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Case Report

Diagnostic Dilemma in Embryonal Rhabdomyosarcoma with Unusual Clinical Manifestation and Positive PAX5 Immunoreactivity: A Case Report and Review of the English Literature

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Abstract

Rhabdomyosarcoma (RMS) is the most frequent soft tissue sarcoma in children and young adults. Rarely, a range of hematological presentations can occur in RMS with an unknown primary tumor site and bone marrow involvement. The clinical manifestations of RMS mimicking acute leukemia are most reported in alveolar RMS (ARMS), which is associated with poor prognosis. Paired-box (PAX) 5 is a frequently used B-lineage marker for the diagnosis of B-cell lymphoma. However, recent reports suggest that PAX5 is immunoreactive to several other malignant tumors, including ARMS but not embryonal RMS (ERMS). This cross-reaction further complicates the differential diagnosis between RMS and B-cell lymphoma/leukemia. Herein, we present a case of ERMS immunoreactive to PAX5 in a young woman with manifestations resembling high-grade B-cell lymphoma/leukemia. This is the first report of PAX5 immunoreactivity in ERMS. The key takeaway from this case is that when children and young adults present with hematological manifestations, differential diagnoses should include RMS in addition to high-grade lymphoma/leukemia. In contrast to the recurrent forkhead box O1 (FOXO1) gene rearrangement found in ARMS, ERMS does not have recurrent structural chromosomal rearrangements but rather has frequent chromosomal gains. Recently, phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha (PIK3CA) mutation has been reported more frequently in ERMS than in ARMS. The present case demonstrated complex karyotype, C-myelocytomatosis oncogene (MYC) amplification, and a PIK3CA (H1047) mutation. Our findings contribute to the understanding of the molecular genetic profile of ERMS and could be valuable for developing potential targeted therapies.

Keywords: Embryonal rhabdomyosarcoma; B-cell lymphoma/leukemia; Tumor lysis syndrome; Mediastinal mass; PAX5; PIK3CA.

Abbreviations: BCL, B-cell lymphoma; BM, bone marrow; CD, cluster of differentiation; FISH, fluorescence *in situ* hybridization; FOXO1, forkhead box O1; H&E, hematoxylin and eosin; Ki-67, a marker of proliferation, protein-coding gene; LDH, lactate dehydrogenase; MYC, myelocytomatosis oncogene; MyoD1, myogenic differentiation 1; PAX, paired-box; PIK3CA, phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha.

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Introduction

Pediatric rhabdomyosarcoma (RMS) is classified into the following two main subtypes: embryonal RMS (ERMS) and alveolar RMS (ARMS). ERMS is a more prevalent subtype, accounting for 60–70% of all RMS cases. ARMS is the second most common subtype (25%) and is predominantly observed in adolescents and young adults between the ages of 10 and 25 years. Spindle cell/sclerosing RMS is a rare subtype of RMS representing 5–10% of RMS cases. RMS arises from mesenchymal tissue at any location in the body. Most cases of RMS present with a primary mass that causes local destruction and obstruction. Fewer than 25% of patients have overt distant metastatic disease at diagnosis; bone marrow (BM) involvement occurs in approximately 30% of cases. While systemic signs and symptoms are rare, RMS involving the BM without an overt primary tumor site can have manifestations similar to those of acute lymphoblastic leukemia. It has been reported that a collection of hematological systemic symptoms presented in RMS can be easily confused with that of acute leukemia, delaying the diagnosis or even causing a misdiagnosis.[1]

Tumor lysis syndrome is a metabolic complication that occurs more frequently in hematolymphoid malignancy and is very rare in patients with solid tumors, which usually manifests after chemotherapy. Rarely, patients with RMS may present with spontaneous tumor lysis syndrome. To date, only two case reports[1] on RMS with an initial presentation of spontaneous tumor lysis syndrome are available in the literature, and they initially suggested lymphoma/leukemia as the diagnosis. The most common sites of occurrence for RMS are the head and neck region or the genitourinary region. There are a few reported cases where RMS primarily presented with a mediastinal mass in ARMS.[2–4] Mediastinal masses are often observed in primary thymic large B-cell lymphoma or T-lymphoblastic lymphoma in young adults and adolescents, respectively. Clinically, distinguishing RMS from

hematolymphoid malignancy can be challenging, especially when a patient initially presents with tumor lysis syndrome and mediastinal mass.

Pathological evaluation is important to distinguish RMS from lymphoma/leukemia in cases with hematological presentations. Both ERMS and ARMS are morphologically characterized as "small round blue cell tumors," which often mimic lymphoma/leukemia. Most of the time, immunohistochemical studies can effectively distinguish between RMS and lymphoma/leukemia, if these mimickers are considered in the differential diagnoses. Paired-box (PAX) 5 is a well-known B-cell lineage marker, which is largely helpful for the diagnosis of B-cell lymphoma/leukemia. However, a diagnostic dilemma was identified in recent years due to the discovery that PAX5 is immunoreactive to several other tumors, including approximately 67% of ARMS cases.[5] In cases where patients present solely with hematological symptoms and BM infiltration of blast-like small round blue cells positive for PAX5, pathologists may misdiagnose B-cell lymphoma/leukemia if they do not consider RMS as a potential diagnosis.

While the majority of the reported cases initially presenting as possible acute leukemia were ARMS, a minority of them are ERMS. The distinction between ERMS and ARMS primarily relies on fluorescence *in situ* hybridization (FISH) and/or molecular testing for forkhead box O1 (FOXO1) gene arrangement, which occurs in approximately 85% of ARMS cases. However, the absence of FOXO1 gene rearrangement in 20% of ARMS cases poses a challenge to distinguish it from ERMS. ERMS includes the typical (or not otherwise specified), dense, and botryoid patterns of RMS. The dense pattern of ERMS shows a hypercellular area composed of small round blue cells with a high nuclear-to-cytoplasmic ratio and minimal rhabdomyoblastic differentiation that may resemble solid ARMS. The focal, rather than diffuse, myogenin immunostaining pattern and testing for *PAX-FOXO1* translocations may assist in this distinction. However, it is difficult to separate the dense pattern of ERMS from the 20% of ARMS cases without FOXO1 gene rearrangement. Recent studies have revealed that myogenic differentiation 1 (*MYOD1*) (L122R) and/or phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha (*PIK3CA*) (H1047) mutations occur in a subset of ERMS, particularly in sclerosing RMS. Sclerosing RMS, a rare subtype, has been recognized as a separate entity from ERMS in the World Health Organization's soft tissue and bone tumor classification since 2013.[6–8]

To date, case reports on RMS presenting initially with tumor lysis syndrome or a mediastinal mass mimicking lymphoma have been rare. Although immunoreactivity to PAX5 has been reported in RRMS, it has not been reported in ERMS. The cytogenetic and molecular profile of ERMS is still unclear. In this report, we describe a case of ERMS with hematological presentation including a high risk for tumor lysis syndrome, mediastinal mass, and PAX5 immunoreactivity, all of which mimicked high-grade B-cell lymphoma in a 22-year-old woman. The manuscript was prepared according to the CARE guidelines, and the associated checklist was completed.

Case report

This was a retrospective analysis of one clinical case and is not considered human research according to the USA's federal policy. Therefore, institutional review board approval was not required. This study was performed in accordance with the Declaration of Helsinki (as revised in 2013) and adhered to the ethical standards of the relevant ethics committee.

Hematological presentation

A 22-year-old woman with a medical history of polycystic ovarian syndrome and hypothyroidism presented with rash, chest pain, palpitations, shortness of breath, dysphagia, low-grade fevers, weight loss, cough, orthopnea, fatigue, and severe unremitting back pain. Physical examination revealed a palpable left supraclavicular lymph node. Chest computed tomography and magnetic resonance imaging revealed a large anterior mediastinal mass measuring 13.4 cm × 9.7 cm × 5.5 cm which compressed/invaded the superior vena cava, small pericardial effusion, enlarged mediastinal and hilar lymph nodes, and left supraclavicular large lymph node measuring 3.7 cm × 2.9 cm. Pelvic magnetic resonance imaging revealed extensive multifocal, patchy to confluent, and nodular areas of BM replacement, consistent with extensive "lymphomatous" involvement. Positron emission tomography-computed tomography scan showed high metastatic activity in the mediastinal mass and left supraclavicular enlarged lymph node, both with a standardized uptake value of 20, and marked diffuse increased metabolic uptake in the axial and central appendicular skeleton, with standardized uptake values up to 14.

Laboratory tests indicated progressive anemia (hemoglobin of 10.4–11.7 g/dL, normal range: 13–16 g/dL), mild thrombocytopenia (platelet count of 125–136 K/mm^3, normal range: 150–400 K/mm^3), progressively elevated lactate dehydrogenase (LDH) (14,059–26,111 U/L, normal range: 84–246 U/L), mildly elevated uric acid (6.8 mg/dL; normal range: 2.6–6.0 mg/dL), aspartate aminotransferase level of 131 U/L (normal range: 15–37 units/L), potassium at 3.4 mg/dL (normal range: 3.5–5.1 mg/dL), calcium at 9.1 mg/dL (normal range: 8.4–10.2 mg/dL), and phosphorus at 4.7–6.1 mg/dL (normal range: 2.5–4.5 mg/dL). Coagulation studies showed markedly elevated d-dimer levels (14.76 mg/L, normal range: 0–0.59 mg/L). The patient was diagnosed with superior vena cava syndrome caused by the anterior mediastinal mass.

Pathological evaluation

An ultrasound-guided core biopsy of the left supraclavicular lymph node was initially performed. Hematoxylin and eosin (H&E)-stained sections from the small core biopsy showed diffuse and discohesive neoplastic cells that were small to medium in size, with degenerative changes and rhabdoid cytology (Fig. 1a). The tumor cells were positive for PAX5 (strong to variable staining) (Fig. 1b), B-cell lymphoma (BCL)-2 (Fig. 1d), BCL-6 (scattered cells, Fig. 1e), and C-myelocytomatosis oncogene (MYC) (Fig. 1f) but negative for cluster of differentiation (CD)3, CD20 (Fig. 1c), CD30, CD117, CD138, cyclin D1, GATA3, melan-A, pancytokeratin, and TTF1 (data not shown). The proliferative index, as determined by a marker of proliferation, protein-coding gene (commonly known as Ki-67), was approximately 50–60%. Given the patient's age, the extensive BM involvement, the mediastinal mass observed in imaging, the positivity for PAX5, and the markedly increased LDH levels, a diagnosis of high-grade B-cell lymphoma was strongly suggested. However, the core biopsy tissue did not provide enough material for additional immunostaining. Subsequently, a BM biopsy revealed a monotonous population of blast-like malignant cells with occasional clusters, and patchy and diffuse infiltration (Fig. 2a). The neoplastic cells were more viable compared to those from the supraclavicular lymph node. They showed a high nuclear-to-cytoplasmic ratio, prominent nucleoli, and scant cytoplasm, all of which resembled lymphoid blasts (Fig. 2b). Considering the clinical presentation, morphological fea-

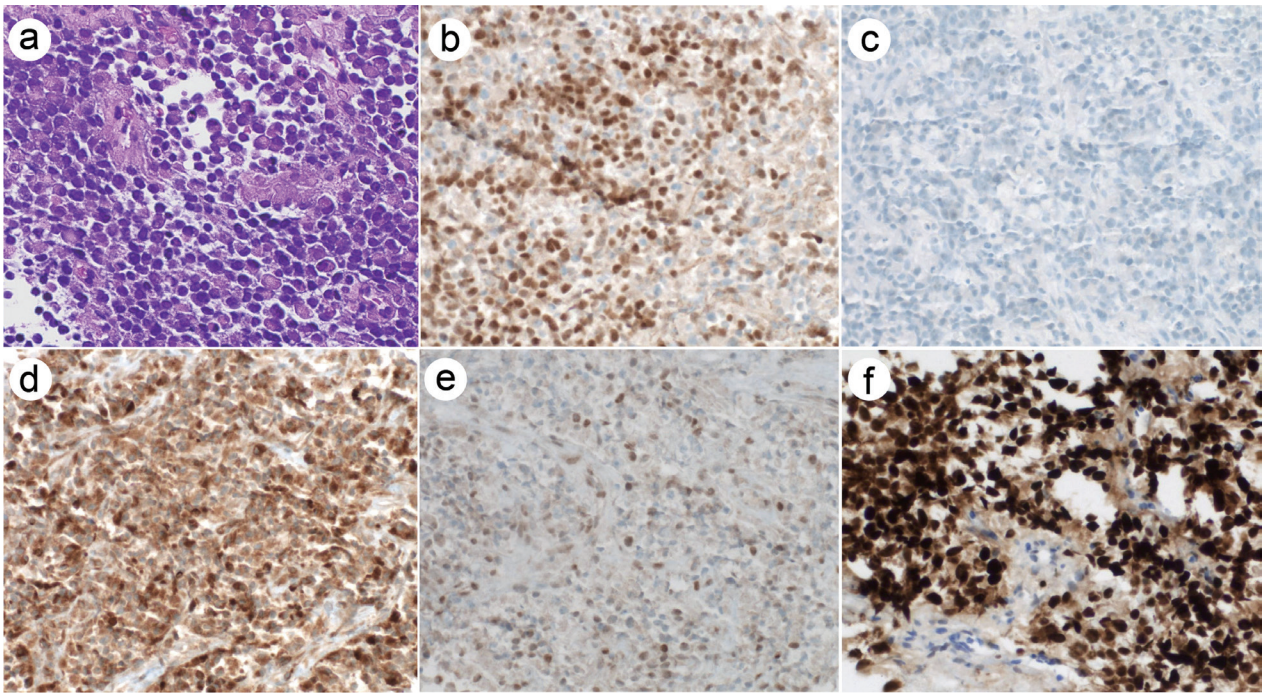

Fig. 1. RMS in the initial supraclavicular lymph node biopsy mimicking B lymphoblastic lymphoma. (a) H&E; (b) PAX5; (c) CD20; (d) BCL-2; (e) BCL-6; (f) C-MYC. Magnification: a: 400x. b-f: 200x. BCL, b-cell lymphoma; CD, cluster of differentiation; H&E, hematoxylin and eosin; MYC, myelocytomatosis oncogene.; PAX5, paired-box 5; RMS, rhabdomyosarcoma.

tures, and PAX5 immunoreactivity from the prior small core biopsy of lymph node, immunohistochemical studies for B-lymphoblastic lymphoma/leukemia were initiated. The immunohistochemical panel showed that the tumor cells were again positive for PAX5 (dim) (Fig. 2c), C-MYC, CD56 (Fig. 2d), and CD99 (patchy and dim), and vimentin but negative for other hematolymphoid markers (ALK, BCl-2, BCL6, BOB-1, CD3, CD4, CD7, CD10, CD19, CD20, CD23, CD30, CD34,

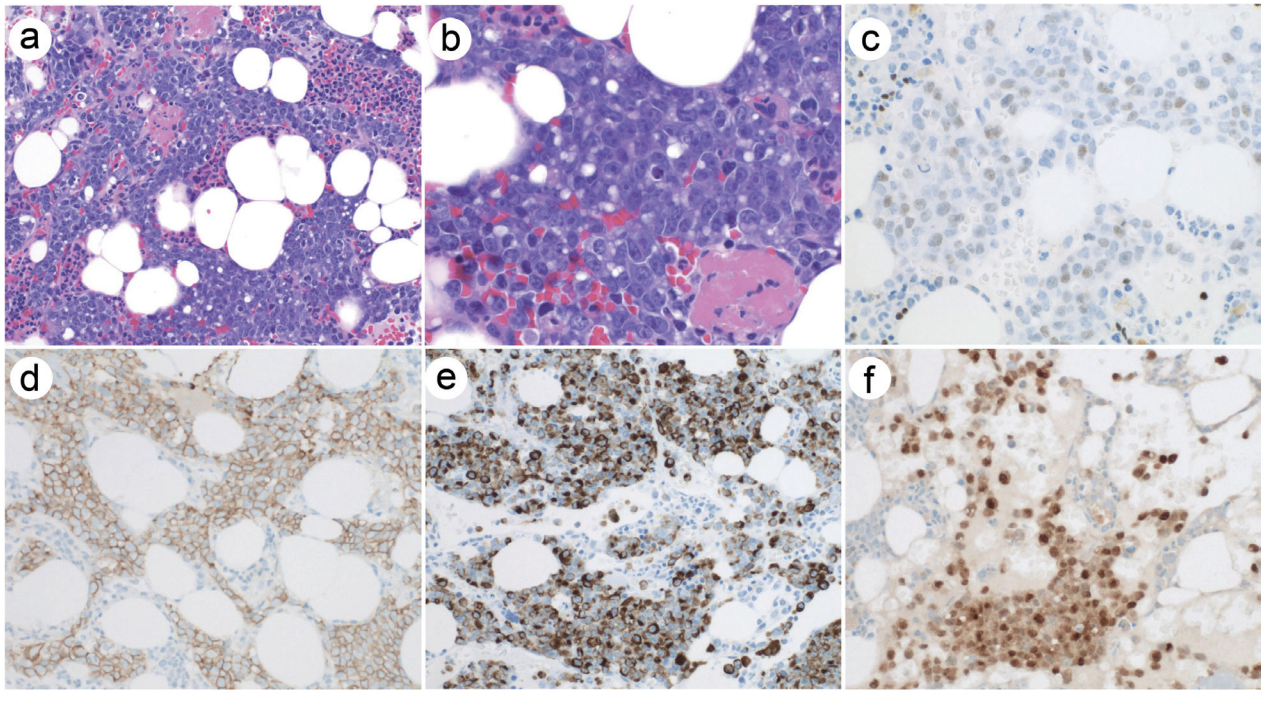

Fig. 2. RMS with BM involvement. (a) H&E; (b) H&E; (c) PAX5; (d) CD56; (e) Desmin; (f) Myogenin. Magnification: a: 200x. b, c: 400×. d-f: 200×. BM, bone marrow; CD, cluster of differentiation; H&E, hematoxylin and eosin; PAX5, paired-box 5; RMS, rhabdomyosarcoma.

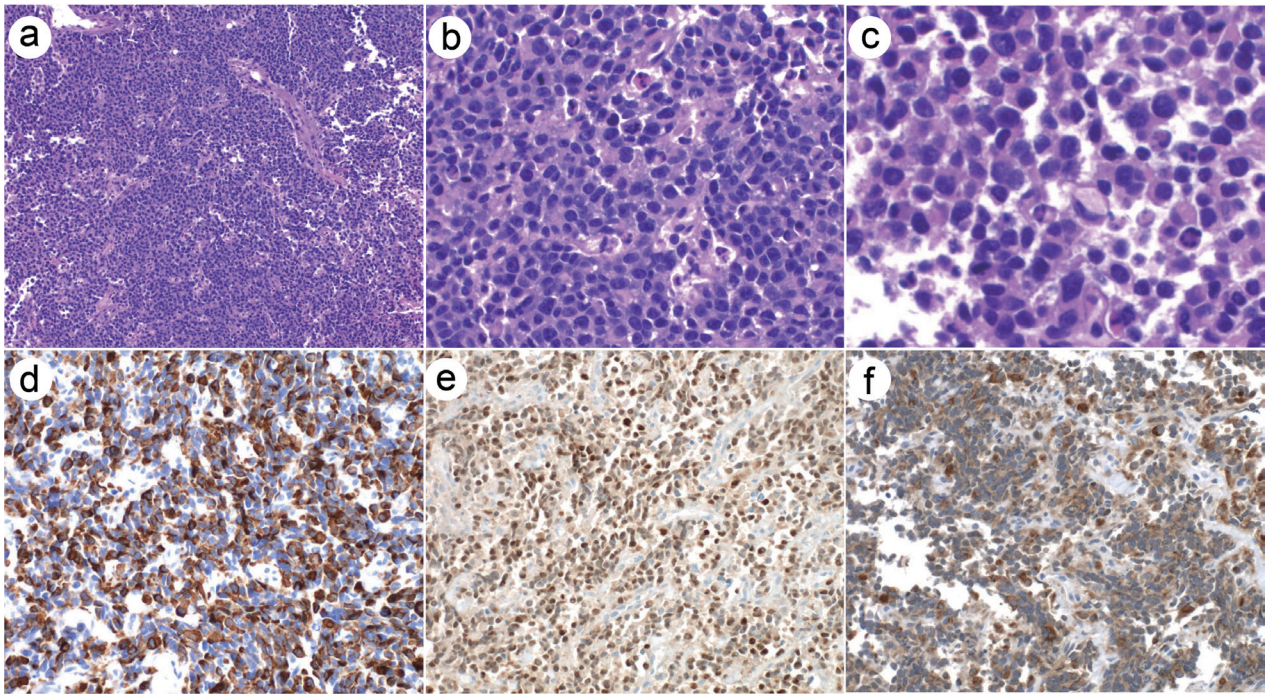

Fig. 3. RMS in the repeat supraclavicular lymph node/mass biopsy. (a) Typical dense pattern of ERMS (H&E); (b). The tumor cells with scant to ample eosinophilic cytoplasm and frequent mitosis and apoptotic bodies (H&E); (c) Rare tumor cells with rhabdoid morphology (H&E); (d) Desmin; (e) Myogenin; (f) MyoD1. Magnification: a: 100x. b: 200x. c: 400×. d-f: 200×. ERMS, embryonal rhabdomyosarcoma; H&E, hematoxylin and eosin; MyoD1, myogenic differentiation 1; RMS, rhabdomyosarcoma.

CD43, CD45, CD117, CD138, cyclin D1, glycophorin, MUM-1, OCT2, TdT), epithelial markers (AE1/AE3, CAM5.2, GATA3, OSCAR, pancytokeratin, and PAX8), melanocytic markers (SOX10, S-100), germ cell tumor markers (PLAP, OCT3/4), and other markers (SMA, WT-1). As PAX5, CD56, C-MYC, and CD99 are not lineage specific, a conclusive diagnosis could not be reached, and further studies were performed. A diagnosis of RMS was finally confirmed based on the tumor cells in the BM specimen being positive for desmin (diffuse) and myogenin (patchy) (Fig. 2e and f). Subsequent repeated supraclavicular lymph node/mass biopsy (Fig. 3) showed an infiltrative tumor comprising closely packed sheets of small round tumor cells with high nuclear to cytoplasmic ratio, round to irregular nuclei, occasional prominent eosinophilic nucleoli, and scant to moderate eosinophilic cytoplasm (Fig. 3a and b). Rare tumor cells demonstrated rhabdoid features, characterized by eccentric nuclei and abundant eosinophilic cytoplasm (Fig. 3c). The sample also showed frequent mitosis and apoptotic bodies (Fig. 3b). The tumor cells were strongly positive for desmin (Fig. 3d), myogenin (patchy to diffuse, Fig. 3e), and MyoD1 (focal to patchy, Fig. 3f), consistent with RMS. However, the alveolar type of RMS was favored in conjunction with the compatible features of discohesive morphology and progressive clinical presentation of BM involvement.

Cytogenetic and next-generation sequencing

Chromosomal analysis of the BM aspirate revealed complex hyperdiploid chromosomal abnormalities, including the gain of chromosomes X, 4, 6, 8, and 17, along with multiple copies of double minutes or small chromosome fragments. Interphase FISH revealed C-MYC (8q24) amplification in 49.5% of the 200 examined nuclei. However, FISH for FOXO1 gene rearrangement showed negative results. Next-generation sequencing was employed to further explore the genetic landscape. The Rhabdomyosarcoma Fusion Profile performed by NeoGenomics Laboratories (Fort Meyers, FL, USA) on the supraclavicular lymph node showed no evidence of expression of fusion RNA or mutations involving the FOXO1, NCOA2, and TEF3 genes; however, FoundationOne Heme panel (406 genes for limited exons using DNA and 265 rearrangement and fusions using RNA; Foundation Medicine, Cambridge, MA, USA) identified a *PIK3CA* H1047R (no variant allele frequency provided) and C-MYC amplification in the same position. No N-MYC amplification was detected. While MyoD1 is not included in this panel, due to the absence of FOXO1 gene rearrangement this case was re-classified as ERMS with a dense pattern.

Discussion

RMS usually presents as a primary mass in children and young adults. Stage IV disease with BM metastasis or dissemination is rare, and more commonly occurs in ARMS,[9] which has a worse prognosis. Very rarely, patients with disseminated RMS with an unknown primary site may present with hematological symptoms such as leukocytosis, cytopenia, disseminated intravascular coagulopathy),[1] tumor lysis syndrome,[1] lymphadenopathy,[10] and mediastinal mass.[2–4] Numerous cases have been misdiagnosed as acute lymphoblastic lymphoma/leukemia due to similar presentations.[9,11] There have been several reports[1] on widespread RMS in adolescents presenting with severe hemorrhage resulting from disseminated intravascular coagulopathy and the laboratory features strongly suggestive of acute hematologic malignancy. For example, Bien *et al*.[1] reviewed 13 cases of childhood RMS metastatic to BM presenting with disseminated intravascular coagulopathy. One of these cases[12] involved a 14-year-old boy presenting with leukocytosis, disseminated

intravascular coagulopathy, and acute tumor lysis syndrome. BM aspirate showed 89.6% of undifferentiated tumor cells, initially suggesting the suspicion of hematological malignancy. The patient was treated with idarubicin and cytarabine. However, further examination revealed that tumor cells were positive for CD56, PAS, HHF35 (muscle-actin-specific), and desmin, and lacked lineage-specific markers of lymphoid or myeloid cells. They were negative for MPO. Reverse transcriptase polymerase chain reaction demonstrated PAX3/FKHR (now called PAX3/FOXO1) fusion transcripts, confirming the diagnosis of ARMS. Another 14-year-old boy presented similarly with acute tumor lysis syndrome and received treatment for malignant non-Hodgkin's lymphoma and Ewing sarcoma 3 days before being diagnosed with probable ERMS of unknown primary site. Despite aggressive therapy, the patient died of disease progression 3 months later. Such hematological presentations show the diagnostic challenge posed by hematological presentations of RMS, leading to misdiagnoses, and affecting clinical management and prognosis. Among these 13 cases, there were 5 cases of ERMS, 5 of ARMS, and 3 of unclassified RMS.

Tumor lysis syndrome consists of a constellation of laboratory findings such as hyperuricemia, hyperkalemia, hyperphosphatemia, and hypocalcemia. The main predictors of its development are tumor burden (reflected by serum LDH levels), initial white blood cell count, tumor size, and extensive BM involvement. Tumor lysis syndrome is particularly common in patients with hematological malignancies characterized by rapid cellular turnover rates, such as acute lymphoblastic leukemia or Burkitt lymphoma; however, it is very rare in patients with solid tumors and extremely rare in RMS,[1,13] especially if it occurs spontaneously without treatment.[14] In the current patient, the large size of the mediastinal mass suggested that the mediastinal mass was the primary site. The large tumor burden, extensive BM involvement, the abnormal laboratory tests with marked elevated LDH, mild hyperuricemia, and hyperphosphatemia indicated that the patient was at risk for tumor lysis syndrome. Consequently, allopurinol was initiated to prevent tumor lysis syndrome before a definitive diagnosis was established. The patient also had anemia, thrombocytopenia, and elevated D-dimer levels. These hematological symptoms led to a strong clinical suspicion of a hematolymphoid malignancy such as high-grade lymphoma. Based on this impression, the initial pathological workup on the lymph node biopsy was biased toward lymphoma/leukemia in the BM on this case.

High-grade B-cell lymphoma usually presents with lymphadenopathy, and symptoms such as weight loss, fever, and fatigue. Although RMS rarely has these presentations, a small number of RMS cases have been reported with initial presentations of lymphadenopathy and mediastinal mass. Primary mediastinal RMS unassociated with germ cell tumors, teratoma, or malignant epithelial components is extremely rare.[4] Because of the presence of lymphadenopathy and mediastinal mass in addition to other hematological symptoms in our young adult patient, there was a high clinical suspicion for high-grade large-cell lymphoma. The initial presentation of a mediastinal mass should prompt pathologists to consider not only lymphoma, germ cell tumors, and thymoma but also RMS, especially in adolescents and young adults.

PAX5 is a transcription activator involved in the development of B-cells. Although PAX5 is a B-cell-specific activator, its expression has also been reported in non-B-cell malignancies, such as a subset of neuroendocrine carcinomas, urothelial tumors, Merkel cell carcinoma, small-cell carcinoma, glioblastoma, and neuroblastoma cell lines. Sullivan *et al*.[5] reported that 34 of 51 (67%) ARMS were PAX5 immunoreactive, whereas none of the 55 ERMS cases showed such reactivity. All of the PAX3 or PAX7-FOXO1 translocation-positive cases showed nuclear reactivity to PAX5, whereas the translocation-negative cases did not. The authors proposed that PAX3 and PAX7 fusion genes characterize the majority of ARMS, making cross-reactivity with these proteins an attractive theory, and suggest that PAX5 immunoreactivity may be specific for translocation-positive ARMS. However, our case is believed to be ERMS without any translocation involving the PAX gene, and it demonstrated PAX5 positivity, which may suggest the possibility of cross-reactivity with other PAX proteins in ERMS. To the best of our knowledge, our case is the first reported instance of PAX5 immunoreactivity in ERMS. Unfortunately, the presence of PAX5 coupled with the hematological presentation and lack of immunohistochemical studies for RMS led to an initial impression of high-grade B-cell lymphoma in the left subclavicular lymph node biopsy. The lesson learned from this case is that the presence of small blue round cells morphologically similar to lymphoblasts and/or myeloblasts in BM in the absence of specific hematopoietic markers should prompt pathologists to consider a possible diagnosis of RMS. The inclusion of desmin, MyoD1, and myogenin in the immunohistochemical panel is essential in such cases.

In contrast to the recurrent chromosomal translocations found in ARMS, ERMS exhibits frequent chromosomal gains rather than recurrent structural rearrangements. The most notable gains in ERMS are chromosomes 2, 8, 11, 12, 13, and 20.[15] Comparative genomic hybridization studies indicated that usual ERMS tumors have a low frequency of amplification (6–10%) and anaplastic ERMS tumors had a higher frequency of amplification (67%).[15] Our case demonstrated gain of chromosomes X, 4, 6, 8, and 17, and amplification of C-MYC by FISH, which corresponded to the presence of multiple copies of chromosome 8. This amplification of C-MYC may indicate the possibility of anaplastic ERMS in our case, although it could not be confirmed due to the small size of the biopsy sample. To the best of our knowledge, C-MYC amplification has not been directly reported in any case of RMS except for the ARMS and ERMS cell lines,[16] although N-MYC[17] and MDM2[18] amplifications have been reported in ARMS and/or ERMS. MYC (C-MYC) encodes a transcription factor that regulates many genes related to cell cycle regulation and growth. MYC amplification has been significantly linked with increased mRNA and protein levels, leading to the dysregulation of a large number of target genes. Studies have shown that cells that overexpress MYC protein may be sensitive to some inhibitors.

While ARMS typically contains translocations generating the PAX3-FOXO1 or PAX7-FOXO1 aberrant transcription factors, which block terminal myogenic differentiation,[7] no functionally comparable genetic event has been found in ERMS. Recent oncogene mutation profiling of pediatric solid tumors revealed significant subsets of ERMS with recurrent mutations, particularly in the myogenic transcription factor MYOD1 and PIK3CA. Three independent studies demonstrated the presence of MYODI (L122R) mutations as the basis to re-classify spindle cell and sclerosing RMSs as distinct typical ERMS.[6–8] For instance, Shukla *et al*.[8] reported PIK3CA mutations in 5% (3/60) of ERMS cases, two of which were re-classified as sclerosing RMS. A study by Kohsaka *et al*.,[7] involving whole exome sequencing of 20 RMS samples from children and young adults (< 25 years-old), discovered a recurrent somatic point mutation Leu122Arg in MYOD1 in a distinctive subset of ERMS characterized by poor outcomes and frequent PI3K/AKT pathway mutations. This recurring point mutation in MYOD1 is found in 10% of ERMS cases.

Mutant MYOD1 cooperates with PI3K pathway alterations to block differentiation and promote transformation. *MYOD1* mutations identify a subset of ERMS that may benefit from high-risk treatment protocols or targeted therapy.

PIK3CA mutation occurs more frequently in ERMS, which may further support the diagnosis of ERMS rather than ARMS in this case. Of note, one study revealed no PIK3CA mutation in a sample of 49 cases of ERMS. Large cohort studies are needed to identify recurrent genetic alteration specific for ERMS, especially considering the 20% of ARMS cases without the FOXO1 gene rearrangement. The presence of PIK3CA mutation and/or C-MYC amplification in this case may be associated with the clinically aggressive features of disseminated disease with BM involvement, resulting in hematologic symptoms, tumor lysis syndrome, lymphadenopathy, and a mediastinal primary site. This finding is similar to a case report in which PI3KCA mutation and MDM2 amplification were associated with progressive course in sclerosing RMS.[18] The PIK3CA mutation and C-MYC amplification identified in our case may benefit from targeted therapy. In addition, understanding the MYOD1 mutation status in this patient may help to understand the molecular pathogenesis and possible targeted therapy, although this case had no evidence of sclerosing RMS.

In summary, we describe the first case of ERMS exhibiting positive PAX5 immunoreactivity. It highlights the importance of considering an immunohistochemical evaluation for RMS in cases involving children and young adults with hematological manifestations and blast-like small round blue cell tumors, to avoid misdiagnosis.

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Conflict of interest

HYW is the Editor-in-Chief of the *Journal of Clinical and Translational Pathology*. The authors have no other conflicts of interest to declare.

Author contributions

Study design and manuscript writing (DHW) and critical revision of the manuscript (HYW). Both authors made a significant contribution to this study and approved the final manuscript.

Ethical statement

This case report was a retrospective analysis of a single clinical case and is not considered human research according to the USA's federal policy. An institutional review board approval was thus deemed unnecessary. This study was performed in accordance with the Declaration of Helsinki (as revised in 2013) and adhered to the ethical standards of the relevant ethics committee. The individual consent for this retrospective analysis was waived.

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