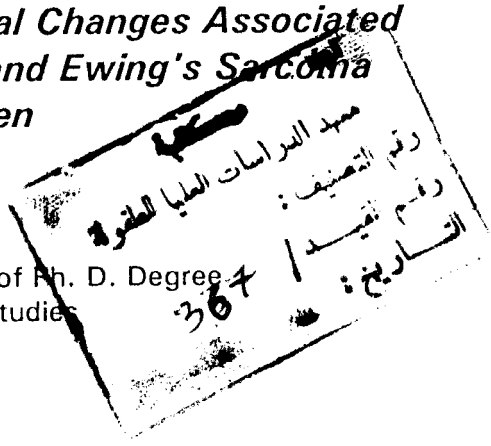
Psychological and Biochemical Changes Associated with Osteogenic Sarcoma and Ewing's Sarcoma in Children



THESIS

Submitted for fulfillment of Ph. D. Degree
in Childhood Studies

BY

Mie Afify Mohammed Khalil
M.B, B.CH., M.SC. (Pediatrics)
Research Assistant, National Research Center

Supervised BY

Prof. Dr. Zeienb Bishry
Prof. of Neurology & Psychiatry
Faculty of Medicine,
Ain Shams University

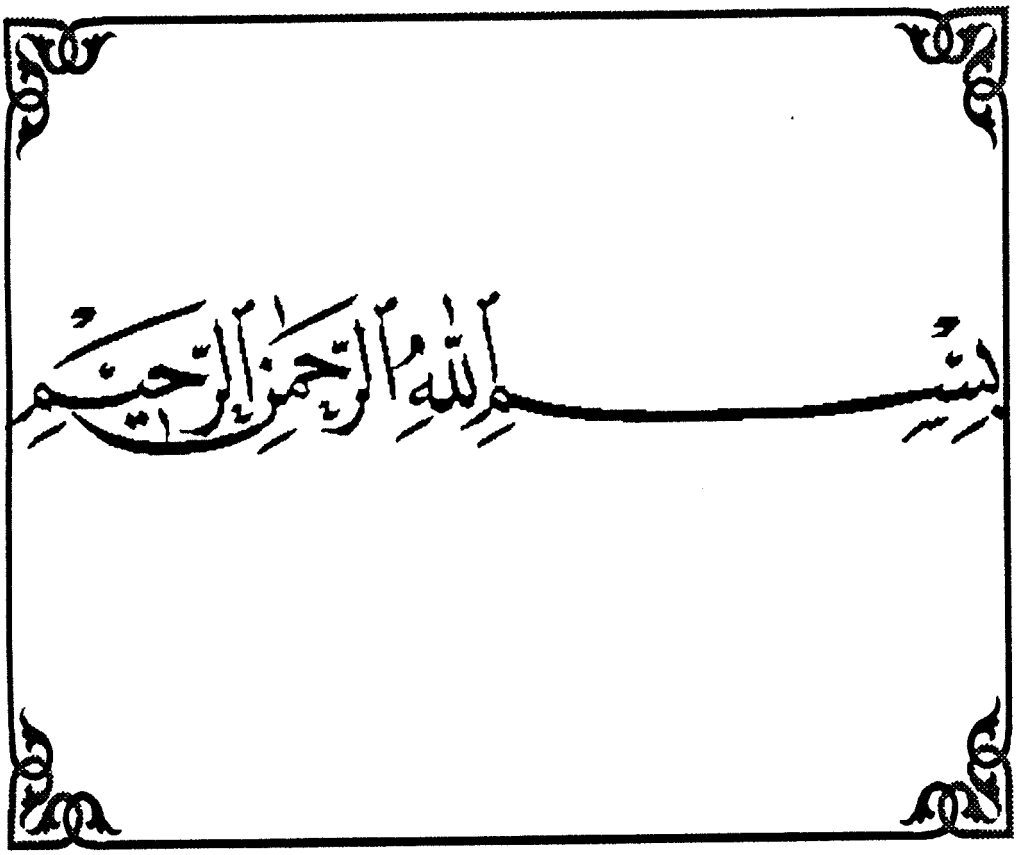
Prof. Dr. Yehia Shaker
Prof. of Clinical Biochemistry
Head of Biochemistry Department
National Research Center

Dr. Mohammed Hany M. Hussein
Assist. Prof. of Pediatric Oncology
National Cancer Institute

Dr. Khaled H. Taman
Lecturer of Pediatrics
Institute of Postgraduate
Childhood Studies
Ain Shams University

Institute of Postgraduate Childhood Studies
Medical Department
Ain Shams University

1996


بسم الله الرحمن الرحيم

TO
MY FAMILY

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First thanks god for blessing this work until it has reached its end as a little part of his generous help throughout our life.

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الكلية: معهد الدراسات العليا للطفولة

شكر

اشكر السادة الأساتذة الذين قاموا بالاشراف

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٣) د. محمد صابر حسين / استاذ مساعد بمعهد الدراسات العليا للطفولة
٤) د. جمال الدين حسين طه / مدرس الطفولة / معهد الدراسات العليا للطفولة

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٢) د. آلفت محمد عزبي / باحث بالمركز القومي للبحوث
٣) .....

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١) معهد الدراسات العليا للطفولة
٢) المركز القومي للبحوث
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اتقدم بالشكر للسادة الأساتذة الذين قاموا بالإشراف

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٢) أ.د. أمين مختار / أستاذ الكيمياء الحيوية / المركز القومي للبحوث

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٢) د. آمال محمد موسى / باحث بالمركز القومي للبحوث

٣) .....

وكذلك الهيئات الآتية :

١) معهد الدراسات العليا للطفولة

٢) المركز القومي للبحوث

٣) .....

Abstract

Psychological and biochemical changes in osteogenic sarcoma and Ewing's sarcoma among children

The study was performed on 21 patients with Ewing's sarcoma (age range 8-16 years), and 35 patients with osteogenic sarcoma(25 children of age range 8-12 years and 10 adolescents of age range 13-20 years), besides 30 healthy persons (15 children with age range 8-12 years, and 15 adolescents with age range 13-20 years) to serve as control.

The psychological assessment of all patients (Ewing's sarcoma and osteogenic sarcoma) and the control groups was done with the standard psychiatric sheet history, anxiety scales sheet and detection of personality of the patients through the E.P.Q.

The serum T.LDH and T.ALP activities were kinetically determined according to the Scandinavian recommended method at 37°C, the LDH iso-enzymes were separated by agarose gel electrophoresis and quantitated by scanning at 570 nm. The ALP iso-enzymes were identified and quantitated by three methods, a) separation on PAGE b) heat inactivation and c) chemical inhibition by L-phenylalanine.

The results revealed a significant increase in the frequency of anxiety symptoms in all patients, and the nervousness, eating troubles, bed wetting, night mares and terrors and thumb suckling were the most common neurotic traits affecting the all patients in order of frequency.

The results revealed, the neuroticism was significant high in both boys and girls. As for extroversion-introversion personality trait in patients with osteogenic sarcoma were significantly high in both boys and girls, and in patients with Ewing's sarcoma it was significant higher in girls than boys. As for the psychcitism trait was low in both boys and girls in all patients. As for lie scores were significant higher in girls than boys in all patients.

The frequency of different psychiatric disorder in all patients revealed that, there were more than one diagnosis could be present at the same time in one patient. The behavioral and emotional disorders were the most prevalent disorders.

The results revealed a significant increment in the serum total LDH levels before therapy as compared to healthy control group. Such increment was mainly due to the increased activities of the iso-enzymes LD_2, LD_3 and LD_4. During the course of therapy, the elevated serum LDH levels decreased gradually in response to therapy, except during radiotherapy when the serum levels increased. These increment were mainly due to the increased activities of the isoenzymes LD_2, LD_3 , LD_4 , and LD_5.

The serum LDH levels reached normal values in 12 patients (57%) who continued treatment and did not develop metastasis. However, there were 4 patients (19%) with increased serum levels despite treatment before the clinical diagnosis of metastasis, and proved to be metastatic later by other

investigations. So, the increase in the LDH anaerobic isoenzyme activities could be used to monitor any changes in the tumor activity before other clear-cut physical signs are manifested.

The results revealed a significant increment in the serum ALP levels in patients with osteogenic sarcoma before therapy as compared to healthy control group. Such increment was mainly due to the increased activity of the bone ALP isoenzyme.

During the course of therapy including the surgery, the elevated serum ALP levels decreased gradually in response to therapy in 24 patients (68.5%) who continued treatment and did not develop metastasis (16 children 64% and 8 adolescents 80%). The serum ALP level increased in 6 patients (17%) (5 children 20% and 1 adolescent 10%) despite of therapy and before the clinical diagnosis of metastasis and proved to be metastatic later by another investigations. So, the increase in the activity of bone ALP iso-enzyme could be used to monitor any changes in the tumor activity before other clear-cut physical sign are manifested.

Key Words

Osteosarcoma, Ewing's sarcoma, Psychological disturbance, Serum LDH, Serum ALP, Iso-enzymes.

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List of abbreviations

%	percentage
1ry	Primary
AJC	American Joint Committee
ALP	Alkaline phosphatase
ANC	Absolute leucocytic count
B.ALP	Bone Alkaline phosphatase
BCG	Bacillus Calmette-Guerin
BMA	Bone marrow aspiration
Ca	Control adolescent
CBC	Complete blood count
Cc	Control children
cm	Centimeter
CT	Computed tomography
d	Decrease
DSA	Digital subtraction Angiography
ECG	Electro cardiography
EDAT	Ethylen-diamine-tetra-acetic acid
EM	Electron microscope
ESR	Erythrocytic sedimentation rate
Fig.	Figure
GA	Pre-treatment stage
GB	Pre radiotherapy or pre-operative
GC	During radiotherapy or post operative
GD	During chemotherapy
GE	At the end of chemotherapy
GF	At follow up after 6 months of cure
GGT	Gamma glutamyltransferase
gm%	Gram percent
GPO	German Society for pediatric Oncology
H	Heart
h.s.	Highly significant
HBD	Alpha-hydroxybutyrate dehydrogenase
HLA	Human lymphocytic antigen
i.	Increase
I.ALP	Intestinal Alkaline phosphatase
IESS-1	First Intergroup Ewing's sarcoma Study

IESS-II	Second Intergroup Ewing's sarcoma Study
L	Lactate
L.ALP	Liver- Alkaline phosphatase
LD	Lactate dehydrogenase
LDH	Lactate dehydrogenase
LFT	Liver function test
M.	Muscle
m.s.	Moderate significant
MDP	Muramyl dipeptide
$mglm^2$	Mil gram per square meter surface area
MIOS	Multi-Istitutional Osteogenic sarcoma Study
MRI	Magnetic resonance imaging
MDP	Muramyl dipeptide
MTP-PE	Muramyl tripeptide-phosphatidylethanolamine
MTS	Musculoskletal Tumor Society
N.	Normal range
N.s.	Non significant
NCI	National Cancer Institute
NSE	Neuron specific enolase
P	Probability
p.	Pyruvate
P.A	Postro-anterior
P.ALP	Placental Alkaline phosphatase
PAS	Periodic acid schiff
Pic.	Picture
PNET	Peripheral neuroectodermal tumor
PSYCOG	Psychological collaborative oncology group
PTSD	Post traumatic stress disorder
RFT	Renal function test
S.D	Standard Deviation
S.E	Standard Error
s.s.	Slightly significant
Ser. No.	Serial number
SGOT	Serum glutamyl oxalacetate transaminase
SGPT	Serum glutamyl pyruvate transaminase
SR	Sedimentation rate
SSS	Surgical Staging System
t.d.s	Three times daily

TLC	Total leucocytic count
TNM	Tumor, Node involvement and metastasis
U/L	Unit per liter
v.h.s.	Very highly significant
VAC	Vincristin, Actynomycin-D and Cyclophosphamide
VACA	Vincristin, Actynomycin-D, Cyclophosphamide and Adriamycin
VS	Versus
WBC	White blood cells
yr.	Years

INTRODUCTION AND AIM OF THE WORK

Introduction and aim of the work

Introduction

Bone tumors are relatively rare. Their incidence varies according to age. While the frequency of osteogenic sarcoma and Ewing's sarcoma is higher in adolescents than adults. The most frequent sarcomas of the bone are osteogenic sarcoma (45%), and Ewing's sarcoma (18%), both tumors of young children and adolescents (Dahlin, 1987).

Osteogenic sarcoma is the most common primary malignant tumor of the bone. Histologically, it is composed of malignant spindle cells and osteoblasts that produce osteoid or immature bone (Acchiapati et al., 1965). While Ewing's sarcoma is a primitive malignant tumor of the bone characterized by uniform, densely packed small cells with rounded nuclei but without distinct cytoplasmic border or prominent nucleoli (Enneking et al., 1980).

Research in psychosocial and behavioral aspect of cancer has shown steady growth since the 1950's, and its course of development has paralleled the history of medical techniques in treating cancer (Holland, 1991).

The diagnosis of cancer creates crisis that requires the patient to adapt quickly to news of catastrophe, the patients try to control the level of emotional distress while taking crucial treatment decisions, major concerns are fear of dependency, disfigurement, disability, and abandonance and disruption in relationships. Also the diagnosis of cancer has an immediate and lasting impact on the family, it presents a major developmental challenge to the family, as well as the child (Hersh and Wiener, 1993).

During the 1980's, there has been an explosion of clinical knowledge and experience in the management of bony neoplasm, the development of centers with specific interest in these tumors have played an important role in the understanding and surgical management of these lesions and in the development of multimodality treatment regimens, which leads to dramatic improvement in the survival rates (Simon et al.,1986). In the light of this medical success, the focus of pediatric oncology now includes a heightened awareness of the multiple psychiatric and psychological implications of cancer in childhood. The child's adaptation during disease and treatment and the quality of life as a long term survivor are of paramount concern (Lansky et al.,1989).

Now, the field of biological markers of malignant bone tumors has been the focus of increased interest for their applicability in the diagnosis, response to therapy and prognosis. The association of elevated bone alkaline phosphatase in the serum of patients with osteosarcoma was the first evidence that the tumor cells themselves produce the enzyme, the amount of phosphatase present gives valuable clinical information as the degree of tumor destruction produced by the therapy, changes in the tumor activity may frequently be detected by this means before other clear-cut physical signs are manifested. The reduction of elevated serum alkaline phosphatase levels following chemotherapy is a valuable guide to administration of therapy (Rosen et al.,1974).

Previous reports firmly established that serum phosphatase values are helpful in determining prognosis, post-operative, or post-chemotherapy follow up examination should include alkaline phosphatase determination, since increasing values herald the presence of a residual, reactivated and spreading osteogenic sarcoma (Aparisi et al.,1982).

Serum alkaline phosphatase is not a single enzyme, but consisted of a family of isoenzymes originating from liver, bone, intestine, and placenta (Fishman,1987). The presence of Regan isoenzyme (carcinoplacental, heat stable fraction) in the serum of some patients with osteogenic sarcoma has been proposed as a prognostic factor indicating the metastatic evolution of the disease, hence, isoenzymes could be used as a marker of relapse in an early subclinical stage (Tucci et al.,1990).

Lactate dehydrogenase is a glycolytic enzyme which catalyzes the reversible alternation of pyruvate to lactate, present nearly in all metabolizing cells. Damage to nearly any tissue can cause elevated serum lactate dehydrogenase. Therefore, the test is rather non specific indicator for tissue destruction. However, in conjugation with its isoenzyme patterns, it can give more specificity of the damaged cells. In recent years the relationship between neoplasm and lactate dehydrogenase has been studied with increasing intensity, rapidly dividing tumors convert to an anaerobic metabolism, so the measurement of lactate dehydrogenase is an indicator of an anaerobic metabolism which occur in aggressive tumors (Bacci et al.,1985).

Serum lactate dehydrogenase levels have been recently found to be useful as prognostic markers in several neoplastic diseases, previous reports proved that serum lactate dehydrogenase could be used as a tumor marker in Ewing's sarcoma (Glaubiger et al.,1980). In 1981, Rosen et al., founded that the initial serum lactate dehydrogenase values correlated with the bulking of the tumor.

Aim of the work

The aim of this study is to :

1- Identify the behavioral and psychiatric disturbances in the children with osteogenic sarcoma or Ewing's sarcoma.

2- Estimation of serum levels of alkaline phosphatase, lactate dehydrogenase and their isoenzymes in patients with osteogenic sarcoma or Ewing's sarcoma respectively, aiming that these enzymes might help in; evaluation if the serum level of these enzymes and isoenzymes, have a prognostic value in those malignant bone tumors, can be used to monitor the response of those patients to therapy, or can be used as a signal of tumor recurrence.

REVIEW OF LITERATURE

Bone Tumors

Malignant tumors arising from the skeletal system are rare, representing only 0.2% of all primary cancers (Silverberg,1987). The incidence of bone tumors is highest during adolescence, with a rate of 3 per 100,000. In spite of the high incidence at this age, bone tumors comprise only 3.2 % of childhood malignancies that occur before the age of 15 years. Osteosarcoma and Ewing's sarcoma are the two most common bone tumors, they occur mainly during childhood and adolescence (Dahlin,1978).

Other mesenchymal (spindle cell) neoplasm (Fibrosarcoma, Chondrosarcoma, and malignant fibrous histocytoma), that characteristically arise after skeletal maturity are less common. There are sometimes associated with underlying benign bony tumors, previous radiation or primary bone disease (Mirra,1989).

Etiology

The observation of high incidence in children supports the assumption that skeletal neoplasm arise in areas of rapid growth (Dahlin,1978). In addition the most common location of primary bone sarcomas is metaphyseal, near the growth plate. This is the region in the bone with the most intense cellular proliferation and remodeling activity during long bone growth. The highest incidence is in the distal femur and proximal tibia, the two areas with the most active growth plates (Malawer,1989).

Prolonged growth or over stimulated metabolism may blend imperceptibly with neoplasm. This may be seen in neoplasm arising from adult tissues affected by metabolic stimulation from long standing Paget's disease (Giant cell tumors or osteosarcoma), Hyperparathyroidism (Brown tumors), chronic osteomyelitis

(Squamous cell carcinoma and osteosarcoma) and fracture callus (Jaffe,1974).

Radiation has been linked to the formation of osteogenic sarcoma, Chondrosarcoma and Fibrosarcoma as a result of both radiation treatment and internal bone -seeking radioisotope from occupational and medicinal use (Miller,1981).

The role of infectious agents, particularly in osteogenic sarcoma has been suggested based on laboratory observations, i.e. induction of Osteosarcoma in mice by extract from human osteosarcoma (Finkel et al.,1970). A consistent cytogenic marker in Ewing's sarcoma of the bone is an 11 : 22 translocation (Aurias et al.,1983).

Detection and Diagnosis

Clinical evaluation

Patients generally present with pain in the area of the lesion and commonly may be worse at night. In more advancer lesions patients may note a mass or swelling (generally only if there is significant periosteal reaction or the tumor has eroded through the bony cortex), and may occasionally present with a pathological fracture. If the lesion is near a joint, a sympathetic effusion or stiffness of the joint also may occur. Systemic or constitutional symptoms such as weigh loss, fever, malaise, or night sweats are quite uncommon with most bone sarcoma, but occur more commonly with Ewing's sarcoma or in cases with multiple metastases at the time of presentation (Mirra,1989).

The history is extremely important for a number of reasons. A past history of carcinoma may suggest a metastatic etiology of a new bone lesion, while previous radiation treatment to an area may

suggest of post radiotherapy sarcoma. In patients with known cartilaginous lesions, e.g. Enchondroma or Osteochondroma, occurrence of further growth in adulthood, or pain may be early indication of malignant transformation (Mirra,1989).

Diagnostic procedures

Roentgenograms are the single important diagnostic tool for diagnosis and prognosis of bone tumors. A number of roentgenographic parameters are considered in evaluation of a bone lesion (Malawer et al.,1989).

Computed tomographic CT scans are extremely helpful in establishing the extent of the tumor within the bone, and in determining the presence of any extraosseus soft tissue mass. The CT also help to delineate the three dimensional anatomy of the lesion, this can be essential to planning either surgical biopsy or definitive treatment. One of the most important strengths of CT scanning is the ability to assess even subtle degrees of cortical destruction quite accurately. Additionally, CT with contrast delineates very accurately the anatomic relationship between pelvic tumors and the bowel and ureters (Mirra,1989).

Magnetic resonance imaging MRI, is an alternative to CT scanning, MRI shows with highly accurate detail the relationship between normal tissues and neurovascular structures and the tumor tissue. This is essential in planning surgical biopsy or treatment. In addition, extent of the reactive zone of the tumor within the bone is also visualized with high sensitivity, as even edema of the marrow adjacent to tumor tissue is apparent (Berquist,1989).

Radionuclide imaging using Technetium 99 (bone scan) has become an essential part of bone tumors evaluation, although bone

scans lack specificity for neoplasms, the studies are extremely sensitive, and can detect tumor foci in the bone not visualized on standard radiographs. Thus, the extent of the tumor within a bone, including the reactive zone and the presence of skip metastases, can be demonstrated accurately (Bragg, et al.,1985).

Angiography is no longer used as commonly in staging work-up due to increasing use of CT with contrast or MRI, however, in pelvic lesions, angiography is quite useful in defining the locations of the major vessels, and in neoplasms with excessive vascularity can facilitate embolization prior to biopsy or definitive surgical treatment to decrease intra-operative blood loss (Bragg et al.,1985). Table (1) showed the imaging modalities for the evaluation of bone cancer.

Table (I) Imaging modalities for evaluation of bone cancer (Mirra., 1989)

Primary tumors and regional lymph nodes:

Method	Diagnosis and staging capability	Recommended for use
CT	Useful to stage tumor locally for medullary and soft extension as well as presence of matrix or cortical disruption.	Yes; essential in planning limb conservation surgery.
MRI	Applications at present are speculative. Main use with soft tissue rather than bone tumors.	No
Conventional roentgenograms	High sensitivity and specificity for primary bone tumors; lower sensitivity and specificity for metastatic lesions in bone.	Yes; metastases tend to involve medullary canal and must be >1cm to be detected.
Angiography	Seldom useful except to serve as a vascular road map for surgery.	No (yes, if considering limb salvage and clear margin from tumor not demonstrated by CT); essential if embolic or infusion therapy contemplated.
Metastatic evaluation:		
Chest roentgenograms	Essential for all tumor types	Yes
Film tomography	Useful to characterize questionable plain film finding	No
CT of lungs	Use in high-risk patient group where treatment decisions hinge on presence or absence of lung metas.	Yes
Radionuclide bone scan	Essential to determine if bone lesion is monostotic or polyostotic.	Yes

CT= Computed Tomography; MRI = Magnetic Resonance Imaging

Laboratory studies have limited usefulness in bone sarcoma evaluation, Alkaline phosphatase concentration may be elevated, particularly in osteosarcoma, and is extremely high levels in secondary osteosarcoma of Paget's disease, multiple myeloma can be diagnosed by serum protein electrophoresis if a significant level of abnormal immunoglobulin is present. The white blood cells, differential, and sedimentation rate may be useful to exclude Osteomyelitis, which can frequently mimic a primary bone tumor. Hyperglycemia may suggest myeloma or disseminated metastatic disease and can also present in Brown tumors of hyperparathyroidism, although with this lesion there is associated hypophoshatemia as well (Mirra,1989).

Bone biopsy is essential in the diagnosis of any primary bone neoplasm, the biopsy should not be performed until staging studies such as CT, MRI and bone scan evaluations of the local extent of the lesion have been completed (Enneking,1983). Because of the reaction of normal bone and periosteal elements to the lesion, sampling error is a potential problem and can make diagnosis difficult, for this reason incisional biopsy is preferred by many, although needle biopsies are also effective and can achieve diagnostic accuracy of 70% to 90 % at institutions with considerable experience with this technique (Dollahite et al.,1989).

Classification and Staging

A primary bone cancer is any neoplasm that arises from the tissues or cells present within bone, and has the capability of producing metastases, bone sarcomas are named in relation to the predominant differentiated tissue type, although obviously multiple cellular elements may be present in any tumor.

Bone forming tumors are referred to as Osteosarcoma, cartilage tumors as Chondrosarcoma, fibroblastic tumors as Fibrosarcoma, fat forming tumors as Liposarcoma, and tumors derived from vascular elements as Angiosarcoma (Dahlin,1978), as shown in table (II)

Table (II) General classification of bone tumors (Dahlin,1978).

Histologic Type	Benign	Malignant
Hematopoietic (41.4%)		Myeloma Reticulum cell sarcoma
Chondrogenic (20.9%)	Osteochondroma Chondroma Chondroblastoma Chondromyxoid fibroma	Primary Chondrosarcoma secondary Chondrosarcoma Different Chondrosarcoma Mesenchymal Chondrosarcoma
Osteogenic (919.3%)	Osteoid osteoma Benign osteoblastoma	Osteosarcoma Parosteol osteosarcoma
Unknown origin (9.8%)	Giant cell tumor	Ewing's tumor Malignant giant cell tumor Adamantinoma
Fibrogenic (3.8%)	Fibrous histiocytoma Fibroma Desmoplastic fibroma	Fibrous histiocytoma Fibrosarcoma
Notochordal (3.1%)		Chordoma
Vascular (1.6%)	Hemangioma	Hemangioendothelioma Hemangiopericytoma
Lipogenic (<0.5%)	Lipoma	
Neurogenic (<0.5%)	Neurilemmoma	

Anatomical staging system using the tumor, node, metastases (TNM) parameters has not been formalized by the American Joint Committee on cancer staging and end result reporting (AJC) as yet, although some TNM staging guidelines have been suggested (Enneking,1986).The Musculoskletal Tumor Society (MTS) adopted a Surgical Staging System (SSS) for bone sarcoma which is based on the concept of compartmental localization of the tumor, thus anatomical T parameter is similar in both staging systems. T1 is within the bone (intracompartmental), and T2 indicate extension beyond the cortex of the bone (extracompartmental). Spread of the bone tumor to lymph nodes is rare, the usual pattern is hematogenous spread to pulmonary and other sites, in addition distant metastases carry a similar poor prognosis, whether the metastases is to the regional lymph node or to the lung, because of this, the MTS staging system does not differentiate N1 or M1 and nodal or other metastases place the patient in stage III. Finally tumor grade is limited to G1 (low grade) and G2 (high grade), as opposed to G1 to G4 in the AJC system, due to frequent disagreement among pathologists on specific grading of tumors, particularly with the intermediate-grade lesion (Presant et al.,1986).

Principle of treatment

Non malignant tumors are best removed surgically by the most direct route possible, large defects in the bone are filled with a suitable bone grafts, and occasionally protective internal fixation may be needed to prevent fracture, intralesional resection (generally curettage) or marginal resection is generally adequate for treatment of these lesions, marginal resection is preferred to curettage for more aggressive benign tumors such as Chondromyxoid fibroma, Giant cell tumor and Aneurysmal bone cyst (Mirra, 1989).

Radiation treatment to benign tumors may eradicate the lesion but radiation doses to the bone and cartilage have a high potential for late malignant changes, both in the lesional tissue and in the normal surrounding bone, for this reason, radiotherapy to benign lesions in the bone is limited to surgically inaccessible tumors or lesions that are responsive to very low doses of radiation, such as Aneurysmal bone cyst and Eosinophilic granuloma, (McCullough,1980). Malignant tumors are managed by a combination of surgery, radiotherapy and chemotherapy (Sim,1987), although this is strongly dependent on the tumor type, lower grade sarcomas (MTS: stage1) are almost always treated with definitive surgery alone such as, secondary Chondrosarcomas arising in Enchondromas and Osteochondromas, central high grade Osteosarcoma is usually treated with surgery and adjuvant chemotherapy, while Ewing's sarcoma may be treated with irradiation and chemotherapy with surgery as the adjuvant treatment (Sailer et al.,1988).

OSTEOGENIC SARCOMA

Definition

Osteogenic sarcoma is a primary malignant tumor of the bone derived from bone-forming mesenchyme and characterized by production of malignant osteoid tissue or immature bone by the malignant proliferating spindle cell stroma (Huvos,1979) . Low grad Osteosarcoma are rarely encountered in the pediatric and adolescent age group (Ritts,1987).

Epidemiology

Excluding plasma cell myeloma, osteogenic sarcoma is the most frequent primary malignant bone tumor. It is approximately twice as common as Chondrosarcoma, three times more frequent than Ewing's sarcoma, and ten times more prevalent than malignant fibrous Histocytoma of bone (Huvos,1990 and Mokhtar,1991) .

Only half of bone tumors in the childhood are malignant, of these osteogenic sarcoma is the most frequent, accounting for approximately 60% of malignant bone tumors in the first two decades of life. Ewing's sarcoma, the second most frequent primary bone cancer is actually more common than osteosarcoma in children younger than 10 years (Dahlin,1978 and Mokhtar,1991).

Osteogenic sarcoma can occur at any age, although it is chiefly an affection of the young and most of the cases presented during the first two decades of life. Adolescents between 10 and 20 years are by far the most commonly affected (Huvos et al.,1983).

Girls develop osteosarcoma earlier and their age peak is steeper than in boys, this corresponds to the more advanced skeletal age and earlier adolescent growth spurt of females (Dahlin and

Unni,1986) . Males are affected slightly more frequently than females (Huvos,1990), this may be related to the longer period of skeletal growth and additional volume of bone produced in males.

Until the cessation of growth period, the long tubular bones are the bones most frequently involved in osteosarcoma. Approximately 50% of the tumors arise around the knee joint, the distal femur, the proximal tibia and proximal humors are respectively, the most frequently involved sites (Dahlin and Unni,1986 and Huvos,1990). After this period the long and the flat bones are about equally affected (Huvos,1990 and Mokhtar,1991).

Etiology

Although the precise cause of osteosarcoma is unknown, many factors such as aberration of normal process of bone growth, infectious agents, ionizing radiation, hereditary disorders and/or pre-existing osseous diseases have been postulated as predisposing factors to osteosarcoma (Huvos,1979).

Osteosarcoma is a disease of the second decade of life, occurring during adolescent growth spurt suggests a relationship between rapid bone growth and osteosarcoma, this has led to speculation that the tumor arises from aberration of the normal process of bone growth and remodeling (Huvos,1990).

A viral cause has been suggested base upon evidence that bone sarcoma can be induced by viruses in certain animals (Friedlander and Mitchell,1976), and in hamsters by the injection of cell free extracts of human osteosarcoma. Moreover, in immunologic studies, anti sarcoma specific antibodies have been found in patients and in close relatives of patients with sarcoma (Singh et al.,1977). Similarly, lymphocytes cytotoxic to

osteosarcoma cells have been found in the blood of patients with osteosarcoma and their parents (Yu et al.,1977).

Antecedent trauma has often been associated with the development of bone tumors, but there is little evidence to support a causal relationship. Rather, injury (particularly pathologic fracture) often brings the patient to medical attention and radiographs reveal the underlying neoplasm (Huvos,1990).

The only environmental agent known to cause bone sarcomas is ionizing radiation, treatment with alkylating agents may also linked to the subsequent development of bone cancer independent of the administration of radiotherapy, and may potentiate the effect of radiation in the development of secondary osteosarcoma (Tucker et al.,1987).

Osteosarcoma is known to occur at sites of pre-existing skeletal lesions, approximately 2% of patients with Paget's disease of bone develop osteosarcoma. Other lesions predisposed to malignant degeneration include solitary and multiple Osteochondroma, Solitary Enchondroma or Enchondromatosis "Oiller's disease", multiple hereditary exostosis, chronic osteomyelitis, non ossifying Fibroma and sites of bone infarcts (Huvos, 1990) .

Detection and diagnosis

Clinical evaluation

Osteosarcoma typically occurs during childhood and adolescence. In patients older than 40 years, it is usually associated with a preexistent condition, such as Paget's disease, irradiated bones, multiple hereditary exostosis, or polyostotic fibrous dysplasia (Dahlin, 1978).

The most commonly involved sites are the metaphyseal portions of the long tubular bones. Approximately 50% of the tumors arise adjacent to the knee joint, the distal femur, proximal tibia, and proximal humerus are respectively the most frequently involved sites. Flat bones of the axial skeleton, notably the pelvis, are involved in 15% to 20% of cases, but less frequently in the pediatric age group (Huvos,1990). Bones of the hands and feet are involved in less than 1%. Tumor arising outside the bone "extraosseous" are extremely rare (Dahlin,1978).

The majority of patients with osteogenic sarcoma initially complain of pain, with or without an associated soft tissue mass over the affected bone. Systemic symptoms are rare in the absence of widespread metastatic disease (Dahlin and Unni,1986). pathological fracture are especially prevalent in osteolytic osteogenic sarcoma, a distinctive subvariant of which is the telangiectatic type, in which pathological fractures occur in 29% of patients (Huvos et al.,1982).

The duration of symptoms before diagnosis averages 3 months. Patients with more indolent varieties of osteogenic sarcoma "particularly parosteal lesions" are more likely to have a longer history of painful symptoms, often more than a year or two (Dahlin and Unni,1986).

Approximately 10% to 20% of patients with osteogenic sarcoma have overt metastatic disease at diagnosis. In the majority of cases, metastases are found in the lung, although a small fraction present with bone metastases, and pulmonary metastases as well. Metastases to bones of the skeleton occur in 15% to 30% of patients. Less common sites of metastases occurring preterminally include pleura, pericardium, kidney, brain, and adrenal glands.

Involvement of the lymph nodes is unusual, but it is a poor prognostic sign (Huvos,1990).

Laboratory evaluation

The blood count and erythrocyte sedimentation rate are often normal in this disease, and are not helpful in diagnosis.

Ceruloplasmine has also been suggested as a tumor marker but is not commonly used (Link et al.,1988).

Serum lactic dehydrogenase (LDH) level is elevated in approximately 30% of patients who lack evidence of metastases (Link et al.,1988).

Serum alkaline phosphatase level is an important biological marker of tumor activity in patients with osteogenic sarcoma (Levin et al.,1975). Serum alkaline phosphatase determination have definite value in establishing the prognosis, patients in whom serum phosphatase levels remain high after amputation should be strongly suspected of harboring metastases (Huvos,1990).

Radiological studies

The radiographic appearance of osteogenic sarcoma is characterized by the interrelationship of three aspects, namely distruction of the pre-existent cortical or medullary bone "osteolysis", calcification and bone production, and periosteal new bone formation (Kesselring and Penn,1982).

Typical findings are increased intramedullary radiodensity (tumor bone or calcified cartilage), an area of radiolucency (nonossified tumor), a pattern of permeative destruction with poorly defined borders, cortical destruction, periosteal elevation

"codman's triangle" (it is a manifestation of extreme periosteal elevation forming an acute angle with the cortex), and extraosseous extension with soft-tissue ossification This combination of characteristics is not seen in any other lesion (Wilner,1982). The radiographic difference between osteogenic sarcoma and Ewing's sarcoma are seen in table (III).

common Bone Tumors (Huvos, 1991).

Feature	Osteosarcoma	Ewing's sarcoma
Location in bone	Metaphyseal	Diaphyseal
Involvement of long bones	Yes	Yes
Involvement of flat bones	Rare	Yes
Diffuse medullary cavity involvement	Rare	Common (moth-eaten or permeative involvement)
New bone formation	Yes	No-only as secondary phenomenon
Periosteal reaction	Yes (Codman's triangle) or speculation Not prominent but may be present	Yes (Onionskin appearance) Yes

Computed Tomographic (CT) scans are extremely helpful in establishing the extent of the tumor within the bone, and in determining the presence of any extra-osseous soft tissue mass. CT provides an excellent method for localization for biopsy. It is best used for guiding biopsy when the lesion is not well visualized on conventional films. It is also considered as the most sensitive modality available for detection of pulmonary nodules, CT of the chest can be used during follow-up to assess response to

chemotherapy when resection of the lung metastasis is not indicated (Rosenthall,1985).

Magnetic Resonance Imaging (MRI) is especially useful for defining the intramedullary extent, soft tissue spread, the presence of skip lesions in the medulla and also the response to chemotherapy. It also shows with highly accurate details, the relationship between normal tissues and neurovascular structures and tumor tissue. This is essential in planning surgical biopsy or treatment "limb sparing surgery". It has also proven helpful in several difficult situations, such as detecting small lesions, evaluating a positive bone scan when the corresponding plain radiograph is negative (Huvos,1990).

The radionuclide bone scan is indicated in the initial diagnostic evaluation to define the extent of the primary tumor. It is not as accurate as CT scanning or magnetic resonance imaging in indicating the soft tissue spread of an osteogenic sarcoma, but may be useful in defining the degree of medullary extension. Because uptake of the radiopharmaceutical will extend beyond the histologic limit of the tumor, it defines a safe margin to use in planning surgery. The sensitivity of radionuclide bone scanning is also useful in the detection of "skip lesions" which are seen infrequently in patients with osteogenic sarcoma (Kesselring and Penn,1982).

Angiography, the role of selective angiography in the diagnosis and management of osteogenic sarcoma is a positive one as the angiographic demonstration of the irregular and marked vascularity of the tumor provides useful confirmatory evidence of roentgenographic diagnosis. More frequently, the Digital Subtraction Angiography (DSA) will be important in the pre-operative planning and the localization of soft-tissue spread, vascular structures displaced by a tumor are best demonstrated by

DSA, and in many situations where limb-preservation surgery will be done the surgeon will require a " road map" of the area whose blood supply will inevitably be bizarre. Angiographic delineation of the tumor facilitate the performance of limited but radical local en-bloc resection instead of amputation in osteogenic sarcoma (Grainger and Allison,1986).

Xeroradiography. It has received less than wide acclaim in the diagnosis of bone tumors in general and of osteogenic sarcoma in particular. However, this technique demonstrates enhanced details of bone destruction, intratumoral bone production, and periosteal reaction as well as soft-tissue extension by tumor (Gold,1975).

Biopsy

The biopsy of the primary lesion is the most important procedure in the diagnostic evaluation of a suspected malignant bone tumor, although the radiographic findings may be highly suggestive, a biopsy is always required to confirm the diagnosis. The biopsy should be performed by an orthopedic familiar with the management of malignant bone tumors, and preferably by the surgeon who will ultimately perform the definitive surgical procedure (Manikin et al.,1982). Although, fine-needle aspiration and core needle biopsy are used at a number of centers, most patients require open biopsy to ascertain that a generous sample of adequate and representative tissue is obtained (White et al.,1988).

Pathology

The diagnosis of osteosarcoma is based on histopathologic criteria and correlation with a confirmatory radiographic appearance, the histologic diagnosis of osteosarcoma depends on the presence of a frankly malignant sarcomata's stroma associated

with the production of tumor osteoid and bone (Dahlin and Unni,1977).

The largest group of osteosarcoma are the high grad conventional osteosarcoma, the variants of which are seen predominantly in children and adolescents. In conventional osteogenic sarcoma, the connective tissue stroma appears as a mixture of a large, atypical, spindle- shaped cells, that are highly malignant, the stroma may be largely anaplastic and is interspersed with area of malignant osteoid production intimately associated with malignant cell. Three categories of conventional osteosarcoma have been defined by Dahlin, based up on the predominant differentiation of tumor cells, approximately 50% of cases of osteosarcoma characterized by abundant production of osteoid and are classified as "Osteoblastic osteosarcoma " in about 25% of cases the predominant differentiation is toward cartilage "Chondroblastic osteosarcoma", and the remaining cases demonstrate a spindle cell stroma, with a herring bone pattern and minimal amount of osteoid " Fibroblastic osteosarcoma ". The value of this subclassification of conventional osteosarcoma is not well established and no significant differences in behavior or outcome can be determined among these subclasses (Dahlin and Unni,1986).

Telangiectatic osteosarcoma is an unusual variant (constitute approximately 3% of osteosarcoma), that characteristically appears as a purely lytic lesion on plain radiographs, with little calcification, this tumor are grossly cystic, and histiologically they demonstrate dilated spaces filled with blood and necrotic tissue (Dahlin and Unni,1977) .

Parosteal osteosarcoma, represents less than 5% of all osteosarcoma, clinically, this lesion tends to occur in older patients who have a relatively long history of symptoms, the most frequent primary site is the posterior aspect of the distal femur, but other long bones may be affected, radiological, the tumor appears intensely ossified, arises from the cortex (juxtracortical) from a broad base without invading the medullary cavity and appears to encircle the involved bone (Huvos,1979).

Histologically, Parosteal osteosarcoma are low grad lesions and behave indolently, tending to recur locally after incomplete excision but not to spread distantly, the management of these tumor is surgical, since radical excision of the tumor is curative for the majority of patients Parosteal osteosarcoma metastasize slowly and has an overall survival rate 75% to 85% (Huvos,1979).

Periosteal osteosarcoma , also arises on the surface of the bone without invading the medullary cavity (juxtracortical), the lesion occurs frequently in the second decade of life, usually involves the upper tibia mataphysis, appearing as an ill- defined radio lucent lesion on the surface, histologically the tumors are relatively high-grad, predominantly Chondroblastic osteosarcoma, the tumor tend to recur locally unless radical resection is performed and rapidly metastasizing, so many investigators treat them with postsurgical adjuvant chemotherapy (Rosen et al.,1982) .

Low grade Intraosseous osteosarcoma, are well differentiated, with minimal cytological atypia, and can be mistaken for benign conditions particularly Fibrous dysplasia, these tumors tend to recur locally after inadequate surgery, distant metastasis are unusual and systemic therapy probably is not indicated (Dahlin and Unni,1977) .

Small cell osteosarcoma is a recently described variant that is easily confused with Ewing's sarcoma the presence of malignant osteoid matrix distinguishes this tumor from Ewing's sarcoma, the behavior of this tumor is some what different from other varieties of conventional osteosarcoma, so an aggressive approach with systemic chemotherapy seems to be advised (Ayala et al.,1989) .

Finally, Extraosseous osteosarcoma is an uncommon variant that arises outside the bone and occurs most frequently in the soft tissues of the lower extremity in the middle-age adults, usually these tumors are seen as a late complication of radiotherapy, local excision of these lesions is inadequate treatment because local recurrence and distant metastases invariably follow limited surgery (Belasco et al.,1982).

Recently, a histological grading of the effect of chemotherapy on the primary tumor has been utilized and reported to have prognostic significance as well as important for treatment planning (Davis et al.,1994) .

Management

Management requires the experience of a multidisciplinary team familiar with the various management options. Patients with suspected diagnosis of osteosarcoma should be referred to centers with treatment programs before biopsy.

The patients with a primary tumor of the extremity without evidence of metastases requires surgery to control the primary tumor and chemotherapy to control micrometastatic disease. The choice between amputation and limb-sparing resection must be made by an experienced orthopedic oncologist.

Surgical management

A) Amputation

An amputation provides definitive surgical treatment in patients in whom a limb-sparing resection is not a prudent option, which permits removal of all gross and microscopic tumor with clean margins and durable local control in the majority of cases. Most surgeons now feel comfortable with less radical amputations and allow a margin of 6 to 7 cm above the most proximal medullary extent of the tumor defined on these scans, curettings from the medullary canal proximal to the amputation margin are examined microscopically to ensure the adequacy of the procedure. Large lesions around the pelvis or proximal femur usually require amputation, but most sarcomas of the shoulder girdle and knee require a limb-salvage procedure (Huvos, 1990).

Rehabilitation of the patient should begin as soon as possible after surgery, and early ambulation enhances the functional and psychological recovery of the patient. Advances in available prosthetic devices have resulted in improved functional results (Huvos, 1990).

B) Limb-sparing surgery

Limb salvage surgery is a safe operation for selected patients, this technique may be used for all spindle cell sarcomas, regardless of histogenesis. Between 30% and 80% of patients with osteosarcoma can be treated successfully with this technique (Malawer et al., 1989) Successful limb-sparing procedures consist of three surgical phases:

1- Resection of the tumor. This strictly follows the principles of oncologic surgery, avoiding local recurrence is the criterion of success and the main determinant of the a mount of bone and soft tissue to be removed.

2- Skeletal reconstruction. The average skeletal defect after adequate bone tumor resection is 15 to 20 cm, techniques of reconstruction vary and are independent of the resection.

3- Soft tissue and muscle transfers. Muscle transfers are performed to cover and close the resection site and to restore motor power, adequate skin and muscle coverage is mandatory.

The surgical guidelines and technique of limb-sparing surgery used by Malawer in 1993, are summarized in :

1) No major neurovascular tumor involvement.
2) Wide resection of the affected bone, with a normal muscle cuff in all directions.
3) En-bloc removal of all previous biopsy sites and all potentially contaminated tissue.
4) Resection of bone 3 to 4 cm beyond abnormal uptake, as determined by CT or MRI and bone scan.
5) Resection of the adjacent joint and capsule.
6) Placement of the tourniquet proximal to the lesion if possible.
7)Adequate motor reconstruction, accomplished by regional muscle transfers.
8) Adequate soft tissue coverage.

Limb salvage procedures are being used with increasing frequency for the management of primary osteogenic sarcoma, but still are not a good option for patients who have primary tumors in the distal tibia or whose primary proximal tibia tumors has extensive extramedullary involvement, because of the difficulty of ensuring adequate soft tissue margins (Martin et al.,1993).

A number of issues remain to be resolved concerning the role of limb salvage surgery in osteogenic sarcoma, the safety of limb salvage in Osteogenic sarcoma has been questioned by investigators

of the German Society for Pediatric Oncology (GPO), who found a significant higher distant failure rate in patients who were treated with en-bloc resection than in those who underwent amputation (Winkler et al.,1984 and 1986).

However, a retrospective comparison by investigators of the Multi-Institutional Osteogenic Sarcoma Study (MIOS) of patients with primary tumors of the distal femur treated by limb salvage and amputation, failed to demonstrate a difference in overall outcome (Link et al.,1988).

Radiotherapy in Osteogenic Sarcoma

In recent years, the use of routine radiotherapy for osteogenic sarcoma has declined in many centers, but in the past it was used extensively. However, osteogenic sarcoma has been found to be highly radioresistant lesion. Radiation doses less than 6000 cGy have been associated with only transient tumor control (Jenkin et al.,1972), and viable tumor has been observed in amputation specimens after doses of 8000 cGy or more (lee and Mckenzie, 1964).

Because the aggressive chemotherapeutic approach combined with surgery is the primary treatment of choice, radiation therapy is recommended only for several clinical situations as non-resectable lesions in the pelvic girdle or in the vertebral column, facial lesions and as a postoperative adjuvant. In occasional well documented patients, disease-free survival 10 years or more after aggressive radiation therapy has been reported (Beck et al.,1976).

The use of wide-field irradiation to the lungs to prevent the development of pulmonary metastases was also investigated at the Westminster Hospital, the results showed a slight increase in the

survival rate of the group receiving radiotherapy, the actuarial survival of the treated group being 43% and the control group 28% at 5 years. By 1978, when the study was published, chemotherapy was producing even better results (Breuer et al.,1978).

Chemotherapy treatment

Adjuvant chemotherapy

The rationale of adjuvant chemotherapy of osteosarcoma derives from experimental evidence that microscopic metastatic disease is present at the time of diagnosis in the majority of patients who do not have overt metastases and that subclinical metastases can be eradicated if the treatment is initiated when the total burden of metastatic tumor is sufficiently low (Schabel,1977). Evidence from two randomized studies evaluating the efficacy of adjuvant chemotherapy in patients who have received postoperative adjuvant chemotherapy demonstrates a definite improvement in disease-free survival compared to those patients who did not receive adjuvant chemotherapy (Meyer's et al.,1992).

However, osteosarcoma is a relatively drug-resistant neoplasm, and results of the activity of single agents and drugs in combination against macroscopic osteosarcoma have been disappointing. Few drugs have produced responses in more than 15% of patients, and most responses are partial. Notable exceptions are the responses observed in trials of doxorubicin (Adriamycin), cis-platinum, high-dose methotrexate with leucovorin rescue, ifosfamide, and the combination of bleomycin, cyclophosphamide and actinomycin D "BCD" (Jaffe et al.,1977 and Marti et al.,1985), with response rates of 25% to 35%. The hopeless prognosis of patients with osteosarcoma led to the enthusiastic application of these agents, singly or in combination, as adjuvant therapy for patients with non-metastatic osteosarcoma after amputation. Because experimental

evidence, however, suggests that eradication of microscopic metastases is possible, even with drugs that are marginally effective or ineffective against gross macroscopic tumors (Eliber et al.,1987).

The responses in the primary tumor have been variable, with favorable responses observed in 30% to 35% of patients. The overall results are excellent, but comparable to adjuvant studied that utilize regimens of equal intensity but without any preoperative chemotherapy (Weiner et al.,1986 and Winkler et al.,1993). With currently available regimens, approximately 60% to 65% of patients with non metastatic osteosarcoma of the extremity survive without evidence of recurrence. The development of adjuvant regimens has been largely impric, and newer more intensive regimens have further improved outcome. Most regimens currently in use incorporate doxorubicin, cisplatin, high-dose methotrexate with leucovorin rescue and the combination of bleomycin, cyclophosphamide, and dactinomycin. Ifosfamide has recently been shown to have activity against macroscopic osteosarcoma and is included in some regimens under study (Marti et al.,1985).

Neoadjuvant chemotherapy

Pre-surgical (neoadjuvant) chemotherapy has been used with increasing frequency in the past decade in the management of patients with osteosarcoma. This strategy involved concurrently with limb-sparing procedures at the Memorial Sloan-Kettering Cancer Center in 1973. During the presurgical delay necessary for production of the prosthesis, chemotherapy was administered to shrink the primary tumor and thus facilitate limited resection. Patients treated with presurgical chemotherapy appeared to fare better than concurrent patients treated with immediate surgery and postoperative adjuvant therapy (Rosen et al.,1976).

Table (IV) shows the advantages and disadvantages for presurgical and postsurgical chemotherapy.

Table (IV) Considerations for presurgical and postsurgical chemotherapy (Rosen et al., 1983).

Timing of Chemotherapy	Advantages	Disadvantages
Preoperative Chemotherapy	* Early institution of systemic therapy against micrometastases. *Reduced chance of spontaneous emergence of drug-resistant clones in micrometastases. *Reduction in tumor size, increasing the change of limb salvage. *Provides time for fabrication of customized endoprosthesis. *less chance of viable tumor being spread at the time of surgery. *Individual response to chemotherapy allows selection of different risk groups.	*High tumor burden (not optimal for first-order kinetics) *Increased probability of the selection of drug-resistant cells in primary tumor, which may metastasize. *Delay in definitive control of bulk disease; increased chance for systemic dissemination. *Psychological trauma of retaining tumor. * Risk of local tumor progression with loss of a limb-sparing option.
Postsurgical chemotherapy	*Radical removal of bulk tumor decreases tumor burden and increases growth rate of residual disease, making S-phase-specific agents more active and optimizing conditions for first- order kinetics. *Decreased probability of selecting a drug-resistant clone in the primary tumor.	*Delay of systemic therapy for micrometasta. *No preoperative in vivo assay of cytotoxic response. *Possible spread of viable tumor by surgical manipulation.

Responsiveness of the primary tumor to preoperative chemotherapy "as assessed by histologic examination of the tumor at the time of resection" was found to be a powerful predictor of tumor recurrence, unfavorable responders were likely to develop distant metastases despite continued use of chemotherapy after surgery (Huvos et al.,1977). The prognostic significance of tumor response to preoperative chemotherapy has been confirmed in studies conducted by the German Society for Pediatric Oncology (GPO). Several theoretic advantages of preoperative chemotherapy in the treatment of all patients with osteosarcoma "whether or not they are candidates for limb-salvage" have been proposed. Administration of chemotherapy soon after biopsy would provide early treatment of micrometastases known to be present in most patients. This would represent a significant advantages over the traditional adjuvant approach, in which the administration of systemic chemotherapy is delayed by a month or more by surgery (Rosen et al.,1979).Table (V) showed the Histologic Grading of the Effect of Preoperative Chemotherapy on Primary osteosarcoma.

In addition, the administration of preoperative chemotherapy provides a window of time for planning definitive therapy of the primary tumor, permitting the fabrication of customized internal prosthetic devices if required to limb-salvage. Finally, prolonged exposure to presurgical chemotherapy might select for drug resistant tumor cells that might metastasize before definitive surgery (Bacci et al.,1993).

One of the most compelling rationales for presurgical chemotherapy is its use as an in vivo drug trial to determine the drug sensitivity of an individual tumor and to customize postoperative chemotherapy. Results of the Memorial Hospital studies suggest that patients whose tumors are responsive to

presurgical chemotherapy are destined to do well when the same therapy is continued postoperative. Patients whose tumors are unresponsive to presurgical regimen have a much less favorable outlook and might benefit from a change in chemotherapeutic agents. Several objections can be raised on theoretic grounds. Considerations of cell kinetics predict that responsiveness of a bulky tumor may not predict responsiveness of micrometastases (Bode and Levine,1982).

Table (V) Histologic Grading of the Effect of Preoperative Chemotherapy on Primary osteosarcoma (Rosen et al.,1983).

Grade	Effect
I	Little or no effect identified
II	Area of a cellular tumor osteoid, necrotic, or fibrotic material attributable to the effect of chemotherapy, with other areas of histologically viable tumor
III	Predominant areas of a cellular tumor osteoid, necrotic, or fibrotic material attributable to the effect of chemotherapy with only scattered foci of histologically viable tumor cells identified
IV	No histologic evidence of viable tumor identified within the entire specimen

Pre-surgical chemotherapy has been increasingly administration via the intra-arterial route directly into the arterial supply of the tumor to maximize drug delivery to the tumor vasculature and drug extraction by the tumor. Adriamycin and cisplatin, in particular, have been utilized for this purpose. High local drug concentrations have been documented by pharmacokinetic studies, and dramatic responses in the primary tumors have been observed (Jaffe et al.,1983 and 1985).

Use of biological response modifiers

Therapeutic trials based on immunologic approaches to osteosarcoma have been stimulated by documentation of tumor-specific humoral and cellular immune responses in patients and animals with osteosarcoma. The presence in osteosarcoma patients of tumor-specific cytotoxic lymphocytes that are inhibited by a concomitant population of "inhibitor lymphocytes" suggests a role for specific and nonspecific immune stimulation in the treatment of osteosarcoma (Singh et al.,1977)

Early trial using the injection of inactivated osteosarcoma cells or tumor cells lysates induced evidence of cellular immune response in patients but produced no definitive therapeutic advantage (Green et al.,1976). Similarly, nonspecific immune stimulation with Bacillus Calmette-Guerin (BCG) vaccine was not effective therapeutically, even when administered in conjunction with adjuvant chemotherapy (Eilber et al.,1975).

Interferon has been demonstrated to inhibit the growth of osteosarcoma cell lines in vitro and was used as an adjuvant to surgery in an controlled trial from the Karolinska Hospital in Sweden (Strander and Einhorn,1977), a significant improvement over historical results was observed. Although trials of biological response modifiers have not yet yielded promising results, hybridoma technology and advances in the technology for cloning T-cells and expanding such clones for therapeutic purposes provide interesting possibilities for further trials (Rosenberg et al.,1985 and 1986).

Muramyl tripeptide-phosphatidylethanolamine (MTP-PE) is a synthetic lipophilic analog of muramyl dipeptide (MDP), the smallest component of the mycobacterium capable of stimulating

the immune system. Animal studies have demonstrated that MTP-PE-containing liposomes localize to the pulmonary microvasculature resulting in activation of pulmonary macrophages to the tumoricidal state. This approach has been found to be efficacious in canine osteosarcoma, currently, a randomized multiagent, multimodality trial is planned to examine the role of liposome-encapsulated MTP-PE in the prevention of pulmonary metastases in pediatric patients (Kleinerman et al.,1992).

Treatment of metastatic disease

Many investigators have recommended adjuvant chemotherapy after thoracotomy for the management of metastatic osteosarcoma to destroy residual microscopic tumor deposits after surgical treatment of overt metastases (Meyer et al.,1987). Thus, when overt metastatic disease is discovered, a systemic approach is recommended. A careful search for all metastatic lesions, usually by thoracic CT scan and radionuclide bone scan is essential, other investigations to search for metastases to other sites should be performed if clinically indicated (Skinner et al.,1992).

The discovery of unresectable extrathoracic metastases or of pulmonary disease that is obviously unresectable " because of hilar involvement, malignant pleural effusion, massive disease or the presence of more than 16 nodules" is a contraindication to aggressive thoracotomy, and the patient should be treated palliatively. Radiotherapy may be useful in the palliative treatment, the use of chemotherapy " with or without radiotherapy" rarely produces complete response of all metastatic disease, but some patients with inoperable metastases may respond sufficiently to permit complete resection of disease at a later date, with a chance of long-term disease control (Rosen et al.,1978 and Rosenberg et al.,1979).

Thus, for the patients with osteogenic sarcoma, presentation with or development of metastases not a hopeless situation, aggressive systemic treatment of metastases offers prolonged survival for many patients and the possibility of cure for a significant fraction (Meyers et al.,1993).

Ewing's sarcoma

Definition

In 1921, James Ewing described a vascular, hemorrhagic bone tumor composed of uniform, densely packed small cells with rounded nuclei but without distinct cytoplasmic border or prominent nucleoli, and without associated osteoid formation. It is usually occurred in the mid shaft of the long bones or in the flat bones of the trunk (Campanacci,1990).

Epidemiology and biology

Ewing's sarcoma is the second most common malignant primary bone tumor of childhood and accounts for 10-15% of all primary malignant bone tumors, the annual incidence is estimated at 0.6 per million population, although classically believed to originate in bone, Ewing's sarcoma arising in soft tissues has been noted (Miser et al.,1989).

Ewing's sarcoma occurs most frequently in the second decade of life and is rare before 5 or after 30 years of age, the incidence in males is equal to that in females until age 13 years, when, as with Osteosarcoma, males predominate. As with osteosarcoma, epidemiological studies demonstrate that taller persons are more likely to develop Ewing's sarcoma, suggesting that its development is in some way linked to growth (Jurgens et al.,1992).

There is a significant difference in the racial incidence of this tumors, blacks in both the United States and Africa are genetically resistant to Ewing's sarcoma and only rarely are they affected by it , but it is rare in Chinese (Glass et al.,1970).

Ewing's sarcoma has not been associated with congenital syndromes, but an association with skeletal anomalies (i.e., Enchondroma, Aneurysmal bone cyst) and genito urinary anomalies (i.e., Hypospadius, Duplication of the renal collecting system) has been reported. Ewing's sarcoma has been associated with retinoblastoma (Israel et al., 1989).

Chromosomal translocation, t(11:22), is a characteristic abnormality of Ewing's sarcoma (Turc-Carel et al.,1988). The translocation is indistinguishable from that reported in the peripheral neuroepithelioma by Whang-Peng and colleagues, suggesting that these entities have a common histogenesis, this is further supported by the demonstration of neuroectoderm associated antigens on Ewing's cell lines in culture and identical patterns of protooncogene expression as in peripheral neuroepithelioma. Because of this realization tumors that once would have been diagnosed as Ewing's sarcoma are now often designated as peripheral neuroepithelioma or synonymously peripheral neuroectodermal tumor (PNET) (McKeon et al.,1988).

Detection and diagnosis

Clinical presentation

Although Ewing's sarcoma most commonly presents in the femur and bones of the pelvis, it can affect any bone. Unlike osteosarcoma, it often originates in the axial skeleton. Most patients with Ewing's sarcoma seek medical attention because of pain and swelling of the affected bone. Systemic symptoms, such as fatigue, weight loss, and intermittent fever, may be present, especially in patients with metastatic disease. The duration of symptoms before presentation may be measured in weeks or months and is often prolonged in patients who have primary sites in the axial skeleton (Campanacci,1990).

Frequently the entire medullary cavity of the affected bone is involved with the tumor, extension through the bony cortex and into the soft tissues often results in a large soft tissue mass that, particularly with axial lesions, may be larger than the interosseous component (Campanacci, 1990).

Less common presentations of Ewing's include primary rib tumor associated with a pleural effusion and respiratory symptoms, mandibular lesions presenting with chin and lip parasthesias, primary vertebral tumor with symptoms of nerve root or spinal cord compression, and sacral primary with neurogenic bladder (Rosen,1988).

The involvement of the craniofacial bones in Ewing's sarcoma is unusual, it is present in less than 4 percent of cases. A better survival rate was noted in one study that compared patients with craniofacial primary sites to all other skeletal locations, patients with primary mandibular lesions have the most favorable disease outcome (Siegal et al.,1987). Periosteal presentation is rare and so is multifocal osseous involvement (Coombs et al.,1986). Extraskeletal primary locations are reported with increasing frequency, cutaneous affection has also been noted (Peters, et al.,1985).

The incidence of metastatic disease at the time of presentation in patients with Ewing's sarcoma ranges from 14% to 15%. Metastasis is predominantly hematogenous, although lymph node involvement may occur. The lung is the most common site of metastatic disease at presentation and the most frequent site of initial relapse. CNS involvement is detected in fewer than 1% of patients and is the site of first relapse in fewer than 5% of patients.

More commonly, the CNS is involved as a result of direct intracranial or intraspinal extension of bony metastatic disease (Cangir, et al.,1990).

Radiological evaluation

In the past, the radiographic image was considered to be pathognomonic of Ewing's sarcoma when, in a patient's in the first two decades of life, the diaphysis of a long tubular bone, especially in the lower limb, showed fusiform outline, soft tissue extension of the tumor, mottled, moth-eaten-appearing internal pattern of bone destruction, poorly defined margins, and parallel, onionskin, periosteal reaction (Dwyer et al.,1982). Many subsequent studies have demonstrated that neither the distribution of the lesions nor their radiographic appearance is absolutely diagnostic for Ewing's sarcoma, and at best, only a presumptive diagnosis can be made, with histologic confirmation required in all patients, pathological fracture is noted in approximately 5 percent of the cases (Aurfane, et al.,1967).

Angiography may be useful in the diagnosis of Ewing's sarcoma. In about 90% of the cases, hypervascularization is noted, with pathologic tortuous arteries present in approximately half of cases examined, the extensive vascular tumor staining diffusely involves the entire lesional tissue (Tzib et al.,1977). Arteriography is especially valuable in defining the extent of the soft tissue involvement (Kittredge,1970).

The computed tomographic scan of the primary lesion often provides important information about the extent of bone destruction, the cortical breakthrough, and the adjacent soft tissue mass. The relationship of the primary tumor to the neurovascular anatomic structures and adjacent facial planes is displayed in some

detail to help in arriving at a rational plan for local surgical excision (Ginaldi and de Santos,1980).

Magnetic resonance imaging (MRI) is a useful diagnostic tool in Ewing's sarcoma, the high-contrast imaging potential of nuclear magnetic resonance imaging permits fine distinctions between tumor and marrow or tumor and soft tissue, which places this diagnostic method in the forefront of sophisticated radiographic diagnosis. In general, MRI appears to be superior to CT in visualizing bone marrow involvement by tumor but of course does not distinguish among the various small cell tumors of the bone, images in sagittal and axial planes can clearly outline not only the bone changes, but also soft tissue alternations, to which MRI is exquisitely sensitive. Multiplaner and thin-slice imaging techniques furnish fine detail and pinpoint localization in addition to defining tumor margins (Ginaldi and de Santos,1980).

Radionuclide bone scans Using Technetium 99 (bone-scan) may yield useful information concerning the extent of intramedullary involvement by the primary sarcoma, and to find bony metastases (Nair,1985).

Laboratory evaluation

Bone marrow aspiration smears may pinpoint the extent of local tumor and confirm the presence of bone-marrow involvement which occurs early, but it more frequently accompanies long-standing unrecognized disease. Bone marrow aspiration and bone trephine biopsies are to be taken from two separate sites from bones which are not affected by the primary tumor to determine if there has been dissemination. Anterior and posterior iliac crest are considered as separate sites (Akhtar et al.,1985). Although there is no specific serum marker for Ewing's sarcoma, lactate

dehydrogenase (LD) is frequently elevated in those with more advanced disease, the clinical usefulness of pretreatment serum (LD) levels in predicting the course in patients with localized Ewing's sarcoma, (to detect the response to therapy and detecting early relapse of the disease) (Gehan,et al.,1981).

The white blood cells (WBC), differential and sedimentation rate (SR) my be useful to exclude osteomyelitis, which can frequently mimic the picture of Ewing's sarcoma (Cserhati,1978).

Biopsy

Biopsy obviously is essential in the diagnosis of any primary bone neoplasm, open biopsy is preferred to provide adequate material. It is often possible to obtain adequate tissue from the extraosseus soft tissue component as it is easy to process, more viable and representative (intramedullary portion is often necrotic) and also there is no violation of bone integrity. The biopsy should be monitored with frozen section to be certain that the diagnostic tissue has been obtained (Huvos,1991).

Pathology

Ewing's sarcoma is an undifferentiated round cell tumor possessing no unique morphologic markers. It is diagnosed only after the exclusion of the other small, round, blue cell tumors of childhood, which include lymphoma, neuroblastoma, rhabdomyosarcoma, peripheral neuroepithelioma, primitive sarcoma of bone, and primary sarcoma of bone (including small cell osteosarcoma and mesenchymal and myxoid Chondrosarcoma) (Hartman et al.,1991).

Refinements in electron microscopic, immunocytochemical, cytogenetic, and molecular genetic techniques have increased the

sensitivity with which these tumors can be identified, consequently shrinking the numbers of cases left in the Ewing's, biopsy should ensure that adequate tissue is obtained for these special studies. By the light microscopy, Ewing's sarcoma is a diffuse mass of homogenous tumor cells, with marked vascularity and widespread coagulative necrosis are typical features. The cytoplasm is pale, the cellular borders are ill defined (Huvos,1991). In 90% of cases, the presence of glycogen can be demonstrated with periodic acid Schiff (PAS) and diastase reactions, routine histological description has now been supplemented by immunocytochemical techniques using antibodies directed toward a variety of cell-specific antigens (NSE, Myosin, CKMM, Myoglobin, MIC2, and others), and cytogenetic studies to allow better distinction from other small round cell tumors (Tsokos et al.,1984 and Ambros et al.,1991). Table (VI) shows the biological difference between the most common small round cell tumors.

Table (VI) compares the biologic features of Ewing's sarcoma and PNET with neuroblastoma (Israel et al., 1989).

Variable	Neuroblastoma	PNET	Ewing's sarcoma
Clinical presentation :			
age	<4 years	adolescence	adolescence
site	abdominal	thoracic, extremity, pelvis	thoracic, extremity, pelvis
Biological Markers			
*Cytologic features of neural differentiation	+	+	-
*EM features of neural differentiation	+	+	-
*Neurotransmitters	Adrenergic	Cholinergic	Cholinergic
*Surface HLA expression	+	-	-
Cytogenetic characteristics			
Chromosomal translocation	-	t(11:22) (q24; q11-12)	t(11:22) (q24; q11-12)
Gene amplification	+	-	-
Oncogene expression			
MYCN	+	-	-
MYC	-	+	+

Staging

There is no uniformly accepted staging system for Ewing's sarcoma. A system based on a TNM concept is more appropriate for this disease than a system based on the extent of the disease after a surgical procedure, because the approach to local control of this tumor is rarely surgical. Experience suggests that the size of tumor has prognostic importance. In several studies, the prognosis of those with lesions less than 5 to 10 cm in the maximal diameter was better than for those with larger tumors (Gobel et al.,1987). Node involvement (N) is rare. The presence of metastatic disease (M) dramatically reduces the likelihood of survival (Huvos,1991).

The most favorable prognostic factors are a distal primary tumor, normal serum lactate dehydrogenase, and absence of metastatic disease at presentation. Pelvic and sacral sites for primary tumors and metastatic disease are the least favorable factors. A partial or complete response to initial chemotherapy is a strong predictor of long term disease control (Cangir et al.,1990).

Treatment

In 1921, James Ewing observed that " small cell sarcoma of bone" was, in contrast to osteosarcoma, responsive to a radium implant. For the next half century, the treatment for Ewing's sarcoma employed local surgery or radiation therapy or both. Even, when local control was effective over 90% of patients died, usually of metastatic disease (Hustu, et al.,1968). Adjuvant chemotherapy has had a major impact on the cure of Ewing's sarcoma. The earliest trials, begun in the mid-1960s at St. Jude Children's Research Hospital and the National Cancer Institute, demonstrated that adjuvant chemotherapy had the potential to cure patients with Ewing's sarcoma. Progress in the multimodality therapy of Ewing's sarcoma over the past 25 years has resulted in the expectation that approximately 50% of patients with localized tumors can be cured (Miser, et al.,1989).

The treatment of Ewing's sarcoma is currently based on promising investigational therapeutic regimens, which include neoadjuvant high-dose chemotherapy, radiation therapy, and surgical excision of the primary tumor (Nesbit et al.,1990). The complex interplay involving chemotherapy and local irradiation is a delicate equation in which the indications for surgery are still being evaluated. The multidrug neoadjuvant chemotherapy, which

precede irradiation, establishes the chemotherapeutic response of the tumor (Rosen,1988).

Surgery

The beneficial role of surgery in Ewing's sarcoma is still unclear, because with surgical therapy alone, the long-term survival rates of patients in most early series were less than 10%, with failure usually caused by distant metastatic disease. The success of adjuvant chemotherapy in preventing distant failure in patients with Ewing's sarcoma and the effectiveness of radiation therapy in controlling the primary site of disease have resulted in abandoning surgery as the sole primary modality of therapy However, there are no controlled data that compare the advantages and disadvantages of surgery and irradiation for the primary treatment of Ewing's sarcoma (Bacci et al.,1982).

More recently Wilkin's and others reported their results of 140 patients with Ewing's sarcoma those 92 patients with localized disease were treated by 3 main modalities; radiation alone (16 patients), radiation + chemotherapy (49 patients) and complete surgical excision (27 patients). The 5 year survival rate for those managed without surgery was 27% VS. 74% for those managed with complete surgical excision, thus suggesting role for surgical resection in the management of Ewing's sarcoma (Wilkins et al.,1986).

Surgery (when indicated) should be performed after 4 courses of chemotherapy (induction) as soon as peripheral blood counts have reached, TLC > 2000, ANC > 1000, and platelets > 100,000, when surgery is undertaken, the next course of chemotherapy will be given as soon as possible post-operatively. Unless there are exceptional circumstances, chemotherapy should resumes within 2

weeks post-operatively, and in most cases, can begin within a few days of surgery. The indications for primary surgical resection of Ewing's sarcoma used at some institutions include a lesion in an expendable bone, such as rib, clavicle, fibula, or individual bones of the feet. Surgery usually follows initial chemotherapy which can be executed to significantly debulk the lesion. Amputation may be indicated if there is an unmanageable pathologic fracture, but many of the fractures heal during initial chemotherapy, allowing subsequent irradiation. If the tumor arises at or below the knee in a young child (<6 years) and a major uncorrectable functional deformity is expected from radiation therapy, amputation or limb-sparing surgery with an expandable prosthesis should be considered (Pritchard,1980).

Radiation therapy

James Ewing initially described the tumor's susceptibility to radium. It has been recognized that this tumor is highly responsive to radiation therapy, before the availability of chemotherapy, local control of Ewing's sarcoma was attained in 44% to 86% of patients with radiation doses greater than 4000 to 5000 cGy, even through long-term survival was low (16-25%) (Ewing,1924).

With the addition of effective chemotherapy and local irradiation, local recurrence of Ewing's sarcoma is approximately 10% for distal extremity lesions and 20% to 40% for patients with proximal extremity or pelvis primaries. Coordinated therapy has increased the control of microscopic systemic disease and markedly increased survival, local control with primary irradiation and chemotherapy appears to depend on tumor size, there are significant differences in local control and survival with lesions with volumes greater or smaller than 100 ml (Nesbit et al.,1990).

Radiotherapy is delivered as local irradiation of the primary tumor region with adequate margins including the whole tumor-bearing compartment. The compartment dose is 45 Gy. In cases of definitive irradiation the area of the tumor including safety margins as defined below are boosted to a total dose of 55 Gy (Nesbit et al.,1990).

The region of radiotherapy has to be planned according to tumor extent at diagnosis. The 45Gy compartment volume has to include the whole tumor-bearing compartment. If long bones of the extremities are affected, the epiphysis distant from the tumor may be spared if a 5cm safety margin can be assured (Miser et al.,1985).

For tumors of the trunk there must be a 2cm safety margin based on the initial tumor extent, an exception are tumors with non-infiltrating extension into cavities, e.g. pelvic or chest wali tumors. For these tumors the region of radiation may be planned according to the intrathoracic and intrapelvic tumor extent at the time of radiotherapy including a 2cm safety margin. However, for infiltration of bones and the soft tissues the volume of radiation must be planned according to the tumor extent at diagnosis (Dunst et al.,1991). The boost is given to the remaining tumor volume following initial chemotherapy and chemotherapy-radiotherapy in selected cases plus a 2cm safety margin in all directions (Dunst et al.,1991).

Acute side effects may occur during radiotherapy in rapidly proliferating tissue such as skin and mucosa in the field of irradiation, they require symptomatic treatment and usually clear within 1-2 weeks following termination of radiotherapy. The radiated skin should be kept dry using a non-perfumed powder, and mechanical irritation should be avoided. When extremities are

irradiated prophylactic physiotherapy should be done to avoid contractures. In patients receiving pelvic irradiation, it is important to consider ovarian transposition in young women, and should be aware of symptoms of radiation enteritis, e.g. spasm and diarrhea. mild cases usually clear with symptomatic treatment such as dietary measures. In sever cases it may be necessary to interrupt radiotherapy (lewis et al.,1977).

The long term functional results in patients treated solely with radiotherapy appear to be related to multiple factors, including tumor site, volume of tissue irradiated, dose of irradiation, age, involvement of a major joint, and presence of open epiphyses. In young patients with an immature skeleton, radiation treatment may result in the subsequent development of limb length inequality. There is concern about the induction of a second malignancy in the irradiated site in 3% to 18% of patients, the incidence appears to be related to total radiation dose, to radiation energy (orthovoltage), and to the use of chemotherapy (Lewis et al.,1977).

Chemotherapy

Before the use of adjuvant chemotherapy, long-term survival of patients with Ewing's sarcoma was rare. Single-agent chemotherapy trials were initiated in the 1960s with highly encouraging results, many agents appear to be active against Ewing's sarcoma, with cyclophosphamide and doxorubicin consistently the most active. Few new agents have been developed for this or other pediatric cancers, possibly because drugs are tested against tumors resistant to multiple drugs and radiation (Nesbit et al.,1990).

In 1973, a multi-institutional randomized trial, the first Intergroup Ewing's Sarcoma Study (IESS-1), was initiated. Patients without evidence of metastatic disease were treated on one of three

treatment regimens. Local tumor control was planned with radiation therapy to the entire involved bone in doses ranging from 4500 to 5490 cGy, depending on the patients age, followed by boost of 1000 cGy to gross radiographically demonstrable tumor. Daily dose fractionation was 200cGy, delivered 5 days each week. In addition it local therapy, patients were randomly assigned to receive adjuvant VAC (Vincristin, Actinomycin-D and Cyclophosphamide) with Doxorubicin (regimen 1), VAC without Doxorubicin (regimen 2), or VAC plus bilateral pulmonary irradiation, which consisted of midplane dose of 1500 to 1800cGy (regimen 3). The duration of chemotherapy was 1.5 to 2 years. At 24 months, the failure-free survival was 74% with VACA, 58% with VAC + lung irradiation, and 35% with VAC. The VACA regimen was more intensive than VAC alone, and it was thought that improved failure-free survival may have resulted from more intense therapy (Burgert et al.,1988).

The second Intergroup Ewing's Sarcoma Study (IESS-II) evaluated the role of a more intensive regimen, which relied heavily on the two most active agents, cyclophosphamide and doxorubicin. In IESS II the " high-dose intermittent" chemotherapy consisted of vincristine ($2mg/m^2$) and doxorubicin ($75mg/m^2$) alternating every 3 weeks with vincristin and cyclophosphamide ($1400mg/m^2$). Although there is as yet no statistically, significant difference in survival between this "high-dose intermittent therapy" and "low-dose continuos therapy" for patients with non pelvic primaries, and the treatment administered on IESS II appear superior to those of IESS 1 (Burgert et al.,1990).

NCI investigators tested a strategy that maximized the use of cyclophosphamide and doxorubicin to achieve complete remission followed by irradiation to the primary and metastatic sites of disease, which was followed by total body irradiation. Improved

outcome was correlated to a higher dose intensity of doxorubicin delivered in that study and autologous bone marrow reconstitution to prevent systemic recurrence. Although most patients achieved a complete response, this has not been maintained in the patients presenting with metastatic disease (Miser et al.,1985).

In 1985, at the NCI, a phase II study evaluating the role of combination Ifosfamide and etoposide in patients who had relapsed begun, in order to develop a new approach to the treatment of patients with metastatic disease (Meyer et al.,1992). The study was based on the fact that Ifosfamide is very similar in structure and mechanism of action to cyclophosphamide, an agent that is one of the most important in the treatment of Ewing's sarcoma. It was also based on the fact that both Ifosfamide and etoposide has significant single agent activity in patients who had failed 1ry therapy with regimens containing both cyclophosphamide and doxorubicin. The preliminary, results of this phase II study have shown that most patients with Ewing's sarcoma will have significant responses to this combination even when resistant to a cyclophosphamide-based regimen (Miser et al.,1987).

Treatment of metastatic disease

Patients with detectable metastasis at diagnosis will not be randomized and will be treated as a high risk patients receiving the investing the Ifosamide, etoposide alternating with VACA. For local control of the primary tumor, guidelines will be as usual.

A-Lung metastases

For treatment of the lung metastases these patients should, in addition to chemotherapy, receive whole lung irradiation as outlined in radiotherapy section. Radiotherapy to both lung should also be delivered in patients who have obtained a complete

remission of their visible lung disease following chemotherapy surgical removal of the remaining lung disease prior to radiotherapy may be attempted if possible (Miser et al.,1987).

B-Bone metastases:

Patients with bone or bone marrow metastases at diagnosis have extremely poor prognosis with <10% survival 2 years following diagnosis. Results consolidation remission with autologous or allogeneic bone marrow transplantation are promising. For this reason patients with primary bone or bone marrow metastases at diagnosis are treated according to the high risk protocol with chemotherapy. Patients who have achieved a documented complete remission should be considered for autologous or allogeneic bone marrow transplantation (Cornbleet et al.,1981).

Psychological disorders of childhood cancer

Childhood cancer today, with the marked improvement of treatment is no longer a model for understanding psychological aspects of terminal illness. Instead, this is now a model for studding chronic illness and long survival (Bond and Wellisch,1990).

In the united states in 1991 it is estimated that nearly 2000 persons will be diagnosed with bone cancer. Osteosarcoma and Ewing's sarcoma account for the majority of these patients which commonly affect the children and adolescent. An adolescent who received a diagnosis of bone cancer (osteosarcoma and Ewing's sarcoma) twenty years ago could expect an 80 percent chance of dying of this disease. This mortality rates has been drastically improved: an individual receiving that diagnosis today could expect a 60 percent -70 percent chance of surviving. Many patients whose only surgical option would have been amputation may now be considered for limb-sparing surgery. The loss of limb adds to the burden of cancer and become a constant reminder of the disease and its consequences (Boring et al.,1991).

In the light of medical success in improvement of the survival rates of children with cancer, the focus of pediatric oncology now includes a heightened awareness of multiple psychiatric and psychological implication of cancer in childhood. The child's adaptation during disease and treatment and the quality of life as a long-term survivor are of paramount concern, for the child with cancer, successful outcome in all psychological arenas hinges on early assessment, prevention, and intervention (Lansky et al.,1993).

Most children with a suspected malignancy are referred to a tertiary care facility for definitive diagnosis and initiation of treatment. This is the period of intense activity and extreme stress. Families are faced with the diagnosis of cancer and the fears that it arouses, they are frightened by the life-threatening illness and by the unfamiliar environment of a large medical center. Guilt is pervasive as parents search of causes, often looking for past errors of omission or commission (Holland and Rowland.,1989).

In the midst of this distress, parents must begin to learn about the many aspects of their child's illness. They will be faced with the prognostic implications and the many necessary diagnostic and treatment procedures. The parents' education includes familiarisation with the names of various tests, chemotherapeutic agents, and side effects, as well as understanding possible surgical interventions. In addition, parents and the child must learn about general hospital and clinic routines (Johnson et al.,1979).

The initial presentation of the diagnosis to the child is also critical. It is this encounter that sets the stage for future relationships among the child, parents, and treatment team. All children should told about the diagnosis by their parents or by the physician with parents present. Later, older children will benefit from private discussions about their disease and treatment with the medical staff as they develop trust in and comfort with their caretakers (Lansky et al.,1993).

The child's and family's ability to effectively assimilate and use illness-related information is also influenced by individual and family characteristics. Early assessment of the whole family is thus important to comprehensive care, evaluation begins with a careful history of the child that includes questions about developmental

tasks, school performance, and school functioning, a description of family demographics outlines family members' ages, sex, and any special problems (Shields et al.,1995). In addition to the new stresses and anxieties imposed by the diagnosis of cancer, both the child and family may be coping with a variety of ongoing pressures, these factors must be identified (Moore et al.,1986). A sample list of possible stress factors is presented in table (VII).

The magnitude of these stresses, individually or in combination, has a significant impact on the family ability to deal with the new illness, it is impossible, however, to identify specific factors that place a particular family or child at risk, each child and family reacts differently. The family's previous manner of coping with major events may best predict their way of coping with cancer (Hersh and wiener 1993).

Table (VII) Stress factors impacting on child and family (Moore et al., 1986)

Illness related	Non-illness related
*Cost	*Job or school change
*Separations	*Move or relocation
*Disruption of usual routines (school, job)	*Marital problems or divorce
	*School problems
*Unfamiliar caretakers	*Family illness, injury, or death
*General burdens (traveling, sleep interruption)	*Financial problems
	*Family problems
*Isolation from peers	
*Threat of death	

The child's Reaction to the Diagnosis and Initiation of Treatment

Developmental Considerations

A diagnosis of cancer is traumatic for all children, but the child's age and developmental level significantly influence the experience of illness-related events. Children will be particularly upset when circumstances interfere with unique developmental needs (Hockenberry et al 1989).

Young children

The young child's immediate concerns revolve around hospitalization, separation from parents, and fear of medical procedures. Toddlers and pre-schoolers are particularly sensitive to separations and changes in familiar routines. Theses young patients may view hospitalisation and disruption of usual daily life as punishment, this perception is further reinforced by the experience of painful and invasive medical procedures. Hospitalised young children need constant reassurance from their parents that they will not be abandoned and that hospitalisation and medical interventions are not a form of punishment (Pefferbaum,1990).

So, before any medical procedure, children should receive a brief, honest description of what the procedure is, how it will be done, and the intensity and duration of any pain involved. Behavioral intervention are also effective in reducing the child's anxiety and distress surrounding medical procedures, a system of positive incentive in combination with strategies of attention distraction can help the child control fear before and during tests or treatments (Manne et al.,1990).

When surgery is planned, the child needs special preparation, visits to the operating and recovery rooms and familiarisation with the surgical personnel help ease the child's fear and lessen the shock of the strange environment. The child also needs to be prepared for the consequences of surgery, discussions about amputation or other resulting disabilities or deformities must begin very early. This kind of trauma is best handled when children have several days to assimilate the information, voice their fears, and adjust expectations in light of further explanation. Other amputees are of considerable assistance in allaying some of the child's anxiety, finally, as with all intensive and painful procedures, physical comforting is reassuring (Holland and Rowland,1989).

The stress of illness and hospitalisation can cause regression in all patients. For the young child, regression might take the form of loss of newly acquired skills (e.g., toilet training, speech, self-feeding). Previously discarded behaviors, such as thumb-sucking or clinging, may re-emerge, parents should be understand this regression as part of the child's efforts to cope. Adaptive regressive behaviors generally subside after the acute phase of illness, at times, however, regressive patterns will become entrenched and can become serious problems. A small child who is separated from parents by hospitalisation experience profound separation anxiety, less commonly, depression can occur which takes the form of withdrawal and lack of emotional involvement in surroundings (Lesko and Holland,1990). Table (VIII) Summarizes the disruption by illness of normal developmental tasks and the basic interventions used to ameliorate these disruptions.

Table (VIII) Disruptions of normal developmental tasks and basic interventions (Lesko and Holland,1990)

Stage	Task	Disruptions	Intervention
Childhood (early)	*Motor *Speech *Cognition *Family bonding Socialization	Developmental slow Regression Separation anxiety Withdrawal	Physical / Social stimulation structured "play" Family contact
Childhood (late)	*Prepubertal peer-relations *↑Intellectual skills *↑Physical prowess	Being " different" School phobia Death fears	Maintain near normal appearance Minimize school absences Discuss illness
Adolescence	*Menarche/ puberty *Peer acceptance *↑Independence *Sexual experimentation	Looking different ↓ School / Physical performance ↑ Dependence Conflicts about self sexuality	Maintain near normal appearance Maintain peer contact Support independence Counseling

School-age children

In the school-age child, diagnosis and initiation of therapy arouse many of the same feelings and fears seen in the pre-schooler. Separation, strange people, an unfamiliar environment, fears of abandonment and punishment, and threats to body integrity are all major concerns of the school-age child (Hockenberry et al.,1989).

School-age children cope with the stress of illness in a variety of ways. They may have a delayed initial reaction or may respond

immediately with acute anxiety or panic, other reactions include psychosomatic complains, nightmares, labile emotions, regression, or "adult acceptance". These children are likely to be verbal about their illness, requesting information about all aspects of the disease and treatment. This is a developmental period of vigorous inquiry, and the diagnosis of a sever illness, with all the anxiety, prompts a barrage of questions. From the outset, these questions should be answered in a simple straightforward approach (Nir,1981). Compared with the pre-schoolers, school-age children have heightened concerns about privacy. Their extreme modesty can interfere with medical examinations and procedures. Within reason, every attempt should be made to respect their need for privacy (Whaley and Wong,1991).

Parents often need to engage in a specific scripting because they are not used to conveying stressful information to their children. It is important to start this process at the first encounter, if the family's usual practice is to avoid issues, the child will learn that asking questions produces discomfort in the parents, which increases the child's personal distress (Moore et al.,1986).

A Particularly difficult behavior pattern to change, once it has been established, is a symbiotic relationship between the sick child and a parent, usually the mother. The pair becomes an inseparable dyed, with the child continuing to regress and becoming progressively dependent on the parent. Extreme separation anxiety occurs in both child and parent. The pair may isolate themselves in the child's hospital room or, after discharge, remain in seclusion at home. They may withdraw from all social contact, including other family members. At times, the child's medical care may be obstructed, as the parent and child resist even the involvement of physicians and nurses. This seriously maladaptive behavior can be

prevented be early attention to and active initiation of mother-child separations (Shields et al.,1995).

The major activity of these children is school. School begins the processes of working toward independence from parents, establishing peer relationships, and acquiring academic skills. The physical and emotional concomitants of life-threatening illness result in a significant disruption in school attendance and performance. The development of friendships and separation from family are key tasks for school-age and adolescent children. The necessary dependence, compliance, and loss of control that accompany illness and treatment may impede the drive to achieve separation in the school-age child and independence in the adolescent (Katz et al.,1988).

Adolescents

For the adolescents, in a state of transition between childhood and adulthood, illness presents unique issues. Focal concerns about independence, appearance, acceptance, sexuality, and future plans are immediately confronted (Zeltzer,1993). The adolescent's strivings toward autonomy and self-determination are inevitably threatened by the forced dependence, compliance, and loss of control accompanying the illness and treatment. Most adolescents are self-conscious about their appearance and emerging sexuality. These issues assume an even greater significance in the face of disease and treatment-related delays in puberty, as well as such physical changes as hair loss, weight alterations, and mutilating surgery (Orr et al.,1984).

Concerns about infertility may be misconstrued or not addressed at all, may result in fears of impotence or frigidity, and may increase the chances that the adolescent will develop a distorted image of his or her own sexuality. The sense of physical weakness

and vulnerability similarly interferes with the adolescent's maintenance of peer interactions, it has been suggested that teens with cancer are more avoidant and guarded in their male-female relationships than their healthy peers (Fritz and Williams,1988).

For the adolescent, close relationships with peers of both sexes are important, which the youth builds while struggling with self-definition in social, moral, and physical domains. If treatment causes striking changes in appearance, adolescents may be devastated, because body image is so central to self-esteem (Heiney et al.,1991). Although these disruptions do not necessarily lead to psychopathology, they have potential long-term and significant implications for the progressive development of the child social confidence and competence. Thus, maintaining contact with friends is important throughout treatment, even during times of low energy (Kazak, and Meadows,1989).

School life is also disrupted. Both social and academic pursuits are interrupted, delayed or critically altered by frequent and prolonged school absences. Feelings of isolation or embarrassing physical changes may make it difficult for the adolescent to return to school or maintain adequate attendance (Lansky et al.,1990). Long-established plans or expectations about career or family may require reconsideration in light of physical limitations, academic difficulties, and questions related to fertility and parenthood, each adolescent responds to these stresses differently, and the same child may react differently at different times. Emotions ranging from depression and withdrawal to agitation are common. For some adolescents, verbal expression is important. For others, behavioral outlets for feelings are more commonly used (Levenson et al.,1982).

The past two decades have seen significant improvement in the therapy and survival of cancer patients. With increased survival, attention has been focused on the quality of life. The definition of quality of life is difficult, and even illusive, since it embodies numerous items and considerations. The meaning of a "good" life appears to be very subjective and cannot be generalised and, despite various methods devised, is difficult to measure (Aaronson,1991).

Lifestyles of amputees often need to change to accommodate the patient's physical limitations and decreased mobility, the degree of change not only depends on the type and level of amputation and prosthesis, but the nature and degree of rehabilitation and the patient's will and desires. Most amputees, if adequately rehabilitated, are capable of independent living in and out of their home (Tebbi et al.,1987). Some studies have found that a greater length of denial and a longer period of time is needed for adaptation for patients undergoing limb-salvage as compared to amputation (Tebbi et al.,1989).

It appears that the former group focuses their emotional efforts on saving their limb, whereas amputees, after a relatively short period of mourning the loss of their limb, go on to deal with their disease and treatment. This may be in part due to the need for a longer period of surgical care and hospitalization after a limb-salvage operation (Weddington et al.,1985).

Hospitalization

Usually children with cancer are hospitalized many times during their illness. Repeated and long-term hospitalization can cause a specific pattern of emotional reactions. The child's reactions were particularly related to the fatal character of the disease. Hospitalization was not only experienced as temporary separation

but also as a reminder of final separation. Clinging, signs of distress, despair, withdrawal, and depression were associated with this separation anxiety (Hollenbeck et al.,1980).

It has been noted that patient care in reverse isolation may increase the risk of psychological disturbance because it implicates a reduction of social contact and sensory stimulation. Although psychological problems are reported including anxiety, depression, and behavioral changes in sleep and play, the change in patients' emotionality seemed to be influenced by the severity of the disease rather than the degree of isolation. It could not be concluded that reverse isolation inevitable leads to psychological problems but this favorable outcome may be due to the high degree of involvement of parents and medical staff, especially with young children, who are less capable in generating their own sensory stimulation than older children (Hutchison & Cockburn,1986).

Child's reaction to hospitalization

Hospitalization, with its separation from home and the various treatment procedures encountered, may cause a variety of reactions depending upon the child's level of psychological development, the family's response, the meaning of the illness and hospitalization, the type of treatment required and other factors. The older the child, the more he will understand the realistic meaning of this experience and the less likely he will be misinterpret its significance. The child from age 4 through the early school age period, although he may experience separation anxiety, is usually more preoccupied with fears of bodily mutilation (Kempe et al.,1978).

Older children are usually able to comprehend the reality of the hospital experience more objectively but may still show signs of mild regression and anxiety over bodily functioning. Fears of

genital inadequacy, muscular weakness, and loss of body control or of helplessness during anesthesia may enhance feelings of anxiety and inferiority that are characteristic of this stage of development. The same trends may be seen in adolescents but in a muted form, they may have difficulty in accepting the authority of the medical staff (Kempe et al.,1978).

Children should be prepared for hospitalization. All children should be told simply and truthfully why they are going to the hospital. They should be given a general impression of what being in a hospital is like and what will be done to make them comfortable. They must be assured that their parents will remain in contact with them (Hutchison & Cockburn,1986).

Outpatient care

With modern cancer treatment patients are less hospitalized and after initiation of treatment medical care will often be provided on an outpatient basis. This is advocated in order to decrease the emotional side-effects associated with hospitalization and to facilitate the patient's return to family, friends, school and community while still undergoing vigorous treatment. There is an emphasis on having the child return to the lifestyle as normal as possible because only a physically and emotionally successful rehabilitation will result in a truly cured child (Kagen Goodheart, 1977).

Pain in pediatric oncology

Pain in cancer patients is a serious problem. Fifteen percent of non-metastatic cancer patients (specially bone cancer), both adult and children complain of pain. This figure rises to 33 percent in metastatic cancer patients and includes almost all patients with advanced disease. Pain has a subjective component that enhanced in

the presence of anxiety or depression, conversely, the presence of uncontrolled pain causes anxiety and depression. Children are even more undermedicated than adult . Very young children cannot make their complaints known clearly. Older children often make complaints in the context of a predictable regression, which is then used to disqualify the complaints (Massie et al.,1987).

Many children became withdrawn as a means of coping with pain, reluctance to medicate adequately, or at all, may result in the child's recall of a painful episode as a traumatic experience, making him or her more vulnerable to anxiety in the future episodes (Ferrell,1995).

Management of stressful and painful procedures. Recent work on Post Traumatic Stress Disorder (PTSD) in children reinforces the principle that aggressive prophylactic treatment of stressful procedures is important not only for the children's current comfort, but also for their long-term adaptation. Emotional support, education and environmental manipulation, anesthesia, psychopharmacology, and especially behavioral methods must all be brought into play (Kuttner et al.,1988).

Long-term survivors

The anxiety felt when therapy is completed dissipates slowly as months pass and the child remains free of disease. As concern about recurrent disease diminishes, other worries take its place. These include fears about long-term sequelae of the illness and treatment. Long-term survivors of childhood cancer face serious future physical and psychological risks, many of which are as yet unknown (Lansky et al.,1993).

The survivor must first cope with medical sequelae, ranging from changes in appearance (e.g., obesity, physical disabilities left by surgery), to defects in major organ systems (e.g., cardiac, liver, renal), to worries about fertility, progeny, and the risk of second malignant neoplasms. Routine follow-up of long-term survivor should include screening for these potential late effects as well as age-appropriate education regarding special lifetime health risks and necessary health maintenance practices (Meadows et al.,1980).

Potential disruptive effects of childhood cancer and its treatment extend into other areas of the survivor's life. Developmental disruptions experienced during treatment have undeniable implications for future psychosocial adjustment. The degree to which these disruptions affect the child's later adjustment varies, increasing time since disease onset and younger age at diagnosis seem to be related to fewer subsequent adjustment problems. Older children and adolescents may be particularly sensitive to interruptions in their developing peer and intimate relationship, school and extracurricular activities, and plans for future life-style and occupation. They are more likely to exhibit agitation, restlessness, and hyperactivity (Koocher et al.,1980).

Specific educational and occupational achievements and choices may be affected by cancer survival, the academic ability of the long-term survivor may potentially be affected by intellectual deficits and learning problems. Cancer therapy, specifically cranial irradiation, has been associated with problems in attention and concentration, performance under pressure, visual and auditory memory, and mathematical skills. Language skills appear to be relatively unaffected. These potential learning problems, coupled with disruptions in school attendance, may limit the child's

educational achievement and occupational attainment (Lansky et al.,1993).

Recommendations for intervention include baseline assessment and periodic monitoring of neuropsychological functioning. Continuous evaluation of academic performance as a part of regular aftercare permits prompt identification of learning disabilities that may not appear foe several years. Once identified, learning problems can be dealt with through an educational program tailored to the individual child's specific areas of strength and weakness (Lansky et al.,1993).

Even when they are successful in overcoming learning and educational barriers, long-term survivors may encounter difficulties in the workplace as a result of their cancer history. Hiring discrimination, ineligibility for health and life insurance, and employers' attitudes about cancer may all complicate the cancer survivor's entry into the work force (Lansky et al.,1986).

Psychological problems of families

Cancer strikes not only the patient, but the family as well. When the course of cancer was brief and relentless, the family's task was to endure a short, anguished period followed by bereavement. Today, the course is more often prolonged and draining, punctuated by crises with alternating periods of hope and despair, often ending ultimately in loss. Even when the outcome is successful, the family may remain scarred because the emotional cost, as well as the cost in time, money, lost resources, and family disruption, has been so great. This is reflected in psychiatric morbidity (Dahlquist et al.,1993).

Caretakers in the family are in double jeopardy : They must provide care for the patient, make up for the loss of the patient's contribution to the family, respond to increased expenses and physical needs in the face of decreased input and resources, and respond to the increased demands of other members. And they must do all this while coping with their own emotional anguish, which usually closely parallels that of the patient, on a decreased amount of sleep and rest, often creating a vicious cycle of fatigue and insufficiency (Hersh and Wiener,1993).

Nevertheless, caretakers must be watched for signs of psycho-pharmacologically treatable anxiety and depression, which do develop and require ongoing support and help to set their priorities. Caretakers must also be helped to remain connected to as broad a support system as possible since beleaguered families may become withdrawn and reclusive, rejecting social supports when they most need them (Mulhern et al.,1992).

Previous psychopathological problems in a member of the cancer patient's family is often exacerbated by the stress of cancer illness in the patient. This situation may seriously hamper the delivery of care or place intolerable pressure on the cancer patient. The family member must be evaluated and referred for treatment; in some cases, crisis intervention may be necessary. One must also set limits and structure the environment to protect the cancer patient as much as possible (Evans,1975).

Prevalence of psychological disorders

There are no psychological, medical, or social reasons to presume that the prevalence rate of psychiatric disorders for patients with bone malignancies is different from that of patients with other forms of cancer (Zeltzer,1993).

The actual incidence of psychiatric disorders in cancer patients has only been determined in the past decade, by assessing ambulatory and hospitalized patients in three cancer centers using standard criteria. 53% of patients interviewed though showing signs of being under stress were coping adequately. The remaining 47% had a diagnosable psychiatric disorder. The most common by far was adjustment disorder with anxious and depressive symptoms. Depression was next, seen in 13% of those with a psychiatric diagnosis, central nervous system complications resulting in organic mental disorders were present in 4%, prior psychiatric disorders account for only 5% of cases (Derogatis et al.,1983).

More recently, Holland and Rowland (1989), studied the spectrum of psychiatric disorders in patients with cancer, adjustment disorders were encountered in 30% of the study group. Common psychiatric disturbances in cancer patients are classified into two major categories:

1-Psychiatric disorders directly related to cancer patients including:

A- Adjustment disorders.

B- Major depression.

C- Delirium

2-Pre-existing psychiatric disorders aggravated by malignancy including:

A- Primary anxiety disorders.

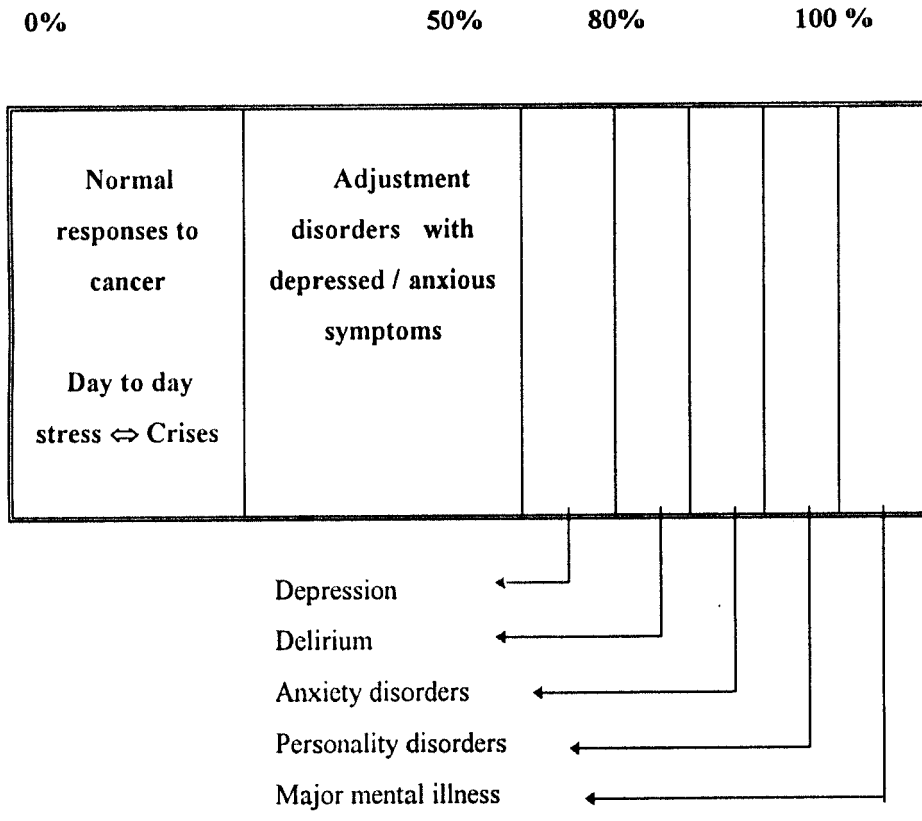
B- Personality disorders.

C- Major mental illness.

The Psychological Collaborative Oncology Group (PSYCOG)

Reported on study of 215 randomly selected hospitalized and ambulatory patients with many cancer diagnosis at three major cancer centers (Johns Hopkins University, University of Rochester, and Memorial Sloan-kettering Cancer Center). Diagnostic interviews revealed that 47 percent had a DSM-III diagnosis, these patients had the following disorders: 32 percent, an adjustment disorder with depressed, anxious, or mixed mood; 6 percent, major depression; 4 percent, organic mental disorder; 3 percent, personality disorder; and 2 percent, anxiety disorder. Nearly 90 percent of the psychiatric disorders observed were reactions to or manifestations of disease and treatment. Less than 11 percent of patients represented prior psychiatric problems, such as personality and anxiety disorders or major mental illness (manic depression or Schizophrenia) (Holland and Rowland,1989) Figure (1)

Figure (1) Spectrum of psychiatric disorders in cancer.



(Holland and Lowland, 1989)

Therefore, the physician treating patients with cancer is generally treating psychologically healthy individuals who have emotional reactions related to issues posed by their illness and its treatment. Patients with malignancies are no more or less depressed than patients with equally physically debilitating non oncology illness, only about 25 percent of all cancer patients, irrespective of their status and diagnosis, experience significant emotional distress that is often depressive in content (Lesko and Holland,1990).

1- Psychiatric disturbances directly related to cancer

Adjustment disorders

Adjustment disorders are an exaggeration of the mixed anxiety and depression seen in self limited stress responses. It is presented by an unusual persistence and undue interference with functioning. Interventions are aimed at helping the patient resume successful coping by use of several modalities. Individual psychotherapy focuses on clarifying the medical situation and the meaning of illness and on reinforcing the patient's positive coping strategies. It is often desirable to include a spouse or family member to enhance support at home. Group therapy, with a focus on illness, is often helpful, as are behavioral methods such as relaxation and hypnosis (Holland,1989).

Couple and family therapy may be helpful when interpersonal issues are prominent. The decision to prescribe a psychotropic drug requires a high level of distress and inability to carry out daily activities. Low doses of alprazolam, lorazepam, or oxazepam control symptoms and need not to cause undue daytime sedation or risk withdrawal or dependence in these psychologically healthy patients (Holland et al.,1991).

Exaggeration and progression of adjustment disorders make them difficult to differentiate from major depression and primary anxiety disorders and may best be treated as if they were in the more serious category so as to give the patient all possible treatment benefits (Holland et al.,1991).

Major depression

Major depression may be either strong reactive component or true major depression. An essential point to diagnose major depression in cancer patients is to omit in its assessment the associated medical illness and to avoid our dependence on such somatic symptoms as lack of energy, and anorexia. The diagnostic symptoms and signs of major depression may include :" Dysphoric or sad mood, feeling of helplessness and hopelessness, loss of self esteem (respection), feeling of worthlessness and guilt, and a wish to die" (Plumb and Holland,1977).

Chronic mild depression mixed with anxiety is often present throughout much of child's cancer treatment, depression may be showed as signs of affective disorder, separation anxiety, fear, and sleep disturbance during treatment. Although mild depression is the commonest to occur, it may occasionally progress to a sever retarded depression with vegetative symptoms (Holland et al.,1987).

Actual suicidal acts are uncommon and studies show the incidence of suicide in cancer patients to be only slightly higher than in the general population. Suicidal risk in cancer patients can be attributed to: sever depression with hopelessness, chronic poorly controlled pain, mild delirium (With impaired judgment and poor impulse control), emotional exhaustion and sense of helplessness, poor or absent family and social support, pre-existing psychiatric or

personality disorders, family history of depression or suicide (Breitbart,1989).

Depressed patients are usually treated with a combination of supportive psychotherapy and antidepressants. When depression is sever or persistent, or when interferes with medical treatment or psychosocial functioning medication should be used. Tricyclic antidepressants (mainly imipramine and amitriptyline,1-2 mg/kg/d) are beneficial for a number of symptoms in this group of patients including: anxiety, depression, sleep disturbance, withdrawn behaviour, and pain. Tricyclic are effective and safe, without hepatic, renal, and cardiac side effects, although these need to be closely monitored (Holland et al.,1987).

Major tranquilizers and Tricyclic antidepressants allay anxiety and facilitate psychotherapeutic intervention in terminal ill children. If the child is receiving cardiotoxic chemotherapy (adriamycin), it is advisable to monitor the heart function by ECG even if you are using low doses of Tricyclic antidepressant (Lederberg et al.,1989).

Delirium from CNS complication

"Delirium is a syndrome of an acute onset of agitation, impaired cognitive function, altered attention and fluctuating level of consciousness". One of the most distressing psychiatric problems among childhood cancer patients is (acute delirium) with psychosis manifestations resulting from CNS complication of the disease either by direct CNS invasion, or indirect through 2ry encephalopathy from CNS infection, radiation, chemotherapeutic agents, steroids, malnutrition of chronic illness, vascular complication, metabolic disturbance, narcotics, and organ failure. The early symptoms are often unrecognized or misdiagnosed by medical and nursing staff as symptoms of depression or poor

coping, early recognition is important since the underlying cause may be treatable complication of cancer (massie et al.,1983).

Many of the chemotherapeutic agents in cancer treatment do not cross the blood brain barrier to any significant degree and consequently have few effects on CNS. However, confusion, disorientation, and delirium have been associated with the use of methotrexate, 5-fluorouracil, the vinca alkaloids, bleomycin, cisplatin, L-asparaginase, procarbazin, cytosine arabinoside (ara-c), and prednisone (Young,1982).

Management of a cancer patients with delirium, includes environmental measures to decrease confusion guarantee patient safety (e.g., use of bed rails, night lights, repeated orientation), and an antipsychotic such as haloperidol (haldol), which is the drug of choice because of its lack of sedation and its overall safety, drug doses must be titrated against symptoms with careful monitoring of vital signs, the dose ranges from 0.5 mg in a cachectic patients to 2 mg in a stronger one (Baldessarini,1985).

2-Pre-existing psychiatric disorders aggravated by cancer

Anxiety disorders

Okasha, 1987 defined anxiety as unpleasant emotion characterized by the term worry, apprehension and dread. Any situation that theartens the well being of the organism is assumed to produce a state of anxiety. Another broad definition of anxiety is a condition of predominant tension and apprehension, experienced mentally and physically, persisting independently of external factors (Okasha,1988).

A phobia is a subtype of anxiety attached to specific object. Okasha, 1988 defined phobia as a special form of fear which:

1- Is out of the proportion to the demands of the situation.

2- Can not be explained or reasoned away.

3- Is beyond voluntary control, and.

4- Leads to avoidance of the feared situation.

Anxiety in the cancer setting can be acute (related to disease symptoms or treatment), chronic, or the consequence of disorders that antedate and are exacerbated by the patient's disease. Acute anxiety usually occurs while awaiting the diagnosis, in anticipation of major and minor tests, treatments and procedures (e.g. bone marrow aspiration / biopsy, chemotherapy, or irradiation), and on anniversary of illness-related events. The distress may lead the child to withdraw and cry or to become agitated, angry, and combative. This may make the painful procedures impossible to performed, it may also add to the problem of non- compliance. More over underlying unresolved anxiety may cause serious disturbances in psychosocial function (Lesko and Holland,1990).

Pre-existing chronic anxiety states that may be exacerbated include generalized anxiety disorders, simple phobia (e.g., claustrophobia during diagnostic scanning procedures or fear of needles), and panic states. These all may require aggressive prophylactic use of anxiolytic medication along with reassurance and support. Behavioral techniques with relaxation, desensitization, or distraction may be useful as adjuvants to pharmacological management. The medication used in the treatment of sever anxiety may be; minor tranquilizer such as diazepam and alprazolam in low doses (0.02mg/kg t.d.s.) for children undergoing painful procedures, major tranquilizer such as phenothiazines (chlorpromazine) may used at time be necessary, and Tricyclic antidepressant such as

amitriptyline and imipramine appear to be helpful in the management of chronic anxiety and if anxiety with depression (Lesko and Holland,1990).

Personality disorders

Patients with " difficult personalities" frustrate and anger those who treat them. The stress of cancer exaggerates their normally maladaptive coping strategies and they become even more difficult than usual. The disorders are recognizable by the exaggeration of common characteristics:

1- The paranoid person who is suspicious and constantly threatens litigation.

2-The obsessive person whose excessive attention to details of care is accompanied by repeated criticism.

3- The dependent person who demand care far beyond objective needs and who may be dependent on drugs

4- Borderline disorder patients who are unable to conform to rules and may disturb other patients as well as staff.

5 Histrionic person who overdramatizes symptoms and distress.

Since personality disorders are not felt by the patients to be a problem, management depends on helping staff to understand the pattern and to contain their behaviour (Lederberg et al.,1989).

Major mental illness

Schizophrenia and manic depressive (Bipolar) illness are rare in the general population and hence uncommon in cancer patients. However, when present, they require careful management to ensure the patient's co-operation with treatment and to prevent escalation of symptoms under stress (Lederberg et al.,1989).

Special issues during care and treatment of a cancer patients

The therapeutic approaches used in the Oncology setting are a combination of psychotherapeutic, pharmacological, and behavioral techniques. Psychotherapeutic approaches include:

1- Individual therapy.

2- Professionally led groups (Cancer care, family support group).

3- Self-help individual meetings (patient-to-patient volunteers).

4-Self-help support groups (Make Today Count, Candle Lighters).

Psychopharmacological management is effective for anxiety syndromes, depression, delirium, schizophrenia, manic depressive illness, pain syndromes, nausea, vomiting, and insomnia. Behavioral interventions, including relaxation, biofeedback, systematic desensitization, hypnosis, and guided imagery, are helpful for pain, anxiety during procedures, nausea and vomiting, and cancer-related eating disorders (Lesko and Holland,1990).

Enzymology

Gamma-Glutamyltransferase (EC. 2.3.2.2.;5-Glutamyl-Peptide:Amino-Acid 5-Glutamyltransferase; γ-Glutamyltransferase; GGT)

γ-Glutamyl transferase (GGT), also known as g-glutamyl transpeptidase (GGTP), catalyses the transfer of a γ-glutamyl group from a γ-glutamyl peptide to an acceptor peptide or an L-amino acid (Goldberg,1980).

γ-GGT activity has been found in the following organs (decreasing order of activity), kidney, seminal vesicles, pancreas, liver, spleen, lactating breast, and brain. In addition, it has been described in biliary epithelium, choroid plexus, small bowel epithelium, pancreatic ductular epithelium, bronchial epithelium, small bowel fluid, bile, pancreatic fluid, milk, and bronchial secretions. It is said to be absent from skeletal and cardiac muscle and mature erythrocytes. The enzyme is membrane-bound, both to plasma membrane and to endoplasmic reticulum (Rosalki,1975).

At the cellular level it is associated with the microsomal fraction and the plasma membrane (Meier et al.,1984). There are several isoenzymes of GGT, but they have found no clinical application. The enzyme occurs normally in serum at similar levels in both sexes (Wenham et al.,1985).

Its physiological role remains to be defined: some data suggest that it participates in transport of amino acids across intracellular membranes as part of the gamma-glutamyl cycle (Meister,1976), other reports suggest that its primary role is the hydrolysis of glutathione (Shaw & neuman,1979).

Clinical significance

GGT present in serum appears to originate primarily from the hepatobiliary system, and GGT activity is elevated in all forms of liver disease and is not elevated in bone disorders. It is highest in cases of intra- or post-hepatic biliary obstruction, reaching levels 5-30 times normal. GGT is more sensitive than alkaline phosphatase in detecting obstructive jaundice, cholangitis, and cholecystitis, and its rise occurs earlier and persists longer. Only moderate elevations (2-5 times normal) are seen in infectious hepatitis, and in this condition GGT determination are less useful diagnostically than other measurements of transaminases (Lum and Gambino,1972). Small increases of GGT activity are observed in patients with fatty livers, and similar but transient increases are seen in cases of drug intoxication (Moss et al.,1994).

The high serum GGT activities reported in patients with myocardial infarction with or without obvious congestive heart failure are probably related to hepatic circulatory changes (Betro et al.,1973).

In acute and chronic pancreatitis and in some pancreatic malignancies (especially if associated with hepatobiliary obstruction), enzyme activity may be 5-15 times the upper limit of normal (Goldberg and Durie,1993), so GGT is the most sensitive enzymatic indicator of hepatobiliary disease available at present; normal values are rarely found in the presence of liver disease. However, GGT is of little value in attempting to discriminate between different kinds of liver disease (Moss,1994).

Normal levels of the enzyme are seen in cases of skeletal disease, in children older than one year of age, and in healthy pregnant women-conditions in which alkaline phosphatase is elevated. Thus,

measurement of GGT levels in serum can be used to ascertain whether observed elevation of ALP are due to skeletal disease or reflect the presence of hepatobiliary disease (Tietz, 1996).

Unexplained is the finding of elevated plasma GGT activity, but not in the cerebrospinal fluid in patients with certain disorders of the central nervous system (Ewen and Griffiths., 1973). Table (IX) Showed lists conditions in which elevated plasma GGT activity has been described.

Table (IX) Diseases, other than hepatobiliary and pancreatic, in which GGT activity has been reported as increased (Goldberg and Martin.,1975) :

1. Myocardial infarction, angina
2. Diabetes
3. CNS disease
4.Renal neoplasm, renal infarction, transplant rejection, nephrotic syndrome
5. Rheumatoid arthritis
6. Chronic obstructive pulmonary disease

Increased levels of GGT are seen in the sera of heavy drinkers and of patients with alcoholic cirrhosis. In patients receiving anticonvulsant drugs such as phenytoin and phenobarbital, increased levels of the enzyme in serum may reflect induction of new enzyme activity and the toxic effects of alcohol and other drugs on microsomal structures in liver cells. Hepatic complications occurring in cystic fibrosis also lead to elevations of GGT (Moss, 1994).

High levels of GGT are present in the prostate, which may account for the fact that the activity of GGT in the sera of males is 50% higher than that in females. Prostate malignancy may at times be the source of an elevated GGT activity in the serum. The irradiation of tumors in

cancer patients may be accompanied by rise in GGT activity. However, in malignant disease in general, an increased serum GGT activity must arouse suspicion that the disease is metastatic to the liver (Tietz, 1995).

Alkaline Phosphatase

(EC. 3. 1. 3. 1; Orthophosphoric - Monoester phosphohydrolase (alkaline optimum); ALP)

Alkaline phosphatase, is a dimeric zinc-containing 34 glycoprotein which catalyses the hydrolysis of organic phosphate esters in an alkaline environment, generating an organic radical and an inorganic phosphate (Posen, 1967).

The serum alkaline phosphatase activity of the new-born infant is rather above the normal adult level, and it rises rapidly to as much as two and a half to three times the adult upper limit of normal during the first year of life. The activity falls somewhat by the end of the second year to values which are of the order of one and a half to two and half times adult levels, these elevated values are maintained approximately constant throughout childhood and early adolescence (Salz et al., 1973).

The serum activity in children and adolescents originates in osteoblast, correlates with the rate of bone growth, and is considerably elevated in both sexes, but perhaps some upturn in the serum ALP level at the onset of puberty, typically declining to adult levels during the later teenage years (Fleisher et al., 1977).

Most human tissues contain alkaline phosphatase (ALP), intestine, liver, kidney, bone, spleen, and placenta being particularity rich sources. Alkaline phosphatase is present practically in all tissues of the body, but it is especially concentrated in cell membranes of the intestinal epithelium, kidney tubules, the osteoblasts of bones, as well as the liver and placenta.

The precise metabolic functions of the enzyme are not yet fully understood (Reichling and Naplan,1988), but it appears:

(a) that the enzyme facilitates transfer of metabolites across cell membranes.

(b) that it is associated with lipid transport.

(c) that it is involved in the calcification process of bone synthesis.

Alkaline phosphatase may be subdivided into three groups see table (X). The unspecified ALP, which may be found in liver, bone, kidney, and other tissues, originates from one gene locus (Moss.,1986). It may be distinguished by electrophoretic migration, heat stability, and antigenic differences, attributable to post-translation changes (McKenna et al.,1979).

Intestinal ALP (I.ALP) arises from a second gene locus and is found in the circulation in the portion of the population who have blood group B or O and who are called secretors, given a normal diet. Another subdivision of ALP is the placental ALP (P.ALP), which arises from a third gene locus (Epstien et al., 1986).

Table (X) Probable genetic origins and expression of multiple forms of human alkaline phosphatase (Moss., 1986).

Isoenzymes encoded by independent genetic loci	Expression
"Tissue-unspecific"	*expressed widely in, for example, bone, Liver, kidney, as variants with tissue-specific properties due to differences in glycosylation of presumed identical protein.
Adult intestinal Fetal intestinal (status as product of a separate structural gene not yet completely proved, but probable).	*Localized to small-intestinal brush border; traces in other tissues, e.g. kidney. *expressed in fetal intestine up to about 30 weeks of gestation
Placental	*highly localized expression in form of numerous allelozymes in term placenta: traces in other tissues, e.g. lung, cervix.
Testicular (status as product of a separate gene or a variant of placental isoenzyme still undecided)	*Similar to placental isoenzyme except with some polycolonal antibodies and in inhibition by L-leucin e; expressed in trace amounts in testis , thymus, and other tissues.

Multiple forms of alkaline phosphatase

Alkaline phosphatase displays considerable inter- and intra-tissue heterogeneity, but there are rarely more than two or three forms in any serum specimen. The common forms of alkaline phosphatase, namely liver, bone, intestinal, placental, and kidney alkaline phosphatase are compared and contrasted. Their catalytic inhibition, antigenic, and structural properties support the conclusion that these forms are coded for at least three separate genetic loci. The liver /bone/"minor" kidney types constitute one isoenzyme group while intestine/"minor" kidney and placental types constitute two further separate isoenzyme groups (Moss.,1986).

Properties of Multiple Forms of Alkaline Phosphatase

Hepatic Alkaline Phosphatase

The distinctive properties of liver alkaline phosphatase include differences in electrophoretic mobility and solubility compared with, e.g., the phosphatase of bone (Peacock et al.,1963). Differences in these respects between bone and liver phosphatase are abolished by complete digestion with neuraminidase, and thus can be attributed to differences in the number, and possibly arrangement of sialic acid residues which dominate the charge-dependent properties of all except intestinal alkaline phosphatase (Samuelson and Moss,1978).

Liver alkaline phosphatase is also distinguishable from the enzyme of bone by its slightly, but significantly, greater stability to heat (Moss and King.,1962). Although unaffected by the removal of sialic acid, the difference in stability between bone and liver phosphatase is reduced by treatment with certain glycosidases, thus, it too can be attributed to differences in the respective carbohydrate moieties of the alkaline phosphatase glycosides (Moss et al.,1987).

Intestinal Alkaline Phosphatase

It may be presumed that the functional needs of the small intestine have led to the evolution of a specific alkaline phosphatase isoenzyme, encoded by a unique, although it is still not clear precisely what these needs are. The intestinal alkaline phosphatase is similar to, but not identical with, an even later evolutionary product, the placental isoenzyme. The intestinal and "tissue-unspecific" alkaline phosphatase differ structurally in both the protein and carbohydrate moieties, differences in protein structures presumably underlie the ability to prepare monospecific anti intestinal phosphatase antibodies (Lehmann,1975), while an obvious difference in carbohydrate components is the absence of terminal

sialic acid residues from intestinal phosphatase (Moss and Henderson, 1994).

Intestinal alkaline phosphatase reaches the circulation in large amounts via the thoracic lymph (Keiding,1964), but normally is detectable in only low activities in serum. Activities are greater, especially after eating, in individuals of blood groups B or O who are secretor-positive (Langman et al.,1966).

These genetically associated differences in intestinal phosphatase levels in serum appear to derive from differences in the rates at which the isoenzyme disappears from the circulation, indeed, since the considerable amounts of intestinal phosphatase which enter the circulation result in only minor changes in its level in serum, it may be concluded that the rate of removal (or inactivation) is the major factor in determining the activity of the intestinal isoenzyme in serum. The absence of sialic acid residues from intestinal phosphatase may facilitate its recognition and binding by galactosyl receptor sites of hepatocytes, compared with sialylated proteins such as other alkaline phosphatase (Moss et al.,1987).

This may account for the lack of correlation of serum intestinal alkaline phosphatase activity with intestinal disease, while elevated activities may be observed in chronic liver disease (Stolbach et al.,1967).

Placental Alkaline Phosphatase

The ectopic re-expression of gene determining placental alkaline phosphatase is not infrequent manifestation of malignant disease, particularly of the reproductive system. analogous ectopic expression of nonplacental alkaline phosphatase has been reported less frequently, partly because of the difficulty of distinguishing ectopic

production of the "tissue-unspecific" phosphatase from the background level of these forms originating in liver and/or bone, which in any case is often increased in malignant disease (Moss,1982).

However, the alkaline phosphatase expressed in cancer cells may be modified compared with its normal counterpart, and this may aid in the identification of an alkaline phosphatase derived from a tumor, e.g. an alkaline phosphatase apparently corresponding to a differently sialylated form of kidney alkaline phosphatase has been identified in primary and secondary renal cell carcinoma tissues (Whitaker et al.,1982).

In the case for true isoenzymes in general, those of alkaline phosphatase (i.e. the placental, intestinal and tissue-unspecific forms) are antigenically distinct. Polycolonal antisera distinguish between these three forms, although there is typically some cross- reaction between the placental and intestinal antisera which can be eliminated by absorbing-out There is little or no cross-reaction between these isoenzymes and the phosphatase of the tissue- unspecific group, but polycolonal antisera fail to differentiate at all between the tissue-unspecific phosphatase (Moss,1982).

Monoclonal antibodies are able to distinguish between the allelozymes of placental alkaline phosphatase. It is also largely through the availability of these reagents that a "placental-like" alkaline phosphatase, resembling true placental phosphatase in all but a few properties, has been recognized in trace amounts in normal testis and certain other tissues, and as the product of some tumors (Epenetos et al.,1985).

The development of monoclonal antibodies specific for bone or liver alkaline phosphatase has proved to be more difficult, as would be expected from the presumed identity of their protein components. However, reports are appearing that even this problem is beginning to yield to the exquisite sensitivity of monoclonal antibodies as structural probes, although the discrimination obtained is by no means complete (Laurell.,1985).

Alkaline Phosphatase Isoenzymes in Normal Healthy Subjects

Serum from healthy subjects almost always contains more than one form of ALP detectable by electrophoretic or selective-in activation techniques. A form identical of these respects to the main zone of liver phosphatase is almost invariably present, as is bone-type ALP, the relative proportion of the latter phosphatase may be low in many adults sera and this, together with its more diffuse and less prominent zone has led to reports that it is absent from these sera when electrophoretic analysis has been used. However, heat inactivation analysis shows that a substantial part of the normal adult serum alkaline phosphatase activity is contributed by phosphatase of presumed bone origin. The activities of bone phosphatase are markedly dependent on age. The effect of bone growth on the bone component of ALP and therefore on total ALP, in the serum of children and adolescents is well known (Moss,1982).

Liver phosphatase activity in serum increases steadily throughout life, and there is also some increase in bone phosphatase in the elderly. There is a small, statistically significant difference between the sexes at same ages with respect to the concentrations of both phosphatase, but this sex-related difference is less important in interpretation of result than are the effects of age. Variations in the amounts of these two phosphatase in diseases are of great significance in diagnostic (Moss,1982).

Isoenzyme analysis has shown that a small amount of intestinal ALP is a component of about 25% of normal serum (Whitaker,1982). The present of this isoenzyme is more probably in individuals of B or O blood groups who are secretor- positive. After eating, the concentration of intestinal ALP in the serum increases in those individuals in whom this occurs (Langman et al.,1966).

Intestinal ALP enters the blood of all individuals in large amounts by way of thoracic lymph, so that its persistence in a small amounts in the plasma of some individuals and not others suggests differences in the rate at which the isoenzyme is removed from the circulation. Some workers have shown that intestinal ALP is bound by erythrocytes of group A but is bound to a lesser degree by these of group B or O. Blood group antigens are present in the intestinal phosphatase molecule, as shown by the ability of individual preparations of the isoenzyme to react with anti-blood group antisera (Moss and Henderson,1994).

Clinical Applications

Since a raised serum alkaline phosphatase activity is characteristic of two main groups of diseases, bone diseases and hepatobiliary diseases, the most common reason for requesting alkaline phosphatase isoenzyme analysis in diagnosis is the need to distinguish between bone and liver as alternative, or co-existing, sources of the raised serum activity (Rosalki,1986).

More than half of all requests probably arise in this way, and can usually be answered by qualitative techniques, such as the visual inspection of zymograms. However, quantitative methods are needed fully to exploit the potential increase in sensitive, as well as organ specificity, offered by isoenzyme analysis. This is especially true when searching for minimum pathological changes, or in following

the progress of disease, for example in patients with metastatic cancer (Moss.,1982).

Indeed, the use of specific, quantitative determination of bone alkaline phosphatase in the diagnosis and treatment of cancers metastatic to bone is possibly the most important clinical application of alkaline phosphatase isoenzyme analysis, since even those cancers which give rise to predominantly osteoclastic metastases usually stimulate an osteoblastic response, detectable by an increase in bone phosphatase in serum provided that suitable methods of analysis are used. Furthermore, isoenzyme determinations are free from the risks associated with repeated applications of imaging techniques (Rosalki,1986).

The association of elevated (bone) alkaline phosphatase in the serum of patient with osteosarcoma was the first evidence that the tumor cells themselves produce the enzyme. The amount of phosphatase present gives valuable clinical information as to the degree of tumor destruction produced by the therapy, changes in the tumor activity may frequently by detected by this means before other clear-cut physical signs are manifested. The reduction of elevated serum alkaline phosphatase levels following chemotherapy is a valuable guide to administration of therapy (Rosen et al.,1974). The presence of Regan isoenzyme (Carcinoplacental) (heat-stable fraction) in the serum of some patients with osteosarcoma has been proposed as a prognostic factor indicating the metastatic evolution of the disease, hence, isoenzymes could be used as a marker of relapse in an early subclinical stage (Tucci et al.,1990).

The occurrence in serum of a high molecular weight, highly negatively charged form of alkaline phosphatase (the 'fast-liver'

particularly of the localized intrahepatic obstruction caused by malignant infiltration of the liver. However, this fraction seems to be neither more sensitive nor more specific in this respect than the main liver fraction itself. The high molecular weight alkaline phosphatase in serum is heterogeneous, part corresponding to enzyme bound to membrane fragments (Koinozymes) and part probably resulting from interaction between the enzyme and lipoproteins (Moss,1982).

The levels of bone and liver phosphatase in serum are governed by their rates of entry into the circulation from the cells in which they are produced. In contrast, the level of intestinal alkaline phosphatase, which reaches the circulation in large amounts via the thoracic lymph (Keiding,1964), is determined largely by rate of removal.

Usually this is extremely rapid, so that only small activities of the intestinal isoenzyme are detectable in serum, even in subjects of blood groups B or O who are secretor positive, in whom clearance of the isoenzyme seems to be somewhat delayed. Therefore, there is a poor correlation between serum intestinal alkaline phosphatase and disease, although increased levels are encountered, e.g. in chronic liver disease, especially in patients of the appropriate blood groups. Recently, a low level of fetal intestinal alkaline phosphatase in amniotic fluid at 16-18 weeks of gestation has been shown to be a reliable indication of cystic fibrosis in the fetus (Moss and Henderson,1994).

Inappropriate expression in cancer tissues of alkaline phosphatase which correspond more or less closely to normal forms of the enzyme is now a well-recognized phenomenon. Placental alkaline phosphatase ("Regan isoenzymes") are present in sera from 5-15 % of patients with cancers of all types, with higher incidences

in, for example, genital tumors, and in tumor tissues (Kellen et al.,1976).

A significant advance in the detection of placental and placental-like alkaline phosphatase in cancer has resulted from the availability of monoclonal antibodies. Like their normal counterparts, tumor- associated placental phosphatase exist as molecular variants, and the use of a panel of monoclonal antibodies is more effective than a single antibody in detecting the isoenzymes in cancer patients. Monoclonal antibodies have also facilitated the recognition of a 'placental-like' isoenzyme in trace amounts in normal tissues such as testis, and in increased amounts ('Nagao isoenzymes') in sera and tissues of patients with certain cancers, particularly dysgerminoma of the ovary and seminoma in which the isoenzyme is a particularly valuable tumor marker (Epenetos et al.,1985).

Monoclonal antibodies directed against placental phosphatase and placental-like phosphatase are opening up new possibilities of specific tumor localization and of targeted cancer therapy with drugs or radioisotopes (Moss.,1986) .

lactate dehydrogenase

(EC.1.1.1.27;L-Lactate : NAD Oxidorductase; LD;LD)

Lactate dehydrogenase (LD, LDH) is an enzyme classified as an oxidoreductase and is widely distributed in nature including mammalian and, specifically, human tissues. Its numerical classification is E.C.1.1.1.27, its systematic name is L-lactate, NAD oxidoreductase. The enzyme is involved in the final step in the glycolytic pathway (Embden-Meyerhof) and is important in glucose-metabolizing tissues such as heart and skeletal muscle (William,1979).

Lactate dehydrogenase is a hydrogen transfer enzyme that catalyzes the oxidation of L-lactate to pyruvate with the mediation of NAD as hydrogen acceptor. The reaction is reversible (equation) and the reaction equilibrium strongly favors the reverse reaction, namely the reduction of pyruvate to lactate (P → L) (McComb.,1983).

The pH optimum for the lactate to pyruvate (L → P) reaction is 8.8-9.8 and an optimal assay mixture at 30 °C contains NAD, 5mmol/L, and L-lactate, 50 mmol/L for the (P → L) assay, at 30 C and optimal concentrations for reactants of NADH, 150 mmol/L, and Pyruvate, 1.5 mmol/L. The pH optimal varies among the different source of enzyme (i.e. with the predominant isoenzymes in the sample) and depends on the temperature, as well as on substrate and buffer concentrations. The enzyme does not act on D-lactate. Its specificity extends to various alpha-hydroxy and alpha-hydroxy gamma- oxoacids, including alpha-hydroxybutyrate. Its reactivity to the latter has led to the designation of an enzyme called alpha-hydroxybutyrate dehydrogenase (HBD), wherein the indicated substrate is oxidized to alpha-oxobutyrate (Henery et. al.,1974).

LD activity can be inhibited by a variety of substances including both lactate and pyruvate (substrate inhibition). The inhibitory effect of pyruvate is greater. Other inhibitory agents include ethylen-diamine tetra-acetic acid (EDAT), oxalate, borate, oxalate and various sulfhydral reagent. Inhibitory actions differ for the various substances, such that, oxalate and borate are though to compete for lactate binding sites and oxalate for those of pyruvate. Cysteine and glutathione are known to reverse the inhibitory effect of sulfhydral reagent (Moss and Henderson, 1994).

The half-life of serum LD has been estimated to be 52 hr. This phenomenon may explain the elevations of serum LD that may be encountered for several days after the initiating mechanism has lost significance (Moss et al., 1987).

Lactate dehydrogenase isoenzymes

LD is a protein of approximately 135,000 molecular weight. It is known to exist in a series of five tetrameric isoenzymes composed of combinations of two different subunits, H (heart) and M (muscle) which are polymerized to form the five isoenzymes of LD (Latner and Skillen, 1968).

The subunits are dissimilar polypeptides and are each under the control of a specific gene. The H subunit contains relatively more aspartate and glutamate than the M subunit, which is richer in arginine and methionine so that the polymer HHHH is most electronegative and the polymer MMMM is most electropositive (Cohen et al., 1962).

The isoenzyme fractions differ in their electrophoretic mobility and may be specifically identified and quantitated. The migration of LD isoenzymes on electrophoresis can be related to the various plasma proteins. And LD isoenzyme fraction is not present in the area where albumin is found. LD-1 (heart "fast") migrates roughly in the region of alpha-1 globulin, LD-2 and LD-3 may be found with alpha-2 globulin and beta globulin, respectively. LD- 4 and LD-5 are found with gamma globulin fraction, LD-5 presenting most often at or cathodal to the point of application (Kaplan and Pesce.,1984).

An additional isoenzyme fraction has been identified migrating in the region of LD-3. It appear between LD-3 and LD-4 on electrophoresis and has been variously called LD-X_1 X_4, etc. It has been identified in testes and spermatozoa. The observation has been of little use clinically because of its rarity but has been seen in serum as a genetic variant and rarely accompanying male genital malignancy (Wilkinson,1970).

It becomes apparent that the heart fraction (LD-1) subunit is geared for the aerobic oxidation of pyruvate and, by the fact that it appears nearest the anode, indicates that it is more negatively charged. The liver, or Skeletal muscle, fraction (LD-5) is most concerned with anaerobic metabolism, reduction of pyruvate, and is most positively charged. The intervening fractions then, by subunit composition possess varying capability for the reduction of pyruvate depending upon the quantity of liver subunit present (Wilkinson,1965).

Patterns of LD Isoenzymes Distribution

All five LD isoenzymes occur in all tissues but the relative proportion for each tissue are significantly different patterns of LDH isoenzymes distribution (Wilkinson.,1970). Table (XI) Shows the different patters of LDH isoenzymes distribution.

Table (XI)The patterns of serum LD isoenzymes distribution (Wilkinson., 1970)

Origin	LD1 (H4)	LD2 (H3M)	LD3 (H2M2)	LD4 (HM3)	LD5 (M4)
Normal serum	25%	35%	20%	10%	10%
Erythrocytes	39%	56%	0%	5%	0%
Leukocytes	6%	19%	29%	19%	22%
Heart	40%	35%	20%	5%	0%
Kidney	35%	30%	25%	10%	0%
Liver	0%	5%	10%	15%	70%
Skeletal muscle	0%	0%	10%	30%	60%
Skin	0%	0%	4%	17%	79%
Lung	5%	10%	35%	35%	15%
Spleen	5%	15%	30%	30%	20%
Brain	25%	35%	30%	10%	0%
Thyroid	5%	10%	30%	30%	25%
Bladder	5%	10%	40%	35%	10%
Uterus	5%	20%	50%	20%	5%
Bowel	5%	30%	45%	10%	10%

Although complete agreement has not been reached as to how far the different patterns of aerobic and anaerobic metabolism of tissues reflect the observed characteristics of their predominate isoenzymes of lactate dehydrogenase, these studies represent important attempts to give functional significance to variations in isoenzyme composition between tissues. Moreover, it remains true that a tendency towards anaerobic patterns of metabolism in tissues is generally associated with the presence of the electrophoretically slower lactate dehydrogenase isoenzymes (Wilkinson,1965).

Clinical Applications

Owing to its widespread distribution in the tissues elevation of the total lactate dehydrogenase in the serum is generally of little value in diagnosis. In the clinical application of LD activity and/or LD isoenzyme fractionation, one is usually searching for evidenced of some form of tissue injury that results in the escape of LD into the serum. Tissue injury may range from simple anoxia to severe cellular necrosis, each inducing varying degrees of elevation of enzyme activity. Increases in total LD suggest tissue injury and the predominant isoenzyme fraction may identify the organ source (Tietz,1987).

Elevations of serum LD may be found accompanying myocardial disease, hemolytic phenomena, liver and renal injury, neoplasm, shock, infection, and other miscellaneous organ involvement. It has also been evaluated in urine, cerebrospinal fluid, and effusions. The total lactate dehydrogenase is of course necessary for the correct interpretation of isoenzyme measurements, whether these are made by electrophoresis and densitometry or by indirect methods involving substrate analogues (HBD reaction), thermal stability or selective inhibitors. It is generally more useful to express the isoenzyme composition in U/Liter rather than as percentages of the total

enzyme activity (Knotinen et al.,1974). Table (XII) reflects several clinical conditions and the resultant LD isoenzyme patterns which would be expected to be obtained.

Table (XII) : LDH isoenzyme patterns in various disease states (Tietz., 1990).

LDH isoenzyme pattern.	Disease state
a. LD1and LD 2 elevated, ratio of LD 1 / LD 2 generally greater than one	Myocardial infarct, Pernicious anemia, acute renal damage, Hemolysis, Muscular dystrophy (later stage).
b. LD 5 elevated	Liver disease, Muscular dystrophy (early stage)
c. LD 3 elevated	Neoplastic disease, Lympho-proliferative disorder
d. LD 2 and LD 3 elevated	Massive pulmonary infarctions.
e. All isoenzymes elevated	Crush syndrome.

a) Liver disease

In liver disease generally the release of lactate dehydrogenase into the serum is somewhat variable, but high activities often occur in diseases such as ineffective hepatitis, (Batsakis et al.,1974), infections mononucleosis and toxic jaundice in which hepato-cellular damage is a predominant feature. In these marked increase in the amount of LD-5 is found (Dito,1973), but there are also indications of a change in the serum isoenzyme towards LD-5, even in chronic liver diseases, such as obstructive jaundice and cirrhosis, even when the total serum lactate dehydrogenase remains within normal limits (Tietz.,1987).

b) Skeletal muscle disease

Increases in serum LD activity having been documented in both genetic and acquired disease or conditions involving skeletal muscle. Most inflammatory myopathies show mild persistent elevations of serum LD which on fractionation is predominantly LD-5. Progressive muscular dystrophy patients also present increased serum LD, which, on isoenzyme fractionation is predominantly LD-1 and LD-2 (fast fraction). This is thought to be due to a loss of LD-5 as disease progresses. Elevations of total LD (LD-5) are noted in the post-ictal state and may persist for 24-36hr following convulsive seizure (Griffiths.,1979).

c) Malignancy

Elevations of LD activity in serum have been reported with a wide variety of neoplasm, including leukemia (Zondag and Klein,1968). They are nonspecific but may be a reflection of the organ involved (e.g. metastatic lesions of liver). Isoenzyme patterns, as well as total LD activity, may also reflect tumor determinants or organ involvement. This may be exemplified by extensive metastases involving lymph nodes where not infrequently, midzone fractions (LD-2, LD-3, and LD-4) increases may be seen. Neoplasm may be expected to be rich in LD-4 and LD-5 because of the more embryonic nature of anaerobic metabolism (Tietz,1990).

Measurement of total serum LD is a nonspecific measurement of tumor activity in bone tumor (Ewing's sarcoma), LD isoenzymes 2 and 3 have been found to be specifically associated with tumor activity in lymphoma. These isoenzymes measured on presentation may be a prognostic factor in patients with Ewing's sarcoma (Rosen et al.,1981).

d) Hematological disease

Various hematological disorders are associated with increases in serum LD activity. These include any disease process associated with hemolysis disordered maturation, or increased red cell destruction Thus various acquired hemolytic anemia's as well as most megaloblastic anemia's, most prominently pernicious anemia, may present moderate to even massive (most likely full) increases of this enzyme in serum. Increases in serum LD activity are seen only irregularly with various white cell disorders, particularly the various leukemia's. Leukopenias are not associated with LD activity alterations unless other organs (e.g. liver) are secondarily involved (Batsakis et al,1974).

e) Renal disease

Persistent elevations of total serum LD may be noted accompanying severe pyelonephritis and acute tubular necrosis. Elevations are of little diagnostic significance but may provide problems in clinical interpretation if one is unaware of renal influence on LD activity. Isoenzyme fractionation will frequently provide evidence of increases in LD-2 and often LD-3 (Tietz,1990).

The serum LD is usually within the normal range in chronic renal disease associated with uremia, but there is frequently an increase after therapeutic dialysis. This enhancement of the serum activity appears to be due to the removal of a number of enzyme inhibitors such as urea and oxalate (Wilkinson et al.,1970).

PATIENTS AND METHODS

PATIENTS AND METHODS

Patients

The study was conducted on 86 cases, 30 healthy controls and 56 histologically confirmed cases with osteogenic sarcoma or Ewing's sarcoma have been observed in the National Cancer Institute, Cairo University. The blood samples were collected from patients in pediatric department of National Cancer Institute, seeking for medical care and treatment .The patients were categorized into two groups.

1- Ewing's sarcoma group

It included 21 patients diagnosed as Ewing's sarcoma with age range from 8 - 16 years, 8 cases were females (8/21 - 38%) and 13 cases were males (13/21 - 62%), the male to female ratio was 1.6:1

2- Osteogenic sarcoma group

It included 35 patients diagnosed as Osteogenic sarcoma, it was subdivided into two subgroups according to the age.

A) Osteogenic sarcoma in children :

It included 25 patients diagnosed as Osteogenic sarcoma with age range from 8 -12 years, 11 cases were females (11/25 - 44%) and 14 cases were males (14/25 - 56%) , the male to female ratio was 1.2:1 .

B) Osteogenic sarcoma in adolescent :

It included 10 patients diagnosed as Osteogenic sarcoma with age range from 13-20 years, 4 cases were females (4/10-40%) and 6 patients were males (6/10-60%), the male to female ratio was 1.5:1.

The control group

The control group included 30 cases, they are normally children, the blood samples and the psychological sheets were taken from the healthy children and adolescents from the school, the control group were devided into two subgroups according to the age.

1- The children group

It included 15 cases with age range from 8-12 years, 6 cases were females (6/15 -40%) and 9 cases were males (9/15 - 60%), the male to female ratio was 1.5 : 1.

2- The adolescent group:

It included 15 cases with age range from 13-20 years, 5 cases were females (5/15 -30%) and 10 cases were males (10/15 -70%), the male to female ratio was 2 : 1.

Collection of blood samples

Fasting blood samples were collected from patients upon admission to the hospital before the initiation of therapy, during the course of therapy (the samples were taken before the initiation of chemotherapeutic cycle to avoid the false transient rises in the serum enzymes caused by the toxicity of chemotherapy), and after the end of therapy by 6 months.

The blood samples were allowed to clot and the sera were separated by centrifugation at 3000 r.p.m. for 10 min., the serum samples were analyzed for LD and its isoenzymatic activities in the same day of sample collection because it is so delicate, sensitive and easily destructive enzyme, and determination of the other enzymatic activities were performed within 24hrs and the serum samples were kept at + 4^0 C.

Patients eligibility

All patients who had localized osteogenic sarcoma or Ewing's sarcoma presented at the pediatric unit of NCI were taken. Patients were eligible for entry in the study if they fulfilled the following criteria:

(1) Histologic confirmation of Osteogenic sarcoma or Ewing's sarcoma.
(2) No evidence of metastases.
(3) No prior history of cancer.
(4) No previous therapy.

Treatment plane for the patients

In osteogenic sarcoma , the patients treated with combined therapy (chemotherapy and surgery). The chemotherapy include 6 courses, in the form of 2 courses pre-operative (neoadjuvant chemotherapy) and 4 courses post-operative (adjuvant chemotherapy). Each cycle taken every three weeks and contains Platinol 90 mg/m^2 at day 1, and adriamycin 25 mg/m^2 for 3 days. Surgical treatment was in the form of amputation for the majority of patients and limb sparing surgery when indicated.

In Ewing's sarcoma, the patients treated with combined therapy (chemotherapy and radiotherapy). Chemotherapy include 17 courses, in the form of 4 courses before the radiotherapy, 3 courses during radiotherapy and the last 10 courses taken after radiotherapy. Each cycle will be taken every three weeks and contains VACA (vincristine 1.5 mg/m^2, cyclophosphamide 1200 mg/m^2, adriamycine 75 mg/m^2, and actinomycin D 1.15 $\mu g/m^2$). The radiotherapy treatment in a dose of 5500 cG within 3 weeks followed by booster dose of 500 cG. The whole treatment take around 52 weeks.

Methodology

All patients presented during the period 1994 - 1995 were subjected to the following studies:

1- **Detailed medical and psychological history.**
2- **Complete clinical examination.**
3- **psychological studies.**
4- **Biochemical studies.**
5- **Laboratory diagnosis in the form of :**
   a- Complete Blood Picture.
   b- Liver function tests, including:
      . Bilirubin.
      . SGOT, SGPT.
      . Alkaline phosphatase.
      . G. G.T.
   c- Kidney function tests:
      . Blood urea.
      . Serum creatinine.

6- **Radiological diagnosis in the form of**
   . Local X- ray.
   . Chest X- ray (PA. & lateral views).
   . CT scan of the chest.
   . Radionucloid Bone Scan.
   . Abdominal Sonography when indicated.
   . CT local or Magnetic Resonance Imaging (MRI) when limb salvage is recommended.

Psychological studies

All cases with Ewing's sarcoma, Osteogenic sarcoma and the control groups were subjected to the following:

1- Detection of the personality traits using Junior Eysenk Personality Questionnaire.

2- Detection of the anxiety using the Children Anxiety Scale (CAS).

3- Detection of associated psychiatric disorders using a sheet of complete psychological history.

Psychological Tools Applied In This Study

1- The Children Anxiety Scale (CAS)

This scale was designed by Abd-Hamied and El-Nail (1991). It is an Arabic version drived from Children's Manifest Anxiety Scale (CMAS) by McCandless, Castaneda and Palermo. This scale consists of 36 statements and measures all the aspects of the anxiety which include the following :

1) Somatic features.
2) Physiological features.
3) Motor features.
4) Emotional features.
5) Mental features, and
6) Social features.

Each category could be assessed by 6-statements. The child after reading each statement by himself or by the interviewer answers either yes or no, if the answer is yes he scores 1 and if it is no he scores 0. The total degree ranged between 0-36, the cut off point of the scale is 18 and above 18 the child is considered to have high anxiety as a state.

2- Eysenk Personality Questionnaire (EPQ)

Eysenk personality questionnaire is a development of various earlier personality questionnaire, it differs from latest of those (The Eysenk Personality Inventory) by including an additional scale and hopeful by having made certain improvements in the other scales. The Junior Eysenk Personality Questionnaire (J.E.P.Q.) measures 4 aspects of the personality extroversion, neuroticism, psychoticism and lie scale. This J.E.P.Q. consists of 96 questions inorder to measure these 4 aspects of the personality. Instructions for literate subjects are printed on each copy of the J.E.P.Q. these should be read aloud to groups of subjects or be read silently by subject tested individually. When the questionnaires are collected after completion, care should be taken to check that all questions have been answered.

The questionnaires are scored by using the appropriate scoring sheet there are four keys for the junior version of the E.P.Q.

Biochemical studies

1- Determination of serum γ- glutamyl transferase activity

This was performed using the method recommended by the Committee on Enzyme of the Scandinavian Society for Clinical Chemistry and Clinical Physiology (1976).

Gamma glutamyl transferase (GGT) catalyses the transfer of the glutamyl group from γ- glutamyl peptides to another peptide or L-amino acids, or to water. The determination of GGT was considerably simplified by the introduction of the directly chromogenic substrate γ- glutamyl-4- nitro -anilide with the use of the glycylglycine as acceptor molecule. The rate of production of 4-nitroaniline was monitored at 405 nm at 37 ^{0}C. This was performed by already commercially available kit from Bio-Merieux Company, France.

2-Determination of Serum alkaline phosphatase and its isoenzymatic activities

A) Total Alkaline Phosphatase Activity in serum

The alkaline phosphatase activity was measured kinetically by p-nitro-phenyl phosphate as substrate in diethanolamine buffer, using the Scandinavian method (1974)

The rate of production of p-nitro-phenol was continuously monitored at 405 nm., with the recording photometer 4010 at 37 C. The test was performed using already available kit from Bio-Merieux company, France.

B) Inhibition of Serum ALP Activity by L-Phenylalanine

The most commonly used selective inhibitor in routine methods for alkaline phosphatase analysis is L-phenylalanine which inhibits intestinal and placental alkaline phosphates, substantially more than liver and bone alkaline phosphatase.

Usually, the activity is measured in the presence of 5mmol/L (0.925 gm/L) L-phenylalanine dissolved in diethanolamine substrate buffer. The percentage of inhibition was determined by the equation,

$$\% \text{ inhibition} = \frac{T - A}{T} \times 100$$

(Fishman et al., 1965)

In the absence of the heat stable ALP (placental or carcinoplacental isoenzymes) and if the I.ALP appeared on the electrophoretogram (see the demonstration of ALP isoenzymes in serum by electrophoresis in the next pages), it could be calculated by the equation.

$$\text{Intestinal (ALP) activity} = \frac{0.85\,T - A}{0.25}$$

(Whitaker., 1982)

Where:- (T) Total ALP in the absence of L.phenylalanine.
(A) Total ALP in the presence of L.phenylalanine.

C) Serum ALP Isoenzyme Activities by Heat inactivation

Quantitative heat inactivation analysis of alkaline phosphatase were done as described by Moss and Whitby,1975. Serum was thawed and kept at + 4 °C overnight. before analysis in order to allow full recovery of the alkaline phosphatase activity (Brojer and Moss,1971).

Heating of serum was performed in a small-walled glass tube in a large-capacity water-bath with rapid circulation of water and controlled by contact thermometer at exactly 56 °C.

One tube per serum pre-heated in the bath and one pre-cooled in ice, with all tubes well immersed. At time zero, 100ul of serum were added to the pre-heated tube. After exactly 15 minutes of incubation 50 UL of serum were removed from the tube and rapidly transfer to the pre-cooled tube. After 25 minutes of incubation the tube was removed from the bath to the ice container.

The residual activities after heating at 56 °C for 15 and 25 minutes were measured kinetically at 37 °C by the Scandinavian recommended method and the liver alkaline phosphatase was calculated from the equation,

Log liver ALP activity = 2.5 Log (a) - 1.5 Log (b)

Where:- (a) and (b) are residual activities (IU/L)
after 15 and 25 minutes respectively.

Then the liver isoenzyme activity was subtracted from the total activity to obtain the bone isoenzyme activity. In all cases a second serum sample was heated in Dryer tube in water-bath at 65 °C for 30 min in order to inactivate all alkaline phosphatase

except the placental type phosphatase (which is the only isoenzyme stable at this temperature) and then cooled in ice.

If the placental type of alkaline phosphatase was present, its activity should be measured and subtracted from the total activity and residual activities after 15 and 25 minutes at 56 °C, and the previously mentioned equation for calculation of the liver isoenzyme was applied (Fishman et al.,1972).

D) Demonstration of ALP Isoenzymes by Electrophoresis

Alkaline phosphatase isoenzyme patterns were performed on vertical 7% (W/V) polyacrylamide slab gel electrophoresis in tris-borate buffer pH 9.5 (0.38 mol/L tris containing 0.5 mmol/L Mg CI2 and adjusted by 2% boric acid till pH 9.5) using a method modified from that of Akroyd,1967.

40 UL of the stained serum samples with bromophenol for control and tested samples were applied to the slits. Another sample from each serum was incubated with neuraminidase for 30 minutes at 37 °C and then applied on the gel. After short time of incubation with this enzyme, it partially digests the sialic acid residue from both liver and bone alkaline phosphatase which permits better separation between the two isoenzymes on the electrophoretogram as recommended by Moss and Edwards,1984.

The slab was placed immediately in the electrophoretic chamber which was cooled to 8-10 °C and 70 volts were applied till the samples entered the gel, then the volt was raised to 130 volts. When the stained serum albumin ran approximately 8 Cm from the application slit (about 1- hr) the run was stopped.

The substrate staining solution (1-naphthyl phosphate (sodium salt) 4mmol/L and fast blue BB 3mmol/L in tris borate buffer (pH 9.5) was poured over the gel slab and incubated at room temperature for 45 minutes. Then the gels were washed in fresh methanol/acetic acid/water (5:1:5) solution. The isoenzyme bands were detected by visual inspection.

3-Determination of serum total lactate dehydrogenase and its isoenzymatic activities

A)- Total LD Activity in Serum

Lactate dehydrogenase activity was estimated by the kinetic procedure according to the Scandinavian recommended method, 1974 using pyruvate as substrate at 37°C. The reaction was monitored at 340 nm. The test was performed by using the commercially available kit from Bio-Merieux Company, France.

B) Serum LD Isoenzymes by Electrophoresis

Lactate dehydrogenase Isoenzyme patterns were performed on agarose gel electrophoresis in barbital buffer at pH 8.6, by the standard method of Rosalki, 1974.

20 ml of 1% agarose gel in barbital buffer pH 8.6 was prepared and poured on glass plate (10x10cm). The slit marker was put immediately in the boiling agarose which solidified after 15 minutes. After that the slit marker was removed from the plate, 5ul of stained serum sample with bromophenol blue of control and tested samples were applied to the slit. The plate was placed immediately in the electrophoretic chamber which was cooled to 8-10 °C and 10 V/Cm were applied. When the stained serum albumin ran approximately 5cm, the run was stopped.

The substrate-staining solution (L lactate 2mmol/L, NAD 10mmol/L, Nitroblue tetrazolium 10 mmol/L, tris buffer pH 8.0, 15 mol/L, phenazine methosulphate 10 mmol/L) was poured over the plate and incubated at 37 °C for 90 minutes. Then the plate was pressed for 30 min, dried in the oven. The bands of the isoenzymes were scanned at 570 nm.

Statistical procedure

The data collected was introduced to a Mackintoch computer (quatra 700), statistical analysis of the whole work was done using the following statistical procedures according to (Knapp & Miller, 1992).

The mean

The arithmetic average

$$\text{The mean "M"} = \frac{\Sigma X}{N}$$

Where, Σ = the sum of,
X = individual values,
N = the number of cases.

Standard deviation (SD)

It is the square root of the variance. It gives an estimate of the average deviation around the mean.

$$SD = \sqrt{\frac{\Sigma X^2 - \frac{(\Sigma x)^2}{n}}{n-1}}$$

Where ΣX^2 = the sum of squares of the individual values, and
$(\Sigma X)^2$ = the square of the sum of the individual values.
n = the number of cases.

T. test

A statistical procedure designed to compare two sets of observations.

$$T = \frac{M1 - M2}{\sqrt{\frac{(SD1)^2 + (SD2)^2}{N1 + N2}}}$$

Where, M1 = the mean of the first group.
M2 = the mean of the second group.
SD1 = the standard deviation of the first group.
SD2 = the standard deviation of the second group.
N1 = number of cases of the first group.
N2 = number of cases of the second group.

Chi-square : X^2

$$X^2 = \sum \frac{(O-E)^2}{E}$$

Where, O = the observed value.
E = the expected value.

RESULTS

Results

Fifty six patients with a histopathologic diagnosis of Osteogenic sarcoma or Ewing's sarcoma presented at the pediatric oncology department of the National Cancer Institute, Cairo University, from April 1994 till May 1996, were studied.

Patients characteristics

Age distribution

In osteogenic sarcoma the age of the patients included into this study ranged between 8 and 20 years with 25 of the 35 patients between the ages of 8 and 12 years, the mean age was 10.2 years, and 10 patients between the ages of 13 and 20 years, the mean age was 15.2 years.

In Ewing's sarcoma the age of the patients included into this study ranged between 8 and 16 years. the mean age was 12 years.

Sex distribution

In osteogenic sarcoma out of 25 patients 11 were females (11/25 - 44%) and 14 were males (14/25 - 56%), the male to female ratio was 1.2 : 1. And out of 10 patients 4 were females (4/10 - 40%) and 6 were males (6/10 - 60%), the male to female ratio was 1.5 : 1.

In Ewing's sarcoma out of 21 patients 8 were females (8/21 - 38%) and 13 were males (13/21 - 62%), the male to female ratio was 1.6 : 1.

All patients were subjected to, a complete history taken, a physical examination, and several chemical laboratory analyses. The primary tumor was evaluated with plain radiographs, bone scan, angiogram, computed tomography scan, or magnetic resonance imaging in three cases where limb salvage surgery were done. The diagnostic procedures carried out in the initial work up to exclude the presence of metastases.

Histopathology

The pathological classification of osteogenic sarcoma used in this study is that proposed by Dahlin and Unni in (1977). All patients had high-grade osteogenic sarcoma. In the 25 children patients, 14 had osteoblastic (14/25% - 56%), 6 had Chondroblastic (6/25-24%), 3 had fibroblastic (3/25-12%) and 2 had telangiectatic osteogenic sarcoma (2/25-8%).

In the 10 adolescent patients, 6 had osteoblastic (5/10-50%), 3 had Chondroblastic (2/10-20%), only 1 patient had fibroblastic (1/10-10%) and 1 had telangiectatic osteogenic sarcoma (1/10-10%).

All patients with Ewing's sarcoma were diagnosed histopathologically as small round cell tumors with its specific histologic criteria for diagnosis.

Primary site of the disease

In osteogenic sarcoma among children the site of the primary lesion was the distal femur in 10 patients (10/25-40%), proximal tibia in 8 patients (8/25-32%), proximal humerus in 3 patients (3/25-12%), and two in fibula (2/25-8%) and one in the following sites: distal tibia, and talus (1/25-4%).

In adolescent patients the site of the primary lesion was the distal femur in 5 patients (5/10-50%), proximal tibia in 3 patients (3/10-30%), and one in each of the following sites: scapula and mandible (1/10-10%).

In Ewing's sarcoma, the site of the primary lesion was the femur in 9 patients (9/21-43%), tibia in 5 patients (5/21-24%), fibula in 3 patients (3/21-14%), humerus in 3 patients (3/21-14%) and the pelvis in one patient (1/21-4%).

Symptomatology

In osteogenic sarcoma, pain and swelling at the site of the primary lesion were recorded from one month to one year prior to the diagnosis in all patients. Sixty five percent of the patients presented within 2 to 4 months of the onset of symptoms (23/35 -65.7%). Pathological fracture was recorded in 14% (5/35) of our patients. The primary lesion varied in size from less than 3x3x5 cm to 16x14x9 cm.

In Ewing's sarcoma, pain and swelling at the site of the primary lesion were recorded from few weeks to 2 months prior to the diagnosis in all patients. Most of the patients (15/21-71%) have fever, anemia, leukocytosis and increased sedimentation rate on admission to hospital, pathological fracture was recorded in one patient (1/21-4.7%).

Table (1) : Osteogenic sarcoma , patients characteristics:-

	Osteosarcoma in children n = 25 pt.		Osteosarcoma in adolescent n = 10 pt.	
Age:-				
Range	8-15	years	16-24	years
Mean	12.68	years	17.8	years
Sex:-				
Male	14	56 %	6	60 %
female	11	44 %	4	40 %
Male to female ratio	1.2 :1		1.5 :1	
Primary site:-				
Distal femur	10	40 %	5	50 %
Proximal tibia	8	32 %	3	30 %
Proximal humerus	3	12 %	0	-----
Fibula	2	8 %	0	-----
Distal tibia	1	4 %	0	-----
Foot bone	1	4 %	0	-----
Scapula	0	-----	1	10 %
Mandible	0	-----	1	10 %
Histopathology:-				
Osteoblastic	14	56 %	6	60 %
Chondroblastic	6	24 %	3	30 %
Fibroblastic	3	12 %	1	10 %
Telangiectatic	2	8 %	1	10 %
Symptomatology:-				
Onset of symptoms				
From 2 to 4 m.	17	68 %	6	60 %
From 1 m.To 1 y.	8	32 %	4	40 %
Pain	All		All	
Swelling	All		All	
Pathological fraction	5/35 - 14% of all		5/35 - 14% of all	
Laboratory tests:-				
Hb%	10.5 - 13.8g/l		9.5 - 13.8g/l	
WBCs	3800 - 11,500		3,500 - 14,500	
ESR	12/45		20/55	
Bilirubin	0.8 - 1.2		0.3 - 0.9	
SGOT	12 - 38		11 - 42	
SGPT	9 - 40		9 - 38	
Urea	21		18	
Cereatinine	0.3 - 1 mg%		0.3 - 0.9 mg%	

Table (2) Patients characteristics of Ewing's sarcoma

total no. of patients :	21 patients.	
Age :		
Range	8 - 16	years
Mean	12	years
Sex :		
Male	13	62 %
Female	8	38 %
male to female ratio	1.6 : 1	
Primary Site :		
Femur	9	43 %
Tibia	5	24 %
Fibula	3	14 %
Humerus	3	14 %
Pelvis	1	4 %
Symptomatology :		
Onset of symptoms	From 3 weeks till 2 months	
Pain	All	
swelling	All	
Leukocytosis	15	71 %
Anemia	15	71 %
Fever	15	71 %
Elevated ESR	15	71 %
Pathological fracture	1	4.7 %
Laboratory tests:		
Hb%	ranged from 14.5 to 15.5 g/l	
WBCs	ranged from 4,300 to 12,600	
ESR	18 / 68	
Bilirubin	0.3 - 1	
SGOT	3 -37	
SGPT	5 - 40	
Urea	8-25	
Cereatinine	0.3 - 1.2	

Results of psychiatric disturbance in children with bone cancer

Table (3) Frequency of anxiety symptoms in the control group.

Variable	Anxiety +ve		Anxiety -ve		Total	Chi^2	P
	no.	%	no.	%			
All child.	3	10%	27	90%	30		
Boys	1	5.2%	18	94.8%	19		
Girls	2	18.2%	9	81.8%	11	1.3	> 0.05

Table (3) shows the frequency of anxiety symptoms in the control group, and the frequency difference between boys and girls was not statistically significant (P>0.05)

>0.05 : Non significant

<0.05 : Significant

<0.01 : Highly significant

<0.001 : Very highly significant

Table (4) Frequency of anxiety symptoms in patients with Ewing's sarcoma at the different stages of therapy

Variable	Anxiety +ve		Anxiety -ve		Total	Chi^2	P
	no.	%	no.	%			
C. group	3	10%	27	90%	30		
At diagnosis	9	38.1%	12	61.9%	21	7.4	<0.01
At admission	12	63.2%	7	36.8%	19	15.5	<0.001
At end of TTT	7	43.8%	9	56.2%	16	6.9	<0.01

Table (4) Shows the frequency of anxiety symptoms in patients with Ewing's sarcoma during the different stages of therapy, there were highly significant increase in the frequency of anxiety symptoms in the patients at diagnosis of the disease and at the end of therapy, while at admission to the hospital the frequency of anxiety symptoms increased and it was very highly significant increase as compared to the control group.

There were no statistically significant difference in the frequency of the anxiety symptoms between the different stages of therapy (Chi^2 =2, P>0.05), but the rate of anxiety positive at admission (63.2%) was higher than the rates in the other stages of therapy (38.1% and 43.8% respectively).

Table (5) Frequency of anxiety symptoms in patients with osteogenic sarcoma at the different stages of therapy

Variable	Anxiet +ve		Anxiety -ve		Total	Chi^2	P
	no.	%	no.	%			
C. group	3	10%	27	90%	30		
At diagnosis	14	40%	21	60%	35	7.5	<0.01
At admission	20	64.5%	11	35.5%	31	19.3	<0.001
At end of TTT	10	38.5%	16	61.5%	26	6.3	<0.05

Table (5) Shows the frequency of anxiety symptoms in patients with osteogenic sarcoma at the different stages of therapy, there were highly significant increase in the frequency of anxiety symptoms at the diagnosis, very highly significant increase at the admission to the hospital and a significant increase at the end of therapy as compared to the control group.

There were no statistically significant different in the frequency of anxiety symptoms between the different stages of therapy (Chi^2= 5.2, P>0.05) , but the rate of anxiety positive at the admission (64.5%) was higher than the rate in the other stages of therapy (40% and 38.5% respectively).

Table (6) Sex factor affecting the production of anxiety symptoms in all patients with Ewing's sarcoma or osteogenic sarcoma

Variable	Anxiety +ve		Anxiety -ve		Total	Chi^2	P
	no.	%	no.	%			
Control boys	1	5.2%	18	94.8%	19		
Ewing's boys	4	30.8%	9	69.2%	13	3.81	<0.05
Osteogenic boys	6	30%	14	70%	20	4.05	<0.05
Control girls	2	18.2%	9	81.8%	11		
Ewing's girls	5	62.5%	3	37.5%	8	3.9	<0.05
Osteogenic girls	8	53.3%	7	46.7%	15	3.3	>0.05

Table (6) Shows the difference in the frequency of anxiety symptoms between boys and girls in patients with Ewing's sarcoma or osteogenic sarcoma.

There were significant increase in the anxiety symptoms among boys and girls ($P<0.05$) with Ewing's sarcoma as compared to the control group, and no statistical significant different between boys and girls ($Chi^2= 2.03$, $P>0.05$) in Ewing's sarcoma patients.

There were significant increase ($P<0.05$) in the anxiety symptoms among boys with osteogenic sarcoma, while the girls there was no significant increase ($P>0.05$) as compared to the control group, and there were no statistically significant different between boys and girls ($Chi^2= 1.94$, $P>0.05$) in osteogenic sarcoma patients.

Table (7) Relation of neurotic traits in patients with osteogenic sarcoma and anxiety symptoms.

The traits	Anxiety positive				Anxiety negative				Chi²	P
	present		absent		present		absent			
	no	%	no	%	no	%	no	%		
Nervous	10	71.4%	4	28.6%	6	28.6%	15	71.4%	6.2	< 0.05
Nail biting	8	57.1%	6	42.9%	5	23.8%	16	76.2%	4	< 0.05
Bed wetting	4	28.6%	10	71.4%	2	9.5%	19	90.5%	2.1	> 0.05
Nightmares & terror	9	64.3%	5	35.7%	3	14.2%	18	85.8%	9.5	< 0.01
Stuttering	2	14.3%	12	85.7%	2	9.5%	19	90.5%	0.2	> 0.05
Thumb suckling	5	35.7%	9	64.3%	2	9.5%	19	90.5%	3.6	> 0.05
Eating trouble	11	78.6%	3	21.4%	5	23.8%	16	76.2%	10.2	< 0.01
Somnambulism	3	21.4%	11	78.6%	1	4.8%	20	95.2%	2.3	> 0.05
Encopuresis	0	0	14	100%	1	4.8%	20	95.2%	0	0

Table (7) : Shows Nervousness, Nail biting, Nightmares & terrors and Eating troubles all were significant among anxiety positive group. Bed wetting, Thumb suckling, Stuttering, Somnambulism and Encopuresis were not significant. From the above data, most of patients with anxiety symptoms experienced one or more neurotic traits at the same time. From the above data, most of patients with anxiety symptoms experienced one or more neurotic traits at the same time.

Table (9) The incidence of neurotic traits in all patients with Ewing's sarcoma or osteogenic sarcoma

The trait	Ewing's sarcoma (21)		Osteogenic sarcoma (35)		Chi^2	p value
	No.	%	No.	%		
Nervous	11	52.4%	16	45.7%	0.2	>0.05
Eating troubles	9	42.8%	16	45.7%	4.3	> 0.05
Night mares & terrors	9	42.8%	12	34.3%	0.4	> 0.05
Nail biting	7	33.3%	13	37.1%	0.1	> 0.05
Thumb suckling	5	23.8%	7	20%	0.1	> 0.05
Bed wetting	2	9.5%	6	17.1%	0.6	> 0.05
Stuttering	0	0	4	11.4%	-----	-----
Somnambulism	0	0	4	11.4%	-----	-----
Encopuresis	0	0	1	2.9%	-----	-----

Table(9) Shows the incidence of the neurotic traits in all patients with Ewing's sarcoma or osteogenic sarcoma in order of frequency, nervousness, eating troubles, bed wetting, night mares & terrors and thumb suckling were the most common neurotic traits affecting the patients.

The difference in the neurotic traits between Ewing's sarcoma patients and osteogenic sarcoma patients were not significant (P>0.05).

Table (8) Relation of neurotic traits in patients with Ewing's sarcoma and anxiety symptoms.

The traits	Anxiety positive				Anxiety negative				Chi 2	P
	present		absent		present		absent			
	no	%	no	%	no	%	no	%		
Nervous	8	88.9%	1	11.1%	3	25%	9	75%	8.4	< 0.01
Nail biting	5	55.6%	4	44.4%	2	16.7%	10	83.3%	1.9	> 0.05
Bed wetting	1	11.1%	8	88.9%	1	8.3%	11	91.7%	4.6	> 0.05
Nightmares & terrors	7	77.8%	2	22.2%	2	16.7%	10	83.3%	7.8	< 0.01
Stuttering	0	0	9	100%	0	0	12	100%	___	___
Thumb sucking	3	33.3%	6	66.7%	2	16.7%	10	83.3%	0.8	> 0.05
Eating troubles	7	77.8%	2	22.2%	2	16.7%	10	83.3%	7.8	< 0.05
Somnambulism	0	0	9	100%	0	0	12	100%	___	___
Encopuresis	0	0	9	100%	0	0	12	100%	___	___

Table (8): Shows Nervousness, Nightmares & terrors, and Eating troubles were significant among anxiety positive group. Nail biting, Bed wetting, Stuttering, Thumb suckling, Somnambulism and Encopuresis were not significant.

Table (10) Junior E. P. Q. in Ewing's sarcoma patients and Control

The traits	Boys		Girls	
	Cases (13)	control (19)	cases (8)	control (11)
Neurocitism No. %	8 61.5% Chi ^{2}= 5.39	4 21.1% P < 0.05	5 62.5% Chi ^{2} = 3.9	2 18.2% P < 0.05
Extroversion No. %	2 15.4% Chi ^{2} = 0.9	2 10.5% P > 0.05	4 50% Chi ^{2} =3.99	1 9.1% P < 0.05
Psychoticism No. %	0 0% Chi ^{2} = ---	1 5.3% P ---	1 12.5% Chi ^{2} = ----	0 0% P ---
Lie score No. %	5 38.5% Chi ^{2} = 2.5	3 15.8% P > 0.05	5 62.5% Chi ^{2} = 3.9	2 18.2% P < 0.05

Table (10) Shows the personality make up of both the patients with Ewing's sarcoma and the control groups.

*Regarding neuroticism was significantly high in both boys (61.5%) and girls (62.5%) than the control (P<0.05). The difference between boys and girls was not statistically significant (P>0.05).

*Regarding the extroversion, it was significantly higher in girls (50%) than the boys (15.4%) and the control group (P< 0.05), the difference between girls and boys was not statistically significant (P>0.05).

*Regarding the psychoticism, it was low in both girls and boys and no difference between them as compared to the control group.

*Regarding the lie scale, it was significantly higher in girls (62.5%) than both boys (38.5%) and the control group (P<0.05), the difference between boys and girls was not statistically significant (P>0.05).

Table (11) Junior E. P. Q. In osteogenic sarcoma patients and control

The traits	Boys		Girls	
	Cases (20)	control (19)	cases (15)	control (11)
Neurocitism No. %	11 55% Chi ^{2}= 4.7	4 21.1% P < 0.05	9 60% Chi ^{2}=4.5	2 18.2% P < 0.05
Extroversion No. %	8 40% Chi ^{2} =4.44	2 10.5% P < 0.05	7 46.7% Chi ^{2}=4.2	1 9.1% P < 0.05
Psychoticism No. %	1 5% Chi ^{2} =0.001	1 5.3% P > 0.05	0 0% Chi ^{2}= ---	0 0% p ---
Lie No. %	5 25% Chi ^{2}=0.877	3 15.8% P > 0.05	9 60% Chi ^{2} = 4.5	2 18.2% P < 0.05

Table (11) Shows the personality make up of both the patients with Ewing's sarcoma and the control groups.

*Regarding neuroticism as significantly in both boys (55%) and girls (60%) than the control (P< 0.05). The difference between boys and girls was not statistically significant (P>0.05).

*Regarding the extroversion, it was significantly higher in both boys (40%) and girls (46.7%) than the control group (P< 0.05), the difference between girls and boys was not statistically significant (P>0.05).

*Regarding the psychoticism, it was low in both girls and boys and no difference between them as compared to the control group.

*Regarding the lie scale, it was significantly higher in girls (60%) than both boys (35%) and the control group (P<0.05), the difference between boys and girls was not statistically significant (P>05).

Table (12) The frequency of psychiatric disorders in patients with Ewing's sarcoma or osteogenic sarcoma

The disorder	Ewing's sarcoma (21) No.	%	Osteogenic sarcoma (35) No.	%	Total
A-Behavior and emotional disorder.					
1-Hyperkinetic disorders	3	14.3%	8	22.8%	11
2-Conduct disorders	2	9.5%	7	20%	9
3-Emotional disorders with onset specific to childhood	5	23.8%	6	17.1%	11
*Separation anxiety disorder	2	9.5%	4	11.4%	8
*Phobic anxiety disorder	2	9.5%	3	8.5%	5
*Social anxiety disorder	6	28.6%	10	28.6%	16
4-Tic disorders	2	9.5%	6	17.1%	8
5-Others	0	0	4	11.4%	4
*Non organic enuresis	11	52.4%	20	57.1%	31
*Stuttering					
*School problems					
B-Behaviorsyndromeassociated with physiological disturbance					
1- Eating disorders	9	42.9%	16	45.7%	25
2- Sleep disorders	9	42.9%	16	34.3%	24
3- Speech disorder	0	0%	4	11.4%	4
C-Mood disorders					
1- Depressive episode	3	14.3%	5	14.3%	8
2- Dysthymia	4	19%	6	17.15	10

Table (12): Shows the frequency of different psychiatric disorders among patients with Ewing's sarcoma or osteogenic sarcoma. From this table it is clear that more than one diagnosis could be present at the same time.The behavioral and emotional disorders were the most prevalent disorders among the patients.

Figure (1)

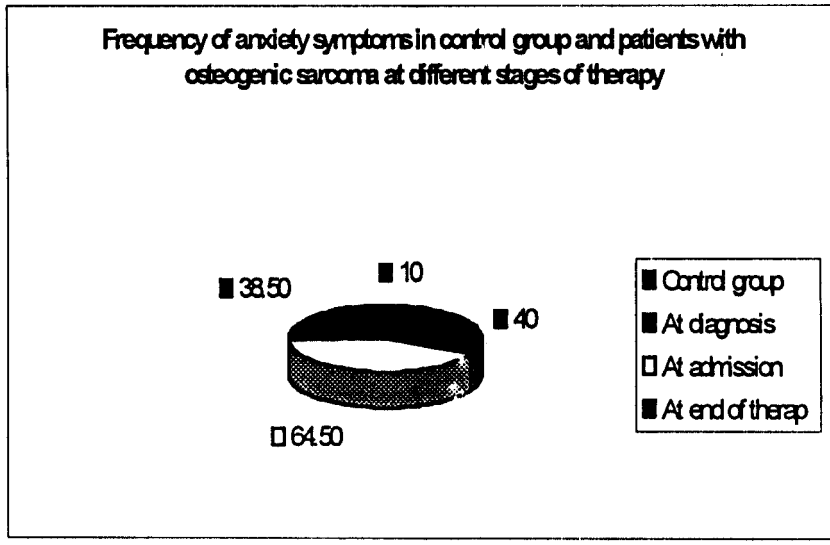


Figure (2)

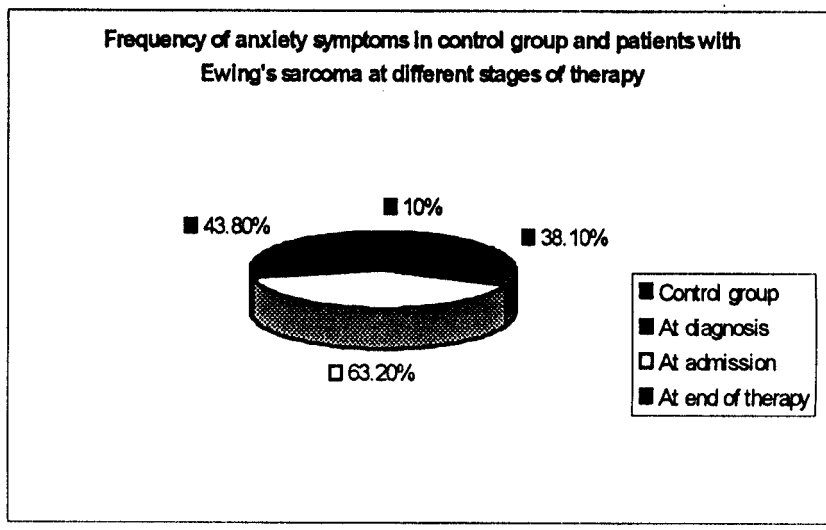


Figure (3)

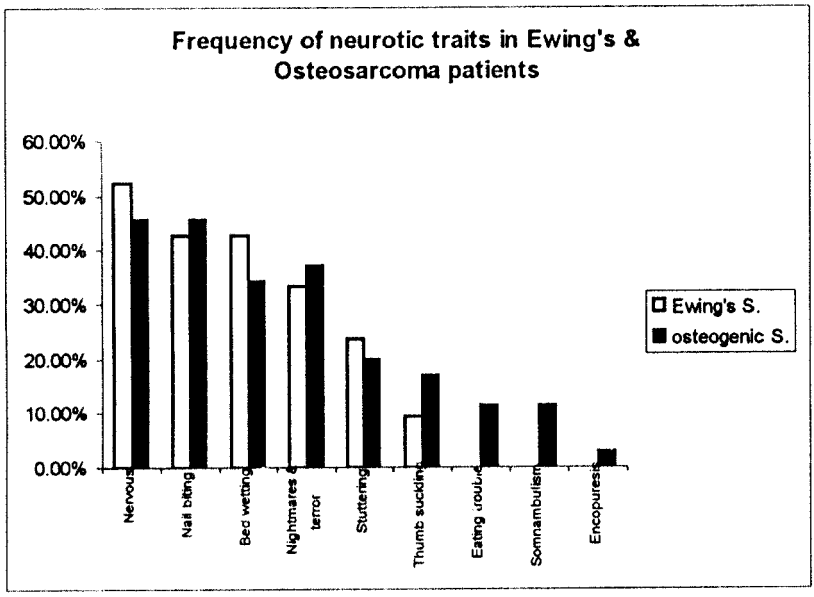


Figure (4)

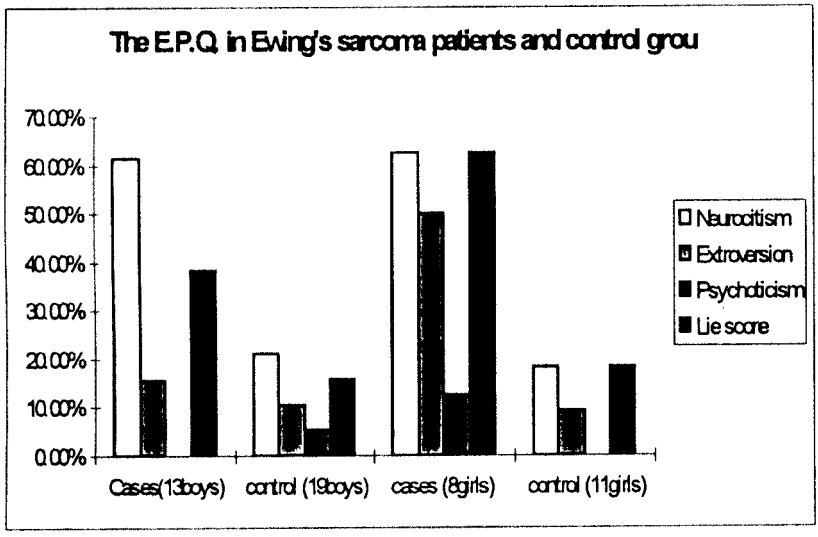
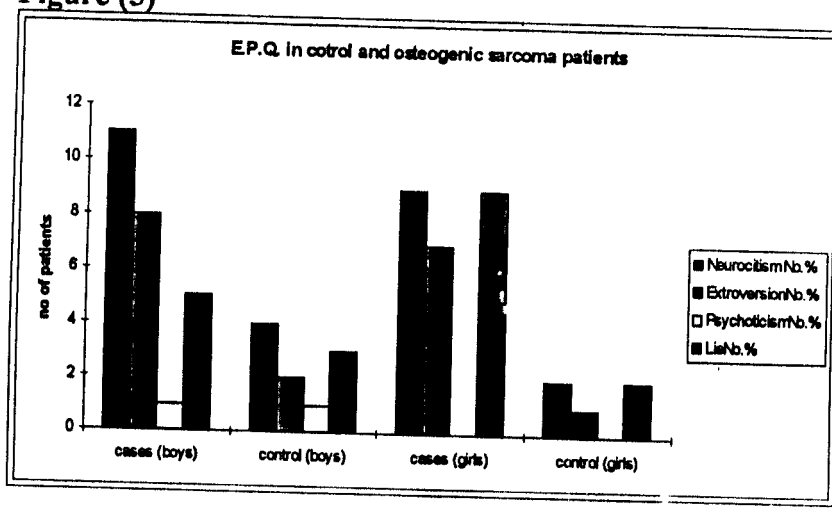


Figure (5)


E.P.Q. in cotrol and osteogenic sarcoma patients
no of patients
12
10
8
6
4
2
0
cases (boys)
control (boys)
cases (girls)
control (girls)
NeuroticismNo.%
ExtroversionNo.%
PsychoticismNo.%
LieNo.%

Results of Serum lactate dehydrogenase and its isoenzymatic activities

A)- In control groups

1- In healthy children group (Cc)

The serum level of T.LDH activity showed a normal range as adolescent healthy group (serum range 245-350 U/L), and the mean value showed a non significant variation (P>0.05) as compared to the mean value of healthy adolescent group, while regarding to the LD isoenzymes activity the data revealed that, there were non significant variations (P>0.05) as compared to mean value of adolescent healthy group. The data shown in tables (13,22,23).

2- In healthy adolescent group (Ca)

The serum level of T.LDH activity showed a normal range as children healthy group (serum level ranged between 269-390 U/L) and the mean value showed a non significant variation (P>0.05) as compared to the mean value of children in healthy group, while regarding to the LD isoenzymes activity the data revealed that, there were non significant variation (P>0.05) as compared to mean value of children healthy group.The data shown in tables (14,22,23).

B)- In Ewing's sarcoma group

1- In the pre-treatment stage (GA) 21 cases

The serum level of T.LDH activity in seven cases (7cases out of 21 cases which represent 33%) showed a normal range as healthy children control group (serum level ranged between 279-329 U/L) and the mean value showed a non significant variation as compared to the mean value of healthy children group. While the serum level of T.LDH activity in the pre-treatment stage of all cases (21 cases) showed a higher values than the upper reference

limit of healthy children control group (serum level ranged between 279-813 U/L). The mean value showed a very high significant increase ($P<0.0005$) as compared to the mean value of healthy children control group.

Regarding to the LD isoenzymatic activity the data revealed that, there are very high significant increase ($P<0.0005$) in the isoenzymes LD-2, LD-3, and high significant increase ($P<0.005$) in LD-4 as compared to the healthy children control group. The data shown in tables (15,22,23).

2- In the pre- radiotherapy stage (GB) 19 cases

The serum level of T.LDH activity in nineteen cases (19 cases out of 21 cases which represent 90% of all cases, two cases are out of the study, one case died during chemotherapy from chest infection and the other one did not continue) showed a lower value than the serum level of the previous stage of therapy (pre-treatment stage), but still had a higher values than the upper reference limit of healthy children control group (serum level ranged between 223 - 489 U/L). The mean value of serum LDH level showed a high significant decrease ($P<0.005$) as compared to the mean value of the enzyme in the previous stage, and still a very high significant increase ($P<0.0005$) as compared to the mean value of healthy children control group.

Regarding to the LDH isoenzymatic activities the data revealed that, there were a very high significant decrease ($p<0.0005$) in LD2, LD-3, LD-4 as compared to the mean value of the previous stage of therapy, and showed a non significant difference ($P>0.05$) in LD2, and still a very high significant increase ($P<0.0005$) in the LD3 and LD4, and there was a high significant increase ($P<0.005$) in LD-5 as compared to the mean

value of the healthy children control group. The data shown in tables (16,22,23).

3-During radiotherapy stage (GC) 18 cases

The serum level of T.LDH activity in eighteen cases (18 cases out of 21 cases which represent 85.7% of all cases, in addition to the two cases there is another one case excluded who did not continue) showed a higher values than the serum level of the previous stage of therapy (pre-radiotherapy stage) and a higher values than the upper reference limit of healthy children control group (serum level ranged between 339-560 U/L). The mean value showed a high significant increase ($P<0.005$) as compared to the mean value of the previous stage, and a very high significant increase ($P<0.0005$) as compared to the mean value of healthy children control group.

Regarding to the LDH isoenzymatic activities the data revealed that, there are a high significant increase ($P<0.005$) in the mean values of LD-1, and a very high significant increase ($P<0.0005$) in LD-2, LD-3, LD-4, and LD-5 as compared to the mean value of healthy children control group, and mild significant increase ($P<0.01$) in LD1 and LD2, slight significant increase ($P<0.025$) in LD3, No significant difference ($P>0.05$) in LD4, and a very high significant increase ($P<0.0005$) in LD5 as compared to the mean value of the previous stage. The data shown in tables (17,22,23).

4-During chemotherapy stage (GD) 16 cases

The serum level of T.LDH activity in sixteen cases (16 cases out of 21 cases which represent 67.2% of all cases, in addition to the three cases there are another one case was excluded who died from chest infection, and one case developed chest metastases) showed a higher values than the upper reference limit of healthy children

control group and lower values than the serum level of the previous stage of therapy (during radiotherapy), (serum level ranged between 300-460 U/L). The mean value showed a very high significant increase ($P<0.0005$) as compared to the mean value of healthy children control group and a high significant increase ($P<0.005$) as compared with the mean value of the previous stage of therapy.

Regarding to the LDH isoenzymatic activities the data revealed that, there are non significant difference ($P>0.05$) in LD1 and LD2, and a very high significant decrease ($P<0.0005$) in LD3, LD4, and LD5 as compared with the mean values of the previous stage of therapy, and also showed high significant increase ($P<0.005$) in LD1 and a very high significant increase ($P<0.0005$) in LD-2, LD-3, LD-4, and LD-5 as compared to the mean value of healthy children control group. The data shown in tables (18,22,23).

5- At the end of chemotherapy stage (GE) 12 cases

The serum level of T.LDH activity in twelve cases (12 out of 21 which represent 57.1% of all cases, in addition to the five cases there are another four cases developed metastases during the treatment) showed a lower value than the serum level in the previous stage of therapy (during chemotherapy stage), and also showed a normal range level as healthy children control group (serum level ranged between 263 - 359 U/L), and the mean value showed a very high significant decrease ($P<0.0005$) as compared to the mean value of the previous stage of therapy, and also showed a non significant variation ($P>0.05$) as compared to the mean value of healthy children group.

Regarding to the LD isoenzymatic activities the data revealed that, there are a very high significant decrease ($P<0.0005$) in LD1, LD2, LD3, and LD4, and also showed a slight significant increase ($P<0.025$) in LD-3, and LD-5 as compared to the mean value of healthy children control group. The data shown in tables (19,22,23).

6- At follow up stage after 6 months (GF) 12 cases

The serum level of T.LDH activity in twelve cases showed a normal range as healthy children control group (serum level ranged between 252-351 U/L), and the mean value showed a non significant variation ($P>0.05$) as compared to the mean value of the previous stage of therapy (end of therapy) and the healthy children group.

Regarding to the LD isoenzymatic activities the data revealed that, there are non significant variation in all isoenzymes activity except LD5 in which there is a slight significant decrease ($P<0.025$) as compared to the mean values of the previous stage of therapy, and also showed a non significant variations ($P>0.05$) in all isoenzymes activity as compared to the mean value of the healthy children control group. The data shown in tables (20,22,23).

The results of the five cases which developed metastases

The five cases which developed metastases during the course of treatment and during follow up (5 cases out of 21 cases which represent 23.4% of all cases), one case no.12 developed bone and chest metastases during the last course of chemotherapy, and cases no. 2,15,16 developed chest metastases during the last course of chemotherapy, case no.13 developed bone metastases during follow up after 6 months. The serum level of T.LDH activity in the

five cases showed a higher values than the upper reference limit of healthy children control group (serum level ranged between 423-999 U/L). The mean value showed a very high significant increase ($P<0.0005$) as compared to the mean value of healthy children control group, while as regarding to the LD isoenzymatic activities the data revealed that, there were very high significant increase in LD-1, LD-2, LD-3, LD-4, and LD-5 as compared to the mean value of healthy children control group. The data showed in tables (21,22).

Correlation between serum LDH levels and the prognosis

At presentation, serum LDH activity elevated in 14 patients (67%) and normal in 7 patients (33%). Among patient with high serum LDH level at presentation, there seems to be a fairly good correspondence between serum level and the incidence of metastases, the percentage of patients who experienced metastases were 23.8%, four patients 80% of all patients who developed metastases had high serum LDH levels and only one patient (20%) had normal serum LDH level.

All changes in the the serum LDH was due to changes in the anaerobic iso-enzymes LD2,LD3, and LD4. While during metastases there were an increase in tha all five isoenzymes. In all patients who experienced metastases, the serum LDH levels at presentation decreased gradually in response to therapy but not reached the normal levels, till they developed metastases the serum levels were highly increased.

Table (13) Total serum LDH (U/L) and its isoenzymatic activities (U/L) in 15 healthy children control group.

Serial no	Age	T.LDH (U/L)	LD1 (U/L)	LD2 (U/L)	LD3 (U/L)	LD4 (U/L)	LD5 (U/L)
1	8	297	121.9	128.64	24.56	12.68	9.89
2	8	314	105.83	148.32	33.13	14.73	11.99
3	8	350	119.1	120.03	70.84	26.18	13.86
4	8	345	120.34	135.31	58.96	21.29	9.11
5	10	270	95.47	106.27	35.26	15.8	17.17
6	10	268	73.3	94.51	52.39	25.51	22.29
7	10	283	72.93	93.42	66.19	27.37	23.12
8	10	298	87.17	106.21	54.68	28.43	18.54
9	11	345	99.33	120.61	68.79	28.60	27.66
10	11	291	79.91	91.17	68.56	27.88	17.63
11	11	254	62.23	93.12	61.32	26.59	10.74
12	12	305	99.64	119.59	47.79	26.78	11.19
13	12	280	89.99	96.74	49.78	25.14	18.31
14	12	272	74.39	94.9	63.83	26.54	12.35
15	12	310	93.78	110.61	59.64	27.22	18.76
Range n=15	8-12	254-350	62.2-120.3	91.1-148.6	24.5-70.8	12.6-28.6	9.11-27.6
Mean	10.02	298.8	91.64	11.96	54.381	24.05	16.17
S.D		29.83	16.811	19.03	14.092	5.314	5.469
S.E		7.70	4.341	4.914	3.648	1.372	1.412
distr.		100%	30.67%	37.47%	18.19%	8.05%	5.41%

Table (14) Total LDH (U/L) and its isoenzymatic activities (U/L) in 15 healthy adolescent control group.

Serial no	Age (yr.)	T. LDH (U/L)	LD1 (U/L)	LD2 (U/L)	LD3 (U/L)	LD4 (U/L)	LD5 (U/L)
1	13	310	87.45	110.27	61.88	27.13	23.28
2	13	297	83.03	104.51	58.89	24.08	21.15
3	13	270	65.18	104.70	58.81	25.97	18.04
4	13	333	100.37	125.81	62.44	25.44	18.95
5	14	390	131.55	150.7	59.9	25.39	22.39
6	14	285	108.73	116.96	36.11	13.19	10.01
7	14	298	85.43	109.52	63.15	24.67	18.18
8	15	282	81.10	105.56	54.91	21.83	18.95
9	15	315	82.40	125.31	65.46	24.03	21.04
10	16	345	125.03	138.86	40.26	20.97	23.05
11	16	299	112.33	123.31	38.51	15.13	9.72
12	17	269	77.96	104.83	52.08	19.63	14.49
13	18	303	115.87	127.14	41.45	12.42	9.15
14	20	320	96.86	146.43	40.61	17.56	18.05
15	20	280	73.19	116.98	60.98	12.93	18.70
Range n=15	13-20	269-390	65.18-131.55	104.51-150.74	36.1-65.46	13.19-27.13	9.15-23.28
Mean	15.4	306.4	95.098	120.73	53.03	20.69	17.68
SD		31.80	19.81	15.24	10.54	5.21	4.75
SE		8.211	5.124	3.932	2.723	1.352	1.231
% distr.		100%	30.95%	39.39%	17.5%	5.57%	5.63%

Table (15) Total LDH (U/L) and its isoenzymatic activities (U/L) among children with Ewing's sarcoma at the pre treatment stage (GA).

Serial no.	Age (yr.)	T.LDH (U/L)	LD1 (U/L)	LD2 (U/L)	LD3 (U/L)	LD4 (U/L)	LD5 (U/L)
1	8	598	113.68	199.97	160.14	107.52	20.57
2	8	615	115.86	208.36	163.59	108.79	18.39
3	8	618	116.69	206.52	164.88	109.57	16.99
4	11	547	111.37	178.65	138.17	98.36	20.46
5	11	279	76.61	99.14	64.67	19.97	15.82
6	11	813	124.71	279.56	232.84	160.01	15.85
7	11	749	106.13	252.93	207.02	155.27	20.15
8	12	329	106.76	115.91	65.73	19.8	14.14
9	12	289	88.02	197.25	53.75	26.5	13.47
10	12	513	103.78	182.27	133.69	72.69	20.57
11	12	647	131.41	205.68	172.62	114.97	16.24
12	12	523	111.97	170.71	136.71	83.16	20.45
13	13	730	106.43	270.39	201.48	136.73	11.32
14	13	498	95.42	158.68	135.75	95.12	18.52
15	13	304	79.36	111.2	62.42	32.81	14.14
16	13	489	98.97	156.19	137.6	69.68	20.55
17	14	318	92.95	118.74	65.37	26.62	16.73
18	14	310	82.83	123.81	64.17	24.93	14.76
19	14	694	120.48	226.66	196.12	124.78	19.06
20	14	656	124.25	224.22	173.77	117.88	16.07
21	16	296	63.49	85.66	73.85	56.36	16.64
Range n=21	8-16	279-813	63.49-131.41	85.66-270.39	53.75-232.89	19.8-160.01	11.32-20.57
Mean	12	515	103.39	179.64	133.54	83.87	17.47
SD		173.69	17.75	55.99	56.29	45.59	3.35
SE		37.9	3.88	12.28	12.28	9.95	0.73
% dist		100%	21.86%	34.04%	25.39%	15.05%	3.79%

Table (16) Total LDH (U/L) and its isoenzymatic activities (U/L) among children with Ewing's sarcoma at the pre-radiotherapy stage (GB).

Serial no	Age (yr.)	T. LDH (U/L)	LD1 (U/L)	LD2 (U/L)	LD3 (U/L)	LD4 (U/L)	LD5 (U/L)
1	8	409	83.52	122.98	102.95	77.46	22.09
2	8	437	101.91	134.06	52.88	26.79	25.07
3	8	449	120.52	178.77	86.03	39.06	24.60
4	11	440	71.21	73.08	54.02	23.86	17.62
5	11	238	61.52	97.53	39.10	21.13	18.71
6	11	466	119.95	143.43	123.49	54.10	27.45
7	11	489	120.35	166.58	128.70	49.14	27.82
8	12	298	69.49	115.15	62.04	29.14	22.17
9	12	223	63.98	75.07	48.17	20.45	17.08
10	12	395	82.04	160.8	85.48	42.74	23.89
11	12	Did not continue					
12	12	396	85.29	124.54	97.77	67.72	20.67
13	13	476	122.05	168.31	107.96	52.12	25.50
14	13	377	98.73	123.24	103.46	38.07	25.33
15	13	317	77.09	122.96	65.75	22.92	28.28
16	13	357	109.06	116.49	93.92	22.02	16.92
17	14	289	86.55	95.22	63.96	28.29	15.84
18	14	347	86.51	111.98	86.61	33.14	28.77
19	14	460	121.07	146.02	112.79	55.16	24.93
20	14	The patient died					
21	16	264	65.63	72.47	55.33	48.71	18.95
Range n=19	8-16	264-489	61.52-126.52	72.47-172.77	39.1-123.49	20.45-77.46	15.84-28.77
Mean	12	375.11	91.91	123.62	82.65	39.58	22.19
SD		83.75	21.64	32.10	27.05	16.68	4.57
SE		19.21	4.96	7.37	6..21	3.83	1.05
% dist.		100%	25.69%	34.26%	22.65%	10.9%	6.49%

Table (17) Total LDH (U/L) and its isoenzymatic activities (U/L) among children with Ewing's sarcoma during radiotherapy stage (GC).

Serial no	Age (yr.)	T. LDH (U/L)	LD1 (U/L)	LD2 (U/L)	LD3 (U/L)	LD4 (U/L)	LD5 (U/L)
1	8	510	109.45	162.08	122.09	84.30	32.08
2	8	405	113.24	122.07	88.69	48.48	32.52
3	8	449	118.89	138.60	74.17	45.17	31.74
4	11	476	120.80	145.13	107.76	57.17	45.74
5	11	501	122.09	162.22	113.43	68.58	34.82
6	11	560	102.27	220.79	126.61	62.72	47.6
7	11	490	122.99	158.71	98.59	65.86	43.86
8	12	345	93.74	106.5	84.11	34.36	26.29
9	12	410	109.02	128.74	95.32	36.74	40.18
10	12	339	84.41	137.13	64.92	31.15	21.35
11	12	Did not continue					
12	12	396	85.29	124.54	97.77	67.72	20.67
13	13	452	107.73	164.46	100.75	38.56	35.97
14	13	397	110.64	122.67	96.47	37.43	29.77
15	13	421	108.07	130.34	107.94	42.02	32.72
16	13	407	82.78	130.77	119.58	34.60	39.31
17	14	472	117.48	166.14	99.07	41.07	47.86
18	14	456	119.47	171.36	69.81	56.86	38.53
19	14	410	97.13	143.71	89.99	37.39	37.68
20	14	The patient died					
21	16	Did not continue					
Range n=18		339-560	82.78-122.99	106.5-220.79	69.81-126.61	31.15-84.30	21.35-47.86
Mean		440.17	106.94	147.15	97.88	49.32	36.19
SD		56.44	13.37	25.79	17.40	15.13	7.19
SE		13.3	3.15	6.08	4.10	3.57	1.69
% dist.		100%	5.3%	32.99%	2.28%	11.06%	8.37%

Table (18) Total LDH (U/L) and its isoenzymatic activities (U/L) among children with Ewing's sarcoma during chemotherapy stage (GD).

Serial no	Age (yrs)	T. LDH (U/L)	LD1 (U/L)	LD2 (U/L)	LD3 (U/L)	LD4 (U/L)	LD5 (U/L)
1	8	Did Not Continue					
2	8	450	117.41	168.71	94.46	44.91	24.48
3	8	300	77.82	110.34	64.95	24.57	19.32
4	11	401	120.95	145.36	77.35	32.32	25.02
5	11	398	103.24	154.14	82.62	37.16	19.06
6	11	390	104.87	151.87	74.41	31.86	23.47
7	11	430	106.55	167.31	89.18	42.78	24.17
8	12	325	93.44	106.24	71.99	31.43	21.90
9	12	347	107.02	123.81	67.49	26.44	22.24
10	12	341	93.70	129.41	71.41	27.59	18.76
11	12	Did not continue					
12**	12	698	153.98	212.54	167.31	110.21	53.95
13	13	410	117.30	140.47	89.18	35.14	32.02
14	13	350	10.6.65	118.13	72.73	28.46	24.05
15	13	409	113.66	158.61	84.82	26.46	21.31
16	13	420	117.85	159.35	84.71	36.71	21.38
17	14	460	123.97	166.93	100.79	42.64	25.35
18	14	415	107.65	164.92	82.92	37.10	22.41
19	14	340	102.65	132.97	63.38	27.23	13.77
20	14	The patient died					
21	16	Did Not Continue					
Range n=16		325-460	93.44-123.97	106.24-186.71	63.38-100.79	24.57-44.91	13.77-32.02
Mean		386.63	107.17	143.66	79.52	33.31	22.42
SD		46.95	11.86	21.16	10.82	6.46	3.92
SE		11.74	2.97	5.29	2.70	1.62	0.98
% dist.		100%	27.53%	36.69%	20.77%	9.08%	5.93%

** = case developed metastases and excluded from the study.

Table (19) Total LDH (U/L) and its isoenzymatic activities (U/L) among children with Ewing's sarcoma at the end of chemotherapy (GE).

Serial no	Age (yr.)	T. LDH (U/L)	LD1 (U/L)	LD2 (U/L)	LD3 (U/L)	LD4 (U/L)	LD5 (U/L)
1	8	Did not continue					
2**	8	999	198	361.54	247.55	94.30	97.31
3	8	283	82.32	109.60	53.14	25.75	12.11
4	11	336	99.52	109.53	72.74	28.86	25.33
5	11	350	105.98	123.34	71.89	28.21	21.01
6	11	359	98.76	110.89	66.42	21.89	22.94
7	11	300	91.92	118.14	56.13	17.10	13.71
8	12	293	84.70	94.19	62.07	26.21	25.69
9	12	263	70.56	82.06	62.62	23.30	24.46
10	12	323	94.22	107.46	67.76	28.94	24.61
11	12	did not continue					
12**	12	the patient developed Metastases					
13	13	390	110.18	129.67	81.03	31.08	38.02
14	13	331	94.69	111.08	68.88	28.83	19.33
15**	13	447	88.86	109.47	112.55	71.21	64.90
16**	13	423	108.88	133.45	106.97	32.02	37.39
17	14	345	106.54	117.16	67.59	29.46	24.56
18	14	262	63.01	101.81	57.67	21.45	18.02
19	14	290	87.29	95.17	69.09	22.42	15.23
20	14	The patient died					
21	16	Did Not Continue					
Range n=13		263-390	70.56-110.18	82.06-129.67	53.14-81.03	17.10-31.08	12.11-38.02
Mean		317.3	91.51	108.47	65.93	25.65	21.92
SD		38.99	13.89	12.84	7.58	4.11	6.69
SE		10.82	3.85	3.56	2.10	1.15	1.85
% dist.		100%	28.84%	34.19%	20.77%	8.08%	6.91%

** = cases developed metastases and excluded from the study.

Table (20) Total LD (U/L) and its isoenzymatic activities (U/L) among children with Ewing's sarcoma at follow up after 6 months of cure (GF).

Serial no.	Age (yr.)	T.LDH (U/L)	LD1 (U/L)	LD2 (U/L)	LD3 (U/L)	LD4 (U/L)	LD5 (U/L)
1	8	Did not continue					
2**	8	The patient developed metastases					
3	8	298	92.02	101.20	58.38	25.21	21.22
4	11	343	94.43	117.82	91.07	24.97	14.71
5	11	337	110.91	128.39	49.77	26.62	21.28
6	11	295	89.23	92.19	67.17	20.59	25.81
7	11	262	77.60	85.41	56.72	22.51	19.75
8	12	271	69.67	82.30	51.07	19.23	18.72
9	12	252	75.37	91.09	50.88	17.58	17.06
10	12	250	74.09	86.92	60.29	22.56	15.13
11	12	did not continue					
12**	12	the patient develop metastases					
13**	13	507	96.09	126.13	103.99	46.28	34.51
14	13	281	94.55	117.26	40.77	14.69	13.71
15**	13	The patient developed metastases					
16**	13	The patient developed metastases					
17	14	351	116.60	132.77	69.60	17.59	17.35
18	14	258	71.85	100.56	42.28	22.91	20.28
19	14	310	88.07	112.69	62.86	27.01	18.10
20	14	The patient died					
21	16	Did Not Continue					
Range n= 12		252-351	69.67-116.66	82.30-132.77	40.77-91.07	19.23-27.01	13.71-25.8
Mean		293.08	87.87	104.05	58.41	21.79	18.59
SD		35.38	15.09	17.32	13.66	3.91	3.38
SE		10.21	4.36	4.99	3.94	1.14	0.98
% dist.		100%	30.12%	35.75%	20.08%	7.56%	6.49%

** = cases developed metastases and excluded from the study.

Table(21) Serum TLD (U/L) and its isoenzymatic activities in 5 cases who developed metastasis during the course of treatment and follow up after 6 months.

	T.LDH (U/L)	LD1 (U/L)	LD2 (U/L)	LD3 (U/L)	LD4 (U/L)	LD5 (U/L)
Range	423-999	88.86-198.20	109.47-361.54	103.99-247.55	32.02-110.21	34.51-97.3
Mean	634.5	129.16	188.63	147.67	70.80	57.61
SD	245.56	46.05	104.51	61.56	32.44	25.42
SE	122.78	20.59	46.74	27.53	14.51	11.37
% dist.	100%	21.03%	30.7%	24.04%	11.53%	9.38%

Table (22):- Range and Mean ± SE of Total LDH (U/L) and Its Isoenzymatic activities (U/L) Control group and Ewing's Sarcoma among children at different stages of therapy.

Group	No. of cases	Stage		T.LDH (U/L)	LD Isoenzymatic Activities (U/L)				
					LD1 (U/L)	LD2 (U/L)	LD3 (U/L)	LD4 (U/L)	LD5 (U/L)
Cc	15	Control Children	Range	254 -350	62.23 -120.34	91.17 -148.64	24.56 -70.84	12.68 -28.6	9.11 -27.66
		Age range 8 - 12 y.	Mean ± SE	298.8 ± 7.70	91.64 ± 4.34	111.96 ± 4.91	54.38 ±3.65	24.05 ±1.37	16.17 ±1.41
GA	21	Pre-treatment	Range	289 - 813	63.49 -131.41	85.66 -270.39	53.75 -232.8	19.8 -160.01	11.32 -26.55
		Age range 8 - 15 y.	Mean ± SE	515 ±37.90	103.39 ±3.88	179.64 ±12.22	133.54 ±12.28	83.87 ±9.93	17.47 ±0.73
GB	19	Pre-radiotherap	Range	264 - 489	61.52 -126.52	72.47 -172.77	39.1 -123.49	20.45 -77.46	15.84 - 28.77
			Mean ± SE	375.11 ±19.21	91.91 ±4.96	123.62 ±7.37	82.65 ±6.21	39.58 ±3.83	22.19 ±1.05
GC	18	During-radiotherap	Range	339 -560	82.48 -122.99	106.5 -220.79	69.81 -126.61	31.15 -84.3	21.35 -47.86
			Mean ±SE	440.17 ±13.3	106.94 ±3.15	147.15 ±6.08	97.88 ±4.10	49.32 ±3.57	36.19 ±1.69

Table (22) continued

Group	No. of cases	Stage		T.LDH (U/L)	LD Isoenzymatic Activities (U/L)				
					LD1 (U/L)	LD2 (U/L)	LD3 (U/L)	LD4 (U/L)	LD5 (U/L)
GD	16	During-radiother.	Range	325 -460	93.44 -123.97	106.24 -186.7	63.38-100.79	24.57 -44.9	13.77 -32.02
			Mean + SE	385.63 ± 11.74	107.17 ±2.97	143.66 ± 5.29	79.52 ±2.70	33.31 ±1.62	22.42 ±0.98
GE	12	At the end of chemothe.	Range	263 -359	70.56 -106.54	82.06 -123.34	53.14 -72.74	17.10 -29.46	12.11 -25.69
			Mean + SE	311.25 ±9.74	89.96 ±3.83	106.70 ±3.36	64.67 ±1.83	25.20 ±1.14	20.58 ±1.41
GF	12	During-follow up after 6 m.	Range	252 -351	69.67 -116.66	82.30 -132.7	40.77 -91.07	19.23 -27.01	13.71 -25.8
			Mean + SE	293.08 ±10.12	87.87 ±4.36	104.05 ±4.99	58.41 ±3.94	21.79 ±1.14	18.59 ±0.98
GM**	5	Develop. Metas. during treatment	Range	423 -999	88.86 -198.2	109.47 -361.54	103.99 -247.55	32.02 -110.21	34.5 -97.3
			Mean + SE	634.5 ±122.78	129.16 ±20.59	188.63 ±46.74	147.67 ±27.23	70.80 ±1451	57.61 ±11.37

Table (23) Statistical significant of serum T.LDH (U/L) and its isoenzymatic activities in control and Ewing's sarcoma among children at different stages of therapy.

(A) Serum T.LDH

	GA (21)	GB (19)	GC (18)	GD (16)	GE (13)	GF (12)
C. child. (15)	5.590 v.h.s.i.	3.687 v.h.s.i	9.198 v.h.s.i	6.2558 v.h.s.i.	1.0027 N.S.	0.4473 N.S.
GA	___	3.292 h.s.d.	1.863 N.S.	3.2354 h.s.d.	5.2068 v.h.s.d..	5.6538 v.h.s.d.
GB	___	___	2.784 h.s.i.	0.5117 N.S.	2.9650 h.s.d.	3.7707 v.h.s.d
GC	___	___	___	3.018 h.s.d.	7.8204 v.h.s.d	8.7726 v.h.s.d.
GD	___	___	___	___	4.9415 v.h.s.d	6.0127 v.h.s.d.
GE	___	___	___	___	___	1.2877 N.S.
GF	___	___	___	___	___	___

GA = Pre- treatment group
GB = After 3 courses of chemotherapy
GC = During radiotherapy
GD = during chemotherapy (Post radiotherapy)
GE = End of therapy
GF = Follow up after 6 months

(B) Serum LD1

	GA (21)	GB (19)	GC (18)	GD (16)	GE (13)	GF (12)
C. child. (15)	2.0184 s.s.i.	N.S.	2.8531 h.s.i	2.9531 h.s.i.	0.2902 N.S.	0.6128 N.S.
GA	—	1.8230 N.S.	0.7103 N.S.	0.7736 N.S.	2.4634 m.s.d.	2.6592 m.s.d.
GB	—	—	2.5580 m.s.i	2.6396 m.s.i.	0.3112 N.S.	0.6118 N.S.
GC	—	—	—	0.1152 N.S.	3.4241 v.h.s.d	3.545 v.h.s.d
GD	—	—	—	—	3.5509 v.h.s.d	3.6584 v.h.s.d
GE	—	—	—	—	—	0.3601 N.S.
GF	—	—	—	—	—	—

(C) Serum LD2

	GA (21)	GB (19)	GC (18)	GD (16)	GE (13)	GF (12)
C. child. (15)	5.1391 v.h.s.i	1.3167 N.S.	4.5029 v.h.s.i	4.3921 v.h.s.i	0.8841 N.S.	1.1299 N.S.
GA	—	3.9256 v.h.s.d	2.3804 s.s.d	2.7020 h.s.d	5.7553 v.h.s.d	5.7267 v.h.s.d
GB	—	—	2.4628 m.s.i	2.2089 s.s.i	2.0889 s.s.d	2.1988 s.s.d
GC	—	—	—	0.4330 N.S.	5.8229 v.h.s.d	5.4796 v.h.s.d
GD	—	—	—	—	5.8977 v.h.s.d	5.4468 v.h.s.d
GE	—	—	—	—	—	0.4405 N.S.
GF	—	—	—	—	—	—

(D) Serum LD3

	GA (21)	GB (19)	GC (18)	GD (16)	GE (12)	GF (12)
C. child. (15)	6.1805 v.h.s.i	3.9274 v.h.s.i	7.9341 v.h.s.i	5.5471 v.h.s.i	2.5257 m.s.i	0.7513 N.S.
GA	——	3.6982 v.h.s.d	2.7544 h.s.d	4.2964 v.h.s.d	5.5471 v.h.s.d	5.8256 v.h.s.d
GB	——	——	2.0467 s.s.d	0.4622 N.S.	2.7772 h.s.d	3.2961 h.s.d
GC	——	——	——	3.7399 v.h.s.d	7.3967 v.h.s.d	6.9413 v.h.s.d
GD	——	——	——	——	4.5528 v.h.s.d	4.4197 v.h.s.d
GE	——	——	——	——	——	1.4410 N.S.
GF	——	——	——	——	——	——

(E) Serum LD4

	GA (21)	GB (19)	GC (18)	GD (16)	GE (12)	GF (12)
C. child. (15)	5.9559 v.h.s.i	3.8179 v.h.s.i	6.6085 v.h.s.i	4.3646 v.h.s.i	0.6452 N.S.	1.2680 N.S.
GA	——	4.1541 v.h.s.d	3.2684 h.s.d	5.0154 v.h.s.d	5.8582 v.h.s.d	6.1986 v.h.s.d
GB	——	——	1.8603 N.S.	1.5077 N.S.	3.5985 h.s.d	4.4519 v.h.s.d
GC	——	——	——	4.0838 v.h.s.d	6.4361 v.h.s.d	7.3460 v.h.s.d
GD	——	——	——	——	4.0941 v.h.s.d	5.8155 v.h.s.d
GE	——	——	——	——	——	2.1151 s.s.d
GF	——	——	——	——	——	——

(F) Serum LD5

	GA (21)	GB (19)	GC (18)	GD (16)	GE (12)	GF (12)
C. child. (15)	0.8188 N.S.	3.4243 h.s.i	9.0961 v.h.s.i	3.6398 v.h.s.i	2.2116 h.s.i	1.4093 N.S.
GA	——	3.6909 h.s.i	10.1688 v.h.s.i	4.0507 v.h.s.i	1.9587 N.S.	0.9165 N.S.
GB	——	——	7.0365 v.h.s.i	0.1601 N.S.	0.9158 N.S.	2.5065 m.s.d
GC	——	——	——	7.0486 v.h.s.d	7.0924 v.h.s.d	9.0091 v.h.s.d
GD	——	——	——	——	1.0716 N.S.	2.7635 h.s.d
GE	——	——	——	——	——	1.1589 N.S.
GF	——	——	——	——	——	——

Figure (6)

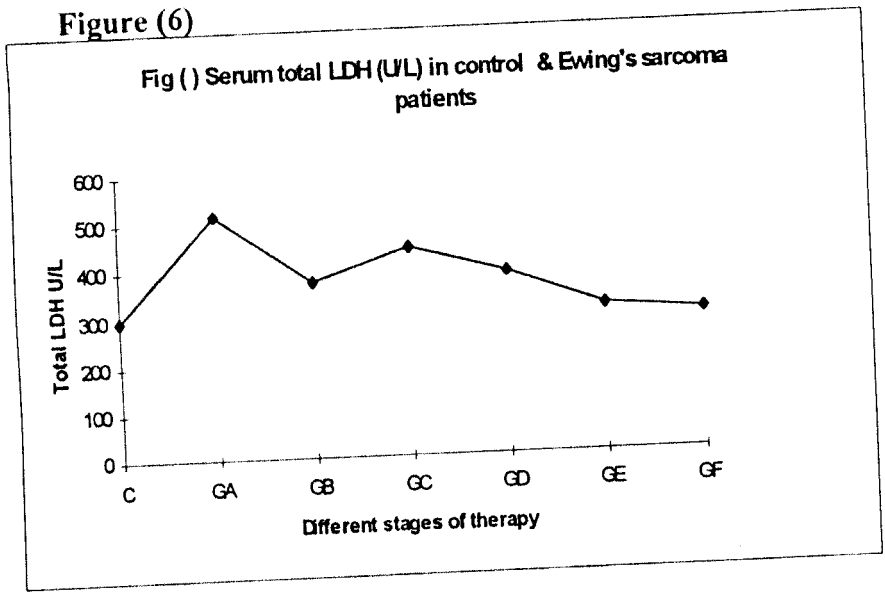


Figure (7)

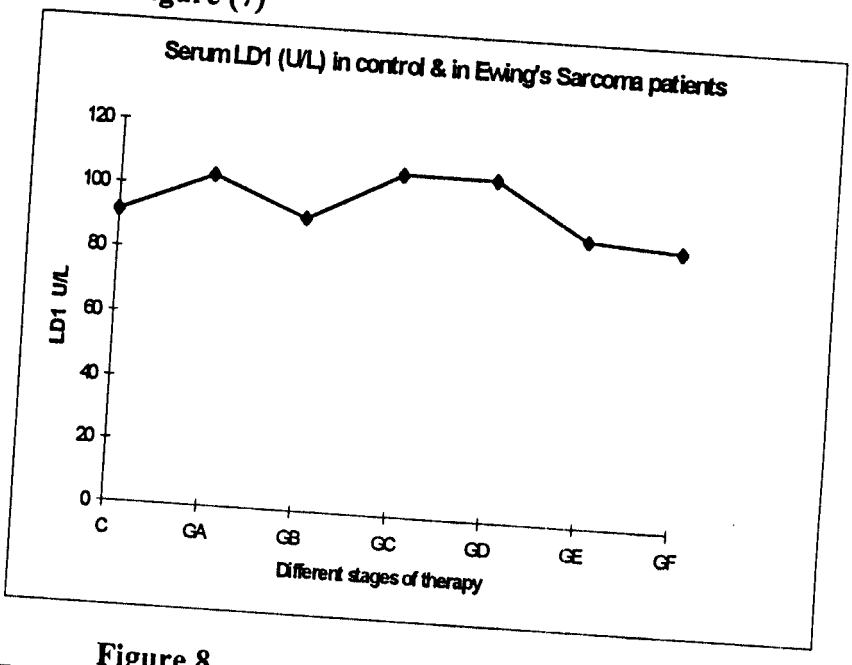


Figure 8

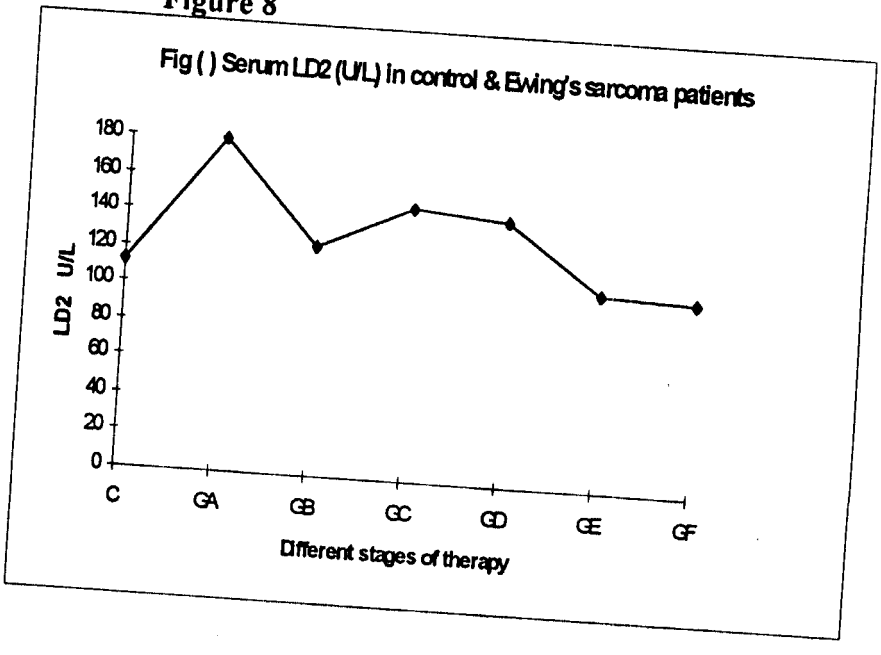


Figure (9)

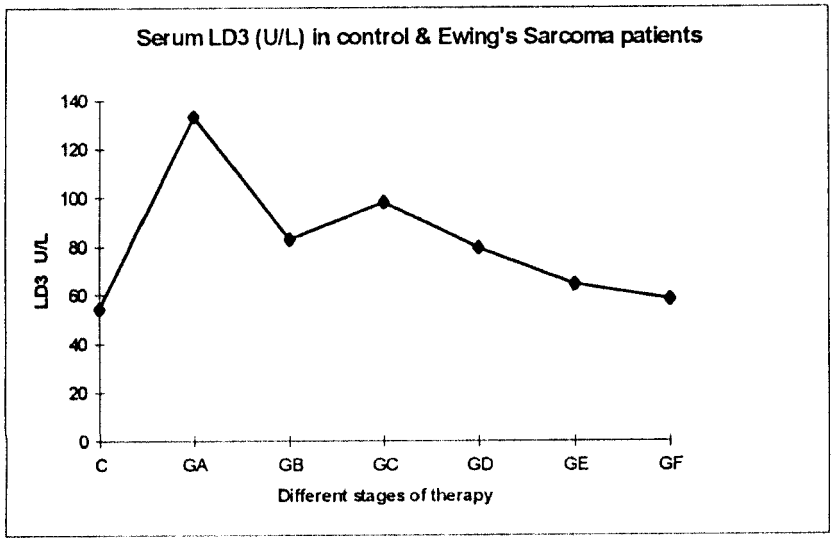


Figure (10)

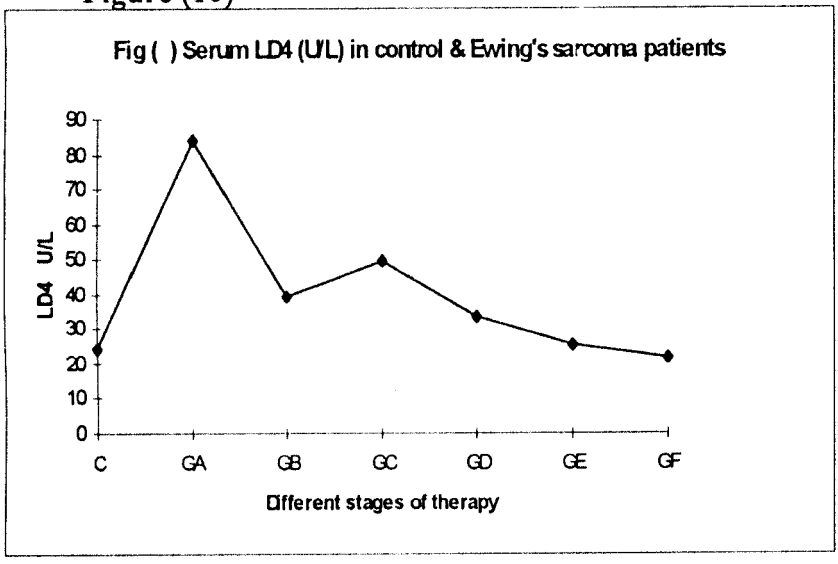


Figure (11)

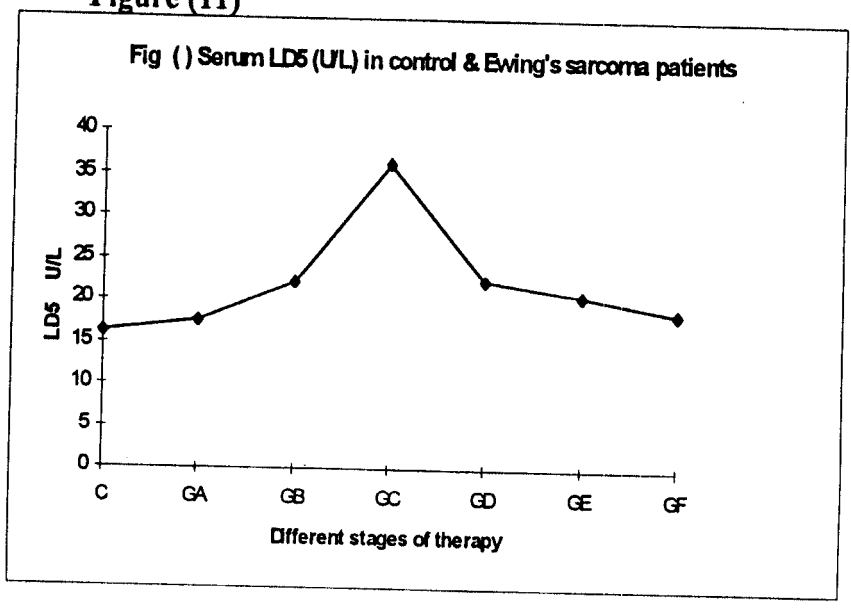
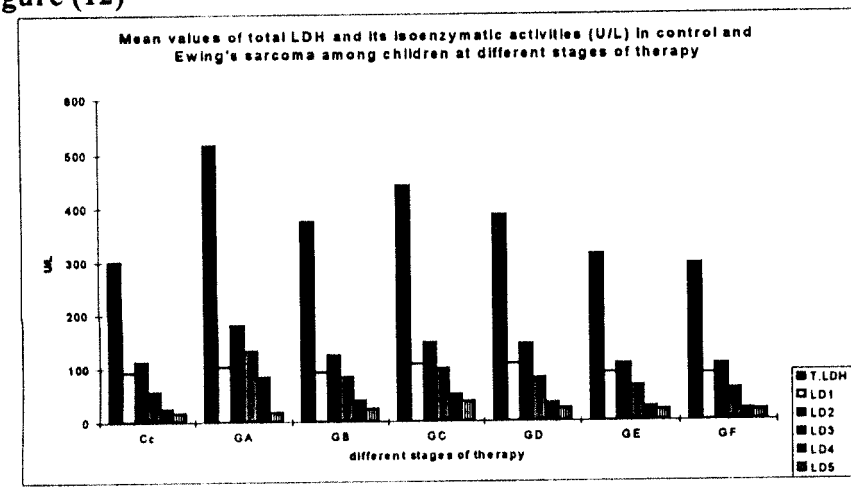


Fig ( ) Serum LD5 (U/L) in control & Ewing's sarcoma patients
LD5 U/L
40
35
30
25
20
15
10
5
0
C
GA
GB
GC
GD
GE
GF
Different stages of therapy

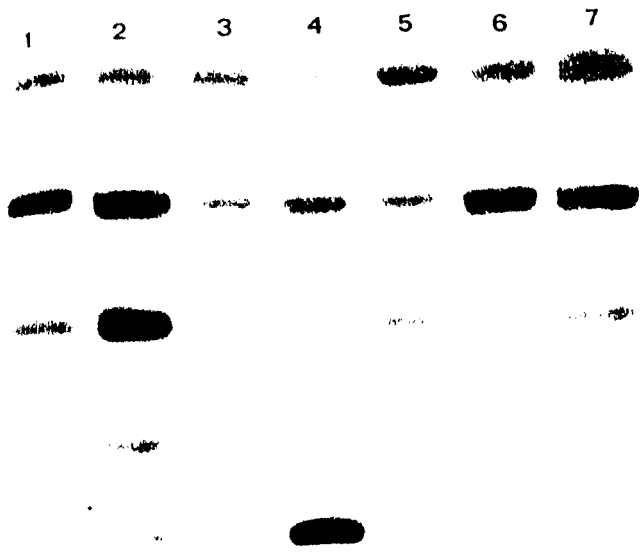
Figure (12)


Mean values of total LDH and its isoenzymatic activities (U/L) in control and Ewing's sarcoma among children at different stages of therapy
800
500
400
300
200
100
0
U/L
Cc
GA
GB
GC
GD
GE
GF
different stages of therapy
T.LDH
LD1
LD2
LD3
LD4
LD5

Serum LDH iso-enzyme patterens (separated on 1% agarose gel. PH 8.6 in patients with Ewing's sarcoma and healthy controls.

Lanes 1, 2, from the left demonstrate cases with Ewing's sarcoma. While lanes 3,4,5,6,7, represent the healthy control cases.

Note the slightly increase in the isoenzyme LD1, and marked increase in LD2, LD3, LD4 in the cases with Ewing's sarcoma as compared with the control cases.



Pic. (2): Serum LDH isoenzyme patterns (separated on 1% agarose gel, pH 6.8) in control and patient with Ewing's sarcoma at different stages of treatment.

Healthy control case is demonstrated in lane 1 from the left. Patient with Ewing's sarcoma at different stage of therapy are demonstrated in lanes 2,3,4,5,6,7 (pre-treatment, pre-radiotherapy, during the radiotherapy, during chemotherapy, at the end of chemotherapy and at follow up after 6 months of cure respectively.)

Note the gradual decrease in the increased iso-enzymes in response to the therapy except during radiotherapy there is increase in the LD5 (lane 4).

The results of Serum Gamma Glutamyl transferase (GGT) activities

1- In control groups (GC)

The serum level of GGT activity in control groups (children and adolescent) showed a normal range (serum level ranged from 10 - 40 U/L) and the mean value of the children group showed a non significant (P>0.05) variations as compared to the mean value of adolescent healthy control group, the data shown in tables (24,25, $38,41_{A-F}$).

In Osteosarcoma among children group 25 cases

The serum level of GGT activity in children with Osteogenic sarcoma group in different stages of therapy and follow up showed a normal range as children healthy group (serum level ranged between 11 - 40 U/L) and the mean value showed a non significant (P>0.05) variations as compared to the mean value of children healthy group, the data shown in tables ($39,41_{A-F}$).

In Osteosarcoma among adolescent group 10 cases

The serum level of GGT activity in adolescent with Osteogenic sarcoma group in different stages of therapy and follow up showed a normal range as adolescent healthy group (serum level ranged between 11 - 40 U/L) and the mean value showed a non significant (P>0.05) variations as compared to the mean value of adolescent healthy group, the data shown in tables ($40,41_{A-F}$).

The results of Serum alkaline phosphatase and its isoenzymatic activities

A)- In control groups

In healthy children and adolescent control group (Cc,Ca) 30 cases, the serum level of T.ALP activity in the healthy children control group (Cc) (serum level ranged between 317-551U/L), showed a higher value than the upper normal reference limit of adolescent healthy group (Ca) (serum level ranged between 131-294 U/L), and the mean value of the children control group showed a very high significant ($P<0.0005$) increase as compared to the mean value of healthy adolescent control group.

Regarding to the serum ALP isoenzymatic activities in healthy children control group, the mean values showed a very high significant ($P<0.0005$) increase in the bone fraction of serum ALP "B.ALP" and also the liver fraction of ALP "L.ALP" showed a non significant ($P>0.05$) variations as compared to the healthy adolescent control group, the data shown in table ($24,25,38,41_{A-F}$)

B) -In Osteogenic Sarcoma among children

1- In pre-treatment stage (GA) 25 children cases

The serum level of T.ALP activity in the 25 cases "serum level ranged between 440-1089 U/L", showed higher value than the upper normal reference limit of healthy children group, and the mean value showed a very high significant ($P<0.0005$) increase as compared to the mean value of healthy children group.

Regarding to the ALP isoenzymatic activities the mean values showed a very high significant ($P<0.0005$) increase in the bone fraction of ALP "B.ALP" and also the liver fraction of ALP "L.ALP" showed a non significant ($P>0.05$) variation as compared

to the healthy children control group, the data shown in tables ($32,39,42_{A-F}$)

2- In pre-operative stage (GB) 25 children cases

The serum level of T.ALP activity in the 25 cases " serum level ranged between 381-968 U/L", showed a lower value than the serum level of the previous stage of therapy (pre-treatment stage) but still has higher value than the upper normal reference limit of healthy children group, and the mean value showed a high significant ($P<0.005$) decrease as compared to the mean value of the previous stage of therapy, and also showed a very high significant ($P<0.0005$) increase as compared to the mean value of healthy children group.

Regarding to the serum ALP isoenzymatic activities, the mean values of bone fraction of ALP "B.ALP" showed a high significant ($P<0.005$) decrease compared to the mean value of the previous stage of therapy, and also showed a very highly significant ($P<0.0005$) increase as compared to the mean value of healthy children control group, and the other isoenzymes liver fraction of ALP "L.ALP" showed a non significant ($P>0.05$) variation as compared to the mean value of the previous stage of therapy and the healthy children control group, the data shown in tables ($33,39,42_{A-F}$)

3- In post-operative stage (GC) 22 children cases

The serum level of T.ALP activity in 22 cases " serum level ranged between 334-625 U/L", (22 cases out of 25 cases which represent 88%, two cases were excluded because they refused the surgery, and one case developed metastases) showed a lower value than the serum level of the previous stage of therapy (pre-operative stage) and showed a higher serum level as healthy children group,

and the mean value showed a high significant ($P<0.005$) decrease as compared to the mean value of the previous stage of therapy, and showed a non significant ($P>0.05$) variation as compared to the mean value of healthy children group.

Regarding to the ALP isoenzymatic activities, the mean value of bone fraction of ALP "B.ALP" showed a high significant ($P<0.005$) decrease as compared to the mean value of the previous stage of therapy, and non significant ($P>0.05$) variations as compared to the mean value of healthy children group, also the liver fraction of ALP "L.ALP" showed non significant ($P>0.05$) variations as compared to the mean value of the previous stage of therapy, and healthy children group. The data shown in tables ($34,39,42_{A-F}$).

4- During chemotherapy stage (GD) 20 children cases

The serum level of T.ALP activity in 20 cases "serum level ranged between 300-710 U/L", (20 cases out of 25 cases which represent 80%, in addition to the first three cases, there were two cases died) showed a slight higher serum value as compared to the previous stage of therapy (post-operative stage), and to the healthy children group, and the mean value showed a non significant ($P>0.05$) variation as compared to the mean value of the previous stage of therapy and to the healthy children group.

Regarding to the ALP isoenzymatic activities, the mean value of the bone fraction of ALP "B.ALP" showed a non significant ($P>0.05$) variations as compared to the mean value of the previous stage of therapy, and of the healthy children group, also the liver fraction of ALP "L.ALP" showed non significant ($P>0.05$) variations as compared to the mean value of the previous stage of

therapy, and healthy children group. The data shown in tables ($35,39,42_{A-F}$).

5- At the end of chemotherapy (GE) 17 children cases

The serum level of T.ALP activity in 17 cases " serum level ranged between 357-610 U/L", (17 cases out of 25 cases which represent 68%, in addition to the five cases , there were another three cases developed metastases) showed a serum level value mostly as the serum value of the previous stage of therapy (during chemotherapy stage) and as the healthy children group, the mean value showed a non significant ($P>0.05$) variation as compared to the mean value of the previous stage of therapy, and to the healthy children group.

Regarding to the ALP isoenzymatic activities, the mean value of the bone fraction of ALP "B.ALP" showed a non significant ($P>0.05$) variations as compared to the mean value of the previous stage of therapy, and of the healthy children group, also the liver fraction of ALP "L.ALP" showed non significant ($P>0.05$) variations as compared to the mean value of the previous stage of therapy, and healthy children group. The data shown in tables ($36,39,42_{A-F}$).

6- During follow up after 6 months stage (GF) 16 cases

The serum level of T.ALP activity in 16 cases " serum level ranged between 360-530 U/L", (16 cases out of 25 cases which represent 64%, in addition to the eight cases , there is another one case developed metastases) showed a normal serum level as the healthy children group as well as the serum level of the previous stage of therapy (end of chemotherapy), and the mean value showed a non significant ($P>0.05$) variation as compared to the

mean value of the previous stage of therapy, and healthy children control group.

Regarding to the ALP isoenzymatic activities, the mean value of the bone fraction of ALP "B.ALP" showed a non significant ($P>0.05$) variations as compared to the mean value of the previous stage of therapy, and of the healthy children group, also the liver fraction of ALP "L.ALP" showed non significant ($P>0.05$) variations as compared to the mean value of the previous stage of therapy, and healthy children group. The data shown in tables ($37,39,42_{A-F}$).

C)- In osteogenic sarcoma among adolescents

1- In pre-treatment stage (GA) 10 adolescent cases

The serum level of T.ALP activity in the 10 cases "serum level ranged between 396 - 833 U/L", showed higher value than the upper normal reference limit of healthy adolescent group and the mean value showed a very high significant ($P<0.0005$) increase as compared to the mean value of healthy adolescent control group.

Regarding to the ALP isoenzymatic activities the mean value of bone fraction of ALP "B.ALP" showed a very highly significant ($P<0.0005$) increase, also the liver fraction of ALP "L.ALP" showed a non significant ($P>0.05$) variation as compared to the mean value of healthy adolescent control group. The data shown in tables ($26,40,41_{A-F}$)

2- In pre-operative stage (GB) 10 adolescent cases

The serum level of T.ALPT activity in the 10 cases "serum level ranged between 239-616 U/L", showed a lower value than the serum level of the previous stage of therapy (pre-treatment stage), but still had a higher value than the upper normal reference

limit of healthy adolescent group. The mean value showed a high significant ($P<0.005$) decrease as compared to the mean value of the previous stage of therapy, and also showed a very high significant ($P<0.0005$) increase as compared to the mean value of healthy adolescent control group.

Regarding to the ALP isoenzymatic activities the mean values of bone fraction of ALP "B.ALP" showed a high significant ($P<0.005$) decrease as compared to the mean value of the previous stage, and showed a very highly significant ($P<0.0005$) increase as compared to the mean value of healthy adolescent control group, also the liver fraction of ALP "L.ALP" showed a non significant ($P>0.05$) variation as compared to the mean value of the previous stage of therapy and to the healthy adolescent control group. The data shown in tables ($27,40,41_{A-F}$).

3- In post-operative stage (GC) 9 adolescent cases

The serum level of T.ALP activity in 9 cases "serum level ranged between 168-512 U/L" (9 cases out of 10 cases which represent 90%, one case was excluded, because he refused surgery) showed a lower value than the serum level of the previous stage of therapy (pre-operative stage), but still had a higher value than the upper normal reference limit of healthy adolescent group, and the mean value showed a slight significant ($P<0.025$) decrease as compared to the mean value of the previous stage of therapy, and showed a slight significant ($P<0.025$) increase as compared to the mean value of healthy adolescent group.

Regarding to the ALP isoenzymatic activities the mean values of bone fraction of ALP "B.ALP" showed a slight significant ($P<0.025$) decrease as compared to the mean value of the previous stage of therapy, and also showed a slight significant ($P<0.025$)

increase as compared to the mean value of healthy adolescent group, also the liver fraction of ALP "L.ALP" showed a non significant (P>0.05) variation as compared to the mean value of the previous stage of therapy, and of the healthy adolescent control group. The data shown in tables (28,40,41$_{A-F}$).

4- During chemotherapy stage (GD) 9 adolescent cases

The serum level of T.ALP activity in 9 cases "serum level ranged between 185-460 U/L" showed a slightly lower value than the serum level of the previous stage of therapy (post-operative stage), but still had a higher value than the upper normal reference limit of healthy adolescent group, and the mean value showed a non significant (P>0.05) variation as compared to the mean value of the previous stage of therapy, and also showed a slight significant (P<0.025) increase as compared to the mean value of healthy adolescent group.

Regarding to the ALP isoenzymatic activities the mean values of bone fraction of ALP "B.ALP" showed a non significant (P>0.05) variation as compared to the mean value of the previous stage of therapy, and also showed a slight significant (P<0.025) increase as compared to the mean value of healthy adolescent control group, also the liver fraction of ALP "L.ALP" showed a non significant (P>0.05) variation as compared to the mean value of the previous stage of therapy, and the healthy adolescent control group.The data shown in tables (29,40,41$_{A-F}$).

5- At the end of chemotherapy stage (GE) 8 adolescent cases

The serum level of T.ALP activity in 8 cases "serum level ranged between 149-310 U/L", (in addition to the first case, there is another one case developed metastases) showed a slight higher serum level than the serum level of the previous stage of therapy

(during chemotherapy), and the healthy adolescent group, and the mean value showed a non significant (P>0.05) variation as compared to the mean value of the previous stage of therapy, and the healthy adolescent group.

Regarding to the ALP isoenzymatic activities the mean values of the bone fraction of ALP "B.ALP" showed a non significant (P>0.05) variation as compared to the mean value of previous stage of therapy, and to the healthy adolescent control group, also the liver fraction of ALP "L.ALP" showed a non significant (P>0.05) variation as compared to the mean value of the previous stage of therapy, and the healthy adolescent control group. The data shown in tables ($30,40,41_{A-F}$).

6- At follow up stage after 6 months (GF) 8 adolescent cases

The serum level of T.ALP activity in 8 cases "serum level ranged between 141-245 U/L", (in addition to the first case, there is another one case was developed metastases) showed a normal serum level as of healthy adolescent group and lower level than the previous stage of therapy (end of therapy), and the mean value showed a non significant (P>0.05) variation as compared to the mean value of the previous stage of therapy, and of the healthy adolescent group.

Regarding to the ALP isoenzymatic activities the mean values of the bone fraction of ALP "B.ALP" showed a non significant (P>0.05) variation as compared to the mean value of the previous stage of therapy, and of the healthy adolescent control group, also the liver fraction of ALP "L.ALP" showed a non significant (P>0.05) variation as compared to the mean value of the previous stage of therapy, and the healthy adolescent control group. The data shown in tables ($31,40,41_{A-F}$).

Correlation between serum ALP levels and the prognosis

At presentation, serum ALP elevated in 19 children patients (76%) and normal in 6 patients (24%), while in adolescents it was elevated in all patients (100%). In children and adolescent patients (29 patients) with elevated serum ALP levels, the values of the enzyme were moderately elevated (less than two times of normal values) in 20 patients (69%) and considerably elevated (more than two times of normal values) in 9 patients (31%),

The percentage of patients with high serum ALP levels was significantly higher in adolescent patients compared with the children patients (100% versus 76%, $P<0.01$)

Among patients with high serum ALP level at presentation, there seems to be a fairly good correspondence between serum ALP levels and the incidence of metastases. The percentage of patients who experienced metastases (local or extended) were 20% in children patients and 10% in adolescent patients, the all patients whom developed metastases had high levels of serum ALP at presentation specially who had a considerable elevated levels of serum ALP.

All changes in the serum ALP was due to changes in the bone fraction of ALP (as determined by heat inactivation method and electrophoresis of ALP isoenzymes) with more confirmation that such increment in the serum ALP was due to bone fraction of ALP, the serum GGT activity concentrations were within the normal reference limits.

In 10 patients (53%) of 19 children patients with localized disease and high serum ALP levels at presentation, the enzyme level returned to normal after surgery. In the remaining 7 patients (37%) the serum levels decreased after surgery but remained elevated more than the normal values, (2 patients did not continued), While in adolescent patients the serum ALP almost returned to the normal values except in one case, in whom the serum levels decreased after surgery but still higher than the normal limits.

The percentage of metastases is 57% in children patients in whom the serum ALP levels remained above the normal values after surgery, and 10% in children patients in whom the serum ALP levels returned to the normal levels after surgery, so there is a significant higher incidence for metastases in patients with high serum ALP levels after surgery.

During post-operative chemotherapy and in follow up by 6 months, The serum levels of ALP and its isoenzymes returned to the normal values in 69% of patients who continued therapy, while in 31% of patients the serum levels showed an elevation in the enzyme activity specially in the bone fraction of ALP before the clinical diagnosis of metastases, so any changes in the tumor activity can be detected by the enzyme levels before other clear-cut physical signs are manifested.

Table(24): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoretic demonstration in healthy children control group.

Serial no	Age (yr.)	GGT (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% Inhibit.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme patterns by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	8	19	497	462	7.04%	383.56	113.44	_	+	++	_
2	8	35	508	452	11.02%	426.81	81.19	_	+	++	_
3	8	18	436	407	6.65%	361.15	74.85	_	+	++	_
4	8	40	486	401	17.48%	380.56	105.44	_	+	++	_
5	10	31	551	503	8.71%	457.04	93.96	_	+	++	_
6	10	23	546	495	9.34%	455.5	90.5	_	+	++	_
7	10	15	529	472	10.77%	418.22	110.78	_	+	++	_
8	10	17	499	399	20.04%	402.01	96.99	_	+	++	_
9	11	27	501	459	8.39%	388.36	112.63	_	+	++	_
10	11	17	463	399	13.82%	337.06	125.94	_	+	++	_
11	11	25	362	332	8.29%	223.76	138.03	_	+	++	_

Table (24) continued

Serial no	Age (yr.)	GGT (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% Inhibit.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme patterns by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
12	12	8	405	327	19.25%	275.8	129.2	_	+	++	_
13	12	26	321	285	11.21%	244.83	76.17	_	+	++	_
14	12	20	317	269	15.15%	217.29	99.71	_	+	++	_
15	12	14	392	334	14.79%	177.71	115.82	_	+	++	_
Range n= 15	8 -12	8-40	317-551	269-503	6.65-20.04	177.71-457.04	74.85-138.03				
Mean	10.02	22.33	454.2	399.7	12.13	343.31	104.31				
S.D		8.59	77.96	75.4	4.38	92.12	19.12				
S.E		2.11	20.13.	19.46	1.13	23.785	4.937				

Table(25): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoreticdemonstration in healthy adolescent control group.

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ I. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	13	42	294	265	9.86%	181.26	112.72	_	+	++	_
2	13	11	230	211	8.26%	143.55	86.45	_	+	++	_
3	13	25	198	166	16.2%	121.59	76.41	_	+	++	_
4	13	14	227	206	9.25%	128.03	98.97	_	+	++	_
5	14	19	195	162	16.9%	88.85	106.15	_	+	+	_
6	14	17	184	165	10.3%	118.2	65.8	_	+	++	_
7	14	36	245	203	17.2%	101.77	143.23	_	+	+	_
8	15	35	154	139	9.74%	72.17	81.83	_	+	+	_
9	15	23	203	165	18.6%	52.4	151	_	+	+	_
10	16	31	178	165	7.1%	46.24	131.86	_	+	++	_
11	16	22	237	214	9.7%	142.32	94.68	_	+	++	_

Table (25) continued

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
12	17	26	198	174	12.1%	128.12	69.98	_	+	++	_
13	17	17	163	144	11.6%	58.3	104.7	_	+	+	_
14	20	13	131	114	12.9%	51.57	79.43	_	+	+	_
15	20	28	151	135	10.6%	91.71	59.29	_	+	+	_
Range n= 15	13-20	11-42	131-294	114-265	7.1-18.63	51.57-181.26	46.24-151				
Mean	15.4	23.93	199.2	175.2	12.02	101.73	97.5				
S.D		9.18	42.49	38.54	3.57	40.38	27.85				
S.E		2.37	10.97	9.95	0.92	10.28	7.14				

Table(26): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoretic demonstration in osteogenic sarcoma among adolescents at the pre-treatment stage (GA).

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	13	18	495	415	16.16%	389.1	105.9	-	+	++	-
2	13	29	432.5	393.6	8.99%	337.62	94.88	-	+	++	-
3	14	11	833	701	15.85%	721.16	111.84	-	+	+++	-
4	14	21	569	476	16.34%	492.55	76.45	-	+	++	-
5	14	12	496	421	15.12%	406.51	89.49	-	+	++	-
6	15	17	418	369	11.72%	298.34	119.66	-	+	++	-
7	15	18	593	500	15.63%	510.33	82.67	-	+	++	-
8	16	40	595	502	15.62%	513.42	81.58	-	+	++	-
9	18	35	532	424	20.30%	427.14	104.86	-	+	++	-
10	20	21	396	343	13.38%	301.63	94.37	-	+	++	-
Range n=10	13-20	11-40	396-833	343-701	8.99-20.3	298.34-721.16	76.45%-119.66				
Mean	15.2	22.2	535.95	454.46	14.91	439.77	96.17				
S.D		9.56	126.29	101.44	3.02	127.25	14.15				
S.E		3.02	39.94	32.08	0.96	40.24	4.47				

Table(27): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoretic demonstration in osteogenic sarcoma among adolescents at the pre-operative stage (GB).

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	13	19	309	183	12.44%	117.11	91.89	_	+	+	_
2	13	20	324.6	294.6	9.24%	230.11	94.49	_	+	++	_
3	14	16	616	506	17.86%	495.02	120.98	_	+	++	_
4	14	25	423.6	381.4	9.96%	336.7	86.4	_	+	++	_
5	14	11	352	319	9.38%	260.75	91.25	_	+	++	_
6	15	26	310	260	16.13%	199.5	110.5	_	+	+	_
7	15	11	405	347	14.32%	334.11	70.89	_	+	++	_
8	16	35	546	434	20.51%	443.39	102.61	_	+	++	_
9	18	19	451.4	392.8	12.98%	331.07	120.33	_	+	++	_
10	20	19	239	216	9.62%	140.89	98.11	_	+	+	+
Range n=10	13-20	11-35	239-616	216-506	9.24-20.51	117.1-495.02	70.89-120.33				
Mean	15.2	20.1	397.6	333.38	13.24	288.87	98.85				
S.D		7.2	116.16	99.68	3.93	122.98	15.50				
S.E		2.28	36.73	31.52	1.24	38.89	4.9				

Table(28): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoretic demonstration in osteogenic sarcoma among adolescents at post-operative stage (GC).

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	13	29	192	165	14.06%	108.01	83.99	-	+	+	-
2	13	21	230.9	191.5	17.06%	146.52	84.38	-	+	+	-
3	14	15	512	450	12.11%	401.02	110.98	-	+	+	-
4	14	35	360	320	11.11%	270.74	89.26	-	+	+	-
5	14	Refuse operation									
6	15	22	168	145	13.69%	68.7	99.3	-	+	+	-
7	15	19	351	292	16.81%	273.36	77.64	-	+	+	-
8	16	15	244	194	20.49%	154.77	89.23	-	+	+	-
9	18	16	300.9	269.2	10.54%	180.47	120.43	-	+	+	-
10	20	28	176	165	6.25%	82.88	93.13	-	+	+	-
Range n=9	13-20	15-35	168-512	145-450	6.25-20.49	68.7-401.02	77.64-120.4				
Mean	15.2	22.22	281.64	243.52	13.57	187.39	94.26				
S.D		7.05	112.08	99.18	4.21	108.44	13.79				
S.E		2.35	37.36	33.06	1.04	36.15	4.61				

Table(29): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoretic demonstration in osteogenic sarcoma among adolescents during chemotherapy stage (GD).

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	13	26	185	161	12.97%	98.06	86.94	_	+	+	_
2	13	20	250	210	16%	143.22	106.78	_	+	+	_
3	14	17	460	431	6.31%	347.51	112.49	_	+	++	_
4	14	27	340	315	7.35%	249.83	90.17	_	+	+	_
5	14	Did not continue									
6	15	15	201	179	10.95%	116.81	84.19	_	+	+	_
7	15	11	320	288	10%	240.02	79.98	_	+	+	_
8	16	25	230	201	12.61%	124.84	105.16	_	+	+	_
9	18	16	270	226	16.31%	177.86	92.14	_	+	+	_
10	20	20	200	165	17.50%	100.86	99.14	_	+	+	_
Range n=9	13-20	11-27	185-460	161-431	6.31-17.5	98.06-347.51	79.98-112.49				
Mean	15.2	19.67	272.89	241.78	12.22	177.67	95.22				
S.D		5.48	88.25	88.48	3.95	85.04	11.2				
S.E		1.83	29.42	29.48	1.32	28.35	3.73				

Table(30): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoretic demonstration in osteogenic sarcoma among adolescents at the end of therapy (GE).

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	13	29	162	135	16.67%	82.34	89.66	_	+	+	_
2	13	19	231	186.3	19.35%	121.33	109.67	_	+	+	_
3	14	25	485	431	11.13%	366.19	118.81	_	+	+	_
4	14	11	310	280	9.68%	224.24	85.76	_	+	+	_
5	14	Did not continue									
6	15	35	158	129	18.35%	69.17	88.83	_	+	+	_
7	15	9	268	243	9.33%	181.62	86.38	_	+	+	_
8	16	19	235	215	8.51%	142.15	92.85	_	+	+	_
9	18	16	149	126	15.44%	94.33	54.67	_	+	+	_
10	20	28	196	175	10.71%	97.79	98.21	_	+	+	_
Range n=9	13-20	9-35	149-485	126-431	9.68-19.35	82.34-366.19	54.67-118.81				
Mean	15.2	23.11	243.78	213.37	13.24	153.24	91.65				
S.D		11.11	105.3	97.32	4.2	94.19	17.84				
S.E		3.7	35.11	32.44	1.4	31.39	5.95				

Table(31): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoretic demonstration in osteogenic sarcoma among adolescents at follow up after 6 months of cure (GF).

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L.Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	13	19	216	201	6.94%	133.08	82.82	_	+	+	_
2	13	34	168	135	19.64%	101.56	66.44	_	+	+	_
3**	14	10	694	560	19.31%	573.05	120.95	_	+	+++	_
4	14	16	245	198.6	18.94%	124.35	120.65	_	+	+	_
5	14	Did not continue									
6	15	19	228	189	17.03%	130.32	97.68	_	+	+	_
7	15	14	210	186	11.43%	112.06	97.94	_	+	+	_
8	16	15	141	129	8.51%	51.34	89.66	_	+	+	_
9	18	14	160	145	9.38%	70.01	89.99	_	+	+	_
10	20	16	210	196	6.67%	111.79	98.21	_	+	+	_
Range n=8	13-20	10-34	141-243	129-201	6.67-19.64	51.34-130.32	66.44-120.65				
Mean	15.2	18.38	197.25	172.45	12.32	104.31	92.94				
S.D		6.61	36.47	30.6	5.4	29.31	15.42				
S.E		2.32	12.89	10.82	1.91	10.36	5.45				

** = cases develop metastases and excluded from the study.

Table(32): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoretic demonstration in osteogenic sarcoma among children at the pre-treatment stage (GA).

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	8	18	984	878	10.77%	873.61	110.39	-	+	++++	-
2	8	20	975	861	11.69%	887.22	97.78	-	+	++++	-
3	8	16	894	729	18.46%	788.53	105.47	-	+	++++	-
4	8	19	1089	968	11.11%	965.54	123.46	-	+	+++++	-
5	8	28	1042	846	18.89%	919.97	122.03	-	+	+++++	-
6	9	17	584	482	17.46%	456.12	127.88	-	+	++	-
7	9	19	745	651	12.62%	660.55	84.45	-	+	+++	-
8	9	17	969	848	12.49%	873.32	95.68	-	+	++++	-
9	9	24	505	412	18.42%	386.06	118.91	-	+	++	-
10	10	19	540	510	5.51%	471.35	72.65	-	+	++	-
11	10	21	550	496	11.43%	474.5	65.5	-	+	++	-
12	10	19	544	514	5.61%	472.35	71.65	-	+	++	-
13	10	25	794	702	11.59%	717.06	76.94	-	+	+++	-
14	11	25	883.8	741.7	16.08%	757.01	126.79	-	+	+++	-
15	11	14	650	540	16.92%	577.26	72.74	-	+	+++	-
16	11	20	856	741	13.43%	735.22	120.78	-	+	+++	-

Table (32) Continued

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
17	11	16	953	887	6.93%	847.26	105.74	_	+	+++	_
18	11	30	867	745.7	13.99%	768.14	98.86	_	+	+++	_
19	12	26	631.6	513.9	18.63%	505.89	125.71	_	+	+++	_
20	12	16	942	816	13.37%	847.15	94.85	_	+	++++	_
21	12	18	440	350	20.36%	310.9	129.1	_	+	++	_
22	12	28	607	477	21.41%	499.01	107.99	_	+	++	_
23	12	11	590	466	21.0%	492.21	97.79	_	+	+++	_
24	12	15	554	477	13.89%	477.28	96.72	_	+	++	_
25	12	18	725	552	10.07%	627.48	97.52	_	+	+++	_
Range n=25	8-12	11-30	440-1089	350-968	5.5- 21.4	310-965.54	65.5-125.7				
Mean	10.2	19.96	756.58	652.17	14.07	655.64	101.88				
S.D		4.8	195.3	177.32	4.59	191.78	19.69				
S.E		0.94	38.35	35.46	0.92	38.35	3.94				

Table(33): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoretic demonstration in osteogenic sarcoma among children at the pre-operative stage (GB).

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	8	19	691.4	637.8	7.75%	582.17	109.23	_	+	+++	_
2	8	11	751	680	9.46%	655.58	95.42	_	+	+++	_
3	8	18	670	530	20.89%	559.73	110.27	_	+	+++	_
4	8	20	879	699	20.48%	758.52	120.48	_	+	+++	_
5	8	21	968	837	13.35%	847.25	120.75	_	+	+++	_
6	9	17	455	383	15.82%	330.98	124.02	_	+	++	_
7	9	16	625	501	19.84%	544.26	80.74	_	+	++	_
8	9	16	645	560.6	13.09%	525.64	119.36	_	+	++	_
9	9	20	381	351	7.87%	288.51	92.49	_	+	+	_
10	10	19	490	430	12.24%	379.43	110.57	_	+	++	_
11	10	23	449	369	17.82%	355.9	93.1	_	+	++	_
12	10	23	381	355	6.82%	301.44	79.56	_	+	++	_
13	10	23	595	487	18.15%	513.22	81.78	_	+	+++	_
14	11	17	615	492	20.01%	494.64	120.36	_	+	+++	_
15	11	13	580	490	15.52%	501.71	78.29	_	+	+++	_
16	11	15	710	650	8.45%	594.25	115.75	_	+	+++	_

Table (33) Continued

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
17	11	17	704	654	7.1%	604.18	99.82	_	+	++	_
18	11	21	609	491	19.38%	4999.22	109.78	_	+	++	_
19	12	16	556	466	16.19%	445.01	110.99	_	+	++	_
20	12	18	693.1	562.6	18.83%	603.85	89.25	_	+	+++	_
21	12	26	410	381	7.07%	284.74	125.26	_	+	+	_
22	12	23	498	412	17.27%	398.01	99.99	_	+	++	_
23	12	21	510	470	7.83%	411.55	98.45	_	+	++	_
24	12	13	505	447	11.49%	419.08	85.92	_	+	++	_
25	12	15	594	488	17.85%	515.68	78.32	_	+	++	_
Range n=25	8-12	11-26	381-908	351-837	6.82-20.1	288.5-847.5	78.3-125.26				
Mean	10.2	18.44	598.58	512.96	14.03	496.58	101.99				
S.D		3.79	144.58	122.71	5.02	141.04	15.84				
S.E		0.74	28.92	24.54	1.0	28.21	3.17				

Table(34): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoretic demonstration in osteogenic sarcoma among children at the post-operative stage (GC).

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	8	21	580	466	19.65%	461.15	118.85	_	+	++	_
2	8	16	610	482	20.98%	511.25	98.75	_	+	++	_
3	8	11	590	485	17.79%	474.67	115.33	_	+	++	_
4**	8	21	984	896	8.96%	758.52	120.48	_	+	++++	_
5	8	30	651	567	12.9%	530.96	120.04	_	+	++	_
6	9	11	336	310	7.73%	236.34	99.66	_	+	++	_
7	9	13	551	441	19.96%	478.25	72.75	_	+	++	_
8	9	19	545.6	508	6.89%	449.61	95.99	_	+	++	_
9	9	25	435	375	13.79%	354.38	80.62	_	+	++	_
10	10	18	334	305	8.68%	244.77	89.23	_	+	++	_
11	10	30	427	393	7.9%	340.05	96.95	_	+	++	_
12	10	21	529	486	8.13%	439.04	89.96	_	+	++	_
13	10	Did not continue refused operation									
14	11	16	366.6	326.2	11.02%	272.61	93.99	_	+	++	_
15	11	17	519	451	13.1%	413.26	105.74	_	+	++	_
16	11	21	625	564	9.76%	532.02	92.98	_	+	++	_

Table (34) continued

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
17	11		Did	not	continue	refused	operat.				
18	11	19	413	367	11.14%	331.81	81.19	_	+	++	_
19	12	21	514	460	10.51%	414.02	99.98	_	+	++	_
20	12	13	559	509.7	8.99%	231.83	95.17	_	+	++	_
21	12	16	411	379	7.87%	290.8	120.2	_	+	++	_
22	12	12	585	501	14.35%	476.19	108.81	_	+	++	_
23	12	19	490	410	16.33%	372.71	117.29	_	+	++	_
24	12	17	481	392	18.5%	398.03	82.97	_	+	++	_
25	12	22	501	453	9.58%	408.26	92.74	_	+	++	_
Range n=22	8-12	11-30	334-625	305-567	7.7-19.96	236.3-532.02	72.75-120.2				
Mean	10.2	18.55	502.42	437.72	12.53	393.72	98.69				
S.D		5.27	92.44	75.12	4.53	95.15	13.69				
S.E		1.12	20.75	16.02	0.97	20.84	2.92				

** = Cases developed metastases and excluded from the study.

Table(35): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoretic demonstration in osteogenic sarcoma among children during chemotherapy stage (GD).

Serial no	Age (yr.)	GGT (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	8	18	550	490	10.9%	434.22	115.78	_	+	++	_
2	8	16	550	495	10%	452.88	97.12	_	+	++	_
3	8	13	525	486	7.43%	419.64	105.36	_	+	++	_
4**	8	the		patient	develop	metast.					
5	8	12	710	658	7.32%	620.21	89.79	_	+	++++	_
6	9	15	329	291	11.55%	233.68	95.32	_	+	++	_
7	9	18	450	390	13.33%	374.26	75.74	_	+	++	_
8	9	19	470	390	17.02%	364.53	105.47	_	+	++	_
9	9	20	390	310	20.51%	298.33	91.67	_	+	+	_
10	10	28	360	305	15.28%	259.74	100.26	_	+	+	_
11	10	21	300	275	8.33%	203.55	96.45	_	+	+	_
12	10	11	450	401	10.89%	333.76	116.24	_	+	++	_
13	10			did	not	continue					
14	11	23	510	472	7.45%	414.57	95.43	_	+	++	_
15	11	17	460	384	16.53%	359.76	100.24	_	+	++	_
16	11	15	560	490	12.5%	467.83	92.17	_	+	++	_

Table (35) continued.

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
17	11	did not continue									
18	11	16	440	398	9.55%	318.22	121.78	_	+	+	_
19	12	14	350	305	12.86%	252.36	97.64	_	+	+	_
20	12	27	381	326	14.44%	281.82	99.18	_	+	+	_
21	12	30	335	301	10.15%	262.82	72.18	_	+	+	_
22	12	21	610	556	8.85%	494.88	115.12	_	+	++	_
23	12	20	480	415	13.54%	369.26	110.74	_	+	+	_
24	12	the patient died									
25	12	the patient died									
Range n=20	8-12	11-30	300-710	275-658	7.32-20.51	203.55-620.21	72.18-121.78				
Mean	10.2	18.7	460.5	406.9	11.92	360.82	99.68				
S.D		5.23	105.29	101.92	3.58	103.20	12.68				
S.E		1.17	23.54	22.79	0.799	23.08	2.84				

** =cases developed metastases and excluded from the study.

Table(36): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoretic demonstration in osteogenic sarcoma among children at the end of treatment stage (GE).

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	8	16	540	460	14.81%	429.15	110.85	-	+	++	-
2	8	16	541	482	10.91%	442.60	98.74	-	+	++	-
3	8	13	420	371	11.67%	313.19	106.81	-	+	++	-
4**	8	the patient develop. metast.									
5**	8	12	997	893	10.43%	916.51	80.49	-	+	+++++	-
6	9	10	395	345	12.66%	304.39	90.61	-	+	++	-
7	9	17	440	356	19.09%	342.81	97.19	-	+	++	-
8	9	19	510	456	10.59%	409.621	100.38	-	+	++	-
9	9	29	390	345	11.54%	300.26	89.74	-	+	++	-
10	10	15	405	372	8.15%	305.84	99.16	-	+	++	-
11	10	18	357	288	19.33%	260.55	96.45	-	+	++	-
12	10	26	498	432	13.25%	386.25	111.75	-	+	++	-
13	10	the patient did not continue									
14**	11	17	696	561	19.46%	597.01	98.99	-	+	+++	-
15	11	20	420	361	14.04%	317.26	102.74	-	+	++	-
16	11	17	610	554	9.18%	514.26	95.74	-	+	+++	-

Table (36) continued

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
17	11	Did not continue									
18	11	23	500.3	431.5	13.75%	378.24	122.06	-	+	++	-
19	12	11	367	340	7.36%	268.32	98.62	-	+	++	-
20	12	24	481.3	394.4	18.06%	383.43	97.87	-	+	++	-
21	12	20	366.6	326.2	11.02%	272.61	98.99	-	+	++	-
22**	12	18	927	732	14.45%	816.25	110.75	-	+	++++	-
23	12	19	360	295	18.05%	239.79	120.21	-	+	+	-
24	12	the patient died									
25	12	the patient died									
Range n=17	8-12	10-29	357-610	288-554	7.36-19.36	239.79-514.26	80.49-120.2				
Mean	10.2	18.41	447.13	388.77	13.15	345.21	102.23				
S.D		5.06	75.99	71.08	3.72	75.0	9.23				
S.E		1.23	18.43	17.24	0.90	18.19	2.24				

Table(37): Serum GGT (U/L), T.ALP (U/L) and its isoenzymatic activities (U/L) by heat inactivation and electrophoretic demonstration in osteogenic sarcoma among children at follow up stage after 6 months of cure (GF).

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
1	8	19	497	462	7.04%	383.56	113.44	-	+	++	-
2	8	13	460	375	18.48%	361.76	98.24	-	+	++	-
3	8	16	410	340	17.07%	304.26	105.74	-	+	++	-
4**	8			the	patient	develop.	Metast.				
5**	8	20	907	801	11.69%	786.55	120.45	-	+	+++++	-
6	9	13	426	391	8.22%	329.33	96.67	-	+	++	-
7	9	19	365	290	20.55%	270.75	94.25	-	+	++	-
8	9	11	516	456	11.63%	407.06	108.94	-	+	++	-
9	9	26	410	345	15.85%	299.22	110.78	-	+	++	-
10	10	23	399	368	7.77%	300.04	98.96	-	+	++	-
11	10	16	405	367	9.38%	275.8	129.2	-	+	++	-
12	10	19	501	459	8.39%	388.36	112.64	-	+	++	-
13	10			did	not	continue					
14**	11	25	1606	909	43.39%	901.3	196.7	508	+	+++++	-
15	11	20	405	359	11.35%	302.3	102.7	-	+	++	-
16**	11	12	950	810	14.74%	862.76	87.24	-	+	+++++	-

Table (37) Continued

Serial no	Age (yr.)	G.G.T (U/L)	T.ALP (U/L)	T.ALP+ L. Ph. A. (U/L)	% inhib.	Isoenzyme activities by heat inactivation (U/L)			Isoenzyme activities by electrophoresis		
						Bone	Liver	Placenta	Liver	Bone	Intestine
17	11	did not continue									
18	12	17	530	431.5	18.59%	411.33	118.67	-	+	++	-
19	12	14	436	407	6.65%	356.02	79.98	-	+	++	-
20	12	40	509	432	15.13%	419.31	89.69	-	+	++	-
21	12	16	486	401	17.49%	380.56	105.44	-	+	++	-
22**	12	18	864	766	11.34%	743.05	120.95	-	+	++++	-
23	12	11	360	315	12.5%	244.46	115.54	-	+	++	-
24	12	the patient died									
25	12	the patient died									
Range n=16	8-12	11-40	360-530	290-462	7.04-18.59	244.5-419.31	79.98-129.2				
Mean	10.2	18.31	444.68	387.41	12.88	339.63	105.05				
S.D		7.09	55.34	51.92	4.71	55.74	12.09				
S.E		1.77	13.83	12.98	1.18%	13.94	3.03				

** = cases developed metastases and excluded from the study.

Table (38):- Range and Mean ± SE of Total GGT (U/L),T.ALP (U/L) and Its Isoenzymatic Activities (U/L) in adolescent and Children Control groups.

Group	Stage		T.GGT (U/L)	T.ALP (U/L)	T.ALP + L.Ph.A. (U/L)	% Inhibit	Isoenzyme Activity By Heat Inactivation (U/L)		
							Liver (U/L)	Bone (U/L)	Intest. (U/L)
Cc	Cont.child.	no.	15	15	15	15	15	15	0
		Range	8-40	317-551	294-503	6.65-20.1	76.2-138.1	177.7-457.1	-
	Age range 8-13y.	Mean + SE	21.66 ± 2.11	448.2 ± 19.05	339 ± 19.46	12.13 ± 1.13	12.02 ± 0.92	93.95 ± 7.14	-
Ca	Cont.adol.	no.	15	15	15	15	15	15	0
		Range	11-42	131-294	114-265	7.1-18.63	46.24-151	51.57-181	-
	Age range 13-20y.	Mean + SE	23.86 ± 2.37	199.2 ± 10.97	175.2 ± 9.95	12.02 ± 0.92	93.95 ± 7.14	105.29 ± 10.28	-

Table (39): Range and Mean ± SE of Total GGT (U/L),T.ALP (U/L) and Its Isoenzymatic Activities (U/L) among Children with Osteogenic Sarcoma at Different Stages of Therapy.

Group	Stage		T.GGT (U/L)	T.ALP (U/L)	T.ALP+ L.Ph.A. (U/L)	% Inhibit.	Isoenzyme Activity By Heat Inactivation (U/L)		
							Liver	Bone	Intest.
Cc	Control-children Age range 8-12y.	no.	15	15	15	15	15	15	0
		Range	8-40	317-551	294-503	6.65-20.1	76.2-138.1	177.7-457.4	-
		Mean ± SE	22.33 ± 2.11	454.2 ± 20.13	339.7 ± 19.46	12.13 ± 1.13	104.31 ± 4.937	343.31 ± 23.785	-
GA	pre-treat. stage.	No.	25	25	25	25	25	25	0
		Range	11-30	440-1089	350-968	5.5-21.4	65.5-123.7	310-965.54	-
		Mean ± SE	19.96 ± 0.94	756.58 ± 38.35	652.17 ± 35.31	14.07 ± 0.92	101.88 ± 3.94	655.64 ± 38.35	-
GB	pre-operative stage.	no.	25	25	25	25	25	25	0
		Range	11-26	381-968	351-837	6.8-20.1	78.3-125.3	288.5-847.5	-
		Mean ±SE	18.44 ± 0.74	598.58 ± 28.92	512.96 ± 24.54	14.03 ± 1.01	101.99 ± 3.17	496.58 ± 28.21	-
GC	post-operative stage	no.	22	22	22	22	22	22	0
		Range	11-30	334-625	305-567	7.7-19.9	72.8-120.2	36.3-532.1	-
		Mean ± SE	18.55 ± 1.12	502.42 ± 20.75	437.72 ± 16.02	12.53 ± 0.97	98.69 ± 2.92	398.27 ± 20.84	-

Table (39):- Continued in children.

Group	Stage		T.GGT (U/L)	T.ALP (U/L)	T.ALP + L.Ph.A. (U/L)	% Inhibit.	Isoenzyme Activity By Heat Inactivation (U/L)		
							Liver (U/L)	Bone (U/L)	Intest. (U/L)
GD	during chemoth. Stage	no.	20	20	20	20	20	20	0
		Range	11-30	300-710	275-658	7.32-20	72.16-121.78	203.5-620.2	-
		Mean ± SE	18.7 ±1.17	460.5 ±23.54	406.9 ±22.79	11.92 ±0.799	99.68 ±2.84	360.8 ±23.08	-
GE	at end of chemoth. Stage	no.	17	17	17	17	17	17	0
		Range	10-29	357-610	288-554	7.4-19.3	80.44-120.2	239.8-514.3	-
		Mean ±SE	18.41 ± 1.23	447.13 ± 18.43	388.77 ± 17.24	13.15 ± 0.90	102.23 ± 2.24	345.21 ± 18.19	-
GF	at follow up stage.	no.	16	16	16	16	16	16	0
		Range	11-40	360-530	290-462	7.1-18.6	79.98-129.2	244.5-419.3	-
		Mean ± SE	18.31 ± 1.77	444.68 ± 13.83	387.41 ± 12.98	12.88 ± 1.18	105.05 ± 3.03	339.63 ± 13.94	-
M**	metast. cases during therapy	no.	5	5	5	5	5	5	0
		Range	12-25	864-1606	766-909	8.96 - 43.4	80.49-196.7	743.1-916.5	-
		Mean ± SE	17.6 ± 2.54	1082.2 ± 133.48	854.8 ± 28.27	17.77 ± 6.47	121.17 ± 20.63	836.42 ± 36.13	-

Table (40):- Range and Mean ±SE of Total GGT (U/L),T.ALP (U/L) and Its Isoenzymatic Activities (U/L) among adolescent with osteogenic sarcoma at different stages of therapy.

Group	Stage		T.GGT (U/L)	T.ALP (U/L)	T.ALP+ L.Ph.A. (U/L)	% Inhibit.	Isoenzyme Activity By Heat Inactivation (U/L)		
							Liver	Bone	Intestine
Ca	Control adolescent Age range 13-20y.	no.	15	15	15	15	15	15	0
		Range	11-42	131-294	114-265	7.1-18.63	46.24-151	51.57-181	-
		Mean +SE	23.86 ± 2.37	199.2 ± 10.97	175.2 ± 9.95	12.02 ± 0.92	93.95 ± 7.14	105.29 ± 10.28	-
GA	pre-treatment stage.	no.	10	10	10	10	10	10	0
		Range	11-40	396-833	343-701	8.99-20	76.5-119.66	298.3-21.2	-
		Mean +SE	22.2 ±3.02	535.95 ±39.94	454.46 ±32.08	14.91 ±0.96	96.17 ± 4.47	439.77 ± 40.24	-
GB	pre-operative stage.	no.	10	10	10	10	10	10	0
		Range	11-35	239-616	216-506	9.2-20.5	70.89-120.3	117.1-495.2	-
		Mean +SE	20.1 ± 2.28	397.60 ± 36.73	333.38 ± 31.52	13.24 ± 1.24	98.75 ± 4.90	288.87 ± 38.89	-
GC	post-operative stage.	no.	9	9	9	9	9	9	0
		Range	15-35	168-512	145-450	6.3-20.5	77.64-120.4	68.7-401.02	-
		Mean +SE	22.22 ± 2.35	281.64 ± 37.36	243.52 ± 33.06	13.57 ±1.40	94.26 ± 4.61	187.39 ± 36.15	-

Table (40):-Continued in adolescent.

Group	Stage		T.GGT (U/L)	T.ALP (U/L)	T.ALP + L.Ph.A. (U/L)	% Inhibit.	Isoenzyme Activity By Heat Inactivation (U/L)		
							Liver	Bone	Intest.
GD	during chemoth. Stage.	no.	9	9	9	9	9	9	0
		Range	11-27	185-460	161-431	6.3-17.5	84.2-112.49	98.06-347.5	-
		Mean ±SE	19.67 ± 1.83	272.89 ± 29.42	241.78 ±29.48	12.22 ±1.32	95.22 ± 3.73	177.67 ± 28.35	-
GE	at the end of chemo. stage.	no.	9	9	9	9	9	9	0
		Range	9-35	149-485	126-431	9.7-19.4	54.67-118.8	82.34-366.2	-
		Mean ±SE	23.11 ± 3.70	243.78 ± 35.11	213.37 ± 32.44	13.24 ± 1.40	91.65 ± 5.95	153.24 ± 31.39	-
GF	at follow up stage after 6m.	no.	8	8	8	8	8	8	0
		Range	10.34	141-245	129-201	6.7-19.6	66.44-120.7	51.34-130.3	-
		Mean ± SE	17.44 ± 2.26	197.25 ± 12.89	172.45 ± 10.82	12.32 ± 1.91	92.94 ± 5.45	104.31 ± 10.36	-
GM**	Metast. case at follow up stage.	No	1	1	1	1	1	1	0
		Range	10	694	560	19.31	120.95	375.05	-
		Mean ±S.E	———	———	———	———	———	———	———

Table (41) Results for Statistical Variations in G.G.T, T.ALP, and its isoenzymatic activities among adolescent in control and osteogenic sarcoma at different stages of treatment

(A) Serum T.GGT in adolescents

Group	GA (10)	GB (10)	GC (9)	GD (9)	GE (9)	GF (8)
Ca (15)	0.432 N.S.	0.836 N.S.	0.491 N.S.	1.399 N.S.	0.171 N.S.	1.960 N.S.
GA (10)	------	0.555 N.S.	0.284 N.S.	1.009 N.S.	0.045 N.S.	1.526 N.S.
GB (10)		------	0.339 N.S.	0.493 N.S.	0.460 N.S.	0.959 N.S.
GC (9)			------	0.853 N.S.	0.203 N.S.	1.466 N.S.
GD (9)				------	0.833 N.S.	0.767 N.S.
GE (9)					-----	1.308 N.S.
GF (8)						------

Group Ca = Control adolescent group

Group (GA) = Pretreatment stage

Group (GB) = Post chemotherapy-pre radiotherapy

Group (GC) = During radiotherapy

Group (GD) = Post radiotherapy- during chemotherapy

Group (GE) = At the end of chemotherapy

Group (GF) = During follow up after 6 months

(B) Serum T. ALP in adolescents

Group	GA (10)	GB (10)	GC (9)	GD (9)	GE (9)	GF (8)
Cc (15)	8.13 v.h.s.i.	5.177 v.h.s.i.	2.117 s.s.i.	2.347 s.s.i	1.212 N.S.	0.115 N.S.
GA (10)	-------	3.733 h.s.d.	4.65 v.h.s.d.	5.303 v.h.s.d	6.192 v.h.s.d.	8.07 v.h.s.d.
GB (10)		--------	2.193 s.s.d.	2.587 m.s.d.	2.958 h.s.d.	4.816 v.h.s.d.
GC (9)			-------	0.184 N.S.	0.738 N.S.	2.135 s.s.d
GD (9)				-------	0.753 N.S.	2.355 s.s.d.
GE (9)					--------	1.244 N.S.
GF (8)						--------

(IIII(Serum T.ALP + L.Ph.A in adolescents

Group	GA (10)	GB (10)	GC (9)	GD (9)	GE (9)	GF (8)
Cc (15)	8.314 v.h.s.i	4.786 v.h.s.i.	1.979 s.s.i	2.139 s.s.i	0.96 N.S.	0.187 N.S.
GA (10)	-----	2.692 m.s.d.	4.579 v.h.s.d.	4.881 v.h.s.d.	5.404 v.h.s.d.	8.329 v.h.s.d.
GB (10)		-----	1.967 N.S.	2.122 s.s.d.	2.775 m.s.d.	4.829 v.h.s.d.
GC (9)			-----	0.039 N.S.	0.017 N.S.	2.043 s.s.d.
GD (9)				-----	0.775 N.S.	2.208 s.s.
GE (9)					-----	1.033 N.S.
GF (8)						-----

(D) Serum ALP % inh. In adolescents

Group	GA (10)	GB (10)	GC (9)	GD (9)	GE (9)	GF (8)
Ca (15)	1.557 N.S.	0.662 N.S.	0.800 N.S.	0.048 N.S.	0.728 N.S.	0.142 N.S.
GA (10)	-----	1.065 N.S.	0.789 N.S.	1.648 N.S.	0.984 N.S.	1.035 N.S.
GB (10)		-----	0.176 N.S.	0.563 N.S.	0.000 N.S.	0.400 N.S.
GC (9)			-----	0.702 N.S.	0.167 N.S.	0.528 N.S.
GD (9)				-----	0.530 N.S.	0.043 N.S.
GE (9)					-----	0.388 N.S.
GF (8)						-----

(E) Serum L.ALP in adolescents

	GA 10)	GB (10)	GC (9)	GD (9)	GE 9)	GF (8)
Ca (15)	0.264 N.S.	0.554 N.S.	0.037 N.S.	0.158 N.S.	0.247 N.S.	0.112 N.S.
GA (10)	-------	0.458 N.S.	0.607 N.S.	0.163 N.S.	0.607 N.S.	.0.458 N.S.
GB (10)		-------	0.667 N.S.	0.573 N.S.	0.921 N.S.	0.793 N.S.
GC (9)			-------	0.162 N.S.	0.347n.s.	0.185 N.S.
GD (9)				-------	0.508 N.S.	0.345 N.S.
GE (9)					------	0.159 N.S.
GF (8)						-----

(F) Serum B.ALP in adolescents

	GA (10)	GB (10)	GC (9)	GD (9)	GE (9)	GF (8)
Ca (15)	8.053 v.h.s.i.	4.563 v.h.s.i.	2.184 s.s.i.	2.4 s.s.i.	1.452 N.S.	0.067 N.S.
GA (10)	------	2.696 m.s.d.	4.666 v.h.s.d.	5.325 v.h.s.d.	5.614 v.h.s.d.	8.073 v.h.s.d.
GB (10)		------	1.911 N.S.	2.311 s.s.d.	2.714 m.s.d.	4.586 v.h.s.d.
GC (9)			------	0.212 N.S.	0.713 N.S.	2.209 s.s.d.
GD (9)				------	0.578 N.S.	2.43 s.s.d.
GE (9)					------	1.48 N.S.
GF (8)						------

Table (42):-Results for Statistical Variations in G.G.T, T.ALP, and its isoenzymatic activities among children in control group and osteogenic sarcoma at different stages of treatment

(A) Serum T. GGT in children

Group	GA (25)	GB (25)	GC (22)	GD (20)	GE (17)	GF (16)
Cc (15)	1.614 N.S.	1.44 N.S.	1.302 N.S.	1.089 N.S.	1.331 N.S.	1.21 N.S.
GA (25)	-----	1.989 N.S.	1.241 N.S.	0.732 N.S.	1.245 N.S.	0.927 N.S.
GB (25)		-----	0.089 N.S.	0.182 N.S.	0.021 N.S.	0.068 N.S.
GC (22)			-----	0.073 N.S.	0.084 N.S.	0.115 N.S.
GD (20)				-----	0.138 N.S.	0.158 N.S.
GE (17)					-----	0.046 N.S.
GF (16)						-----

Group Cc = Control children group

Group (GA) = Pretreatment stage

Group (GB) = Post chemotherapy-pre radiotherapy

Group (GC) = During radiotherapy

Group (GD) = Post radiotherapy- during chemotherapy

Group (GE) = At the end of chemotherapy

(B) Serum T. ALP in children

Group	GA (25)	GB (25)	GC (22)	GD (20)	GE (17)	GF (16)
Cc (15)	7.161 v.h.s.i.	4.342 v.h.s.i.	1.925 N.S.	0.406 N.S.	0.04 N.S.	0.164 N.S.
GA (25)	-----	3.289 h.s.d.	5.802 v.h.s.d.	6.549 v.h.s.d.	7.231 v.h.s.d.	7.599 v.h.s.d.
GB (25)		-----	2.702 h.s.d.	3.703 v.h.s.d.	4.416 v.h.s.d.	5.312 v.h.s.d.
GC (22)			-----	1.336 N.S.	1.992 N.S.	2.315 s.s.d.
GD (20)				-----	0.447 N.S.	0.599 N.S.
GE (17)					-----	0.106 N.S.
GF (16)						-----

(C) Serum T.ALP + L.Ph.A in children

Group	GA (25)	GB (25)	GC (22)	GD (20)	GE (17)	GF (16)
Cc (15)	6.299 v.h.s.i	3.639 v.h.s.i.	1.526 N.S.	0.264 N.S.	0.393 N.S.	0.495 N.S.
GA (25)	-----	3.256 h.s.d.	5.551 v.h.s.d.	4.579 v.h.s.d.	6.724 v.h.s.d.	7.059 v.h.s.d.
GB (25)		-----	2.567 m.s.d.	3.167 h.s.d.	4.141 v.h.s.d.	4.522 v.h.s.d.
GC (22)			-----	1.106 N.S.	2.079 s.s.d.	2.440 m.s.d.
GD (20)				-----	0.634 N.S.	0.743 N.S.
GE (17)					-----	0.063 N.S.
GF (16)						-----

(D) Serum ALP % inh. In children

Group	GA (25)	GB (25)	GC (22)	GD (20)	GE (17)	GF (16)
Cc (15)	1.331 N.S.	1.259 N.S.	0.269 N.S.	0.152 N.S.	0.692 N.S.	0.459 N.S.
GA (25)	-----	0.029 N.S.	0.862 N.S.	1.764 N.S.	0.175 N.S.	0.795 N.S.
GB (25)		-----	1.077 N.S.	1.648 N.S.	0.654 N.S.	0.774 N.S.
GC (22)			-----	0.485 N.S.	0.469 N.S.	0.229 N.S.
GD (20)				-----	1.022 N.S.	0.674 N.S.
GE (17)					-----	0.182 N.S.
GF (16)						-----

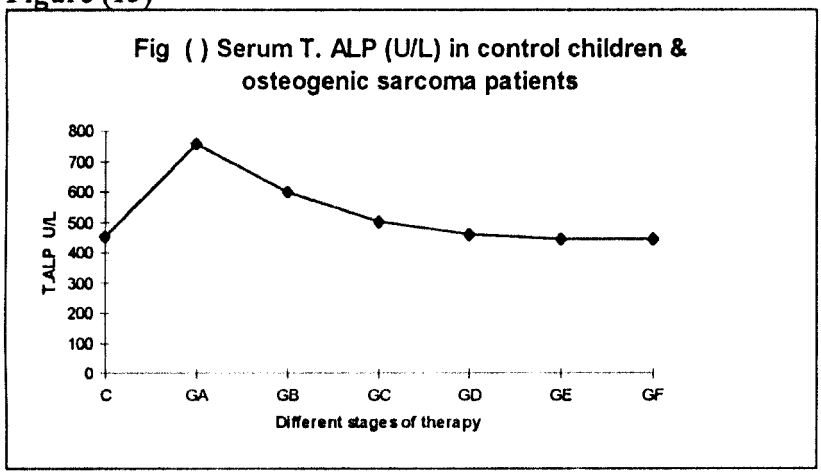
(E) Serum L.ALP in children

Group	GA (25)	GB (25)	GC (22)	GD (20)	GE (17)	GF (16)
Cc (15)	0.445 N.S.	0.395 N.S.	0.979 N.S.	0.813 N.S.	0.383 N.S.	0.139 N.S.
GA (25)	-----	0.098 N.S.	0.567 N.S.	0.370 N.S.	0.162 N.S.	0.712 N.S.
GB (25)		-----	0.766 N.S.	0.543 N.S.	0.062 N.S.	0.698 N.S.
GC (22)			-----	0.243 N.S.	0.962 N.S.	1.511 N.S.
GD (20)				-----	0.705 N.S.	1.293 N.S.
GE (17)					-----	0.748 N.S.
GF (16)						–

(F) Serum B. ALP in children

Group	GA (25)	GB (25)	GC (22)	GD (20)	GE (17)	GF (16)
Cc (15)	6.943 v.h.s.i.	4.154 v.h.s.i.	1.738 N.S.	0.528 N.S.	0.064 N.S.	0.134 N.S.
GA (25)	-----	3.351 h.s.d.	5.917 v.h.s.d.	6.608 v.h.s.d.	7.340 v.h.s.d.	7.775 v.h.s.d.
GB (25)		-----	2.803 m.s.d.	3.725 v.h.s.d.	4.510 v.h.s.d.	4.988 v.h.s.d.
GC (22)			-----	1.204 N.S.	1.918 N.S.	2.339 s.s.d.
GD (20)				-----	0.531 N.S.	0.786 N.S.
GE (17)					-----	0.244 N.S.
GF (16)						-----

Figure (13)



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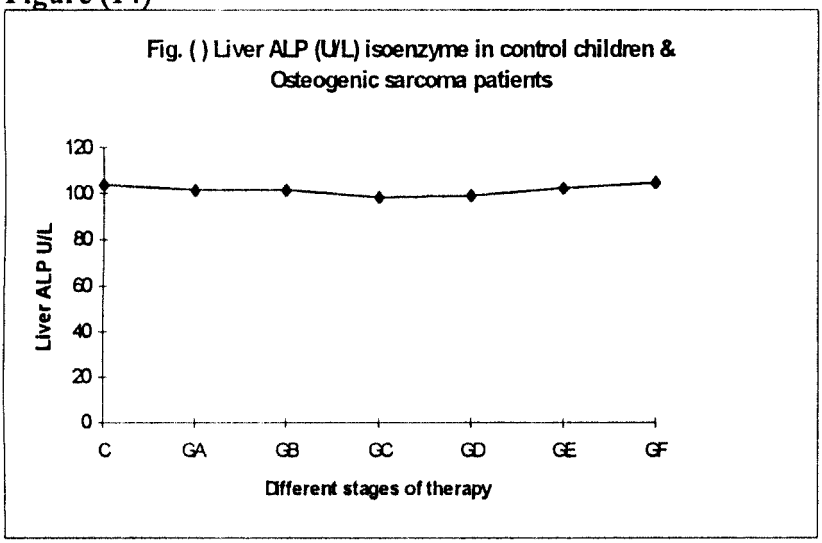

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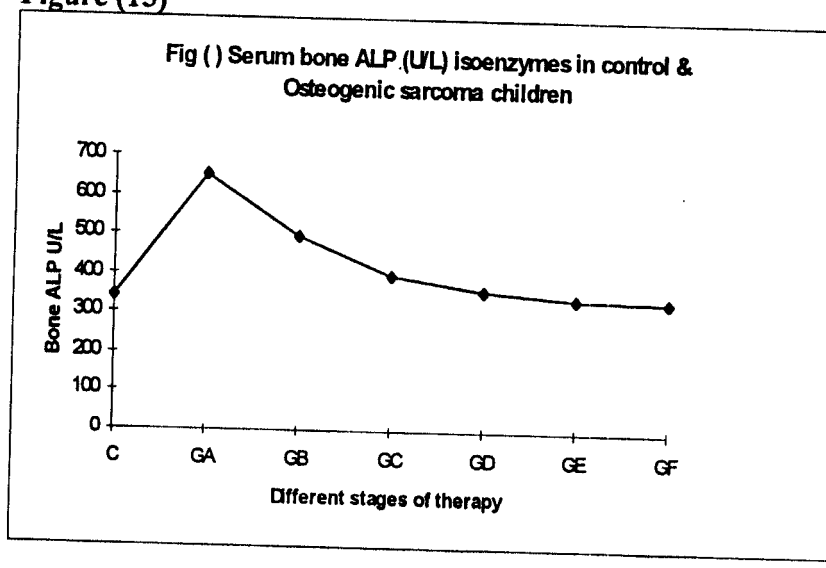

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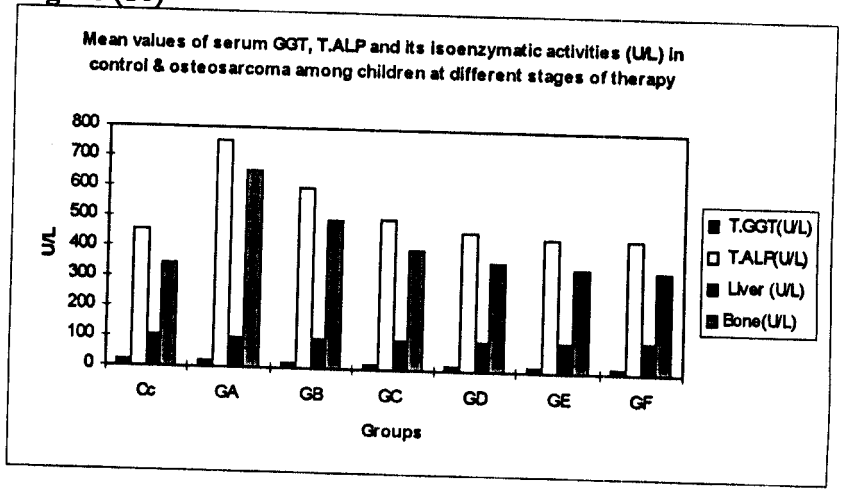


17)

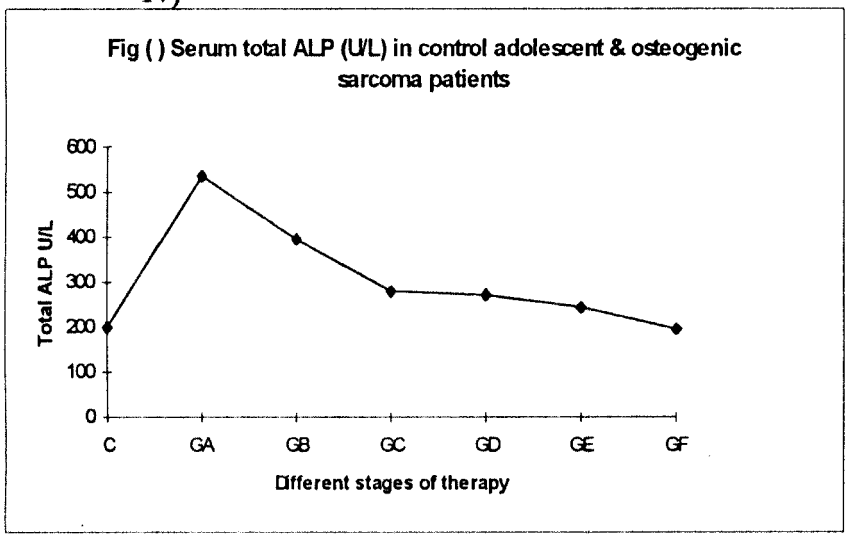


Fig ( ) Serum total ALP (U/L) in control adolescent & osteogenic sarcoma patients
Total ALP U/L
600
500
400
300
200
100
0
C
GA
GB
GC
GD
GE
GF
Different stages of therapy

Figure (18)

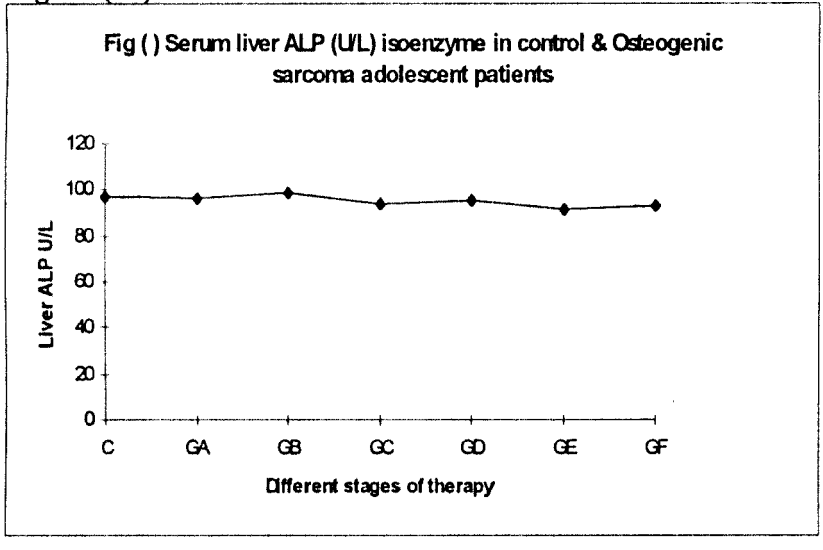


Fig ( ) Serum liver ALP (U/L) isoenzyme in control & Osteogenic sarcoma adolescent patients
Liver ALP U/L
120
100
80
60
40
20
0
C
GA
GB
GC
GD
GE
GF
Different stages of therapy

Figure (19)

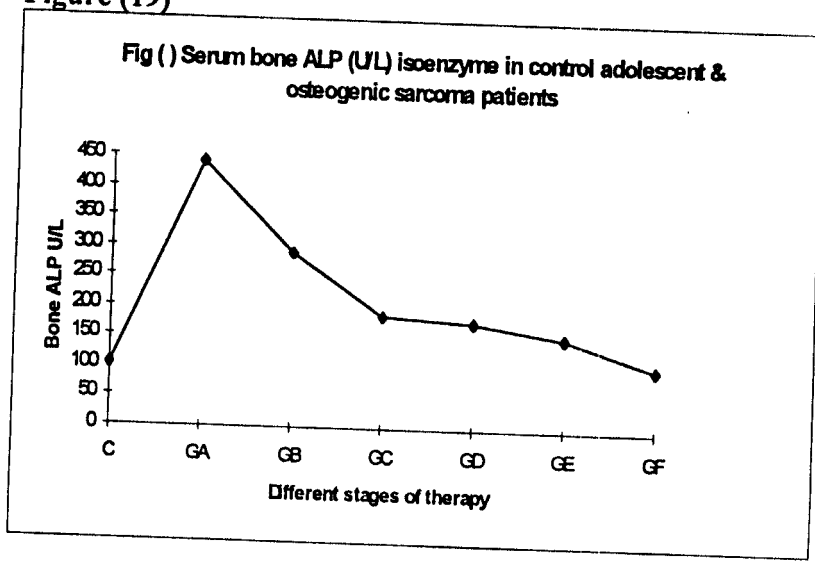
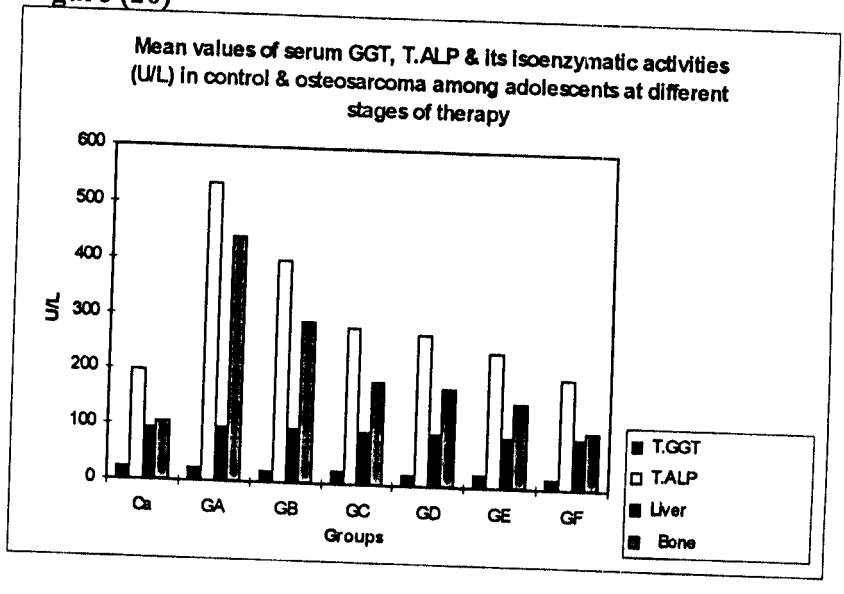


Figure (20)



Effect of partial digestion by neuraminidase on electrophoretic mobilities of L.ALP and B.ALP.

Lane 1, shows serum sample of a child (aged 10 years) with osteosarcoma without neuraminidase treatment it consists mainly of B.ALP. **Lane 2,** shows the same sample after partial digestion of the sialic acid residue by neuraminidase treatment.

Lane 3, shows serum sample of adolescent patient with osteosarcoma without neurominidase treatment, it consists mainly of B.ALP. **Lane 4,** shows the same sample after partial digestion of the sialic acid residue by neuraminidase treatment.

Lane 5, shows serum sample of healthy child without neurominidase treatment, it consists mainly of B.ALP. **Lane 6,** shows the same sample after partial digestion of the sialic acid residue by neuraminidase treatment.

Lane 7, shows serum sample of cirrhotic adult patient(as a marker for I.ALP) without neuraminidase treatment, it appeared L.ALP and B.ALP as one band and I.ALP separatly. **Lane 8,** shows the same sample after partial digestion of the sialic acid residue by neuraminidase treatment, the anodal stiuated zone is traces of L.ALP, while the less anodal is the B.ALP with I.ALP (as one band due to the I.ALP is not affected by neuraminidase treatment)..

Effect of partial digestion by neuraminidase on electrophoretic mobilities of L.ALP and B.ALP

Lane 1, shows serum sample of a child (aged 10 yaers) with osteosarcoma (before therapy) without neurominidase treatment, it consists mainly of B.ALP. **Lane 2**, shows the same sample after partial digestion of the sialic acid residue by neuraminidase treatment. **Lane 3**, shows serum sample of the same patient (before operation) without neurominidase treatment, it consists mainly of B.ALP. **Lane 4**, shows the same sample after partial digestion of the sialic acid residue by neuraminidase treatment. **Lane 5**, shows serum sample of the same patient (at the end of therapy) without neurominidase treatment, it consists mainly of B.ALP. **Lane 6**, shows the same sample after partial digestion of the sialic acid residue by neuraminidase treatment. **Lane 7**, shows serum sample of the same patient (during chemotherapy post operative) without neurominidase treatment, it consists mainly of B.ALP. **Lane 8**, shows the same sample after partial digestion of the sialic acid residue by neuraminidase treatment. **Lane 9**, shows the seum sample of healthy adult control (as a marker of L.ALP and B.ALP) without neurominidase treatment both L.ALP and B.ALP apeared as one band. **Lane 10**, shows the same sample after partial digestion of the sialic acid residue by neuraminidase treatment the anodal stiuated zone of L.ALP while the less anodal zone is the B.ALP.

DISCUSSION AND CONCLUSION

Discussion & Conclusion

The psychological aspects of childhood cancer

The diagnosis of childhood cancer forces a child to adapt to a new way of life. Coping with the social and emotional impact of cancer can be as difficult as contending with the medical treatments. Pain and discomfort associated with the illness and treatment, alteration in the physical appearance, social isolation, and fear of death create emotional reactions in the child and threaten normal development. The child's schooling may be blampered by absences or side effects of the disease and treatment. Support and guidance from the family and physician help the child adjust to illness, when sever problems arise or the child is experiencing special difficulties, referral to the psychologist or psychiatrist can be helpful (Copel and Worchel,1986).

The absolute number of long term survivors of childhood cancer is increasing at a truly remarkable rate. This represents major advances in surgery, radiation therapy, and chemotherapy treatment programs. Surviving the disease itself however, does not necessarily signal an end to the difficulties and stresses faced by these young patients and their families (Fochtman,1995).

The late medical sequel of cancer and its treatments can be frightening and restrictive, and the financial burdens incurred during treatment can be prohibitive, extending beyond the time of illness. Equally significant are the alterations in the life style, relationships, future plans, and intrapsychic equilibrium consequent to the living with a life-threatening disease. So there has been an increasing focus on issues relating to the quality of life and the psychological aspects of cancer care (Bond and Wellisch,1990).

The results of the present study revealed that, there were significant increase in the frequency of anxiety symptoms during the different stages of therapy (at diagnosis - at admission - at end of therapy) as compared to the control group, and there were no statistically significant different in the frequency of anxiety symptoms between the three stages, although the rate of anxiety symptoms in all patients at admission were higher (63.2% in Ewing's sarcoma & 64.5% in osteosarcoma) than the other stages of therapy, due to the small child who is separated from parents by hospitalization experience profound separation anxiety.

These results go with (Achte and Vauhkonen,1989) who interviewed 100 patients suffering from cancer, they found that tenseness, depression, and anxiety were the psychic symptoms that were discovered to be of most frequent occurrence. They were found to be present in more than half of total patients.

Lansky et al., in 1993 noted that, the child with cancer experiences more anxiety with the progression of the disease, and with each clinic visit. It is assumed that this increase in anxiety makes the child more liable to psychological problems. While (Lansky and Gendel, 1978) observed that in children with malignancies an extreme separation anxiety with regressive behavior which resulted in a symbiotic relationship with mother.

Bartholome in 1979 noted that, children in remission indicated that they had overcome the threat of death, it appeared that the anxiety associated with it was not dispelled. It is this chronic uncertainly about relapse and survival that poses a significant threat for the child living with cancer. Also (Copland and Worchel,1986) noted that, ending treatment for cancer also produces anxiety in children and their families. During treatment, they tend to feel

protected against the disease and understandably, they become emotionally attached to the medical staff and medication. Because they know that cancer may recur, stopping treatment makes them feel more vulnerable.

These results were not go with (Kellerman et al.,1980) in their study 349 adolescents were compared with 168 adolescents with various chronic or serious disease including cancer on standardized measures of trait anxiety, self-esteem, and health locus of control, no difference in anxiety or self esteem were found between healthy and ill groups or between various ill groups.

As regard the neurotic traits the results showed high incidence of neurotic traits among the anxiety positive group of all patients with Ewing's sarcoma or osteosarcoma which were statistically significant. The most common trait in Ewing's sarcoma and osteogenic sarcoma respectively was nervousness (88.9% & 71.4%), then eating troubles (77.8% & 78.6%), and night mares and terrors (77.8% & 64.3%), other traits like bed wetting, thumb sucking, stuttering, somnambulism and encopuresis were not significant in the anxiety group. From this results most of patients with anxiety symptoms, experienced one or more neurotic traits.

While the incidence of neurotic traits in all patients with Ewing's sarcoma or osteogenic sarcoma were estimated respectively, Nervousness (52.4% & 45.7%), then eating troubles (42.8% & 45.7%), and night mares and terrors (42.8% & 34.3%), nail biting (33.3% & 37.1%) were the most common neurotic traits affecting the patients in order of frequency and there were no statistically significant difference in the frequency of the neurotic traits between Ewing's sarcoma and osteogenic sarcoma patients.

These results go with Zeltzer (1980), who noted that any child diagnosed with cancer may at times appear depressed. The child may have troubles in sleeping or eating, be excessively anxious or sad, or be silent and withdrawn. These behaviors often reflect the shock, fear, distress, and exhaustion accompanying the diagnosis of cancer. However, if symptoms are persistent, continuing after remission is achieved, and include uncharacteristic feeling of hopelessness and worthlessness, further evaluation is wanted.

Lesko & Holland in 1990 noted that when individual receive a diagnosis of leukemia, they experience a characteristic but normal period of "shock" disbelief, denial, emotional unrest with depressive and or anxious symptomatology, irritability, and disruption of appetite or sleep are common, these normal psychological symptoms usually resolve in two weeks. But some patients continue to exhibit greater than normal levels of anxious and depressive symptoms, which can persist for weeks to several months, such persistent reactive distress frequently requires psychiatric treatment.

Lansky et al., in 1993, noted that in the school age child, diagnosis and initiation of therapy arouse many feelings and fears. Separation, strange people, an unfamiliar environment, fears of abandonment and punishment, and threats to body integrity are all major concerns of school age child. They cope with the stress of illness in a variety of ways, they may have a delayed initial reaction or may respond immediately with acute anxiety or panic. Other reactions include psychosomatic complaints, nightmares and terrors, labile emotion, regression in the form of loss of newly acquired skill (e.g. Toilet training, speech and self feeding) and previously discarded behaviors, such as thumb-sucking or clinging, may re-emerge.

Daud et al., 1994, noted that children with non-neurological physical illness are particularly prone to emotional symptoms and eating troubles, rather than antisocial behavior. While eating anomalies may be explained by poor appetite and heightened maternal concern, especially in pre-school children.

Redd ,1994 noted that, in addition to fears of recurrence, a major issue for children who have been treated for cancer is social isolation and reintegration. In a recent longitudinal study of adaptation in adolescents with cancer, he found that young survivors differed from peers primarily in the level of social anxiety and shyness. Teenage survivors appeared to be more socially isolated than were their peers. However they were in different in terms of overall popularity, depression, loneliness, or self concept.

In this study, we have tried to assess the different personality traits in patients with Ewing's sarcoma or osteogenic sarcoma. The comparison was done between boys, girls and control group regarding the four aspects of the JEPQ. Neuroticism in all patients (Ewing's sarcoma & osteosarcoma patients) was significantly high in both boys (61.5% & 55%) and girls (62.5% & 60%) respectively, than the control group.

The finding of high neuroticism in patients with cancer is of great importance as it confirmed that the patients had high score of emotionality, also it confirmed the Eysenck and Eysenck,1964 description of child with high neuroticism score as being anxious, worrying individual, moody, frequently depressed, likely to suffer from various psychosomatic disorders, with strong emotional reactions that interfere with the proper adjustment and that make the individual to react irrational and some time with rigidity.

As regard to extroversion in patients with Ewing's sarcoma, it was higher in girls (50%) than both boys and control group, and in patients with osteogenic sarcoma, it was high in both boys (40%) and girls (46.7%) than the control group, and the difference between boys and girls in all patients with Ewing's sarcoma or osteogenic sarcoma was not statistically significant. This could be explained on bases that extroversion is affected more as physical impairment become worse (i.e. as lost limb or deformed limb) becomes deviant from normal standard.

Psychocitism was non significant in both boys and girls in all patients with Ewing's sarcoma or osteogenic sarcoma compared with the control group.

While as regard the lie scale in all patients with Ewing's sarcoma or osteogenic sarcoma, it was significant higher in girls (62.5% & 60%) respectively than both boys and control group, this could explained on the fact that there is a close association between increasing impairment and increasing lie scores as denial or ignoring of the unpleasant symptoms or traits may be frequently used by the impaired patients to enable them to carry on with their life despite the serious obstacles present. We revealed from the previous results that, there were more than one traits could be present in the patient at the same time.

These results go with Achte & Vauhkonen in 1989, who interviewed 100 patients suffering from cancer, they found tenseness, depression and anxiety were the psychic symptoms that were discovered to be most frequent occurrence. They were found to be present in more than half of total patients. Aggressiveness and paranoid attitude were also quite frequent, a fear of death was

present in half of patients in the series, phobic, obsessive or neurotic symptoms were not very common.

As regard to the frequency of psychiatric disorders in patients with Ewing's sarcoma or osteogenic sarcoma, the results revealed that, the behavioral and emotional disorders were the most prevalent disorders among the patients, and there were more than one diagnosis of psychiatric disorders could be present at the same time in one patient.

These results go with Katz,1980 who noted that, the outcome of the child's coping efforts with total set of threats associated with cancer has drown mush attention because of its role in the development of psychosocial problems. Behavioral problems and emotional disturbances have often been reported such as anxiety, fear, depression, extreme dependency on the parents, sleep disturbances, regression, anger and withdrawal.

Derogatis et al.,1983 found that, by assessing ambulatory and hospitalized patients in three cancer centers using standard criteria, 53% of patients interviewed through showing signs of being under stress were coping adequately. The remaining 47% had a diagnosable psychiatric disorders, the most common by far was adjustment disorder with anxious and depressive symptoms. Depression was next, seen in 13% of those with a psychiatric diagnosis, central nervous system complications resulting in organic mental disorders were present in 4%, prior psychiatric disorders account for only 5% of cases. More recently, Holland & Rowland in 1989, studied the spectrum of psychiatric disorders in patients with cancer adjustment disorders were encountered in 30% of the study group.

There is no question that the study of psychosocial factors is important in pediatric oncology. Research during the last 10 years has documented this fact and has resulted in major changes in how treatment is given and how medical treatment and psychosocial support are provided. Clinician have become increasingly alert to the multiple and varied implications of childhood cancer, the child with cancer is now recognized as a child at risk for future difficulties-medical, psychiatric, and psychosocial. Coping with and adjusting to life-threatening disease significantly alerts a child's life. The child and family are challenged by the immediate crises and developmental disruptions as well as the spectrum of long-term sequelae.

This study highlights the need for a comprehensive and preventive approach that begins with early assessment and continues with ongoing evaluation of stress throughout all phases of illness. Each phase raises unique concerns, ranging from acute anxiety and fear at diagnosis to difficulty in resuming previous activities during treatment and remission to the uncertainties of long-term survival. The medical care team's attention to these changing needs and use of appropriate psychological and psychiatric support measures will minimize immediate distress and facilitate the child's future adjustment.

Serum alkaline phosphatase and its isoenzymatic activities in osteogenic sarcoma

Malignant tumors that arising from the skeletal system are rare. Their incidence varies according to age, osteosarcoma is the most frequent primary malignant bone tumor, it is approximately three times more frequent than Ewing's sarcoma. Only half of bone tumors in childhood are malignant of these osteogenic sarcoma which is the most frequent, accounting for approximately 60% of malignant bone tumors in the first two decades of life. Ewing's sarcoma is relatively common and accounts for 10-15% of all primary malignant bone tumors, Ewing's sarcoma, the second most frequent bone cancer is actually more common than osteosarcoma in children younger than 10 years (Mokhtar,1991).

Now, the field of biological marker of malignant bone tumors has been the focus of interest for their applicability in the diagnosis, response to treatment and in prognosis of the disease (Kaplan et al.,1995). The total plasma alkaline phosphatase level has long been recognized as an indicator of osteoblastic activity, but lack of specificity makes it an insensitive index of the progress of disease and the response to treatment, measurement of plasma bone-specific alkaline phosphatase activity is a sensitive and reliable measure of osteoblastic activity (Leung et al.,1993).

In osteosarcoma, with the introduction of adjuvant and neoadjuvant chemotherapy combined with surgery substantially increased the cure rate of patients with osteosarcoma of the extremities (Bacci et al.,1990). Despite the success of this combined treatment, metastases developed in approximately 30-40% of patients who then die of the disease. Therefore, to improve the disease-free survival, it would be important to be able to

identify patients with a poor prognosis who could be treated with more aggressive therapy (Campanacci et al.,1981).

Prognostic clinical factors previously analyzed, such as size and location of the tumor, age and sex of the patients, duration of the symptoms before diagnosis, histologic features, soft tissue invasion, haemoglobin values and lymphocytic counts gave contradictory results (Hudson et al.,1990), so it is surprising that recent articles concerning the study of prognostic factors in osteosarcoma did not consider pre-treatment values of serum alkaline phosphatase (ALP). It has been reported that in patients with osteosarcoma, the high pre-treatment levels of this enzyme were associated with a worse prognosis and that during follow up, an increase in the serum ALP (which usually declines to normal levels when the tumor is surgically removed) heralds the appearance of local or distant metastases (Bacci et al.,1993).

The aim of the present work is to evaluate the use of pre-treatment of serum total ALP levels as a prognostic indicator in 35 patients with a localized osteosarcoma, treated with combined surgery and chemotherapy (adjuvant and neoadjuvant chemotherapy), serum ALP levels were also evaluated after surgery and during the course of chemotherapy to verify whether persistent high levels of serum ALP herald the appearance of a recurrence.

The results of this study showed that, serum ALP levels varies by the age in the control groups. The serum ALP levels in the children group (aged from 8 to 12 years) was 317-551 U/L and in adolescents group (aged from 13 to 20 years) was 131-294 U/L, and there is no significant difference in the serum ALP between the males and females and it may be due to the small number of the cases in the control groups. Such changes in the serum ALP were

rapid skeletal growth, these values estimated according to the Scandinavian recommended method at 25 °C.

In 1988 Dolland found that, the alkaline phosphatase activity in serum increases during the first few months of life to reach a level up to 2.5 times the upper reference limit for adults. This level is maintained throughout childhood. With the onset of the adolescent growth spurt there is a gradual rise in ALP activity followed by a fall to adult levels towards the beginning of the third decade. This process starts at approximately 11 years old in females and is completed by the age of 20. The changes occur later in males and they do not reach adult level till well after the age of 20. During puberty, peak ALP activities show a better correlation with sex-maturity ratings than with chronological age.

As the units for a method vary with the conditions of the reaction such as type of substrate and its concentration, the type of buffer, pH, temperature and concentration of activator, it will be evident that for alkaline phosphatase there will be quite a number of normal ranges, laboratories are recommended to develop reference ranges for the populations they serve (Doland,1988) .

In this study at presentation, the serum levels of ALP in 25 children with osteogenic sarcoma were elevated in 19 patients (76%), with a serum levels ranged from 584 to 1089 U/L and normal in 6 patients (24%), with a serum level ranged from 440 to 554 U/l as compared to the serum levels of ALP in the healthy children control group. While in 10 adolescent patients the serum levels of ALP were elevated in all patients (100%) with a serum level ranged from 396 to 833 U/L as compared to the serum level of ALP in the healthy adolescent control group.

This increment in the serum ALP was mainly due to increase in the bone fraction of the ALP, and the serum γ- glutamyl transferase activity concentrations were within the normal range (serum levels ranged from 11 to 30 U/L), so the data revealed that the increase in the serum total ALP was mainly due to the increase in the bone fraction of ALP as confirmed by " heat inactivation and electrophoretic separation". while the liver fraction of ALP was in the normal ranges.

These results were in agreement with many authors, Bacci et al (1993), who found that at presentation, serum ALP levels were elevated in 66% of the patients and normal in 34% of the patients, the percentage of patients with high serum ALP levels was significantly higher in patients 20 years of age and older compared with younger patients (92.3% versus 58.9%). The fact that in the group of patients 19 years of age and younger the percentage of cases with high levels of serum ALP was lower than in the group of older patients has to be cautiously considered, because the activity of serum ALP varies with skeletal growth, in the younger patients the range of the normal values of the enzyme is much wider than in the older patients, this means that a serum ALP value in a patients with osteosarcoma probably has a different meaning in a 10-year-old patient and in a 20-year-old patient.

Thorpe et al.,(1979), found that elevated serum ALP levels have been reported in 40% to 80% of patients with osteosarcoma, and the increase of this enzyme has been referred to the osteoblastic activity of the tumor cells. Cultured osteosarcoma cells from humans and animals have been shown to produce large amounts of alkaline phosphatase that are released in the culture medium .

Van Straalen et al.,(1991), found that bone alkaline phosphatase is an enzyme localized in the plasma membrane of osteoblastic cells and may be considered as a marker of osteoblastic activity and of bone formation, respectively. Previously, it has been shown that bone ALP (as determined by electrophoretic separation) is superior to total alkaline phosphatase activity in the detection of osteoblastic activity.

Withold et al.,1995, reported that there is an increased in the bone ALP values in osteosarcoma patient's with high level of serum ALP and had a serum γ- glutamyl transferase activities within the normal reference limits, an increased fraction of bone ALP could be confirmed by electrophoretic separation.

in the pre-operative stage, most of children patients (15 cases) with high serum ALP levels at presentation and treated with neoadjuvant chemotherapy, the serum ALP levels decreased (the serum levels ranged from 968 to 580 U/L) but still higher than the normal reference limits, but in 10 cases the serum ALP levels returned to the normal limits (the serum level ranged from 556 to 381 U/L). While in adolescent patients, most of them in the pre-operative stage showed that the serum ALP levels decreased (the serum level ranged from 310 to 616 U/L) but still higher than the normal levels, except one case in whom, the serum ALP level returned to the normal value.

In the post-operative stage, the serum ALP levels in the children patients decreased (the serum level ranged from 580 to 651 U/L) but not returned to the normal limits in 6 patients with no evidence of metastases, and in most of patients (16 cases) the serum ALP levels decreased and nearly returned to the normal levels (the serum level ranged from336 to 559 U/L), one case (case no. 4) had

an elevated serum ALP post operatively with a serum level of 984 U/L which proved to be metastatic later.

While in adolescent patients, most of patients showed that, the serum ALP levels decreased (the serum level ranged from 351 to 512 U/L) but not returned to the normal values with no clinical evidence of metastasis, except 3 patients in whom the serum ALP levels returned to the normal values(the serum level ranged from 168 to 300.9 U/L).

These increments in the enzyme levels may be due to the tumor activity or due to normal bone reaction to the surgical procedures.

During chemotherapy post-operatively till the end of chemotherapy, the serum ALP levels returned to the normal values in the children patients whom continued therapy (16 cases) except 4 cases which had an increase serum level of ALP irrespective the treatment with chemotherapy, they proved to develop metastases later.

While in the adolescent patients, the serum ALP levels returned to the normal values in most patients, except one case (case no. 3) in whom the serum ALP increased with no evidence of metastasis but developed metastases later.

During follow up, the serum ALP in the remaining children patients returned to the normal values in all patients except one patients (case no.16) who had an elevated serum ALP levels and proved to develop metastasis. While in adolescent patients, the serum ALP levels returned to the normal values in the remaining patients.

These changes in the serum ALP levels were mainly due to changes in the bone fraction of ALP as detected by heat inactivation and confirmed by electrophoreses, and all patients had normal γ glutamyl transferase levels. While the liver fraction of ALP was in the normal ranges in all patients.

These result were in agreement with Bacci et al 1993, who reported that, in 304 of 337 patients with localized disease and high serum ALP levels at presentation, the enzyme level returned to normal after surgery. In most patients treated with neoadjuvant chemotherapy, the values were normal after preoperative chemotherapy. In the remaining 33 patients, serum ALP levels decreased after surgery but remained elevated more than the normal levels. It is not easy to establish whether the elevated serum ALP levels were due to the tumor or to the reaction of the normal bone cells to the surgical devices or to the grafts in patients who underwent limb-sparing surgery.

While Rosen et al 1974, reported that the association of elevated (bone) ALP in the serum of patients with osteosarcoma was the first evidence that the tumor cells themselves produce the enzyme. The amount of phosphatase present gives valuable clinical information as to the degree of tumor destruction produced by the therapy, changes in the tumor activity may frequently be detected by this means before other clear-cut physical signs are manifested. The reduction of elevated serum ALP levels Following chemotherapy is a valuable guide to administration of therapy.

The prognosis appeared to be influenced by the pre-treatment values of serum ALP, our results revealed that, the percentage of patients who experienced metastases were 26.3% in children patients and 10% in adolescent patients, and all patients had a

significant high serum ALP levels at presentation. So we can conclude that the pre-treatment serum ALP could be used as a prognostic tool for early relapse in osteosarcoma.

These results were in agreement with Bacci et al 1993, and Rosen et al 1974, they reported that the patients with elevated pre-treatment serum ALP levels had a worse prognosis than those with normal values (55% of relapse versus 26%) and among patients with elevated pre-treatment serum ALP levels, there also appeared to be a correlation between serum levels of the enzyme and prognosis (i.e. the higher the serum ALP levels, the greater the risk of relapse).

While the percentage of metastases in children patients with elevated serum ALP levels and normal serum ALP levels after surgery were 57%, versus 0.06%, and the percentage of metastases in adolescent with elevated serum ALP post operatively was 10%, so there were also a significant higher incidence for metastases in patients with elevated serum ALP post operatively.

These results due to its small sample size were not in agreement with Bacci et al 1993, Thorpe et al 1979, and Bentzen et al 1988, they reported that in almost patients with elevated pre-treatment serum ALP levels, a decrease to the normal levels of the enzyme was observed after surgery, but they observed that the percentage of relapses in patients with elevated serum ALP after surgery was not significantly higher than the percentage observed in the remaining patients in whom the enzyme levels returned to normal levels after surgery. The percentage of relapses was 51.9% in 304 patients who had normal serum ALP levels after surgery and 84.8% in 33 patients with still-elevated serum levels after surgery, this difference was not statistically significant.

In the view of these results, the clinical usefulness of pre-treatment serum ALP levels in predicting the course of the disease in patients with localized osteosarcoma should be carefully considered, and the response to the treatment can be assessed by the evaluation of the serum ALP levels after each course of chemotherapy. Serum ALP levels should certainly be analyzed when comparing the results achieved in this tumor with different therapeutic protocols in unrandomized studies or in planning new randomized clinical trials. Perhaps a more aggressive treatment program, although implying a higher toxicity risk, could be justified for patients with high levels of serum ALP (Bacci et al., 1993).

Serum Lactate Dehydrogenase and its isoenzymatic activities in Ewing's sarcoma

Recently, the relationship between neoplasm and lactic dehydrogenase (LDH) has been studied with increasing intensity. The glycolytic enzyme LDH is a marker for the cytosol in numerous tissues and serum levels can become elevated in many disease. High serum levels of the enzyme have been observed in patients with solid tumors, leukaemia and lymphoma (Bacci et al.,1985).

Serum total LDH levels have been recently found to be useful as prognostic markers in several neoplastic diseases, particularly in non-Hodgkin's lymphoma. Controversy around the use of LDH as a tumor marker In Ewing's sarcoma, two studies of a patient population with Ewing's sarcoma at the National Cancer Institute have shown that serum LDH levels have prognostic value in determining the probability of survival in those patients (Glaubiger et al.,1980 and Brereton et al.,1975). Two other studies, from Memorial Hospital, showed that the initial LDH level had no prognostic value in patients with Ewing's sarcoma treated with adjuvant chemotherapy (Rosen et al.,1981 and Farley et al.,1987).

The aim of the present work is to evaluate the use of serum LDH levels and its isoenzymatic activities in 21 patients with localized Ewing's sarcoma as a prognostic factor and to monitor the response of those patients to therapy.

The results of this study showed that the serum total LDH activity in the children control group (age range 8 to 12 years) was 254 to 350 U/L at 37 °C and in the adolescent control group (age range 13 to 20 years) was 269 to 390 U/L by using the

Scandinavian recommended method. The mean percentage distributions for serum LDH isoenzymes which were separated on agarose gel in children control group revealed values of 30.67% for LD1, 37.47% for LD2,18.19% for LD3, 8.05% for LD4 and 5.41% for LD5, and in adolescent control group revealed values of 30.95% for LD1, 39.39% for LD2, 17.46% for LD3, 6.57% for LD4 and 5.63% for LD5. As shown there were no statistical significant difference between the two control groups neither the total serum LDH nor in its isoenzyme activities.

Tietz 1995, who reported that The total serum LDH levels in healthy group aged from 2 years to 12 years was 110 to 295 and at 37 °C and in group aged from 13 years to 60 years was 110 to 190 U/L and the isoenzymatic distribution which were separated on starch gel showed values of 14% to 26% for LD1, 29% to 39% for LD2, 20% to 26% for LD3, 8% to 16% for LD4 and 6% to 16% for LD5.

Afify 1990, reported that the total serum LDH levels in healthy children ranged from 247 to 410 U/L at 37°C and the mean percentage distribution for LDH isoenzymes which were separated on agarose gel revealed values of 32.41% for LD1, 44.48% for LD2, 16.06% for LD3, 5.13% for LD4 and 3.92% for LD5.

Tietz 1990, reported that, values for serum LDH activity vary considerably, depending on the direction of the enzyme reaction, the type of method used, and the experimental parameter, for the P $\Rightarrow$ L reaction at 30 °C and at PH 7.4 a range of 95 to 200 U/L represent the experience of most workers. Using the Scand. Assay the total serum LDH levels in healthy adult group ranged from 195 to 250 U/L and the mean of percent distribution for LDH isoenzymes which were separated on cellulose acetate revealed

values of 27% to 35% for LD1, 34% to 44% for LD2, 16% to 22% for LD3, 4% to 8% for LD4 and 3% to 7% for LD5.

It was observed that different values were obtained either the mean or range, the difference between these values could be attributed to some reasons:

(a) Different supporting media used such as cellulose acetate, agarose, and starch gel.

(b) collection and storage of samples, and

(c) different population.

So, it is advisable that each laboratory should determine its own normal range.

At presentation, the results of the this study showed that. Among the 21 patients with localized Ewing's sarcoma, the serum total LDH level was normal in 7 cases (33%) (serum level ranged from 279 to 329 U/L), as regards to the isoenzymes there were a normal distribution and values of all the isoenzymes. And the serum LDH levels were elevated in the other 14 cases (67%) (serum level ranged from 489 to 813 U/L). This such increment was mainly due to the increased in the serum LDH isoenzyme activities of LD2, LD3, and LD4 with mean values of 179.64U/L (34.04%), 133.54 U/L (25.39%), and 83.87U/L (15.05%) respectively.

The explanation for an elevated serum LDH level in absence of liver metastases is unclear, it may reflect the burden of neoplastic cells or may be due to basic differences in proliferation and turnover of malignant cells in the different cases. There is some evidence for this latter hypothesis and leakage or secretion of LDH into general circulation by tumor cells has been reported (Wilkinson., 1970).

These results were in agreement with Farley et al 1987, reported that, among 56 patients with localized Ewing's sarcoma 32 (57%) had normal serum LDH level and 24 (43%) had elevated serum LDH level. The explanation for the elevated serum LDH in patients with Ewing's sarcoma was that, rapidly dividing tumors convert to an anaerobic metabolism, the measurement of serum LDH is an indicator of anaerobic metabolism, and serum LDH levels are elevated in patients with aggressive tumors.

Bacci et al 1985, reported that, elevated levels of serum LDH were observed in 26 out of 59 (44%) patients with localized Ewing's sarcoma at admission, and 33 (56%) patients had normal serum LDH.

Zondag and Klein 1968, reported that , the serum LDH isoenzymatic distributions in Ewing's sarcoma remains essentially normal with the principal activity in LD2, LD3, and LD4 .

Rosen et al.,(1981), suggested that, measurement of total serum LDH is a non-specific measurement of tumor activity. LDH isoenzymes LD2 and LD3 have been found to be specifically associated with tumor activity in lymphoma. These isoenzymes measured on presentation may be a prognostic factor in patients with Ewing's sarcoma.

Talageri et al.,(1971), reported that , the serum LDH isoenzyme pattern may reflect the primary tumor or as is more often the case, the release of isoenzymes from damaged normal tissue adjacent to the tumor. Studies on the LDH isoenzyme distribution of different types of malignant tumors have shown that these tissues, irrespective of tissues of origin contain mainly cathodic isoenzymes namely LD3, LD4, and LD5. However, the serum isoenzyme

distribution in Ewing's sarcoma showed activity mainly confined to LD2, LD3 and LD4 with no significant increase in LD5.

During chemotherapy before the local control, the results of this study showed that, the elevated serum LDH level in patients with Ewing's sarcoma at presentation decreased (serum level ranged from 264 to 489 U/L) after the 3 courses of chemotherapy before the local control of the tumor, but still higher than the normal limits, and as regards to the isoenzymes, there were decreased in the activity of LD2, LD3, and LD4 with the mean values of 123.62 U/L for LD2, 82.65 U/L for LD3, and 39.58 U/L for LD4 respectively.

During radiotherapy, the decreased serum LDH level after the 3 courses of chemotherapy showed an increase in the serum LDH level (serum level ranged from 339 to 560 U/L), These increments were due to the destructive effect of radiotherapy on the tumor cells as well as the adjacent normal tissues. These such increment was mainly due the increase in the LDH isoenzymes LD2, LD3, LD4, and LD5 with the mean values of 147.15 U/L for LD2, 97.88 U/L for LD3, 49.32 U/L for LD4, and 36.19 U/L for LD5. The increase in LD5 isoenzyme although there is no liver metastases may be due to the destructive effect of the radiotherapy to the normal adjacent tissues, usually it is skeletal muscles which are rich with LD5 isoenzyme.

During chemotherapy after the local control of the tumor (radiotherapy), the results of this study showed that, the serum LDH levels decreased gradually till reached the normal levels at the end of treatment. There were 12 cases 71% from the 17 cases who continue the treatment cured and had a normal serum LDH level at the end of treatment and during follow up after 6 months, and 5

cases 29% showed persistent elevation of serum LDH level during and after the treatment which developed metastases later.

It is worth mentioning that, Farley et al.,(1987), reported that during treatment, liver function tests, including the serum LDH levels secondary to the chemotherapeutic agents, show a transient rise. This false-positive rise in the serum LDH levels can not be correlated with the tumor activity. By the end of chemotherapy, serum LDH levels fall to normal values.

Bacci et al.,(1988), reported that, in some patients during the treatment a transient rise in the liver function tests including serum LDH levels . This false positive rise in LDH levels, unrelated to the tumor activity, always returned to normal within 3-10 days after the end of the cycle of chemotherapy. Also they reported that, in 89% of patients who presented with elevated serum LDH levels at presentation, the enzyme reached a normal value after the local control of the tumor (radiotherapy or surgery). In the remaining 11% of patients, the serum LDH level fell after local control of the tumor but never reached a normal level, all of these patients developed metastatic disease within 3-15 months from the beginning of the therapy.

The prognosis of this study, the incidence of relapse in the patients with normal serum LDH levels at presentation was 14.28% (one patient out of 7 patients developed chest metastases), while in patients with elevated serum LDH levels at presentation was 28.85% (4 patients out of 14 patients, 3 patients developed chest metastases during the course of treatment and one patient developed bone metastases during follow up after the end of treatment by 6 months). So serum LDH levels at presentation could be used as a prognostic tumor marker.

These results were in agreement with Breterton et al 1975, reported that 10 of 32 patients (31%) with normal initial serum LDH levels had tumor recurrence and 5 of 15 patients (33%) with elevated serum LDH levels had recurrent disease, there is no statistical difference in the recurrence rate between the two groups, with sample size of 32 patients and 15 patients, however, the ability to detect small to moderate differences in disease recurrence was limited.

The importance of serum LDH values at presentation is controversial, Glaubier et al 1980, reported a 6% rate of disease free survival in 31 patients with high serum LDH levels at presentation compared with a 60% rate in 45 patients with normal serum LDH levels. On the contrary, Rosen et al 1981, who compared 25 patients with high serum LDH levels with 42 patients with normal serum LDH levels, and Farley et al 1987, who studied 13 patients with high serum LDH levels and 32 patients with normal serum levels, found no difference in the prognosis.

Bacci et al.,(1988), reported that among the patients with localized Ewing's sarcoma 117 patients (58.7%) had normal serum LDH levels and among these patients 46 patients (39.3%) relapsed, while there were 82 patients (41.3%) had elevated serum LDH and among these patients 56 patients (68.2%) relapsed. The difference in the relapse rate between the two groups of patients (normal versus elevated enzyme level at admission) was statistically significant, so they reported that in non metastatic Ewing's sarcoma, among the patients with elevated pre-treatment LDH serum level there was a fairly good correlation between the amount of the enzyme and the prognosis, i.e. the high the serum LDH level, the higher the rate of relapse.

In the view of these results, we feel that the clinical usefulness of pre-treatment of serum LDH levels in predicting the course in the patients with localized Ewing's sarcoma should be considered carefully. In patients with elevated serum LDH levels there is usually a return of the enzyme to normal levels after the treatment, the lack of this response in our patients was always followed by an early recurrence. In addition, rising serum LDH levels after transitory normalization following the treatment often observed in patients who subsequently relapsed. We conclude that in Ewing's sarcoma the initial serum LDH levels is an important prognostic factor and could be used both to monitor the response of the patients to therapy and in comprehensive post-treatment follow up program to signal early tumor recurrence (Bacci et al., 1988).

SUMMARY

Summary

The present study dealt with estimation of serum ALP, LDH, and their isoenzymatic activities in patients with osteogenic sarcoma and Ewing's sarcoma respectively, at the presentation of the disease, during the course of treatment but before the administration of the cycle of chemotherapy and at follow up after 6 months, since the determination of serum iso-enzymatic activities of these enzymes proved to be of greater organ specificity and increased sensitivity in comparison to total activity.

Also this study dealt with the impact of cancer and its treatments on the psychological and social functioning of the patients, and the range of normal and abnormal psychological responses to cancer is a guide for the oncologist in diagnosis, management, and referral.

Our aim was to study the serum ALP isoenzymes in osteosarcoma and serum LDH isoenzymes in Ewing's sarcoma diseases, to give an idea on the prognosis and to provide an easy, less costly means for the detection of the response to therapy. And to study the psychological impact of cancer on the patients.

This study was performed on 86 cases who were classified into the following groups.

****Control group**

It included 30 healthy controls, who were classified according to age into (15 children with age range 8-12 yr., 15 adolescents with age range 13-20 yr.)

**Osteosarcoma group

It included 35 patients, who were classified according to the age into (25 children patients, and 10 adolescent patients).

**Ewing's sarcoma group

It included 21 patients with age range of 8-16 years.

All patients were subjected to the following

*full clinical examination, laboratory diagnosis (C.B.C, L.F.T, R.F.T, B.M.A, E.S.R), Histopathological analysis of the bone biopsy, radiological diagnosis (bone and chest X ray, C.T scan of the bone and chest, M.R.I of the bone when indicated).

*Psychological assessment with standard psychiatric sheet history, anxiety scales sheet and detection of personality traits of the patients through the Junior E.P.Q.

*Serum ALP activity in patients with osteogenic sarcoma was kinetically determined according to the Scandinavian recommended method at 37°C. The ALP isoenzymatic activities were identified and quantitated by three methods; a) separation on PAGE b) heat inactivation, and c) chemical inhibition by L-phenylalanine.

*Serum LDH activity in patients with Ewing's sarcoma was kinetically determined according to the Scandinavian recommended method at 37°C. The LDH isoenzymes were separated by agarose gel electrophoresis and quantitated by scanning at 570 nm.

The results revealed a highly significant increase in the frequency of anxiety symptoms in patients with osteogenic sarcoma, and Ewing's sarcoma at the diagnosis of the disease and at the end of therapy, while the frequency of anxiety at admission to the hospital revealed a very highly increased, because a small child who is separated from parents by hospitalization experiences profound separation anxiety, and there were no significant difference between boys and girls.

Regarding to the neurotic traits in all patients with Ewing's sarcoma and osteogenic sarcoma, the results revealed the nervousness, eating troubles, bed wetting, night mares, and terrors and thumb suckling were the most common neurotic traits affecting the patients in order of frequency.

Regarding to the personality make up of the patients with osteogenic and Ewing's sarcoma the results showed that, the neuroticism was significant high in both boys and girls as compared to the control group, this could be explained on the bases of that the patients had high score of emotionality. As for extroversion-Introversion personality trait in patients with Ewing's sarcoma was higher in girls than in boys, and it was high in both boys and girls in patients with osteogenic sarcoma as compared to the control group, this could be explained on bases that extroversion is affected more as physical impairment becomes worse, i.e. as lost limb or deformed limb becomes more deviant from the normal standards. As for the psychoticism was low in both boys and girls in all patients. As for the lie scores in all patients were significant higher in girls than boys as compared to the control group, this could explained on the fact that there is a close association between increasing impairment and increasing lie scores as denial or ignoring of the unpleasant symptoms or traits may be frequently

used by the impaired patients to enable them to carry on with their life despite the serious obstacles present.

The frequency of different psychiatric disorders in patients with Ewing's sarcoma and osteogenic sarcoma showed that, there were more than one diagnosis could be present at the same time, the behavioral and emotional disorders were the most prevalent disorders.

Regarding the total serum LDH levels in patients with Ewing's sarcoma the results showed that, the serum levels increased significantly than the normal level of the control group, this increments were mainly due to increased activity of the iso-enzymes LD2, LD3, and LD4. These increment was mainly due to the secretion of LDH into the general circulation by the tumor cells because of the rapidly dividing tumors convert to an anaerobic metabolism, and measurement of serum LDH is an indicator for anaerobic metabolism, while the isoenzymatic distributions remains essentially normal in Ewing's sarcoma with the principal activity in LD2, LD3, and LD4

During the course of therapy, the elevated serum LDH levels decreased gradually in response to therapy except during the radiotherapy, the serum level was increased, this increment was mainly due to the increased activity of the isoenzymes LD2, LD3, LD4, and LD5. This could be explained on bases of the destructive effect of the radiation on the bone and the adjacent normal skeletal muscles which were rich in LD5.

The serum LDH level reached the normal values in 12 patients (57%) who continue the treatment and not developed metastasis, and there were 4 patients showed increased serum levels despite of

RESEACH DESIGN OF THE STUDY

PSYCHOLOGICAL AND BIOCHEMICAL CHANGES ASSOCIATED WITH OSTEOGENIC SARCOMA AND EWING'S SARCOMA IN CHILDREN

Type of the study: this is a descriptive study on the psychological impact of the diagnosis and management of the bone cancer on the patients. Evaluation of serum enzymes and isoenzymes activities in patients with osteosarcoma and Ewing's sarcoma in National Cancer Institute and National Research center.

Back ground: A diagnosis of cancer creates crisis that requires the patients and their families to adapt quickly to the news of catastrophe. So, for the child with cancer, successful outcome in all psychological arenas hinges on early assessment, prevention, and intervention. Now, the field of biological markers of malignant bone tumors has been the focus of increased interest for their applicability in the diagnosis, response to therapy, and prognosis.

Aim of the study:

1- Identification the behavioral and psychiatric disturbances in the children with osteogenic sarcoma and Ewing's sarcoma.
2- Evaluation of serum total alkaline phosphatase and its isoenzymatic activities in patients with osteosarcoma, and serum total lactate dehydrogenase and its isoenzymatic activities in patients with Ewing's sarcoma, throughout the stages of therapy and follow up after 6 months of cure. And the possibilities of their usage as prognostic tools for the detection the response to therapy and prediction of early relapse of the tumors before other clear cut physical sign to be manifested.

Criteria for inclusion:

1- Histopathologic confirmation of osteogenic sarcoma or Ewing's sarcoma
2- No evidence of metastases.
3- No prior history of cancer
4- No previous therapy.

Criteria for exclusion:

1- Undiagnosed bone tumors.
2- Metastatic case.
3- Past history of previous cancer.
4- History of previous therapy.

Limitation of the study:

1- Rarity of the disease.
2- Time consuming.
3- Lack of cooperation from some patients and their families.

Time table:

- Registration: April, 1994
- Sample: May, 1994
- Questionnaire: May, 1994
- Literature: Sep., 1995
- Analysis: Jul., 1996

Questionnaire

*Personal history.
*Family History.
*Past history.
*Psychological history.
*Eysenk personality questionnaire.
*Children anxiety scale.

Evaluation of the serum

*Total γ glutamyl transferase.
*Total lactate dehydrogenase and its isoenzymatic activities.
*Total alkaline phosphatase and its isoenzymatic activities.

Complete clinical examination.
laboratory and radiographic diagnosis.
bone biopsy and pathological analysis.

Psychological sheet

Name: Age: Job: General Appearance:

System Review:

Digestive System:

Overfeeding

Food refusal

Faddiness

Pica

Nausea

Vomiting

Diarrhea

Abdominal pain

Constipation

Fecal soiling

Urinary System:

Bed Wetting

Painful Micturition

Overflow

Frequent

Sleep:

Difficulty to sleep

Night mares

Night Terrors

Cirulatory & Respiratory Systems:

breathlessness

Cough

Palpitation

Motor System:

Restlessness

Overactivity

Underactivity

Clumsiness

Abnormal gait

Motor weakness

Right or left handed

Habitual manipulation of the body:

Nailbiting

Thumbsucking

Nosepicking

Headbanging

Speech:

Over-talkativeness

Mutism

Faulty speech

Thought Processing:

Poor concentration

Distractibility

Disorder thought

Day dreaming

Vision & Hearing:

any defect

Hallucination

Attack disorders:

Epilepsy

Fainting

Alternation of consciousness

Personality Traits & behaviour:

Happy or unhappy

Aggressive

relation with siblings & freinds

Fearfullness

Tearfullness

Negativism

Lying

Wandering from home

Staying out late

Submissive

Calm

Excitable

Anxiety

Disobedience

Stealing

Smoking

Drinking

Drug taking

Sexual Difficulties:

Onset of menstruation

Dysmenorrhea

Masturbation

Sex Education

Mood & emotional states:

Happy

Unhappy

Elated

Frankly depressed

Anxious

Hostile

Resentful

Suspicious

Has he ever wished he was dead

Suicidal attempts

Does he cry? Why?

School problems:

Attitude to school

Behaviour at school

Progress in school

Osteogenic sarcoma

Initial Sheet

-Name : ..... Male ☐ Female ☐

- Hospital no : ..... Code no : .....

- Birth date ☐☐/☐☐/☐☐

- Adress : .....

-Date of first symptom : ☐☐/☐☐/☐☐

- Measurments : Wt. : ..... Ht. : .....

- Symptomatology :

	yes	no
1 - Pain	☐	☐
site :		
2 - Mass	☐	☐
site :	Size	cm
3 - Pathologic Fracture	☐	☐
4 - Chest symptoms	☐	☐
describe :		
5- Others :	☐	☐

.....

- Pathology :

* *Osteoblastic* ☐
* *Chondroblastic* ☐
* *Fibroblastic* ☐
* *Telengictaic* ☐
* *others :* .....

	I	II	III
* Grade	☐	☐	☐

Investigations

(Initial)

- Name : .....

- Hospital no: ..... Code No. : .....

1 - CBC : TLC : ... HCT : PLT :

2 - LFT : Bilirubin : ..... SGOT : ..... SGPT :

AP : GT : .....

3 - KFT : BUN : ..... s. creatinine : .....

C. Clearance :

4 - Local X - ray : .....

5 - CXR : +ve ☐ - ve ☐ not performed ☐

6 - C.T.Chest : ☐ ☐ ☐

if :

7 - Bane Scan : ☐ ☐ ☐

8 - Two-dimentional echocardiography :

9 - Audiometry : .....

10 - Biopsy : ☐ ☐

Date : |__|__|__|__|__|__|

11 - Surgery : ☐ ☐

Date |__|__|__|__|__|__|

Type :

Lag period between 1 st symptom and biopsy :

Lag period between biopsy and definit surgery : .....

Lag period between surgery and chemotherapy : .....

- First day chemotherapy : |__|__|__|__|__|__|

Follow up Sheet No. : .....

- Name : .....
- Hospital nº : ..... - Code No : .....
- Date : |_|_|_|_|_|_|
- Last document :

Investigations :

* CBC : TLC : Hct : Plt :
* LFT : Bilirubin : SGOT : SGPT :
AP : G.T. : .....
* KFT : BUN : S.creatinine : .....
Creatinine Clearance : .....
* CXR + ve ☐ - ve ☐ not performed ☐

* C.T. Chest : +ve ☐ - ve ☐ not performed ☐

*Bone Scan :

* Echo-cardiography :

* Audiometry :

	yes	no	
- Remession :	☐	☐	
- Relapse :	☐	☐	Date : |_|_|_|_|_|_|
- Alive :	☐	☐	

- Date of death : |_|_|_|_|_|_|

Symptoms & Signs

		رقم المستشفى:	_____	:سم

Wt.: _____ Ht. _____ S.A. _____

- **Date of 1st Symptom:** _____
- **Date of 1st Presentation:** _____

	No	Yes
- **Pain:**	☐	☐
- **Mass:**	☐	☐
• Site	_____	
• Max. Diameter	_____	
• Diameter of other Limb	_____	

- Local Sings:	No	Yes
• Redness	☐	☐
• Hotness	☐	☐
• Tenderness	☐	☐
• Bleeding	☐	☐
• Fungation	☐	☐

	No	Yes
- **Pathologic Fracture:**	☐	☐
• Site	_____	

	No	Yes
- **Chest Symptoms:**	☐	☐
• Describe	_____	

- Systemic Symptoms:	No	Yes
• Fever	☐	☐
• Wt. loss	☐	☐
• Fatigue	☐	☐
• Anorexia	☐	☐

	No	Yes
- **Others:**	☐	☐
• Describe	_____	

Laboratory Investigations

(Initial & at weeks 9-18-30 & 54)

Week ☐

1- CBC:

- TLC _____
- HCT _____
- Plt. _____
- ANC. _____

2- Liver Function Tests:

- Bilirubin _____
- SGOT _____
- SGPT _____
- Alk. ph. _____

3- Renal Function Tests:

- Uric acid _____
- S. creatinine _____
- C. clearance _____

4- LDH: _____

5- ESR: 1st hr. _____ 2nd hr. _____

6- B.M. Aspirate: +ve ☐ -ve ☐

_____

7- B.M. Biopsy: +ve ☐ -ve ☐

_____

_____

8- Urine Analysis:

_____

Radiologic Evaluation

(Initial & at weeks 9-18-30 & 54)

Week ☐

	+ve	-ve	Not Performed
1- Chest X-ray	☐	☐	☐
2- Local X-ray	☐	☐	☐
3- Abdominal U/S	☐	☐	☐
4- C.T Scan Chest	☐	☐	☐
5- C.T. Scan Local	☐	☐	☐
6- Skeletal Survey	☐	☐	☐
7- Bone Scan	☐	☐	☐
8- MRI Local	☐	☐	☐
9- Echo Cardiography	☐	☐	☐
10- ECG	☐	☐	☐

Biopsy

I- Initial Diagnostic Biopsy:

- Pathology No. ☐☐
- Date ☐☐☐
- Site _____
- Lag period between 1st symptom and biopsy _____ days
- Pathologic Findings:
  - *Ewing's sarcoma* ☐
  - *Peripheral primitive neuro-ectodermal tumour (PPNET)* ☐
  - *Extra-osseous Ewing's sarcoma* ☐
- Results of cytogenetics: _____

  _____
- E/M findings: _____

  _____

II- Post-Induction Biopsy:

- Pathology No. ☐☐
- Date ☐☐☐
- Nature of biopsy
  - *Open biopsy* ☐
  - *Needle biopsy* ☐
- **Response to chemotherapy:**
  - ***Good response (≤ 10% viable tumour)*** ☐
  - ***Poor response (≥ 10% viable tumour)*** ☐

Follow-up Sheet No. ☐ Date ☐☐☐

☐☐ رقم المستشفى: _____ الأسم:

1- Complete blood count:

TLC _____ HCT _____ Plt _____

2- Liver function tests:

Bilirubin _____ SGOT _____ SGPT _____

Alk. phosph _____

3- Renal function tests: _____

Uric acid _____ S. creatinine _____ Urea _____

4- ESR: _____

1st Hr. _____ 2nd hr. _____

5- LDH: _____

	+ve	-ve	Not Performed
6- CXR	☐	☐	☐
7- Local X-ray	☐	☐	☐
8- Bone Scan	☐	☐	☐
9- Echo-card.	☐	☐	☐

Please give details of pathologic data if present

_____

10- Others _____

11- Clinical complications: _____

_____

_____

Status:	**Yes**	**No**	
• Remission	☐	☐	
• Relapse	☐	☐	Date ☐☐☐
• Alive	☐	☐	Date ☐☐☐
• Loss of F.U.	☐	☐	Date ☐☐☐

treatment before the clinical diagnosis of metastasis, and proved to be metastatic later by another investigations. So that, the increase in the LDH anaerobic isoenzyme activities could suggest any changes in the tumor activity before other clear cut physical sign are manifested.

The total serum ALP levels in patients with osteogenic sarcoma in children and adolescents, were very highly significant increased as compared to the serum control levels. These increments were mainly due to the increased activity of the bone fraction of ALP as proved by electrophoretic separation, heat inactivation, normal serum level of γ glutamyl transferase, and normal liver fraction of ALP levels. This could be explained on the basis of increased osteoblastic activity of osteosarcoma tumor cells.

During the course of therapy including the surgery, the elevated serum ALP levels decreased gradually in response to therapy in 24 patients (68.5%) who continue the treatment and not developed metastasis (16 children 64% and 8 adolescents 80%), the serum ALP level increased in 6 patients (5 children and 1 adolescent) despite of therapy and before the clinical diagnosis of metastasis and proved to be metastatic later by another investigations. So that, the increase in ALP isoenzyme activities could suggest any changes in the tumor activity before other clear-cut physical sign are manifested.

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ARABIC SUMMARY

الملخص العربى

تتضمن الرسالة إجراء دراسات على نشاط إنزيم الفوسفاتيز القلوى الكلى وشبيهاته فى مصل دم المرضى المصابين بسرطان العظام **osteogenic sarcoma** وإنزيم اللاكتيك ديهيدرجيناز وشبيهاته فى مصل دم المرضى المصابين بسرطان العظام **Ewing's sarcoma** فى مراحل علاج المرض المختلفة (قبل بداية العلاج - أثناء العلاج- فى نهاية العلاج - أثناء متابعة المريض بعد شفائه بستة أشهر) وذلك بهدف إعطاء فكرة عن تطور المرض وإستجابته للعلاج وكذلك إيجاد طرق سهلة وقليلة النفقات للإكتشاف المبكر لإنتكاس المرض أثناء متابعة المريض. حيث أن تقدير نشاط شبيهات الإنزيم أثبتت أن لديها خصوصية وحساسية لتأثر العظام مقارنة بـا لنشاط الكلى للإنزيم.

كما تضمنت الرسالة دراسة تأثير مرض السرطان وعلاجه على الحاله النفسية والسلوك الاجتماعى للمريض والاستجابة النفسية الطبيعية والغير طبيعية لمرضى السرطان كمؤشر للطبيب للتشخيص والعلاج والتحويل.

أجريت الدراسة على ٨٦ حالة تم تقسيمهم إلى المجموعات التالية:-

١- **مجموعة الاصحاء:**- وتشمل ٣٠ حالة تم تقسيمهم حسب العمر إلى

(أ) - ١٥ طفل يتراوح أعمارهم من ٨-١٢ سنة

(ب)- ١٥ شخص فى سن البلوغ يتراوح أعمارهم من ١٣-٢٠ سنة

٢- **مجموعة osteogenic sarcoma** :-

وتشمل ٣٥ حالة تم تقسيمهم حسب العمر إلى:-

(أ)- ٢٥ طفل مريض يتراوح أعمارهم من ٨-١٢ سنة

(ب)- ١٠ مرضى فى سن البلوغ يتراوح أعمارهم من ١٣-٢٠ سنة

٣- مجموعة Ewing's Sarcoma :-

وتشمل ٢١ مريض يتراوح أعمارهم من ٨-١٦ سنة

تم تشخيص المرضى بالطرق الاكلينيكية والتحاليل المعملية والفحوصات بالاشعة بجانب أخذ عينة من العظام للفحص الباثولوجى.

تم تقدير النشاط الكلى لكل من إنزيم الفوسفاتيز القلوى واللاكتيك دهيدروجينياز و الجاما جلوتاميل ترانسفيراز (ALP, LDH & GGT) بالطريقة الاسكندينافيه (Scandinavian Method). وتم تقدير نشاط شبيهات إنزيم الفوسفاتيز القلوى بثلاثة طرق وهم الفصل الكهربى على مادة PAGE و التثبيط الكيميائى بمادة Phenyl alanine و التثبيط الحرارى. كما تم تقدير نشاط شبيهات إنزيم الاكتيك دهيدروجيناز (LD isoenzymes) بطريقة الفصل الكهربى على مادة Agarose gel

وتم دراسة الإضطرابات النفسية المصاحبة لمرضى السرطان عن طريق التقييم النفسى والسلوك الإجتماعى ومعرفة معدل القلق النفسى عندهم بدراسة إستبيان مقياس القلق فى الأطفال . كما تم تعيين سمات شخصيات المرضى عن طريق إستبيان إيزنج.

وقد أوضحت الدراسة مايلى:-

وجود زيادة معنوية فى معدل القلق النفسى فى المرضى المصابين بسرطان العظام Osteogenic Sarcoma & Ewing's Sarcoma أثناء تشخيص المرض وعند نهاية العلاج. وتكون أكثر وضوحا عند دخولهم المستشفى للعلاج . ويرجع السبب فى هذه الزيادة هو خوف المرضى من إبعادهم عن أهلهم . وكذلك أوضحت الدراسة

بأنه لايوجد فرق إحصائى فى معدل القلق النفسى بين الأولاد والبنات المرضى وذلك بالمقارنة بالعينة الضابطة.

أشارت النتائج إلى زيادة السمات العصبيه بين المرضى وعلى الآخص العصبيه وإضطرابات الأكل والتبول اللاإرادى والآحلام المزعجة والكوابيس وكذلك مص الاصبع مقارنة بالعينة الضابطة.

وبدواسة سمات الشخصية للمرضى فقد أشارت النتائج إلى زيادة معنوية فى عدد الحالات الذين لديهم إستعداد للعصاب فى كل من الاولاد والبنات فى المرضى المصابين بسرطان العظام **Ewing's Sarcoma &Osteogenic Sarcoma** وذلك مقارنة بمعدل العينة الضابطة ويرجع هذا إلى أن المرضى لديهم معدل عالى من العاطفه.

وقد وجد زيادة معنويه فى عدد الحالات الذين عندهم السمه الإنطوانية فى المرضى المصابين بسرطان العظام **Ewing's Sarcoma** ، وكانت هذه الزيادة عاليه فى الاولاد عنهم فى البنات .أما بالنسبة للمرضى المصابين بسرطان العظام **Osteogenic Sarcoma** فكانت نسبة السمه الانطوائية مرتفعة فى كل من الاولاد والبنات مقارنة بالعينة الضابطة ويرجع ذلك إلى التأثير النفسى للاطفال للاعاقة الفسيولوجية التى تصاحب المرض (بتر الساق).

وقد وجد زيادة معنوية فى عدد المرضى الذين عندهم إستعداد عالى للكذب ، وكانت هذه الزياده مرتفعة فى البنات عنهم فى الاولاد فى المرضى المصابين بسرطان العظام **Osteogenic Sarcoma & Ewing's Sarcoma** وذلك مقارنة بالعينة الضابطة ويرجع ذلك إلى تجاهل المريض للاعاقة لكى يساير الحياه الطبيعيه.

كما أشارت النتائج إلى عدم وجود زياده معنويه فى عدد المرضى ذات الاستعداد للمرض العقلى (الذهان) فى المرضى المصابين بسرطان العظام **Osteogenic Sarcoma & Ewing's Sarcoma** فى كل من الاولاد والبنات مقارنة بالعينة الضابطة.

أما بالنسبة للاضطرابات النفسية المصاحبة لمرضى السرطان فقد أشارت النتائج إلى وجود أكثر من تشخيص للمريض الواحد وكانت أكثر الاضطرابات شيوعا هما الاضطرابات السلوكيه والاضطرابات العاطفية.

أشارت النتائج إلى وجود زيادة معنوية فى نشاط إنزيم اللاكتيك دهيدروجيناز **LDH** الكلى فى مصل دم المرضى المصابين بسرطان العظام **Ewing's Sarcoma** قبل بداية العلاج وهذه الزياده كانت ممثلة فى زيادة نشاط شبيهات الانزيم اللاهوائية LD_2 , LD_3 and LD_4 مقارنة بمجموعة الاصحاء ويرجع ذلك إلى إفراز الانزيم من الخلايا السرطانية فى الدوره الدمويه وذلك لتحول الخلايا السرطانية السريعة الانقسام إلى التمثيل الاهوائى الذى تعتبر قياس نشاط شبيهات الانزيم الكاثودية مؤشرا لها .

وقد وجد أثناء العلاج إنخفاض ملحوظ فى نشاط إنزيم اللاكتيك دهيدروجيناز وشبيهاته كاستجابة إيجابيه للعلاج الكيماوى ولكن أثناء العلاج الاشعاعى فقد لوحظ زيادة معنوية فى نشاط إنزيم اللاكتيك دهيدروجيناز وشبيهاته LD_2, LD_3, LD_4 and LD_5 ويرجع ذلك إلى التأثير التهتكى للعلاج الاشعاعى على العضلات السليمه المحيطه بالورم السرطانى والغنية بشبيه الانزيم LD_5.

كما لوحظ إستمرار إنخفاض معدل نشاط إنزيم اللاكتيك دهيدروجيناز LDH الكلى وشبيهاته اللاهوائيه فى مصل دم المرضى إلى المعدل الطبيعى للاصحاء فى ١٢ مريض (٥٧٪) الذين استكملوا العلاج وتماثلوا للشفاء ولكن لوحظ زياده معنويه فى نشاط إنزيم اللاكتيك دهيدروجينياز وشبيهاته فى أربعة من المرضى (١٩٪) مع إستمرار العلاج و لم تظهر عليهم دلالات إنتكاس المرض ولكنها ظهرت عليهم فى وقت لاحق بفحوص أخرى .

وفى المجمل يمكن إستخدام معدل تغيير شبيهات إنزيم اللاكتيك دهيدروجيناز اللاهوائية الكاثودية كمؤشر لاى تغييرات تحدث فى نشاط الورم السرطانى قبل ظهور الدلالات المرضية على المريض.

أشارت النتائج إلى وجود زيادة معنوية فى نشاط انزيم الفوسفاتيز القلوى الكلى فى مصل دم المرضى المصابين بسرطان العظام **Osteogenic Sarcoma** قبل بداية العلاج . وهذه الزيادة كانت ممثلة فى زيادة نشاط شبيه الانزيم الناتج من العظام مقارنة بمجموعة الاصحاء ويرجع السبب فى هذه الزياده إلى زيادة النشاط التيبسى لخلايا **Osteogenic Sarcoma** الخبيثه مدعما لذلك عدم وجود أى زيادة معنوية فى نشاط كل من شبيه انزيم الفوسفاتيز القلوى الناتج من الكبد وإنزيم الجاما جلوتاميل ترانسفيراز **(GGT)** فى مصل دم المرضى عنه فى الاصحاء .

كما لوحظ أثناء العلاج إستمرار إنخفاض معدل نشاط انزيم الفوسفاتيز القلوى الكلى وشبيهه الناتج من العظام كاستجابة إيجابيه للعلاج الكيماوى والعلاج الجراحى إلى المعدل الطبيعى للاصحاء فى ٢٤ مريض (٦٨,٥٪) (١٦ طفل مريض - ٨ مرضى فى سن البلوغ) الذين إستكملوا العلاج وتماثلوا للشفاء ولكن لوحظ زيادة

معنوية فى نشاط انزيم الفوسفاتيز القلوى الكلى وشبيهه الناتج من العظام فى ستة من المرضى (١٧٪) (٥ أطفال مرضى - ١ مريض فى سن البلوغ) مع إستمرار العلاج ولم تظهر عليهم دلالات إنتكاس المرض ولكنها ظهرت عليهم فى وقت لاحق بفحوص أخرى .

وفى المجمل يمكن إستخدام معدل تغير إنزيم الفوسفاتيز القلوى الكلى وشبيهه الناتج من العظام كمؤشر لاى تغييرات تحدث فى نشاط الورم السرطانى قبل ظهور الدلالات المرضية على المريض.

المستخلص العربى

تتضمن الرسالة إجراء دراسات على نشاط إنزيم الفوسفاتيز القلوى الكلى وشبيهاته فى مصل دم المرضى المصابين بسرطان العظام **osteogenic sarcoma** وإنزيم اللاكتيك ديهيدرجيناز وشبيهاته فى مصل دم المرضى المصابين بسرطان العظام **Ewing's sarcoma** فى مراحل علاج المرض المختلفة.

كما تضمنت الرسالة دراسة تأثير مرض السرطان وعلاجه على الحاله النفسية والسلوك الاجتماعى للمريض والاستجابة النفسية الطبيعية والغير طبيعية لمرضى السرطان.

أجريت الدراسة على ٨٦ حالة تم تقسيمهم إلى ٣٠ حالة أصحاء كمجموعة مقارنة (١٥ طفل يتراوح أعمارهم من ٨-١٢ سنة- ١٥ شخص فى سن البلوغ يتراوح أعمارهم من ١٣-٢٠ سنة)و٣٥ مريض بسرطان العظام **osteogenic sarcoma** (٢٥طفل مريض يتراوح أعمارهم من ٨-١٢ سنةو ١٠ مرضى فى سن البلوغ يتراوح أعمارهم من ١٣-٢٠ سنة). ٢١ مريض **Ewing's Sarcoma** يتراوح أعمارهم من ٨-١٦ سنة.

وقد أوضحت الدراسة وجود زيادة معنوية فى معدل القلق النفسى فى المرضى المصابين بسرطان العظام **Osteogenic Sarcoma & Ewing's Sarcoma** أثناء تشخيص المرض وعند نهاية العلاج.وكذلك زيادة السمات العصبيه بين المرضى وعلى الآخص العصبيه وإضطرابات الأكل والتبول اللاإرادى والآحلام المزعجة والكوابيس وكذلك مص الاصبع مقارنة بالعينة الضابطة.وبدواسة سمات الشخصية للمرضى فقد أشارت النتائج إلى زيادة معنوية فى عدد الحالات الذين لديهم إستعداد للعصاب وكذلك السمه الإنطوانية والإستعداد العالى للكذب .كما أشارت النتائج إلى عدم وحود زياده معنويه فى عدد المرضى ذات الاستعداد للمرض العقلى (الذهان) مقارنة بالعينة الضابطة. أما بالنسبة للاضطرابات النفسية المصاحبة لمرضى السرطان فقد أشارت النتائج إلى وحود أكثر من تشخيص للمريض الواحد وكانت أكثر الاضطرابات شيوعا هما الاضطرابات السلوكيه والاضطرابات العاطفية.

أشارت النتائج إلى وجود زيادة معنوية فى نشاط إنزيم اللاكتيك دهيدروجيناز **LDH** الكلى فى مصل دم المرضى المصابين بسرطان العظام **Ewing's Sarcoma** قبل بداية العلاج وكانت هذه الزياده ممثلة فى زيادة نشاط شبيهات الانزيم اللاهوائية **LD_2, LD_3 and LD_4** مقارنة بمجموعة الاصحاء.وقد وجد أثناء العلاج إنخفاض ملحوظ فى نشاط إنزيم اللاكتيك دهيدروجيناز وشبيهاته كاستجابة إيجابيه للعلاج الكيماوى ولكن أثناء العلاج الاشعاعى فقد لوحظ

فقد لوحظ زيادة معنوية فى نشاط إنزيم اللاكتيك دهيدروجيناز وشبيهاته LD_2, LD_3, LD_4 and LD_5. كما لوحظ إستمرار إنخفاض معدل نشاط إنزيم اللاكتيك دهيدروجيناز LDH الكلى وشبيهاته اللاهوائيه إلى المعدل الطبيعى للاصحاء فى ١٢ مريض (٥٧٪) الذين استكملوا العلاج وتماثلوا للشفاء ولكن لوحظ زياده معنويه فى نشاط إنزيم اللاكتيك دهيدروجينياز وشبيهاته فى أربعة من المرضى (١٩٪) مع إستمرار العلاج و لم تظهر عليهم دلالات إنتكاس المرض ولكنها ظهرت عليهم فى وقت لاحق بفحوص أخرى .

أشارت النتائج إلى وجود زيادة معنوية فى نشاط انزيم الفوسفاتيز القلوى الكلى فى مصل دم المرضى المصابين بسرطان العظام **Osteogenic Sarcoma** قبل بداية العلاج . وكانت هذه الزيادة ممثلة فى زيادة نشاط شبيه الانزيم الناتج من العظام مقارنة بمجموعة الاصحاء كما لوحظ عدم وجود أى زيادة معنوية فى نشاط كل من شبيه انزيم الفوسفاتيز القلوى الناتج من الكبد وإنزيم الجاما جلوتاميل ترانسفيراز **(GGT)** فى مصل دم المرضى عنه فى الاصحاء .

كما لوحظ أثناء العلاج إستمرار إنخفاض معدل نشاط انزيم الفوسفاتيز القلوى الكلى وشبيهه الناتج من العظام كاستجابة إيجابيه للعلاج إلى المعدل الطبيعى للاصحاء فى ٢٤ مريض (٦٨,٥٪) الذين إستكملوا العلاج وتماثلوا للشفاء ولكن لوحظ زيادة معنوية فى نشاط انزيم الفوسفاتيز القلوى الكلى وشبيهه الناتج من العظام فى ستة من المرضى (١٧٪) مع إستمرار العلاج و لم تظهر عليهم دلالات إنتكاس المرض ولكنها ظهرت عليهم فى وقت لاحق بفحوص أخرى .

وفى المجمل يمكن إستخدام معدل تغير إنزيمين الفوسفاتيز القلوى واللاكتيك دهيدروجينازوشبيهاتهما كمؤشر لاى تغيرات تحدث فى نشاط الورم السرطانى قبل ظهور الدلالات المرضية على المريض.

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الكليـــة:

رسالـــة ماجستير / دكتــوراه ✓

اسم الطالب: من عفيفي محمد خليل
عنوان الرسالة: التغيرات الكيميائية الحيوية المصاحبة لسرطان العظام في الأطفال الأستيوجينيك والإيونج ساركوما

اسم الدرجة: (ماجستير / دكتوراه)

لجنـة الإشـراف
١- الاسم/ أ.د. زينب بشري — الوظيفة/ أستاذ الأمراض الصدرية — جامعة عين شمس
١- الاسم/ أ.د. صفي شاكر محمد — الوظيفة/ أستاذ الكيمياء الحيوية — المركز القومي للبحوث
١- الاسم/ أ.د. صلاح الدين مصطفى — الوظيفة/ أستاذ ورئيس قسم الدراسات الطبية — معهد الدراسات العليا للطفولة
١- الاسم/ أ.د. أحمد السيد خلف الله — الوظيفة/ أستاذ الكيمياء الباثولوجية — جامعة القاهرة

تاريخ البحث: ٢١ / ٢ / ١٩٩٤

الدراسات العليا
ختم الإجازة:
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موافقة مجلس الكلية
١٨ / ٢ / ١٩٩٧

موافقة مجلس الجامعة
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القسم التابع لـــــــه : ..... الدراسات العليا .....
أســـم الكليـــــــة : ..... معهد الدراسات العليا للطفولة .....
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سنـــة المنـــــح : ..... مايو ١٩٩٧ .....

شـــــروط عامـــــه

يوضع شعـــار الجامعـــة علـــى الغـــلاف الخارجـــى.

"بسم الله الرحمن الرحيم"

قالوا سبحانك لا علم لنا الا ما علمتنا إنك أنت العليم الحكيم

﴿صدق الله العظيم﴾

سورة البقرة آية ﴿٣٢﴾

التغيرات النفسيه والبيوكيميائيه المصاحبه لسرطان العظام في الاطفال (الاستيوجينك والايونج ساركوما)

رسالة مقدمه

للحصول على درجة دكتوراة الفلسفه

فى دراسات الطفولة

مقدمة من

الطبيبة / مى عفيفى محمد خليل

بكالوريوس الطب والجراحه - جامعة القاهرة

ماجستير الاطفال - جامعة القاهره

باحث مساعد - المركز القومى للبحوث

تحت إشراف

أ.د. زينب بشرى عبد الحميد

استاذ الامراض النفسيه والعصبيه

كلية الطب

جامعة عين شمس

أ.د. يحيى شاكر محمد حامد شاكر

أستاذ الكيمياء الحيويه الاكلينيكيه

رئيس قسم الكيمياء الحيويه

المركز القومى للبحوث

د. محمد هانى محمد حسين

أستاذ مساعد فى طب أورام الاطفال

معهد الاورام القومى

جامعة القاهرة

د. خالد حسين طمان

مدرس فى طب الاطفال

معهد الدراسات العليا للطفوله

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معهد الدراسات العليا للطفوله

القسم الطبى

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١٩٩٦