Abstract. Primitive neuroectodermal tumors (PNETs) are malignant tumors comprised of small round cells of neuro-ectodermal origin. Current evidence indicates that peripheral PNETs (pPNETs), which arise in the non-central nervous system, possess histological similarity to Ewing's sarcoma. Though the occurrence of pPNETs in the head and neck region is rare, these are aggressive malignant tumors, and long-term survival rates following diagnosis remain poor. The current report presents a case of pPNET and evaluates its significance with regard to previous studies. In the present case, a tumor was located in the larynx of the patient, and was diagnosed as pPNET. Immunohistochemical analysis indicated that tumor cells were positive for cluster of differentiation 99. The patient was treated with surgery, multiagent chemotherapy and radiotherapy. Five years subsequent to treatment, the patient had survived and demonstrated no evidence of disease recurrence. In existing literature concerning pPNET located outside the head and neck region, it is recommended that patients are treated with a combination of resection with a wide surgical margin, multiagent chemotherapy and radiotherapy. The present case report concluded that the combination of surgery, systematic chemotherapy and radiotherapy, offers an improved outcome for pPNET localized to the head and neck region, compared with any of these therapies alone.

Introduction

Primitive neuroectodermal tumor (PNET) is a small round cell sarcoma that primarily develops in the central nervous system (CNS) and soft tissues of children (1). In particular, peripheral PNET (pPNET) has been hypothesized to originate from the neural crest (2). Previously, PNET was suggested to be a neoplasm of the central nervous system (3). Since then, this concept has been extended to encompass the periphery, and these non-central nervous system tumors are referred to as pPNETs (3). pPNET was initially described by Stout in 1918 (4). In addition, Ewing's sarcoma (ES) of the bone was initially reported by J. Ewing in 1921 (5). Subsequent to this initial description, a diagnosis of extraskeletal ES was reported by Angervall and Enzinger in 1975 (6). In 1979, Askin et al (7) described a ‘malignant small cell tumor of the thoracopulmonary region’ (Askin tumor). An additional significant discovery occurred in 1984, when a neuroectodermal tumor of the bone was identified by Jaffe et al (8).

At the time of diagnosis, these aforementioned tumors were recognized as being pathologically distinct. However, it has since become apparent that these tumors share a number of pathological features (7,8). ES and pPNET (including Askin tumor) are proposed to arise from the neural crest (9). Currently, there are two opposing opinions that recognize ES and pPNET as separate entities, or alternatively group them within a single category known as the ‘ES/pPNET group’ (3) or the ‘ES family of tumors’ (10). Pathologically, when ES and pPNET are recognized as separate entities, these distinctions are based primarily on the more neural differentiation exhibited by pPNET (11).

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Typically, when considered from a histopathological perspective, pPNETs possess similarities to a number of neoplasms, including rhabdomyosarcoma and small cell carcinoma (4).

Within the head and neck region, the larynx is a rare primary site for pPNET to arise (12). To the best of our knowledge, only a small number of cases have previously been reported (13-15). In these cases, follow-up periods were short and the details of treatment were not discussed. The current report presents a case of pPNET arising in the larynx, and subsequently reviews the associated literature.
A previously healthy 33-year-old female presented with the symptom of hoarseness ~6 months prior to admission. This symptom was progressive, and the patient was unable to speak during the initial consultation. Following clinical examination using a fiberscope, a slightly red tumor covered with white tissue was identified on the right false vocal cord. A punch biopsy was performed under fiberscopic view, and the tumor was initially diagnosed as being neuroendocrine. However, this diagnosis was unreliable due to the small size of the biopsy specimen. In addition, the initial report indicated that the tumor possessed low malignant potential. Computed tomography (CT) and magnetic resonance imaging (MRI) studies were unable to detect the tumor due to its small size. Consequently, microscopic surgery was performed under general anesthesia after admission to Nagoya Daini Red Cross Hospital on June 28, 2001. The base of the tumor mass was identified in the right laryngeal ventricle (Fig. 1), and dissected from the mucosa. The tumor was resected, and subsequently a potassium titanyl phosphate laser beam was applied to the tumor resection and the mucosa around the tumor. A diagnosis of pPNET was reached following pathological examination. Paraffin sections revealed sheets of small round cells, which possessed relatively regular nuclei and sparse cytoplasm. The nuclei demonstrated a salt and pepper chromatin pattern, and the nucleoli were obscured. Abortive Homer-Wright rosettes were rare, and no other specific patterns were identified. A relatively high frequency of mitotic figures was observed, while a Periodic Acid Schiff stain indicated that no glycogen was present (Fig. 2A). Immunohistochemical studies revealed that tumor cells were positive for cluster of differentiation 99 (CD99), neuron-specific enolase (NSE), S100 protein and vimentin, and negative for cytokeratin (AE1/AE3, CAM5.2), epithelial membrane antigen, muscle-specific actin, α-smooth muscle actin, desmin, leucocyte common antigen, chromogranin A and synaptophysin (Fig. 2B; Table I). An ultrastructural study revealed that the tumor cytoplasm contained a reduced number of mitochondria. Other organelles were not well developed; no electron dense core granules were detected, and nuclei were round with obscured nucleoli.

Following diagnosis, CT scans of the chest, abdomen and pelvis, an X-ray of all bones, an isotope bone scan, and an MRI scan of the brain and bone marrow, were normal. Hematological tests revealed a slight increase in the levels of lactate dehydrogenase and creatine phosphokinase.
Additional surgery with a wide marginal resection, including a total laryngectomy, was recommended; however, the patient declined. The patient was therefore treated with multiagent chemotherapy (ifosfamide, 50 µg/kg; vincristine, 2 µg/kg; doxorubicin, 500 µg/kg). The patient also received radiotherapy of a total dose of 60 Gy over 6 weeks.

The patient currently remains alive and healthy. Five years subsequent to treatment, the patient has exhibited no local disease recurrence or evidence of metastases.

**Discussion**

The concept of PNET has been the subject of considerable discussion in multiple cases (8,9). Initially, the term PNET was limited to tumors of the CNS. However, non-CNS tumors possessing a similar histological origin have been reported, and are known as pPNETs (3).

pPNET, with a primary site in the head and neck region, is rarely diagnosed (12,16-18). The most common primary site of pPNET is the chest wall, which is known as Askin tumor. The second most common primary site of pPNET is the pelvis (19,20). To the best of our knowledge, among the associated literature, the present case is only the fourth report of laryngeal pPNET (13-15). In addition, to the best of our knowledge, in the 5 years following the presentation of this case, no similar reports have been documented. Previous reported cases of pPNET arising in the larynx are summarized in Table II (13-15).

Histologically, typical pPNET is comprised of small round, blue-stained cells possessing hyperchromatic nuclei, with a high mitotic rate (21). The cytoplasm is indistinct, except in regions where cells are more mature, and the elongated hair-like cytoplasmic extensions coalesce to form rosettes. The majority of the rosettes contain a central solid core of neurofibrillary material known as a lobular or pseudorosette, for example, the Homer-Wright rosette (13,22).

The ultrastructural characteristics of a typical pPNET are primarily the presence of junctional complexes and confluent cell processes, which contain occasional neurosecretory type granules, microtubules and intermediate filaments (23).

Immunohistochemically, a number of authors have reported that typical pPNETs are positive for NSE and S100 protein staining, as well as additional markers, including Leu-7, synaptophysin, neurofilament (24), cytokeratin and chromogranin, which are positive at varying rates, suggesting variation in neural differentiation (25). ES and pPNET apparently arise in association with a translocation between chromosomes 11 and 22, specifically t(11;22)(q24;q12). These two tumors also express glycoprotein p30/32, coded for by the CD99 gene (HBA71 antigen or MIC2), and are recognized by commercially available antibody, O13 (19).

Anti-CD99 antibodies have been recognized as specific markers for the differential diagnosis of ES and pPNET, as these tumors demonstrate strong positive expression of the CD99 gene (26). However, a number of studies have reported that cases of bone and soft tissue tumors, including T-lymphoblastic lymphoma (27), poorly differentiated synovial sarcoma (28), small cell osteosarcoma (29), rhabdomyosarcoma (26), desmoplastic small round cell tumor (30), small cell carcinoma (31), Merkel cell carcinoma (32) and mesenchymal chondrosarcoma (33), also express CD99 gene products (24-31).

Therefore, differential diagnoses of ES and pPNET must be obtained from overall microscopic, ultrastructural and immunohistochemical findings. In addition, identification of a common cytogenetic abnormality t(11;22)(q24;q12) in these tumors provides strong support for diagnosis.

A number of previous studies have reported that pPNET is an aggressive disease, demonstrating high rates of local recurrence and distant metastases at an early stage. The most frequently observed sites of metastasis are the lungs and bone (34). Numerous patients exhibiting distant metastases at the time of diagnosis of pPNET possessed poor prognoses (35). The treatment for these pPNET cases was similar to that for rhabdomyosarcoma and Ewing's sarcoma; however, a highly effective treatment strategy for pPNETs remains to be established (36,37). In previous years, the prognosis of pPNET was poor, but has improved due to the administration of combined treatment strategies consisting of surgery, radiotherapy and chemotherapy (34,38).

The range of surgical resection is controversial as pPNET arises at a variety of sites, and the method of resection and surgical margin is site-specific. For tumors located in the head and neck, a wide surgical margin cannot be achieved. It has been reported that when pPNETs were resected with a surgical margin >10 cm, postoperative radiotherapy was not required (34). Regarding the larynx and with consideration of the anatomy in the present study, it was hypothesized that if the tumor did not invade the adjacent tissue, including the pharynx and thyroid, laryngectomy was a wide enough resection of pPNET, as the mucosa of the larynx was enclosed by thyroid and cricoid cartilage. In the present case, the patient refused a laryngectomy due to the fact that the resection was close to the surgical margin and further surgery may have been

**Table II. Review of reported cases of laryngeal peripheral primitive neuroectodermal tumors and the present case.**

<table>
<thead>
<tr>
<th>Study</th>
<th>Year</th>
<th>Patient age, years</th>
<th>Patient gender</th>
<th>Treatment</th>
<th>Metastases</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Jones et al</td>
<td>1995</td>
<td>9 months</td>
<td>M</td>
<td>S+C</td>
<td>None</td>
<td>(13)</td>
</tr>
<tr>
<td>Yang et al</td>
<td>2004</td>
<td>74</td>
<td>M</td>
<td>S+R</td>
<td>None</td>
<td>(14)</td>
</tr>
<tr>
<td>Lynch et al</td>
<td>2014</td>
<td>45</td>
<td>F</td>
<td>C+R</td>
<td>None</td>
<td>(15)</td>
</tr>
<tr>
<td>Present case</td>
<td></td>
<td>33</td>
<td>F</td>
<td>S+C+R</td>
<td>None</td>
<td></td>
</tr>
</tbody>
</table>

C, chemotherapy; R, radiation therapy; S, surgery; M, male; F, female.
required. A number of previous studies have recommended resection of pPNETs, with a wide surgical margin. It has been reported that patients who underwent a resection with a wide surgical margin, demonstrated improved overall survival, compared with those who underwent a less-than-wide resection (39). Additionally, for patients with localized tumors, wide resection may facilitate favorable prognoses. Additional postoperative radiotherapy may be capable of controlling local disease when microscopic surgical margins remain pathologically positive (34). The resection of all gross pPNET tumors within 3 months of diagnosis is correlated with significantly improved disease-free survival rates (20).

Furthermore, multiagent chemotherapy has been recommended for the control of distant micrometastases by numerous previous studies. Since the 1980s, preliminary reports have suggested vincristine, ifosfamide, cyclophosphamide and doxorubicin as chemotherapeutic agents for the treatment of metastasis of Ewing's sarcoma. Identical chemotherapeutic strategies have been performed on pPNETs, and the responses were similar to those identified in Ewing's sarcoma, resulting in improved rates of survival (10,35,38,40). However, the optimal chemotherapeutic agents for the treatment of pNET remain under investigation.

In previous studies, the prognosis of pPNET has been demonstrated to be poor, although certain studies have demonstrated improvements (34,39). Current literature indicates that the overall 5-year survival rate is ~40-60%, whereas this figure was 48% subsequent to 1970, and 28% prior to this (34). In recent reports, 5-year survival rate is around 60% (41-43). However, an improved prognosis has been demonstrated by patients exhibiting pPNET arising in the head and neck region, compared with those patients with tumors at alternative sites (35,44,45). In a previous study, 5 cases of pPNET were reported to have arisen at head and neck sites. In these cases, an improved prognosis was demonstrated, compared with tumors that arose at alternative sites, and all 5 patients had survived 18 months later (46). This result suggested that complete tumor resection with a wide margin, followed by local irradiation and systemic chemotherapy may be concluded to provide an improved prognosis for patients exhibiting pPNET, compared with that of alternative treatment strategies.

In conclusion, pPNET in the head and neck region is rare, particularly in the larynx. pPNET treatment strategies consisting of combinations of surgery, chemotherapy and radiation remain controversial. Further study is required in order to provide clearer evidence regarding the optimal treatment strategy for pPNET.

References


