Ewing Sarcoma/Primitive Neuroectodermal Tumor of the Kidney

A Rare and Lethal Entity

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Ewing sarcoma/primitive neuroectodermal tumor (ES/PNET) is a group of rare, primitive, biologically aggressive tumors derived from the neuroectoderm. Although the tumor typically arises in the soft tissue and bones of children and young adults, it may rarely present as a tumor arising from the kidney. It is important to distinguish ES/PNET from other entities that represent a renal mass because of its dire prognosis as well as treatment implications.

Since its first description in 1975,1 there have been only a few case reports and small case series of primary renal ES/PNET, with a recent meta-analysis of the literature by Risi et al2 summarizing the clinicopathologic characteristics of 116 cases. This rare entity exhibits a unique set of clinical features, gross/microscopic pathology, and genetic signature that should be used to distinguish it from other small round blue cell tumors of the kidney.

CLINICAL FEATURES

Ewing sarcoma/primitive neuroectodermal tumor typically arises in adolescents and young adults, with a few series and one meta-analysis reporting a median age at diagnosis of 27 to 28 years old.2–4 The majority of patients present with back pain (most common) and hematuria.2,5 This is in contrast to most renal tumors, which are incidental imaging findings. Patients typically present at an advanced tumor stage.6 One-third of patients present with tumor thrombi in the renal vein or inferior vena cava at the time of diagnosis.6,7 The most common sites of metastasis are the lungs, followed by the liver and bone.6,7

RADIOLOGIC FEATURES

Imaging study findings in patients with ES/PNET include a large, ill-defined renal mass, often with heterogeneous contrast enhancement with intermixed areas of necrosis and hemorrhage (Figure, A).3,5,6,8 Focal calcifications are occasionally seen.3,9 The imaging features are in general nonspecific and the diagnosis, although rare, should be entertained whenever a young patient presents with a large renal mass. The differential diagnosis based on imaging findings may include rhabdomyosarcoma, Wilms tumor, carcinoid tumor, neuroblastoma, lymphoma, and desmoplastic small round blue cell tumor.3,6

GROSS AND MICROSCOPIC PATHOLOGY

Grossly, tumor size ranges from 3.3 to 18 cm, with 13 cm as the median diameter in the recent meta-analysis of the literature.2,3,7 Confluent areas of necrosis and hemorrhage are characteristic (Figure, A, B, C, and F).10 Renal vein invasion can be grossly identified in a number of cases.10 Microscopically, most tumors are composed of uniform small round cells, but in others, the tumor cells are larger. There is infiltration into the normal renal parenchyma (Figure, E) in broad sheets (Figure, D and F) and/or narrow projections10 (Figure, D). The tumor cells have a high nuclear to cytoplasmic ratio and the nuclei are hyperchromatic, round to ovoid, with condensed chromatin and few small nucleoli (Figure, G). There are cases that show cells with a small amount of clear-appearing cytoplasm.11 In the largest single review of cases collected by one group, 44 of

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79 cases showed Flexner-Wintersteiner rosettes with well-defined central lumina. In 27 of 79 cases Homer Wright rosettes were present, with fibrillary cores. Both rosette patterns were seen together in 3 of the 79 cases. The cases with rosette formations were traditionally reserved the title PNET, whereas those cases without were termed ES. It is now recognized that both represent a morphologic spectrum of the same underlying process and that the presence of rosettes is not a feature required for diagnosis. Increased and atypical mitotic figures are commonly seen, as well as microscopic evidence of angiolymphatic invasion.

Imaging, gross, microscopic, immunohistochemical, and molecular features of Ewing sarcoma/primitive neuroectodermal tumor of the kidney. A, Axial noncontrast magnetic resonance imaging shows a complex mass with hematoma arising from inferior renal pole. B, Gross nephrectomy specimen reveals a 14-cm paracortical mass with central hemorrhage and necrosis. The white arrow indicates residual kidney and the black arrow indicates the tumor. C, Cut section of the kidney with attached mass. The white arrow indicates residual kidney and the black arrow indicates the tumor. D, Tumor grows in solid sheets as well as in an infiltrative pattern around normal renal parenchymal structures. E, Small round blue tumor cells infiltrating sclerotic renal cortical tissue. F, Tumor is composed of sheets of viable cells (left) intermixed with areas of necrosis (right). G, Tumor cells are uniformly small and exhibit high nuclear to cytoplasmic ratio, with nuclei demonstrating condensed chromatin and small nucleoli. H, Tumor cells show strong membranous staining with CD99. I, Dual-probe interphase fluorescence in situ hybridization study shows cells with split red and green signals (yellow arrow) confirming EWS gene rearrangement. Note that there are other nontumor cells present without separation of the signals (white arrow) (hematoxylin-eosin, original magnifications ×10 [D and F] ×20 [E], and ×40 [G]; original magnifications ×40 [H] and ×120 [I]).
**Immunohistochemistry**

The immunohistochemical pattern of renal ES/PNET is similar to that seen in other locations. Proteins encoded by the MIB2 gene, most commonly CD99 or O-13, are the most commonly expressed markers.\textsuperscript{10,13} CD99 immunohistochemistry is positive in more than 90% of ES/PNET.\textsuperscript{14,15} Tumor cells typically stain in a diffusely membranous pattern (Figure, H). CD99 has been found to be largely nonspecific, however, showing expression in nonneuroectodermal tumors such as lymphoblastic lymphoma, synovial sarcoma, and rhabdomyosarcoma, albeit in a weaker, cytoplasmic staining pattern.\textsuperscript{14,16} Friend leukemia virus integration 1 (FLI-1), a DNA-binding transcription factor, has been found to be overexpressed in a majority of ES/PNET (as a result of the gene rearrangement it is involved in, described below).\textsuperscript{17} In one study by Folpe et al.,\textsuperscript{15} which included 41 cases of ES/PNET, antibody to FLI-1 showed strong nuclear positivity in 71% of cases. The use of this antibody is limited also because of nonspecificity, staining a small proportion of rhabdomyosarcomas, desmoplastic small round cell tumors, and synovial sarcomas,\textsuperscript{14,15} and a majority of lymphoblastic lymphomas.\textsuperscript{14} FLI-1 also stains endothelial cells and normal lymphocytes, which can serve as internal positive controls. Together, CD99 and FLI-1 constitute a useful albeit incomplete immunohistochemical panel in the workup of small round blue cell tumors of the kidney.

Vimentin and neuron-specific enolase have been reported positive in more than 80% of cases.\textsuperscript{6,13} Cytokeratins can be positive (less than 10%-20% of cases).\textsuperscript{2,11} S100 and synaptophysin have been reported as positive in 30% to 40% of cases.\textsuperscript{2} WT-1 is generally negative; however, one series reported positivity in 3 cases showing EWS rearrangement with fluorescence in situ hybridization (FISH).\textsuperscript{18}

**Molecular Studies**

The most common genetic alteration seen in ES/PNET, including those arising from the kidney, is the fixed chromosomal translocation t(11;22) between the genes EWS (22q12) and FLI-1 (11q24). The chimeric gene product localizes to the nucleus and binds DNA at the same site as normal FLI-1, activating sets of genes that FLI-1 alone cannot.\textsuperscript{19} In the kidney, ES/PNET has been shown to be positive for t(11;22) in 72% of the cases.\textsuperscript{2} Other members of the ETS (erythroid differentiation–associated transforming sequences) family of genes (such as ERG)\textsuperscript{20} have less commonly been reported as variant translocation partners with EWS (22q12). Our review of the literature did not find any mention of ES/PNET of the kidney positive for these secondary mutations. The mutation has traditionally been detected in the clinical setting by FISH or direct sequencing with reverse transcriptase–polymerase chain reaction. Dual-color, break-apart FISH (used at our institution) includes fluorescently labeled probes specific to the sequences flanking the EWS gene. Break-apart of the signals signifies a positive gene rearrangement (Figure, I). This method is sensitive yet not specific for ES/PNET because there are other entities that demonstrate EWS gene rearrangement, including desmoplastic small round cell tumor, clear cell sarcoma, extraskeletal myxoid chondrosarcoma, and myxoid liposarcoma. Direct sequencing following reverse transcriptase–polymerase chain reaction is more specific but requires frozen tumor tissue. Patients carrying the translocation have been shown to have a slightly better disease-free survival, although the difference is not significant (P = .23).\textsuperscript{2}

Other cytogenetic abnormalities have rarely been tested for and detected. In a study by Parham et al.\textsuperscript{12} of 146 cases of renal ES/PNET, 5 cases were karyotyped. Among those, 3 had additional abnormalities beyond t(11;22). Two of the 3 showed atypical morphologic features. One demonstrated plump, spindle-shaped streaming cells in an abundant myxoid stroma, and the other had large undifferentiated cells.

**Differential Diagnosis**

Because of its atypical location at presentation, ES/PNET of the kidney should be diagnosed only following a thorough investigation of histomorphology, multiple immunostains, and, whenever possible, confirmation using molecular–genetic testing. There are many other entities composed of small round blue cells that may be primary to or secondarily involve the kidney. The list includes blastemal predominant Wilms tumor, lymphoblastic lymphoma, synovial sarcoma, solid variant of alveolar rhabdomyosarcoma, clear cell sarcoma of the kidney, neuroblastoma, desmoplastic small round blue cell tumor, and small cell carcinoma. Complicating matters, there are a number of positive immunohistochemical markers that are shared among these entities. For this reason, a careful selection and interpretation of immunohistochemical markers is recommended (Table).

Hematologic malignancies, primarily lymphoblastic lymphoma, are close mimics of ES/PNET. These are the only other tumors in the differential that are also typically positive for both CD99 and FLI-1. The diagnosis of ES/PNET should not be made without ruling out lymphoblastic lymphoma or other hematologic malignancies. This means that there should be a documented negative CD45, terminal deoxynucleotidyl transferase, and/or CD43. A limited panel using these stains as well as CD99 and FLI-1 can be useful.

<table>
<thead>
<tr>
<th>Diagnosis</th>
<th>CD99</th>
<th>FLI1</th>
<th>CD45</th>
<th>TdT</th>
<th>WT-1</th>
<th>Cytokeratin</th>
<th>Desmin</th>
<th>Myogenin</th>
<th>Genetic Alterations</th>
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</thead>
<tbody>
<tr>
<td>ES/PNET</td>
<td>+</td>
<td>+</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>Rare</td>
<td>Rare</td>
<td>−</td>
<td>t(11;22)(q24;q12) EWS-FLI1</td>
</tr>
<tr>
<td>LBL</td>
<td>Variable</td>
<td>Variable</td>
<td>+</td>
<td>+</td>
<td>−</td>
<td>Rare</td>
<td>−</td>
<td>NA</td>
<td></td>
</tr>
<tr>
<td>RMS</td>
<td>−/−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−/−</td>
<td>−</td>
<td>−</td>
<td>t(11;19)(p11.2;q13.3) EWS-WT1</td>
</tr>
<tr>
<td>SS</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>Variable/−</td>
<td>Rare</td>
<td>−</td>
<td>t(X;18)(p11.23;q11) SS18-SSX1</td>
</tr>
<tr>
<td>NB</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>−</td>
<td>+</td>
<td>−</td>
<td>t(2;13)(q35;q14) PAX3-FKHR</td>
</tr>
<tr>
<td>DSRCT</td>
<td>Variable</td>
<td>Variable</td>
<td>+</td>
<td>+</td>
<td>−</td>
<td>+</td>
<td>−</td>
<td>−</td>
<td>MYCN gene (2p24) amplification (25%)</td>
</tr>
</tbody>
</table>

Abbreviations: DSRCT, desmoplastic small round cell tumor; ES/PNET, Ewing sarcoma/primitive neuroectodermal tumor; LBL, lymphoblastic lymphoma; NA, not applicable; NB, neuroblastoma; RMS, rhabdomyosarcoma; SS, synovial sarcoma; TdT, terminal deoxynucleotidyl transferase; WT, Wilms tumor; +, positive staining; −, negative staining.

\* Data derived from Folpe et al.,\textsuperscript{15} Arvid and Denny,\textsuperscript{19} Carpentieri et al,\textsuperscript{20} Zhang et al,\textsuperscript{21} and Hasegawa et al.\textsuperscript{22}
when suggesting this differential diagnosis based on morphologic analysis.

Given its location and its classic small blue cell appearance, blastemal predominant Wilms tumor should be high on the differential diagnosis. An important reason to distinguish these 2 entities is that Wilms tumor responds well to a standard regimen of multiagent chemotherapy and thus has a much better prognosis than ES/PNET of the kidney. Blastemal predominant Wilms tumor should be negative for CD99 (although rare positivity has been reported) and FLI-1 and positive for WT-1. Although supporting the diagnosis of Wilms tumor, WT-1 positivity alone is insufficient to rule out ES/PNET. In a series of 30 cases diagnosed as ES/PNET, 8 were positive for WT-1, although only 3 of the 8 were also positive for EWS rearrangement by FISH.

Other potential differential diagnoses include solid variant of alveolar rhabdomyosarcoma, desmoplastic small round blue cell tumor, neuroblastoma, small cell carcinoma, and synovial sarcoma. Rhabdomyosarcoma should have cells with brightly eosinophilic cytoplasm and show skeletal muscle differentiation by immunohistochemistry. Desmoplastic small round blue cell tumor has a unique stereotypic histologic appearance characterized by nests of malignant small cells embedded in a highly vascular, desmoplastic stroma. The cells should coexpress vimentin, desmin, and cytokeratin and may only focally express CD99. Neuroblastoma almost always presents in infants and the small blue cells are CD99 negative. Additionally, patients tend to have increased catecholamine metabolites in blood. Small cell carcinoma should show cytokeratin and chromogranin and/or synaptophysin positivity and CD99 negativity, and may present at multiple locations. Synovial sarcoma should typically have at least focal areas of classic monophasic/biphasic pattern, with cytokeratin positivity and FLI-1 negativity.

Poorly differentiated small round blue cell tumors with conflicting immunophenotypes can be notoriously difficult to diagnose, and genetic/molecular techniques can be very helpful in establishing the correct diagnosis.

**TREATMENT AND PROGNOSIS**

Because of the rarity of this tumor, there is no standardized treatment strategy. The primary modality is surgical excision. The 2-year overall survival of patients who undergo surgery is 80%, compared with 30% for those who do not (statistically significant, P = .02). Approximately half of patients receive neoadjuvant or adjuvant chemotherapy. Because of biologic similarities to ES/PNET at other sites, the cases primary to the kidney are treated in a similar fashion. It is common for 5 or more chemotherapeutic agents to be used at once. Among those used are doxorubicin, vincristine, cyclophosphamide, ifosfamide, and etoposide. Before the use of chemotherapy, the 5-year survival rate was less than 10%. Now the 5-year survival rate for patients treated with chemotherapy has been reported at 45% to 55%. For patients who receive nonsurgical therapy (presurgery or postsurgery), there is a mild increase in 12-month overall survival (93% versus 75%, P = .92). Radiation has shown some success as salvage therapy, for example as targeted treatment of positive lymph nodes following surgery, but is not recommended as a primary modality of treatment.

Despite aggressive treatment strategies, the prognosis of primary renal ES/PNET remains dismal. Median overall survival has been reported at 26.5 months. Patients with metastatic disease have more than a 4-fold increase in relative risk of death. Importantly, in the largest meta-analysis, 40% of patients who presented without metastasis subsequently developed metastasis following surgery. The most common metastatic sites are lung, liver, bone, and lymph node. 

**SUMMARY**

Primary renal ES/PNET is a rare and lethal entity. Because of the prognostic and therapeutic implications of this diagnosis, consideration must be made of the other differential diagnoses of small round blue cell tumors of the kidney. Whenever possible, correlation must be made among the gross pathology, histomorphology, immunostains, and molecular/genetic testing. Knowledge of the genetics of this lesion can guide gross room protocol, because saving fresh frozen tissue is necessary for gene-sequencing studies. In particular, ES/PNET of the kidney typically presents as a large renal tumor with a heterogeneous appearance including areas of hemorrhage and necrosis. Tumor cells are small and round and have a high nuclear to cytoplasmic ratio. Rosettes may or may not be seen. Tumor cells typically coexpress the following antigens: CD99, FLI-1, and vimentin. By FISH the tumor cells demonstrate rearrangement of the EWS gene, and by direct sequencing the most common alteration is the EWS:FLI-1 translocation. Although the tumor uniformly manifests aggressive clinical behavior, its accurate diagnosis facilitates the prolongation of survival with appropriate treatments.

**References**


