# Nuclear medicine techniques provide unique physiologic characterization of suspected and known soft tissue and bone sarcomas

Rodney J Hicks

Department of Nuclear Medicine, Peter MacCallum Cancer Institute, Locked Bag No. 1 A'Beckett Street, Melbourne VIC 3000, Australia. Tel + 61-3-9656-1020. Fax + 61-3-9656-1406

*Primary bone and soft tissue tumors have a range of physiologic, biochemical and genetic characteristics which differentiate them from benign tumors and normal tissues. These "fingerprints" are amenable to noninvasive detection and quantification using nuclear medicine techniques. Functional characterization using radiotracers imaged using a gamma camera or positron emission tomography (PET) can provide unique but complementary information to that provided by anatomically-based imaging modalities such as plain radiography, computed tomography (CT) and magnetic resonance imaging (MRI). This is particularly important following therapy when residual mass lesions may cause a dilemma regarding the need for further treatment. Lack of understanding of the role of functional imaging has impeded more rational use of nuclear medicine in this field. This review addresses the role of nuclear medicine techniques in the primary evaluation, staging, and therapeutic monitoring of soft tissue and bone sarcomas. The potential of delivering targeted radiotherapy to these tumors with radiopharmaceuticals which exploit the unique pathophysiology of each individual malignant lesion is also addressed.*

Nuclear medicine techniques are often overlooked in the assessment of soft tissue and bone tumors with heavy reliance placed on anatomical imaging including radiography, ultrasound, computer x-ray tomography (CT) and, more recently, magnetic resonance imaging (MRI). Despite this, radionuclide studies potentially offer unique information about tumor physiology and biology which may be complementary or incremental to anatomical imaging. This is particularly true following therapy when residual structural abnormality often persists both with and without residual tumor.

## *Physiological principles of tumor imaging*

There are a number of physiological characteristics of malignant soft tissue and bone tumors which can potentially be used to differentiate them from normal tissues and from benign diseases. These characteristics can be as simple and non-specific as an increase in perfusion or as specific as a genomic mutation. Qualitative and quantitative differences in tissue physiology, biochemistry, and genomic structure all provide potential means for targeting tumor diagnosis and therapy.

Due to neovascularisation, most malignant soft tissue and bone tumors are hypervascular with the pattern of perfusion abnormality potentially helpful in differentiating benign from malignant processes. Increased perfusion around the periphery of a lesion with decreased or absent blood flow to the central regions occurs particularly in the presence of central tumor necrosis which is seen primarily in aggressive tumors. Soft tissue, third space edema is also a common but non-specific physiological feature of malignant soft tissue and bone tumors, often reflecting disruption or loss of integrity of the endothelium. As with lesion perfusion, the pattern of alteration of blood volume or third space fluid can provide important information regarding tumor biology.

Bone forming tumors, particularly including osteosarcoma but also some high grade chondrosarcomas, are characterised by malignant osteoid formation. Other tumors may arise in the bone marrow space or in extraosseous soft tissues and secondarily invade the medullary bone trabeculae and cortical bone. While locally destructive, these tumors usually invoke reactive osteoblastic activity at the periphery of the lesion. Increased incorporation of calcium and phosphates into bone characterises both bone formation by the tumor itself and reactive osteoblastic activity.

Heightened proliferative activity is a particular feature of malignant lesions and is associated with a wide range of metabolic consequences. Increased mitochondrial numbers, altered cell membrane potentials, increased protein synthesis, accelerated glycolysis, alteration in glucose transporter protein expression are but a few of the biochemical and physiological correlates of this process. Scar tissue that forms after surgery, radiotherapy or successful chemotherapy has decreased metabolic activity potentially allowing differentiation between scar tissue and residual tumor by functional imaging.

Finally, there are features of the cell type involved in malignant transformation which characterise the tumor. These include cell surface receptor and transport protein complexes, cytosolic enzymes, cytoskeletal proteins and genetic material including DNA. The specific ways in which these physiological and biological processes are exploited in nuclear medicine diagnosis and therapy are detailed below.

## *Technical aspects of tumor imaging and therapy*

Relative differences in radiotracer uptake by normal tissues, benign lesions and malignant tumors are fundamental to both diagnostic and therapeutic nuclear medicine techniques. Single photon agents, so called to differentiate them from positron emitting radionuclides which produce 2 photons for each radioactive decay, are generally now imaged with a gamma camera. The modern gamma camera is a digital imaging device which can supply planar images in a number of projections and can perform single photon emission computed tomography (SPECT). The latter yields a volume of information which allows three-dimensional evaluation of radiotracer distribution. By removing the overlap of tissues with differing radiotracer uptake characteristics it also increases the contrast with which lesions are seen. SPECT imaging is more technically demanding than planar imaging but provides substantial incremental information in evaluating bone and soft tissue tumors (Podoloff et al. 1992). The tomographic nature of the study lends it to direct visual comparison or image coregistration with CT or MRI.

The spatial resolution of both planar and SPECT imaging is dependent on the characteristics of the gamma camera and particularly on the type of collimator used. This, in turn, is partly determined by the energy of the radioactive emissions being detected. For example the higher energy of Ga-67 than Tc-99m requires use of a medium energy collimator as opposed to a low energy collimator. The higher the energy rating of a collimator, the lower the spatial resolution. For any given energy rating, there is also an inverse relationship between resolution and sensitivity of the gamma camera for detection of emitted photons. The higher the resolution, the lower the sensitivity. Thus, acquiring statistically adequate images requires longer with high resolution collimators. Within practical limits the goal of bone tumor imaging should always be to obtain the highest possible spatial resolution. For local bone pathology in the appendicular skeleton, pin-hole imaging provides excellent spatial resolution albeit at the cost of prolonged imaging times.

Positron emission tomography (PET) does not require use of a lead collimator, relying instead on electronic collimation by way of coincidence detection. The physics of PET are beyond the scope of this article but suffice it to say that the spatial resolution of current PET scanners is half to one quarter that of SPECT systems while their sensitivity is dramatically greater (Jones, 1996). Recent advances in PET scanners have made whole body scanning feasible allowing more accurate tumor staging.

The type of radioactive emission is important both to the imaging of radiotracer distribution within the body and determining the radiation dose received by various target organs. Gamma rays and x-rays which are highly penetrating have much less radiobiologic effects than particulate emissions such as beta particles (electrons released from the atomic nucleus), conversion and Auger electrons (released from orbiting electron shells of the atoms) and alpha particles (the equivalent of a helium atomic nucleus expelled from the nucleus of large, radioactive nuclides). Agents which have particulate emissions have a potential therapeutic role, delivery high radiation doses in the immediate vicinity of where the radiopharmaceutical localizes. Depending on the type and energy of emission, the distance that these therapeutic emissions travel in tissue may vary from a few microns to several millimeters (Volkert et al. 1991).

## *Nuclear medicine targets tumor pathophysiology*

### Available imaging techniques

Nuclear medicine is ideally suited to characterisation of the physiological and biochemical "fingerprints" of tumors since it is based on the tracer kinetic model. This model, developed by George Hevesy early this century states that minute quantities of a radioactive chemical will pass through or "trace" the same steps as the non-radioactive form of this chemical without itself altering the process being studied (Hevesy,

1962). Although the term "radiopharmaceutical" is often used for these "radiotracers", the concentration of the carrier compound is usually in the nanogram to microgram range and well below the amount required for a pharmacological effect. Administration of only a minute amount of tracer chemical makes nuclear medicine extremely safe with an extremely low incidence of allergic reactions or other side effects (Keeling, 1994). The radiation exposures involved with most nuclear medicine studies are comparable to other diagnostic radiology procedures. While radiographs, CT, and MRI may define the location, size and tissue relations of tumor masses and biopsy findings (usually) provide a definitive diagnosis, functional imaging can also provide additional information which can non-invasively guide patient management, monitor therapy and assess prognosis.

## *Specific nuclear medicine investigations*

### Vascular phase imaging

Administration of any radiotracer in a sufficient activity and with suitable imaging characteristics can allow assessment of regional perfusion. The blood flow phase of bone scanning is an example of this technique but flow imaging can also be obtained with agents such as technetium-99m (Tc-99m) diethyltriamine pentacetic acid (DTPA), a renal imaging radiotracer, and by using positron emission tomography (PET) with oxygen-15 (O-15) water or nitrogen-13 (N-13) ammonia. Early uptake of thallium-201 (Tl-201) and other cardiac imaging agents is also largely dependent on regional perfusion.

Due to neovascularisation, most malignant soft tissue and bone tumors are hypervascular. Unfortunately many benign lesions also have this characteristic. The classic example of a benign bone tumor with hypervascularity is the osteoid osteoma which has extremely high capillary phase blood flow. Osteomyelitis, fractures and healing avascular necrosis and bone infarcts can all have increased local perfusion. The pattern of perfusion abnormality can however be helpful in differentiating benign from malignant processes. Increased perfusion around the periphery of a lesion with decreased or absent blood flow to the central regions occurs particularly in the presence of central tumor necrosis which is seen primarily in aggressive tumors. A recent study of three-phase bone scanning demonstrated that incorporation of the blood flow and blood pool images substantially improves specificity and overall accuracy of bone scanning in the detection of bone tumors (Caluser et al. 1994).

Tumor vascularity will generally decrease with effective therapy either returning to similar levels seen in adjacent bone or soft tissue or becoming relatively avascular if healing fibrosis occurs. Peripherally increased blood flow with central hypoperfusion is typically seen in aggressive tumors which undergo central necrosis as they outgrow their vascular supply.

Although often called blood pool scanning, the primary physiologic process detected on early imaging after equilibration of radiotracer in the vascular space is often also reflective of third space fluid. Agents which remain for a variable period in the intravascular space and which enter third space are quite diverse and include bone scanning agents such as Tc-99m methylene diphosphonate (MDP) early after injection, the renal imaging agent Tc-99m DTPA and O-15 water which can be imaged by PET. Blood pool activity is often concordant with changes in regional perfusion but there are many exceptions to this. Cavernous hemangioma of bone for example usually have low blood flow but increased blood volume. As with perfusion imaging findings, a decrease in blood pool activity following treatment generally reflects a favourable therapeutic response but uncontrolled tumor growth may lead to central tumor necrosis and an associated decrease in blood pool activity leading to a "doughnut" appearance. This is not, however, a specific finding since central tumor necrosis can also occur following successful radiotherapy of osseous and soft tissue malignancy following radiotherapy. In this setting peripherally increased vascularity can occur in an organising fibrous capsule early after treatment. This process may lead to diagnostic difficulties when using blood flow imaging techniques but is also problematic for MRI and CT scanning which also show peripheral contrast enhancement in this setting. Chondrosarcomas may also have relatively low central vascularity relating to the presence of more mature stromal elements in the centre of the lesion while the active tumor growth generally occurs in the periphery.

## *Bone scanning—more than a low-resolution radiography*

The most widely used and well recognised nuclear medicine technique in evaluation of bone and soft tissue tumors is the bone scan. The whole body screening capability of this scan provides cost-effective screening for metastatic spread or multifocal bone pathologies. Unlike CT scanning or skeletal survey, radionuclide scanning of the entire skeleton does not involve increased radiation exposure above that involved in evaluation of the primary lesion alone. Formation of malignant osteoid by primary bone forming tumors or the reaction of adjacent normal bone to locally invasive soft tissue tumors increases local osteo-

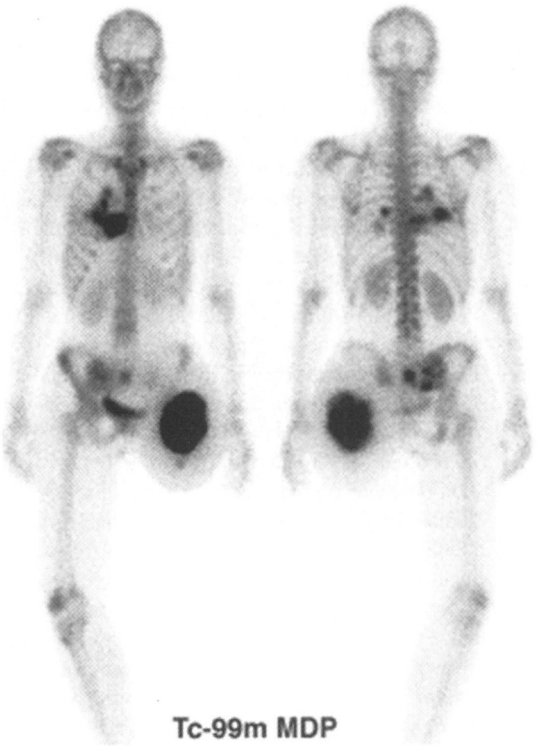


*Figure 1. Bone scanning with Tc-99m MDP demonstrates intense but heterogeneous osteoblastic activity in relationship to recurrent osteosarcoma of the left amputation stump. Soft tissue metastases in the right lung also demonstrate active radiotracer accumulation. A photopenic defect in the right sacrum represents a destructive bone metastasis without significant osteoblastic activity.*

blastic activity and leads to concentration of bone seeking radiotracers including Tc-99m methylene diphosphonate (MDP) and uptake of other bone seeking radiopharmaceuticals. However, because of the wide range of pathologies that can affect bone turnover, correlative techniques are often required to make a diagnosis of bone malignancy. While a decrease in bone turnover throughout a lesion following therapy strongly suggests therapeutic response, the converse is not as predictive of lack of response. Increased osteoblastic activity may accompany bone healing with successful treatment of lesions which have caused significant bone destruction. This early healing phenomenon is often termed a "flare" response.

Radiographs provide information about the calcium content of bone at a particular time. Despite representing the balance between osteoblastic and osteoclastic activity over a prolonged period it provides no information about the current nature of this balance. The bone scan, on the other hand, provides information about the process of bone formation and relatively little about current calcium content.

In osteosarcoma high uptake of bone tracers is usually noted in both the primary tumor and secondary deposits but, as with other aggressive tumors, central tumor necrosis often leads to a photopenic central region in large lesions. High Tc-99m MDP uptake in soft tissue tumors is uncommon except in osteosarcoma metastases (Figure 1) or in primary neuroblastomas of childhood but can be seen in some adenocarcinomas with mucinous histology. The intensity of osteoblastic activity with Ewing's sarcoma is variable but tends to be less than with osteosarcoma. High bone tracer uptake in relationship to chondroid tumors in our experience increases the likelihood of malignant transformation. In chondroid tumors, relative or absolute photopenia in the central part of the mass lesion does not necessarily reflect central tumor necrosis since mature chondroid matrix usually does not accumulate Tc-99m MDP. Malignant fibrous histiocytoma in bone usually invokes moderately intense osteoblastic activity at the bone tumor interface but the tumor itself usually doesn't take up bone seeking radiopharmaceuticals. Lack of abnormality on the bone uptake phase of the study strongly favours a benign or mature bone process but lesions arising in the medullary cavity may have relatively little osteoblastic activity related to them. MRI scanning is the most sensitive technique for detecting such lesions.

In addition to information regarding the rate of bone matrix formation, further physiological data can be obtained in conjunction with bone scanning which can be used to further characterise the nature of bone and soft tissue disease. The amount of radioactivity administered with a bone scan is sufficient to obtain a

dynamic sequence of images or movie representing the perfusion of a particular body part. This allows assessment of the arterial and capillary phase blood flow to tumors analogous to angiographic studies but with lower spatial resolution. Increased vascularity extending beyond the normal bony limits suggests an associated soft tissue mass and is consequently a more ominous finding than changes confined to the bone. As the radiotracer becomes dispersed in the vascular space and leaks across damaged capillaries into third space, fluid images can be obtained that correspond physiologically to the extracellular fluid space. Images obtained early after radiotracer injection reflect this phase and are termed "blood pool" images. This information provided by these images are often similar to that of contrast studies used for CT and MRI scanning, albeit with lower spatial resolution and anatomical detail. Osteosarcoma, chondrosarcoma, Ewing's sarcoma, malignant fibrous histiocytoma, rhabdomyosarcoma all tend to be highly vascular. When soft tissue sarcomas abut the periosteum this local hyperperfusion may increase delivery of bone seeking radiotracers to the nearby bone and increased cortical activity in the adjacent bone does not necessarily reflect tumoral invasion. As mentioned previously "doughnut" patterns of hyperperfusion are suggestive of malignant lesions and tend to be somewhat more specific than diffuse hypervascularity.

The combination of vascular phase and delayed bone uptake imaging provides several complementary pieces of information about the physiology of the lesion in question. Used together and in concert with data obtained from anatomical imaging, these can help to limit the potential differential diagnoses and to provide more accurate follow-up of therapeutic response or disease progression. For example, a generalised decrease in vascularity despite increased or persisting osteoblastic activity on the delayed imaging phase of the bone scan is more likely to reflect favourable therapeutic response than is the case if there is persisting abnormality on the vascular phases.

### *Metabolic imaging*

Metabolic imaging is often incorrectly considered to be solely the province of PET. To some extent most nuclear medicine investigations are dependent on a metabolic process. The term metabolic imaging is however most pertinent to radiotracers which have active uptake into tumors based on basal metabolic rates or proliferative activity.

Thallium-201 (Tl-201) is a particularly good example of such an agent. As an monocation, it is an analog for potassium and is concentrated in tumor cells by the sodium-potassium ATPase pump (Sehweil et al. 1989) and by a cotransport system mediated by a calcium-dependent ion channel. Enhanced metabolic activity often increases activity of this pump and therefore malignant tumors frequently concentrate this tracer more intensely than normal soft tissues or bone. Increased perfusion also plays a role in the increased uptake of Tl-201 in tumors.

Thallium-201 chloride was first reported as a tumor-imaging radiotracer following serendipitous discovery of uptake in a coincidental lung cancer during myocardial perfusion scanning (Cox et al. 1976). Tl-201 imaging has been evaluated in a range of tumors including lung cancer, glioma, breast carcinoma and thyroid carcinoma. High affinity for Tl-201 has been observed in various bone and soft tissue sarcomas. The Cedars-Sinai group (Ramannah, 1990) studied 38 patients with surgically proven bone and soft tissue sarcomas and found increased uptake in all cases. Similarly, a recent Japanese study demonstrated abnormal Tl-201 uptake in all 30 patients with osteosarcoma at diagnosis (Ohtomo et al. 1996).

It has been shown in several tumors that the intensity of Tl-201 uptake is correlated with proliferative activity, tumor grade and viability (Black, 1989; Kim, 1990; Ando, 1987). In glioma, for example, high Tl-201 concentration is associated with high grade histology and a poor prognosis. Scar tissue has extremely low uptake of Tl-201 while benign lesions also generally have low uptake. Thallium-201 scanning can help to differentiate between benign and malignant bone processes identified by the presence of increased osteoblastic activity and lacking clear features of malignancy on anatomical correlation. Recently, Tl-201 has been shown to be help in demonstrating osteosarcoma involving pagetic bone (Colarinha et al. 1996). This is an important observation since this diagnosis is extremely difficult with other imaging modalities (Figure 2). Thallium-201 imaging may have a role in differentiating between chondromas and chondrosarcoma which can be difficult on structural imaging and even on pathology. Soft tissue tumors including leiomyosarcoma (Shuke et al. 1995) and malignant hemangioendotelioma (Kida et al. 1987) have been identified on Tl-201 imaging.

Since heterogeneity of proliferative activity often exists within tumors, Tl-201 and other metabolic imaging approaches can be helpful to guide the most representative area of tumor. This is important since the behaviour of the tumor, its prognosis and the most appropriate management is usually determined by the highest grade of tumor histology within the lesion.

Increased Tl-201 uptake in a lesion is not however unequivocal evidence of a malignant aetiology. Vari-

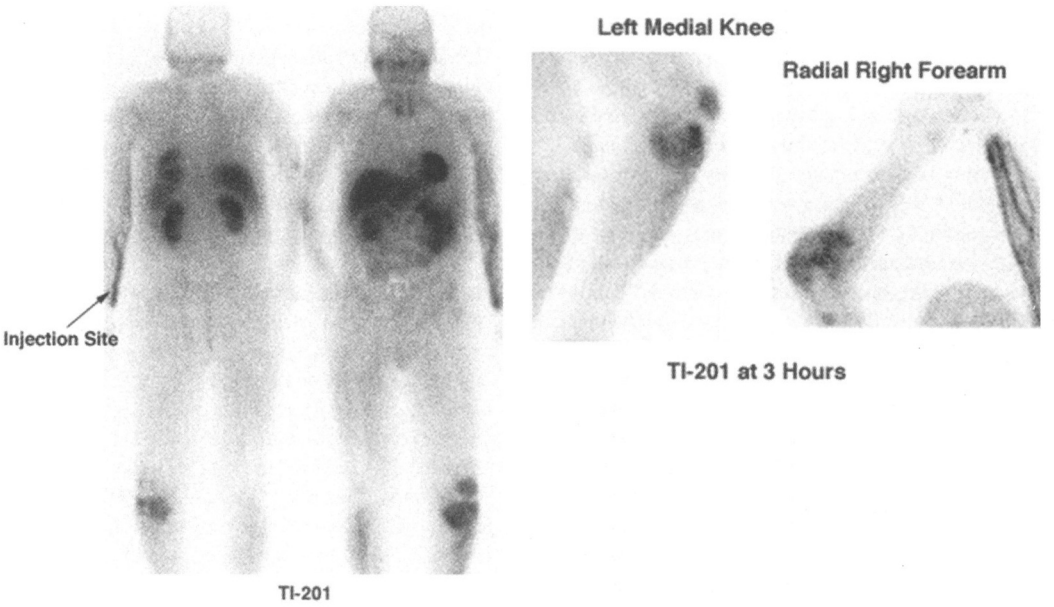


*Figure 2. Thallium-201 imaging demonstrates several focal areas of increased radiotracer uptake in this patient with multifocal osteosarcoma complicating polyostotic Paget's disease.*

ous granulomatous diseases can have increased uptake early, possibly reflecting third space trapping, but retention is usually quite low and therefore delayed imaging may provide more specific identification of malignancy than early scans. Tuberculosis and sarcoidosis of bone are potential causes of high early uptake of Tl-201 and hence either both early and delayed or only delayed imaging is recommended for assessing the likelihood of malignancy in bone lesions with equivocal anatomical features.

The development of new Tc-99m based myocardial blood flow imaging agents with similar biodistributions to Tl-201 has logically led to evaluation of these tracers for tumor imaging. The shorter half-life of Tc-99m (6 hours) than of Tl-201 (73 hours) provides more favourable radiation dosimetry allowing higher administered activity. The higher energy of the gamma photons of Tc-99m and their greater abundance also help to provide superior images. However, diagnostically there appears to be little difference in the uptake of these tracers in sarcomas. Tc-99m sestamibi (MIBI), an isonitrile, has been shown to be actively concentrated in tumor cells with uptake reflecting transmembrane electrical potentials. Owing to the high transmembrane potential of mitochondrial membranes a significant proportion of this radiopharmaceutical is localised within these organelles. Since malignant cells often have increased numbers of mitochondria, enhanced uptake of Tc-99m sestamibi is commonly observed in malignant tumors. Damage to tumor cells following chemotherapy or radiotherapy is usually associated with increased cytosolic calcium levels which, in turn impairs mitochondrial function. Thus, a decrease in Tc-99m MIBI uptake may reflect favourable therapeutic response (Caner et al. 1992). Unfortunately, there is also recent data showing that efflux of this radiopharmaceutical reflects expression of p-glycoprotein, a membrane-associated channel which is involved in exclusion of various chemotherapeutic agents from cancer cells. This is an important mechanism for multi-drug resistance (MDR). Consequently, a decrease in Tc-99m MIBI retention in tumors may reflect either loss of tumor viability or developing MDR. It may be that comparison of Tl-201 or fluorine-18 FDG uptake in such cases could help to differentiate between these possibilities. Tc-99m tetrofosmin is an even newer cardiac imaging agent also actively concentrated in malignant tissues but unlike Tc-99m MIBI, it does not accumulate significantly in mitochondria (Arbab et al. 1996) and therefore may provide different information regarding cell viability following therapy. It is, however, also a substrate for p-glycoprotein and therefore has a potential as a probe for evaluating the likelihood of multidrug resistance (Ballinger et al. 1996).

The use of metabolic tracers imaged with PET is

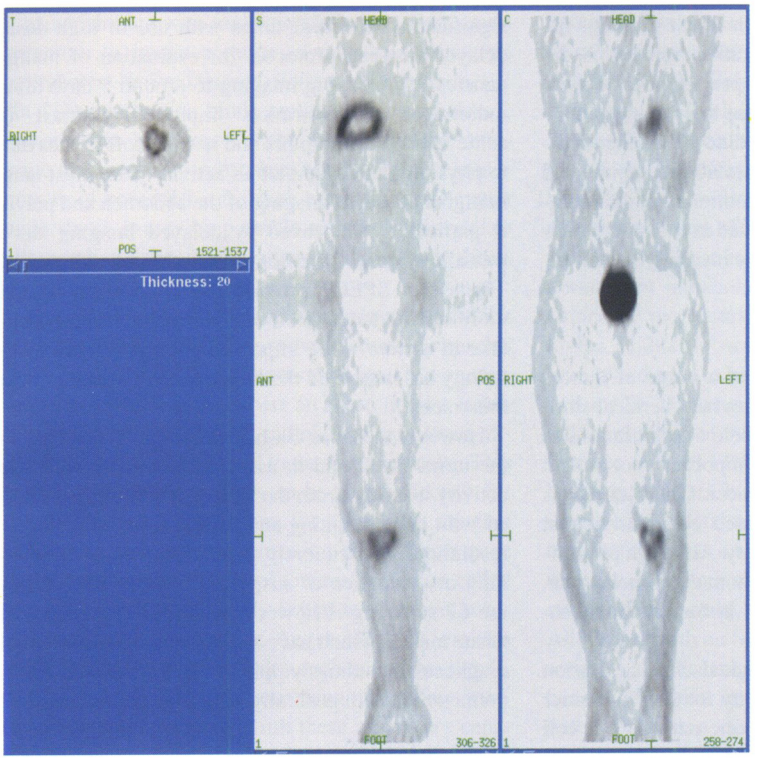


*Figure 3. PET scanning with F-18 FDG demonstrates intensely increased activity at the periphery of a biopsy proven osteosarcoma of the proximal left tibia. Whole body screening demonstrates no evidence of metastatic spread.*

playing an increasing role in oncology (Leskinen-Kallio 1994). The most widely used PET tracer is fluorine-18 fluorodeoxyglucose (F-18 FDG). F-18 FDG is an analog of glucose and is transported into cells by a range of glucose transporter proteins. It is a substrate for the hexokinase enzyme which phosphorylates it into F-18 FDG-6-phosphate. This species is, however, metabolically inert, not being a substrate for other enzymes involved in glycolytic metabolism. Since it has low membrane permeability it accumulates in cells lacking glucose-6-phosphatase. Enhanced metabolic activity in malignant tissues was simplistically considered the mechanism for the empiric observation of increased F-18 FDG uptake in malignant tissues. Although this may well reflect increased glycolytic metabolism in tumor cells there is increasing evidence that it may also be a manifestation of increased expression of glucose transporter proteins, specifically including the Glut 1 and 3 proteins (Yamaoto et al. 1990), on the cell membrane of tumor cells, and of alteration of the activity of the hexokinase enzyme which is the rate-limiting enzyme of FDG trapping in cells. Relative hypoxia in tumors may be the stimulus for upregulation of both GLUT 1 and hexokinase expression (Kubota et al. 1994). F-18 FDG has been shown to concentrate in a wide range of tumors and the intensity of uptake has, like Tl-201, been shown to correlate with histologic grade, proliferative activity (Okada et al. 1992) and prognosis in a range of tumor types (Figure 3).

The ability to quantify regional radiotracer kinetics of positron-emitting radiotracers allows determination of the glucose metabolic rate within tumors. This parameter is a potential means of following therapeutic response with a decrease suggesting beneficial effects. A simpler method involves calculation of lesion to soft tissue activity ratios or of the standardised uptake value (SUV) which relates lesion activity to the administered dose of radioactivity and the patient's body weight or surface area.

F-18 fluoromisonidazole (FMISO) is an imidazole compound which is trapped within hypoxic cells. Since many malignant tumors outgrow their blood supply, cellular hypoxia is common in such tumors. This is important since hypoxia decreases the sensitivity of tissues to radiation and thus lessens the efficacy of radiotherapy. PET scanning with FMISO may provide useful information in selecting the most appropriate form of adjuvant therapy for bone and soft tissue tumors (Koh et al. 1992).

Increased protein synthesis in tumors can be assessed using a range of radiopharmaceuticals based on naturally occurring amino-acids. These can be labeled with single photon agents or with positron emitting radionuclides. The most intensively evaluated agent is carbon-11 methionine, a PET agent (Kubota

et al. 1993). Incorporation of this agent in tumor tissues is correlated with proliferative activity. A decrease in protein synthesis may precede and be more predictive of therapeutic response than changes in F-18 FDG uptake and retention. Iodine-123 alpha-methyl tyrosine represents a new radiolabeled amino acid suitable for imaging with a conventional gamma camera. It has primarily been evaluated in malignant melanoma and its role in bone tumor imaging is unclear.

### *Gallium-67 scanning*

Gallium-67 citrate is taken up in a range of tumors including sarcomas. Since gallium has chemical similarities to iron, iron-binding protein concentrations in sarcomas are thought to be an important mechanism of tumor uptake of this radiotracer. Ga-67 citrate is also a bone tracer with increased concentration at sites of active osteoblastic activity. Although primarily used for the staging of lymphoma and melanoma, gallium scanning has been used in for functional assessment of sarcomas.

Even more specific physiological characterisation is provided by tracers with affinity for specific tumor cell-associated receptors, metabolic pathways or cell surface markers. Monoclonal antibodies have been raised to a number of cell-associated antigens.

Gallium-72 (Ga-72) lactate was initially developed as a bone imaging agent but was serendipitously noted to concentrate also in a range of soft tissue tumors and in infection. The development of Ga-67 citrate with its more favourable, though still suboptimal, imaging characteristics led to use of this agent in the evaluation of suspected and known soft tissue tumors and bone pathology. Although the use of gallium-67 imaging has been proposed for assessment of bone tumors, this agent is now primarily used for the staging and evaluation of residual masses in lymphoma, for staging malignant melanoma and to evaluate suspected infectious or inflammatory processes.

Gallium has chemical similarities with iron. The mechanism of retention of Ga-67 in malignant tissues is unclear but it has been suggested with some justification that it relates to increased expression of transferrin receptor on the surface of malignant tumors. The concentration of iron-binding proteins with tumors may also play a role in retention of gallium-67 within malignant lesions. Uptake of this radiotracer in relationship to osteoblastic activity leads to similar problems with specificity observed with bone scanning. Osteomyelitis and heterotrophic ossification are examples of benign conditions which can have intense Ga-67 uptake and retention.

Techniques for gallium-67 imaging have improved significantly in recent times with use of high-dose, delayed imaging protocols for evaluation of malignancies. By delaying imaging to around 5 days after radiotracer administration improved contrast is achieved between tumors and normal soft tissues due to physiological clearance of activity from most non-malignant tissues. Imaging of the abdomen and pelvis is particularly improved by delayed imaging since this allows time for colonic excretion of radiotracer. The use of SPECT provides better localisation and more sensitive detection of abnormal gallium-67 uptake in tumors while improved gamma camera technology has improved the image quality obtained with this tracer.

However, because Ga-67 uptake can occur both in the tumor itself and in association with osteoblastic activity it suffers from the lack of specificity associated with bone scanning and suffers from even poorer resolution. Consequently, I seldom use Ga-67 scanning in evaluation of suspected bone tumors. Gallium-67 scanning is however useful in assessing soft tissue masses which have a wider range of differential diagnoses including lymphoma and metastatic melanoma which both generally have high gallium avidity.

### *New radiopharmaceutical approaches to diagnosis of soft tissue and bone tumors*

Advances in the understanding of the molecular biology of tumor cells coupled with improved methods for radiolabeling of biological compounds will increasingly allow development of specific radiotracers targeting the unique characteristics of tumor cells.

Monoclonal antibodies to cell surface antigens on tumor cells are an example of this type of approach. The anti-3F8 monoclonal antibody directly against gangliosides has affinity for a range of sarcomas including malignant fibrous histiocytoma. The relatively large molecular size of monoclonal antibodies however limits tissue penetration and therefore contrast between blood and tumor activity. The fact that most monoclonal antibodies are currently produced using murine hybridoma technology also limits their use due to the development of human anti-mouse antibody (HAMA) responses. New techniques for humanised antibody production and development of small variable domain fragments will hopefully improve diagnostic and therapeutic efficacy.

Peptides which act as receptor ligands and cell messenger proteins offer attractive prospects for tumor imaging if radiolabeling can be performed without significantly altering structural characteristics of the active binding site. The prototypic example of this approach is the use of radiolabeled somatostatin ana-

logs which bind with high specificity to somatostatin receptors to image a range of neuroendocrine and neural crest-derived tissues. For example, indium-111 (In-111) pentreotide binds particularly to subclass 2 somatostatin receptors. As the growth factors, autocrine and paracrine substances which regulate tumor growth of bone and soft tissue tumors become better understood, it may be possible to radiolabel these to allow radiodiagnosis and targeted radionuclide therapy.

Oligonucleotides including antisense codons which form triplexes within the groove of double-stranded DNA potentially allow detection of specific genetic sequences within cells. Where an aberrant gene is implicated in tumorigenesis a radiolabeled antisense oligonucleotide could be used to target tumor cells. This approach is however likely to be more effective for therapeutic intervention than for diagnostic imaging due to the very low concentration of the target gene within tumor cells. A type of ionising radiation called an Auger electron is ideally suited to therapeutic application of this type. The Auger electrons emitted from a range of radionuclides such as iodine-125 (I-125) travel only a few microns from where they arise. Oligonucleotides labeled with these agents are capable of inducing lethal double-stranded DNA breaks at specific sites in the chromosome.

## *Clinical applications of nuclear medicine techniques to bone and soft tissue tumor imaging*

### Tumor staging and grading

Bone scanning is a useful technique to stage primary bone and soft tissue sarcomas since it allows whole body screening for sites of metastatic bone disease as well as assessing the extent of local bony involvement. In osteosarcoma soft tissue metastases may also be evident by virtue of malignant osteoid formation. Thallium-201 can also be used to stage sarcomas. Evaluation of the abdominal region is however compromised by the presence of normal uptake in abdominal organs and the intestines.

A recent comparison of bone scanning with Tl-201 chloride scanning demonstrated slightly higher specificity and overall accuracy of Tl-201 than delayed bone uptake images (Caluser et al. 1994). A total of 40 paired scans were verified by biopsy and a further 7 paired scans by long-term follow-up. Although the sensitivity of bone scanning was similar to Tl-201 at around 88%, the specificity was substantially lower (38% versus 69%). Incorporating evaluation of the vascular phases improved diagnostic performance of the bone scan to values similar to those on Tl-201 imaging. The methodology used for Tl-201 was however suboptimal since imaging was performed only 10 minutes after radiotracer injection. The use of only early imaging likely accounts for the similarity of Tl-201 results with blood pool imaging on bone scan and the relatively poor specificity of Tl-201 in this particular study. As outlined above there is evidence that delayed retention of Tl-201 is a better predictor of viable tumor than is early activity. Indication of the importance of metabolic activity was however demonstrated by the observation that all 12 patients with a higher Tl-201 to background activity ratio than bone scan blood pool to background ratio had sarcoma.

Both Tl-201 imaging and F-18 FDG PET may provide evaluation of the grade of a range of tumors. The first use of F-18 FDG PET for tumor grading was in brain tumors (Di Chiro 1986) PET has been evaluated in musculoskeletal tumors (Adler et al. 1991). In 25 tumors there was a high correlation (Rho = 0.83) between a quantitative measure of FDG retention (the standardised uptake value or SUV) and NCI grade. All tumors with an SUV >1.6 were high grade in this series. A subsequent smaller study (Jones et al. 1996) has found similar concordance between tumor grade and SUV with a cut-off of 1.6 stratifying high and low-grade tumors.

An early study from Japan suggested that the ratio of Tl-201 to gallium-67 uptake in tumors may reflect the proliferative activity of tumors those lesions with higher Ga-67 than Tl-201 having a higher doubling rate than tumors with the converse pattern (Togawa et al. 1986). Although this study did not specifically evaluate sarcomas, the principle of using more than one physiologic tracer to characterise different aspects of tumor biology is an attractive one and may increase the specificity of diagnosis.

## *Differentiation of scar from residual or recurrent tumor*

Following surgery, radiotherapy and, less often with chemotherapy there may be scarring in the region of the tumor which may distort normal anatomical relations and cross normal tissue planes. Further, scar may co-exist with tumor tissue. While tumor tissue will generally demonstrate contrast enhancement on CT or MRI scanning, this is not uncommonly seen also with organising scar tissue. There are number of fundamental differences between scar tissue and tumor. Low metabolic activity and cellularity is seen in chronic scar tissue whereas most malignant tumors have high cellularity and metabolic activity.

Tl-201 imaging has been evaluated for detection of suspected residual or recurrent bone and soft tissue

sarcomas at the Memorial Sloan Kettering (Kostakoglu et al. 1995). In this series, 29 sarcomas were evaluated using Tl-201 and a conventional imaging modality (MRI in 17, CT in 6 and contrast angiography in 6). Tl-201 detected all 21 histologically-proven tumors while there was 1 false negative on conventional restaging. More importantly, Tl-201 was negative in 7/8 patients without recurrence and equivocal in the remaining patient while conventional imaging was false positive in 4/8 including 3 cases with MRI.

The overall accuracy of Tl-201 scintigraphy was 97% versus 83% for conventional approaches.

In a series of 24 patients with suspected recurrent tumor, including 9 soft tissue tumor primaries and 3 bone tumors, F-18 FDG and C-11 methionine PET were compared as markers of metabolically active, residual tumor. Overall sensitivity was similar for both tracers but slightly higher with F-18 FDG than with C-11 methionine (Inoue et al. 1996). The intensity of uptake was also greater on the F-18 FDG studies. The results were poor for lesions less than 1.5 cm in diameter but this likely reflects the relatively low spatial resolution of the scanner used for this study.

### *Therapeutic monitoring*

Because uptake of a range of radiotracers into malignant tumors reflects the metabolic and proliferative activity of the cells within them, these radiotracers can be potentially used to monitor the response of tumors to therapy. The most widely used single photon agent for this application is Tl-201 while PET using F-18 FDG or C-11 methionine is being increasingly widely used (Jansson et al. 1995).

The percent tumor necrosis found histologically following chemotherapy has been shown to be a predictor of outcome in osteosarcoma in particular. Prognosis is substantially better if the percentage tumor necrosis is >90%. Although its role in other tumors is less clear, Tl-201 uptake has been shown to correlate with this parameter in soft tissue sarcomas including rhabdomyosarcoma (Maini et al. 1994). Ga-67 scanning and Tc-99m MDP bone scanning were found to be less accurate in assessing therapeutic response. In particular, 4/8 patients with >95% tumor necrosis had unchanged or increased Ga-67 uptake. Similar results were obtained by a more recent study of 30 patients with osteosarcoma treated with chemotherapy (Ohtomo et al. 1996).

There are substantial data on the accuracy of PET for therapeutic monitoring in a range of tumors. Decreased uptake of F-18 FDG (Haberkorn et al. 1991; Wahl et al. 1993) and C-11 methionine (Huovinen et al. 1993) have both been shown to be accurate markers of therapeutic response. An early increase in F-18 FDG uptake may however occur following radiotherapy and therapeutic monitoring in this setting may be more accurate with C-11 methionine. C-11 methionine is a marker of protein synthesis. An influx of inflammatory cells into the radiation field is the most likely explanation for the local increase or preservation of FDG retention despite lack of residual viable tumor in some cases. This phenomenon seems much less common following chemotherapy with an early reduction in F-18 FDG being the norm after successful therapy. Results with combined chemoradiotherapy seem to be similar to those with chemotherapy alone.

Impairment of cellular viability preceding cell death can be detected by a range of radiotracers, Tc-99m MIBI is reduced in prenecrotic cells with impaired mitochondrial function. Although changes in F-18 FDG, Tl-201 and Tc-99m MIBI uptake all appear to some degree reflect therapeutic response, the cellular processes which they trace clearly differ and they may each provide differing information regarding the effect of chemotherapy on cellular metabolism and viability (Slosman and Pugin. 1994). A recent study (Garcia et al. 1996) comparing PET with F-18 FDG and Tc-99m MIBI SPECT demonstrated both higher sensitivity and specificity for PET (98% versus 82% and 90% versus 80%, respectively). The visual grade for confirmed recurrent tumors was also higher for PET than Tc-99m MIBI (2.1 versus 1.6). Interestingly, 4 of 9 patients with F-18 FDG but not Tc-99m MIBI uptake failed to respond to multidrug therapy. These data may reflect the ability of Tc-99m MIBI to demonstrate p-glycoprotein expression and to thereby predict for multidrug resistance.

### *Potential therapeutic applications*

The goal of highly selective binding of radiopharmaceuticals to tumor cells relative to normal cells is as important, or more important, to therapeutic applications of radionuclides than it is to diagnostic applications. High target (tumor) to background (non-tumor) radioactivity ratios potentially allow administration of targeted radiotherapy directly to tumor deposits with low toxicity to the remainder of the body.

The high extraction of bone-seeking radiopharmaceuticals by various sarcomas offers the potential for delivering high local radiation doses to these tumors if radionuclides with particulate emissions are used. Agents such as strontium-89 and samarium-153 EDTMP (Figure 4) have been recently introduced into clinical practice for the palliation of pain related to osteoblastic bone tumors. Radiolabeled monoclonal

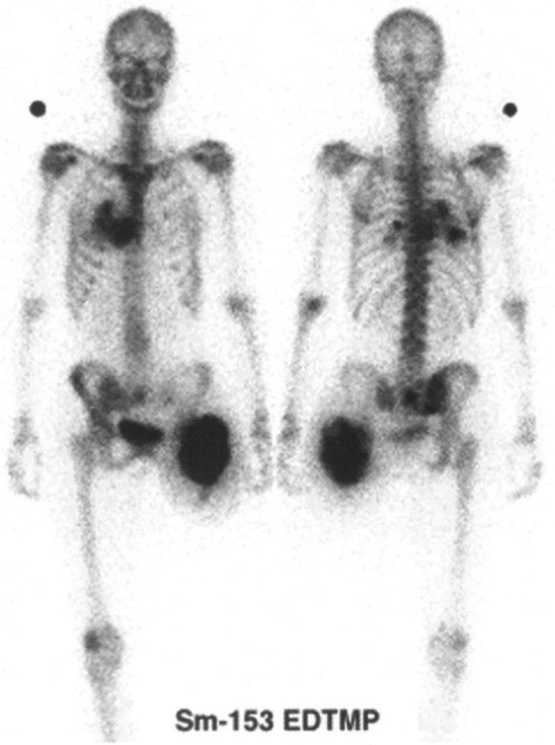


*Figure 4. Samarium-153 EDTMP is a bone seeking radiopharmaceutical with both gamma and beta emissions allowing imaging of this agent when administered for palliative therapy of bone pain related to actively osteoblastic lesions such as osteosarcoma. The simularity of this scan following a radionuclide therapy to the bone scan on the same patient (Figure 2) is clearly apparent.*

antibodies provide a potential means of specifically targeting the tumor cells. Access of these antibodies to tumor antigens may be limited by the large size of monoclonal antibody molecules and attempts to improve tissue penetration including use of variable domain fragments may improve therapeutic efficacy.

Approaches to improving therapeutic efficacy of radionuclides are possible. These potentially include; use of radiosensitising chemotherapy in combination with these systemic radionuclide administration, the use of regional hyperthermia to increase radiopharmaceutical delivery to the tumour, use of a combination of radiopharmaceuticals with differing physical characteristics to optimise tissue penetration and radiation dose rates, and adoption of supportive strategies now routinely used in combination with chemotherapy and external beam radiotherapy in order to allow high administered activities in situations where non-target tissue toxicity is dose limiting.

## *Summary*

The cellular derangement which produces a malignant tumor most likely begins with altered cell cycle regulation related to genetic damage. This in turn will often alter cellular metabolic function and ultrastructural characteristics including enzymatic, cytosolic and cell-surface protein expression. These changes alter biochemical characteristics of the tumor cell which affect tissue physiology. It is only when the volume of abnormal cells becomes large enough to be detected by an imaging technique, direct visualisation or palpation, that a mass lesion becomes evident. Although this is a very simplistic account of tumorigenesis, it makes the point that anatomical imaging can only be expected to detect the more advanced stages of malignancy while nuclear medicine techniques, which are ideally suited to non-invasively evaluating the physiological, biochemical and genetic features of tumors ought to have an important role in the early detection and treatment of bone and soft tissue tumors.

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