CORRESPONDENCE

Malignant transformation of fibrous dysplasia into osteosarcoma confirmed with *TP53* somatic mutation and mutational analysis of *GNAS* gene

Sir,

The World Health Organization (WHO) classification of bone and soft tissue tumours describes fibrous dysplasia (FD) as a neoplastic fibro-osseous lesion composed of randomly distributed irregular, curvilinear trabeculae of woven bone, without prominent osteoblastic rimming set in a background of bland fibroblastic cells.¹ It is associated with activating missense mutations in the *GNAS* gene (20q13) resulting in inhibition of intrinsic GTPase activity of G protein and upregulation of intracellular cAMP. *GNAS* mutations are specific to FD among other fibro-osseous lesions of bone and have been identified in both monostotic and polyostotic types.^{2,3}

Malignant transformation of FD into osteosarcoma is rare. Only two cases in the literature have documented point mutations in *GNAS* in both the benign and malignant components, confirming a clonal relationship between FD and osteosarcoma.^{4,5} Herein, we report an additional case of FD with transformation to high grade osteosarcoma, diagnosed preoperatively on a small biopsy supported by *GNAS* and *TP53* mutations detected with next generation sequencing (NGS).

A 21-year-old man with suspected FD of the right maxilla (incidental discovery on imaging 8 years prior) presented with sudden right maxillary swelling, pain and paraesthesia. Computed tomography (CT) scan revealed destruction of the anterior maxillary wall by an enhancing soft tissue mass, 31×24×22 mm. A pre-lacrimal endoscopic biopsy was performed, showing an atypical fibro-osseous lesion composed of a moderately cellular population of plump spindle cells in vague storiform patterns set in fibrocollagenous stroma. Associated with the stroma were randomly placed, short, irregular spicules of variably mineralised woven bone, without osteoblastic rimming. The spindle cells demonstrated ovoid, mild to moderately pleomorphic and focally hyperchromatic nuclei with coarse chromatin, small nucleoli and moderate volumes of pale eosinophilic cytoplasm. Mitotic activity was increased in areas, with up to 6 mitoses counted per 10 consecutive high power fields (HPF; field area 0.23 sq mm) (Fig. 1). Immunohistochemistry showed diffuse staining of the spindle cells for p53 and weak, patchy staining for smooth muscle actin. Negative markers included CK8/18, EMA, S100, desmin, CD34, STAT6 and MUC4. The Ki-67 proliferation index was elevated, estimated at 25.0% in foci of highest density staining.

NGS was performed using the TruSight Tumour 26 panel (Illumina, USA) showing a point mutation in *GNAS* at codon 201 (Arg201Cys) at an allele frequency of 30.0% and a separate point mutation in *TP53* at codon 281 (Asp281Asn)

at an allele frequency of 46.0%. Interphase FISH analysis showed no evidence of *MDM2/CDK4* amplification.

Whilst definitive diagnosis was challenging on purely morphological grounds, the presence of both *GNAS* and *TP53* mutations allowed for a more confident diagnosis of FD with malignant transformation to osteosarcoma. The patient was commenced on a chemotherapy protocol specific for osteosarcoma. However, neoadjuvant therapy was abandoned due to symptomatic rapid growth of the tumour with further local destruction of the maxillary sinus wall on imaging and an urgent right maxillectomy was performed (Fig. 1). Histopathology confirmed FD with malignant transformation to high grade osteosarcoma. Only small areas of conventional residual FD were seen composed of the typical curvilinear arrays of woven bone set in a mildly cellular stroma of bland spindled fibroblastic cells. For the most part, the mass was formed by osteosarcoma, in which a moderate to highly cellular population of malignant spindle cells were randomly placed and occurred in vague storiform patterns. The spindle cells demonstrated moderate to highly pleomorphic and hyperchromatic nuclei with coarse chromatin, small nucleoli and moderate volumes of pale eosinophilic cytoplasm. Mitotic activity was brisk, with up to >25 mitoses counted per 10 consecutive HPF (field area 0.23 sq mm) (Fig. 1).

The *TP53* variant previously detected in the tumour was not detected in matched normal tissue (submandibular gland) consistent with somatic (not germline) mutation. The patient is currently well with no signs of recurrence at 10 months post-surgery.

FD is a benign, medullary, fibro-osseous lesion which may involve one or more bones. FD is characterised histologically by randomly distributed irregular, curvilinear trabeculae of woven bone, without prominent osteoblastic rimming set in a background of bland fibroblastic cells. FD occurs in children and adults with equal sex distribution. The monostotic form is 6–10 times more common than the polyostotic form.³ Sites affected include femur, pelvis, skull and tibia. Malignant transformation accounts for 0.4–1.0% of all FD cases.^{6,7} A case series of 266 patients with a definitive diagnosis of FD in a Chinese population showed that malignant transformations only occurred in three patients with polyostotic forms.⁷ Another case series from Mayo Clinic of 1122 cases with a histological diagnosis of FD had malignant transformations in 19 cases of monostotic FD and 9 cases of polyostotic FD.⁸ The most common malignant changes are osteosarcoma, chondrosarcoma and fibrosarcoma.⁶ Two case series have suggested a relationship with prior radiation, but most of the other case series have not investigated this relationship.^{8,9}

FD is associated with activating missense mutations in *GNAS* (20q13). These mutations are specific to FD among other fibro-osseous lesions of bone and have been identified in both monostotic and polyostotic types. Up to 95% of *GNAS* mutations are point mutations that replace arginine at codon 201 with either cysteine or histidine. In the remaining

Fig. 1 (A) CT scan of the face in 2011 showed expansion of the right maxillary sinus with ground glass density consistent with fibrous dysplasia. (B) Repeat CT scan in 2019 showed significant interval progression with increasing size and perforation through the anterior maxillary wall into the facial soft tissues. (C) Fibrous dysplasia (left of field) with malignant transformation to high grade osteosarcoma (right of field), and corresponding P53 immunohistochemistry (top right of field). (D) Areas of fibrous dysplasia showed typical arrays of woven bone surrounded by a bland spindle cell stroma. (E,F) Osteosarcomatous transformation comprised cellular populations of malignant spindle cells associated with spicules of mineralised osteoid matrix and (E) corresponding P53 immunohistochemistry (top right of field).

5%, glutamine replaces leucine at codon 227.^{1,2} The sensitivity of detecting *GNAS* mutations in FD, depending on the molecular technique, varies from 45% up to 100%.²

In contrast to other forms of fibro-osseous lesions, the specificity of *GNAS* mutations in fibrous dysplasia is high. In a larger study encompassing an array of fibro-osseous lesions of bone, *GNAS* mutations were found only in fibrous dysplasia.^{2,3} They do not occur in low grade osteosarcoma.^{2,3} Malignant transformation in fibrous dysplasia of the maxilla is rare, a recent review identifying 26 cases. Of the 26 cases, only two case reports confirmed the presence of *GNAS* mutations in the malignant component.⁶ Thus far,

there are only two published reports addressing the presence or absence of *GNAS* mutations in the malignant component of transformed fibrous dysplasia, one in the femur and one in the maxilla.^{4,5} Our case represents the third, also affecting the maxilla.

It is important to confirm the *GNAS* status in cases of FD as well as in the malignant component so as to confirm the initial diagnosis and to prove that the malignant tumour was derived from FD.⁶ The authors have found limited published studies that address this.

TP53 mutations were detected in up to 88% of osteosarcomas¹⁰ and one study demonstrated this mutation in low

grade osteosarcoma arising from FD in the femur.⁵ Immunohistochemically, this FD was negative for p53 and MDM2, whereas the low grade osteosarcomatous component was positive for both p53 and MDM2.⁵ Both studies, including ours, demonstrated *TP53* mutations. *TP53* mutation was central to the diagnosis of osteosarcoma, which would have been difficult on morphology alone, especially so in small biopsies. This was important as neoadjuvant chemotherapy is the standard of care for osteosarcoma as it has been shown to improve event free survival and overall survival.¹¹

Other factors that limited the diagnosis of osteosarcoma in small biopsies were small sample size, not being representative of the entire lesion and sampling underdeveloped atypia instead. In our case, *TP53* mutation was detected in the small biopsies taken, which was used to support the diagnosis of osteosarcoma.

In summary, this is a rare report of high grade osteosarcoma arising from FD of the maxilla supported by *GNAS* and *TP53* mutation analysis. Our findings suggest that *GNAS* and *TP53* mutations can be used to support osteosarcomatous transformation in fibrous dysplasia.

Conflicts of interest and sources of funding: The authors state that there are no conflicts of interest to disclose.

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DOI: <https://doi.org/10.1016/j.pathol.2020.08.027>

Two cases of parosteal osteosarcoma with unusual histological and molecular features

Sir,

Osteosarcomas arising from the cortical surface of bone can be subdivided into parosteal, periosteal, and high grade surface osteosarcomas, with parosteal osteosarcomas being the most well differentiated.¹ Clinically, parosteal osteosarcomas show female predominance and generally occur in the third decade of life, with the distal femur, proximal tibia, and proximal humerus being the common sites of origin.² In general, they are low grade tumours with good prognosis.³ Radiologically, they appear as a lobulated mass with a broad base of attachment to the bone, and irregular mineralisation¹; a cleavage plane between the tumour and the bone is sometimes noted.⁴ On gross examination, they appear as ossified masses, which may have a cartilaginous cap; soft areas representing dedifferentiation may be present.¹ Classically, they are described as having a biphasic histological appearance with parallel trabeculae of woven bone, with intervening spindle cells showing moderate atypia and collagenous stroma; this is the so called ‘streamer pattern’. Occasionally, other histological patterns are encountered, which may resemble desmoplastic fibroma or fibrous dysplasia.¹ Dedifferentiation (presence of high grade areas arising from a classic parosteal osteosarcoma) may occur, and is associated with a poorer prognosis.⁵ Molecular studies show parosteal osteosarcomas to be associated with amplification of Cyclin dependent kinase 4 (*CDK4*) and Mouse double minute 2 (*MDM2*), with 87–100% of cases positive for either (or both) of these markers by immunohistochemical staining in two studies,^{6,7} and amplification of *MDM2* by fluorescent *in situ* hybridization (FISH) seen in 83% of cases examined in a 2012 work.⁸ As such, cases which present without the classical histological pattern or without typical molecular changes may mimic benign lesions and present a diagnostic challenge. We present two cases of parosteal osteosarcoma with atypical histological features diagnosed recently in our institution, and expand the morphological spectrum of parosteal osteosarcoma.

The first case is that of a 21-year-old Chinese female, with no relevant past medical history, who presented with a 2 year history of posterior left knee pain, swelling, and limited range of motion. She had tried acupuncture to no effect, and underwent magnetic resonance imaging (MRI) in a neighbouring country, revealing a 3.1 cm lobulated mass apparently arising from the left posterior knee joint. She presented to our institution seeking a second opinion where plain radiographs were performed, demonstrating a bony lesion with circumferential sclerosis arising from the posterior aspect of the left lower femur, with ill-defined cortical margins and no association with any soft tissue mass. Radiological differential diagnoses considered were chondromyxoid tumour and parosteal osteosarcoma (Fig. 1A,B). Computed tomography scan of the leg revealed a 2.8 cm exophytic lesion at the posterior aspect of left lower femur with a rim of ossification. At this point the differential diagnoses considered by the clinical team included osteochondroma, parosteal osteosarcoma, chondromyxoid tumour, or chondrosarcoma. She underwent *en bloc* resection of the mass, and the post-operative