immunoscintigraphy. Tricot et al. (*26*) described the dislocation of immature granulopoietic cells during myelodysplastic syndrome, including blasts in the bone marrow, as an abnormal localization of immature precursors (ALIP). They also reported that ALIP is an adverse prognostic finding that is very accurate in predicting which patients will have a short survival time and which will have early transformation to acute leukemia (*26,27*). We expect that the immunoscintigraphic technique described here can provide useful information to diagnose and classify this complex syndrome.

In the myelogenous leukemia patients, myelocytes expressed NCA-95 to a variable degree, and normal or increased uptake of the anti-NCA-95 antibody was observed in all four patients. Of course, because of the lack of lymphoid cell binding sites, there was markedly decreased uptake in acute lymphocytic leukemia subjects.

In patients with other disorders, such as iron deficiency anemia, pure red cell aplasia, thalassemia minor and Evan's syndrome, immunoscintigraphy revealed bone marrow expansion. Peripheral bone marrow expansion can also be evaluated with bone-seeking radiopharmaceuticals (*1*). Findings of marrow expansion in ^{99m}Tc-labeled phosphate bone scans are not specific or clear. In comparison, the immunoscintigraphy protocol described in this article provides more precise images and can be used to evaluate bone marrow functional states.

CONCLUSION

Until now, immunoscintigraphy with antigranulocyte antibody has been used to detect bone marrow metastasis and to localize inflammation (*10,11,28,29*). This preliminary report suggests that it may also be effectively used to evaluate the functional status of bone marrow during disease and treatment.

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EDITORIAL

Noninvasive Evaluation of the Bone Marrow

The study reported by Chung et al. (*1*) in this issue reflects the evolution of sophisticated, noninvasive technology for the evaluation of bone marrow anatomy, function and disease. Such methods offer the major advantage of visualizing the entire marrow, or at least a major portion of the bone marrow, as compared to the small sampling obtained by bone marrow aspiration and biopsy. Each of these new technologies, however, has its own shortcomings that limit it as a sole method of studying the marrow. Such methods alone are not likely to replace bone marrow aspiration or biopsy and pathologic review. In combination, however, they may offer complementary information to the aspirate and biopsy and to each other, and help to improve patient management. There are two major methods of noninvasively studying the

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bone marrow: (a) MRI and spectroscopy and (b) radiopharmaceuticals.

Nuclear magnetic resonance (NMR) imaging utilizes the difference in proton relaxation properties (T1 and T2 relaxation times) of fat and water (cells) to create a map of the cellularity of the bone marrow. It thus provides an assessment of its *anatomy* (*2*). Patterns of bone marrow disorders that can be evaluated by MRI include recruitment of fatty marrow by increased demand for hematopoiesis, marrow infiltration or replacement with leukemia, metastases, etc. and bone marrow failure (iatrogenic or spontaneous) resulting in loss of cellularity and increase in fat. Some investigators (*3*) have even suggested that particular patterns of cellular distribution in hypocellular marrows (homogenous versus heterogeneous) may be helpful in distinguishing, for example, aplastic anemia from hypoplastic myelodysplastic syndrome. MRI offers the advantage of being sensitive, though nonspecific for pathology, and it can assess a much larger sample than can be studied by the usual invasive techniques. MRI has become the technique of choice for imaging selected regions of the bone marrow.

NMR spectroscopy can be used as an adjunct to MRI to obtain information, noninvasively, about the intracellular metabolites of a biologic sample. Although it has not been well utilized in studies of the bone marrow, and is still considered investigational, it can potentially provide information about the *biochemistry* and *physiology* of the marrow. Such spectroscopic studies have been performed in vivo on the brain and breast and in vitro using leukemic cell lines to determine differences in normal and malignant tissue, as well as between mature and immature cells, respectively (*4*). Jansen et al. (*5*) even reported differences in T1 relaxation times of the water spectrum in vivo for leukemic versus normal marrow. Both the imaging and spectroscopic techniques require specialized equipment and training.

Radionuclide imaging of the bone marrow targets specific cellular components of the normal bone marrow. It can be divided into three categories: (a) erythroid, (b) macrophage/histiocytic (RES) and (c) myeloid. The earliest studies of bone marrow utilized ^{52}Fe, a positron-emitting isotope, and ^{59}Fe, a long-lived, high-energy photon emitter. Both are incorporated into the erythroid precursors. Their utility has been limited by the equipment required to generate or analyze the isotope, the cost, and, in the case of ^{59}Fe, the radiation exposure to the marrow. Although PET is becoming more widely available, production of the appropriate isotope is still difficult. The RES has been visualized previously by ^{198}Au and more recently with ^{99m}Tc-colloid. The colloid particles attach to the membrane of the cell, are then endocytosed and degraded by the lysosomes within the cell. Because these particles are distributed predominantly to the Kupffer cells of the liver and the phagocytic cells of the spleen and in much smaller amounts to the RES of the marrow, uptake in the marrow is relatively reduced. In addition, visualization of the lumbar and thoracic spine is impaired because of the high hepatosplenic uptake. Anatomic patterns change significantly with underlying systemic hematologic conditions, neoplastic hematologic disorders and tumor infiltration (*6*). The development of ^{99m}Tc-labeled nanocolloid has resulted in higher concentrations of particles localizing to the bone marrow compared to the conventional colloids, but limitations to visualization are similar.

Technetium-99m-labeled monoclonal antibodies, such as the NCA-95 antibody described by Chung et al., have been developed recently to target the myeloid elements of the bone marrow and peripheral blood. The NCA-95 antibody, under clinical investigation, binds to the target cells by interacting with the nonspecific cross-reacting antigen 95 (NCA-95) (*7*). This antigen is expressed on the membrane of granulocytes and other myeloid precursors. Other myeloid antibodies such as M195, p67 and a genetically engineered humanized version, HuM195 (all directed against the CD33 antigen), bind to myeloblasts and early myeloid precursors, but not to mature granulocytes. The anti-CD33 antibodies rapidly target leukemia cells throughout the marrow, liver and spleen (*8,9*); when labeled with ^{131}I these antibodies can be used to characterize biodistribution of leukemia cells and to quantitate target cells (*10*). When labeled with modest amounts of ^{131}I (80–140mCi), this targeting may reduce residual leukemia cells in patients in clinical remissions. Dosimetry modeling has revealed significant heterogeneity of uptake within the marrow (*11*). When labeled with larger amounts of ^{131}I (200–360mCi), the entire marrow, normal and neoplastic cells as well, can be ablated (*8,12*). Such cytoreduction may be applicable to pre-bone marrow transplantation preparation.

A major advantage to using the antibody as a carrier for the ^{99m}Tc is its specificity for hematopoietic elements. Like MRI, classic scintigraphy, using radioactive nuclides or nuclides attached to carriers, antibody targeting is most useful in providing information about the *anatomy* of the marrow. Its limitations lie in the distribution of the antigen to which the antibody has been developed and its value will most likely be as a tool for monitoring a disease or localizing a site for an invasive procedure. For example, marrow replaced by acute myelogenous leukemic may appear "falsely" hypocellular if the radiolabeled antibody used is expressed only on maturing myeloid cells; a hypoplastic bone marrow due to aplastic anemia may appear the same as that due to a hypoplastic myelodysplastic syndrome, or a hypoplastic phase preceding the development of full blown hairy cell leukemia or acute lymphocytic leukemia. On the other hand, an antibody targeting myeloblasts may prove useful in following patients in remission and in diagnosing early relapse. Conversely, bone marrow involvement by solid tumors allows detection by demonstrating photopenic areas.

Finally, PET, which also relies on selective accumulation of administered radiolabeled materials, has been developed to study functional and metabolic changes in normal and abnormal tissue. Because of the physics of positron emission and detection, its performance in diagnosing malignancy has been found superior to that of CT and MRI, with sensitivity *and* specificity greater than 90% in the detection of some tumors. Like NMR spectroscopy, it focuses on differences in the biochemistry and metabolism of normal and diseased tissue (*13,14*). Dahlbom et al. (*15*) have recently developed a whole-body PET method that permits evaluation of the entire body, and have opened the door to broader applications in hematology and assessment of bone marrow. Its application, unfortunately, has been limited by cost and the availability of equipment and labeled radiotracers.

Noninvasive imaging and spectroscopic techniques for the marrow are still in their infancy. Therefore, carefully conducted clinical studies that include correlation between the results of imaging and the reference standard—bone marrow aspiration and biopsy—are critical to the definition of their utility in hematology and oncology. As these noninvasive methods become more sophisticated, as they are used in combination and as

larger databases are developed, the need for the invasive procedures may lessen.

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FDG-PET to Evaluate Response to Hyperthermic Isolated Limb Perfusion for Locally Advanced Soft-Tissue Sarcoma

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We investigated FDG-PET in patients undergoing hyperthermic isolated limb perfusion (HILP) with rTNF-α, rIFN-γ and melphalan for locally advanced soft-tissue sarcoma of the extremities. **Methods:** Twenty patients (11 women, 9 men; aged 18–80 yr, mean age 49 yr) were studied. FDG-PET studies were performed before, 2 and 8 wk after HILP. After the final PET study, the tumor was resected and pathologically graded. Patients with pathologically complete response (pCR) showed no viable tumor after treatment. Those with pathologically partial response (pPR) showed various amounts of viable tumor in the resected specimens. **Results:** Seven patients showed a pCR (35%) and 12 patients showed a pPR (60%). In one patient, pathological examination was not performed (5%). The pre-perfusion glucose consumption in the pCR group was significantly higher than in the pPR group ($p < 0.05$). Visual analysis of the PET images after perfusion showed a rim of increased FDG uptake around a core of absent FDG uptake in 12 patients. The rim signal contained a fibrous pseudocapsule with inflammatory tissue in the pCR group, viable tumor was seen in the pPR group. The glucose consumption in the pCR group at 2 and 8 wk after perfusion had decreased significantly ($p < 0.05$) in comparison to the glucose consumption in the pPR. **Conclusion:** Based on the pretreatment glucose consumption in soft-tissue sarcomas, one could predict the probability of a patient achieving complete pathological response after HILP. FDG-PET indicated the pathologic tumor response to HILP, although the lack of specificity of FDG, in terms of differentiation between an inflammatory response and viable tumor tissue, hampered the discrimination between pCR and pPR.

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Malignant soft-tissue sarcomas are a heterogeneous group of lesions that all arise from tissue of mesenchymal origin and are characterized by aggressive local growth and hematogenic metastases. They account for 1% of all malignant tumors and have an incidence rate of 2 per 100,000. About 60% of these tumors occur in the extremities and are often quite large at diagnosis (*1*). Limb-saving treatment of extremity soft-tissue sarcomas is a multidisciplinary matter, with surgery and radiotherapy as the usual treatment protocol (*2,3*). This combination therapy has avoided ablative surgical procedures in the majority of patients.

The majority of locally advanced extremity soft-tissue sarcomas are treated by amputation. Intra-arterial chemotherapy with adriamycin, combined with preoperative radiotherapy, surgery and postoperative radiotherapy is effective in the treatment of locally advanced soft-tissue sarcoma, but significant morbidity does occur (*4*). Recently Eilber et al. (*5*) reported a complete response rate of 49% and a limb-saving rate of 98% with neo-adjuvant chemotherapy and radiation for high-grade extremity soft-tissue sarcoma with low-treatment morbidity. Hyperthermic isolated limb perfusion (HILP) also proved to be of value in the treatment for locally advanced extremity soft-tissue sarcoma (*6–8*). With HILP, chemotherapeutic tissue concentrations may be up to 20 times higher than can be attained with systemic administration (*9*). The intro-