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On the Natural History of Osteosarcoma

Aspects on diagnosis, prognosis and endocrinology

BY

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Dignity is inherent in the state of being human,
for no man can by his fellow man be used only as a means,
but must always constitute the purpose as well,
and just therein lies his dignity.

Immanuel Kant

The present work is based on the following papers, which will be referred to by their Roman numerals:

I. **Broström, L.-Å., Harris, M., Simon, M., Cooperman, D., Nilsonne, U.**: The effect of biopsy on survival of patients with osteosarcoma. J. Bone Jt. Surg. 61B: 209–212, 1979.

II. **Broström, L.-Å., Aparisi, T., Ingimarsson, S., Lagergren, C., Nilsonne, U., Strander, H. Söderberg, G.**: Can historical controls be used in current clinical trials in osteosarcoma? Analysis of prognostic factors in a historical and a contemporary group. Int. J. Rad. Oncol. Biol. Phys. (In press).

III. **Broström, L.-Å., Aparisi, T., Ingimarsson, S., Lagergren, C., Nilsonne, U., Strander, H., Söderberg, G.**: Can historical controls be used in current clinical trials in osteosarcoma? Metastases and survival in a historical and a contemporary group. Int. J. Rad. Oncol. Biol. Phys. Accepted for publication.

IV. **Broström, L.-Å., Ingimarsson, S., Strander, H., Eklund, G.**: Correlation between prognostic factors and blood variables in osteosarcoma. Acta med. scand. (In press).

V. **Broström, L.-Å., Adamson, U., Filipsson, R., Hall, K.**: Longitudinal growth and dental development in osteosarcoma patients. Acta Orthop. Scand. Accepted for publication.

VI. **Adamson, U., Broström, L.-Å., Efendić, S., Hall, K.**: Glucose tolerance, growth hormone and somatomedin levels in osteosarcoma patients. Acta endocr. (Kbh). Accepted for publication.

CONTENTS

CHAPTER 1 – INTRODUCTION

History

Primary malignant bone tumours account for less than 1 % of all malignant tumours in man in all ages.[26,131] Osteosarcoma occurs predominantly during the first three decades of life and is the most common primary malignant bone tumour in this age group.[14,26,61,67,77,79,86,104,116,118,131,147]

Smith and Dawson[123] reported in 1924 that evidence has been found dating the occurrence of primary bone tumours as far back as the fifth Egyptian dynasty. The term osteosarcoma was used by Boyer[6] as early as 1807 and a first attempt at classifying bone tumours into malignant and benign forms was presented by Nelaton[92] in 1860. More systematic classifications have later been published by Codman[20] in 1925, Scarff[117] in 1937, Ewing[39] in 1939, and Cade[13] in 1947, and more recently by Lichtenstein[79] in 1952, Dahlin[26] in 1957, and Jaffe[61] in 1958.

The term osteogenic sarcoma is encountered early in the literature without any clear distinction from chondrosarcoma and fibrosarcoma, which were often included in this concept. Some authors have used the term osteogenic sarcoma as a collective designation for different malignant primary tumours of bone,[81,82,83] whereras others use the term as synonymous to osteosarcoma.[28,61,79,86,94,97,136] In recent years the term osteogenic sarcoma has been virtually ousted by osteosarcoma to designate the specific primary malignant bone tumour which histopathologically is characterized by osteoid-producing malignant bone cells.[28,78,118,136] An annual incidence of 2 cases per million inhabitants has been reported.[74,103]

Pathology

It is often difficult to make a diagnosis of osteosarcoma on the basis of the histopathological findings, especially as the bone tissue contains cells of many different types and varying stages of maturity. It has been suggested that the tumour cells originate from immature osteogenic mesenchymal cells, in combination with often completely anaplastic connective tissue cells of spindle-shaped type.[14, 28, 29, 36, 80, 82] Giant cells of both malignant and benign type are frequently seen, but an absolute condition for a diagnosis of osteosarcoma is the presence of osteoid.[26,28,61,79,81,83,136]

Tumours designated as osteosarcoma may be further distinguished according to the predominant cell type into an osteoblastic, chondroblastic or fibroblastic variety. In large series, such as those presented by Dahlin,[26-29] about 50 % of the tumours are of the osteoblastic type, while the chondroblastic and fibroblastic types account for about 25 % each. Different portions of the solid tumour may present an entirely different histological picture, which makes it difficult to differentiate osteosarcoma from chondrosarcoma or fibrosarcoma, and sometimes also from wholly benign conditions like aneurysmal bone cyst, fibrous dysplasia or chronic osteomyelitis.[26,61,79]

The degree of malignancy can be assessed by examining the degree of differentiation of the tumour cells as reflected by nuclear appearance and mitotic frequency. Broders *et al.*[9] have described such a grading system based on a four-grade scale in which grade IV represents the least differentiated tumour type. Another malignancy grading system based on mitotic frequency has been presented by Price.[101]

Diagnosis

The diagnosis of osteosarcoma is based on both clinical, radiological and histopathological findings. Somewhat varying meanings have been attached to the term classical osteosarcoma in the literature, but in the present work it is used to define intraosseous osteosarcoma in young individuals, usually under 30 years of age, with a peak incidence during the second decade of life often coinciding with the pubertal growth spurt. The most common site of the tumour is the metaphyseal region of the long bones and a predominance has been noted among males. The radiological findings often present a typical pattern with destructive tumour growth through the cortex. Frequent findings are osteolysis and osteosclerosis, as well as the formation of bony spicula and the separation of the periosteum producing what is known as Codman's triangle.[26,61,79]

Parosteal and periosteal variants of osteosarcoma have an entirely different clinical course with a higher survival rate.[30,36,81,144,145] The histopathological picture in these tumour forms is marked by considerably higher differentiation.[144, 145] Such tumours, as well as extraosseous,[107] multiple,[2,43] and secondary forms of osteosarcoma such as those resulting from irradiation or Paget's disease are usually excluded from the concept of classical osteosarcoma in the literature.[81]

Material

The various investigations comprising the present study were partly based on different clinical series, which are presented in Table I. The term Contemporary Group indicates that this group of patients is contemporary to the adjuvant therapy initiated in the 1970's, whereas the term Historical Group refers to the group of patients predating the introduction of such treatment.

Table I. Clinical series reported in Papers I–VI.

Paper no	Time period	Geographical area	No of patients	Group	Adjuvant therapy	
I	1938–1959	Karolinska Hospital	105	–	–	
II	1952–1972	"	35	Historical	–	
	1972–1974	Sweden	44 (23+21)	Contemporary	interferon	(21)
III	1952–1972	Karolinska Hospital	35	Historical	–	
	1972–1974	Sweden (excl. Karolinska Hospital)	23	Contemporary	–	
IV	1972–1978	Karolinska Hospital	33	–	interferon	(33)
V	1972–1974	Sweden	44	Contemporary	interferon	(21)
VI	1976–1979	Karolinska Hospital	15	–	interferon	(15)

Patients with classical primary osteosarcoma were included in all series. Diagnosis was consistently based on both clinical and radiological findings and on light microscopic examination of tumour specimens. In every case included in the series histological examination was carried out by two independent pathologists, both thoroughly familiar with tumour diagnostics.

No patient included in the series had any signs of metastases at the time of diagnosis. The X-ray films of the initial lung examination were re-examined in every single case. Lung tomography or scanning was not performed in any series to exclude patients who could be expected to have a poorer prognosis.

The size of the primary tumour was evaluated in collaboration with a radiologist by measuring the greatest diameter visualized by X-ray films in different views. Type and duration of symptoms were elicited from the clinical records. Blood tests were performed on admission to hospital, before treatment was initiated.

Background and aims of the study

For the past several decades the 5-year survival rates for patients with primary osteosarcoma have ranged between 10 % and 20 %.[15,49,81,115] Treatment has for decades consisted in surgery or radiation, or a combination of both.[49] In the early 1970's various kinds of adjuvant therapy were introduced in an attempt to improve survival.[24,64,128] Initially excellent results were reported for chemotherapy, but these were based on very small and often selected series with a short period of observation.[42] With increased observation periods different centres have reported highly varying effects of this treatment.[15]

Starting in 1972, patients admitted to the Karolinska Hospital without signs of metastatic spread at the time of diagnosis have been given human leukocyte interferon as adjuvant therapy for a period of 18 months.[129] The project was started as a pilot study and the initial results showed a considerably higher survival rate in comparison with a historical control group.[128] The study has since been extended and at present comprises 42 patients.

No studies have been published using randomized treatment protocols to compare adjuvant treatment of osteosarcoma (high-dose cytostatics or interferon) with non-adjuvant treatment. Evaluation of the metastasis-free rate and survival has consequently been based on historical control series.[15,24,49,56,64,115]

During recent years the validity of such historical control series has been called in question.[50,91,112,122,139] One reason is that diagnosis in recent years presumably is made at an earlier stage and that more active treatment has been adopted especially in cases with metastatic spread of the disease. Some centres have moreover reported findings suggesting that the natural history of osteosarcoma itself may be changing.[8,42,139]

We are consequently faced with considerable difficulties in evaluating the effect of different kinds of adjuvant therapy in osteosarcoma.[50,91,122] The present work was initiated in an attempt to test the reliability of historical controls in clinical trials in osteosarcoma and to elucidate the possible influence of a number of different factors on its course.

The following factors were studied: Biopsy; certain prognostic factors (age, sex, type and duration of symptoms, tumour size and site, tumour pathology, primary tumour treatment, treatment delay, treatment of metastases); hormonal and metabolic factors (longitudinal growth, dental development, growth hormone and somatomedin levels, glucose tolerance); blood variables (haemoglobin, erythrocyte sedimentation rate, leukocyte and thrombocyte count, albumin, haptoglobin, immunoglobulin, aspartate aminotransferase, alanine aminotransferase and alkaline phosphatase).

The primary aims of the present study were:

1. To investigate the risks of open biopsy in the diagnosis of osteosarcoma.
2. To study the value and reliability of historical control series by an analysis of factors assumed to affect prognosis.
3. To compare a historical and a contemporary group with respect to metastasis-free rate and survival in relation to prognostic factors.
4. To study the internal and mutual relationship between certain prognostic factors and blood variables.
5. To study longitudinal growth and dental development in osteosarcoma patients.
6. To study glucose metabolism in osteosarcoma patients by the glucose infusion test, and to determine growth hormone and somatomedin levels.

CHAPTER II – RESULTS

The effect of biopsy on survival of patients with osteosarcoma (I)

Between 1938 and 1959 a consecutive series of 105 patients with primary osteosarcoma without evidence of metastatic spread at the time of diagnosis was treated at the Karolinska Hospital. The histopathological and radiological findings in this series have been reported in an earlier study.[80] The patients selected from this series for the present study were all under 30 years of age, and had a tumour located in the metaphyseal region of a long bone which had been removed by radical surgery, i.e. amputation of the limb or disarticulation. No patient had received any kind of adjuvant chemotherapy.

Fifty-seven patients selected by these criteria could be classified as having classical primary osteosarcoma. These patients were divided into two groups. Group I includes 24 patients who had an amputation without previous biopsy. In these cases diagnosis was based only on clinical and radiological findings and subsequently verified by analysis of operation specimens. In Group II, comprising 33 patients, diagnosis was based on analysis of biopsy material. In 8 of these cases frozen sections were obtained with or without vascular occlusion by a proximally placed tourniquet immediately prior to amputation. In the remaining 25 cases a two-stage procedure was adopted with a mode of 4 days between open biopsy and radical surgery.

The two main groups were wholly comparable with respect to such factors as age (mean age 16.3 and 15.7 years, respectively) and sex (42 % females in both groups), and similar with regard to tumour site and treatment. Seven patients in Group I and as many in Group II had received preoperative radiation therapy. The radiation dose varied individually and no standard regimen had been followed.

All of the patients had been followed up for more than 5 years and there was no difference in the survival rate for the two groups at this time. The 5-year survival rate was 21 % in both groups, regardless of whether or not biopsy had been done. Analysis of the patients who survived for 5 years would seem to indicate radical surgery as a common denominator.

Can historical controls be used in current clinical trials in osteosarcoma?

Analysis of prognostic factors (II)

Two groups of patients with osteosarcoma of the long bones without evidence of metastases at the time of diagnosis were analyzed with respect to factors assumed to influence prognosis. One was a historical group comprising 35 patients treated at the Karolinska Hospital during the years 1952–1972, the other a contemporary group including all of the 44 patients treated for osteosarcoma in Sweden during the period 1972–1974. This latter group includes 21 patients admitted to the Karolinska Hospital who received adjuvant therapy with human leukocyte interferon.

The clinical records, tumour slides and X-ray films of both primary tumour and lungs were reviewed independently by the pathologists and clinicians included in a National Cancer Institute (NCI) team visiting Sweden in May, 1976. A unanimous diagnosis of primary osteosarcoma was reached for the 35 patients subsequently included in the historical group and the 44 patients comprising the contemporary group.

An analysis of prognostic factors showed the two series to be comparable in age, with

a mean age of 17 years for the historical and 18 years for the contemporary group. Female patients were proportionally fewer in the contemporary group (1: 2.4 as compared to 1: 1.7), but the difference was statistically not significant. Pain was the most frequent presenting symptom in both groups, noted for all patients in the historical and 86 % of the contemporary group. The average duration of symptoms was 3 months in both groups, but the incidence of symptoms was significantly higher in the historical group. Similarly, a significantly larger tumour size (13 cm as compared to 9 cm) was found in the historical group. A tumour size of more than 10 cm was found in no less than 64 % of the patients in this group. A significantly higher incidence of poorly differentiated tumours was also noted in the historical group, which moreover showed a tendency toward a more proximal location of the tumour.

The study consequently disclosed differences between the two groups with respect to factors which might influence the prognosis. The incidence of unfavourable prognostic factors was higher in the historical than in the contemporary group.

Metastases and survival (III)

The historical and contemporary groups described in the preceding study (II) did not permit a valid comparison of the metastasis-free and survival rates since the contemporary group included 21 patients who had received adjuvant interferon therapy. Their exclusion left a contemporary group of 23 patients who did not receive any adjuvant therapy for comparison with the 35 patients in the historical group.

All patients were followed up for at least 2.5 years at which time the metastasis-free rate was assumed to have attained steady state. The incidence of patients without metastases in the contemporary group was 31 % at this time, as against 14 % in the historical group. A similar relationship could be noted with regard to the survival rate, which was twice as high for the contemporary group after 2.5 years' follow-up. In addition a delayed plateau phase was noted for the contemporary group, producing a displacement of the survival curve as compared to the historical group. This can be ascribed to the considerably more active approach adopted in the contemporary group in the treatment of lung metastases with both pulmonary surgery and high-dose chemotherapy. Half of the patients in this group, but none in the historical group had received surgical treatment for such metastases. Furthermore, 4 patients in the contemporary group had received high-dose chemotherapy as compared to none in the historical group. Radiation therapy was given to some patients in either group and in this respect the two groups were comparable.

Mean survival time from biopsy to death from the tumour was 19 months for the contemporary group and 13 months for the historical group, corresponding to the difference observed between the two groups with respect to the metastasis-free rate.

Analysis of prognostic factors disclosed that the historical group contained larger (13 cm as compared to 6 cm) and more malignant tumours (71 % osteoblastic Grade IV tumours as compared to 48 %). Similarly a proximal tumour site was more common in the historical group, even though about 75 % of all tumours in both groups were located in the knee region. Both the sex ratio, showing about one-third of the patients to be female, and the age distribution were comparable in the two groups. A higher incidence of symptoms was noted for patients in the historical group, who also had shorter intervals between lung X-ray check-ups.

Primary treatment of the tumour was largely the same in the two groups, with the exception of a higher incidence of pre-operative radiation therapy in the contemporary

group when the tumour dose exceeded 45 Gy. The percentage of patients treated with this dose was twice as high in this group. Disarticulation was more often performed in the historical group, whereas amputation was more common in the contemporary group.

The differences that could be noted between the two groups with respect to prognostic factors corresponded with the observed differences in metastasis-free rate and survival.

The results of this study indicate that there may be a risk involved in using historical controls when evaluating the effect of adjuvant therapy in osteosarcoma, especially when the series are small and the observation period is short.

Correlation between prognostic factors and blood variables (IV)

Prognostic factors and blood chemistry were recorded at the time of diagnosis, prior to any treatment, in a consecutive series of 33 patients with primary classical osteosarcoma and scheduled for an 18-month course of adjuvant interferon therapy at the Karolinska Hospital. All patients with clinical evidence of metastases on admission were excluded from the series.

Analysis of certain blood variables such as haemoglobin, leukocyte and thrombocyte counts, albumin, haptoglobin, aspartate aminotransferase (Asat), alanine aminotransferase (Alat) and immunoglobulins produced values within the normal range. Both the erythrocyte sedimentation rate and the alkaline phosphatase activity were slightly increased, but for this latter blood variable it is very difficult to define reliable normal values for growing individuals.

Analysis of prognostic factors showed that one-third of the patients were female, while the average age for the total series was 20 years and the average duration of symptoms 4 months. The tumours averaged 10 cm in diameter and in 13 patients were located distally to the knee or elbow. The analysis moreover showed a prevalence of osteoblastic high-grade tumours.

The recorded prognostic factors were subsequently related to each other and to the analyzed blood variables. The results of this study show that female patients had a higher incidence of less malignant, more differentiated, fibroblastic tumour types ($p < 0.05$), which are assumed to have a better prognosis. A distal tumour site was more common for smaller tumours and was found to have less influence on the recorded blood variables ($p < 0.001$).

Endocrinology

Longitudinal growth and dental development (V)

Between the years 1972–1974 a total of 44 patients (31 males and 13 females) were registered in Sweden under the diagnosis of osteosarcoma without signs of metastatic spread at that time. Information on longitudinal growth from birth until the age of diagnosis was obtained from the records of child health centres and school health services where body height is registered at regular intervals. These data provided complete growth curves for 19 patients (9 boys and 10 girls).

Analysis of height measurements at 8.0 years, when growth is expected to be linear, and at the time of diagnosis – which occurred at a mean age of 13 years for both boys and girls in the series – showed entirely normal growth on comparison with growth curves for other Swedish children at those ages.[137] At age 8 a mean height of 128.4 cm was noted for male patients as compared to 128.6 cm for their controls. The corre-

sponding figures for girls were 129.1 cm and 127.4 cm, respectively. A significant correlation was noted between the standardized height at 8.0 years and the standardized height at the age of diagnosis ($r = 0.88$, $p < 0.001$).

A relationship exists between longitudinal growth and tooth eruption.[41] Dental maturity was analyzed by a method reported by Filipsson.[40] Sufficient data on tooth eruption to permit analysis could be obtained from national and school dental services for 24 patients (16 males and 8 females). Dental development was entirely normal in these osteosarcoma patients, as evidenced by the fact that the mean age noted at the reference point on the tooth eruption curve was 8.67 years for male patients as compared to an age of 8.75 years calculated for controls.[40] Mean age for female patients and their normal controls[40] was 8.36 and 8.44 years, respectively.

Both longitudinal growth and dental maturity were consequently found to be normal in this analysis of osteosarcoma patients.

Glucose tolerance, growth hormone and somatomedin (VI)

A study of glucose tolerance, serum levels of growth hormone and somatomedin, and glucose-induced insulin secretion was undertaken in a consecutive series of 15 patients with classical osteosarcoma admitted to the Karolinska Hospital. All tests were carried out before any treatment was initiated. All patients were in good general condition at the time. Both the mean duration of symptoms (3 months) and the age and sex distribution were typical for classical primary osteosarcoma. High-grade osteoblastic tumours predominated in the series.

Analysis of blood samples showed that somatomedin A and growth hormone levels were entirely within the normal range for age[17,54,55] in all patients at the time of diagnosis. This finding is consistent with the normal growth rate observed for osteosarcoma patients in the preceding study (V).

Glucose intolerance has been reported for osteosarcoma patients.[53] Glucose infusion tests (GIT)[18] were performed in all 15 patients to study the mechanism underlying the reported glucose intolerance in osteosarcoma. No difference in basal glucose and insulin levels was noted between the osteosarcoma patients and healthy controls[32] matched by weight, height and age. However, the disappearance rate of glucose[60] ("K-value") after glucose infusion was significantly decreased in the patient group. This difference was not related to age or sex.

A typical biphasic insulin response to the GIT was noted both in the osteosarcoma patients and in the control group. There was no difference in initial insulin response between the two groups, but during the latter part of the test significantly higher insulin levels were noted for the osteosarcoma patients. Decreased glucose tolerance might be explained by reduced insulin sensitivity and analysis of glucose and insulin response to the GIT by means of a mathematical model[19] did in fact demonstrate reduced insulin sensitivity in the osteosarcoma patients.

The results of this study showed that somatomedin A and growth hormone levels were within the normal range for age in the osteosarcoma patients. The decreased glucose tolerance noted in these patients may be explained by reduced insulin sensitivity.

CHAPTER III – GENERAL DISCUSSION

Biopsy and diagnosis

The diagnosis of osteosarcoma may present considerable difficulties even when aided by histologic examination of tumour tissue.[28] As a rule a diagnosis of primary classical osteosarcoma will have to be based on the combined evidence provided by the patient's history, the X-rays of both primary tumour and lungs, and histologic analysis of a representative specimen of tumour tissue.[14,77,136] The often polymorphous character of the tumour[36,79] urges the need of obtaining specimens from different parts of the neoplastic growth at the time of biopsy.[14] For this reason open biopsy is to be preferred, since aspiration or drilling[78,96,135,136] usually fails to provide sufficient material for analysis from different parts of the bone tumour, such as the periosteum, cortex or marrow, and when indicated extra-osseous soft tissue elements.

The advisability of obtaining specimens from several portions of the tumour by surgical biopsy prior to definitive surgery has been questioned on the suspicion that manipulation of the tumour might entail a risk of tumour cell embolisation,[34,44,45,95,109,142] which in its turn is suspected to increase the risk of metastasis. This theory is supported by animal studies.[71,89,143] Although no evidence has been reported of increased metastasis in man,[136] tumour cells have been demonstrated in the blood of patients with malignant tumours subjected to surgery.[44,96,134,141]

In our first study (I) we investigated whether open biopsy and a delay between biopsy and definitive surgery had any effect on the long-term prognosis. Although small, the series included in this study are comparable to the clinical series reported in connection with studies concerning adjuvant therapy in osteosarcoma in recent years.[15] No patient had any signs of metastases at the time of diagnosis and in all cases the tumour was removed by radical surgery, i.e. amputation or disarticulation of the affected limb.

Our study demonstrated no difference in the 5-year survival rate between patients who had an amputation without prior biopsy and those who had a biopsy. The survival rate of 21 % noted for both groups is consistent with the 5-year figures reported for large series.[27,49,56,76,81,83,86,101,136] Moreover we found no survivors among patients whose definitive surgery was delayed for more than 30 days after biopsy.

The radiological and clinical features of benign conditions like osteomyelitis may simulate osteosarcoma and the necessity of confirming the diagnosis by open biopsy has in recent years been strongly emphasized.[15] Considering the important diagnostic benefits to be gained from open biopsy and the fact that our study failed to demonstrate any negative effect on the long-term prognosis, it would seem that this procedure is to be recommended. However, the series will have to be enlarged to permit more reliable conclusions. Although the observed difference between the two groups is 0 %, a confidence interval (95 %) of 0±21 % can be calculated.

An alternative biopsy procedure involving proximal vascular occlusion by one or two tourniquets and analysis of frozen sections of tumour tissue during operation has been discussed by some authors.[27,36,72,80,135,138] However, many pathologists feel that frozen sections are unsatisfactory for a reliable diagnosis of osteosarcoma, since analysis of bone tissue often requires special methods of preparation. It has moreover been demonstrated that a proximally applied tourniquet primarily serves to occlude the venous flow but does not prevent intra-osseous circulation, and consequently cannot be claimed to provide a wholly reliable safeguard against tumour cell embolisation.[77]

Surgery

Authors discussing the level of amputation seem to be agreed that tumours located distally to the elbow or knee should be removed by transmedullary amputation of the proximal part of the limb, i.e. the thigh or upper arm.[36,135] Concerning the most common tumour site, the distal femur, opinions are more divided. Some authors feel that this tumour site demands disarticulation at the hip joint whereas others recommend proximal femoral amputation.[14,27,36,63,76,86,134] Underlying the demand for extreme radicality are some reports demonstrating the occurrence of so-called "skip lesions", that is, neoplastic foci at a different site from the primary tumour but within the same limb, usually in the same long bone but occasionally with a transarticular location.[14,35,37] Although these findings have been called in question,[29,66,78] the intramedullary encroachment observed in amputation specimens in some cases suggests that this may be more extensive than can be visualized by the preoperative X-rays.[134,146] Bone scans and tomography have regrettably proved to be of limited value as an additional aid in the preoperative assessment of tumour growth.[36,52,134] Angiography is primarily useful in demonstrating extraosseous tumor growth.[36,73,134,150]

The introduction in recent years of adjuvant therapy, which is believed to have an effect on micrometastasis, has led many surgeons to revise their views on amputation surgery and en bloc resection of the tumour has been frequently adopted instead.[1,65,85,93,110] This implies removal of the tumour with a smaller safe margin, often allowing preservation of an adequately functioning limb. The defect left by the tumour is replaced either by foreign implants such as plastic or metal prostheses or by autologous tissue such as bone grafts.[93]

If local resection is to satisfy the demand for complete radicality, it will be necessary to extirpate the site of any previous biopsy incision in one single piece with the rest of the tumour. This places additional demands on an adequate biopsy technique including selection of the right site for incision and caution in manipulating tumour tissue.[80,93,135] The same surgeon will often be required to perform both biopsy and definitive surgery.

At the Karolinska Hospital adjuvant therapy with human leukocyte interferon was introduced in 1971 and so far a consecutive series of 42 osteosarcoma patients without radiological evidence of metastases has been treated. Fifteen of these 42 patients were operated upon with local resection of the tumour following open biopsy.[93] Six of these patients have so far been followed up for at least five years and four are alive without evidence of tumour recurrence, while two have died from other causes.

Prognostic factors

Prior to the 1970's the survival rate for osteosarcoma patients was low – often less than 20 % after a 5-year follow-up.[27,49,56,77,80,81,83,86,97,135] In 1972 the first report on the positive results of chemotherapy (methotrexate, adriamycin) in metastatic osteosarcoma were published,[23,62] followed by the introduction of systematic treatment by various cytostatics as adjuvant therapy in patients with osteosarcoma without evidence of metastatic spread at the time of diagnosis. Early reports showed promising results, but were based on small series and short observation periods.[24,64,128] With longer observation periods the results were somewhat less favourable and today combined treatment with different drugs, often administered in maximum doses, yields a 5-year survival rate of 40–50 %.[25,38,48,57,66,69,99,112,133]

Around 1976 the question was raised whether the natural history of osteosarcoma might be changing, since some centres in recent years had noted higher survival rates

for patients treated only by conventional methods like surgery and irradiation in comparison with earlier series of similarly treated patients.[8,138] In this context the selection of patients was discussed, since patients with a presumably better prognosis might be more liable to be admitted to one centre than to another.[8,42,81] Another question raised was whether refined radiological diagnostic techniques including lung tomography and scintigraphy might contribute to the exclusion of patients with metastatic spread at the time of diagnosis, implying a poorer prognosis.[138] The higher survival rate in patients with lung metastases could to some extent be explained by the more active approach adopted in the treatment of such metastases. All this has cast doubt on the reliability of historical controls.

In the study undertaken at the Karolinska Hospital of adjuvant interferon treatment in patients with classical osteosarcoma without evidence of metastatic spread, we used a historical control group of 35 patients treated at this hospital between 1952 and 1972. To make evaluation of the effect of interferon treatment more reliable and to reduce the risk of selecting patients characterized by factors tending to influence prognosis in a positive or negative way, we collected all patients in Sweden who during the years 1972–1974 received a diagnosis of classical primary osteosarcoma without evidence of metastatic spread at that time and who did not receive adjuvant therapy. Together with the patients treated with interferon during the same period, this produced a contemporary group of 44 patients. A comparative analysis of a number of prognostic factors disclosed appreciable differences between the historical and the contemporary group.

Sex

It has earlier been suggested that female osteosarcoma patients have a better prognosis[45, 80,102,106,119,138] and the question has been raised whether this might be related to the female sex hormone, estrogen.[84,120] In large series of primary osteosarcoma the proportion of females is usually reported as one-third.[26] In our study of prognostic factors (II) the historical group included slightly more female patients than expected, while the ratio between females and males in our study on metastases and survival (III) was 1:2. In our analysis of prognostic factors and blood variables (IV) we found a longer duration of symptoms for female patients and a higher incidence of less malignant, i.e. low-grade fibroblastic tumours than among the males. Moreover it may be noted that two-thirds of the 12 patients surviving without tumour recurrence at the 5-year follow-up in our study on biopsy and survival (I) were female, whereas the sex distribution was reversed in the original series of 57 patients.

Age

One of the criteria commonly used to define the concept of classical primary osteosarcoma is that it affects patients under 30 years of age.[139] The Karolinska Hospital material includes 3 patients in the historical group and 4 in the contemporary group who were over 30, but otherwise manifested a clinical picture wholly typical of primary osteosarcoma. In some earlier studies a more favourable prognosis for older patients has been discussed.[27, 80, 83, 101] In our study correlating prognostic factors and blood variables (IV) we noted that the duration of symptoms increased with age, which might be ascribed to slower tumour growth in older patients. No correlation was found between age and tumour pathology, size or location..

Type and duration of symptoms

Pain is the most common symptom, followed by swelling.[26,81,83,136] Pathological fracture of the involved bone was noted in a few cases, but generally no prognostic importance has been attached to this in the literature.[81] The incidence of symptoms was higher in the historical than in the contemporary group but their duration, which is reported to be correlated to prognosis,[28,81,83,118] was the same in both groups. The more favourable prognosis associated with a longer duration of symptoms may be ascribed to a better individual reaction to the tumour resulting in slower tumour growth.[81,125] This assumption would also seem to be consistent with the positive correlation observed in our study on prognostic factors and blood variables (IV) between duration of symptoms and low-grade tumours as well as higher serum levels of immunoglobulin G.

Size and location of tumour

In our study on prognostic factors and survival (III) we found that tumour size, as measured by the greatest diameter on the X-rays, on the average was twice as large in the historical group as in the contemporary group. A significant difference in tumour size between these two groups was also observed in our analysis of prognostic factors (II), which yielded a tumour size exceeding 10 cm in 64 % of the patients in the historical group. A considerably poorer prognosis has been reported for tumours exceeding 10 cm in diameter.[81,83,118,136] In addition, our study on prognostic factors and blood variables (IV) disclosed a correlation between tumour size and its location, since we found distal tumours to be smaller. They were also associated with higher levels of haemoglobin and albumin, a lower erythrocyte sedimentation rate and a lower haptoglobin serum level. Although it should also be easier to recognize distally located tumours at an earlier stage this study (IV) failed to demonstrate a positive correlation between tumour size and duration of symptoms. Similarly, there was no correlation between tumour size and tumour pathology.

The prognostic significance of tumour location has been a subject of discussion in the literature. Some authors consider distal location a favourable sign,[27,29,49,75,81,147,148] whereas others fail to establish such a relationship.[80,83,102,118,119,136] Although the difference was statistically not significant, the historical group contained a greater number of proximally located tumours.

Tumour pathology

Several authors classify osteosarcoma according to the dominant cell type in an osteoblastic, chondroblastic and fibroblastic variety.[28,36,102] The value of this classification, however, has been called in question.[80,81,101,136] We have mentioned earlier that it is important to base histological examination on serial sections since different parts of the tumour may present varying features.[26,36] Some authors feel that the chondroblastic type is associated with the poorest prognosis,[101,119] whereas Dahlin's studies identify the osteoblastic type as the most malignant form of osteosarcoma.[27,28]

A predominance of osteoblastic Grade IV tumours was found in the historical group and this group totally included significantly more Grade IV tumours. Earlier reports have questioned the value of malignancy grading on the basis of tumour cell differentiation.[27,81,83,101,136,141,148] The literature provides no information to suggest a better prognosis for fibroblastic tumours, but our correlation study (IV) demonstrated that this type was more common in low-grade tumours and had a higher incidence among female

patients. Ten of the 33 patients included in this study were followed up for 5 years, and 5 of them survived without signs of tumour recurrence. Four of these 5 patients (80 %) had a fibroblastic tumour as compared to 8 of the 33 patients (24 %) in the original series. All 33 patients received adjuvant interferon therapy for an equally long period. The presence of osteoid in the tumour tissue could be clearly demonstrated and histologically the tumours could be clearly differentiated from fibrosarcoma.

The historical group consequently showed a higher incidence of large-sized, proximally located and high-grade osteoblastic tumours than the contemporary group. Such factors are assumed to be indicative of a poorer prognosis. This was investigated in our study on metastases and survival (III) in which patients who had received adjuvant interferon therapy were excluded from the contemporary group. A comparison between the two new groups showed that the differences with respect to prognostic factors persisted.

A clear difference could be noted both in the incidence of metastases and in the survival rate after 2.5 years' follow-up. At this point of time the incidence of patients without metastases was twice as high in the contemporary group and the same relation was observed with respect to the number of survivors. In an attempt to explain these differences we also studied the methods of treatment adopted in the two groups.

Treatment

Radiation therapy. For decades the treatment of primary osteosarcoma has consisted in surgery or irradiation, or a combination of both.[49] The extremely low 5-year survival rate observed in patients treated only by surgery led Cade[13,14] to introduce treatment of primary tumours with preoperative irradiation in high doses of 60–90 Gy, followed by a 6-month period of observation prior to definitive surgery. This was intended to avoid surgery for those patients who developed metastases during the 6-month interval. Micrometastases are assumed to be present in a majority of patients – according to some authors up to 80 % – at the time of diagnosis. Studies of large series have shown that lung metastases can be radiologically verified in more than half of the patients within 6 months of diagnosis.[15,36,86,115,136]

Osteosarcoma is not considered to be particularly sensitive to radiation[63] and treatment of the primary tumour by irradiation only, especially with suboptimal doses, often leaves a viable tumour which in due time produces pain, pathological fractures and an impaired general condition.[36,68,76,82,132] The 5-year survival rate for patients treated by irradiation only is in many studies reported as lower than that resulting from surgical treatment only. Some studies report no better results from a combination of surgery and irradiation,[21,36,49,80,83,118,136] but it may be suspected that these findings are biased by the selection of patients not suited for surgical treatment.

With the introduction of adjuvant therapy, which is most effective when there is limited tumour involvement and the primary tumour is removed at an early stage, preoperative radiation therapy has been virtually discarded and the treatment is today primarily used as a palliative procedure for inoperable tumours.[21] Cade claimed that high radiation doses were required to achieve antitumoral effects.[14] In the contemporary control group radiation therapy with doses exceeding 45 Gy was given to twice as many patients as in the historical group. A comparable number of patients in either group was scheduled for a 6-month observation period on conclusion of irradiation before surgical treatment was to be initiated. These patients are included in the study. Two patients in the contemporary group and 5 in the historical group developed metastases during this 6-month interval and had no surgery. This should not influence a prognostic comparison,

since both groups originally included an equal number of patients scheduled for this form of treatment.

Prophylactic irradiation of the lungs to destroy micrometastases has been tested in prospective randomized studies without providing a conclusive answer.[7,12,105] No patient in any of the groups included in the present study has received such adjuvant treatment.

Surgery. Disarticulation was considerably more common among patients in the historical than in the contemporary group. This operation was performed in almost half of the patients in the historical group, partly because of a higher incidence of proximally located tumours and probably partly owing to the prevailing opinion of the day that disarticulation was necessary to ensure radicality. This will scarcely influence a prognostic analysis, however, since macroscopically radical removal of the tumour was carried out in all patients subjected to surgery.

Treatment of lung metastases. Metastasis of osteosarcoma is hematogenous and predominantly involves the lungs.[14,26,61,67,79] In spite of the fact that the average duration of symptoms is less than 6 months it is assumed that the majority of patients[14] – probably up to 80 % – have lung metastases at the time of diagnosis.[8,27,56,64,77] Prior to the 1970's the approach towards treatment of lung metastases was highly conservative and as a rule any treatment given was purely palliative.

The reports claiming successful results with methotrexate or adriamycin therapy for macroscopic metastases in osteosarcoma marked the beginning of a more aggressive approach to the treatment of such metastases. Optimal antitumoral effect was observed when there was limited tumour involvement.[36,48,56,110] Adjuvant therapy has also produced a somewhat changed course of the disease in that fewer and often solitary metastases have been noted to occur and sometimes at a later stage.[4,10,63,98,99,130] As a consequence surgical removal of lung metastases by lobe resection, lobectomy and sometimes total pulmectory seemed more justified and in recent years promising results have been reported of such surgery.[3,12,59,87,111,113,121,124,140]

That a similar change in attitude with respect to the treatment of lung metastases had taken place in Sweden was evident from our study comparing the historical and contemporary groups in this respect (III). In almost half of the patients in the contemporary group who developed lung metastases these were removed by surgery, while 25 % of these patients received high-dose chemotherapy. No patient in the historical group had pulmonary surgery, while high-dose chemotherapy was not available at the time. The difference in the two groups with respect to the treatment of lung metastases to some extent serves to explain the difference in the survival rates noted at the 2.5 year follow-up. Whether active treatment of lung metastases delays the plateau phase in the survival curve cannot be definitely concluded from our study after 2.5 years' follow-up. According to earlier studies lung metastases were noted in 80 % of the patients within one year and since all patients in our study had been followed up for at least 2.5 years, this was the time selected for comparison of the two groups. A combination of adjuvant therapy and active treatment of lung metastases might be expected to produce a further delay of the plateau phase in the curves expressing metastasis-free and survival rates, and thus urge the need for an increased follow-up period.

Adjuvant therapy. Dissemination of the tumour prior to diagnosis and initial treatment is considered of crucial importance for prognosis and various forms of treatment to

counteract this microscopic tumour spread have been suggested.[5,48] During the 1970's various cytostatics have been introduced as adjuvant therapy.[15] Following initial reports of highly successful results, various centres throughout the world have subsequently reported results based on larger series and longer follow-up periods providing an average survival rate of about 50 %, but the results of different centres vary widely.[15] Treatment with a combination of different cytostatics – primarily adriamycin and methotrexate – in doses frequently approaching the maximum dose has been associated with severe side-effects. Even lethal effects have been reported.[88]

Attempts have also been made to treat osteosarcoma patients with anticoagulants, transfer factor and various other regimens of immunological therapy.[11,22,33,51,58,112,114] As a rule such treatment is recommended only as a supplement to radical surgery and adjuvant chemotherapy. In many cases adjuvant therapy of different kinds is founded on in vitro and animal experiments,[16,47,48,126,127] and some authors have succinctly described the state of adjuvant therapy in osteosarcoma today as presenting "more questions than answers".[90]

To summarize, our comparison of prognostic factors in a historical and a contemporary group in several studies (II–IV) has demonstrated a number of differences between the two groups (Table II).

Table II. Difference in prognostic factors between the historical and the contemporary group.

Prognostic factors	Historical versus contemporary group
Symptoms	Higher incidence
Tumour size	Larger
Tumour pathology	Higher incidence of osteoblastic and Grade IV tumours
Radiation therapy	Lower incidence
Surgery	Higher incidence of disarticulation
Treatment of pulmonary metastases	Less active (no surgery or high-dose chemotherapy)

The historical group features a higher incidence of unfavourable prognostic factors such as a proximal tumour location, larger and more malignant tumours. In addition a more aggressive approach to the treatment of metastases presumably resulting in increased survival was noted in the contemporary group. Up-to-date curves for the two groups studied in our analysis of metastases and survival (III) are presented in Fig. 1 and 2, which also show the results for the 42 interferon patients to date. It remains to be seen whether the observed differences between the two groups with respect to prognostic factors persist when the series are enlarged. There is an obvious risk, however, in evaluating the effect of adjuvant therapy by comparison of small clinical series with a short follow-up. An analysis of prognostic factors in the series to be compared is imperative especially in non-randomized studies.

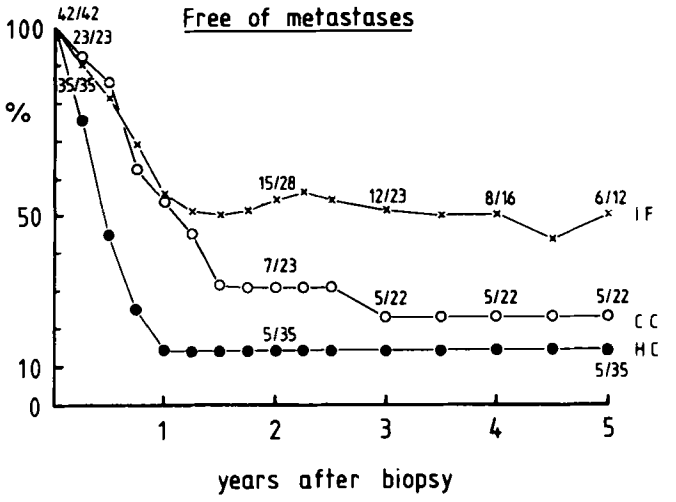

Fig. 1. Up-to-date metastasis-free rate in interferon-treated patients (x—x, N=42), contemporary control group (○—○, N=23) and historical control group (●—●, N=35).

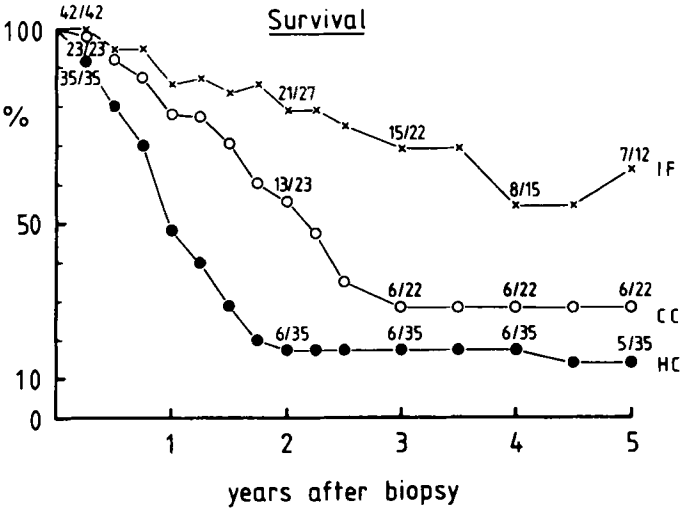

Fig. 2. Up-to-date survival rate in interferon-treated patients (x—x, N=42), contemporary control group (○—○, N=23) and historical control group (●—●), N=35).

Endocrinology

Growth

Osteosarcoma is most common in young individuals and its incidence is almost twice as high among males, with a peak usually coinciding with the pubertal growth spurt.[27, 74, 81, 100, 119, 147] It has been suggested that both pathogenesis and prognosis are influenced by hormonal factors.[53] Female patients are claimed to have a better prognosis[45, 80, 102, 106, 138] and in vitro studies have demonstrated estrogen to have a positive antitumoral effect.[120] The predominant site of the tumour is the metaphyseal region of the long bones and in the majority of patients the tumour is located in the region of the knee.[27, 36]

Earlier studies have reported a predominance of tall individuals,[21, 46, 74, 100, 120] but no analysis has been performed to determine whether osteosarcoma is associated with an abnormal rate of growth. Such an analysis has been attempted in our study of individual growth curves of osteosarcoma patients (V). In contrast to the earlier studies, which were based on cross-sectional growth data instead of individual growth curves, our analysis failed to demonstrate any deviations in longitudinal growth. A relationship exists between longitudinal growth and tooth eruption.[41] In our analysis dental maturity was found to be normal and consequently supported our findings regarding normal growth in osteosarcoma patients. The average duration of symptoms, reported as 3–10 months in earlier studies,[49, 81, 136] was 3 months for the patients in this study. The possibility of the tumour itself stimulating growth could therefore not be excluded, since the period from the presumed onset of the disease until diagnosis was too short to allow detection of growth rate increment. The sex distribution in this study was not wholly typical for osteosarcoma patients. The proportion of female patients was larger than might be expected, but since comparisons were made within each sex this could not influence the results.

Additional evidence to support our finding of normal growth in osteosarcoma patients was provided by a subsequent study (VI), which showed serum levels of somatomedin A and growth hormone to be within the normal range for age for these patients. Earlier studies reporting increased levels of these hormones may have failed to take sufficient account of variations in age, especially with respect to somatomedin levels.

Glucose tolerance

A high incidence of diabetes mellitus has been reported for tumour patients.[70] Glucose intolerance may be ascribed to stress and malnutrition factors in cancer patients in poor general condition.[31, 149] Glucose homeostasis may also be influenced by polypeptide hormone secretion by the tumour tissue.[108] Earlier studies have reported glucose intolerance in osteosarcoma patients in combination with hyperinsulemia.[53] In our study (VI) glucose tolerance was analyzed in 15 osteosarcoma patients. This study provided evidence of glucose intolerance, consistent with earlier reports, and showed that this could most likely be ascribed to decreased peripheral insulin sensitivity. At present we can only speculate whether this decreased sensitivity should be attributed to hormones or hormone homologues secreted by the tumour tissue. In order to analyze this, glucose infusion tests were performed in several patients after removal of the tumour and no change in glucose tolerance or insulin sensitivity was noted in these cases (Unpublished observations).

There was considerable individual variation in insulin sensitivity, but all patients manifesting extremely low insulin sensitivity developed metastases. The follow-up period

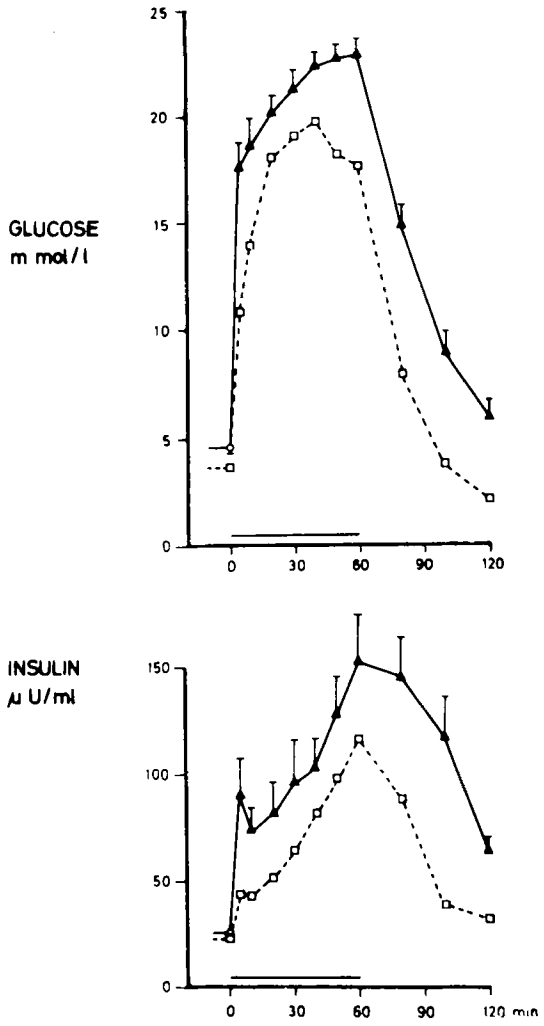

Fig. 3. Glucose and insulin levels during GIT in single patient free of metastases after 3.5 years (□—□) and 8 patients with metastases within one year (▲—▲; vertical bars indicate SEM).

varies for the patients included in this consecutive series, but the patient without metastases after the longest follow-up (3.5 years) belonged to the group manifesting normal insulin sensitivity. Insulin and glucose response to the glucose infusion test for this patient, as compared to the 8 patients who developed metastases within one year (Fig. 3), showed a tendency to lower serum glucose levels during the test and a more rapid decline of the serum glucose level on conclusion of the test. Both the insulin level and the calculated "K-value" were within the normal range for this patient, whereas "K-values" were significantly decreased for the patient group as a whole.

Whether insulin sensitivity may be useful as a prognostic factor in osteosarcoma will be investigated in further studies based on a larger number of patients with a sufficiently long follow-up to permit more reliable prediction of the long-term outcome.

CHAPTER 4 – GENERAL SUMMARY AND CONCLUSIONS

The present study permits the following observations and conclusions:

1. Open biopsy is advisable in the diagnosis of osteosarcoma in view of the many advantages offered by this procedure and the fact that no negative effect on long-term survival was demonstrated. On the other hand, too long a delay between biopsy and definitive surgery would not seem to be advisable.

2. Comparison between a historical and a contemporary group demonstrated a predominance of unfavourable prognostic factors in the historical group. This group was characterized by a higher incidence of symptoms, larger and histologically more malignant tumours, and a higher incidence of proximally located tumours.

3. A contemporary and a historical group without adjuvant tumour treatment manifested clear differences with respect to the incidence of metastases and the survival rate after 2.5 years' follow-up. The percentage of survivors and patients without metastases was twice as high in the contemporary group at this time. The increased survival rate for patients with lung metastases in the contemporary group could partly be ascribed to the more aggressive approach adopted in the treatment of such metastases.

4. Evaluation of the effect of any therapy in non-randomized studies requires an analysis of prognostic factors to disclose possible differences between the clinical series. There would seem to be a definite risk in using historical controls in such studies based on small series with a short period of observation.

5. The blood variables studied in osteosarcoma patients provided values within the normal range, with the exception of the erythrocyte sedimentation rate.

6. Correlation analysis disclosed that female patients had a higher incidence of low-grade tumours and that a distal tumour site was more common in tumours of smaller size.

7. Normal longitudinal growth and normal dental development were found in osteosarcoma patients. This finding is supported by the results of our study demonstrating normal serum levels of somatomedin A and growth hormone.

8. Decreased glucose tolerance was demonstrated in our osteosarcoma patients and could most likely be attributed to decreased insulin sensitivity.

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