Fine-needle aspiration is an integral component of the team approach to managing bone and soft-tissue sarcomas.

Adrienne Anderson. *Giardino: Farfalla,* 1997. Mixed media on linen, 24″ × 72″.

Fine-Needle Aspiration Biopsy of Sarcomas and Related Tumors

William G. Ward, MD, Paul Savage, MD, Carol A. Boles, MD, and Scott E. Kilpatrick, MD

Background: *Largely due to a lack of experience, familiarity, and/or confidence, few centers rely on simple fine-needle aspiration biopsy (FNAB) for the diagnosis of sarcomas and related tumors.*

Methods: *The authors have reviewed their own experience in more than 200 cases of FNAB of bone and soft-tissue tumors, as well as cases reported in the literature.*

Results: *FNAB has proven to be accurate and useful in 8 consecutive years of clinical experience. No serious complications have occurred.*

Conclusions: *FNAB is recommended as an integral part of the initial evaluation of amenable orthopaedic tumors, including sarcomas, especially in cases with classic clinical and radiographic findings.*

Introduction

Fine-needle aspiration biopsy (FNAB) of previously undiagnosed masses or suspected orthopaedic tumors has multiple advantages compared to open biopsy.[1-7] It is quick, inexpensive, and minimally invasive, and it can often be performed on the day of the initial office visit. It has been shown to establish the diagnosis of osteosarcomas at less than one fourth the cost of open biopsy.[5] FNAB can generally be performed without any anesthesia, it causes minimal to no discomfort, and it is well tolerated by patients. FNAB is performed using a 23- or 25-gauge needle. When offered a choice of open vs needle biopsy, most patients prefer the simpler needle biopsy. Diagnostic aspiration biopsies obviate the need for open biopsies.

FNAB is an integral component of an overall team approach to the diagnosis and treatment of bone and soft-tissue tumors. Our team includes an orthopaedic oncologist, a medical oncologist, radiologists, and pathologists. The pathologists must be skillful in the cytopathologic interpretation of orthopaedic tumor aspirates. Due in part to both the shortage of cytopathologists with experience in orthopaedic tumors and the lack of exposure of orthopaedic oncologists to the FNAB procedure, the technique is utilized at only a few orthopaedic tumor centers throughout the United States. However, its use is increasing as experience is gained with the technique. It allows operating room time to be more appropriately utilized for tumor resections rather than unnecessary open biopsy procedures. FNAB is perhaps most accepted for the diagnosis of locally recurrent and metastatic

From the Departments of Orthopaedic Surgery (WGW), Hematology/Oncology (WGW, PS), Diagnostic Radiology (CAB), and Pathology (WGW, SEK) at the Wake Forest University Baptist Medical Center, Winston-Salem, NC, and the Department of Pathology and Laboratory Medicine (SEK), University of North Carolina Hospitals, Chapel Hill, NC.

Address reprint requests to William G. Ward, MD, Professor, Department of Orthopaedic Surgery, Wake Forest University Baptist Medical Center, Medical Center Boulevard, Winston-Salem, NC 27157-1070.

No significant relationship exists between the authors and the companies/organizations whose products or services may be referenced in this article.

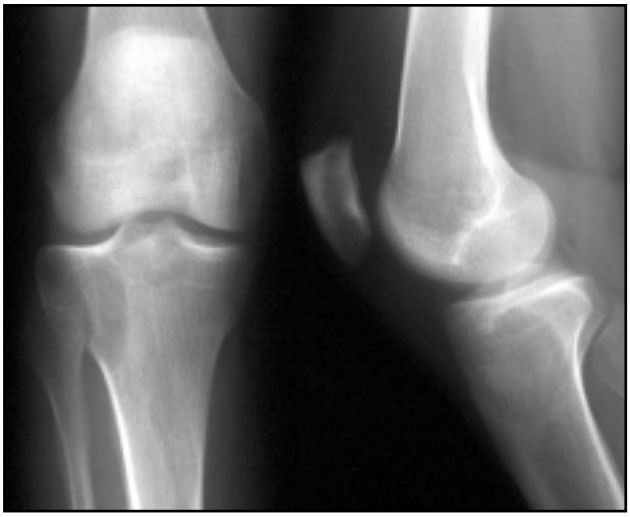


Fig 1A. — AP and lateral radiograph of a classic giant cell tumor. (Figs 1A-B used by permission of W. G. Ward, MD.)

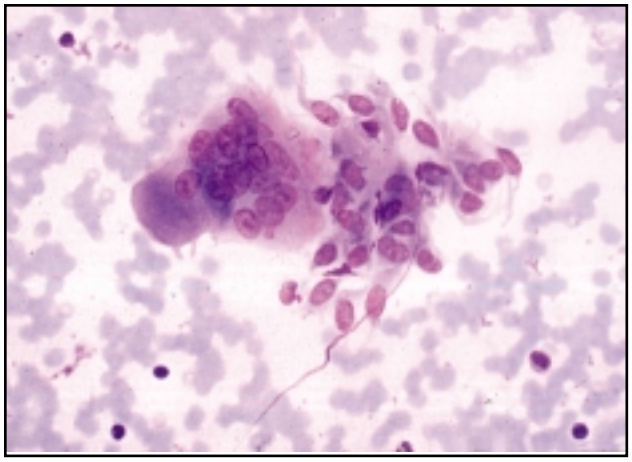


Fig 1B. — FNAB aspirate of giant cell tumor.

lesions in which a diagnosis of sarcoma has been previously established. It is gaining increasing acceptance for the diagnosis of primary tumors of bone and soft tissues, both benign and malignant.

We initially began utilizing FNAB on tumors that had a classic radiographic appearance such as high-grade osteosarcoma. Its role was that of a confirmatory test. We have expanded our use of FNAB to include most previously undiagnosed orthopaedic masses and tumors. FNAB is most successfully utilized when it is simply confirming the clinicoradiographic diagnosis in a patient whose tumor has classic presentation. For example, a patient in the 20- to 40-year-old age group with a distal femoral or proximal tibial lytic, eccentric, geographic, subchondral epiphyseal lesion almost certainly has a giant cell tumor (Fig 1A). FNAB is excellent at confirming the diagnosis of giant cell tumor of bone (Fig 1B). Appropriate laboratory tests may be required to rule out associated hyperparathyroidism, but FNAB provides great reliability in diagnosing giant cell tumors.[5] This allows surgery to proceed expeditiously with a confirmed preoperative diagnosis and avoids the need to wait for the pathologists to process the frozen section intraoperatively. Alternatively, should an osteosarcoma be diagnosed at FNAB, then the appropriate systemic evaluation and chemotherapy can be initiated quickly, without having to schedule and perform an open procedure.

FNAB is successful in the diagnosis of bone malignancies. Previous studies have shown it to be highly accurate in diagnosing osteosarcoma, myeloma, and Ewing's sarcoma as well as other bony sarcomas. In our experience, FNAB can correctly identify bony sarcomas in 93% of cases in which adequate aspiration specimens are obtained. Histogenetic subtyping can be achieved in approximately 82% of these same cases.[7] Few centers would be willing to treat or consider treating a patient with a high-grade osteosarcoma, even with a classic radiographic and clinical presentation, without histologic confirmation. FNAB is well suited to provide histologic confirmatory tests in such situations, allowing chemotherapy to be initiated within 2-3 days of the initial office visit. Reliance on FNAB for diagnosis avoids an open biopsy with its potential systemic seeding, local tumor spread, and tissue tumor contamination. Although complications from local tumor spread are avoidable with proper technique in many open biopsies, some tumor spread and tissue contamination will of necessity occur in even the most skillful hands. The hazards of open biopsy have been well documented, especially when performed with suboptimal technique.[8]

FNAB is also useful in the management and evaluation of both benign and malignant soft-tissue tumors. Inappropriate or incomplete surgical excision of presumed benign lesions ultimately found to be malignant may be avoided by the judicious use of FNAB. The management of benign lesions can be simplified following a diagnostic FNAB. For example, in several cases, unsuspected large gouty tophi that were mimicking sarcomas were readily diagnosed by FNAB.[3] Similarly, unsuspected abscesses can be identified with FNAB, obviating the need for a more expensive workup and allowing initiation of appropriate treatment. The diagnosis of certain benign lesions, such as ganglions and popliteal cysts, can be cytologically confirmed. When the FNAB findings are considered in conjunction with classic radiographic findings, many benign entities can safely be clinically observed. The physician gains a greater sense of confidence that a more worrisome lesion, such as a low-grade myxoid sarcoma, has not been overlooked following a diagnostic FNAB. However, an FNAB that is "negative for malignancy" or that "contains no malignant cells" does not absolutely confirm the absence of malignancy. It

simply means that no identifiable malignant cells were aspirated and expelled onto the glass slide. This could be due to nonrepresentative sampling, to the needle having missed the lesion altogether, or to the absence of a malignant entity. Therefore, a "negative" FNAB should be accepted only as one component of an overall diagnostic picture. Particularly with deeper lesions, imaging-guided FNAB can avoid a false-negative finding due to the needle missing the lesion. If the cytologic findings and interpretations are not consistent with the clinical and radiographic findings and a malignancy is suspected, then open biopsy or core needle biopsy is indicated.

Malignant soft-tissue lesions are likely to be appropriately identified with FNAB. They often can be differentiated into histologic subtypes such as myxoid liposarcoma, extraskeletal mesenchymal chondrosarcoma, and malignant fibrous histiocytoma, but many can be identified only as a pleomorphic sarcoma. This may not be problematic because such an interpretation essentially equates to high-grade sarcoma, which may be sufficiently diagnostic for current treatment regimens. In the setting of an adequate FNAB aspiration specimen, approximately 86% of soft-tissue sarcomas can be correctly identified as sarcomas by FNAB, and approximately 54% can be properly classified into their histologic subtype.[7]

The success of FNAB is somewhat dependent on the diagnostic specificity required by the individual oncology team. At our institution, we treat large, high-grade soft-tissue sarcomas with preoperative chemotherapy followed by wide resection, additional chemotherapy, and ultimately radiation therapy. The postoperative chemotherapy regimen is partly dependent on the tumor's response to the preoperative chemotherapy. This is determined by both the histologic evaluation of the resected specimen and the use of other clinical parameters of tumor response such as shrinkage or necrosis detected clinically and documented by magnetic resonance imaging (MRI).

Our general approach to low-grade malignant lesions is to do a wide resection without any preoperative therapy. It could be argued that at our institution, essentially all malignant orthopaedic soft-tissue tumors are ultimately treated with wide resections. Thus, although the FNAB diagnosis of malignancy is established preoperatively, most soft-tissue tumors will warrant a wide resection. However, the use of FNAB allows selection of patients with high-grade malignancies for preoperative chemotherapy.

FNAB is generally not utilized to differentiate lipoma from low-grade lipoma-like liposarcoma. This differentiation can be difficult on standard histology. Unless the lesion dedifferentiates, the primary clinical concern with well-differentiated lipoma-like liposarcoma is only local recurrence. We treat both lipomatous lesions (lipoma and lipoma-like liposarcoma) essentially the same, with an extracapsular wide resection. Since extensive sampling is often required to differentiate between the two entities and since both are best treated with complete resection, we wait on final pathology to differentiate these lesions. We have been able to correctly predict the pathologic diagnosis preoperatively in most cases by careful analysis of the MRI or computed tomography (CT) images. If there is internal signal streaking or inhomogeneity, lipoma-like liposarcoma is suspected. If a pure homogeneous, fat-appearing image is present, lipoma is suspected, and we resect less margin tissue at surgery, opting for a true capsular resection. Thus far, we have not encountered a problem with this approach.

FNAB Surgical Technique

We routinely perform FNAB on appropriate bone and soft-tissue lesions in both adults and children. The procedure is most often performed in the outpatient setting, but it is occasionally performed on inpatients as well. A mobile cart containing the appropriate stains, glass slides, syringes, syringe holder, and double-headed microscope is brought to the patient's room or the clinical office by the cytotechnologist or pathology resident. The attending pathologist, surgeon and musculoskeletal radiologist typically review the plain radiographs and any additional study films, including CT scans, MRI scans, bone scans, and other appropriate clinical, laboratory, and radiographic data. The area in which the biopsy is to be performed is prepared in a standard fashion with povidone iodine. No local anesthetic is utilized. After antiseptic skin preparation, a 25-gauge needle on a 20-cc syringe containing 5 cc of air is advanced through the skin into the tumor. The plunger on the syringe is then withdrawn fully, creating an additional 15 cc of vacated potential air space within the syringe and a vacuum at the needle tip. The needle is then rapidly and sequentially advanced and withdrawn within the tumor mass in multiple directions to provide a representative sample from multiple areas within the tumor. Ideally, this procedure is performed through one puncture site within the tumor wall. Once the aspiration passes within the tumor have been completed, the negative pressure is released and the plunger returns to the 5-cc position in the syringe with 5 cc of air remaining in the syringe. The needle is then withdrawn from the patient. An assistant maintains pressure on the area for a minimum of 5 minutes to prevent tumor bleeding and local tumor spread. The aspirated specimen typically fills only the hub of the

needle. Rarely, with a bloody specimen, a small amount of bloody tissue sufficient to reach to the channel of the syringe is obtained. The sample of aspirated material is expressed onto glass slides utilizing the retained 5 cc of air in the syringe to expel the specimen. The material is then smeared with a second glass slide, and one smear is air dried and stained with Diff-Quik (Fisher Scientific Biomedical Sciences, Inc, Swedesboro, NJ), and the other slide is immediately immersed in 95% ethanol for subsequent staining by the Papanicolaou method.[5,6]

The Diff-Quik–stained specimen is immediately examined by the cytopathologist and surgeon on site. When the specimen is clearly diagnostic, no further passes are made. In many instances, however, additional passes are necessary for appropriate studies. An additional specimen is often obtained for cell block analysis with hematoxylin and eosin staining and other ancillary stains or studies as deemed appropriate, including flow cytometry, electron microscopy, cytogenetics, ploidy analysis, cultures, and immunohistochemical staining. A saline rinse of the aspiration syringe and needle is performed. This rinse can be centrifuged and the pellet can be stained and studied by any of the above techniques. Details of the technique have been published elsewhere.[1-7]

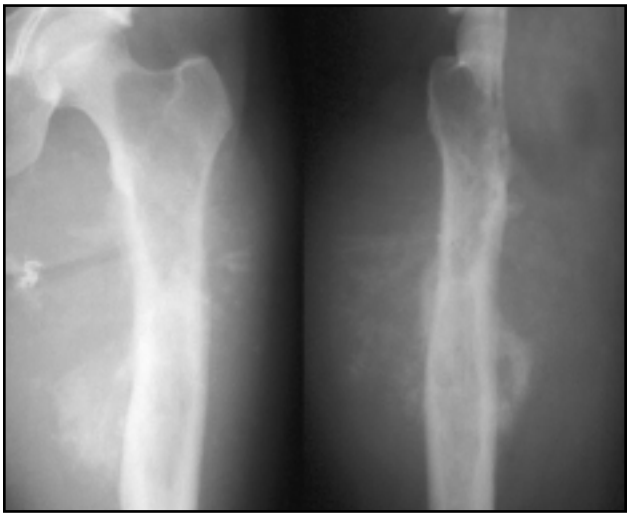


Fig 2A. — AP and lateral radiographs of the proximal femur reveal cortical destruction and a large bone lesion with soft-tissue extension and extensive spiculated periosteal reaction. (Figs 2A-C used by permission of W. G. Ward, MD.)

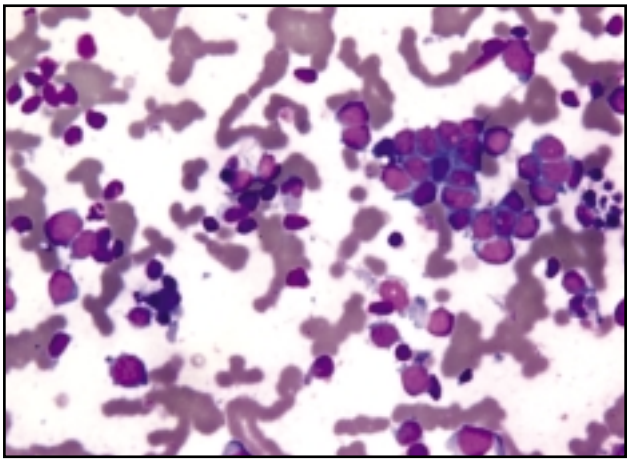


Fig 2B. — A Diff-Quik stain of an initial aspiration revealed solitary and small clusters of mostly uniform cells with high nuclear to cytoplasmic ratios (small blue cells).

Results

In a prior study,[2] we reported that sarcoma was correctly recognized in 61 (84%) of 73 consecutive aspirations from 67 patients with soft-tissue sarcoma, obviating the need for open biopsy in most. In another study,[5] we reported that in primary bone tumors, 48 of 66 (73%) consecutive FNABs of primary bone tumors were diagnostic. We have empirically noted that FNAB is best utilized in lytic bone lesions and in those lesions that have cortical destruction with soft-tissue extension. Biopsy of these areas generally provides diagnostic material. Needle biopsy of sclerotic bone lesions has been less rewarding. It is technically difficult to obtain any specimen from a hard, sclerotic, bony lesion. The same can be said of some mixed lesions. If a lesion is predominantly lytic, we have empirically noted that we can usually derive a diagnosis from FNAB, but if it is primarily sclerotic, the aspirates are less likely to be diagnostic. We seldom perform FNAB on a sclerotic or a predominantly sclerotic lesion unless there is a soft-tissue extension. This is usually the case with osteosarcoma; most cases can be satisfactorily diagnosed either by FNAB due to the soft-tissue extension or by a biopsy of the associated osteolytic tumor regions. Imaging-guided aspiration biopsy is usually performed on nonpalpable lesions.

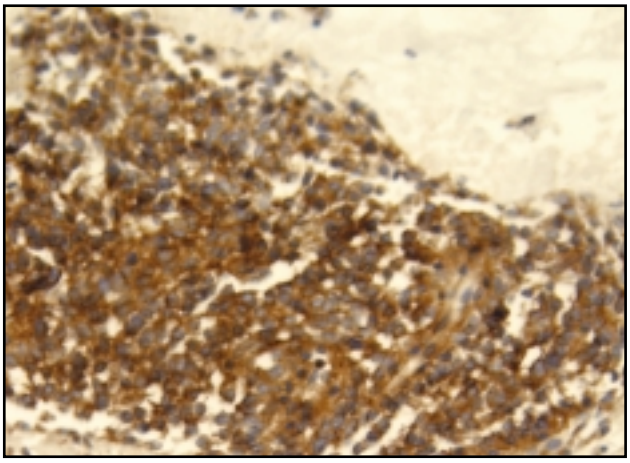
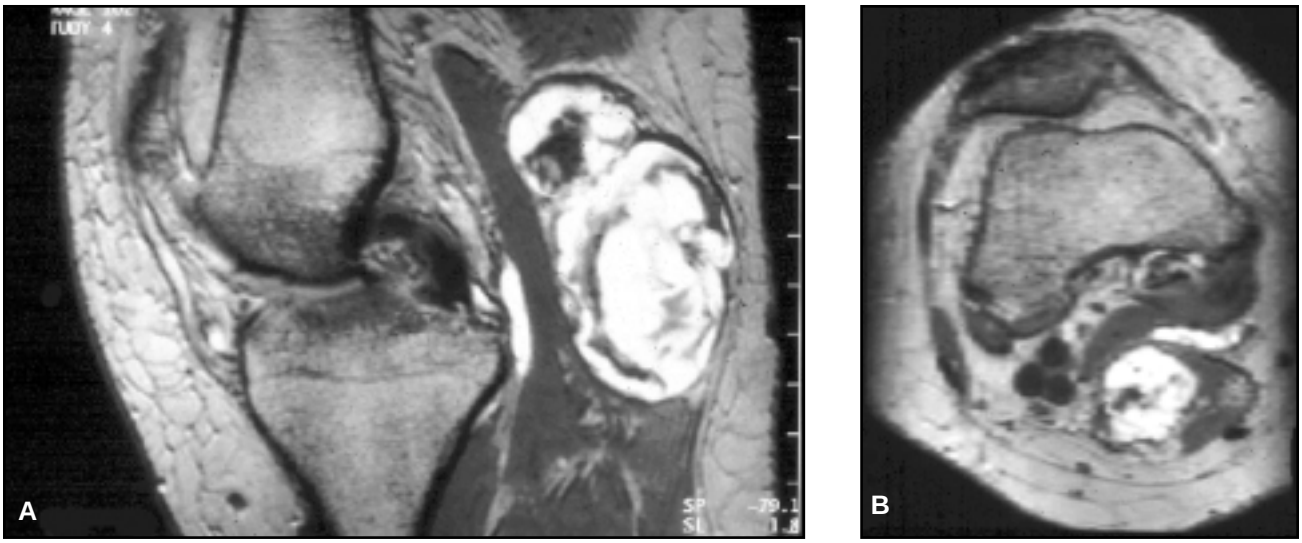


Fig 2C. — Cell block material from FNA of Ewing's sarcoma with positive CD99 (brown membranous material), which is considered diagnostic of Ewing's sarcoma in this clinical setting.



Figs 3A-B. — Coronal (A) and axial (B) MRI scans reveal an inhomogeneous popliteal fossa mass. The small lesion deep to the mass is a true Baker's cyst. (Figs 3A-C used by permission of W. G. Ward, MD.)

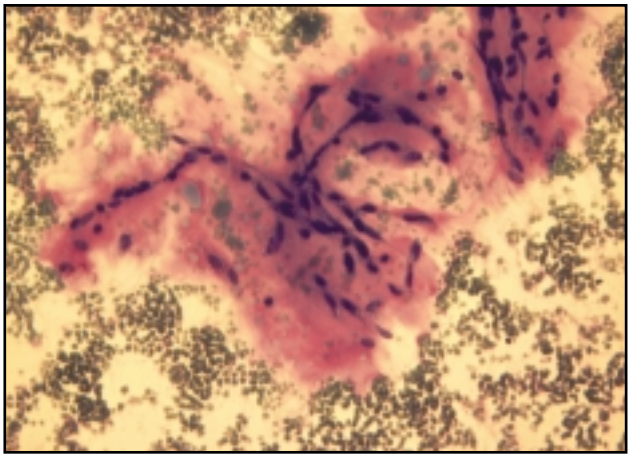


Fig 3C. — FNAB of the lesion in 3A-B reveals extraskeletal myxoid chondrosarcoma.

We have yet to encounter a serious complication resulting from FNAB. We have seen only one local recurrence of a bone tumor diagnosed by FNAB. This occurred in a patient with a proximal tibial giant cell tumor. Approximately 8 months following his intralesional curettage, resection, and treatment with local adjuvants (phenol, hydrogen peroxide, and cauterization), a small subcutaneous recurrence developed. This could have been related to his needle biopsy or to his open intralesional curettage procedure. During the same time period, we had multiple patients who had local recurrences and significant local complications from poorly performed or inappropriate open biopsies. These included patients who had their tumors rendered unresectable because of tumor spread at the time of biopsy, as well as patients who had their limb-sparing operations rendered more difficult due to tissue contamination and tumor spread along intermuscular and neurovascular planes. In some patients, this tumor dissemination has led to death from locally recurrent disease.

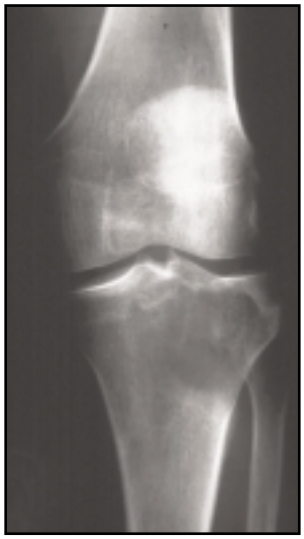


Fig 4A. — AP knee radiograph revealing a destructive lytic lesion in the proximal tibia.

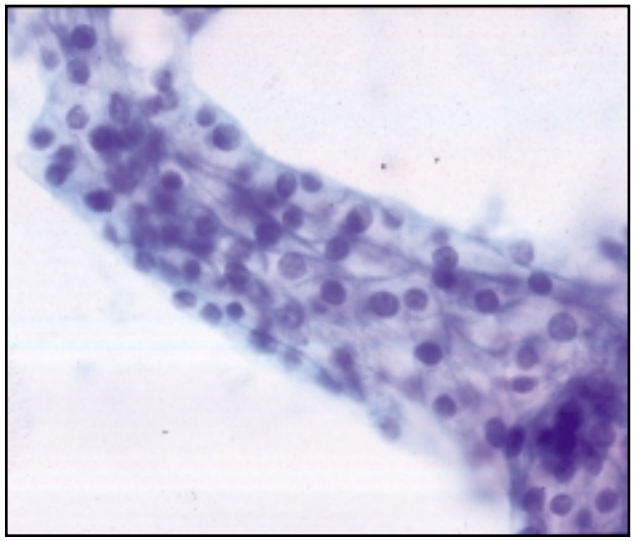


Fig 4B. — The FNAB reveals sheets and aggregates of cells with abundant clear cytoplasm due to metastatic renal cell carcinoma. (Figs 4A-B used by permission of W. G. Ward, MD.)

There has been one significant misdiagnosis. A retarded patient presented with a mass over the greater trochanter. The preliminary FNAB report suggested soft-tissue malignancy due to the presence of atypical

cells. It was resected before the final report was prepared. The mass turned out to be a hematoma with reactive soft-tissues rather than a sarcoma. This final diagnosis was properly appreciated on the final reading of the FNAB. Due to his retardation, he was unable to give any history of trauma, which might have facilitated the proper diagnosis initially. The patient, nonetheless, did well following resection of the subcutaneous hematoma.

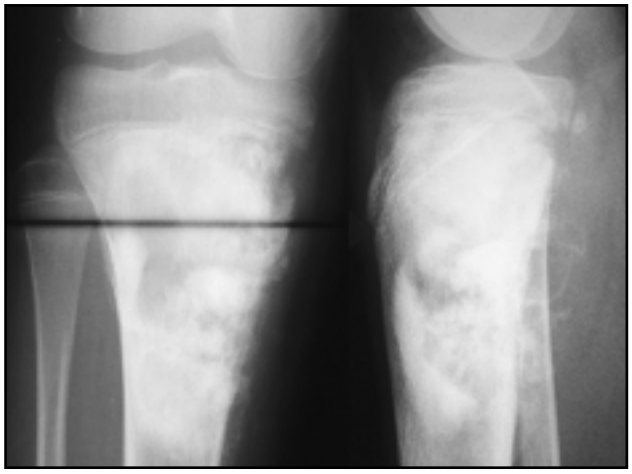


Fig 5A. — AP and lateral radiograph of the proximal tibia reveals a combined osteolytic and osteoblastic process involving the proximal tibial metaphysis with cortical disruption and soft-tissue extension. (Figs 5A-F used by permission of W. G. Ward, MD.)

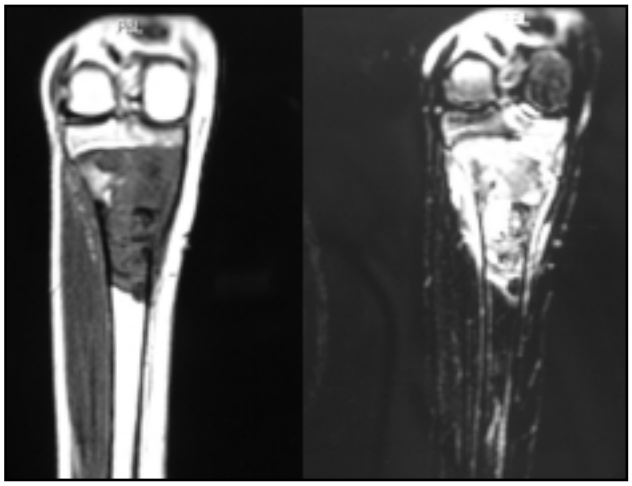


Fig 5C. — Coronal T1- and T2-weighted images reveal the metaphyseal osteosarcoma with extension beyond the cortices into the soft-tissues as well as into the proximal tibial epiphysis.

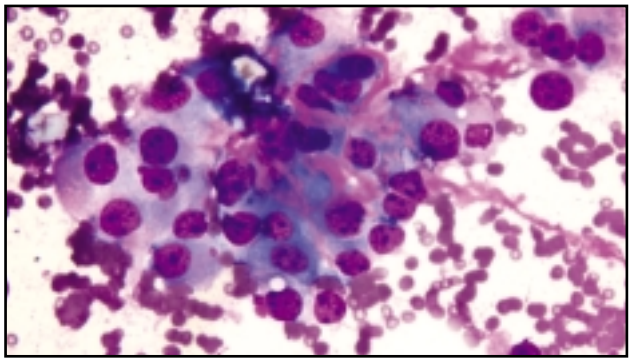


Fig 5E. — This Diff-Quik stain of osteosarcoma FNAB material reveals numerous round to spindled cells and a background of osteoid matrix material.

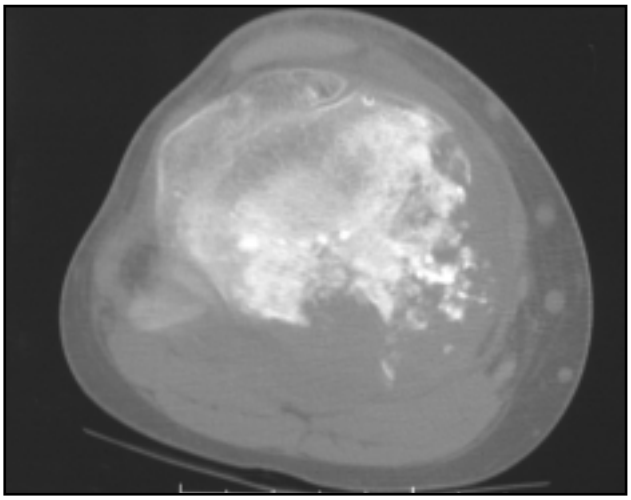


Fig 5B. — Axial CT scan reveals an osteolytic and osteoblastic process with soft-tissue extension beyond the disrupted posteromedial cortex.

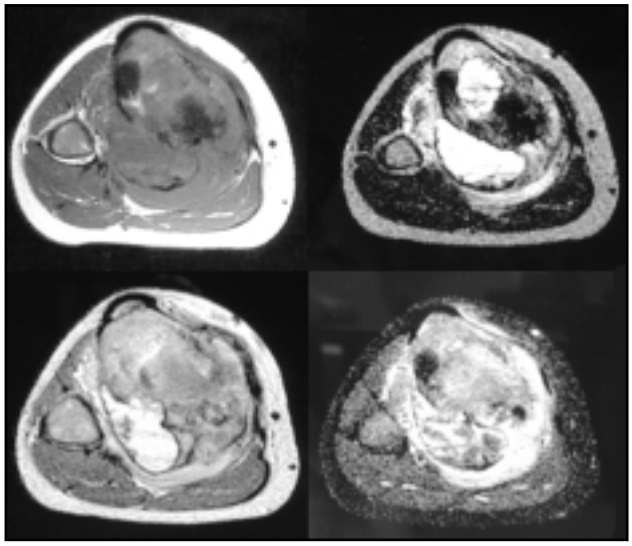


Fig 5D. — Composite of four axial MRI images reveals the medullary and soft-tissue extension of the proximal tibia lesion.

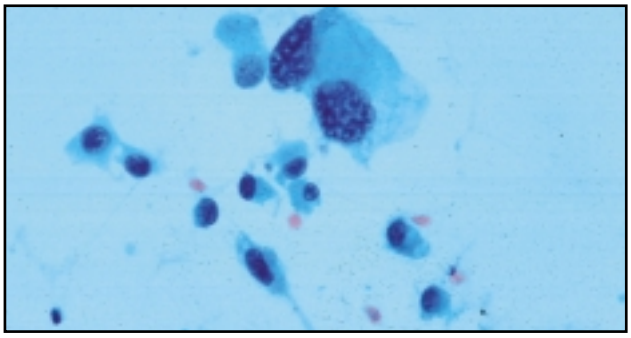


Fig 5F. — Papanicolaou stain of osteosarcoma FNAB material reveals large atypical cells. The finding of cells such as in Figs 5E-F are virtually diagnostic of osteosarcoma in children and adolescents with typical radiologic features of osteosarcoma.

Illustrative Case Presentations

Case 1

A 15-year-old boy presented to the orthopaedic oncology clinic for evaluation of an upper thigh mass. He and his family had noted the mass over 6 months but thought it was related to a sports injury. The mass and the pain persisted. He sought medical attention and plain radiographs were obtained. The anteroposterior and lateral proximal femoral radiographs revealed extensive cortical destruction and periosteal reaction with a spiculated periosteal pattern (Fig 2A). FNAB revealed multiple small, round, blue cells consistent with Ewing's sarcoma (Fig 2B). A CD99 immunoperoxidase stain was supportive of the diagnosis (Fig 2C). Chemotherapy was initiated within several days of his initial office visit without the need for open surgical procedure.

Case 2

A 65-year-old woman presented to her physician with complaints of a popliteal fossa mass. MRI demonstrated a large popliteal fossa mass with homogeneous signal (Fig 3A-B). The radiologists interpreting the case diagnosed a hemorrhagic Baker's cyst. The orthopaedic surgeon was concerned that a neoplasm was present and referred the patient to the orthopaedic oncology clinic. FNAB was performed in the clinic and revealed an extraskeletal myxoid chondrosarcoma (Fig 3C), which was therefore resected. The patient remains free of disease at 1 year of follow-up.

Case 3

A 65-year-old man presented with knee pain. Radiographs revealed a lytic lesion in the proximal tibia (Fig 4A). FNAB was performed in the clinic and allowed a diagnosis of metastatic renal cell carcinoma to be diagnosed within 1 hour of clinic arrival (Fig 4B). He remains healthy 3 years later following embolization, curettage, cementation, internal fixation, and radiation.

Case 4

A 12-year-old boy presented with pain in the proximal tibia. Plain radiographs revealed a combined osteolytic and osteoblastic process involving the proximal tibial metaphysis with soft-tissue extension and cortical disruption (Fig 5A). A CT scan confirmed the osteolytic and osteoblastic changes and soft-tissue extension (Fig 5B). An MRI scan further demonstrated the destructive process involving the metaphysis with epiphyseal extension as well as soft-tissue extension (Fig 5C-D). FNAB confirmed the diagnosis of osteosarcoma, thus allowing him to begin chemotherapy within days (Fig 5E-F). He had an excellent response to chemotherapy and subsequently underwent proximal tibial resection with allograft reconstruction, which continues to function well. He remains free of disease as of latest follow-up.

Conclusions

FNAB facilitates the appropriate care of patients when it is coupled with a treatment regimen utilizing preoperative chemotherapy for high-grade sarcomas and wide resection for all sarcomas. We believe it leads to a lower local recurrence rate due to the lesser degree of local tumor dissemination in comparison to open biopsy. We have encountered no life- or limb-threatening complications in 8 years of use, a statement that cannot be made about patients treated during the same time period who were diagnosed by open biopsy. However, it must be remembered that in any case wherein the cytologic diagnosis does not match the suspected clinicoradiographic diagnosis, an open biopsy should be performed before any major surgical procedure is undertaken. In our experience, FNAB has been useful in the workup and management of patients with orthopaedic tumors.

All figures in this article are copyrighted images from William G. Ward, MD

References

1. Kilpatrick SE, Koplyay PD, Ward WG, et al. Epithelioid hemangioendothelioma of bone and soft-tissue: a fine-needle aspiration biopsy study with histologic and immunohistochemical confirmation. *Diagn Cytopathol*. 1998;19:38-43.
2. Kilpatrick SE, Ward WG, Cappellari JO, et al. Fine-needle aspiration biopsy of soft-tissue sarcomas: a cytomorphologic analysis with emphasis on histologic subtyping, grading and therapeutic significance. *Am J Clin Pathol*. 1999;112:179-188.
3. Nicol KK, Ward WG, Pike EJ, et al. Fine-needle aspiration biopsy of gouty tophi: lessons in cost-effective patient management. *Diagn Cytopathol*. 1997;17:30-35.
4. Nicol KK, Ward WG, Savage PD, et al. Fine-needle aspiration biopsy of skeletal versus extraskeletal osteosarcoma. *Cancer*. 1998;84:176-185.
5. Ward WG, Kilpatrick S. Fine-needle aspiration biopsy of primary bone tumors. *Clin Orthop*. 2000;373:80-87.
6. DeMay RM. Stains. In: DeMay RM, ed. *The Art and Science of Cytopathology*. Vol 1. Chicago, Ill: ASCP Press; 1996:14-15.
7. Kilpatrick SW, Cappellari JOP, Ward WG, et al. Is fine-needle aspiration biopsy a practical alternative to open biopsy for the primary diagnosis of sarcoma? Experience with 140 patients. *Am J Clin Pathol*. 2001;115:56-68.
8. Mankin HJ, Lange TA, Spanier SS. The hazards of biopsy in patients with primary bone and soft-tissue tumors. *J Bone Joint Surg Am*. 1982;64:1121-1127.