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Editorial

Small round cell tumours of bone

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Most diagnostic histopathlogists must be familiar with the controversy associated with small round cell tumours of bone. Although past confusion in the classification and diagnosis of the malignant round-cell tumours in bone has been largely resolved by histochemical, ultrastructural, immunohistochemical, cytogenetic and tissue culture studies, the differential diagnosis of this type of tumour is still one of the most difficult problems in bone tumour pathology. The non-osteogenic so-called "round-cell" sarcoma of bone was first described by Lücke in 1866. Almost no histogenetic attention was paid to this tumour until 1921 when Ewing applied the term "diffuse endothelioma of bone." The "endothelioma" designation persisted as late as 1933 when Melnick called the tumour Ewing's sarcoma. The nature and histogenesis were regarded as uncertain hence by many the eponymous title Ewing's sarcoma has been preferred until today.

Confusion between primary round-cell sarcoma of bone and metastatic neuroblastoma stemmed from the well-known papers of Colville and Willis (1933), and Willis (1940). They strongly questioned the existence of such a tumour entity referring to the "Ewing's syndrome". Willis's adverse criticism was based upon the results of autopsies in which lesions thought to be osseous primaries were shown to be metastatic. Large series of Ewing's sarcoma, however, have been reported during the last 50 years mostly confirming the existence of so-called "round-cell" sarcoma primary in bone.

Another interesting aspect is the problematic relationship between Ewing's sarcoma and so-called reticulosarcoma of bone. Clinical and pathological differences between the two tumours were emphasized by Parker and Jackson in 1939, drawing attention to the better prognosis for "reticulosarcoma" of bone. This was confirmed by later papers (Coley et al. 1950; Francis et al. 1954; Ivins and Dahlin 1963), while an even worse response to treatment was reported for osseous metastatic neuroblastoma (Bhansali and Desai 1963). These differences of prognosis and the clinical need to distinguish between primary and metastatic bone tumours, together with the possible future development of divergent forms of treatment, made it necessary to explore accurate differential diagnosis between "reticulosarcoma", Ewing's sarcoma and neuroblastoma.

The diagnosis of Ewing's sarcoma was greatly facilitated by a paper written by Schajowicz in 1959. He showed that the histochemical demonstration of glycogen granules in the cytoplasm of tumour cells of Ewing's sarcoma, and their absence in reticulosarcoma, proved to be an easy and efficient method of differential diagnosis, provided that the specimens were properly fixed in alcohol before staining with periodic acid-Schiff (PAS). Conflicting results were obtained for the presence of glycogen in neuroblastoma. Price (1973) reported that 5 of 27 neuroblastomas were glycogen positive, while negative results were reported by Schajowicz (1973) for 38 cases of neuroblastoma as well as 78 cases of reticulosarcoma. Price's results, however, were confirmed by Triche and Ross (1978) demonstrating glycogen by the electron microscope in a case of metastatic osseous neuroblastoma primary in the adrenal.

Demonstration of reticulin by silver impregnation methods was considered useful for differentiation between "reticulosarcoma" of bone and Ewing's sarcoma. The reticulin network is usually thought to be diminished or absent in Ewing's sarcoma just as in lymphosarcoma, whereas recticulin fibres often surround the tumour cells in reticulosarcoma (Pice 1973; Shoji 1971; Rappaport 1966). This concept may now be ignored, due to scientific developments in the field of immunology in recent years. Most of the cases formerly diagnosed as reticulosarcoma are the tumour of neither reticulum cells nor histiocytes, but of T or B lymphocytes (Rapapport and Bradylan 1975). Thus, in agreement with Dahlin (1973) the term "malignant lymphoma" is preferred to the older name "reticulosarcoma".

Tumour cell nuclei are large and irregular in shape with prominent nucleoli and peripherally clumped chromatin in some cases of Ewing's sarcoma (Llombart-

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Bosch et al. 1978). The differential diagnosis of these atypical variants of Ewing's sarcoma from malignant lymphoma or osteosarcoma with extremely small cells seems to be more difficult than in conventional forms. A distinct group of osteosarcoma with histological features of both Ewing's sarcoma and osteosarcoma has been reported by Sim et al. (1979). It is especially important to identify small-cell osteosarcoma for which ablation by surgery is necessary rather than radiation as used for Ewing's sarcoma and malignant lymphoma.

The endothelial origin of Ewing's sarcoma received more support in 1980 due to the report by Roessner et al. demonstrating the presence of type IV collagen, factor VIII related protein and basement membrane-like material in the tumours. Several recent studies, however, have argued against an endothelial origin for this tumour, on the basis of collagen production and lack of specific endothelial markers (Miettinen et al. 1982; Stern et al. 1980). It has been shown by tissue culture studies that Ewing's sarcoma cells synthesize several collagen types including I, III, IV, V and VIII (Dickman et al. 1982; Harvey et al. 1982; Sage et al. 1984). Although some of these collagens are synthesized by endothelial cells, they are also produced by other cells as well as several mesenchymal and epithelial tumours. These data support the conclusion that the origin of the tumour is from multipotential, primitive mesenchymal cells (Dickman et al. 1982).

Rosette or pseudorosette formation in Ewing's sarcoma has been reported by some authors for 50 years (Foote and Anderson 1941; Gharpure 1941). Although differences in the rosettes between neuroblastoma and Ewing's sarcoma have been identified by Jaffe (1964), it is usually difficult to differentiate them histologically. Recent ultrastructural and tissue culture studies suggested that some small round cell tumours of bone in children, which presented as Ewing's sarcoma and had rosettes when sampled extensively were of neuroectodermal origin. Jaffe et al. (1984) suggested these lesions should be called primitive neuroectodermal tumours of bone. They are less prone to have light-microscopic evidence of neuropil or neurite production and neurosecretory granules are generally absent, as is catecholamine production. Neuroblastoma generally have evidence of neurite formation and contain neurosecretory granules or have other evidence of catecholamine production. The primitive neuroectodermal tumours described by Jaffe and his colleagues fall into the category of peripheral neuroectodermal tumour, previously called malignant neuroepithelioma or peripheral neuroblastoma of soft tissues. Another related group is those described by Askin et al. (1979) as malignant small cell tumours of the thoracopulmonary region, some of which have recently been shown to have neural features.

Recent cytogenetic studies of Ewing's sarcoma disclosed a reciprocal translocation t(11; 22) (q24; q12) in 90% of the cases (Turc-Carel et al. 1984; Aurias et al. 1984; Whang-Peng et al. 1986; Sandberg et al. 1988). It is very interesting that an identical translocation was described in peripheral neuroepithelioma (Whang-Peng et al. 1984) as well as in Askin's tumour (Whang-Peng et al. 1986; DeChadarevian et al. 1984). A recent tissue culture study of Ewing's sarcoma cell lines demonstrated no morphological evidence of neural differentiation under normal culture conditions, but after treatment with either cAMP or 12-myristate 13-acetate (TPA), the cells had morphological evidence of neural differentiation such as filaments, microtubules and uraniffin-positive dense core granules by electron microscopy, and neuron-specific enolase, cholinesterase and neural filament triplet protein by immunofluorescence (Cavazzana et al. 1987). Neural markers including rosette formation, dense-core granules, well-developed microtubules, abundant 10-nm filaments and immunohistochemically positive neuron-specific enolase were also reported for a cultured cell line from a "neuroectodermal tumour of bone" (Isayama et al. 1990). As a matter of fact, the clinical, morphological and histochemical distinction between peripheral neuroepithelioma, Askin's tumour and Ewing's sarcoma is extremely sutle, raising the hypothesis that Ewing's sarcoma could be a less differentiated form of tumour of neuroectodermal origin. It is, however, still controversial whether Ewing's sarcoma is a separate entity from neuroectodermal tumour and it is uncertain whether the previously diagnosed Ewing's sarcoma with rosettes should be separated from true Ewing's sarcoma or not (Ushigome et al. 1989).

The first electron microscopic study of Ewing's sarcoma performed by Friedman and Gold (1968) demonstrating glycogen granules and principal and dark cells was not helpful in elucidating its histogenesis. Intercellular junctions have been described in many cases of Ewing's sarcoma since its first report by Takayama et al. (1970). Their morphological description varied from focal membrane thickening to well-developed desmosomes (Dickman et al. 1982; Navas-Palacios et al. 1984; Povysil and Matejovsky 1977; Miettinen et al. 1982; Mahoney and Alexander 1978; Llombardt-Bosch et al. 1978; Machinami 1989). The presence of these desmosome-like junctions in Ewing's sarcoma may explain the positive immunostaining for desmoplakin as reported by Gould et al. (1987). Intracytoplasmic intermediate-type filaments were described as being dispersed or isolated in the cytoplasm and rarely forming parallel bundles (Navas-Palacios et al. 1984; Llombart-Bosch et al. 1978). Some of the rescent ultrastructural studies demonstrated the presence of intracytoplasmic filaments with the appearance of tonofilaments. This epithelial differentiation was confirmed by immunocytochemical studies showing positive keratin staining (Navas-Polacios et al. 1984; Greco et al. 1988). Until recently, immunohistochemical studies in Ewing's sarcoma using monospecific antibodies against intermediate filament proteins of keratin type has been negative (Miettinen et al. 1982). It was only in 1986 and 1987 that keratin protein expression was described as positive in small groups of cells (Katzin et al. 1986; Gould et al. 1987). Moll et al. (1987) reported an immunocytochemical analysis of 11 cases of Ewing's sarcoma for the expression of intermediate filaments and cell junction proteins. All tumours expressed vimentin and 9 cases showed variable expression of cytokeratin in addition to reactivity to desmoplakins. The

presence of epithelial differentiation is extremely interesting. The only primary bone tumours known to have epithelial elements are chordomas, adamantinomas of long bones (Peres-Atayde et al. 1985) and rare cases of primary multipotential bone neoplasms (Hutter et al. 1966; Ling and Steiner 1986). Recently, unusual types of adamantinomas have been published showing, in addition to the epithelial components, features of Ewing's sarcoma (van Haelst and de Haas van Dorsser 1975; Lipper and Kahn 1983).

Is it possible to divide Ewing's sarcomas into two groups, namely those with neural differentiation and the others with epithelial differentiation? We should wait until further studies which demonstrate how often epithelial and neural differentiation is seen in bone tumors otherwise considered to be conventional Ewing's sarcoma.

References

Askin FB, Rosai J, Sibley RK, Dehner LP, McAlister WH (1979) Malignant small cell tumor of the thoracopulmonary region in childhood. A distinctive clinicopathologic entity of uncertain histogenesis. Cancer 43:2438–2451

Aurias A, Rimbaut C, Buffe D, Zucker J-M, Mazabraud A (1984) Translocation involving chromosome 22 in Ewing's sarcoma: A cytogenetic study of four fresh tumors. Cancer Genet Cytogenet 12:21–25

Bhansali SK, Desai PB (1963) Ewing's sarcoma. J Bone Joint Surg 45-A:541–553

Cavazzana AO, Miser JS, Jefferson J, Triche TJ (1987) Experimental evidence for a neural origin of Ewing's sarcoma of bone. Am J Pathol 127:507–518

Coley BL, Higinbotham NL, Groesbeck HP (1950) Primary reticulum cell sarcoma of bone – summary of 37 cases. Radiology 55:641–658

Colville HC, Willis RA (1933) Neuroblastoma metastasis in bones, with criticism of Ewing's endotheliomas. Am J Pathol 9:421–430

Dahlin DC (1973) Primary lymphoma (reticulum cell sarcoma) of bone. Proc Symp Colsten Res Soc 24:207–215

DeChadarevian J-P, Vekemans M, Seemayer T (1984) Reciprocal translocation in small-cell sarcoma (Letter). N Engl J Med 311:1702

Dickman PS, Liotta LA, Triche TJ (1982) Ewing's sarcoma. Characterization in established cultures and evidence of its histogenesis. Lab Invest 47:375–382

Ewing J (1921) Diffuse endothelioma of bone. Proc NY Pathol Soc 21:17–24

Foote FW Jr, Anderson HR (1941) Histogenesis of Ewing's tumor. Am J Pathol 17:497–501

Francis KC, Higinbotham NL, Coley BL (1954) Primary reticulum-cell sarcoma of bone: Report of 44 cases. Surg Gynec Obstet 99:142–146

Friedman B, Gold H (1968) Ultrastructure of Ewing's sarcoma of bone. Cancer 22:307–322

Gharpure VV (1941) Endothelial myeloma (Ewing's tumor of bone). Am J Pathol 17:503–509

Gould VE, Moll R, Berndt R, Roessner A, Franke WW, Lee I (1987) Immunohistochemical analysis of Ewing's tumors. Lab Invest 56:28A

Greco MA, Steiner GC, Fazzini E (1988) Ewing's sarcoma with epithelial differentiation: Fine structural and immunocytochemical study. Ultrastruct Pathol 12:317–325

Harvey W, Squier MV, Duance VC, Pritchard J (1982) A biochemical and immunohistochemical study of collagen synthesis in Ewing's tumor. Br J Cancer 46:848–855

Hutter RVP, Foote FW Jr, Francis KC, Sherman RS (1966) Primitive multipotential primary sarcoma of bone. Cancer 19:1–25

Isayama T, Iwasaki H, Kikuchi M, Yoh S, Takagishi N (1990) Neuroectodermal tumor of bone. Evidence for neural differentiation in a cultured cell line. Cancer 65:1771–1781

Ivins JC, Dahlin DC (1963) Malignant lymphoma (reticulum cell sarcoma) of bone. Proc Mayo Clin 38:375–385

Jaffe HL (1964) Tumors and tumorous conditions of the bones and joints. Lea & Febiger, Philadelphia

Jaffe R, Agostini RM Jr, Santamaria M, Medina J, Yunis EJ, Goodman M, Tanery NH (1984) The neuroectodermal tumor of bone. Am J Surg Pathol 8:885–898

Katzin W, Tubbs R, Bauer TW (1986) Coexpression of intermediate filament proteins in small cell tumors. Lab Invest 54:30A

Ling LL-L, Steiner GC (1986) Primary multipotential malignant neoplasm of bone: Chondrosarcoma associated with squamous cell carcinoma. Hum Pathol 17:317–320

Lipper S, Kahn LB (1983) Case report 235. Skeletal Radiol 10:61–66

Llombardt-Bosch A, Blache R, Peydro-Olaya A (1978) Ultrastructural study of 28 cases of Ewing's sarcoma: Typical and atypical forms. Cancer 41:1362–1373

Lücke A (1866) Beiträge zur Geschwulstlehre. Virchows Archiv Pathol Anat 35:524–539

Machinami R (1989) Ultrastructure of small round cell tumors of soft tissue. J Clin Electron Microscopy 22:5–6

Mahoney JP, Alexander RW (1978) Ewing's sarcoma. A light- and electron-microscopic study of 21 cases. Am J Surg Pathol 2:283–298

Melnick PJ (1933) Histogenesis of Ewing's sarcoma of bone; with postmortem report of a case. Am J Cancer 19:353–363

Miettinen M, Lehto VP, Virtanen I (1982) Histogenesis of Ewing's sarcoma. An evaluation of intermediate filaments and endothelial cell markers. Virchows Archiv B Cell Pathol 41:277–284

Moll R, Lee I, Gould VE, Berndt R, Roessner A, Franke WW (1987) Immunocytochemical analysis of Ewing's tumors. Patterns of expression of intermediate filaments and desmosomal proteins indicate cell type heterogeneity and pluripotential differentiation. Am J Pathol 127:288–304

Navas-Palacios JJ, Aparicio-Duque R, Valdes MD (1984) On the histogenesis of Ewing's sarcoma. An ultrastructural, immunohistochemical and cytochemical study. Cancer 53:1882–1901

Parker F Jr, Jackson H Jr (1939) Primary reticulum cell sarcoma of bone. Surg Gynec Obstet 68:45–53

Peres-Atayde AR, Kozakewich HPW, Vawter GF (1985) Adamantinoma of the tibia. An ultrastructural and immunohistochemical study. Cancer 55:1015–1023

Povysil C, Matejovsky Z (1977) Ultrastructure of Ewing's tumor. Virchows Archiv A Pathol Anat 374:303–316

Price CHG (1973) A critique of Ewing's tumour of bone. Proc Symp Colsten Res Soc 24:177–187

Rappaport H (1966) Tumors of the haematopoietic system. In: Atlas of tumor pathology, section III, Fascicle 8. Armed Forces Institute of Pathology, Washington, DC

Rapapport H, Braylan RC (1975) Changing concepts in the classification of malignant neoplasms of the hematopoietic system. In: Rebuck JW, Berard CW, Abell MR (eds) International Academy of Pathology Monograph: The reticuloendothelial system, pp 1–19. The Williams & Wilkins, Baltimore

Roessner A, Voss B, Rauterberg J, Immenkamp M (1980) Ewing's sarcoma. A comparative electron and immunofluorescence microscopical study. Abstracts, XIIIth International Congress, International Academy of Pathology, Paris

Sage H, Balian G, Vogel AM, Bornstein P (1984) Type VIII collagen. Synthesis by normal and malignant cells in culture. Lab Invest 50:219–231

Sandberg AA, Ture-Carel C, Gemmill RM (1988) Chromosomes in solid tumors of and beyond. Cancer Res 48:1049–1059

Schajowicz F (1959) Ewing's sarcoma and reticulum cell sarcoma of bone. With special reference to the histochemical demonstra-

tion of glycogen as an aid to differential diagnosis. J Bone Joint Surg 41-A:349–356
Schajowicz F (1973) Differential diagnosis of Ewing's sarcoma. Proc Symp Colsten Res Soc 24:189–202
Shoji H, Miller TR (1971) Primary reticulum cell sarcoma of bone. Significance of clinical features upon the prognosis. Cancer 28:1234–1243
Sim FH, Unni KK, Beabout JW, Dahlin DC (1979) Osteosarcoma with small cells simulating Ewing's tumor. J Bone Joint Surg 61-A:207–215
Stern R, Wilczek J, Thorpe WP, Rosenberg SA, Cannon G (1980) Procollagens as markers for the cell of origin of human bone tumors. Cancer Res 40:325–328
Takayama S, Sugawa I (1970) Electron microscopic observations of Ewing's sarcoma. A case report. Acta Pathol Jpn 20:81–101
Triche TJ, Ross WE (1978) Glycogen-containing neuroblastoma with clinical and histological features of Ewing's sarcoma. Cancer 41:1425–1432
Turc-Carel C, Philip I, Berger M-P, Philip T, Lenior GM (1984) Chromosome study of Ewing's sarcoma (ES) cell lines. Consistency of a reciprocal translocation t(11; 22) (q24; q12). Cancer Genet Cytogenet 12:1–19
Ushigome S, Shimoda T, Takaki K, Nikaido T, Takakuwa T, Ishikawa E, Spjut HJ (1989) Immunohistochemical and ultrastructural studies of the histogenesis of Ewing's sarcoma and putatively related tumors. Cancer 64:52–62
Van Haelst UJGM, de Haas van Dorsser AH (1975) A perplexing malignant bone tumor. Highly malignant so-called adamantinoma or non-typical Ewing's sarcoma. Virchows Archiv A Pathol Anat 365:63–74
Whang-Peng J, Triche TJ, Knuten T, Miser J, Douglass EC, Israel MA (1984) Chromosome translocation in peripheral neuroepithelioma. N Engl J Med 311:584–585
Whang-Peng J, Triche TJ, Knutsen T, Miser J, Kao-Shan S, Tsai S, Israel MA (1986) Cytogenetic characterization of selected small round cell tumor of childhood. Cancer Genet Cytogenet 21:185–208
Willis RA (1940) Metastatic neuroblastoma in bone presenting the Ewing's syndrome, with a discussion of "Ewing's sarcoma". Am J Pathol 16:317–333