Adamantinoma-like variant of Ewing sarcoma in the metatarsal bone after chemotherapy: Report of a case successfully treated with an osteocutaneous fibular transfer.

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Case Report

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Abstract

Adamantinoma-like Ewing sarcoma is a rare variant of Ewing sarcoma known to have histologic and immunohistochemical evidence of squamous differentiation. This variant most commonly occurs in the head and neck region with a few cases reported in long bones of the limbs. It may be associated with poorer clinical outcome and could pose a diagnostic challenge(s) particularly if it occurs in older patients or as a metastatic lesion. We present a case of Ewing sarcoma in the metatarsal of an 11 year old boy that manifested adamantinoma-like morphology after neo-adjuvant chemotherapy. Chemotherapy has been reported to induce neuronal maturation and rhabdoid morphology in cases of Ewing sarcoma, but no reports of treatment induced squamous differentiation with P40/P63 expression have been demonstrated. This is also the first documented case to use a pedicled osteocutaneous fibular transfer in a metatarsal malignancy, which is usually treated by either ray or below knee amputation.

Background

Ewing sarcoma is a small round cell sarcoma typically arising in the diaphyseal and metadiaphyseal regions of long bones, predominantly in children and young adults, but can also be seen in soft tissues (particularly of the trunk). It shows gene fusions involving one member of the EFT family of genes and a member of the ETS family of transcription factors. Up to 85% of cases harbour a t(11;22)(q24;q12) resulting in EWSR1-FLI1 gene fusion and up to 25% of cases express cytokeratins (1–6). Numerous histological variants exist, namely classic, atypical/large cell, sclerosing, spindle cell sarcoma-like and also one that shows squamous differentiation, called adamantinoma-like Ewing sarcoma. This latter variant, presents more commonly in the head and neck region with only a few reported cases which involved the appendicular skeleton (7,8,17–23,9–16). With this case report we present a metatarsal tumour that showed adamantinoma-like features with P40/P63 expression after neo-adjuvant chemotherapy.

Case Presentation

An eleven-year-old male patient presented with the history of a painless mass involving the first metatarsal of the left foot that was gradually enlarging over the preceding year. Local examination confirmed a firm, 80 x 30mm mass involving the first metatarsal of the left foot. Ankle range of motion was full and painless. Systemic examination did not reveal any abnormalities and laboratory investigations were unremarkable.

Plain radiographs showed mixed lytic and sclerotic changes involving the entire first metatarsal, with an indistinct permeative appearance of the cortex, associated periosteal reaction and a subtle soft tissue component (Figure 1A). A MRI scan showed a diffuse aggressive destructive process involving the entire first metatarsal, with heterogeneous medullary cavity enhancement, an aggressive periosteal reaction and breach of the cortices. The lesion appeared heterogeneous, hypo- and isointense on T1 weighted images, and heterogeneous hyperintense on T2 weighted images. An associated soft tissue component encased
the metatarsal and illustrated post contrast enhancement (Figure 1B). Systemic staging included an F-18 FDG PET/CT scan that showed several skeletal lesions including to the left humerus, lumbar spine and pelvis. An incisional biopsy confirmed the diagnosis of Ewing sarcoma with EWSR1 rearrangement with FISH.

The patient received emergency radiotherapy of the spinal column prior to commencement of neo-adjuvant chemotherapy after developing symptoms related to spinal cord compression secondary to skeletal metastases. Post radiotherapy, neo-adjuvant chemotherapy was commenced according to the Children's Oncology Group Ewing Sarcoma Protocol (AEWS0031). Duration of this chemotherapy regimen spans 48 weeks and compromises courses of Vincristine (V) (1.5mg/m²/dose), Doxorubicin (D) (75mg/m²/dose), Cyclophosphamide (C) (1.2g/m²/dose) (VDC) alternating at intervals with courses of Ifosfamide (1.8g/m²/day for 5 days per course) and Etoposide 100mg/m²/day for 5 days per course) (IE). The patient received 5 cycles of neo-adjuvant chemotherapy comprising VDC/IE before local and systemic staging was repeated to assess response to the chemotherapy. A repeat MRI scan confirmed the permeative destructive process of the first metatarsal with interval decrease in size of the associated soft tissue component (Figure 1C). The follow-up PET/CT showed evidence of residual disease in the known primary of the left foot with no evidence of disease elsewhere.

**Figure 1.** Antero-posterior radiograph showing a poorly defined mixed lytic and sclerotic lesion of the left first metatarsal (A). Magnetic resonance imaging scan showing a permeative destructive process involving the first metatarsal and large soft tissue component encasing the metatarsal (B) and subsequent interval decrease in size following neo-adjuvant chemotherapy (C).

Definitive surgical management consisted of wide excision of the first metatarsal through a dorso-medial approach including resection of the biopsy tract (Figure 2A). Reconstruction of the bone and soft tissue defect was accomplished by an ipsilateral pedicled osteocutaneous fibula flap (Figure 2B). Although amputation was considered given the high risk of positive margins, the decision to perform wide resection and reconstruction was based on extensive discussion between the patient, orthopaedic oncological surgeon and paediatric oncologist. Another contributing factor was that the patient presented with several skeletal metastases that showed interval decrease in size on the repeat MRI. Once all wounds had healed adjuvant chemotherapy, consisting of VDC and IE, was re-commenced and weight-bearing was allowed in a supportive boot. Clinical review at 3 months found a plantigrade sensate foot with no instability of the hallux (Figure 2C). On completion of the chemotherapy regimen, the patient will receive adjuvant radiotherapy for positive surgical margins.

**Figure 2.** Wide resection of the first metatarsal through an antero-medial approach (A). Soft tissue and bony defect reconstructed with an ipsilateral pedicled osteocutaneous fibula flap (B). Plantigrade foot with healed cutaneous flap, three months after surgery (C).

**Histology**

Initial biopsy:
The pre-treatment incisional biopsy consisted of a single fragment measuring 16 x 9 x 7mm and showed a lesion composed of invasive nests of uniform small round cells with round nuclei containing finely stippled chromatin and inconspicuous nucleoli, scant clear to eosinophilic cytoplasm and indistinct cytoplasmic membranes (Figure 3A). Immunohistochemical studies showed membranous expression of CD99 and nuclear expression of FLI-1 in the tumour cells. FISH revealed rearrangement of the EWSR1 gene.

Resection specimen:

Macroscopically the specimen consisted of the left first metatarsal with overlying skin and surrounding soft tissue and measured 60 x 50 x 35mm. On cut section a white-grey lesion was present in the periosteal soft tissue with areas of haemorrhage. Microscopically residual tumour was mostly present in the soft tissue inferio-medial to the metatarsal with scant microscopic foci of residual tumour in the medullary cavity of the metatarsus. Histologically it showed typical features of Ewing sarcoma as seen on the initial biopsy but now with wide-spread squamous differentiation in the form of prominent eosinophilic cytoplasm and frank keratin pearl formation (Figure 3B). Areas of necrosis, hemosiderin-laden macrophages, foamy macrophages, calcification and stromal fibrosis were observed which related to treatment effect/response. Immunohistochemical studies showed CD99 and FLI-1 expression as seen in the previous biopsy. Additional immunohistochemical stains were performed on both (pre- and post- chemotherapy) specimens and included: AE1/AE3, CK5, 34βE12, P63 and P40 (Table 1). AE1/AE3 showed positive staining in both specimens but the P63 and P40 (Figure 4) were only positive in the resection specimen on both decalcified and non-decalcified tissue. CK5 and 34βE12 showed reactivity in single isolated cells in the initial biopsy but was diffusely positive in the resection specimen. Desmin, WT1, ERG and S100 immunohistochemical stains were negative and ruled out the possibility of other small round cell lesion such as a desmoplastic small round cell tumour that can also express CD99 and cytokeratins.

**Figure 3.** Microscopic image of the initial biopsy. Ewing's sarcoma showing a 'classic' growth pattern of nests of small blue round cells; H&E 100x (A). Microscopic image of the post neo-adjuvant chemotherapy resection specimen. Adamantinoma-like Ewing's sarcoma with squamous differentiation and keratin pearl formation; H&E 100x (B).

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<tr>
<th>Immunohistochemical stain</th>
<th>Initial biopsy</th>
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<tr>
<td>CD99</td>
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<td>FLI-1</td>
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<td>34βE12</td>
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**Table 1.** Immunohistochemical staining profile: + (positive in single cells), ++ (diffusely positive), - (negative).
**Discussion**

In this case the initial incisional biopsy showed morphological features in keeping with 'classic' Ewing sarcoma, but with morphological and immunophenotypical changes to an adamantinoma-like Ewing sarcoma variant post chemotherapy. We considered that this change may be due to one of the following: 1. Under sampling of the tumour on the initial incisional biopsy; 2. Emergence/persistence of a more chemotherapy resistant clone; or 3. Alterations in the tumour morphology &/or immunophenotype caused by chemotherapy.

To address the question of chemotherapy related alterations: Chemotherapy response of Ewing sarcoma is graded according to the Paediatric Oncology Groups Study (24) using the typically seen changes of necrosis, haemorrhage, cystic degeneration, calcification, ossification and fibrosis. The grading of chemotherapeutic response has prognostic and therapeutic significance as it plays a role in chemotherapy agent selection for the continuation chemotherapy (24–27).

Other reported morphological changes in Ewing sarcoma post chemotherapy include: neuronal maturation with rosette formation and gangliocytic phenotype differentiation but rhabdoid change has also been described though the tumour maintained its immunophenotype (28,29). Knezevich et al. described loss of EWS/FLI-1 gene fusion in the recurrent tumour (30), while Smith et al. described a different genetic alteration in one of the tumour nodules (31). No case reports of squamous differentiation (including keratin pearl formation) with P40/P63 expression post chemotherapy could be found on a literature search (32).

In our case the original tumour biopsy, even though it might not have been representative of the entire tumour, was P40 & P63 negative and only showed expression of these immunomarkers on the resection specimen (post neo-adjuvant chemotherapy). CK5 and 34\βE12 showed diffuse staining in the resection specimen while only single isolated cells were reactive in the initial biopsy. Classic Ewing sarcoma, in 25% of cases, may express keratins such as CAM5.2, AE1/AE3 and MNF-116 (3,4).

EWS fusion was positive on the initial biopsy using FISH, but the translocation partner remains uncertain as dual fusion FISH probes and PCR are not available in our setting.

It would hence appear that this phenotype and immunophenotype was not present initially and only appeared/persisted post-chemotherapy.

One research paper suggested that adamantinoma-like Ewing sarcoma may be associated with a more aggressive clinical behaviour and poorer outcome (7). If in our case, the ALES is indeed due to
persistence of a more resistant clone, this could support this claim but additional research is needed. Apart from the unique histological findings of this case, a novel reconstructive strategy was also employed. Reconstruction of the first metatarsal is more critical than the lesser metatarsals given its role in progression through the gait cycle. In the context of trauma, first ray reconstruction using both free and pedicled fibula flaps was described as early as 1991(33). Wang et al. described a series of four patients who had metatarsal reconstruction, two of the first ray and two of the lesser, using pedicled fibula flaps (34). More recently Hilaire (35) described computer assisted virtual surgical planning in a case of first metatarsal reconstruction using a free fibula flap. Malignancy of the metatarsals, however, has limited options in terms of salvage surgery. Management of primary bone tumours of the lesser metatarsals usually consist of ray (36) or below knee amputation (34,35). Few reports exist on the use of autologous vascularised fibula grafts, either free or pedicled, for forefoot reconstruction. Borthakur et al. described vascularised free fibula transfer post resection of a first ray osteosarcoma (37). Toriyama et al. reported concomitant first and second metatarsal reconstruction following resection for chondrosarcoma using a free vascularised double-barrelled fibula graft (38), and Toma et al. reported a series of six cases of metatarsal primary bone tumours resected and reconstructed with free vascularised fibula graft, including the first and lesser rays(39). To our knowledge, our case is the first description of a pedicled vascularised fibula graft used for the reconstruction of the first metatarsal post resection for malignancy.

**Conclusion**

This phenotypical and immunophenotypical change from the ‘classic’ variant to the ‘adamantinoma-like’ variant (with squamous differentiation and P63/P40 expression) is very interesting, and the mechanism of this change is not fully understood in regards to its relationship to neo-adjuvant chemotherapy. There is also some literature that suggests that the latter variant may carry a poorer prognosis.

Such a lesion may cause diagnostic dilemmas with a number of neoplasms that also show small cell morphology with squamous differentiation such as basaloid variant of squamous cell carcinoma or NUT-midline carcinoma. This scenario might arise especially if confronted by a small biopsy from a metastatic lesion of such a tumour either in an older patient, in the head and neck region or if there was a history of a Ewing sarcoma for which they received chemotherapy.

Thus an adequate awareness of this variant, the potential chemotherapy related changes, immunohistochemical staining patterns and use of appropriate molecular testing to establish a correct diagnosis is advised.

**Abbreviations**

EWSR1, EWS RNA binding protein 1; FLI1, friend leukaemia integration 1 transcription factor; EFT, Ewing family tumors; ETS, erythroblast transformation specific; MRI, magnetic resonance imaging; FDG,
Declarations

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The dataset supporting the conclusions of this article is included within the article.

Authors’ contributions:
YAM wrote the manuscript. AS, NF, KR, AZ and NR were involved in the treatment of the patient. SDZ, NF and PS revised the manuscript. All authors read and approved the final manuscript.

Declaration of conflicting interests:
The Authors declare that there is no conflict of interest.

Consent for publications:
Written informed consent was obtained from the parent of the patient for publication of this case report and any accompanying images. A copy of the written consent is available for review by the Editor-in-Chief of this journal.

Ethics approval and consent to participate:
Ethical approval was obtained from the Stellenbosch University Health Research Ethics Committee – C20/09/030.

References


