Optimizing Ewing Sarcoma and Osteosarcoma Biopsy Acquisition: A Children's Oncology Group Bone Tumor Committee Consensus Statement

Matthew S. Dietz, MD[1]; Alyaa Al-Ibraheemi, MD[2]; Jessica L. Davis, MD[3]; C. Matthew Hawkins, MD[4]; Brian T. Craig, MD[5]; Roshni Dasgupta, MD[6]; David S. Geller, MD[7]; David S. Shulman, MD[8]; Sarah Cohen-Gogo, MD, PhD[9]; Ajay Gupta, MD[10]; Susan L. Whiteway, MD[11]; Emily K. Slotkin, MD[12]; Christine M. Heske, MD[13]; Safia K. Ahmed, MD[14]; Daniel J. Indelicato, MD[15]; Catherine M. Albert, MD[16,17]; Nicole Montgomery, MD[18]; Jesse K. Sandberg, MD[19]; Holcombe E. Grier, MD[8]; Mark Krailo, PhD[20]; Michael S. Isakoff, MD[21]; Elyssa Rubin, MD[22]; Elizabeth R. Lawlor, MD, PhD[16,17]; Steven G. DuBois, MD[8]; Leo Mascarenhas, MD[23]; Patrick J. Grohar, MD, PhD[24]; Odion Binitie, MD[25]; Damon Reed, MD[12]; Katherine Janeway, MD[8]; Ryan D. Roberts, MD, PhD[26,*]; and Kelly M. Bailey, MD, PhD[27,*]

Abstract

Trends in diagnostic biopsy sample collection approaches for primary bone sarcomas have shifted in the past 2 decades. Although open/incisional biopsies used to be the predominant approach to obtain diagnostic material for Ewing sarcoma and osteosarcoma, image-guided core needle biopsies have increased in frequency and are safe for patients. These procedures are less invasive and reduce recovery times but have potential limitations. The quantity and quality of tissue obtained through these procedures vary between institutions. Acquired viable tissue volumes can be low, limiting the conduct of downstream expanded clinical workup, molecular analyses, and research. Patients with advanced Ewing sarcoma and osteosarcoma continue to have overall poor outcomes despite dose-intensive cytotoxic chemotherapy. The biology of treatment resistance is not currently well understood, partly due to limited availability of relevant tissue to study. There is a need for access to quality tumor specimens for molecular and other analyses to identify high-risk tumor subsets and drive discovery to improve patient outcomes. Given broad variability in bone tumor tissue procurement and processing across member institutions, the Children's Oncology Group Bone Tumor Committee convened a multidisciplinary group of experts to outline the current and near-future tissue needs for optimal clinical care and access to research platforms. The goal of this working group was to provide high-level guidance on biopsy practices that safely meet these evolving needs. Harmonizing tissue collection practices is paramount to improving the care of children, adolescents, and young adults diagnosed with Ewing sarcoma and osteosarcoma.

J Natl Compr Canc Netw, doi:10.6004/jnccn.2024.7063
Published online December 27, 2024

Ewing sarcoma and osteosarcoma are high-grade sarcomas of bone and bone/soft tissue, with peak incidence in adolescents and young adults. Although metastatic disease currently remains the most meaningful prognostic indicator, efforts are underway to better delineate biologic subgroups associated with treatment response and resistance in both Ewing sarcoma and osteosarcoma. Novel risk-stratified treatment approaches for bone sarcomas are emerging and will inform disease management and future clinical trial enrollment.[1] Molecular profiling is an essential component of the emerging proposals for bone sarcoma risk stratification, similar to other pediatric (eg, Wilms tumor, neuroblastoma, soft tissue sarcomas) and adult solid tumors (eg, breast cancer, lung adenocarcinoma) for which molecular biomarker status now guides standard-of-care therapy.[2–7] Indeed, international clinical guidelines for bone sarcoma now recommend the routine acquisition of snap frozen and fresh tissue for clinical molecular studies.[8–10] To achieve this vision for patients with Ewing sarcoma and osteosarcoma, the field must ensure standard clinical practices for the expert procurement, processing, and evaluation of tumor biomaterials for all patients.

However, patients diagnosed with Ewing sarcoma and osteosarcoma receive cancer care at a variety of institutions, including adult and pediatric hospitals and academic and community centers, and care is delivered by pediatric and medical oncology specialists.[11] In fact, the diagnostic center may differ from the facility where the patient receives treatment. Significantly more

[1]University of Utah and Primary Children's Hospital, Salt Lake City, UT; [2]Boston Children's Hospital, Boston, MA; [3]Indiana University, Indianapolis, IN; [4]Emory University School of Medicine, Department of Radiology and Imaging Sciences, Children's Healthcare of Atlanta, Atlanta, GA; [5]Children's Wisconsin, Medical College of Wisconsin, Milwaukee, WI; [6]University of Cincinnati, Department of Surgery, Cincinnati Children's, Cincinnati, OH; [7]Children's Hospital at Montefiore, Montefiore Medical Center, The University Hospital for Albert Einstein College of Medicine, Bronx, NY; [8]Dana-Farber/Boston Children's Cancer and Blood Disorders Center, Boston, MA; [9]The Hospital for Sick Children, Toronto, Ontario, Canada; [10]Division of Pediatric Oncology, Department of Pediatrics, Roswell Park Comprehensive Cancer Center, University at Buffalo Jacobs School of Medicine and Biomedical Sciences, Buffalo, NY; [11]Walter Reed National Military Medical Center, Bethesda, MD; [12]Memorial Sloan Kettering Cancer Center, New York, NY; [13]Pediatric Oncology Branch, National Cancer Institute, National Institutes of Health, Bethesda, MD; [14]Department of Radiation Oncology, Mayo Clinic Arizona, Phoenix, AZ; [15]Department of Radiation Oncology, University of Florida, Gainesville, FL; [16]Ben Towne Center for Childhood Cancer Research, Seattle Children's Hospital, Seattle, WA; [17]Department of Pediatrics, University of Washington, Seattle, WA; [18]Texas Children's Hospital, Baylor College of Medicine, Houston, TX; [19]Department of Pediatric Radiology, Lucile Packard Children's Hospital, Stanford University, Stanford, CA; [20]Department of Population and Public Health Sciences, Keck School of Medicine of the University of Southern California, Los Angeles, CA; [21]Center for Cancer and Blood Disorders, Connecticut Children's Medical Center, Hartford, CT; [22]Children's Hospital of Orange County, Orange, CA; [23]Cedars-Sinai Medical Center, Los Angeles, CA; [24]Center for Childhood Cancer Research, Children's Hospital of Philadelphia, University of Pennsylvania, Perelman School of Medicine, Philadelphia, PA; [25]Moffitt Cancer Center, Tampa, FL; [26]Nationwide Children's Hospital, Columbus, OH; and [27]University of Pittsburgh School of Medicine, UPMC Children's Hospital of Pittsburgh, Pittsburgh, PA.

*R.D. Roberts and K.M. Bailey are co–senior authors.

errors and complications have been reported when biopsies are performed outside of the treatment center.[12] Decentralized and fragmented care threatens consistency in procurement and processing practices of diagnostic biopsy/biomaterials. Variability in the amount and usability of material acquired from biopsies of primary bone tumors limits advances in clinical care and research. Furthermore, sample processing requirements of bone tumors relative to other tumor types heighten the need for a standardized approach to tissue collection. Although open/incisional biopsies are considered the gold standard technique, image-guided core needle biopsies are increasingly used for the acquisition of diagnostic biopsy material for bone tumors, and in many centers have become the dominant approach to biopsy.[13] Given the potential for variable amounts of tissue procured through this approach, components of clinical care, including pathologic assessment, diagnostic accuracy, and molecular evaluations, along with additional research studies with leftover tissue (following informed consent), can be negatively impacted if inadequate tissue is obtained. Indeed, a recent review of Ewing sarcoma and osteosarcoma specimens in the Children's Oncology Group (COG) biorepository identified quality assurance failures due to diagnostic discordance (4% of cases) or lack of viable tumor (7% of cases). Among cases with diagnostic concordance, variable volumes of tumor were present, including cases with scant viable tumor tissue available for additional testing.[14] In addition to the variability noted in the amount of viable tumor acquired at diagnosis, there is often variability in tissue processing and allocation practices between institutions, resulting in the unintended procurement of nondiagnostic, acid-degraded, or insufficient volumes of Ewing sarcoma and osteosarcoma tumor material. The allocation and processing of tumor biopsy material affect the available options for subsequent clinical and research analyses, importantly including quality molecular analysis.

To address these points, the COG Bone Tumor Committee convened a multidisciplinary group of experts to evaluate available literature and institutional practices to generate the following guidance for the acquisition of Ewing sarcoma and osteosarcoma biopsy material. Current and near-future biopsy tissue requirements in the field are highlighted, with a lens toward ensuring that patients maintain access to clinical trial opportunities, which is a standard-of-care offering for patients with Ewing sarcoma and osteosarcoma. This guidance may require practice changes at institutions, with the understanding that clinical decisions about tissue acquisition at the individual case level must be based on tumor location and patient condition.

Biopsy Planning for Suspected Ewing Sarcoma and Osteosarcoma

When Ewing sarcoma or osteosarcoma is suspected, prebiopsy planning with pediatric oncology, radiology, interventional radiology, pathology, and orthopedic oncology or pediatric surgery is necessary to ensure that the necessary tissue specimens are obtained and delivered to pathology promptly (ie, coordinating with the on-call pathologist or arranging a STAT courier to minimize warm ischemia time) while prioritizing patient safety (Figure 1). This may result in practice or protocol changes at individual institutions. Given the importance of multidisciplinary planning for patients with Ewing sarcoma and osteosarcoma, referral to specialty institutions is recommended.[12] As with any procedure, patients or family must provide informed consent for the invasive collection of tumor material. The recommended tissue volumes described in the following discussion ensure equivalency regardless of biopsy approach and tissue collection necessary for modern tumor

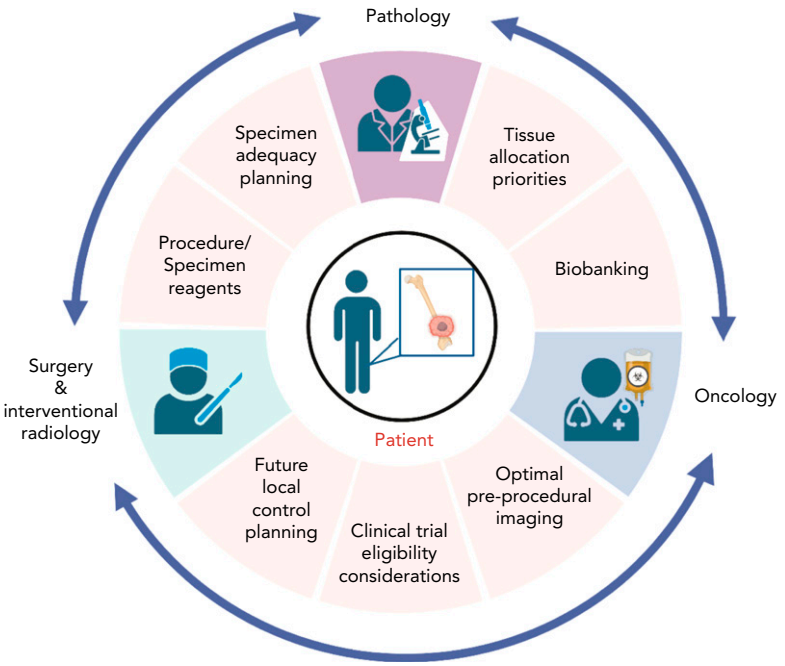

Figure 1. Prebiopsy multidisciplinary discussion recommendations. A multidisciplinary discussion for the management of patients undergoing biopsy of a suspected Ewing sarcoma or osteosarcoma is needed to coordinate appropriate preoperative imaging, future oncologic/local control surgical planning, pathology workflow including timely tissue processing and allocation (eg, histology, molecular and research studies), clinical trial eligibility considerations, and acquisition of biopsy tissue specimen reagents (eg, liquid nitrogen, dimethyl sulfoxide [DMSO], media).

assessment and emerging disease stratification. Of note, efforts to obtain adequate clinical specimens should be differentiated from those of research-only biopsies, for which the tissue is not processed as a diagnostic specimen and which require separate informed consent for enrollment on specific Institutional Review Board (IRB)–approved research protocol(s). Collection of residual tissues for biobanking or submission to tissue repositories likewise requires IRB-approved informed consent. Recently the Fight Osteosarcoma Through European Research (FOSTER) and EURO EWING Consortium (EEC) provided guidance on biologic sample collection to specifically advance research and highlight patients' appetite for participating in these studies.[15]

Establishing the quantity of tumor tissue material required prior to the procedure is instrumental in ensuring that all desired tissue-based analyses can be performed. This is especially important when using image-guided core needle biopsy techniques, because they typically yield smaller tumor samples compared with open or incisional biopsies. In both North America and Europe, there is consensus that bone sarcomas with an associated large soft tissue component are generally safe to biopsy, and for such cases, procuring diagnostic material is typically quite feasible, as has been demonstrated in soft tissue sarcomas.[16,17] However, there are cases in which tumors arise in anatomically sensitive locations and biopsy can carry greater risk. For example, for a patient with Ewing sarcoma arising from a vertebral body or rib, it may not be possible or safe to increase the amount of tumor material obtained. Although studies of soft tissue sarcomas have shown equivalent rates of local recurrence when comparing core versus open biopsies,[18] the rates of biopsy tract seeding are much lower with core needle biopsies. We still advocate for careful biopsy site planning, including limiting to one biopsy site, with the tract ideally placed in the same location as the planned incision for eventual definitive resection or in a location where the tract can be safely resected to reduce the risk of local recurrence. The specific anatomy of the biopsy tract will vary based on whether the primary tumor is in the extremity or the trunk, and should be explicitly discussed in a multidisciplinary fashion. These cases underscore the need for prebiopsy planning and coordination with pathology to ensure specimens that can be obtained undergo appropriate processing and allocation to maximize specimen utility.

Image-Guided Core Needle Biopsies

Image-guided techniques for the acquisition of biopsy material of suspected Ewing sarcoma and osteosarcoma are often performed by interventional radiologists.[19] The safety of image-guided core needle biopsy has been established in prospective and retrospective studies without additional safety events.[20–22] When core biopsy is used for sample collection, the volume of viable tissue obtained for future use is determined by the number of cores collected, gauge of needle used,[23] length of the core (>10 mm superior to <5 mm[24]), and viability of tumor in the area sampled. However, this method of tissue acquisition lacks standardization. Discussions with members of the COG Bone Tumor Committee revealed differences across institutions in sampling techniques, such as location sampled and the number and length of biopsy cores obtained. Standard recommendations are needed to ensure this diagnostic approach results in sufficient tissue for the patient's clinical needs as well as desired research purposes.

Ideally, specimens are acquired from an area presumed to contain viable tumor tissue. Intraoperative tumor viability checks are not available at every institution, in which case viability is assessed by pathology postprocedurally (see later section on "Pathology/Specimen Processing Plan"). Intraprocedure imaging is often used to guide sampling of different portions of the tumor to increase the likelihood of acquiring viable material.[19] Given that fine-needle aspirates (FNAs) are inadequate alone for the diagnosis of osseous lesions,[25,26] we do not recommend using FNAs for the diagnosis of suspected Ewing sarcoma or osteosarcoma.

When targeting a suspected viable tumor site, we provide the following recommendations. For bone tumors with a soft tissue component, obtain biopsies from the soft tissue whenever feasible, with the aim of collecting a total of 15 to 20 cores measuring 1.5 to 3 cm using a 16-gauge needle. We recognize that some institutions prefer alternative gauge sizes. The overarching goal of this recommendation is to acquire an equivalent volume of tissue as that acquired through open biopsies (1–3 cm^3 or 1–3 g of tissue). Automated or vacuum-assisted biopsy devices are available at many institutions and can increase the speed at which multiple biopsies can be obtained.[27–29] Because many downstream testing applications require the presence of viable tumor (sometimes >50%), the recommended ≥15 cores help to achieve adequate viable tumor acquisition even when some cores are small (<1 cm) and mostly necrotic. For bone tumors without a soft tissue component (or the soft tissue component cannot be adequately accessed), we recommend collecting 5 to 7 cores for bone biopsies using a 12- to 13-gauge needle. This number of cores is feasible and does not require a second skin incision (although a second cortical hole is needed in rare cases). When osteosarcoma is suspected, acquire an additional 2 to 3 cores of the underlying osteoid using a 12- to 13-gague needle. These core totals would cover all clinical and research/clinical trial–based needs, including 2 to 4 blocks with 2 to 3 cores per block for clinical diagnostic needs, snap frozen tissue (~0.5 g per vial), and fresh or viably frozen tumor material. Additional allocation details are provided in Figure 2. An overview of clinical and downstream uses of Ewing sarcoma and osteosarcoma biopsy tissue is provided in Figure 3. Specimens should ideally be submitted to pathology on saline-dampened gauze/Telfa or, if not available, in a dry container (no formalin). Effective coordination with pathology is crucial so that tissue does not desiccate in the container and can be promptly processed within the pathology laboratory.

Open Biopsy by Pediatric or Orthopedic Surgical Oncology

Following basic open biopsy surgical principles, an open biopsy may be performed, preferably by the treating orthopedic oncologist or pediatric surgeon. Adhering to principles for safe open biopsies is critical. For extremity tumors, a small longitudinal incision that is in line with the planned resection incision should be used to allow for resection of the biopsy tract at the time of primary tumor resection. Similarly, for chest wall tumors, an incision that overlies the planned incision for definitive resection is recommended, generally along the course of the primarily affected rib. Other safety considerations include maintenance of hemostasis, minimizing dissection, limited drain use, and avoidance of neurovascular structures. If there is no soft tissue component, creating a bone defect during biopsy can increase the risk of pathologic fracture, so this should be considered carefully during open biopsy planning. A minimum of 1 cm^3 (ideally 1–3 cm^3 or 1–3 g of tissue) should be obtained. Identification of viable tumor tissue by frozen section, touch preparation, or other preferred method is recommended

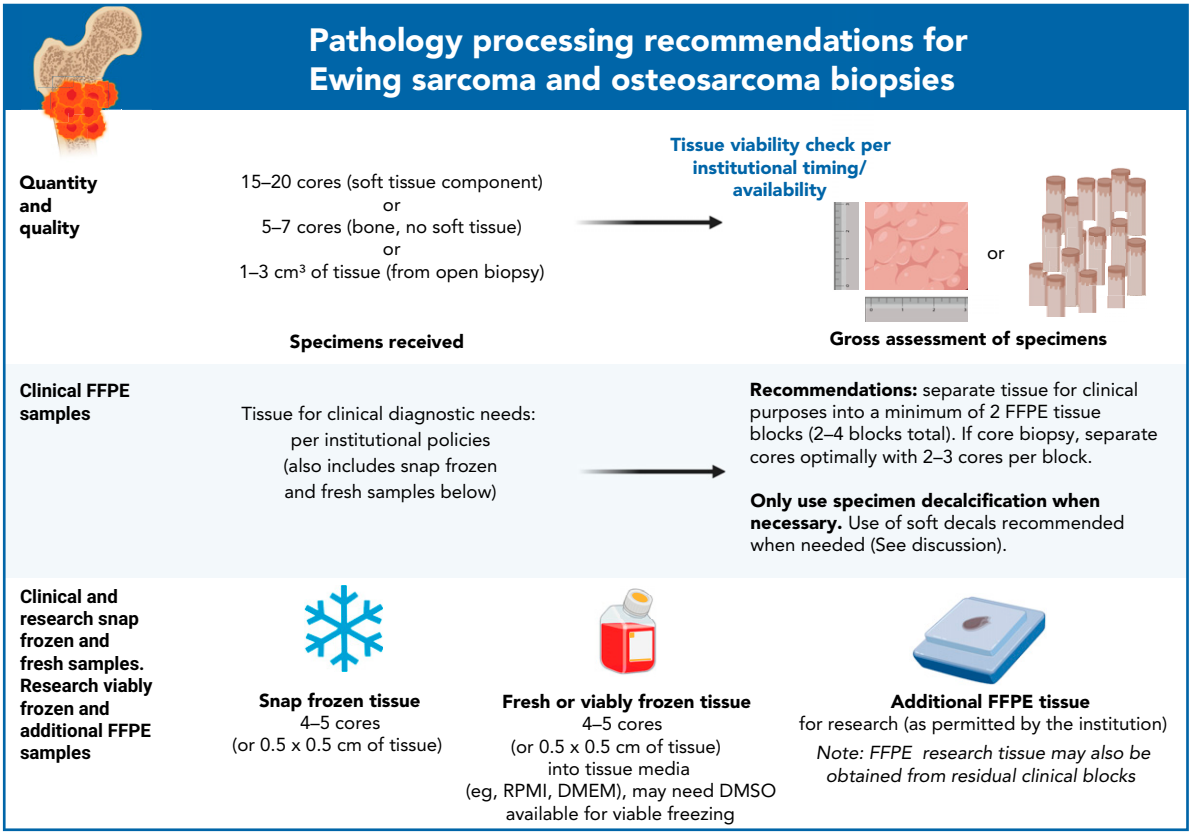

Figure 2. Pathology processing recommendations for Ewing sarcoma and osteosarcoma biopsies. The material should encompass all clinical needs as well as those related to consented clinical and translational research. Tissue allotment can be modified as needed to address the needs of specific institutional protocols.
Abbreviations: FFPE, formalin-fixed paraffin-embedded; DMEM, Dulbecco's Modified Eagle Medium; DMSO, dimethyl sulfoxide; RPMI, Roswell Park Memorial Institute medium.

when institutionally available.[30] If frozen section is used, care should be taken to minimize the amount of tissue used in order to preserve sufficient material for permanent sections or other purposes. Specimens should ideally be submitted on saline-dampened gauze/Telfa or, if not available, in a dry container (no formalin). See Figure 2 for biopsy processing and allocation recommendations.

Pathology/Specimen Processing Planning

Handling of biopsy specimens for histologic and subsequent molecular testing requires thoughtful timing, processing, and tissue prioritization. In addition, specific biospecimens are now a standard requirement for enrollment in many therapeutic clinical trials. Moreover, a growing number of registry, biomarker, and biorepository studies are available in which patients may be interested in participating.

Pathology Processing

After acquisition, specimens should be handled in an expeditious manner and not left unprocessed for more than a few minutes, because degradation begins immediately ex vivo. Upon arrival to the pathology department, the tissue may either undergo viability assessment via touch preparation and/or frozen section or be allocated for testing or further studies. Tissue viability assessment may help guide tissue adequacy and triaging. For routine diagnostic processing, clinical tumor tissue specimens are fixed in formalin and embedded in paraffin for histologic processing (known as *formalin-fixed paraffin-embedded* [FFPE] tissue). Although much improvement has occurred in the ability to extract nucleic acids from FFPE, there are some limitations to testing postfixation tissue, which can be avoided with snap frozen tissue. Therefore, it is beneficial to preplan the allocation of tissue for FFPE processing, snap freezing,[31] viable freezing, and/or fresh tissue applications. Preplanning allows time to request the materials for snap freezing (eg, liquid nitrogen, dewar flask, and cryogenic specimen storage container) and/or viable freezing (eg, 10% dimethyl sulfoxide [DMSO]–containing media) if not routinely on hand.[32] Frozen tissue used for diagnostic testing must be kept in a freezer in a CLIA-certified space with appropriate sample tracking mechanisms. Figure 2 details processing recommendations for bone tumor diagnostic biopsies.

Decalcification Recommendations for Bone Sarcoma Diagnostic Biopsies

Decalcification is used to make hard, mineralized tissues more amenable to subsequent cutting and analysis. Soft tissue biopsies generally do not need to undergo decalcification, and not all bone biopsies require decalcification. The feasibility of cutting a bone core or open biopsy should be checked prior to placing a specimen in decalcification solution. Pure acid decalcification (eg, hydrochloric acid or formic acid) should be avoided in biopsy samples because these agents can affect the histology for

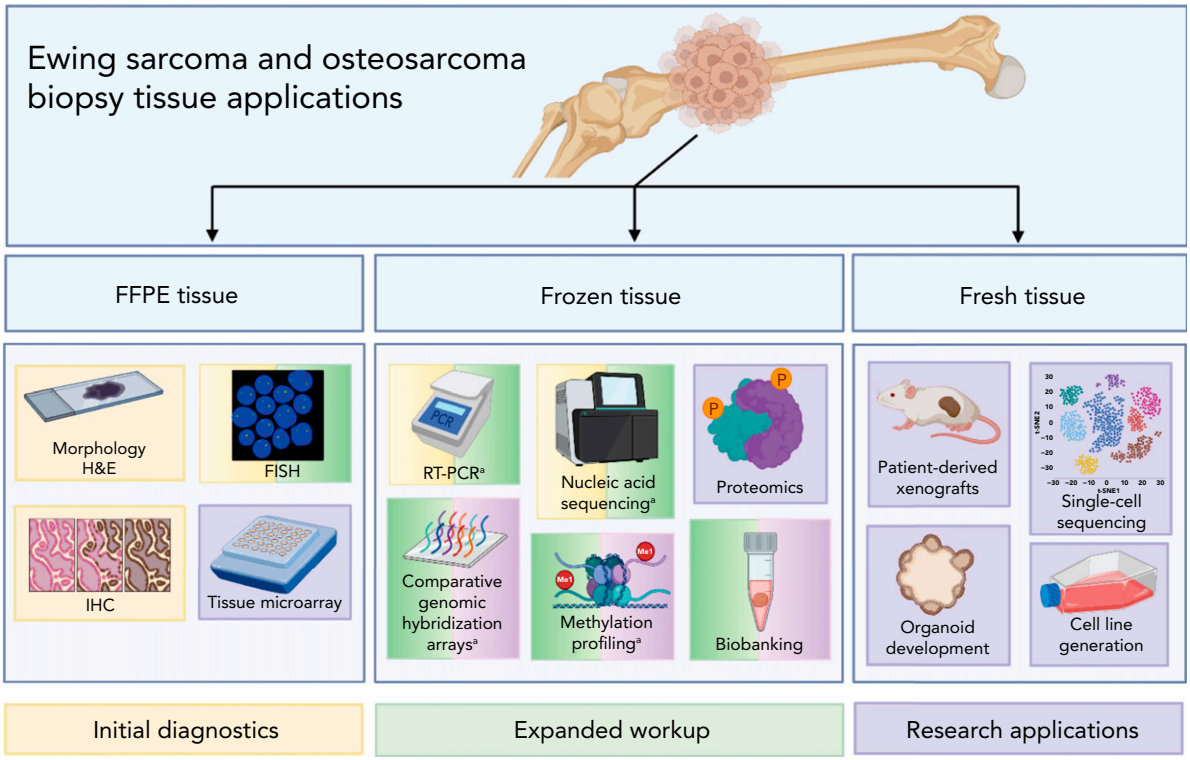

Figure 3. Clinical and downstream uses of Ewing sarcoma and osteosarcoma biopsy tissue. Tissue analysis methodologies are organized by suitable tissue preservation/processing method (FFPE, snap frozen tissue, or fresh tissue) and identified as components of initial diagnosis (yellow), expanded workup (green), or research applications (purple).
Abbreviations: decals, decalcification solutions; FISH, fluorescence in situ hybridization; FFPE, formalin-fixed paraffin-embedded; H&E, hematoxylin-eosin staining; IHC, immunohistochemistry.
[a]Indicates tissue analysis methodologies where FFPE specimens are also suitable.

primary diagnosis, interfere with immunogenicity of tissue, and denature nucleic acids, thus rendering the material less useful for molecular assays or downstream research.[33] "Soft decals" such as ethylenediaminetetraacetic acid (EDTA) or EDTA/formic acid combinations (ie, Formical 2000TM [Fisher Scientific] or similar products) are critically important in the age of molecular profiling, because nucleic acids are better preserved through this processing method.[34]

We suggest decalcifying tissue only if necessary. In the case of an open biopsy, a portion of the tissue may be able to be separated and soft enough to not require decalcification. Every effort should be made to generate fresh and frozen samples before placing biopsy material in either formalin or a decalcifying agent. If specimens are overly calcified or ossified and require decalcification for sampling, we recommend submitting at least one block of tissue in formalin, followed by a "soft decalcification" solution (and consider submitting the entire specimen). It is important to first "fix" the tissue in formalin before placing the tissue in decalcification solution. Tissues should be checked regularly and ideally should not remain in decalcification solution for longer than 2 to 3 hours (small samples may require only 30 to 60 minutes). Even with the use of "soft" decalcification methods, prolonged decalcification may affect the histology and/or preservation of nucleic acids. To help clearly identify potential downstream material issues, we recommend clearly stating the type of decalcifying agent used in the gross pathology report.

Future Directions

It is anticipated that future Ewing sarcoma and osteosarcoma clinical trials will incorporate molecular biomarkers into treatment risk stratification, and some, if not all, of these proposed biomarkers will require prospective validation on future clinical trials.[1] Identification of requisite tumor biopsy volumes and harmonization of pathology processing is paramount to prospective efforts to advance bone sarcoma clinical care and research. Furthermore, novel therapeutic agents are routinely introduced with companion biomarkers, thus it is expected that molecular biomarker testing will become the standard of care. Beyond diagnostic biopsy approaches, image-guided core needle biopsy and/or liquid biopsies may be increasingly used for on-therapy tumor response evaluations, already a common practice for early-phase clinical trials in adult-onset cancers.[35,36] Biological correlate analyses exploring tumor evolution, response, and resistance are not commonly conducted in the pediatric setting. However, as targeted therapies are developed and evaluated, it is anticipated that implementation of on-therapy biopsies will yield clinically actionable results. Although on-treatment biopsy is not on the immediate horizon, developing consensus recommendations for biopsy tissue sampling, as well as for pathology tissue processing and preservation, is immediately relevant for diagnostic tissue in bone sarcoma. These same tissue acquisition principles should also be considered with resection specimens, including both local control and surgical metastatic control, and when biopsies

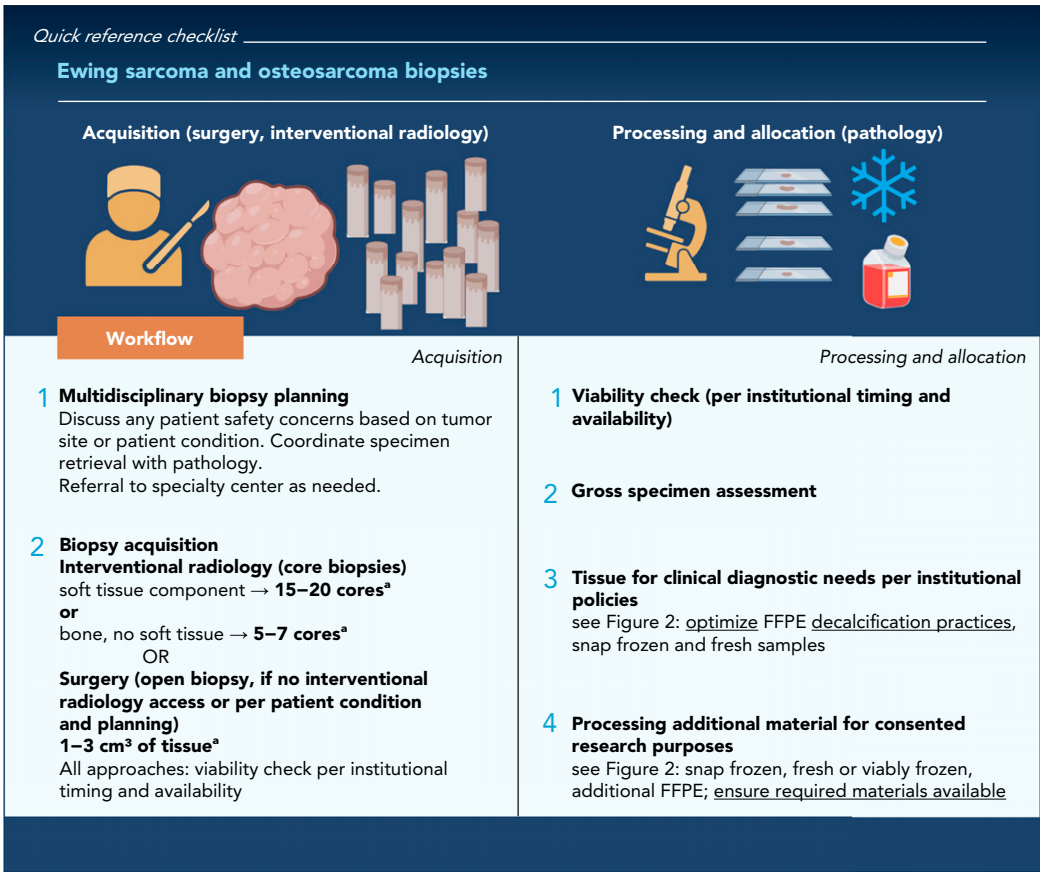

Figure 4. Quick reference checklist for Ewing sarcoma and osteosarcoma biopsies.
Abbreviation: FFPE, formalin-fixed paraffin-embedded.
[a]Goal if tumor site and patient condition permits to acquire approximately equivalent tissue volumes regardless of approach.

or resections are performed at the time of suspected disease relapse.

Conclusions

The COG Bone Tumor Committee recognizes the great need for safely improving the amount and usability of diagnostic biopsy material obtained from patients with Ewing sarcoma and osteosarcoma to continue to advance the field and improve care. Importantly, this patient population is cared for by both pediatric and medical oncologists in academic and community settings, underscoring the need to improve their decentralized care through collaboration. The clinical use of less invasive, safe, and accurate diagnostic biopsy techniques will continue to grow, as will the required tumor tissue volume to meet clinical and research needs. The diagnostic biopsy and processing recommendations for clinical management of bone sarcomas described herein reflect the perspective of clinical and scientific experts in the field in North America and aim to serve as a reference to facilitate harmonization in tissue acquisition and processing algorithms for Ewing sarcoma and osteosarcoma specimens (Figure 4).

Submitted November 20, 2023; final revision received July 24, 2024; accepted for publication August 2, 2024. Published online December 27, 2024.

Disclosures: The authors have disclosed that they have not received any financial considerations from any person or organization to support the preparation, analysis, results, or discussion of this article.
Funding: This research was supported by funding from the National Clinical Trials Network (NCTN) Operations Center (grant number U10CA180886) and NCTN Statistics & Data Center (grant number U10CA180899) from the National Cancer Institute of the National Institutes of Health.
Disclaimer: The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health.
Correspondence: Kelly M. Bailey, MD, PhD, University of Pittsburgh School of Medicine, UPMC Children's Hospital of Pittsburgh, Rangos Research Building, 4401 Penn Avenue, Pittsburgh, PA 15224.
Email: Kelly.Bailey@chp.edu

References

1. Shulman DS, Whittle SB, Surdez D, et al. An international working group consensus report for the prioritization of molecular biomarkers for Ewing sarcoma. NPJ Precis Oncol 2022;6:65.
2. Ambros IM, Tonini GP, Pötschger U, et al. Age dependency of the prognostic impact of tumor genomics in localized resectable MYCN-nonamplified neuroblastomas. Report from the SIOPEN Biology Group on the LNESG trials and a COG validation group. J Clin Oncol 2020;38:3685–3697.
3. Dix DB, Fernandez CV, Chi YY, et al. Augmentation of therapy for combined loss of heterozygosity 1p and 16q in favorable histology Wilms tumor: a Children's Oncology Group AREN0532 and AREN0533 study report. J Clin Oncol 2019;37:2769–2777.

4. Drilon A, Laetsch TW, Kummar S, et al. Efficacy of larotrectinib in TRK fusion-positive cancers in adults and children. N Engl J Med 2018;378:731–739.
5. Hong DS, DuBois SG, Kummar S, et al. Larotrectinib in patients with TRK fusion-positive solid tumours: a pooled analysis of three phase 1/2 clinical trials. Lancet Oncol 2020;21:531–540.
6. Irwin MS, Naranjo A, Zhang FF, et al. Revised neuroblastoma risk classification system: a report from the Children's Oncology Group. J Clin Oncol 2021;39:3229–3241.
7. Trahair T, Gifford AJ, Fordham A, et al. Crizotinib and surgery for long-term disease control in children and adolescents with ALK-positive inflammatory myofibroblastic tumors. JCO Precis Oncol 2019;3:PO.18.00297.
8. Bovée JVMG, Webster F, Amary F, et al. Datasets for the reporting of primary tumour in bone: recommendations from the International Collaboration on Cancer Reporting (ICCR). Histopathology 2023;82:531–540.
9. Strauss SJ, Frezza AM, Abecassis N, et al. Bone sarcomas: ESMO-EURACAN-GENTURIS-ERN PaedCan clinical practice guideline for diagnosis, treatment and follow-up. Ann Oncol 2021;32:1520–1536.
10. Biermann JS, Hirbe A, Ahlawat S, et al. NCCN Clinical Practice Guidelines in Oncology: Bone Cancer. Version 1.2025. For the most recent version, visit https://www.nccn.org
11. Parsons HM, Harlan LC, Schmidt S, et al. Who treats adolescents and young adults with cancer? A report from the AYA HOPE study. J Adolesc Young Adult Oncol 2015;4:141–150.
12. Mankin HJ, Mankin CJ, Simon MA. The hazards of the biopsy, revisited. Members of the Musculoskeletal Tumor Society. J Bone Joint Surg Am 1996;78:656–663.
13. Cooke-Barber J, Brungardt JG, Sorger M, et al. Pediatric and young adult image-guided percutaneous bone biopsy-a new standard of care? Ann Surg Oncol 2023;30:3658–3665.
14. Chen S, Shenoy A, Al-Ibraheemi A, et al. A report on the review of archived osteosarcoma and EWING sarcoma specimens at the Biopathology Center, BONE Sarcoma Committee, Children's Oncology Group. J Clin Oncol 2022;40(Suppl):Abstract 11524.
15. Green D, van Ewijk R, Tirtei E, et al. Biological sample collection to advance research and treatment: a Fight Osteosarcoma Through European Research (FOSTER) and Euro Ewing Consortium (EEC) statement. Clin Cancer Res 2024;30:3395–3406.
16. Birgin E, Yang C, Hetjens S, et al. Core needle biopsy versus incisional biopsy for differentiation of soft-tissue sarcomas: a systematic review and meta-analysis. Cancer 2020;126:1917–1928.
17. Pohlig F, Kirchhoff C, Lenze U, et al. Percutaneous core needle biopsy versus open biopsy in diagnostics of bone and soft tissue sarcoma: a retrospective study. Eur J Med Res 2012;17:29.
18. Binitie O, Tejiram S, Conway S, et al. Adult soft tissue sarcoma local recurrence after adjuvant treatment without resection of core needle biopsy tract. Clin Orthop Relat Res 2013;471:891–898.
19. Tomasian A, Hillen TJ, Jennings JW. Bone biopsies: what radiologists need to know. AJR Am J Roentgenol 2020;215:523–533.
20. Crenn V, Vezole L, Bouhamama A, et al. Percutaneous core needle biopsy can efficiently and safely diagnose most primary bone tumors. Diagnostics (Basel) 2021;11:1552.
21. Mitton B, Seeger LL, Eckardt MA, et al. Image-guided percutaneous core needle biopsy of musculoskeletal tumors in children. J Pediatr Hematol Oncol 2014;36:337–341.
22. Puri A, Shingade VU, Agarwal MG, et al. CT-guided percutaneous core needle biopsy in deep seated musculoskeletal lesions: a prospective study of 128 cases. Skeletal Radiol 2006;35:138–143.
23. Black JO, Al-Ibraheemi A, Arnold MA, et al. The pathologic diagnosis of pediatric soft tissue tumors in the era of molecular medicine: the Sarcoma Pediatric Pathology Research Interest Group perspective. Arch Pathol Lab Med 2024;148:107–116.
24. Wu JS, Goldsmith JD, Horwich PJ, et al. Bone and soft-tissue lesions: what factors affect diagnostic yield of image-guided core-needle biopsy? Radiology 2008;248:962–970.
25. Patel K, Kinnear D, Quintanilla NM, et al. Optimal diagnostic yield achieved with on-site pathology evaluation of fine-needle aspiration-assisted core biopsies for pediatric osseous lesions: a single-center experience. Arch Pathol Lab Med 2017;141:678–683.
26. Ferreira FBMD, Puchnick A, Garcia DL, et al. Image-guided percutaneous needle biopsy for benign and malignant bone tumors: systematic review and meta-analysis. J Vasc Interv Radiol 2023;34:623–632.e2.
27. Barthelemy F, Woods JD, Nieves-Rodriguez S, et al. A well-tolerated core needle muscle biopsy process suitable for children and adults. Muscle Nerve 2020;62:688–698.
28. Hesh CA, Gill AE. Percutaneous core needle biopsy: considerations in the pediatric patient. Tech Vasc Interv Radiol 2021;24:100779.
29. Mohr Z, Hirche C, Klein T, et al. Vacuum-assisted minimally invasive biopsy of soft-tissue tumors. J Bone Joint Surg Am 2012;94:103–109.
30. Kraft AO. Specimen acquisition: ROSEs, gardeners, and gatekeepers. Cancer Cytopathol 2017;125:449–454.
31. National Cancer Institute. NCI best practices for biospecimen resources. Accessed May 1, 2024. Available at: https://biospecimens.cancer.gov/bestpractices/2016-NCIBestPractices.pdf
32. Mattar M, McCarthy CR, Kulick AR, et al. Establishing and maintaining an extensive library of patient-derived xenograft models. Front Oncol 2018;8:19.
33. Miquelestorena-Standley E, Jourdan ML, Collin C, et al. Effect of decalcification protocols on immunohistochemistry and molecular analyses of bone samples. Mod Pathol 2020;33:1505–1517.
34. Duncan I, Danziger N, Duncan D, et al. Acid-based decalcification methods compromise genomic profiling from DNA and RNA. Blood 2019;134:4659.
35. Levit LA, Peppercorn JM, Tam AL, et al. Ethical framework for including research biopsies in oncology clinical trials: American Society of Clinical Oncology research statement. J Clin Oncol 2019;37:2368–2377.
36. Merker JD, Oxnard GR, Compton C, et al. Circulating tumor DNA analysis in patients with cancer: American Society of Clinical Oncology and College of American Pathologists joint review. J Clin Oncol 2018;36:1631–1641.