Original Article
Myxoid and reticular angiomatoid fibrous histiocytoma: a case confirmed by fluorescence in situ hybridization analysis for EWSR1 rearrangement

Li-Xiong Ying, Xiao-Dong Teng

Department of Pathology, The First Affiliated Hospital, Zhejiang University School of Medicine, Hangzhou, P. R. China

Abstract: Angiomatoid fibrous histiocytoma (AFH) is a tumor of intermediate malignancy and undefined lineage, mostly arising in the extremities of young patients. However, AFH may rarely display uncommon clinical and morphologic features, such as older age at presentation, occurrence outside somatic soft tissues and alterations in the architectural patterns, stromal matrix and cytomorphology, causinga great diagnostic challenge for practicing pathologists. Herein, we present a case of AFH with unusual histologic features arising in the right hip of a 37-year-old man. The tumor exhibited a reticular growth pattern and myxoid stroma mimicking myoepithelioma, extraskeletal myxoid chondrosarcoma, or myxoid liposarcoma. The tumor cells focally expressed desmin with a dendritic-like cell process staining pattern and CD68. Fluorescence in situ hybridization analysis confirmed the rearrangement of the EWSR1 gene. This report further expands the clinicopathologic spectrum of AFH and underscores the value of integrating morphologic, immunophenotypic, and molecular findings in the identification of its unusual morphologic variants.

Keywords: Angiomatoid fibrous histiocytoma, AFH, myxoid, reticular, FISH, differential diagnosis

Introduction

Angiomatoid fibrous histiocytoma (AFH), also referred as angiomatoid "malignant fibrous histiocytoma", is an uncommon soft tissue tumor of intermediate malignancy and undefined lineage, most commonly occurring in the deep dermis or subcutis of the extremities in children and young patients [1-3]. Histologically it is characterized by (1) Multinodular proliferation of oval histiocytoid to spindly cells with syncytial growth, forming sheets, whorls or fascicular growth patterns, (2) Central pseudoangiomatous spaces lined by tumor cells rather than true endothelium, with stromal hemorrhage or hemosiderin deposition, and (3) A thick fibrous pseudocapsule and pericapsular lymphoplasmacytic cuffing in variable proportions [1-3]. The tumor cells of AFH are generally bland-appearing with a low rate of mitotic activity, and only occasionally present with hyperchromatic nuclei, significant nuclear atypia, or increased mitotic activity; however, these features have not been shown to be associated with aggressive outcome [4]. The diagnosis of AFH is usually straightforward when it exhibits a classical histology. However, AFH may rarely display uncommon clinical and morphologic features, such as older age at presentation, occurrence outside somatic soft tissues, and alterations in the architectural patterns, stromal matrix, and cytomorphology. This is a great diagnostic challenge for practicing pathologists [4-11]. This is further complicated by the lack of diagnostic immunohistochemical markers for AFH. Although epithelial membrane antigen (EMA), desmin, CD99, and CD68 have been reported to be variably positive in around 50%-75% of cases, these are not specific for this entity [2, 3, 5].

Recently, molecular genetic analyses from several studies have clarified that AFH is characterized by EWSR1 rearrangement, reflecting the *EWSR1-CREB1* fusion gene resulting from t(2;22)(q33;q12) in the majority of cases and less frequently by *EWSR1-ATF1* from t(12;22)(q13;q12) or *FUS-ATF1* from t(12;16)(q13;p11) [12-15]. Therefore, molecular testing for *EWSR1* rearrangement is of great diagnostic utility for AFH, especially in the cases of atypical clinical
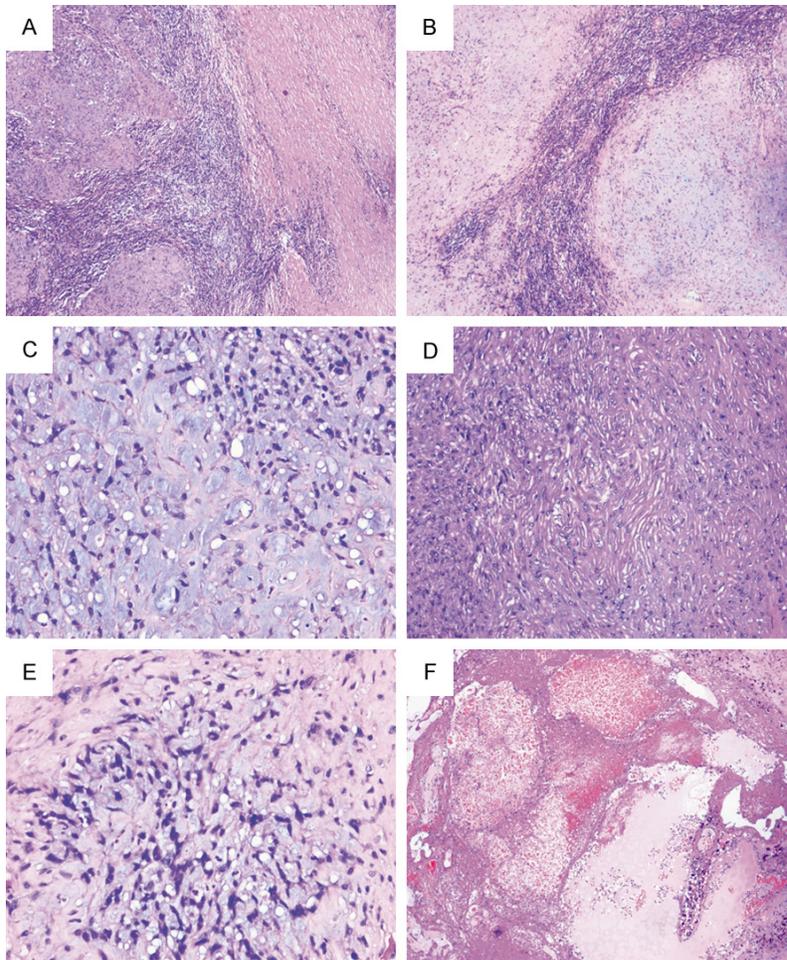
Figure 1. Microscopic features of myxoid AFH. A: Tumor is circumscribed, surrounded by a fibrotic pseudocapsule and shows a dense peripheral lymphocytic infiltrate. B: Nodular growth pattern with reticular and intersecting strands and cords of cells set in a variably myxoid to collagenous stroma. C: Spindle cells are oval to slightly elongated with indistinct borders. The cytoplasm is moderate, pale to faintly eosinophilic, with frequently intracytoplasmic paranuclear vacuole formation. D: Minor areas consisting of densely packed histiocytoid cells are noted. E: Foci of pleomorphic and bizarre nuclei with slight hyperchromatin and prominent nucleoli are observed. F: The cystic portion shows large areas of stromal hemorrhage, fibrosis, and hemosiderin deposition.

nance imaging analyses revealed a 54 mm × 52 mm, round to oval, heterogeneously solid and cystic mass located in the spatium intermusculare of the right hip, posteriorly to the lesser trochanter of the right femur. The mass was radiologically well-demarcated without evidence of erosion into cortical bone or extension into surrounding muscle tissues. The patient's medical history was unremarkable., Physical examination and computed tomography scan did not reveal tumor elsewhere in the body. With the suspicion of a giant cell tumor of tendon sheath, a needle core biopsy of the mass was performed, a descriptive diagnosis was rendered, and a myofibroblastic origin, low-grade tumor or tumor-like lesion was suspected on the basis of the spindly morphology and the lack of any specific differentiation of the lesional cells. A subsequent total tumorectomy was performed and the patient was well with no evidence of recurrence or metastasis at a follow-up of 30 months.

and histologic manifestation [6-11]. Herein, we present an unusual case of AFH with prominent myxoid change and reticular growth pattern from the hip soft tissue of a 37-year-old woman, with molecular genetic comfirmation.

Case presentation

A 37-year-old woman presented with difficulty in bending up for 2 weeks. Routine laboratory tests revealed elevated erythrocyte sedimentation rate (92 mm/h, normal range, 0-20 mm/h). Computed tomography and magnetic reso-

The resected specimen was fixed in 10% buffered formalin. Tissue sections were routinely processed and stained with hematoxylin and eosin. Immunohistochemistry (IHC) analysis was performed using avidin-biotin-complex technique with a panel of commercially available primary antibodies to the following antigens: cytokeratin AE1/AE3 (AE1/3, Dako, Denmark), EMA (E29, Dako), cytokeratin 8/18 (Cam5.2, Dako), smooth muscle actin (SMA) (1A4, Dako), desmin (D33, Dako), melan-A (A103, Dako), HMB45 (HMB45, Dako), S100 protein (polyclonal, Dako), CD31 (JC/70A, Dako), CD34 (QBEnd/10, Dako), ERG (9FY, Biocare Medical,
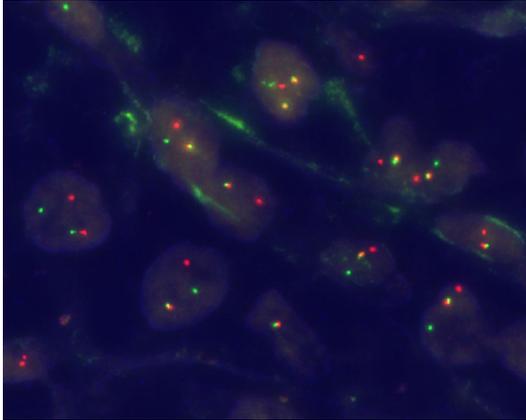
Figure 2. Molecular genetic features of myxoid AFH. EWSR1 fluorescence in situ hybridization assay showed 1 set of fusion signals and 1 set of green and red split signals, indicating evidence of a EWSR1 gene rearrangement.

USA), CD68 (KP-1, Dako), CD99 (O13, Dako), glial fibrillary acidic protein (GFAP) (polyclonal, Dako), P63 (BC4A4, Dako), CD21 (1F8, Dako), CD35 (Ber-MAC-DRC, Dako), and Ki67 (MIB-1, Dako). Appropriate positive and negative controls were run concurrently for all the markers tested. Fluorescence in situ hybridization (FISH) analysis was performed on a 4-mm-thick paraffin section according to standard protocols. *EWSR1* rearrangements were evaluated using a commercially available dual color break-apart probe (Abbott Molecular, USA), spanning the known common breakpoints of *EWSR1*. Written informed consent was obtained from the patient.

Grossly, the tumor was well-circumscribed and encapsulated, measured 6 cm in the greatest diameter. Cut surface showed a partially cystic and solid tumor of a gray-red to yellow color. The cystic wall was thick and the cystic spaces were filled with bloody fluid. Microscopic examination showed a thick, fibrous pseudocapsule accompanied by a dense peripheral lymphoplasmacytic infiltrate with occasional germinal center formation (**Figure 1A**). The solid portion consisted predominantly of a multiple nodular growth pattern with reticular and intersecting strands and cords of cells set in a variably myxoid to collagenous stroma (**Figure 1B**, **1C**). The tumor cells were oval to slightly elongated with indistinct border, the cytoplasm was moderate, pale to faint eosinophilic, with frequent intracytoplasmic paranuclear vacuole formation (**Fig-**

ure **1C**). Minor areas composed of more cellular sheets of uniform histiocyte-like cells with storiform and swirling growth patterns, were also noted at the periphery of the tumor. These areas occupied less than 20% of the tumor (**Figure 1D**). The nuclei were bland-appearing, oval to stellate with fine chromatin and indistinct nucleoli. Occasionally, pleomorphic and bizarre nuclei with slight hyperchromasia and prominent nucleoli were observed (**Figure 1E**). Mitoses were scant (2/10 high power filed), and necrosis was absent. The cystic portion were composed of large, dilated, vascular-like spaces lined by one to several layers of inconspicuous tumor cells, and large areas of stromal hemorrhage, fibrosis, and hemosiderin deposition (**Figure 1F**).

By IHC, the tumor cells focally expressed desmin with a dendritic-like cell process staining pattern and CD68. All the other markers detected were negative. Ki67 labeled less than 5% tumor cells. FISH assay of the tumor cells showed a single interphase nucleus with split red and green signals observed in more than 10% of tumor nuclei, indicating the presence of *EWSR1* gene rearrangement involving chromosome 22 (**Figure 2**).

Discussion

AFH is a neoplasm of obscure lineage of differentiation with an intermediate malignant potential. It was originally described by Enzinger [1] as a fully malignant sarcoma, namely angiomatoid "malignant fibrous histiocytoma", but has subsequently been shown to have a relatively low recurrence rate (2-12%) and a very low metastatic rate (1-5%) with only isolated cases of death owing to disease. Thus, the term 'malignant' has been dropped from the name in order to more accurately reflect the biological behavior, in the current World Health Organization classification [2]. Classically, lesions are slow growing and occur predominantly in children and young adults in the subcutaneous tissue and deep dermis of the extremities, with reports of congenital cases and in rare locations including skeletal muscle, bone, orbit, lung, vulva, retroperitoneum, ovary and mediastinum [4, 5]. Systemic symptoms including anemia, weight loss, chills and pyrexia may occur and often subside after resection. This is believed to be a cytokine-mediated phenomenon [1-3].

Histologically, AFH is typically composed of a bland spindle and/or round cell proliferation of cells with abundant eosinophilic cytoplasm forming a syncytium, often surrounded by a dense fibrous pseudocapsule and lymphoid infiltrate. However, several large series of studies have shown that AFH has a much broader range of histologic features than is widely appreciated [4, 5, 8-11]. AFH with focal myxoid change is not uncommon and has been reported to be observed in up to 14% of cases [1, 4, 5]. However, cases with prominent myxoid deposition (defined as myxoid areas occupying at least 60% of the tumor) seem to be very rare, representing less than 5% of all AFHs according to recently published the largest series of this subtype [11]. As with conventional AFH, the myxoid variant of AFH most often develops in the extremities of young adults with half arising in the upper extremity [5, 7-11]. The age distribution, median size, and slight female predominance are the same as in conventional AFH. Classic morphologic characteristics of AFH such as a fibrous pseudocapsule, peritumoral lymphoplasmacytic infiltrates, and blood-filled cystic spaces were observed in almost all of these cases of myxoid AFH [7-11]. Expression of desmin and EMA in approximately half of the cases, as well as *EWSR1* rearrangement in a subset of the cases, further reflects their close similarity to conventional AFH [11]. Metastasis in AFH in general is rare, being observed in <5% of cases, and local recurrence is reported for 2% to 12% [1-4]. Myxoid AFH seemed to be less aggressive than conventional AFH, according to the largest series reported to date Approximately 14% of such tumors recurred, but none has developed metastases [11]. Our case showed no evidence of recurrence or metastasis at a follow-up of 30 months, further providing supports to therelatively benign clinical course.

The differential diagnosisof myxoid AFH is broad and includes other soft tissue tumors with a prominent myxoid matrix, such as myoepithelioma, extraskeletal myxoid chondrosarcoma, and myxoid liposarcoma [11]. The diagnosis can only be made after careful histologic analysis and the incorporation of ancillary cytogenetic and molecular genetic studies. The identification of areas at the periphery of the tumor with histologic features classic for AFH provide initial clues for the correct diagnosis. Myoepitheliomas typically arise in the extremities with a wide age distribution. They show a multinodular growth pattern and a variety of morphologic appearances, including epithelioid, ovoid, or spindled cells with a chondromyxoid stroma, and may be confused with myxoid AFH because of their expression of EMA in approximately half of the cases. However, in contrast to myxoid AMFH, they often express myoepithelium-associated markers particularly S-100 protein and SOX10 but only rarely desmin and generally show greater cytologic polymorphism and more architectural variability [11, 16, 17]. Extraskeletal myxoid chondrosarcoma typically does not show convincing cartilaginous differentiation and shares with myxoid AFH a pseudocapsule, multinodularity, and a prominent myxoid matrix often with prominent stromal hemorrhage. Extraskeletal myxoid chondrosarcoma tends to arise in older patients with a male predominance. In contrast to myxoid AFH, these tumors lack a peritumoral lymphoplasmacytic cuff, are negative for EMA and desmin, and most commonly show *NR4A3* gene rearrangement [11, 18]. Myxoid liposarcoma, commonly arising in the extremities of young adults, shows multinodular growth and a prominent myxoid matrix, typical arborizing "chicken-wire" vasculature, mucin pools, and variable transition from myxoid to hypercellular areas. In contrast to myxoid AFH, myxoid liposarcoma is not demarcated by a fibrous pseudocapsule and does not display a lymphoplasmacytic cuff or blood-filled cystic spaces. Lipoblasts are usually seen. Myxoid liposarcomas are negative for desmin and EMA but, like myxoid AFH, harbor rearrangements involving *FUS* and/or *EWSR1* [2, 11].

Conclusions

In summary, AFH may present with prominent myxoid stroma, making the correct diagnosis difficult and potentially leading to confusion with other myxoid soft tissue tumors. Our case illustrates the value of rationally integrating morphologic, immunophenotypic, and molecular findings for the identification of unusual presentations of AFH.

Disclosure of conflict of interest

None.

Address correspondence to: Dr. Xiao-Dong Teng, Department of Pathology, The First Affiliated Hospital, School of Medicine, Zhejiang University, 79 Qingchun Road, Shangcheng District, 310003, Hangzhou, Zhejiang Province, P. R. China. E-mail: teng1723@163.com; 1102069@zju.edu.cn

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