

Role of Tc-99m MIBI and Tc-99m DMSA-V in Evaluation of Bone Tumors

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ABSTRACT

Background: Bone sarcomas are usually aggressive tumors treated by multimodality approaches. Early detection of residual or recurrent tumor tissue after treatment is important in the management of these patients. This is currently achieved by clinical examination, diagnostic radiological procedures, radionuclide imaging techniques and detailed histological examination. Tumor response to preoperative chemotherapy in osteosarcoma as defined by the degree of tumor necrosis, has been found to be one of the best prognostic indicators of disease control. However, this information can be only obtained by detailed histologic examination after tumor resection. Recently, there have been attempts to predict the tumor response to chemotherapy before surgical excision by radionuclide imaging techniques. This enables the clinician to consider alternative chemotherapeutic regimens or surgery at an earlier stage.

Purpose: Evaluate and compare Tc-99m pentavalent DMSA to Tc-99m sestamibi and Tc-99m MDP blood pool image in patients with malignant bone tumors for the purpose of:

- 1- Detection of the primary lesion.
- 2- Evaluating treatment response of osteosarcoma after 4 cycles of preoperative chemotherapy.
- 3- Prediction of response of osteosarcoma after the first cycle only of preoperative chemotherapy.
- 4- Detection of residual or recurrent viable tumor after treatment.

Materials and Methods: Ninety-three patients with different types of bone sarcomas were studied. They were divided into 3 groups:

Group I: Newly diagnosed patients with different bone sarcomas (40cases)

Group II: Fourteen patients from group I with definite pathological diagnosis of osteosarcoma were studied to evaluate the response of the primary lesion to preoperative chemotherapy (according to the treatment protocol adopted in NCI).

Group III: Patients with different types of bone sarcomas at different stages of treatment and under follow up (53 patients). All patients were imaged following IV injection of 20 mCi (740 MBq) of ^{99m}Tc-MDP, ^{99m}Tc-MIBI and ^{99m}Tc (V) DMSA with time interval of 48 h between each scan.

Results: The sensitivity of ^{99m}Tc(V)DMSA for detection of primary lesion was nearly 100% versus 92% for BP and 85% for ^{99m}Tc-MIBI scanning in group I. Regarding the assessment of response to preoperative chemotherapy, the patients were classified into 3 sub groups according to percentage of tumor necrosis obtained from histopathologic examination when correlated with scintigraphic findings into partially responding disease, stable disease and progressive disease. Quantitative analysis revealed cut off values of tumor to background ratios (to differentiate between viable and non viable tumor tissue) for ^{99m}Tc(V)DMSA, ^{99m}Tc-MIBI and BP of 1.7, 1.5 and 1.5 respectively. Nineteen patients in group 3 had equivocal findings for local tumor recurrence on CT and/or MRI and the sensitivity for detection of tumor recurrence by ^{99m}Tc-DMSA, BP and ^{99m}Tc-MIBI was 94%, 73% and 68% respectively.

Conclusions: Tc-99m (V) DMSA has high avidity for primary malignant bone tumors with no false negative results. The evaluation of response to preoperative chemotherapy in osteosarcoma after the first cycle may predict the final response where partial responders with significant declining ratios should continue the same treatment while those with progressive disease and rising ratios should change their chemotherapy regimen or do surgery at an earlier stage saving their time and money. This issue needs confirmation with large number of patients and multi-center trials. The higher sensitivity of Tc-DMSA compared to Tc-MIBI in the detection of primary lesions as well as recurrent tumors suggests that Tc-99m pentavalent DMSA is not affected by multi-drug resistance. Further studies using both tracers and confirmed with immunohistochemical staining of P-glycoprotein from tumor samples are recommended to confirm this issue.

Key Words: Bone tumors - Osteosarcoma - Chemotherapy - Tc-99m MIBI - Tc-99m DMSA.

INTRODUCTION

Bone sarcomas are usually aggressive tumors treated by multimodality approaches. Early detection of residual or recurrent tumor tissue after treatment is important in the management of these patients. This is currently achieved by clinical examination, conventional diagnostic morphologic procedures, radionuclide functional imaging techniques and if possible by histological examination [1].

Evaluation of treatment response in bone sarcomas is of utmost importance in planning further management. Anatomical imaging modalities as C.T. and conventional MRI define anatomical details and are helpful in identifying the extent of the lesions. Following surgery, chemotherapy and/or radiotherapy the accuracy of anatomical images, in differentiating post therapy changes from residual or recurrent tumor, is limited [2]. Functional images or metabolic images using single photon radioisotopes or PET tracers are more accurate in solving this problem. [3,4] ^{99m}Tc -MIBI is now a well established tumor imaging agent and is effectively used for detection of early bone & soft tissue tumor recurrence and to differentiate residual/ recurrent, viable tumor from post therapy changes [2].

The tumor response to preoperative chemotherapy in osteosarcoma which is defined by the degree of tumor necrosis, has been found to be one of the best prognostic indicators of disease control. However, this information can be only obtained by detailed histological examination after tumor resection. There have been attempts to predict the tumor response to either ongoing chemotherapy or before surgical excision by radionuclide imaging techniques. This enables the clinician to consider alternative chemotherapeutic regimens or surgery at an earlier stage [5].

^{99m}Tc pentavalent DMSA is a nonspecific tumor imaging agent that was initially used in imaging of medullary cancer thyroid. Recently it has been tried in the imaging of small cell and non small cell lung cancer [6], imaging patients with bone metastases [7] hepatocellular carcinoma [8] and bone and soft tissue tumors [9].

Aim of the work:

We aimed to evaluate Tc-99m DMSA, compared to Tc-99m sestamibi and skeletal blood

pool images, in patients with primary malignant bone tumors for the purpose of:

- 1- Detection of the primary lesion.
- 2- Evaluation of treatment response of primary osteosarcoma to preoperative chemotherapy (according to the adopted protocol of the NCI).
- 3- Prediction of response of primary osteosarcoma to ongoing preoperative chemotherapy after the first cycle only.
- 4- Detection of residual or recurrent tumor after treatment for different malignant bone tumors.

PATIENTS AND METHODS

Ninety-three patients with different bone sarcomas were included in the current study. Their age ranged from 6-34 years (15.7 ± 5.3) including 60 males and 33 females, (Table 1).

They were divided into three groups:

• *The first group:*

Forty patients with newly diagnosed untreated, peripherally located bone sarcomas were studied to test the reliability of ^{99m}Tc (V)DMSA in the detection of the primary lesion compared to ^{99m}Tc -MIBI scan and conventional dual phase bone scan. The early blood pool (BP) image was only included in the quantitative assessment. All patients did not have metastatic disease from the start. The size of the lesions ranged from 3 to 15 cm as was estimated by regional MRI.

• *The second group:*

Fourteen patients (from group I) with definite pathological confirmation of being osteosarcoma. All patients were non metastatic with lesion size ranging from 5 to 15 cm. They were studied to evaluate the response of osteosarcoma to preoperative chemotherapy (according to the treatment protocol adopted at the NCI), using ^{99m}Tc (V) DMSA and ^{99m}Tc -MIBI after the 1st cycle and 4 cycles of chemotherapy.

Patients in this group were operated by limb salvage as decided by their treating physicians according to the treatment protocol of the NCI. The degree of tumor necrosis (TN) was evaluated pathologically by one of our competent pathologists.

• *The third group:*

Fifty-three patients with different bone sar-

comas at different stages of treatment were evaluated scintigraphically to detect any residual or recurrent tumor tissue.

Technical procedures of Nuclear Medicine studies:

The three groups were evaluated by ^{99m}Tc (V)DMSA, dual phase bone scan and ^{99m}Tc -MIBI scanning using 20 mCi (for each) with time interval of 48 hours between each study.

^{99m}Tc (V)DMSA whole body blood pool (W.B.S) with additional localized spot views to the desired site were acquired after 3 hours post injection (PI) while in ^{99m}Tc -MIBI scan, Early (15 min PI) W.B.S and local spot views and late (2 hours) local spot view were acquired.

Lastly dual phase skeletal imaging with local BP image for the site of primary lesion was done.

Quantitative analysis: was done by calculation of tumor uptake ratio (L/Bg) on the local spot view image for ^{99m}Tc (V)DMSA, early and late ^{99m}Tc -MIBI & BP images.

Correlation with other imaging modalities like CT and/or MRI as well as clinical correlation were performed for the third group which included 34 disease free patients and 19 patients with suspected recurrence.

RESULTS

Cut off values (to differentiate between viable and non viable tumor tissue) for ^{99m}Tc (V)DMSA, ^{99m}Tc -MIBI and BP were calculated and were found to be 1.7, 1.5 and 1.5 respectively.

In the first group: ^{99m}Tc (V)DMSA had the highest sensitivity for lesion detection approaching 100% with no false negative results whereas lower sensitivity for ^{99m}Tc -MIBI and BP were found accounting for 85% and 92% respectively.

It was found that the range of tracer uptake ratio for ^{99m}Tc (V) DMSA was 1.9 to 9.5 with a mean value of (4.03 ± 1.74) which was significantly higher than that for ^{99m}Tc -MIBI that ranged from 0.6 to 4.6 with a mean value of (1.84 ± 0.81) with a significant statistical difference ($p < 0.0001$). The mean uptake ratio for blood pool imaging was 2.88 and ranged from 1.3 to 5., with p value < 0.001 (Table 2) with p value < 0.001 .

In the Second Group: This group of 16 patients were classified according to the percentage of tumor necrosis (TN) obtained from histopathological examination when correlated with scintigraphic findings into 3 groups:

TN 10-35% was considered as a mark for progressive disease.

TN 35-50% was considered as a mark for stable disease.

TN $> 50\%$ was considered as a mark for partially responding disease. Unfortunately, there were no patients showing good response ($>90\%$) in this group.

1- *Partially responding disease: (Tumor Necrosis $> 50\%$ but less than 80%)*

• ^{99m}Tc (V)DMSA scanning:

The mean value of uptake ratios in the base line study was 4.74, it decreased to 4.4 after the 1st cycle of chemotherapy. More decrease occurred after the last preoperative chemotherapy cycle to 4.05. In spite of being declining throughout the 3 studies, all values were higher than the cut off value for ^{99m}Tc (V)DMSA (> 1.7) denoting the presence of large amount of viable malignant tumor tissue in the partially responding tumor, (Fig. 3).

• ^{99m}Tc -MIBI scanning:

The mean value of uptake ratios in the baseline study was 2.16 and decreased to 2.1 after the 1st cycle and more decrease occurred after the last preoperative chemotherapy cycle to 1.55. The values were higher than the cut off value for ^{99m}Tc -MIBI (>1.5). The descent of mean values among the 3 studies was more evident in ^{99m}Tc -MIBI than in ^{99m}Tc (V)DMSA, (Table 3).

2- *Stable Disease: (Tumor Necrosis: 35-50%)*

• ^{99m}Tc (V)DMSA scanning:

Revealed minimal rise in mean values with time, starting by 4 and increased to 4.4 after the first cycle and then increased to 5.7 after the fourth chemotherapy cycle.

• ^{99m}Tc -MIBI scanning:

Also showed minor increase in the mean values for the 3 studies done and they were equal to 2.08 (baseline), 2.02 (after the first cycle) and 2.53 (after the fourth cycles) respectively, with no statistically significant difference between the 3 studies done, (Table 4).

3- Progressive Disease: (Tumor Necrosis: 10-35%)

- ^{99m}Tc(V)DMSA scanning:

The mean uptake value was 4.5 for the baseline study, increased to 5.8 after the 1st cycle and showed more rise to 6.03 after the fourth preoperative chemotherapy cycle (Fig. 4).

- ^{99m}Tc-MIBI scanning:

The mean uptake value was 1.28 for the baseline study, 1.1 after the 1st cycle and 1.38 after the fourth preoperative chemotherapy cycle. In spite there was progressive disease, the mean values were less than the cut off value for ^{99m}Tc-MIBI (1.5). This was likely attributed to the phenomenon of MDR, (Table 5).

Clinical outcome of patients who received neoadjuvant chemotherapy (16 patients).

Clinical outcome of patients with partially responding disease (4 patients) (TN > 50%)

Three patients (non metastatic) ended chemotherapy protocol (OSGIII) (Fig. 1), did limb salvage and were put under follow up. The fourth patient developed lung metastases after end of chemotherapy (OSGIV) (Fig. 2). Metastectomy was done and he is receiving another line of chemotherapy and is still living up till now.

Clinical outcome of patients with stable disease (5 patients) (TN = 35%-50%). Two patients received chemotherapy protocol for osteosarcoma (OSGIII). One of them did amputation due to progressive local disease, the other one did limb salvage. For both of them chemotherapy was intensified by more platinum containing compound, due to poor tumor necrosis. Both patients are under follow up. Another two patients received chemotherapy protocol (OSGIII). Post limb salvage, they developed lung metastases, where metastectomy was done, they received another line of chemotherapy and they are under follow up till now.

The last patient was diagnosed to have Retinoblastoma at first. Eye enucleation was done followed by chemotherapy and radiotherapy. The patient was put under follow up for two and half years, thereafter the patient developed osteosarcoma, received neoadjuvant chemotherapy followed by limb salvage. Unfortunately the

patient developed bone and lung metastases and died 6 months after diagnosis of osteosarcoma.

Clinical outcome of patients with progressive disease (7 patients) (TN = 10%-35%).

Three patients received four cycles of cisplatin/doxorubicin. Limb salvage was done to all of them, they had progressive disease (bone and lung metastases). They were given palliative chemotherapy.

Another three patients received 4 cycles of chemotherapy (2 cycles of cisplatin/ doxorubicin and 2 cycles of ifosfamide/doxorubicin. One of them had a local recurrence post salvage, disarticulation was done, followed by uncontrolled lung metastases. Death was the inevitable end. The second patient developed lung nodules, metastectomy was done. Another line of chemotherapy was given. Thereafter, the patient developed lung metastases where metastectomy was done for both lung field for three times. Then the patient had wide spread metastases (lung-bone-suprarenals) and was considered for palliative chemotherapy. He is still alive up till now.

The third patient developed lung metastases after end of chemotherapy. He is receiving another line of chemotherapy and is disease free upto now.

The last patient received 3 cycles of cisplatin/doxorubicin, did limb salvage and lost to follow up.

In the third group:

A- *The controlled group* included 34 patients with no evidence of local recurrence or residual tumor tissue based on clinical and radiologic (CT &/or MRI) findings.

1- ^{99m}Tc(V)DMSA scanning: 30 patients showed low ^{99m}Tc(V) DMSA uptake ratio < 1.7, which is the cut off value for tumor viability and their values ranged from 0.9-1.6. These were considered as true negative cases. The other 4 patients showed high ^{99m}Tc(V)DMSA uptake ratio > 1.7 and they were considered as false positive cases in view of absence of any clinical or radiologic evidence of tumor recurrence. This false positive results were found in osteosarcoma patients 3-6 months after limb salvage and were attributed to the presence of infection

in 2 cases and postoperative sequelae in the other two patients.

2- ^{99m}Tc-MIBI scanning: All 34 patients showed lower ^{99m}Tc-MIBI uptake ratio < 1.5 which is the cut off value for viable tumor tissue and their values ranged from 0.3 to 1.2. They were considered as true negative cases with no false positive results. All false positive cases by Tc-99m DMSA were true negative by MIBI.

3- Blood pool imaging: 33 patients were truly negative with low mean uptake ratio < 1.5 ranging from 0.5 to 1.7. Only one patient showed uptake ratio > 1.7 and was considered false positive result due to the presence of infection.

B- Patients with suspected residual or recurrent viable tumor tissue (19 patients)

1- ^{99m}Tc(V)MDSA scanning: 18 patients showed high mean uptake ratios (> 1.7) ranging from 1.8 to 8.6 and they were considered as true positive cases for tumor recurrence. One patient had low uptake ratio < 1.7 and was considered as false negative result.

2- ^{99m}Tc-MIBI scanning: 13 patients had high mean uptake ratios (> 1.5) ranging for 1.7 to 2.9 and were considered as true positive cases for tumor recurrence, (Fig. 5). The other 6 patients had low uptake ratios < 1.5, giving false negative results. This was likely attributed to multidrug resistance (MDR) phenomenon, (Fig. 6)

3- Blood pool imaging: 16 patients had high uptake ratios (> 1.5) ranging for 1.7-4.5 and they were considered as true positive cases for recurrence while the remaining 3 patients had low uptake ratios < 1.5 and were considered as false negative cases, (Table 6).

Sensitivity, Specificity, Accuracy, Positive and Negative predictive values (for 93 patients with different bone sarcomas).

The highest sensitivity was for ^{99m}Tc(V) DMSA and it approached 98.3% followed by that of ^{99m}Tc-MIBI (88.2%) and BP. (86%). While the highest specificity was for ^{99m}Tc-MIBI (97%) followed by that of BP. (94%) and of ^{99m}Tc(V)DMSA 88.2% (Table 7).

Table (1): Types of pathology in 93 patients with different bone sarcomas.

Type	No.	%
Osteosarcoma	68	73
Ewing's sarcoma	19	21
Chondrosarcoma	2	2
PNET	3	3
MFH	1	1
Total	93	100

Table (2): The range, mean value, true positive and false negative results for ^{99m}Tc(V)DMSA, ^{99m}Tc-MIBI and Tc-99m MDP blood pool scans in 40 patients of untreated bone sarcomas. (group I).

	^{99m} Tc(V)DMSA	^{99m} Tc-MIBI	BP.
Range	1.9 - 9.5	0.6 - 4.6	1.3 - 5
Mean ± SD	4.03±1.74	1.84±0.81	2.88±1.15
T P	40 (100%)	34 (85%)	37 (92%)
F N	0 (0%)	6 (15%)	3 (8%)

p- value (^{99m}Tc(V)DMSA, ^{99m}Tc MIBI) < 0.0001***
 p- value (^{99m}Tc(V)DMSA, BP) < 0.001**
 p- value (^{99m}Tc MIBI, BP) < 0.0001***

Table (3): The mean value of tracer uptake ratio after each cycle of chemotherapy compared between ^{99m}Tc(V)DMSA and ^{99m}Tc-MIBI in partially responding cases.

	^{99m} Tc(V)DMSA	^{99m} Tc-MIBI
Base line study	4.74 ± 2.73	2.16 ± 0.71
After 1 st cycle	4.4 ± 2.36	2.1 ± 0.72
After 4 cycles	4 ± 2	1.55 ± 0.56

Table (4): The mean value of tracer uptake ratio after each cycle of chemotherapy compared between ^{99m}Tc(V)DMSA and ^{99m}Tc-MIBI in stable cases.

	^{99m} Tc(V)DMSA	^{99m} Tc-MIBI
Base line study	4.0 ± 1.47	2.08 ± 1.45
After 1 st cycle	4.4 ± 1.72	2.02 ± 1.27
After 4 cycles	5.7 ± 1.93	2.53 ± 1.46

Table (5): The mean value of tracer uptake ratio after each cycle of chemotherapy compared between ^{99m}Tc(V)DMSA and ^{99m}Tc-MIBI in progressive cases.

	^{99m} Tc(V)DMSA	^{99m} Tc-MIBI
Base line study	4.5 ± 1.3	1.28 ± 0.63
After 1 st cycle	5.84 ± 2.63	1.1 ± 0.46
After 4 cycles	6.03 ± 2.19	1.38 ± 0.78

Table (6): Comparison between the mean values of tracer uptake ratio for ^{99m}Tc(V)DMSA, ^{99m}Tc-MIBI and BP. in equivocal and controlled groups.

	^{99m} Tc(V)DMSA	^{99m} Tc-MIBI	BP.
Equivocal group (n=19)	3.56 ± 2.14	1.79 ± 0.71	2.34±1.24
Controlled group (n=34)	1.28 ± 0.25	0.82 ± 0.35	1.0±0.24
Cut off value	1.7	1.5	1.5
p-value	< 0.0001***	< 0.0001***	< 0.0001***

Table (7): Sensitivity, Specificity, Accuracy, Positive and Negative predictive values of ^{99m}Tc(V)DMSA, ^{99m}Tc-MIBI and BP.

	^{99m} Tc(V)DMSA	^{99m} Tc-MIBI	BP.
Sensitivity	98.3%	88.2%	86.2%
Specificity	88.2%	97.1%	94.3%
Accuracy	94.6%	92%	89.2%
PV +	93.5%	97%	96.2%
PV -	96.8%	88.7%	80.5%

OSG (3) 4 X 30																	
Pre-operative therapy																	
Week	↑ 0	1	2	3	4	5	6	7	8	9	10	11	↑ 12	13			
Agent	I/A			I/A			P					P		Surgery ± metastatectomy			
Post-operative therapy																	
Week	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30↑
Agent		MTX	MTX	MTX	MTX	I/A			MTX	MTX	MTX	MTX		I/A			I/A
<p>Key: A: Adriamycin 75mg/m² over 24hrs infusion (max. cum. dose 300mg/m² < 6yrs., 375mg/m² > 6 yrs.)</p> <p>I : Ifosfamide, 1.8gm/m² d x5 + Mesna.</p> <p>P: Cis-platinum - 150mg/m² over 2 hours, mannitol diuresis employed.</p> <p>MTX: Mtx-LV: mtx, 12.5gm/m² over 6 hr = with L.V. 15mg/m² q 3h commencing 24hrs, after initiation of Mtx.</p> <p>↑* CBC-LFTs-KFTs + creatinine clearance test- Alkaline phosphatase Serum electrolytes (Na⁺, K⁺, Mg⁺⁺ and Ca⁺⁺).</p>																	

Fig. (1) : Treatment plan of osteosarcoma.

Preoperative						
Week	0	3	6	9	11	
	ap	ap	P	P	surgery	
Postoperative						
Week	12	15	18	21	24	27
GR	ap	ap	A	a		
SR	ai	ap	Ai	ap	ai	ai

GR = good pathological response ≥ 95% necrosis
 SR = standard pathological response < 95% necrosis
 a = adriamycin 37.5 mg/m² days 1 & 2 with DXR
 p = platinol 45 mg/m² days 1 & 2
 P = platinol 60 mg/m² days 1 & 2
 i = ifosfamide 1800 mg/m² days 1,2,3,4 & 5 with uromitexan

Fig. (2): Osteosarcoma study group (OSG) IV.

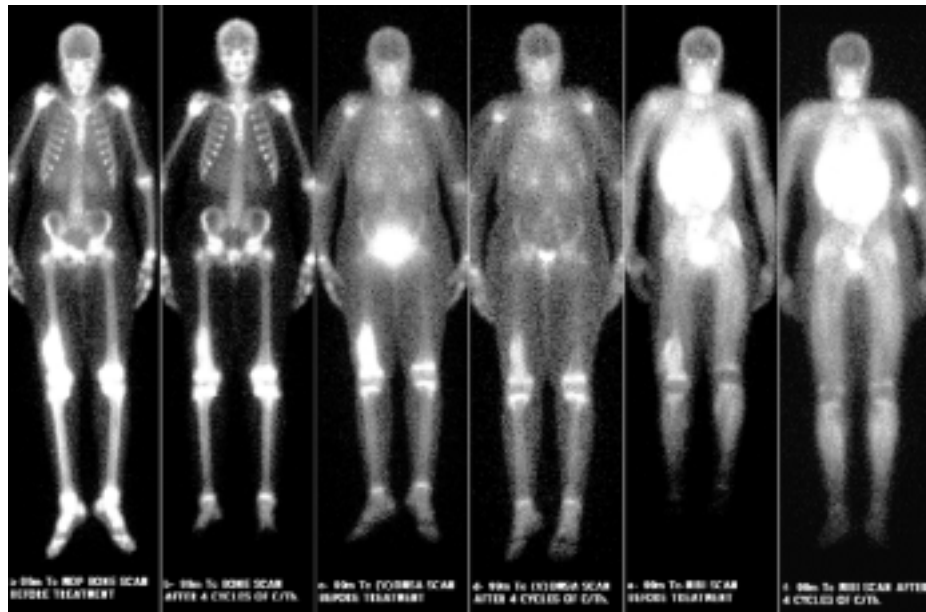


Fig. (3): Bone scan and ^{99m}Tc(V)DMSA scan revealed marked regression of the detected primary lesion while ^{99m}Tc-MIBI revealed total regression of the lesion. TN>70%.



Fig. (4): ^{99m}Tc(V)DMSA and ^{99m}Tc-MIBI scans at base line and after 4 cycles of chemotherapy revealed disease progression.

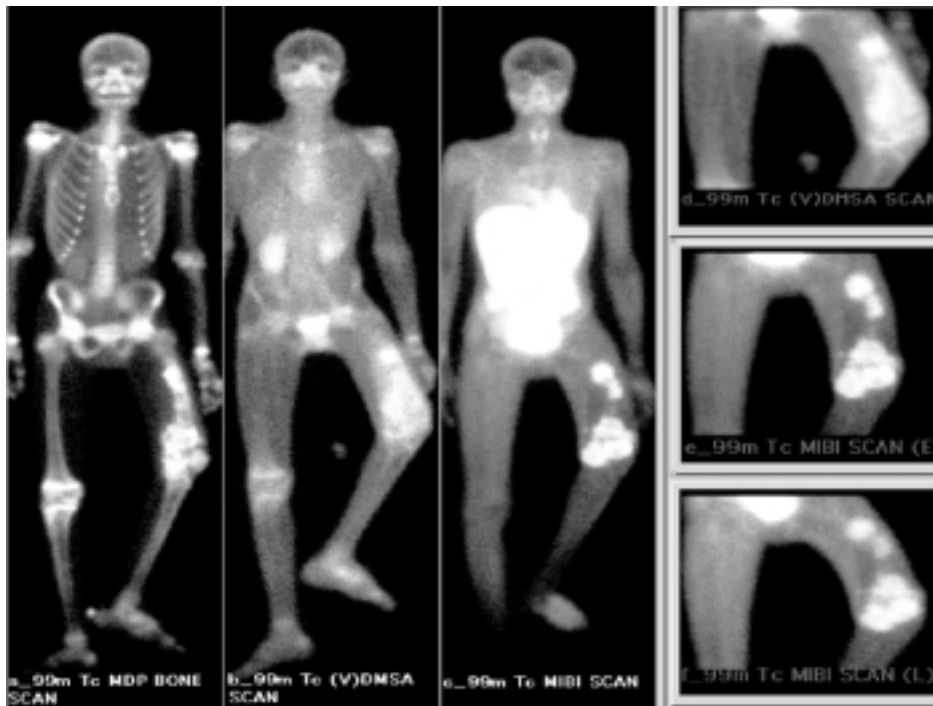


Fig. (5): Bone scan, ^{99m}Tc (V)DMSA and ^{99m}Tc -MIBI revealed local tumor recurrence.

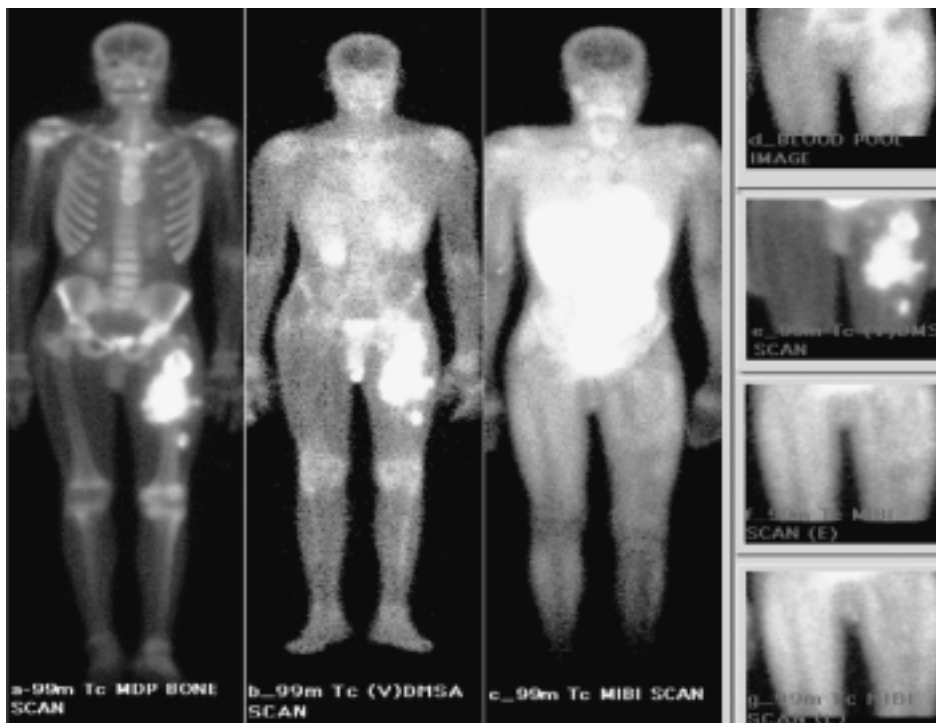


Fig. (6): Blood pool skeletal imaging and ^{99m}Tc (V)DMSA revealed a large residual tumor tissue while ^{99m}Tc -MIBI revealed absence of any tumor tissue attributed to MDR phenomenon.

DISCUSSION

The administration of preoperative and post-operative chemotherapy is nowadays considered the standard treatment for high grade osteosarcoma [10]. The strategy initially evolved from early attempts at limb salvage surgery at the Memorial Sloan Kettering Cancer Centre where endoprosthetic devices were used for limb reconstruction. Since fabrication of these devices required two to three months, patients were treated with chemotherapy following initial biopsy while waiting definitive surgery. Tumor shrinkage in response to chemotherapy appeared to facilitate limb salvage and patients treated with presurgical chemotherapy appeared to fare better than concurrent patients treated with surgery and postoperative chemotherapy alone [11]. Moreover, histologic evaluation of the excised tumor after treatment with chemotherapy was found to be a powerful prognostic factor for tumor recurrence. Unfavorable responders were more likely to develop distant metastases despite continuation of chemotherapy after surgery [12]. The prognostic value of responsiveness of the primary tumor to preoperative chemotherapy has been confirmed in several trials [13-15]. Several systems for grading the effect of presurgical chemotherapy have been proposed based on histological assessment of cellularity and necrosis in the excised specimen. In the Huvos grading system, favorable responses (grade 3 and 4) indicate extensive to complete (90 % or greater) response in the primary tumor [16], whereas unfavorable histologic responses (grade 1 and 2) imply minimal tumor necrosis and persistence of viable tumor [16]. In most studies patients with a favorable response fare extremely well, while those with unfavorable response are likely to develop distant metastases [10,11].

The main applications of nuclear medicine imaging in oncology are assessment of response to therapy and detection of residual/recurrent viable tumor tissue after therapy. Three phase bone scan define flow in the early dynamic phase, soft tissue extent in the blood pool phase and the degree involvement in the late static phase. However, this method can not be used accurately to assess the response to therapy as MDP uptake reflects the osteoblastic activity and bone remodelling rather than tumor viability.

Thallium-201 chloride and Tc-99m sestamibi are the two well established bone and soft tissue

tumor imaging agents used for evaluation of tissue tumors as well as other tumors [2]. Thallium - 201 is not readily available at the nuclear medicine department, relatively expensive with low administered dose and has high skeletal muscle uptake compared to MIBI and pentavalent DMSA. However it has the advantage of being not affected by the MDR phenomenon. Tc-99m MIBI is readily available any time at the nuclear medicine department and is relatively cheaper than thallium. However its main disadvantage is the affection by the MDR phenomenon. Tc-99m pentavalent DMSA has the advantages of MIBI (being cheaper and readily available with no skeletal muscle uptake) and it seems that it is not affected by the MDR phenomenon. The exact mechanism of Tc-99m pentavalent DMSA in tumor cells is not exactly understood [17,18]. It is postulated that pentavalent DMSA resembled the phosphate ion, and suggested that this was the mechanism by which the tracer accumulated in tumors. Tc-99m DMSA is likely taken up by the tumor cells in a similar manner to phosphate molecules that accumulates at sites of increased protein metabolism [17,18]. Yokohama et al. [19] believed that increased blood flow and acidic PH of tumor cells are additional parameters relevant to the accumulation of Tc-99m pentavalent DMSA in tumor cells.

They also reported that tumor/blood ratio of 3.7 and 6 were obtained at 3 and 24 hours post injection respectively.

In the current study, Tc-99m pentavalent DMSA was able to diagnose the malignant bone tumor in the 40 patients studied in this group. It gave the highest sensitivity of 100% with no false negative results. Whereas lower sensitivity was noted for ^{99m}TcMDP blood pool images and ^{99m}Tc-MIBI scans accounting for 92% and 85% respectively. The lower sensitivity for ^{99m}Tc-MIBI could be attributed to the presence of multidrug resistance (MDR) which occur due to transfection of MDR-1 gene that encodes P-glycoprotein (P-gp) into drug sensitive cells resulting in acquisition of resistance to drugs in the MDR family. The latter includes vinca alkaloids as vinblastine, anthracyclines as adriamycin, epipodophyllotoxins as etoposide, antibiotics as actinomycin D & taxol [20]. However MDR cells are not cross resistant with alkylating agents as methotrexate, cytarabine, cisplatin and 5-

fluorouracil [21]. P-glycoprotein is an energy dependent drug efflux pump responsible for the MDR phenomenon and its expression results in concomitant increase in drug efflux and consequently drug resistance [22].

Multiple in vivo studies proved the utility of Tc-99m MIBI in the detection of MDR in various tumors [23-25], they compared early (15-20 min) and late (2-3 hrs) Tc-MIBI images in lung cancers compared to chemotherapy response. They all found that Tc-MIBI SPECT images (either positive with good response or negative with bad response) correctly predicted the chemotherapy response and significantly correlated with survival rate. They concluded that Tc-MIBI images had the potential to predict MDR related P-gp expression and consequently patient prognosis and expected response to chemotherapy.

Kostakuglu et al. [26] prospectively studied 48 patients with breast and lung tumors with Tc-MIBI imaging and immunohistochemical analysis using monoclonal antibody developed against the internal epitope of P-gp. Their results showed an inverse correlation between the tumor to background ratios and the density of P-gp expression in tumor tissue. They concluded that Tc-MIBI imaging was useful to determine the overexpression of P-gp in patients with malignant tumors.

Regarding bone & soft tissue tumors Garcia et al. [27] compared the uptake of F-18 FDG and Tc-MIBI in 48 patients with recurrent bone & soft tissue tumors. The results were correlated with histopathological findings or long-term follow-up. They found that FDG PET images had higher sensitivity and specificity as well as better quality images and higher tumor to normal uptake ratio. They also found 4 out of 9 patients with positive FDG and falsely negative MIBI failed to respond to the given therapy. They concluded that FDG PET images were superior to MIBI for detection of recurrent bone tumors and that a positive FDG and negative MIBI might suggest the presence of MDR with bad prognosis.

^{99m}Tc(V)MDSA appeared in our study not to be dependent on MDR phenomenon with evident higher ratios than Tc-MIBI and no false negative results for primary untreated lesions. This advantage of higher sensitivity for detection of malignant bone tumors can help to decrease

the number of missed, recurrent cases if evaluated by ^{99m}Tc-MIBI alone. However further studies are needed to document this finding using both tracers and immunohistochemical detection of the P-gp by monoclonal antibodies.

In the present study, patients were classified scintigraphically according to their response to preoperative chemotherapy into partially responding, stable and progressive disease in comparison to their histopathological results where TN: > 50% represented partially responding disease, TN: 35-50% represented a stable disease and TN: 10-35% represented a progressive disease. Unfortunately there were no cases with good response (i.e. TN > 90%) in the studied group. It is widely accepted that tumor necrosis of less than 90% denotes a poor response to chemotherapy [11]. A lower percentage of tumor necrosis indicates the presence of hundreds of thousands of viable, chemotherapy resistant tumor cells and correlates with a greater potential of distant metastases and local recurrence. However, tumor necrosis of more than 90% indicates good response to preoperative chemotherapy. This is important, not only for prognosis but also for selection of alternative chemotherapeutic regimens in patients who fail to demonstrate good response to preoperative chemotherapy and in operative planning as well [1,10,11,13,15,16,28].

On the other hand, the histologic interpretation of a good or bad response, which must be done after tumor resection and is based on the percentage of tumor necrosis, is a subjective procedure and may be difficult to quantify among pathologists. In addition, examination of the entire resected specimen in order to fully evaluate the amount of necrotic and viable cells, is a very tedious process and may take several days to be completed. In addition, the criteria of cell viability may be different among different competent pathologists [29].

Ohtomo et al. [30] evaluated the response of preoperative chemotherapy in 30 patients with high grade osteosarcoma by using thallium-201. They compared the scintigraphic results with the percentage of tumor necrosis after surgery and classified their patients into 3 grades. Grade 1 patients (Tumor necrosis less than 60%) showed an increase of post chemotherapy tumor to background ratios of 67.1%, grade 2 patients (Tumor necrosis from 60 to 89%) showed a

decrease of post chemotherapy tumor to background ratios of 38% and grade 3 patients (Tumor necrosis of more than 90%) showed a significant decrease of tumor to background ratios of 106%. They concluded that scintigraphic evaluation of tumor response of osteosarcoma to preoperative chemotherapy by using thallium-201 is an easy and reliable method as compared to pathological assessment of tumor necrosis. Similar data was obtained by Mostafa et al. [5], for 28 patients with osteosarcoma and Ewing's sarcoma evaluated by early and late Tc-99m MIBI images to assess response to preoperative chemotherapy compared to the percentage of tumor necrosis by histo-pathology. Twelve patients with complete response (Tumor necrosis more than 90%) had a decrease of tumor to background ratio from 2.75 and 2.07 for early and late images to 0.95 and 0.89 respectively. Partial responders (Tumor necrosis of 50 to 90%) showed a decrease of T/N ratio from 5.03 & 5.02 to 2.77 & 2.43 after chemotherapy. While bad responders (Tumor necrosis less than 50%) had an increase in tumor to background ratios from 2.93 & 2.96 to 3.8 & 2.8 respectively for early & late images.

Recently, Fluorine-18 FDG PET was effectively used to monitor response of osteosarcoma to preoperative chemotherapy. Consequently Schulte et al. [31] evaluated the response of 27 patients with osteosarcoma to preoperative chemotherapy by using F-18 FDG PET. According to their scintigraphic results compared to histopathology, they graded their patients into 6 grades. Their method discriminated responders (grades 1-3) and non responders (grades 4-6) that properly correlated to the percentage of tumor necrosis. In addition, lung metastases were detected in 4 patients of the poor responder group.

Nair et al. [32], similarly evaluated 16 patients with osteosarcoma by F-18 FDG PET and compared the change in T/N ratios with the percentage of tumor necrosis after surgery. Baseline T/N ratios ranged from 2.5 to 8.7. After treatment patients were classified into two groups. Good responders with T/N ratios less than 0.7 and tumor necrosis of more than 90% and bad responders with higher T/N ratios and less than 90% tumor necrosis.

Ongolo-Zogo et al. [33], evaluated the response to preoperative chemotherapy in 12 patients with surgically resectable osteosarcoma

by using dynamic (contrast enhanced) MRI and three phase bone scan. They evaluated their patients twice, after two and 4 cycles of preoperative chemotherapy and compared the results to the percentage of tumor necrosis after surgery. They found that after two cycles only, the two imaging modalities failed to predict the final histopathological results, but after the end of treatment (4 cycles) they were able to correctly identify responders from non-responders.

Unfortunately, the literature is poor in scintigraphic data that evaluates the response of osteosarcoma after one or two cycles of chemotherapy. However in our opinion we still believe that this is possible and if succeeded this will save time and money for patients as those who are likely to respond will continue 3 or more cycles of the same treatment protocol while non responders are likely to change their treatment regimen or go straightforward to surgery saving time and money in useless treatment.

Computed tomography and MRI are unable to accurately differentiate between residual or recurrent viable bone tumors and post therapy fibrosis in view of the substantial amount of anatomic distortion which take place following treatment [4].

In a study performed by Mostafa et al. [5] 6 false positive cases among 24 patients of adequately controlled bone and soft tissue tumors were falsely diagnosed by C.T and/or MRI as possible recurrent tumor which were not evident on ^{99m}Tc-MIBI scan and histopathologic biopsy. Additionally 2 cases with false negative results were reported as post operative radiation changes by CT among 16 patients with positive scintigraphic evidence of recurrent viable tumor by Tc-MIBI that were proved by histopathology. Thus the missing rate in follow up of bone, soft tissue tumors by CT and MRI in their study was evident in 8 out of 40 patients (20%) versus (5%) only by ^{99m}Tc-MIBI scanning. The sensitivity, specificity and accuracy of ^{99m}Tc-MIBI in detection of recurrent bone, soft tissue tumors in their study were 93%, 95% and 92% as compared to 86%, 75% and 84% for CT and/or MRI respectively.

In our study, 19 patients with equivocal findings on CT and/or MRI for local bone recurrence were evaluated by ^{99m}Tc(V)DMSA, ^{99m}TcMIBI and blood pool imaging. Eighteen patients stud-

ied with $^{99m}\text{Tc(V)}$ DMSA showed high uptake ratios ranging from 1.8-6.8 with high L/N ratio > 1.7 which was the cut off value for $^{99m}\text{Tc(V)}$ DMSA for viable tumor tissue and they gave a true positive result with a sensitivity of 94% while only one patient with low grade osteosarcoma and inadequate treatment showed uptake ratio < 1.7 giving a false negative result.

In the same group of 19 patients studied with $^{99m}\text{Tc-MIBI}$. 13 patients were truly positive and gave uptake ratios > 1.5 with a sensitivity of 68%. While the other 6 patients were falsely negative and they showed uptake ratios < 1.5 , this could be attributed to MDR. These 6 patients were truly positive on $^{99m}\text{Tc(V)}$ DMSA. These findings reinforce the conclusion that $^{99m}\text{Tc(V)}$ DMSA uptake is not affected by MDR which may result in increasing number of missed patients on $^{99m}\text{Tc-MIBI}$ scanning.

In the blood pool imaging 14 out of 19 patients showed uptake ratio > 1.5 with a sensitivity of 73% while the other 5 patients were falsely negative and showed low uptake ratios. Four out of those patients gave true positive results on $^{99m}\text{Tc(V)}$ DMSA scanning.

So the sensitivity in the equivocal group of our study for recurrent viable tumor tissue was found to be highest for $^{99m}\text{Tc(V)}$ DMSA (94%) followed by blood pool imaging 73% and lastly for $^{99m}\text{Tc-MIBI}$ (68%).

On the other hand, 30 out of 34 patients of adequately controlled bone tumors with no radiologic evidence of recurrence, showed by $^{99m}\text{Tc(V)}$ DMSA scan L/N ratios < 1.7 and were considered as true negative cases (87%). The other 4 patients had L/N ratios > 1.7 and were considered as false positive cases (13%). The possible causes for this false positive results were the presence of infection in 2 cases and recent limb salvage surgery in the other 2 patients with attempts of healing evident by increased uptake at both ends of the bony graft. In contrast $^{99m}\text{Tc-MIBI}$ scan showed no false positive results in the 34 patients studied in this group and all were considered as true negative cases (100%).

Our results were also comparable to those of Zaher et al. [34] in their series of 32 patients with suspected residual/recurrent bone & soft

tissue tumors. They used thallium-201 and Tc MIBI as well as three phase bone scan. Their patients were classified into 3 groups. The first group included 16 patients with no clinical or radiological evidence of recurrence. They all showed low T/N ratios with blood pool, thallium and Tc-MIBI less than 1.5. The second group included 6 patients with post operative signs of infection with equivocal results by CT and/or MRI. Blood pool images showed high ratios in all cases due to increased vascularity while thallium and Tc-MIBI showed falsely elevated ratios in only 2 patients and high true positive ratios for the other 4 patients that all proved later to have tumor recurrence with superimposed infection. The third group included the remaining 10 patients with pathological evidence of tumor recurrence. Thallium showed 2 false negative cases while Tc-MIBI showed only 1 false negative case with true positive results in 8 and 9 patients respectively. They reported the sensitivity, specificity and accuracy of thallium and Tc-MIBI in the detection of recurrent viable tumor to be 81.8% and 87.6%, 87.6% and 87.6%, 84.4% and 87.6% respectively

In our study, the specificity in the controlled group was found to be the highest for $^{99m}\text{Tc-MIBI}$ (100%) followed by blood pool imaging (97%) and lastly for $^{99m}\text{Tc(V)}$ DMSA (87%). This is likely attributed to high uptake of pentavalent DMSA at sites of bone remodelling at the healing ends of the applied bone graft.

Fluorine-18 FDG PET studies proved to be effective in the follow-up of patients with bone tumors to detect both local tumor recurrence and distant metastatic deposits in a single study. Franzius et al. [35] evaluated 27 patients with osteosarcoma and Ewing's sarcoma in order to detect local recurrence and/or distant metastatic deposits, compared to local MRI, CT chest and bone scan. Their results were confirmed by histopathological analysis and/or long-term follow-up. They accurately diagnosed 52 sites of viable recurrent tumor (local=7, distant osseous=22, distant pulmonary=13 and distant soft tissue=10). They found that F-18 FDG PET studies had a sensitivity of 96%, specificity of 81% and accuracy of 90% While corresponding values for conventional imaging were 100%, 56% and 82% respectively. They concluded that FDG PET had the advantage of high specificity and accuracy compared to other imaging modal-

ities as well as the ability to screen the whole body in a single study.

Similar results for FDG PET were found by Bredella et al. [36]. They compared FDG PET images with Gadolinium enhanced MRI images in 12 patients with bone and soft tissue tumors after treatment. They found that MRI was unable to accurately differentiate residual viable tumor from post therapy fibrosis in 9 patients and MRI images were inadequate for interpretation in two patients after limb salvage who had metallic plates. On the other hand FDG images correctly diagnosed 5 patients with tumor recurrence and were confirmed by biopsy and excluded the presence of viable tumor in other 4 patients that were clinically free by long-term follow-up. They concluded that FDG PET images were very helpful in these patients after treatment specially those with metallic implants that cause many artifacts on MRI images.

Finally, the 93 patients with different bone sarcomas included in this study showed a sensitivity of 98.3%, 88.2% and 86.2%, specificity of 88.2%, 97.1% and 94.3%, accuracy of 94.6%, 92.2% and 89.2%, positive predictive value of 93.5%, 97% and 96.2% and negative predictive value of 96.8%, 88.7% and 80.5% for $^{99m}\text{Tc}(\text{V})\text{DMSA}$, $^{99m}\text{Tc}\text{-MIBI}$ and blood pool imaging respectively.

Conclusion:

- $^{99m}\text{Tc}(\text{V})\text{DMSA}$ is a sensitive radiopharmaceutical used for evaluation of primary malignant bone tumors in comparison to $^{99m}\text{Tc}\text{-MIBI}$ and skeletal blood pool imaging.

- Scanning with $^{99m}\text{Tc}(\text{V})\text{DMSA}$ may give an early highlight on the response to chemotherapy following the first cycle, being not affected by MDR phenomenon as $^{99m}\text{Tc}\text{-MIBI}$.

- Evaluation of response to chemotherapy after the first cycle may help to select patients with partial response for continuation of chemotherapy to 3 or 4 cycles. Whereas those with progressive disease and continuously rising quantitative ratios may need either changing the protocol of treatment or early surgical interference.

- $^{99m}\text{Tc}(\text{V})\text{DMSA}$ scanning has higher sensitivity in detection of locally recurrent, peripherally located bone tumors whereas $^{99m}\text{Tc}\text{-MIBI}$

scanning has higher specificity. So, if possible, it is clinically useful to combine both radiopharmaceuticals to get the advantage of both high sensitivity and specificity.

Recommendations:

- Multi-center trials for evaluating the response of primary osteosarcoma lesion to the first cycle of preoperative chemotherapy using thallium-201 chloride, $\text{Tc-}^{99m}\text{MIBI}$ or $\text{Tc-}^{99m}\text{DMSA}$ are recommended aiming to guide further management of primary bone sarcomas.

- Combined techniques of $^{99m}\text{Tc}(\text{V})\text{DMSA}$ and $^{99m}\text{Tc}\text{-MIBI}$ are recommended for follow up of patients with bone tumors to gain the highest sensitivity of $^{99m}\text{Tc}(\text{V})\text{DMSA}$ and highest specificity of $^{99m}\text{Tc}\text{-MIBI}$.

- Further studies using Tc-^{99m} pentavalent DMSA and $\text{Tc-}^{99m}\text{MIBI}$ must be done to prove that pentavalent DMSA is not affected by multidrug resistance. The results should be confirmed with Immunohistochemical analysis of P-glycoprotein in tumor samples.

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