Are Biopsy Tracts a Concern for Seeding and Local Recurrence in Sarcomas?

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Abstract

Background A biopsy is the final step in the diagnosis of sarcomas. Complete resection of the biopsy tract traditionally has been recommended in musculoskeletal oncology guidelines, as that tract is considered potentially seeded with tumor cells. However, to our knowledge, the frequency and implications of contamination of the biopsy tract—specifically with respect to the likelihood of local

recurrence—and the factors that affect cell seeding are not well described.

Questions/purposes We asked: (1) How often are biopsy tracts contaminated with pathologically detectable tumor cells at the time of tumor resection? (2) What factors, in particular biopsy type (open versus percutaneous), are associated with tumoral seeding? (3) Is biopsy tract contamination associated with local recurrence?

Methods This is a retrospective study of a database with patient data collected from a single center between 2000 and 2013. We treated 221 patients with sarcomas. A total of 27 patients (12%) were excluded and 14 (6%) were lost to followup. One hundred eighty patients finally were included in the analysis who either had biopsies at our center (112) or biopsies at outside institutions (68). Of those performed at our center, 15 (13%) were open and 97 (87%) were percutaneous; of those at outside centers, those numbers were 47 (69%) and 21 (31%) respectively. Median followup was 40 months (range, 24–152 months). During the study period, we generally performed percutaneous biopsies as a standard practice for the diagnosis of bone and soft tissue sarcomas and open biopsies were done when the percutaneous procedure failed to provide a histologic characterization. The mean age of the population was 48 years (range, 7–87 years); 60% were male; 42% had bone sarcomas. Nineteen patients had preoperative radiotherapy and 56 had postoperative radiotherapy. Fifty-seven patients received neoadjuvant chemotherapy and 73 had

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Each author certifies that his or her institution has or waived for the human protocol for this investigation and that all investigations were conducted in conformity with ethical and confidentiality principles of research.

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adjuvant chemotherapy. We determined what proportion of biopsy tracts were contaminated by pathologic analysis of the biopsy tract specimen; during the period in question, our routine practice was to excise the biopsy tract whenever possible at the time of the definitive resection. Using the logistic regression test and Mantel-Haenszel test, we compared open with percutaneous biopsies in terms of the proportion of those that were contaminated at our site and for outside referral biopsies separately, because we do not assume the level of expertise was the same (our site is a referral tumor center). We compared the local recurrence-free survival between patients with and without contamination and between open and percutaneous biopsies using the Kaplan Meier test, again separating those performed at our site from those referred for purposes of this analysis.

Results Twenty-one of 180 biopsy tracts were contaminated (12%). Twenty of 62 (32%) of the open biopsies and one of 118 (0.8%) of the percutaneous core needle biopsies had cell seeding (odds ratio [OR], 56; 95% CI, 7–428; $p < 0.001$). One of 97 (1%) percutaneous biopsies performed in our center, and none of the 21 (0%) percutaneous biopsies performed in other centers had contaminated biopsy tracts ($p = 0.047$). Two of 15 (13%) open biopsies performed at our center and 18 of 41 (38%) open biopsies performed at other centers had contaminated biopsy tracts (OR, 4; 95% CI, 1–7; $p = 0.001$). Four of 74 (5%) bone sarcomas and 18 of 106 (17%) soft tissue sarcomas had biopsy tract contamination (OR, 3; 95% CI, 1–10; $p = 0.023$). The local recurrence-free survival was longer for patients without contaminated tracts (mean, 107 months; 95% CI, 74–141 months) than for those with biopsy tract seeding (mean, 11 months; 95% CI, 1–20 months; $p < 0.001$).

Conclusions Open biopsies were associated with an increased risk of tumoral seeding of the biopsy site, and tumoral seeding was associated with an increased risk of local recurrence. However, it is possible that other factors, such as increased complexity of the tumor or a difficult location, influenced the decision to obtain an open biopsy. Even so, based on these results, we believe that higher risk of local recurrence may be caused by an incomplete biopsy tract resection. In our opinion, the percutaneous biopsy with neoadjuvant or adjuvant therapy is the preferred method of biopsy at our center.

Level of Evidence Level III, therapeutic study.

have recommended contamination of a biopsy tract as an indication for amputation when the biopsy tract is impossible to include in the surgical approach, affects the neurovascular bundle, or wide margins are not possible [14, 15]. Oncologic surgeons now use different types of biopsies, which create different kinds of biopsy tracts. The scar from a percutaneous core needle biopsy is a puncture of the surface, but in deep tissues it has a tridimensional shape that is difficult to predict reliably. The accuracy of core needle biopsies ranges from 77% to 97% in published series [16, 17, 22, 28–34]. It is excellent for bone sarcomas (77%–96%) and possibly less so for soft tissue sarcomas (76%–78%). However, the sensitivity in soft tissue sarcomas improves with image-guided core needle biopsy (special contrast-enhanced ultrasound) and with a pathologist with extensive experience working with limited samples [7, 17, 19]. The accuracy of open biopsies is close to 100% in some reported series [1, 17, 21], but the cost and the possibility of complications are higher [1, 2, 17, 21]. They are used more commonly in centers without experienced sarcoma experts [1, 2, 17]. An open biopsy produces a longitudinal scar that may be variable in morphologic features and direction.

Some authors have suggested that complete resection of the percutaneous biopsy tract rarely is achieved [10]. Although resection of the biopsy tract is recommended by numerous authors [5, 13, 14], neither we nor Oliveira et al. [20] could find a study that definitively confirmed the frequency of biopsy tract seeding or that there is increased risk of local recurrence when the biopsy tract is not resected.

We performed a pathologic analysis of the musculoskeletal sarcoma biopsy tracts of patients treated at our institution during 2000 to 2013. The main objective was to identify factors associated with contamination. We define “contaminated biopsy tracts” or “biopsy tract seeding” as those in which the pathologists actually find tumor (viable or necrotic) cell nests anywhere in the resected biopsy tract of the main tumor at the time of definitive resection. The secondary objective was to detect the relevance of tumor seeding in local recurrence-free survival of the series. We asked the following questions: (1) How often are biopsy tracts contaminated with pathologically detectable tumor cells at the time of tumor resection? (2) What factors, in particular biopsy type (open versus percutaneous), are associated with tumoral seeding? (3) Is biopsy tract contamination associated with local recurrence?

Introduction

It commonly is assumed in musculoskeletal oncology [3, 5, 14] that the biopsy tract must be resected together with the tumor in limb-sparing surgery for sarcomas. Some authors

Patients and Methods

After institutional ethical board approval, we performed a study in which we included 221 patients with bone and soft tissue sarcomas treated at our tertiary referral

musculoskeletal oncology center from March 2000 until March 2013. This is a retrospective observational study of prospectively collected data of the medical records, surgical protocols, radiologic studies, and pathologist reports. Forty-one of the 221 patients were excluded; 14 were lost to followup and therefore not included for analysis. The samples of four patients were ulcerated or infected and therefore these four patients were not eligible for inclusion in the study. Twenty-three biopsy tracts were not resected because either the biopsy was performed at another hospital and we were not able to find the scar, or the scar resection would have led to an ablative surgery and the patient rejected amputation. Apart from these cases just mentioned, during this period, our practice was to excise and histologically examine the biopsy tract at the time of definitive resection of the sarcoma. We included all the patients with a diagnosed musculoskeletal sarcoma who had their final surgery at our hospital even if the biopsy had been performed elsewhere before the referral (Table 1). Median followup was 40 months (range, 24–152 months).

During this study period, of the biopsies performed at our center and included in this series, 15 of 112 (13%) were open and 97 of 112 (87 %) were percutaneous; of those at outside centers, those numbers were 47 of 68 (69%) and 21 of 68 (31%), respectively. During the study period, we generally performed percutaneous biopsies as a standard practice for the diagnosis of bone and soft tissue sarcomas and open biopsies when the percutaneous procedure failed to provide a histologic characterization. We compared open with percutaneous biopsies, in terms of the proportion of those that were contaminated, for our institution and for outside referral biopsies separately, as we do not assume the level of expertise was the same (our institution is a referral tumor center). We compared local recurrence-free survival between patients with and without contamination and between open and percutaneous biopsies, again separating those performed at our institution from those referred, for purposes of this analysis.

If the patient was referred from another center, our pathology team reviewed the samples to determine if a new biopsy was needed. We routinely performed core needle biopsies as standard procedure for histologic diagnosis of bone and soft tissue tumors. Two senior musculoskeletal radiologists performed all the biopsies (DB-T, MTV). Bone biopsies were performed in most cases using CT guidance, but also targeted to the periphery of the tumor. In some cases with cortical breakout, ultrasound guidance was used. Bone biopsy was performed using a T-LokTM ultrasound-guided (Argon Medical Devices, Plano, TX, USA) or a BonoplyTM (AprioMed, Uppsala, Sweden) coaxial needle.

For soft tissue malignancies biopsy, we use ultrasound Doppler color guidance targeted to the most vascularized area and in some cases we also use contrast-enhanced

Table 1. Baseline epidemiologic population characteristics

Characteristic	Totals
Age of patients (years)	Median, 48.17 (range, 7–87 years)
Gender	106 males (60%); 74 females (40%)
Location of tumor	
Upper limbs	20%
Lower limbs	70%
Extracompartmental	8%
Axial skeleton	2%
Laterality	
Right	52%
Left	47%
Axial	2%
Bone sarcomas	42%
Soft tissue sarcomas	58%
Biopsies performed at authors' institution	62%
Biopsies done at other centers	38%
Biopsies done at both	3%
Percutaneous biopsy	66%
Open biopsy	26%
Both	9%
Diagnosis	
Chondrosarcoma	15%
Leiomyosarcoma	8%
Liposarcoma	14%
Osteosarcoma	19%
Others	15%
Primitive neuroectodermal tumor/Ewing	4%
Pleomorphic sarcoma	16%
Synovial sarcoma	14%
Grade	
1	9%
2	1%
3	66%
4	4%
Margins	
Marginal resection	19%
Wide resection	79%
Other	2%

ultrasound. In a soft tissue sarcoma, this area usually corresponds to the periphery of the tumor. The majority of soft tissue biopsies were performed using a coaxial technique, that is, by inserting an introducer or sheath to the edge of the lesion or just inside the lesion and then advancing a cutting needle through the introducer or sheath into the lesion to obtain the biopsy samples. From 2000 to 2010 a

TruCore-II™ (Argon Medical Devices) was used; since 2010 we use the Speedybell coaxial biopsy system (Biopsybell Srl, Mirandola, Italy) to infiltrate the periosteal layer.

Needle size depends on the difficulty in obtaining a representative tumor sample (myxoid, heterogeneous tumors, or necrotic tumors), but usually a 16-gauge needle is used to get thick cylinders of tumor and only two or three passes are made. Samples usually were placed in a 10% formalin solution and hand-delivered to the pathology unit for analysis. However, each case is reviewed during weekly meetings before the biopsy to determine the number of samples, the approach [12], and the delivery (fresh or formalin). At our center, the same oncologic orthopaedic team performing surgery on the patient also performs all the open biopsies when they are needed. The patients who are referred are discussed during a weekly meeting [9, 27]; if the open biopsy was performed in line with the predicted incision and the surgeon was aware of the possible neoplastic nature of the lesion, the biopsy was considered to have been correctly performed. If the open biopsy is not considered appropriate (ie, not performed correctly) and limb-sparing surgery is still possible, we plan a wider soft tissue resection (often together with plastic surgery).

Nineteen patients had preoperative radiotherapy and 56 had postoperative radiotherapy. We considered preoperative radiotherapy as a possible bias in the adjusted analysis because some authors [4] point to radiotherapy and chemotherapy as being probable influences to biopsy tract contamination. They did not resect the core needle biopsy in 59 patients with neoadjuvant therapies.

The biopsy tract was resected together with the tumor following the standard of treatment for a wide oncologic resection. We could identify the scar of the percutaneous biopsy tract because it was labeled with India ink in the dermis. In the final surgery, we use the images (ultrasound, CT-guided biopsies) if they are available and manual palpation to include the deep tract as completely as possible. The margins of the main tumor resection were evaluated. We consider the margins wide when the distance between the closest border and the pseudocapsule is wider than 1 cm or 2 mm with anatomic barriers (periosteal layer, epineurium, fascia, or vessel adventitia). The sample is sent to the same pathologists (FLB, JJPK) and reviewed independently macro- and microscopically (Fig. 1). The technique used in pathologic analysis consists of cutting 2-mm parallel blocks of all tissue surrounding the macroscopic scar and the 2-mm blocks were cut in 4- μ thick sections, stained with hematoxylin and eosin, and the pathologist specifically searched for a tumoral cell nest in the sample. Immunohistochemical tests were used if necessary for histologic characterization. The tumor stage is recorded according to the American Joint Committee on

Fig. 1 This microscopic image of a biopsy tract shows an osteosarcoma cell nest (Stain, hematoxylin & eosin; original magnification, $\times 10$).

Cancer 2010 and Enneking classification for bone sarcomas [8]. The patients were evaluated with MRI and CT every 3 months for 2 years, then every 6 months until 5 years, and finally annually. Bone sarcomas with prosthesis reconstruction also are evaluated with a bone scan and ultrasonography every 6 months. Median followup was 40 months (range, 24–152 months).

Descriptive statistics were analyzed. The statistical analysis was performed using SPSS software, Version 16.0 (SPSS, Chicago, IL, USA). We used a univariate chi-square test for each variable. We analyzed separately, with the Mantel-Haenszel test, contamination of the open and percutaneous biopsies performed at our center and those performed in other hospitals. We performed a multivariable analysis using the logistic regression proportional model to detect the factors that can affect biopsy tract contamination. We also compared the local recurrence-free survival in patients with and without biopsy tract seeding using the Kaplan-Meier method using the log-rank test. Statistical significance was accepted as a probability less than 0.05.

Results

Twenty-one of the 180 samples (12%) were contaminated, with macroscopically detectable cell nests in two and microscopically detectable cell nests in 19. One of the 21 contaminated biopsy tracts was from a percutaneous biopsy (0.8% contamination rate). One of 97 (1%) percutaneous biopsies performed at our center and none of the 21 (0%) percutaneous biopsies performed at other centers had tumoral cell seeding ($p = 0.047$). This one patient with a contaminated needle biopsy had a Grade 1 chondrosarcoma and underwent extended intralesional resection with high-

Table 2. Logistic regression proportional multivariable analysis

Variable	Odds ratio (95% CI)	p Value
Open biopsies	51,047 (5389–483,506)	< 0.001
Percutaneous biopsies		
Bone sarcomas	5907 (1659–21,035)	0.006
Soft tissue sarcomas		
Biopsy performed at: other hospital	2070 (448–9555)	0.351
our institution		
Radiotherapy or radiotherapy + chemotherapy	*	0.094
Chemotherapy alone or no adjuvant therapies		

*Radiotherapy odds ratio is not valuable.

speed burring and phenol. The biopsy was performed to confirm that this patient had a low-grade, rather than high-grade, chondrosarcoma. The patient has 4 years of followup and no evidence of disease. The other 20 of 21 contaminated biopsy tracts were from open biopsies (32%) (mean, 55 months; 95% CI, (7–428 months; $p < 0.001$).

Open biopsies were more likely to be contaminated than percutaneous biopsies, among the patients whose biopsies we performed and those whose biopsies were performed at referral centers. Two of 15 (13%) open biopsies performed at our center and 18 of 41(38%) open biopsies performed at other centers had contaminated biopsy tracts ($p = 0.001$; odds ratio [OR], 4; 95% CI, 1–7). Other factors associated with an increased risk of biopsy tract contamination were soft tissue sarcoma and biopsies performed at other centers before the patient was referred to us (Appendix 1. Supplemental material is available with the online version of *CORR*[®]). Four of 74 (5%) bone sarcomas and 18 of 106 (17%) soft tissue sarcomas had biopsy tract contamination (OR, 3; 95% CI, 1–10; $p = 0.023$). We could not find differences in the following variables: age, sex, location, grade, stage, neoadjuvant chemotherapy, number of percutaneous core needle biopsies, tumor resection margins, or time from the biopsy until definitive surgery. None of the patients with preoperative radiotherapy had a contaminated biopsy tract. The center where the biopsy initially was performed was not a factor for biopsy tract contamination in the multivariable analysis. However, the bone or soft tissue sarcoma and the type of biopsy are independent factors (Table 2).

The local recurrence-free survival was longer for patients without contaminated tracts (mean, 107 months; 95% CI, 74–141 months) than in those with biopsy tract seeding (mean, 11 months; 95% CI, 1–20 months; $p < 0.001$) (Fig. 2). The local recurrence-free survival did not show statistical differences between patients with

Fig. 2 The Kaplan-Meier curve shows the survival without local recurrence in patients with and without biopsy tract contamination. Blue line = no contaminated biopsy tracts; green line = contaminated biopsy tracts.

percutaneous and open biopsies, neither at our center nor in patients whose biopsies were performed at another hospital. With the numbers we had, we could not show a difference in local recurrence-free survival in percutaneous biopsies (mean, 102 months; SD, 12 months; 95% CI, 77–127 months) compared with open biopsies (mean, 98 months; SD, 10 months; 95% CI, 80–117 months; $p = 0.252$) (Fig. 3). We also did not observe a difference in local recurrence-free survival between patients who had their biopsy at our hospital or at other hospitals.

Discussion

The biopsy is a key step in the diagnosis of sarcomas as incorrectly performed biopsies might result in more ablative final operations or might compromise the oncologic resection [14, 15]. Guidelines recommend biopsy tract resection in the definitive surgery to achieve complete elimination of malignant cells [5, 13, 14]. To our knowledge, and as reported by Oliveira et al. [20], contamination of the tract and the factors that can affect seeding have not been reported. We detected contamination with tumoral cells in 12% of the biopsy tracts. Tumoral cell seeding increased in open biopsies performed at referral centers or at our center. The local recurrence-free survival was longer in patients without contamination of the biopsy tract.

There are limitations in this study. First, the retrospective design may have resulted in some selection bias in terms of which patients received open biopsies and which

Fig. 3 The Kaplan-Meier curve shows the survival without local recurrence in patients with percutaneous and open biopsy tract contamination. Blue line = percutaneous biopsy tracts; green line = open biopsy tracts.

patients underwent percutaneous biopsies. At our center, open biopsies were performed when we did not achieve a diagnosis after at least two percutaneous procedures. Thirty-eight percent of the biopsies were performed elsewhere, consequently we cannot confirm the biopsy technique in those patients. We obviously do not know exactly why patients whose biopsies were performed at outside centers received an open or a percutaneous biopsy, but it is possible that patients with more-complicated or less-accessible tumors underwent open biopsies, and this could have accounted for some of the increased contamination we observed with open biopsies. Even so, the open biopsies performed at our center also showed a higher contamination rate and the multivariate analysis showed that the type of biopsy procedure (open or percutaneous) was associated with the contamination, whereas the center where the biopsy was performed did not correspond to the contamination rate. Second, there was a relatively small and heterogeneous group of patients with disparate diagnoses that prevents statistical analysis of the effect of the histologic diagnosis, and we also looked at bone and soft tissue tumors together. In this series, we observed higher contamination in soft tissue sarcomas compared with bone sarcomas. Nevertheless, a larger study comprised of all bone tumors or all soft tissue tumors might provide different results. Finally, we did not have enough patients who received preoperative radiotherapy to determine if it might have had an effect on contamination of the biopsy tract. However none of the patients who received preoperative

radiation had contamination of the biopsy tract. This variable should be cleared in further study.

The rate of biopsy tract seeding in our series was 12%. There are some published cases of tumor recurrence in the path of no resected or incompletely resected percutaneous and open biopsy tracts [6, 9, 24, 35]. Ribeiro et al. [23] reported contamination of 32% of the biopsy tracts in the largest series with 25 bone and soft tissue sarcomas.

Our study shows that the possibility of contamination is present in open and percutaneous biopsies, but the likelihood is greater with open than with percutaneous procedures. This difference is independent of the hospital where the biopsy is performed, as the possibility remained when we compared percutaneous and open procedures done at our center and at other centers. Other studies also show a high risk of contamination of open biopsies [18, 23]. Saghieh et al. [25] reported 10 cases in pediatric patients in which the percutaneous core needle biopsy tract was not resected and the patients did not have any recurrence. Li et al. [12] suggested decreasing contamination of the percutaneous biopsy tracts in patients with osteosarcoma by using Adriamycin sponges.

When we looked for other factors associated with contamination, we found that soft tissue sarcomas showed higher biopsy tract contamination than bone sarcomas. This factor may be related to the biopsy technique or to the histologic characteristic of each tumor that could not be analyzed separately in this study. Although in our study this factor did not reach statistical significance, other studies have indicated that radiotherapy may decrease tumor tissue contamination near the biopsy pathway [6, 18, 26]. Nevertheless more studies would be necessary to define the influence of adjuvant therapies (chemotherapy/radiotherapy). Binitie et al. [4] did not address the relative effect of each of the adjuvant treatments or the effect of these treatments in open biopsy tracts. Moreover, they did not compare the local recurrence rate in their series with that of patients without resected biopsies who did not receive any adjuvant therapy.

In our study, the local recurrence-free survival was higher in patients with contaminated biopsy tracts, although with the numbers we had, we could not show a difference in local recurrence-free survival in patients who had percutaneous compared with open biopsies. We did not show a difference, when considered separately, biopsies performed at our institution and at other hospitals. It may be attributable to the low number of open biopsies performed at our center, but local recurrence is a complex event that can be affected by other factors, therefore more specific studies are needed to clarify these results.

Percutaneous core needle biopsies can cause contamination in a resected biopsy tract, but the percentage of

contaminated samples was much lower in our series than with open biopsy, at our center and among patients referred to us from outside centers for care. We found that patients with a contaminated biopsy tract has shorter local recurrence-free survival than those with a noncontaminated biopsy tract. This might indicate that wider margins are needed around open biopsy tracts than was done in our patients. Resecting the biopsy tract completely may be a difficult task because its tridimensional shape may be difficult to predict reliably during the definitive surgery. Jalgaonkar et al. [11] reported that the biopsy tract was impossible to find after 3 months in 22 of 45 patients (48.8%) when they did not mark it with India ink. With open biopsies, the tridimensional shape of the scar is not always as expected, and the hematoma, which may have tumoral cells, can reach distant tissues. In our opinion, although we can ascertain this proposition, we suspect we are not able to completely resect a substantial number of biopsy pathways.

The treating surgeon should assess the biopsy tract carefully when planning definitive resection. More study is needed to assess the potential benefits of adjuvant treatments in preventing local recurrence, especially in patients who have had an open biopsy. Percutaneous core needle biopsies appear to be a safer approach to establishing the diagnosis of a musculoskeletal neoplasm.

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