Case report

Biphenotypic sinonasal sarcoma with PAX3/FOXO1 fusion

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Biphenotypic sinonasal sarcoma (BSNS), previously known as low-grade sinonasal sarcoma, is a rare tumour of the sinonasal tract, first described in 2012. It involves both myogenic and neural differentiation and is characterized by PAX3 rearrangement. MAML3 is the most frequent fusion partner of PAX3; however, its partner remains unidentified in a subset of cases. These tumours have significant local recurrence rates but lack metastatic potential. Here, we report a case of BSNS with PAX3/FOXO1 fusion and discuss its clinicopathological features and differential diagnosis.

Key words: biphenotypic sinonasal sarcoma, PAX3/FOXO1, low-grade neoplasia.

Introduction

Biphenotypic sinonasal sarcoma (BSNS), previously known as low-grade sinonasal sarcoma, is a rare tumour of the sinonasal tract, which was first described in 2012 [1]. There are case series consisting of 28 patients, 44 patients, 41 patients, 11 patients, 15 patients, and 3 patients that have been reported in the literature to date [1–6], of which there were 5 cases showing PAX3/FOXO1 fusion; 3 cases in a series of 44 patients, and one case in a series of 41 patients. In addition to these, there is also one case report in the literature [2, 3, 7] (Table I). It expresses both neural and myogenic markers, affects middle-aged adults, and is more common in females than in males [8]. A recurrent PAX3-MAML3 fusion event is present in most BSNS cases, with a few cases showing alternative fusion of PAX3 with NCOA1, NCOA2 WWT-1, and FOXO1 [2, 3]. Here, we report a particular case of BSNS with a PAX3/FOXO1 fusion in the nasal cavity.

Case report

A 66-year-old female presented with a mass in the left choanal area of the nasal cavity. The medical history revealed that she had undergone bilateral mastectomy, radiotherapy, and chemotherapy for breast carcinoma. The patient also had diabetes mellitus, hypertension, and hypothyroidism. Magnetic resonance imaging (MRI) revealed a mass in the left nasal cavity measuring approximately 3.5 cm, filling the entire left frontal sinus, and extending to the left maxillary sinuses. Positron emission tomography showed minimal or mild uptake of the mass, with an SUVmax of 3.3. Intraoperatively, the mass was found to fill the left nasal cavity, and intracranial extension was suspected. Frozen pathology revealed a spindle cell neoplasm without severe atypia or mitotic activity. The decision was made to end the initial procedure and await final surgical pathology. Microscopic evaluation of paraffin blocks revealed a monotonous spindle cell lesion (Fig. 1). Uniform spindle cells with bland nuclear features

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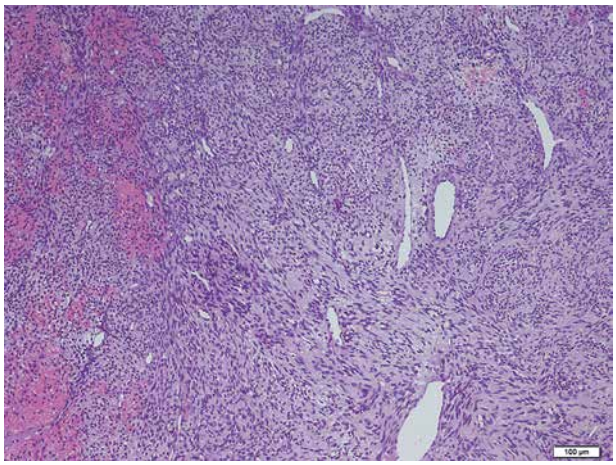

Fig. 1. Spindle cell lesion consisting of monotonous cells

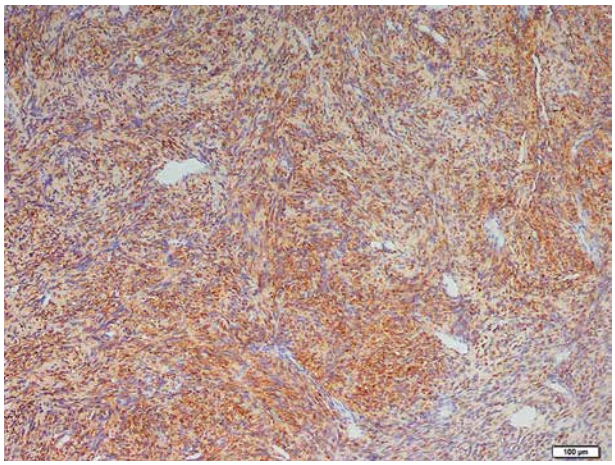

Fig. 2. S100 positivity in tumour cells

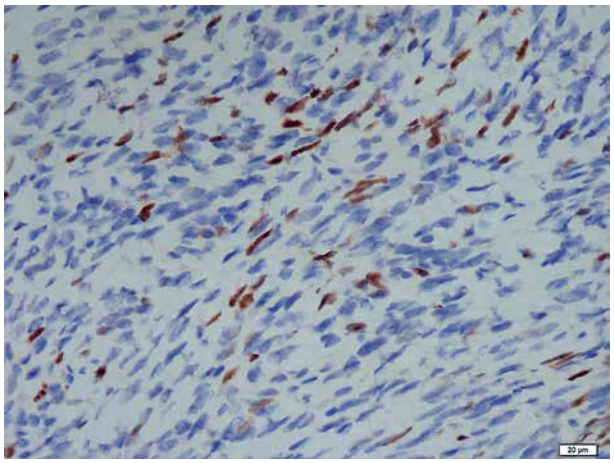

Fig. 3. Focal myo-D1 positivity in tumour cells

Fig. 4. The resection material was measured at 3.8 × 2.5 × 1.8 cm

were arranged in short fascicles with no mitotic figures, cellular atypia, or necrotic areas. Immunohistochemical staining revealed that the tumour cells expressed S100 (Fig. 2), focal desmin, focal myo-D1 (Fig. 3), diffuse TLE-1, and focal nuclear β-catenin. No expression of myogenin, pan cytokeratin, SOX10, STAT-6, CD34, Melan-A, or HMB-45 was observed. Synovial sarcoma was excluded based on negative SS18 FISH and SS18/SS-X1-SS-X2 fusion real-time polymerase chain reaction analyses. Due to focal MyoD1 positivity, FOXO1 molecular analysis was performed to exclude alveolar rhabdomyosarcoma. Fluorescence in situ hybridization (FISH) was performed using a ZytoLight SPEC FOXO1 Dual Color Break Apart Probe, Z-2139-50, (Zytovision, Bremerhaven, Germany). The orange fluorochrome direct-labelled probe hybridizes distally, and the green fluorochrome direct-labelled probe hybridizes proximally to the *FOXO1* gene (13q14.11). Deparaffinization, prehybridization, and hybridization were performed according to the manufacturer's datasheet. One hundred tumour cells were analysed using a fluorescence microscope (Leica DM 2500). The cells were captured using a computer system with a digital camera (ArgenitAKAS Imaging, Turkey). Signals from the tumour cell nuclei were counted, and the presence of redgreen breakapart signals was recorded. FISH analysis was positive for *FOXO1* gene rearrangement.

Real-time polymerase chain reaction was performed to detect the fusion partner of FOXO1 and PAX3-FOXO1 [t (2;13) (q35; q14)] translocation in the patient's sample. A 205 bp amplification product containing a fusion between PAX3 exon 7 (NM_181458.4) and FOXO1 exon 2 (NM_002015.4) was confirmed by Sanger sequencing. Due to co-expression of neural and myogenic markers, the tumour was diagnosed as BSNS, which was confirmed by molecular testing. After diagnosis, the mass was excised totally. The resection material measured 3.8 × 2.5 × 1.8 cm (Fig. 4). Histopathological evaluation revealed monotonous spindle cell proliferation without atypia, necrosis, or mitosis. We performed desmin and S100 immunohistochemical staining again and found the same immunohistochemical profile as that of the biopsy material. The diagnosis was confirmed.

Table I. Biphenotypic sinonasal sarcoma cases showing PAX3-FOXO1 fusion reported in the literature to date

Case	Age	Gender	Fusion	Localization
1. Fritchie *et al*. [2]	31	F	*PAX3-FOXO1*	Nasal/ethmoid
2. Wong *et al*. [7]	33	M	*PAX3-FOXO1*	Nasal/sphenoid
3. Le Loarer *et al*. [3]	39	M	*PAX3-FOXO1*	Nasal cavity
4. Fritchie *et al*. [2]	47	F	*PAX3-FOXO1*	Nasal cavity
5. Fritchie *et al*. [2]	35	M	*PAX3-FOXO1*	Sinonasal
6. Tosun *et al*. (present case)	66	F	*FPAX3-FOXO1*	Nasal/frontal

Discussion

Biphenotypic sinonasal sarcoma, previously known as low-grade sinonasal sarcoma, is a rare tumour of the sinonasal tract. It is a rare entity that affects middle-aged adults and is more common in females than in males [9]. It was included in the most recent edition of the World Health Organization classification of head and neck tumour – β-version in 2022 [10]. Only 6 series have been published with 28, 44, 41, 11, 15, and 3 cases, respectively [1–6]. Among these cases, 5 showed PAX3/FOXO1 fusion. Biphenotypic sinonasal sarcoma is a challenging diagnosis because of its rarity. Differential diagnoses include low-grade spindle cell neoplasms, such as monophasic synovial sarcoma, low-grade malignant peripheral nerve sheath tumour (MPNST), schwannoma, solitary fibrous tumour, and benign reactive proliferation. It is a large spectrum that sometimes requires molecular techniques for diagnosis. Monophasic synovial sarcomas may exhibit histological and immunohistochemical features that are similar to those of BSNS. However, synovial sarcomas in the sinonasal tract are extremely rare, with only 12 cases reported to date [11]. TLE1 positivity may also lead to confusion. Our case was diffusely positive for TLE-1; thus, we needed to perform molecular studies on SS18 rearrangements. However, the results were negative. Schwannomas are S100-positive tumours that can mimic BSNS. Schwannomas are characterized by hypocellular and hypercellular Antoni A–B areas, nuclear palisading (Verocay bodies), and perivascular hyalinization. Although schwannomas are diffusely positive for S100, they also exhibit diffuse SOX10 expression [12]. Our case was positive for S100 but negative for SOX-10. Distinguishing between BSNS and low-grade MPNST in the sinonasal region is extremely difficult and requires further molecular studies. S100 positivity was observed for both entities. Limited SOX10 staining would favour a malignant peripheral nerve sheath tumour because BSNS should be negative for this marker [13]. Our case was negative for SOX10. Solitary fibrous tumours (SFT) have similar uniform spindle cell proliferation and a staghorn vascular pattern but can be distinguished from BSNS by the presence of ropey bundles of collagen rather than the delicate collagen strands that are seen in BSNS [6]. Moreover, SFTs are positive for CD34 and are characteristically positive for STAT6 (nuclear) [14]. Like BSNS, spindle cell rhabdomyosarcoma has a herringbone/fascicular architecture, but the mitotic rate and degree of atypia in the latter are usually more significant [13]. Spindle cell rhabdomyosarcoma shows more extensive desmin and MYO-D1 expression than BSNS [15]. A recurrent PAX3-MAML3 fusion event is present in most BSNS cases, with a few cases showing an alternative fusion of PAX3 with NCOA1, NCOA2, WWT-1, and FOXO1 [3]. In the largest series, 24 cases were positive for PAX3-MAML3 (55%), whereas 15 showed rearrangements of PAX3 without MAML3 involvement (34%). Of the 15 cases with PAX3 involvement only, 5 were found to harbour PAX3-FOXO1. Histomorphologically, cases with PAX3/FOXO1, PAX3/NCOA1 fusion, and no detectable PAX3 fusions have demonstrated the typical spindle-shaped cell infiltrative pattern of BSNS, characterized by high cellularity and a fascicular architecture. All cases with the PAX3/FOXO1 fusion showed positive expression of S100, SMA, and MYO-D1, whereas desmin and myogenin were negative. In contrast, cases with the PAX3/MAML3 fusion showed positive expression of myogenin [2]. In the second largest study, by Le Loarer *et al*. [3], molecular analysis revealed a PAX3-MAML3 transcript in 37 cases (90%). RNA sequencing was performed in 4 cases that were negative for the PAX3-MAML3 fusion and one case harboured the PAX3-FOXO1 fusion [4]. Our case showed PAX3/FOXO1 fusion. Identification of the PAX3-FOXO1 fusion in both BSNS and alveolar rhabdomyosarcoma is challenging for differential diagnosis. The presence of the PAX3/MAML3 fusion in BSNS is diagnostic, but PAX3/FOXO1 and PAX3/NCOA1 fusions are also observed in alveolar rhabdomyosarcoma [9, 16, 17]. The discovery of the PAX3/FOXO1 fusion in both BSNS and alveolar rhabdomyosarcoma indicates that these 2 diseases share a common genetic basis, despite arising from distinct cellular precursors [7].

The reprogramming of PAX3/FOXO1 in alveolar rhabdomyosarcoma leads to the expression of early myogenic markers such as myogenin and MYOD1. Conversely, PAX3/MAML3 reprogramming induces the expression of genes involved in neurogenic differentiation, which is characteristic of BSNS [7, 18]. Alveolar rhabdomyosarcoma is an extremely aggressive malignancy of primitive-appearing cells that most commonly arises in the extremities of adolescents and young adults [19]. Biphenotypic sinonasal sarcoma are low-grade spindle cell malignancies that present in middle adulthood. Rhabdomyosarcoma consistently expresses the rhabdomyoblastic regulatory proteins, myogenin and MYO-D1. In BSNS, positivity for MyoD1 is more common than that for myogenin [3, 18]. Our case was also focally positive for MYO-D1 and negative for myogenin. Finally, a group of NTRK-rearranged spindle cell mesenchymal neoplasms has recently been identified in the literature. Co-expression of S100 protein and CD34 is characteristic [13]. Pan-TRK immunostaining appears to be a sensitive marker for this group of tumours but is not specific because BSNS may also show pan-TRK expression [20].

Conclusions

Biphenotypic sinonasal sarcoma, a recently recognized tumour, is a low-grade spindle cell sarcoma with neural and myogenic differentiation and PAX3 rearrangement with MAML3 or other partners, such as FOXO1, WWTR1, NCOA1, and NCOA2. PAX3-FOXO1 fusion is rare. Despite its rarity, it has many differential diagnoses that should be considered. Further research and cases are necessary to identify the distinct morphological and immunohistochemical features, as well as their prognostic factors of BSNS harbouring a PAX3/FOXO1 fusion.

The authors declare no conflict of interest.

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