Ultrastructural Diagnosis of Soft Tissue Tumors: Avoiding Pitfalls

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Introduction

The use of immunohistochemistry and, more recently, molecular methods has seen a sharp reduction of the role of electron microscopy in the diagnosis of soft tissue sarcomas (1,2). Ultrastructural analysis, however, does continue to play a useful role in selected cases. The focus in this presentation is on adult soft tissue tumors; Dr. John Hicks will follow with a presentation on avoiding pitfalls in the ultrastructural diagnosis of pediatric soft tissue tumors.

Before moving to the issue of pitfalls, consideration should be given to the current role of electron microscopy in diagnosis of soft tissue tumors. One of the most obvious types of cases of adult soft tissue tumors for which electron microscopy can prove useful is in the definitive classification of malignant peripheral nerve sheath tumors. Only 50 to 70 percent of these cases will demonstrate any S-100 staining, yet the ultrastructure is generally diagnostic (1). Without ultrastructural study, many of these cases fall in an unclassified status or are classified as malignant fibrous histiocytomas. Another obvious example is alveolar soft part sarcoma cases not identified by light microscopy.

Other instances include cases where the sample size is too small to permit adequate immunohistochemical study, including fine needle aspirates (3). Additional examples are poorly differentiated lesions with unclear immunohistochemical results in which the goal is to distinguish a sarcoma from a sarcomatoid carcinoma (3,4).

Many other applications for electron microscopy in the evaluation of adult sarcomas arise, but generally in a small subset of any given entity or in some rare variants (1,4). These include:

1. Epithelioid fibrosarcoma (which may be EMA positive) and some typical cases of fibrosarcoma (which have no specific markers).

2. Low grade myofibroblastic sarcoma (for definitive distinction from a smooth muscle tumor).

3. Smooth muscle tumors with negative or very focal immunohistochemical staining.
4. Very rare cases of liposarcoma, particularly pleomorphic liposarcoma, when the diagnosis is in doubt (e.g., distinguishing the epithelioid variant of pleomorphic liposarcoma from adrenal cortical carcinoma (5)).

5. Angiosarcomas lacking definitive immunohistochemical findings.

6. Translocation-negative monophasic synovial sarcomas.

7. Epithelioid sarcoma vs. epithelioid angiosarcoma with a CD34+, CD31-, cytokeratin+ immunophenotype.

8. Malignant perineurial cell tumors (rare).

**Pitfalls**

Within this selected set of cases for which electron microscopy remains highly useful, however, certain pitfalls do arise. And as electron microscopy is less frequently used for tumor analysis than in the past, individuals performing the electron microscopy are more and more likely to have primary ultrastructural expertise in an area other than tumors, and awareness of these relatively few pitfalls may prove helpful.

**Malignant Peripheral Nerve Sheath Tumor vs. Monophasic Synovial Sarcoma**

Again, one of the most common applications for electron microscopy in the evaluation of adult soft tissue tumors may be in definitive classification of malignant peripheral nerve sheath tumors. These tumors may be recognized ultrastructurally by interdigitating cell processes and discontinuous external lamina. While the pattern is fairly specific, it can be mimicked. Monophasic synovial sarcoma classically features bipolar tapering processes, but cases can demonstrate similar interdigitating cell processes. Further, while monophasic synovial sarcoma generally lacks a true external lamina, a flocculent amorphous substance is characteristically identified between cells. As this material presses against the plasma membrane, it can closely mimic a discontinuous external lamina. In the absence of rudimentary epithelial elements – or failing to look for them given what appears to be a classical pattern of a malignant peripheral nerve sheath tumor – one may conclusively classify the lesion as a malignant peripheral nerve sheath tumor ultrastructurally and fail to explore the classic translocation. A key to avoiding this pitfall is the extent of the intercellular amorphous material, though this is a subjective measure. Fortunately, most cases of monophasic synovial sarcoma will not manifest the interdigitating pattern of cell processes.
Crystal-Deficient Alveolar Soft Part Sarcoma

Another ultrastructural pattern which is usually entirely straightforward is that of alveolar soft part sarcoma. Indeed, these tumors sometimes exhibit intracytoplasmic, PAS-positive crystals which are several microns in maximum dimension and readily seen by light microscopy; such cases require no ultrastructural study. Other cases represent a classic application of electron microscopy for tumor analysis, as large rhomboidal crystals with a 10 nm periodicity permit a specific diagnosis. The pitfall is that, despite this classical description, some cases of alveolar soft part sarcoma lack this feature (6). In these, the key feature instead is the presence of numerous secretory granules (as well as prominent rough endoplasmic reticulum and frequent mitochondria). With luck, one may still identify the typical early crystallization within some of the granules, but in some cases, even this may not be found (7).

Smooth Muscle Tumors with Few Filaments

As noted in the list of applications of electron microscopy for soft tissue tumors, occasionally a diagnosis of a smooth muscle tumor may be made ultrastructurally in a case in which immunohistochemistry for actin was negative or too limited to be conclusive. The pitfall in this case is to avoid falsely excluding a smooth muscle tumor ultrastructurally. While some cases will have numerous filaments and dense bodies as well as extensive external lamina and pinocytotic vesicles, such cases will tend to give reliable immunohistochemical results. For the preselected cases which will present for electron microscopy, only scanty filaments may be present, usually peripherally, especially if the lesion is epithelioid (8,9). The presence of numerous mitochondria reinforces the concept of smooth muscle differentiation and should prompt further search for filaments. And in contrast to myofibroblasts, rough endoplasmic reticulum should be scanty.

Myofibroblasts

By immunohistochemistry, myofibroblasts cannot be reliably distinguished from smooth muscle cells. Electron microscopy, then, can be highly valuable for definitive classification of low grade myofibrosarcoma. Identifying tumor cells with myofibroblastic differentiation, however, is not specific for a diagnosis of myofibrosarcoma; myofibroblastic differentiation is also seen in tumor cells of fibromatosis, fibroma of tendon sheath, myxofibrosarcoma, malignant fibrous histiocytoma, and other tumors (7). Identifying myofibroblasts in a tumor, though, does not indicate that it is one of these lesions. Myofibroblasts are very common reactive cells in a large variety of tumors, both soft tissue and epithelial (10). Further, the nuclei of reactive myofibroblasts are not readily distinguished in many cases from neoplastic ones. Care must be taken to establish that the cells are representative of the lesion.
Lipid

One might think that ultrastructure would never have a role in the diagnosis of liposarcoma, and indeed the use of electron microscopy in the diagnosis of liposarcoma is quite rare. Some cases of pleomorphic liposarcoma, however, can express epithelial or myogenic markers, and in such cases, electron microscopy may prove useful for definitive diagnosis (4,5). It may also prove useful in some cases in which vacuolated cells are seen, but no definite lipoblasts are identified (4). In this latter example, unless the vacuoles are explained, negative results are meaningless given the sampling issues. And finding lipid droplets in tumor cells is a highly nonspecific finding, as lipid can accumulate in tumor cells damaged by intrinsic anoxia or therapy, and neoplastic cells infiltrating adipose tissue may contain lipid from phagocytosis of the fat (7).

Desmosomes

Very infrequently, electron microscopy is undertaken simply to determine whether an anaplastic lesion is a sarcoma or a carcinoma, such as when immunohistochemistry is unsatisfactory or inconclusive or insufficient tissue is present for adequate analysis. Such studies are often frustrating, as one might expect. A positive finding that generally indicates carcinoma rather than sarcoma is the detection of desmosomes. One should bear in mind, however, that desmosomes may be found in synovial sarcoma and epithelioid sarcoma (7).

Conclusion

While electron microscopy is much less commonly employed in the diagnosis of adult soft tissue tumors than previously, in a subset of these lesions, ultrastructural study remains highly beneficial for diagnosis. Relatively few pitfalls exist. Knowledge of the current applications of electron microscopy for the diagnosis of these lesions and of the pitfalls in those examinations permits optimal utilization of the electron microscopy laboratory in contributing to the care of these patients.
References


