.

13 Satonaka H, Kusuzaki K, Matsubara T, Shintani K, Nakamura T, Matsumine A, Iino T and Uchida A: *In vivo* antitumor activity of photodynamic therapy with intravenous administration of acridine orange, followed by illumination with high-power flash-wave light in a mouse osteosarcoma model. Oncol Lett *1*: 69-72, 2010.

14 Ueda H, Murata H, Takeshita H, Minami G, Hashiguchi S and Kubo T: Unfiltered xenon light is useful for photodynamic therapy with acridine orange. Anticancer Res *25*: 3979-3984, 2005.
15 Satonaka H, Kusuzaki K, Matsubara T, Shintani K, Wakabayashi T, Nakamura T, Matsumine A and Uchida A: Flash-wave light strongly enhanced the cytocidal effect of photodynamic therapy with acridine orange on a mouse osteosarcoma cell line. Anticancer Res *27*: 3339-3344, 2007.
16 Asai T, Ueda T, Itoh K, Yoshioka K, Aoki Y, Mori S and Yoshikawa H: Establishment and characterization of a murine osteosarcoma cell line (LM8) with high metastatic potential to the lung. Int J Cancer *76*: 418-422, 1998.
17 Kimura M, Kashikura K, Yokoi S, Koiwa Y, Tokuoka Y and Kawashima N: Photodynamic therapy for cancer cells using a flash-wave light xenon lamp. Opt Rev *12*: 207-210, 2005.
18 Kusuzaki K, Suginoshita T, Minami G, Aomori K, Takeshita H, Murata H, Hashiguchi S, Ashihara T and Hirasawa Y: Fluorovisualization effect of acridine orange on mouse osteosarcoma. Anticancer Res *20*: 3019-3024, 2000.
19 Kusuzaki K, Minami G, Takeshita H, Murata H, Hashiguchi S, Nozaki T, Ashihara T and Hirasawa Y: Photodynamic inactivation with acridine orange on a multidrug-resistant mouse osteosarcoma cell line. Jpn J Cancer Res *91*: 439-445, 2000.
20 Ferrari S, Briccoli A, Mercuri M, Bertoni F, Picci P, Tienghi A, Del Prever AB, Fagioli F, Comandone A and Bacci G: Post-relapse survival in osteosarcoma of the extremities: prognostic factors for long-term survival. J Clin Oncol *21*: 710-715, 2003.
21 Palussiere J, Italiano A, Descat E, Ferron S, Cornelis F, Avril A, Brouste V and Bui BN: Sarcoma lung metastases treated with percutaneous radiofrequency ablation: results from 29 patients. Ann Surg Oncol : 2011.
22 Hioki Y, Seto M, Tomoda R, Yamazaki T, Sonoda J and Uchida A: Effects of biopsy on lung metastasis. Oncol Rep *7*: 837-840, 2000.
23 Brahimi-Horn MC, Chiche J and Pouyssegur J: Hypoxia and cancer. J Mol Med *85*: 1301-1307, 2007.
24 Matsubara T, Kusuzaki K, Matsumine A, Shintani K, Satonaka H and Uchida A: Acridine orange used for photodynamic therapy accumulates in malignant musculoskeletal tumors depending on pH gradient. Anticancer Res *26*: 187-194, 2006.
25 Mohamed MM and Sloane BF: Cysteine cathepsins: multi-functional enzymes in cancer. Nat Rev Cancer *6*: 764-775, 2006.
26 Egeblad M and Werb Z: New functions for the matrix metalloproteinases in cancer progression. Nat Rev Cancer *2*: 161-174, 2002.
27 Li JP: Heparin, heparin sulfate and heparanase in cancer: remedy for metastasis? Anticancer Agents Med Chem *8*: 64-76, 2008.
28 Gollnick SO and Brackett CM: Enhancement of antitumor immunity by photodynamic therapy. Immunol Res *46*: 216-226, 2010.
29 Chaput N, Taïeb J, Schartz NE, André F, Angevin E and Zitvogel L: Exosome-based immunotherapy. Cancer Immunol Immunother *53*: 234-239, 2004.
30 Litle VR, Christie NA, Fernando HC, Buenaventura PO, Ferson PF and Luketich JD: Photodynamic therapy for endobronchial metastases from nonbronchogenic primaries. Ann Thorac Surg *76*: 370-375, 2003.
31 Prosst RL, Winkler S, Boehm E and Gahlen J: Thoracoscopic fluorescence diagnosis (TFD) of pleural malignancies: experimental studies. Thorax *57*: 1005-1009, 2002.
32 Zaak D, Karl A, Knuchel R, Stepp H, Hartmann A, Reich O, Bachmann A, Siebels M, Popken G and Stief C: Diagnosis of urothelial carcinoma of the bladder using fluorescence endoscopy. BJU Int *96*: 217-222, 2005.
33 Shintani K, Matsumine A, Kusuzaki K, Morikawa J, Matsubara T, Wakabayashi T, Araki K, Satonaka H, Wakabayashi H, Iino T and Uchida A: Decorin suppresses lung metastases of murine osteosarcoma. Oncol Rep *19*: 1533-1539, 2008.
34 Tomoda R, Seto M, Hioki Y, Matsumine A, Kusuzaki K and Uchida A: Low-dose methotrexate inhibits lung metastasis and lengthens survival in rat osteosarcoma. Clin Exp Metastasis *22*: 559-564, 2005.

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The adverse effect of an unplanned surgical excision of foot soft tissue sarcoma

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Abstract

Background: Malignant soft tissue tumors of the foot are extremely rare and thus can be prematurely excised without appropriate preoperative evaluation. The present study compares adverse effects between unplanned and planned surgical excisions.

Methods: We retrospectively reviewed the clinical records, radiographs, pathology reports and pathological specimens of 14 consecutive patients with soft tissue sarcoma of the foot among 592 with sarcomas between 1973 and 2009. We then compared the incidence and clinical outcomes after unplanned (UT; n = 5) and planned (PT; n = 9) surgical excisions of foot sarcomas.

Results: The most frequent diagnosis was synovial sarcoma (n = 4; 28.6%). The overall 5-year survival rates of the PT and UT groups were 65.6% and 60.0%, respectively, and the event-free 5-year survival rates were 63.5% and 40.0%, respectively. Event-free and overall survival rates did not significantly differ between the two groups. However, tumors were significantly larger in the PT group than in the UT group ($p < 0.05$).

Conclusions: Unplanned resection lead to a relatively worse prognosis and a likelihood of recurrence despite additional resections. We recommend that soft tumors of the foot should only be excised after appropriate preoperative evaluation regardless of the size of the tumor.

Background

Soft tissue sarcomas are rare malignancies that account for < 1% of all adult malignancies that annually develop in the United States of America [1]. Among them, malignant soft tissue tumors of the foot are extremely rare, occurring at a rate of 2% to 5% of soft tissue malignancies [2], whereas benign soft-tissue tumors, such as schwannoma, lipomas and hemangiomas, arise more frequently [3-5].

Soft-tissue sarcomas of the foot and benign tumors are notoriously difficult to clinically differentiate [6-8], as their similar presenting features include a palpable mass, swelling, increased warmth, limping and pain. Furthermore, soft-tissue sarcomas of the foot tend to be relatively smaller than those of the proximal limbs, because discomfort while wearing shoes results in seeking medical attention. These non-specific clinical features coupled with the relative rarity and small size, frequently lead to inadequate treatment of soft tissue sarcomas of the foot compared with sarcomas at other anatomical sites [8].

The initial choice of treatment for soft-tissue sarcoma influences the final oncological outcome [6,9]. Thus, wide surgical excision of a sarcoma of the foot after diagnostic imaging, and accurate biopsy is crucial to achieve a good clinical outcome. However, many potential treatment errors are associated with soft tissue sarcomas of the foot. Unplanned surgical excision of a foot sarcoma without appropriate diagnostic evaluation is quite common [6,9,10]. Unplanned surgical excision can result in an inappropriate surgical margin that will require further wide surgical excision and residual disease that can cause repeated local recurrences that can result in limb amputation or be life-threatening.

The present study compares the prognostic outcomes of unplanned and planned surgical excisions of soft-tissue sarcomas of the foot.

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Methods

We retrospectively reviewed the medical records, radiographs and pathology reports of 592 patients who presented at our hospital with soft tissue sarcoma between 1973 and 2009. Among these, we enrolled 14 consecutive patients with foot sarcoma. The exclusion criteria were benign tumors, bone tumors, myeloma, lymphoma, metastatic carcinoma, and tumors arising from the skin. The mean follow-up period was 56.5 months (range 6 - 170 months) after the initial consultation. The patient group included 10 males and four females with a mean age of 35.8 (range 2 - 67) years at the first presentation. Clinical records, radiographs, pathology reports, and pathological specimens were retrospectively reviewed to compare incidences and clinical outcomes of unplanned (UT group) and planned (PT group) surgical resections. All histological diagnoses were made by our institutional pathologists. The tumor grade was assessed according to the updated version of the FNCLCC system based on tumor differentiation, the mitotic count and necrosis [11]. Unplanned surgical excision was defined as that proceeding without preoperative imaging evaluation or staging studies. The cases initially treated by a general surgeon or general orthopedic surgeon without considering the possibility of malignancy were classified in the UT group. Among the UT group patients, extensive hematoma and inappropriate drainage and improper skin incision were observed in 2, 1 and 5 patients, respectively. In contrast, planned surgical resection was defined as that proceeding with appropriate preoperative imaging evaluation. The cases treated by orthopedic oncologists or treated under the supervision of an orthopedic oncologist were classified in the PT group. As a result, there were no patients who showed extensive hematoma, inappropriate drainage or an improper skin incision in the PT group. Variables included patient age, gender, tumor location, grade, surgical margin of initial surgery, tumor size (≥3 cm, group B; < 3 cm, group S), complications, overall survival rates, event-free survival rates and limb preservation (yes or no) between the UT and PT groups. The overall survival was defined as the time from the initial treatment to the date of death attributed to sarcoma. The event-free survival was defined as the time from the initial treatment to the date of clinically documented local recurrence/distant metastasis. The locations of soft-tissue sarcomas in the foot were classified according to the zones described by Kirby et al. [12]. A transverse plane was drawn from the mid-point of the metatarsal heads to the level of the insertion point of the Achilles tendon, and two vertical planes were drawn from the mid-tarsal point to the posterior end of the longitudinal plantar arch and another from the metatarsophalangeal joints down to the sole of the foot. These three lines form five zones, including the region of the ankle (zone 1), heel (zone 2), dorsum of foot (zone 3), sole (zone 4) and toes (zone 5). Soft-tissue sarcomas arising in zone 1 were excluded. Sarcomas were staged according to the Enneking staging system [13].

The Ethics Committees of Mie University approved the study.

Statistical analysis

Data were statistically analyzed using SPSS Statistics version 18.0 (SPSS Inc, Chicago, IL). P values below 0.05 were considered significant. All data are expressed as means ± standard deviation (SD). Associations in data between the UT and PT groups were determined using an unpaired t-test or the χ^2 test. Actuarial data for overall and event-free survival rates at the final follow-up were calculated using Kaplan-Maier analysis.

Results

Table 1 shows details of the 14 patients who were treated for soft-tissue sarcoma of the foot. The most common diagnoses were synovial sarcoma in 4 patients and clear cell sarcoma and rhabdomyosarcoma in one patient each. Five patients underwent unplanned resection elsewhere before referral (UT group), and nine underwent biopsy and surgical excision at our institution (PT group).

Seven patients had wide excision of the tumors or additional wider excision of lesions that had been inadequately excised. Six patients had undergone amputation. One patient died of the disease before undergoing wide resection. Another patient did not require additional excision because the estimated surgical margin after the initial excision was negative. Four patients died due to the sarcoma and all of them had pulmonary metastasis. One patient died of other causes. The mean time from surgery to death was 13.8 months (range 11 - 38).

Chemotherapy was administered to 5 patients with synovial sarcoma, rhabdomyosarcoma, fibrosarcoma and leiomyosarcoma. Radiation therapy was administered to 2 patients. Only one patient with high-grade sarcoma (rhabdomyosarcoma) received both chemotherapy and radiation therapy. Five did not receive adjuvant chemotherapy after resection because the histological grade was low or intermediate.

Figures 1 and 2 show Kaplan-Meier analysis for event-free and overall survival rates, respectively. The overall 5-year survival rate of all patients was 66.8%. The overall 5-year survival rates for the PT and UT groups were 65.6% and 60.0%, respectively. The overall event-free 5-year survival rate of all of our patients was 55.0%. The event-free 5-year survival rates for the PT and UT groups were 63.5% and 40.0%, respectively. Two developed local recurrence and six developed lung metastases. Only one patient with clear cell sarcoma had

Table 1 Patients' data

Case	Age	Sex	Diagnosis	LC	CC	Stage	FNCLCC grade	Size	FSM	LP	Chem	RT	Follow-up (months)	Prog	Group
1	25	M	RMS	4	Uncertain	II-B	3	S	amp	No	No	Yes	6	DOD	UT
2	46	F	SS	5	Mass	II-B	3	S	amp	No	Yes	No	7	DOD	UT
3	50	M	LPS	3	Mass	I-B	1	B	wide	Yes	No	No	24	CDF	PT
4	2	F	RMS	4	Swelling	III-B	3	S	no op	Yes	Yes	Yes	11	DOD	PT
5	33	M	CCS	5	Mass	II-B	2	B	amp	No	No	No	128	CDF	PT
6	41	M	SS	4	Mass	I-B	2	S	ad	Yes	No	No	89	CDF	UT
7	10	M	FS	3	Mass	II-B	2	B	wide	Yes	Yes	No	170	CDF	PT
8	67	M	CSSP	4	Uncertain	II-B	2	B	amp	No	No	No	7	DOC	PT
9	42	M	LMS	4	Pain, mass	II-B	2	B	amp	No	Yes	No	38	DOD	PT
10	39	F	SS	4	Tenderness	II-B	2	B	ad	Yes	Yes	No	92	AWD	UT
11	47	M	SS	4	Mass	II-B	3	B	amp	No	No	No	110	CDF	PT
12	19	F	CCS	2	Tenderness	II-B	2	S	ad	Yes	No	Yes	84	NED	UT
13	66	M	ESMCS	4	Uncertain	I-B	1	B	wide	Yes	No	Yes	12	AWD	PT
14	14	M	DFSP	3	Mass	II-A	2	B	wide	Yes	No	No	13	CDF	PT

Patients are in chronological order. LC, location; CC, chief complaint; FNCLCC grade, Fédération Nationale des Centres de Lutte Contre le Cancer Grading System; FSM, final surgical margin; LP, limb preservation; Chem, chemotherapy; RT, radiation therapy; Prog, Prognosis. CCS, Clear cell sarcoma; CSSP, chondrosarcoma of soft part; DFSP, dermatofibrosarcoma protuberance; ESMCS, extraskeletal myxoid chondrosarcoma; FS, fibrosarcoma; LMS, leiomyosarcoma; LPS, liposarocoma; RMS, rhabdomyosarcoma; SS, synovial sarcoma. Tumor size: S, < 3 cm; B, ≥3 cm; amp, amputation; wide, wide resection; no op, no operation; ad, additional wider resection; AWD, alive with disease; CDF, complete disease free; NED, no evidence of disease; DOD, died of disease; DOC, died of other causes. PT, planned excision; UT, unplanned excision.

inguinal node metastases. Event-free and overall survival rates did not significantly differ between the PT and UT groups.

Among patient age, gender, tumor location, histological grade, tumor size, complications and limb preservation, only tumor size significantly differed between the UT and PT groups ($p < 0.05$).

Discussion

Soft tissue tumors or tumor-like lesion of the foot are usually histologically benign [4]. Ganglions comprise about one-third of all soft tissue tumors [5]. Because patients with asymptomatic small tumors do not often receive appropriate medical care, the literature does not always reflect the true proportion of soft-tissue tumors that are foot sarcomas. However, Berlin [3] identified 449 sarcomas in an analysis of 307,601 tumors and tumor-like conditions of the foot. Kirby et al. [4] reviewed 83 patients with soft-tissue tumors and tumor-like conditions of the foot and ankle and identified 11 (13%) malignant tumors of which synovial sarcoma was the most frequent. Temple et al. [8] described 12 (34.3%) synovial sarcomas among 39 soft tissue sarcomas of the foot and ankle. Thacker et al. [14] described 17 (32.7%) among 52 soft tissue sarcomas and Cribb et al. [15] found 12 (44.4%) among 27 soft tissue sarcomas. We found here that synovial sarcomas (n = 4; 28.6%) comprised the most frequent soft tissue sarcoma of the foot, even after tumors originating in the ankle were excluded.

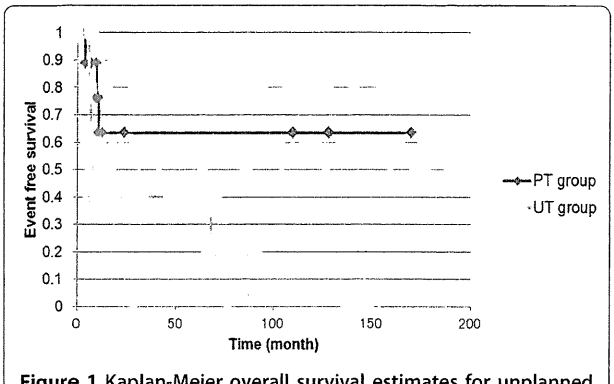

Figure 1 Kaplan-Meier overall survival estimates for unplanned and planned excisions of foot sarcoma. No significant differences between PT and UT groups. PT, planned excision; UT, unplanned excision.

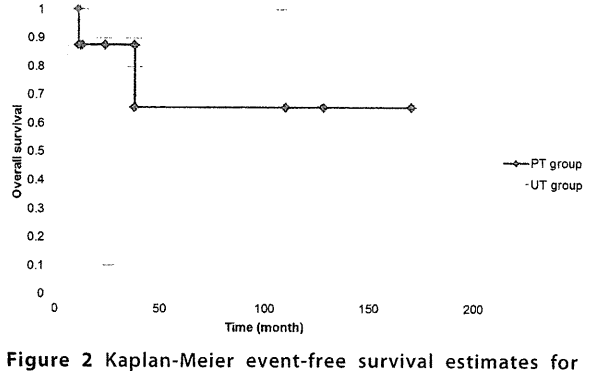

Figure 2 Kaplan-Meier event-free survival estimates for unplanned and planned excisions of foot sarcoma. No significant differences between PT and UT groups. PT, planned excision; UT, unplanned excision.

The prognosis of sarcomas is better when they are located in the distal extremity compared with the proximal extremity. Zeytoonjian et al. [16] described that the overall death rate all sarcomas was 26.6% (630/2367 patients), whereas that of foot sarcoma was 10.3% (18/170 patients). Cribb et al. [15] reported a rate of 14.8% (4/27 patients). Thacker et al. [14] indicated a 5-year survival rate of 78.2% (11/52 patients) among 52 patients. The 5-year survival rate of 66.8% (4/14 patients) in the present study was relatively lower than those described by others [14-16]. However, the 5-year survival rate of latest five patients was 100%. We speculated that improvements in diagnostic imaging technology including MRI enabled accurate preoperative evaluation of tumors. MRI is useful to know tumor size, tumor location and tumor margin. And, we believe that preoperative MRI is useful for evaluating and diagnosing foot tumors, because most of the benign soft-tissue tumors that frequently occur on the foot, such as schwannoma, epidermal cysts, ganglion cysts, lipomas and hemangiomas, can be diagnosed using MR imaging [3-5]. For the diagnosis of schwannoma, Tinel's-like sign with specific MRI findings (target sign) may be useful. In the case of epidermal cysts, T1-weighted images after gadolinium administration show rim-enhancement with subcutaneous involvement. Ganglion cysts can be easily diagnosed because of the homogenous intensity of the lesion. Lipomas can also be easily diagnosed due to their homogeneous high-intensity area on both T1-and T2-weighted images. Although hemangioma is sometimes difficult to distinguish from malignancy, the MR findings often show lobulation, septation, and central low-signal-intensity dots with small fat intensity foci. On the contrary, when a definite imaging diagnosis can not be obtained, the surgeon should carefully excise the tumor after considering the possibility of malignancy, even if the tumor size is small. These preoperative imaging analyses might have been associated with a good clinical result.

Five (35.7%) of 14 patients in the present study had unplanned excisions. However, Davis et al. [6] and Goodlad et al. [10] reported that 43.5% and 40% of patients with soft-tissue sarcomas in their series, respectively, underwent unplanned resections; our findings were comparable to these results. The physical features of soft tissue sarcomas of the foot, including palpable mass, swelling, local warmth, and pain, are similar to those of benign lesions, and thus many smaller lesions tend to undergo simple unplanned excisions without appropriate diagnostic imaging and biopsies. These non-specific features might in part facilitate the high incidence of unplanned surgical excisions. Indeed, tumors were significantly smaller in the UT group than in the PT group in the present series. This result might paradoxically show that smaller tumors throw clinicians off guard. An excisional biopsy may be justified when the tumor is less than 3 cm. However, the problem is whether the initial surgery was performed while considering the possibility of malignancy. Unfortunately, in this study, all of the UT group patients were initially treated without considering the possibility of malignancy, and this led to extensive hematoma formation, inappropriate drainage and improper skin incisions. Therefore, we classified the cases initially treated by general surgeons or general orthopaedic surgeon without considering the possibility of malignancy as part of the UT group.

Unplanned excisions of soft tissue sarcomas are frequent, but their influence on local recurrence, distant metastasis and patient survival remains controversial [17-19]. Davis et al. [6] compared the clinical outcomes between patients primarily treated at an institution specializing in cancer and those who were referred following unplanned excision at a general hospital. They found a higher local recurrence rate in the group with unplanned surgical excisions, especially those with residual tumors in re-resected specimens. Clasby et al. [9] reviewed 377 patients with primary soft-tissue sarcomas, and estimated that 21.3% (80/377 patients) of them had been inadequately treated. Furthermore, they noted a poorer outcome among patients with tumors that recurred after marginal excision. Thacker et al. [14] reported 5-year oncological survival rates of 80.4% (21/23 patients) and 73.4% (22/29 patients) in patients who underwent planned and unplanned excisions, respectively, and 5-year event free survival rates of 68.1% (9/23 patients) and 60.8% (11/23 patients), respectively. Temple et al. [8] reported 4-year oncologic survival rates of 76.5% (13/17 patients) and 77.8% (12/18 patients) in the PT and UT groups, respectively, and local recurrence rates of 11.8% (2/17 patients) and 16.7% (3/18 patients), respectively.

In the present series, 5-year overall survival rate in the PT group was 65.6% (7/9 patients) and that in the UT group was 60.0% (3/5 patients). The 5-year event free survival rates were 63.5% (6/9 patients) and 40.0% (2/5 patients) in the PT and UT groups, respectively. Rates of tumor recurrence or tumor death did not significantly differ between the UT and PT groups. However, tumors were significantly larger in the PT group than in the UT group in this study. Larger tumors significantly and negatively affect prognosis in patients with soft tissue sarcoma [20,21]. These findings suggested a relatively worse prognosis for the UT group than for the PT group. Because the absence of a statistical difference was probably due to the low number of recurrences and metastasis in the present study, further large-scale studies are warranted.

In point of the anatomical site of foot sarcoma, dorsal side (location 3) sarcomas of the foot were all big size (> 3 cm), On the other side, plantar side (loation 2 and 4) sarcomas of the foot were 4/9 cases small size and 5/9 big size. Thus, dorsal side sarcomas of the foot tend to be neglect until the tumor grew big size. Patients who have

plantal sarcomas of the foot tend to be seen in hospital earlier, because they might feel discomfort and pain of their sole.

The major shortcoming of this study was the limited patient population, because soft tissue sarcoma of the foot is extremely rare. Therefore, the statistical power of this study is very low.

Most soft tissue tumors of the foot are small and benign, which results in frequent unplanned excisions. Although benign tumors would not pose a clinical problem, malignant tumors lead to a higher incidence of local recurrence even after additional surgery and result in a poor prognosis. We emphasize that unplanned excisions should be averted by appropriate preoperative evaluation such as by MRI, early referral of all potential malignancies to a cancer center, biopsy assessment and wide excision of malignant lesions.

Conclusions

Sarcomas of the foot are rare and thus tend not to be suspected by either patients or clinicians. Event-free and overall survival rates did not significantly differ between the PT and UT groups, but tumors were significantly larger in the PT group. Larger tumors negatively affect the prognosis in patients with soft tissue sarcoma. Therefore, we concluded that the UT group had a relatively worse prognosis than the PT group. Therefore, we conclude that unplanned resection leads to a relatively worse prognosis and a likelihood of recurrence despite additional resections. We recommend that even small soft tumors of the foot should be preoperatively evaluated by imaging to avoid repeated local recurrence.

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Authors' contributions

AM was the lead author and surgeon for all of the patients. KA, TM and TN were the co-surgeon on the cases and contributed patients and information on the patients. AU reviewed paper and technique of surgery. KK contributed to writing of the paper. AS reviewed paper. All authors read and approved the final manuscript.

Competing interests

The authors declare that they have no competing interests.

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References

1. Westbury G: **The management of soft tissue sarcomas.** *J Bone Joint Surg Br* 1989, 71:2-3.
2. Papagelopoulos PJ, Mavrogenis AF, Badekas A, Sim FH: **Foot malignancies: a multidisciplinary approach.** *Foot Ankle Clin* 2003, 8:751-763.
3. Berlin SJ: **Statistical analysis of 307,601 tumors and other lesions of the foot.** *J Am Podiatr Med Assoc* 1995, 85:699-703.
4. Kirby EJ, Shereff MJ, Lewis MM: **Soft-tissue tumors and tumor-like lesions of the foot. An analysis of eighty-three cases.** *J Bone Joint Surg Am* 1989, 71:621-626.
5. Ozdemir HM, Yildiz Y, Yilmaz C, Saglik Y: **Tumors of the foot and ankle: analysis of 196 cases.** *J Foot Ankle Surg* 1997, 36:403-408.
6. Davis AM, Kandel RA, Wunder JS, Unger R, Meer J, O'Sullivan B, Catton CN, Bell RS: **The impact of residual disease on local recurrence in patients treated by initial unplanned resection for soft tissue sarcoma of the extremity.** *J Surg Oncol* 1997, 66:81-87.
7. Simon MA, Enneking WF: **The management of soft-tissue sarcomas of the extremities.** *J Bone Joint Surg Am* 1976, 58:317-327.
8. Temple HT, Worman DS, Mnaymneh WA: **Unplanned surgical excision of tumors of the foot and ankle.** *Cancer Control* 2001, 8:262-268.
9. Clasby R, Tilling K, Smith MA, Fletcher CD: **Variable management of soft tissue sarcoma: regional audit with implications for specialist care.** *Br J Surg* 1997, 84:1692-1696.
10. Goodlad JR, Fletcher CD, Smith MA: **Surgical resection of primary soft-tissue sarcoma. Incidence of residual tumour in 95 patients needing re-excision after local resection.** *J Bone Joint Surg Br* 1996, 78:658-661.
11. Guillou L, Coindre JM, Bonichon F, Nguyen BB, Terrier P, Collin F, Vilain MO, Mandard AM, Le Doussal V, Leroux A, Jacquemier J, Duplay H, Sastre-Garau X, Costa J: **Comparative study of the National Cancer Institute and French Federation of Cancer Centers Sarcoma Group grading systems in a population of 410 adult patients with soft tissue sarcoma.** *J Clin Oncol* 1997, 15:350-362.
12. Kirby EJ, Shereff MJ, Lewis MM: **Soft-tissue tumors and tumor-like lesions of the foot. An analysis of eighty-three cases.** *J Bone Joint Surg Am* 1989, 71:621-626.
13. Enneking WF, Spanier SS, Goodman MA: **A system for the surgical staging of musculoskeletal sarcoma.** *Clin Orthop Relat Res* 1980, 106-120.
14. Thacker MM, Potter BK, Pitcher JD, Temple HT: **Soft tissue sarcomas of the foot and ankle: impact of unplanned excision, limb salvage, and multimodality therapy.** *Foot Ankle Int* 2008, 29:690-698.
15. Cribb GL, Loo SC, Dickinson I: **Limb salvage for soft-tissue sarcomas of the foot and ankle.** *J Bone Joint Surg Br* 2010, 92:424-429.
16. Zeytoonjian T, Mankin HJ, Gebhardt MC, Hornicek FJ: **Distal lower extremity sarcomas: frequency of occurrence and patient survival rate.** *Foot Ankle Int* 2004, 25:325-330.
17. Lewis JJ, Leung D, Espat J, Woodruff JM, Brennan MF: **Effect of reresection in extremity soft tissue sarcoma.** *Ann Surg* 2000, 231:655-663.
18. Potter BK, Adams SC, Pitcher JD, Temple HT: **Local recurrence of disease after unplanned excisions of high-grade soft tissue sarcomas.** *Clin Orthop Relat Res* 2008, 466:3093-3100.
19. Rougraff BT, Davis K, Cudahy T: **The impact of previous surgical manipulation of subcutaneous sarcoma on oncologic outcome.** *Clin Orthop Relat Res* 2005, 438:85-91.
20. Nakanishi H, Tomita Y, Ohsawa M, Naka N, Araki N, Ochi T, Aozasa K: **Tumor size as a prognostic indicator of histologic grade of soft tissue sarcoma.** *J Surg Oncol* 1997, 65:183-187.
21. Mandard AM, Petiot JF, Marnay J, Mandard JC, Chasle J, de Ranieri E, Dupin P, Herlin P, de Ranieri J, Tanguy A, *et al*: **Prognostic factors in soft tissue sarcomas. A multivariate analysis of 109 cases.** *Cancer* 1989, 63:1437-1451.

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Extraskeletal subcutaneous osteosarcoma of the upper arm: A case report

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Abstract. Extraskeletal osteosarcoma (ESOS) occurs in approximately 1% of soft tissue sarcomas and 2-4% of all osteosarcomas. In particular, subcutaneous osteosarcoma is extremely rare, occurring in less than 10% of ESOS cases. This report presents a case of a subcutaneous tumor in the upper arm of a 79-year-old male. Imaging and pathological findings led to the conclusion that the soft tissue tumor should be diagnosed as subcutaneous osteosarcoma. Additionally, this case report documented the clinicopathological findings of the extraskeletal subcutaneous osteosarcoma in this case and discussed its clinical features by reviewing cases previously described in the literature.

Introduction

Extraskeletal osteosarcoma (ESOS) is a rare malignancy that accounts for approximately 1% of all soft tissue sarcomas and for 2-4% of all osteosarcomas (1-3). ESOS usually occurs in the deep soft tissue of the extremities of adults (4). It typically arises in the deep soft tissue of the thigh. Other less frequent sites include the buttock, shoulder, trunk and retroperitoneum. Approximately 24% of cases have been associated with previous radiotherapy or trauma (5). In contrast to primary osteosarcoma of the bone, this variant typically develops after the fifth decade of life, and the prognosis is uniformly poor (5,6). The present report documented the clinicopathological findings in a patient who had an ESOS arising from the subcutaneous tissue of the upper arm and reviews previous cases of subcutaneous ESOS.

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Key words: osteosarcoma, subcutaneous tissue, extraskeletal

Case report

A 79-year-old male was referred to the Mie University Hospital due to an enlarged, slightly painful mass in the left upper arm. The patient first noted the mass 3 years prior to presentation. No history of trauma or therapy had previously occurred at this site. Moreover, the patient had experienced no recent health problems and recalled no family history of cancer. A physical examination confirmed the presence of a lobulated hard mass with a diameter of 4 cm at the lateral side of the left upper arm. Radiographs of the left upper arm revealed a mass with ossification which appeared to be separated from the humerus (Fig. 1). Magnetic resonance images (MRI) showed a soft tissue mass above the fascia of the triceps with a low signal intensity on T1-weighted images and a heterogeneous signal intensity on T2-weighted images (Fig. 2). Computed tomography (CT) of the chest did not demonstrate any pulmonary masses. All routine laboratory data were normal. The long clinical course from the first awareness of the tumor and the clinical findings, suggested that the tumor was benign or a malignant calcifying epithelioma with ossification. The patient was treated with wide resection and a skin graft from an inguinal lesion. Gross sectioning of the specimen showed a 4x4x2.5 cm firm and solid mass in the deep dermis and subcutaneous tissue (Fig. 3). A microscopic examination showed the presence of numerous spindle and atypical cells often exhibiting pronounced nuclear atypia or multinucleated giant cells with bone and osteoid formation. A high mitotic activity with numerous atypical mitoses was noted (Fig. 4). These findings led to the conclusion that this soft tissue tumor was a subcutaneous ESOS. The patient had no evidence of local recurrence and distant metastasis 1 year following resection.

Discussion

Since ESOS was initially described by Wilson in 1941, approximately 300 cases have been reported thus far (5,6). ESOS is defined as a malignant mesenchymal neoplasm composed of cells producing osteoid, bone and/or chondroid material, with no attachment to bone or periosteum (7). It occurs most often in the deep soft tissues of the extremities of adults, at an average age of 50 years. Clinically, ESOS usually carries an extremely poor prognosis. The 5-year survival rate ranges

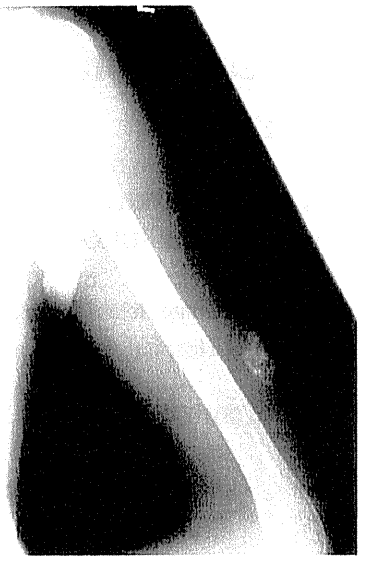

Figure 1. Radiographs revealed a subcutaneous soft tissue mass with ossification in the upper arm.

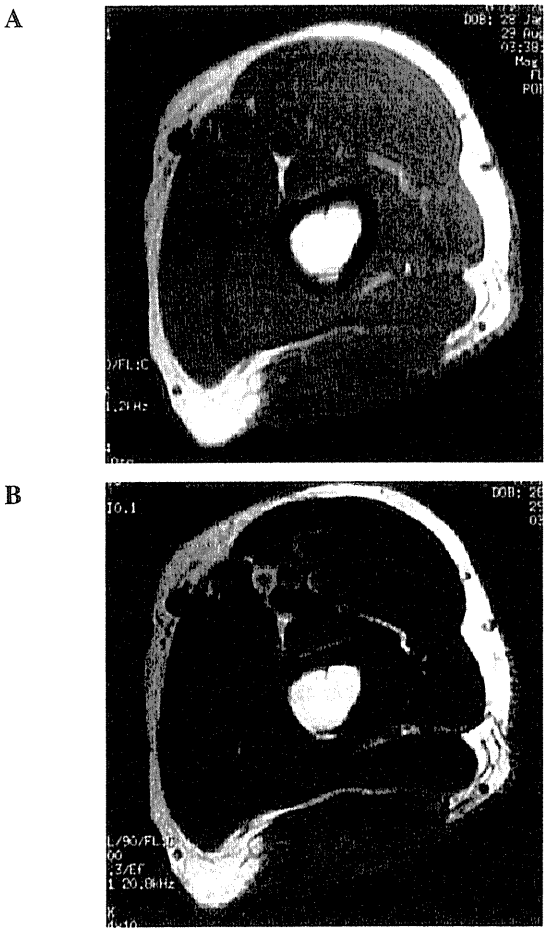

Figure 2. MRI showed a soft-tissue mass above the fascia of the triceps with (A) a low signal intensity on T1-weighted images and (B) a heterogeneous signal intensity on T2-weighted images.

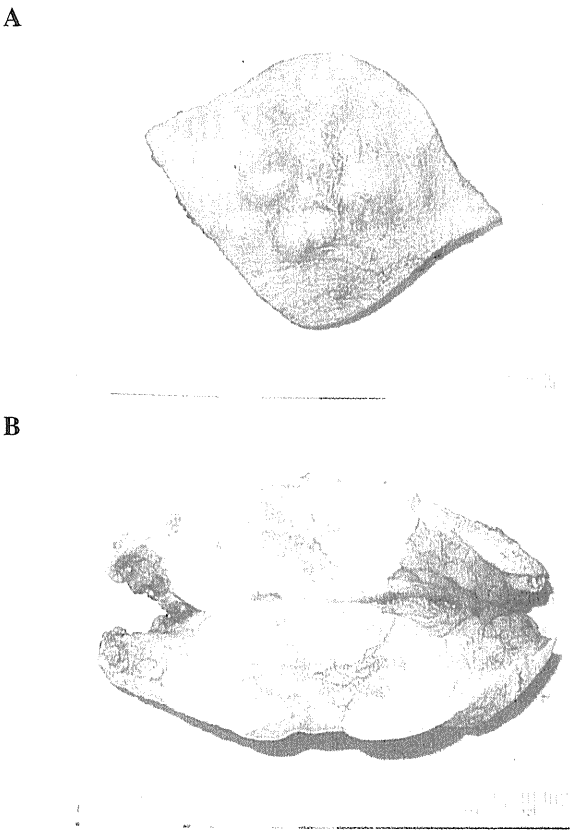

Figure 3. Gross appearance of the tumor (A, surface; B, cut surface). (A) Gross appearance of the resected tumor showed a lobulated hard mass without cutaneous ulceration. (B) The cut surface of the specimen showed a 4x4x2.5 cm firm and solid mass in the deep dermis and subcutaneous tissue.

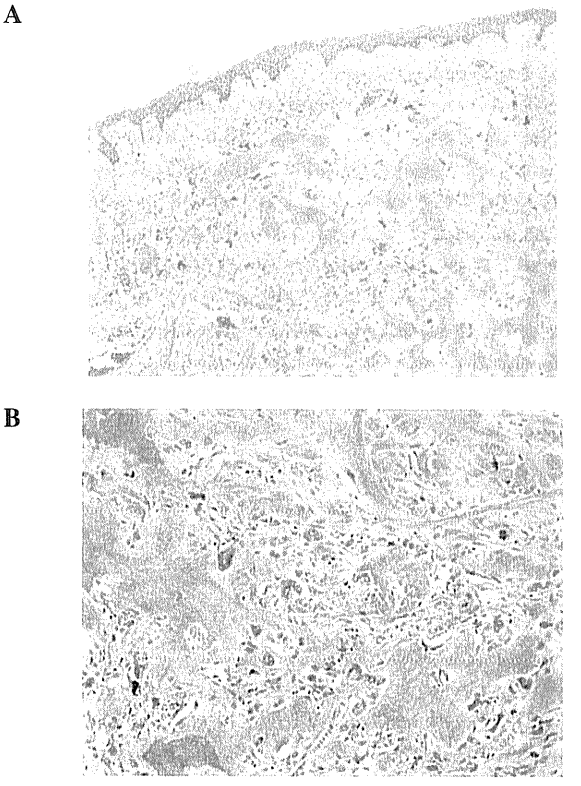

Figure 4. Microscopic findings showed numerous spindle and atypical cells with one or more nuclei, occasional multinucleated giant cells and osteoid and bone formation. H&E staining (A, magnification, x10; B, magnification, x40).

from 25 to 37% for ESOS as previously reported (3,4). The tumor size appears to be the only reliable prognostic variable in that tumors greater than 5 cm have a poor prognosis (3).

Table I. Review of the literature describing cases of primary subcutaneous extraskeletal osteosarcoma.

Author	Age/gender	Location	Size (cm)	Therapy	Outcome
Fang *et al* (8)	59/female	Abdominal wall	1.0	Surgery	CDF, 16 years
Yamakage *et al* (9)	62/male	Forehead	3.0	Surgery	DOD, 2 months (brain metastasis)
Dubec *et al* (10)	75/female	Lower leg	15.0	Surgery	CDF, 12 months
Pillay *et al* (11)	56/male	Scalp	10.0	Surgery and chemotherapy	Unknown
Oonuma *et al* (6)	55/female	Buttock	1.0	Surgery and chemotherapy	CDF, 48 months
Matsumoto *et al* (12)	68/ female	Buttock	1.0	Surgery	CDF, 16 months
Hatano *et al* (13)	25/male	Jaw	1.5	Surgery and chemotherapy	CDF, 16 months
Nakamura *et al* (Present study)	79/male	Upper arm	4.0	Surgery	CDF, 12 months

CDF, continuously disease-free; DOD, dead of disease.

Subcutaneous ESOS was rarely reported. Only eight cases, including the present one, were found in the literature (6,8-13) (Table I). Patients in those studies included 4 males and 4 females, ranging in age from 25 to 79 years. Lesions were located in the buttock in 2 cases and in the scalp, forehead, jaw, abdominal wall, lower leg and upper arm each in 1 case. In general, ESOS develops in the lower extremities, with the thigh being involved most; however, these 8 cases developed ESOS in various anatomical sites. The tumor size was less than 5 cm in all but 2 cases. A surgical resection was performed in all cases. The consequent surgical margin was wide in 7 patients, including an additional wide resection in 2 cases and an intralesional margin in 1 case. A total of three patients received chemotherapy. A wide surgical resection was performed in the present case. No chemotherapy was administered as a result of the advanced age of the patient. A wide margin is generally recommended for ESOS, as for other high-grade sarcomas (14). Lee *et al* reported that recurrence is common in ESOS and usually occurs in more than half of the patients (3). However, a wide (or radical) resection should decrease the recurrence of ESOS.

The role of adjuvant chemotherapy in ESOS is unclear. A recent series (14,15) found that the 5-year survival rate of patients with ESOS receiving chemotherapy showed an obvious improvement in comparison to what was described in previous reports (3,4). The two most recent reports found that the 5-year survival rate of patients with chemotherapy was approximately 70% (14,15). Although adjuvant therapy for ESOS remains controversial, chemotherapy may be useful in an aggressive multimodality approach to this tumor.

The 5-year survival rates associated with ESOS are relatively poor. However, 7 of the 8 cases of subcutaneous ESOS were continuously disease-free. The prognostic significance of the tumor location with respect to its relationship to the superficial fascia of the extremity or trunk was incorporated into the staging system of soft tissue sarcoma in 1998 (16). Although only 9 patients with primary subcutaneous ESOS were previously reported in the literature, these reports may indicate that subcutaneous ESOS has a more favorable prognosis than their more deeply situated counterparts.

References

1. Allan CJ and Soule EH: Osteogenic sarcoma of the somatic soft tissue. Clinicopathologic study of 26 cases and review of literature. Cancer 27: 1121-1133, 1971.
2. McCarter MD, Lewis JJ, Antonescu CR and Brennan MF: Extraskeletal osteosarcoma: analysis of outcome of a rare neoplasm. Sarcoma 4: 119-123, 2000.
3. Lee JY, Fetsch JF, Wasdhal DA, Lee BP, Pritchard DJ and Nascimento AG: A review of 40 patients with extraskeletal osteosarcoma. Cancer 76: 2253-2259, 1995.
4. Kransdorf MJ and Meis JM: From the archives of the AFIP. Extraskeletal osseous and cartilaginous tumors of the extremities. Radiographics 13: 853-884, 1993.
5. Wilson H: Extraskeletal ossifying tumors. Ann Surg 113: 95-112, 1941.
6. Oonuma M, Hatori M, Hosaka M and Kokubun S: Extraskeletal osteosarcoma arising in the buttock. Upsala J Med Sci 106: 211-215, 2001.
7. Enzinger FM and Weis SW: Osseous soft tissue tumors. In: Soft Tissue Tumors. Enzinger FM and Weiss SW (eds). CV Mosby Company, St. Louis, pp1389-1417, 2001.
8. Fang Z, Yokoyama R, Murai K, Beppu Y and Fukuma H: Extraskeletal osteosarcoma: a clinicopathologic study of four cases. Jpn J Clin Oncol 25: 55-60, 1995.
9. Yamakage A, Kohnoike N, Shiraishi M, *et al*: A case of extraskeletal osteosarcoma, which has been suspected to be malignant fibrous histiocytoma. Skin Cancer (in Japanese) 11: 161-166, 1996.
10. Dubec JJ, Munk PL, O'Connell JX, *et al*: Soft tissue osteosarcoma with telangiectatic features: MR imaging findings in two cases. Skeletal Radiol 26: 732-736, 1997.
11. Pillay P, Simango S and Govender D: Extraskeletal osteosarcoma of the scalp. Pathology 32: 154-157, 2000.
12. Matsumoto K, Sakai S, Iijima M, Matsumoto K, Saida T and Kanai S: Subcutaneous tumor on the left buttock. J Jpn Dermatohistopathol Assoc (in Japanese) 19: 18-21, 2003.
13. Hatano H, Morita T, Kobayashi H, Ito T, Segawa H and Hasegawa S: Extraskeletal osteosarcoma of the jaw. Skeletal Radiol 34: 171-175, 2005.
14. Torigoe T, Yazawa Y, Takagi T, Terakado A and Kurosawa H: Extraskeletal osteosarcoma in Japan: multiinstitutional study of 20 patients from the Japanese Musculoskeletal Oncology Group. J Orthop Sci 12: 424-429, 2007.
15. Markin HJ, Hornicek FJ, Rosenberg AE, Harmon DC and Gebhardt MC: Survival data for 648 patients with osteosarcoma treated at one institution. Clin Orthop 429: 286-291, 2004.
16. Fleming ID, Phillips JL, Menck HR, Murphy GP and Winchester DP: The National Cancer Data Base report on recent hospital cancer program progress toward complete American Joint Committee on Cancer/TNM staging. Cancer 80: 2305-2310, 1997.

Retrospective analysis of metastatic sarcoma patients

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Abstract. Numerous studies have reported the survival of metastatic sarcoma patients who have undergone either a lung metastasectomy or chemotherapy. However, little is known with regards to the clinical course of patients with bone or soft tissue sarcomas who have succumbed to disease. This study aimed to analyze the metastatic patterns of sarcoma patients and to describe the clinical course after the detection of distant metastasis. We reviewed the clinical records of 255 patients with a diagnosis of sarcoma who were referred to our institution, and found 63 patients who succumbed due to metastasis. We examined the clinical features of the initially detected distant metastases, the subsequent clinical course up to the time of patient death and the survival time of patients who died of lung metastasis. Of the 63 patients who died of distant metastasis, 52 (83%) developed lung metastasis as the first metastatic site, while 22 (35%) developed extra-pulmonary metastasis. The majority (77%; 49 of 63 patients) died of primary metastasis. While all 18 bone sarcoma patients died of lung metastasis, 11 of the 45 soft tissue sarcoma patients died of extra-pulmonary metastasis. Six patients died of brain metastasis. The survival of the patients with lung metastasis was only approximately 6 months following the cessation of treatment, regardless of the type of treatment used. These results indicate that planned follow-up and treatment of sarcomas require a precise knowledge of tumor clinical behavior, particularly of the preponderant activity.

Introduction

Although the prognosis for patients with bone or soft tissue sarcomas has shown marked improvement over the past three decades, those who develop local recurrence or metastatic disease continue to have high mortality rates. Of all patients diagnosed with malignant musculoskeletal tumors, 5-30% (1-3) have a recurrence and 10-38% of patients present with clinically detectable metastases (4-6). A number of studies have reported the survival of patients who have undergone either a lung metastasectomy or chemotherapy (4,5,7-9).However, little is known about the clinical course of patients with bone or soft tissue sarcomas who have succumbed to the disease. For example, only a few reports are available regarding the clinical course, such as the metastatic site, which led to the cause of death or the survival time following initial detection of metastases (10). Thus, this study aimed to analyze the metastatic patterns of sarcoma patients and to describe the clinical course following the detection of distant metastasis.

Patients and methods

We retrospectively reviewed the medical records of newly diagnosed patients with either bone or soft tissue sarcoma referred to Mie University Hospital between January 1999 and December 2008. Well-differentiated liposarcomas were excluded from this study. During this period, 255 patients with a diagnosis of sarcoma were referred to our institution. We reviewed the clinical records of these patients and found 63 patients who died of metastasis.

We examined the clinical features of the initially detected distant metastases, the subsequent clinical course up to the patient's death and the survival time of the patients who succumbed to lung metastasis.

Statistical analysis. Overall survival was estimated using the Kaplan-Meier method. Factors affecting patient survival were examined based on the log-rank test. The StatMate III program (ATMS Co., Tokyo, Japan software) was used to conduct statistical analysis. Post-metastatic survival was defined as the duration between the first detection of distant metastases and patient death. The post-treatment survival time was defined as the duration between the final treatment [surgery, lung radiofrequency (RF) ablation and chemotherapy] for metastases and death.

Results

Patient and tumor characteristics and follow-up. A total of 34 male and 29 female patients were included in our study. The average age was 51 years (range 6-80), including 17 patients who were ≥70 years of age. The follow-up periods ranged from 1 to 82 months (average 16) after the initial detection of metastasis.

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Key words: cause of death, metastasis, sarcoma

The histological diagnoses of the primary bone tumor were osteosarcoma (n=10), malignant fibrous histiocytoma (MFH) of the bone (n=3), chondrosarcoma (n=2), Ewing's sarcoma (n=2) and others (n=2). Soft tissue tumor included malignant peripheral nerve sheath tumors (n=9), MFH (n=9), leiomyosarcoma (n=6), myxoid liposarcoma (n=4), synovial sarcoma (n=3), alveolar soft part tumor (ASPS) (n=3), extra-skeletal myxoid chondrosarcoma (n=2), epithelioid sarcoma (n=2) and others (n=6). According to the American Joint Commission on Cancer classification and stage grouping of bone sarcomas, 9 were classified as stage 2B, 6 as 3A and 1 as stage 3B. Of the soft tissue sarcomas, 3 were classified as stage 2, 26 as stage 3 and 18 as stage 4.

Post-treatment follow-up was performed at 3-month intervals for 2 years, then every 4-6 months for 5 years, and every year subsequently. At follow-up, a chest X-ray was routinely carried out, and an examination using computed tomography (CT) (X-Vigor or Aquilion; Toshiba, Tokyo, Japan; or HiSpeed Advantage Qx/I; GE Healthcare, USA; or Asteion; Toshiba) was performed at 3- to 6-month intervals to detect distant metastasis. A CT scan of the abdomen and pelvic cavity was performed at 6-month intervals for the patients with myxoid liposarcoma.

Treatment of the primary tumor. A surgical resection was performed in 56 of the 63 patients. Local recurrence occurred in 26 of the 56 patients. In 10 of the 26 patients, the local recurrence had developed prior to the first detection of distant metastasis. Local recurrence was treated with surgical resection (n=20), radiotherapy (n=4) and chemotherapy (n=3). In total, 7 of the 63 patients were not treated with surgery for their primary tumor. Four patients underwent radiotherapy due to their advanced age (n=2), or an unresectable tumor size and location (n=2). One patient with post-radiation MFH at a sacro-iliac lesion underwent carbon ion radiotherapy since it was considered to be less invasive. One patient received only palliative therapy for multiple metastatic lesions. A 6-year-old female with osteosarcoma in her femur was treated only with chemotherapy as her family rejected surgical excision.

Clinical features of the initially detected distant metastases and the clinical course until patient death. The location of the initially detected distant metastases was investigated. In 53 of the 63 patients, lung metastasis developed (83%) and in 21 patients (33%) extra-pulmonary metastasis developed. The average size of the maximum diameter of the lung metastases was 10 mm (3-40 mm), and the average number of lung metastases was 5 (1-20) upon detection.

The sites of extra-pulmonary metastasis were bone (n=10), liver (n=5), soft tissue (n=3), brain (n=2), heart (n=2), pelvic cavity (n=1) and stomach (n=1). The occurrence of lung and extra-pulmonary metastases was observed in 11 patients. Ten patients had only extra-pulmonary metastasis without any co-existing lung metastasis in the following sites: bone (n=5), liver (n=1), brain (n=1), heart (n=1), subcutaneous tissue (n=1) and pelvic cavity (n=1) (Fig. 1).

Of the 53 patients who had lung lesions at the first detection of distant metastasis, 46 died of progression of lung metastasis, 2 of liver metastasis, 4 of brain metastasis and 1 of adrenal metastasis. Of the 5 patients who had bone lesions at the first

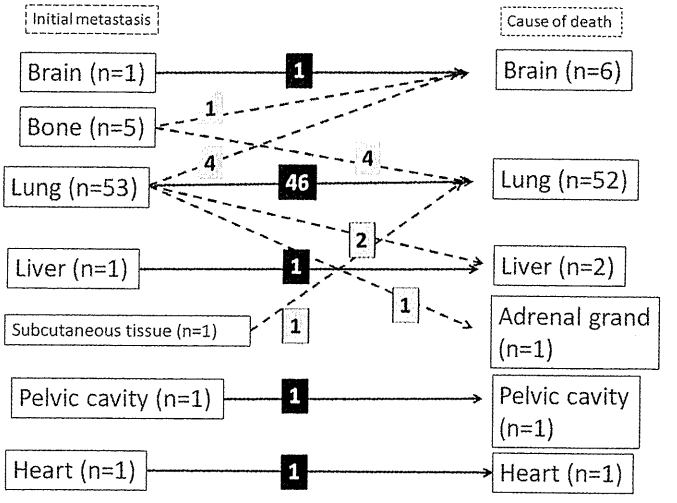

Figure 1. Relationship between the primary metastatic site and the cause of death: left section, initial metastatic site; right section, focus with cause of death.

detection of distant metastasis, 4 died of lung metastasis and 1 of brain metastasis. One patient who had distant metastasis in the subcutaneous tissue at the first detection of metastasis died of lung metastasis. Each patient who had metastasis in the brain, liver, heart or pelvic cavity at the first detection of the metastasis died of aggravation of the initial metastasis (Fig. 1). As a result, of the 63 patients, 52 died of lung metastasis, 6 of brain, 2 of liver and 1 of heart, pelvic cavity or adrenal metastasis. A total of 77% of patients (49 of the 63) succumbed to the primary metastasis.

While all 18 bone sarcoma patients died of lung metastasis, 11 of the 45 soft tissue sarcoma patients succumbed to extra-pulmonary metastasis. Seven of these 11 patients died of other additional distant metastases. The 3 patients with ASPS died of brain metastases.

The 10 patients that survived >2 years following the initial detection of distant metastasis developed other additional distant metastases (Table I). The sites of additional distant metastasis were: retroperitoneum (n=2), bone (n=2), soft tissue (n=1), lymph node (n=1), radial nerve (n=1), lung (n=1), adrenal grand (n=1) and brain (n=1).

Length of survival of the 52 patients who succumbed to lung metastasis. The length of survival of the 52 patients who died of lung metastasis was examined. The median post-treatment survival of the 20 patients who underwent treatment for metastasis, including metastasectomy, RF ablation and chemotherapy, was 6 months (Fig. 2). The following treatments for the lung metastases were performed: lung RF ablation alone (n=5), lung RF ablation and chemotherapy (n=6), metastasectomy and chemotherapy (n=4), chemotherapy alone (n=4), and metastasectomy and lung RF ablation (n=1).

The median post-metastatic survival of the 32 patients who received no treatment for their metastasis was 7.2 months. By contrast, the median post-metastatic survival in the 20 patients who underwent treatment for metastasis was 16 months. This represents a significant difference in the survival rate (P=0.003) (Fig. 3).

Table I. Pattern of metastases in patients who survived more than 2 years following the initial metastasis.

Diagnosis	Initial metastatic site	Other metastatic site	Cause of death	Follow-up periods (months)
ASPS	Lung	Brain	Brain	45
Synovial sarcoma	Lung	Adrenal gland	Adrenal gland	32
Ewing's sarcoma	Lung	Retroperitoneum	Lung	85
MGCT	Lung	Soft tissue	Lung	63
MFH	Subcutaneous	Lung	Lung	43
ESCS	Lung	Lymph node	Lung	59
Synovial sarcoma	Lung	Radial nerve	Lung	29
Myxoid sarcoma	Lung	Retroperitoneum	Lung	25
Osteosarcoma	Lung	Bone	Lung	35
Rhabdomyosarcoma	Lung	Brain	Brain	24

ASPS, alveolar soft part sarcoma; MGCT, malignant granular cell tumor; MFH, malignant fibrous histiocytoma; ESCS, extra-skeletal chondrosarcoma.

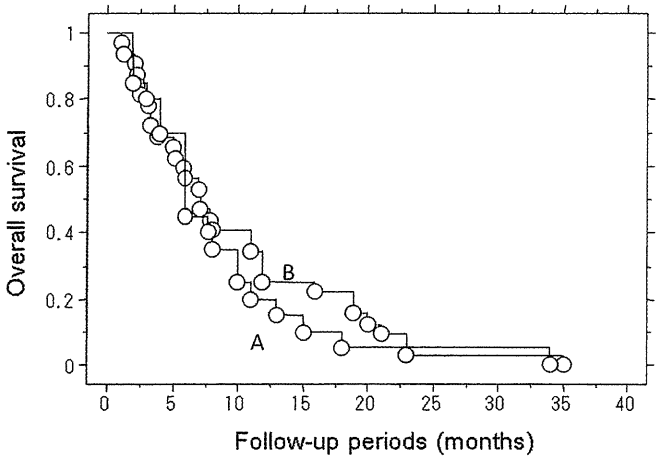

Figure 2. Kaplan-Meier curves showing the post-treatment survival of 20 patients receiving treatment for the metastasis (A), and the post-metastatic survival of 32 patients who did not receive any treatment for the metastasis (B).

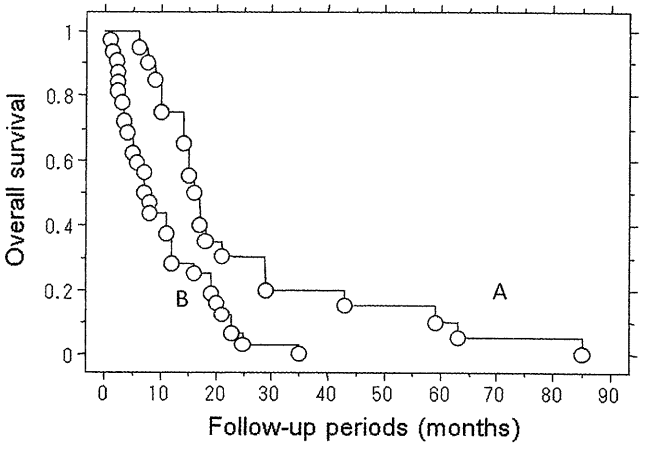

Figure 3. Kaplan-Meier curves showing post-metastatic survival in patients who received treatment (A) and patients who did not receive any radical treatment (B).

Discussion

Regarding the location of metastases, 62-82% of sarcoma patients had lung metastasis and 50-70% had isolated lung metastases in previous studies (4,11). In our series, 83% (52 of 63 patients) developed lung metastasis as the initial metastatic site. Therefore, the present results are consistent with previous reports. Although numerous reports exist regarding the initial metastatic site in sarcoma patients and the treatment of patients with metastasis, few reports are available regarding the location of the metastatic lesion that ultimately caused patient death (4,5,7-11). In the present study, 82.5% of patients succumbed to lung metastasis and 10% to brain metastasis. Other patients succumbed to liver (3%), heart (1.5%), adrenal grand (1.5%) and pelvic metastasis (1.5%).

Brain metastases from soft tissue sarcoma are believed to occur in a minority of soft tissue sarcoma patients, with a reported prevalence ranging between 1 and 6% (12,13). In osteosarcoma, the incidence of brain metastasis is reported to be 2-6.5% (14). Particularly in the absence of lung metastases, brain metastasis is thought to be a relatively uncommon event in the natural history of bone and soft tissue sarcomas. In our cases, only 1 patient with epithelioid sarcoma had brain metastasis without lung metastasis. ASPS has been reported to metastasize to the brain more commonly than other types of high-grade sarcoma, although the majority of studies concerning ASPS are in the form of case reports and small corrective series (15-17). In our present series, brain metastasis developed within 4 years of the primary surgery in the 3 patients with APSP. We therefore recommend the regular use of intracranial CT imaging for patients with ASPS.

Although the prognosis of sarcoma patients has improved, extra-pulmonary metastases should be further investigated. In our series, 11 of the 45 soft tissue sarcoma patients died of extra-pulmonary metastasis. In addition, 7 of the 11 patients succumbed to other additional distant metastases. Although follow-up chest CT and physical examinations were routinely