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Malignant fibrous histiocytoma: similarities to the "fibrohistiocytoid cells" in chronic inflammation

Yutaka Imai[1], Mitsunori Yamakawa[1], Toshihiro Sato[1], and Akio Suda[2]

[1] Second Department of Pathology,
[2] Department of Orthopedic Surgery, Yamagata University School of Medicine, Zao-Iida, Nishinomae, Yamagata 990-23, Japan

Summary. Ultrastructural, enzyme histochemical and immunohistochemical studies were performed on tissue obtained from eight cases of malignant fibrous histiocytoma (MFH) and five cases of sacral decubitus ulcer. The MFH was composed of two major tumour cell types: fibroblast-like and histiocyte-like cells. Both cell types demonstrated abundant branching, fragmented rough endoplasmic reticulum (rER), many free ribosomes, occasional small mitochondria, an oval, elliptical or irregularly shaped nucleus with one or two prominent nucleoli and often a few dense bodies. However, pseudopodial projections, multivesicular bodies and phagosomes, common histiocyte organelles, were not seen. With little difference between cases or selection sites, the MFH cells reacted to acid phosphatase (AcP) and α-naphtyl butyrate esterase (ANBE) by enzyme histochemistry and with ferritin (Fer), α1-antitrypsin (AT), α1-antichymotrypsin (ACT), fibronectin (FN), HLA-DR, HLA-DP, Leu 10 and OKT 9 in immunohistochemical studies. MFH tumour cells did not immunostain with monocyte/macrophage markers (Leu M1, Leu M3, Mo 1, Mo 2 and Macrophage) although non-neoplastic histiocytes did react to these markers. In addition, granulation tissue, such as that found in sacral decubitus ulcers, was examined and the existence of a specific cell type called the "fibrohistiocytoid (FH) cell" was documented. The FH cell was short, spindle shaped and elliptical. Ultrastructurally, it had fragmented rER distributed in a branching pattern, dispersed free ribosomes, small mitochondria and a few dense bodies, but lacked diverse fused lysosomes and distinct pseudopodial cytoplasmic extensions. The FH cells reacted with AcP, alkaline phosphatase and ANBE but not with peroxidase using enzyme histochemistry and with Fer, AT, ACT, FN, HLA-DR, HLA-DP, Leu 10 and OKT 9 but not with monocyte/macrophage markers, C3d receptor, C3bi receptor in immunohistochemical studies. The FH cells had morphological, enzyme histochemical and immunohistochemical characteristics intermediate between fibroblasts and histiocytes. Similarities between MFH cells and the FH cells seen in chronic inflammation are discussed.

Key words: Malignant fibrous histiocytoma – Ultrastructure – Enzyme histochemistry – Immunohistochemistry – "Fibrohistiocytoid cell"

Offprint requests to: Y. Imai

Introduction

The malignant fibrous histiocytoma (MFH) is composed of cells which, on morphological examination, resemble fibroblasts and histiocytes (Rosai 1981; Weiss 1982). The actual differentiation of constituent cells has been controversial since Stout and co-workers first reported the MFH to be a histiocytic neoplasm exhibiting facultative fibroblast-type differentiation (Ozello et al. 1963; O'Brien and Stout 1964). Later, the predominant cells were noted to exhibit a fusiform or spindle shape and to resemble fibroblasts. Subsequently, rival theories contended that MFHs were derived from either fibroblasts or primitive mesenchymal cells (Churg and Kahn 1977; Alguacil-Garcia et al. 1978; Hoffman and Dickersin 1983; Roholl et al. 1985a; Wood et al. 1986; Brecher and Franklin 1986).

The debate has centered around the interpretation of morphological and tissue culture data (Rosai 1981; Enzinger and Weiss 1988), and, more recently, the expression of "histiocytic markers"

by MFH cells (Iwasaki et al. 1982; Boulay 1982; Kindblom et al. 1982). However, several factors complicate the interpretation of these data.

We have previously reported morphological changes in fibroblasts in chronic lymphadenitis (Imai et al. 1983), and the existence of a specific cell type, the "fibrohistiocytoid (FH) cell". This was found in a variety of chronic inflammatory tissues including chronic peritonitis, chronic osteomyelitis, gastric ulcer and the stroma of oesophageal cancers. These FH cells appear to be closely related to cells which resemble histiocytes, in vivo in the rabbit condition (Kojima and Imai 1965), in granulation tissues (Imai et al. 1987) and in the stroma of oesophageal cancer (Sato et al. 1988) and in in vitro culture of human dermal fibroblasts (Takagi et al. 1988). We investigated tissue samples from eight cases of MFH and five cases of sacral decubitus ulcer using ultrastructural analysis, enzyme histochemistry and immunohistochemistry to evaluate the apparent cytological similarity between MFH tumour cells and FH cells.

Materials and methods

Tissues from eight MFHs (Table 1) and five sacral decubitus ulcers were used for light and electron microscopic, enzyme histochemical and immunohistochemical studies. The case selection was made from cases registered in our laboratory from 1979 until 1988.

Tissue samples for light microscopy were sliced into approximately 5 mm slices which were fixed in 10% formalin, embedded in paraffin and cut into 4 μm thick sections. The sections were stained using H&E and Mallory's trichrome stains. Part of the paraffin-embedded specimen was cut into 2 μm thick serial sections for detailed evaluation of individual cell morphology.

Tissue specimens for electron microscopy were processed in the usual manner: fixation in ice-cold 1.25% glutaraldehyde buffered with pH 7.4 cacodylate buffer for 2 h, post-fixation in osmium tetroxide for 1 h, followed by dehydration and embedding in Epon 812. Sections 1 μm thick were stained with toluidine blue, and ultrathin sections were then cut from selected areas, stained with uranyl acetate and lead citrate, and examined with a Hitachi HS-9 transmission electron microscope.

A portion of the tissue specimen was immersed in periodate-lysine-paraformaldehyde (PLP) fixative for 6 h, rinsed in graded sucrose-phosphate-buffered saline, embedded in Tissue-Tek II OCT compound (Miles Lab. Inc., Kankakee, IL), and stored at −80° C until cryostat sectioning for enzyme histochemical and immunohistochemical studies. The remaining fresh tissues for enzyme histochemistry and immunohistochemistry were trimmed and immediately frozen in OCT compound.

Enzyme histochemical stains were performed using alkaline phosphatase (ALP) (Nanba et al. 1977), acid phosphatase (AcP) (Barka and Anderson 1962), α-naphthyl butyrate esterase (ANBE) (Yam et al. 1971) and peroxidase (Kaplow 1975). The antisera used for immunohistochemical staining are listed in Table 2. Representative cryostat sections of fresh-frozen and PLP-fixed material were stained using an indirect immunoperoxidase technique as described previously (Farr and Nakane 1981; Imai et al. 1986).

Table 1. Malignant fibrous histiocytoma; Clinical data

Case	Localization	Age	Sex	Histological type
1	Left femur bone	53	M	Pleomorphic
2	Left fibra	49	M	Pleomorphic
3	Chest wall	88	F	Pleomorphic
4	Right thigh	58	M	Pleomorphic
5	Adjacent to scapula	48	M	Storiform
6	Chest wall	52	F	Storiform
7	Retroperitoneum	56	M	Storiform
8	Retroperitoneum	65	F	Storiform

Table 2. List of antisera used

Antiserum	Source
Rabbit anti-human	
Ferritin, Lysozyme, α1-antitrypsin, α1-antichymotrypsin, Fibronectin	DAKO patts
Mouse monoclonal anti-human	
Macrophage, CR 1, HLA-DR	DAKO patts
Leu M1, M3, M5, Leu 10, HLA-DP, CR2, CR3	Becton-Dickinson
Mo 1, Mo 2	Coulter Clone
HLA-A,B,C	Selatech
OKT 9	Ortho Diag.
Peroxidase-conjugated swine anti-rabbit Ig	DAKO patts
Peroxidase-conjugated F(ab')2 fragment sheep anti-mouse Ig	Amersham Japan
Peroxidase-conjugated goat anti-rat Ig	EY Lab.

Fig. 1. Malignant fibrous histiocytoma, storiform type. Tumour cells are composed of two major cell types, fibroblast-like cells and histiocyte-like cells. The stroma is occupied by dense collagen fibers. HE stain, ×40

Figs. 2–7. Electron micrographs show a clearly identifiable variety of cell types in malignant fibrous histiocytoma

Fig. 2. A low-power view shows several tumour cells, including a lower undifferentiated cell (*arrow*), an upper differentiated cell (*arrow head*) and a fibroblast/histiocyte-like cell (*double arrows*). ×2000

Fig. 3. A fibroblast-like cell has a large elliptical shaped nucleus with a prominent nucleolus. The nuclear envelope has complicated folds. Note the cytoplasm containing well-developed, fragmented rough endoplasmic reticula and a few dense bodies. ×3000

Fig. 4. A high-power view shows a fibroblast-like cell. Note the peripheral condensations of filaments (*arrows*). ×7000

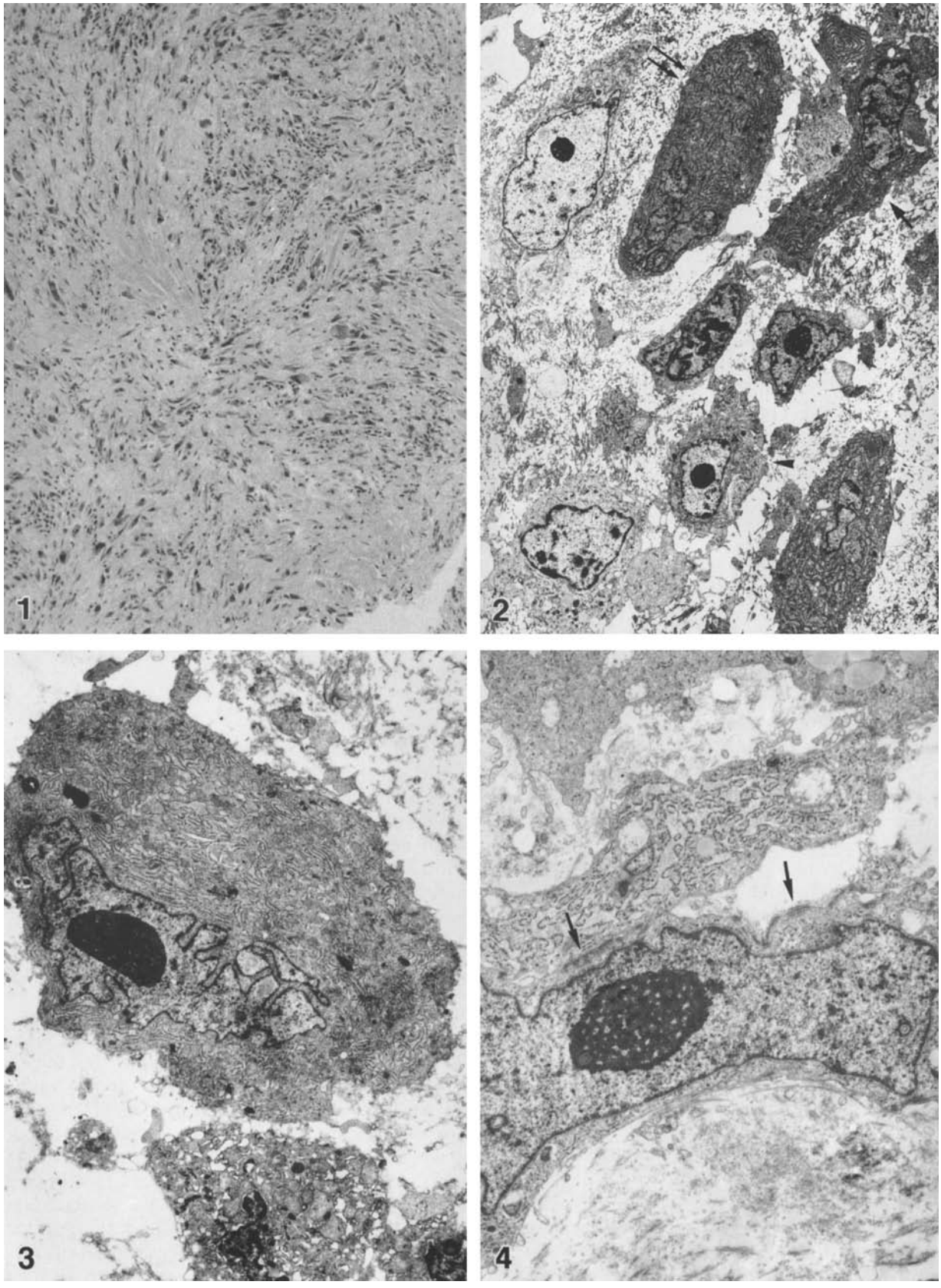
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Table 3. Ultrastructural characteristics of the fibroblast, fibrohistiocytoid cell and histiocyte in granulation tissues, and of the fibroblast-like cell and histiocyte-like cell in malignant fibrous histiocytomas

Cell Organella	Granulation Tissue			Malignant Fibrous Histiocytoma	
	Fibroblast	Fibrohistiocytoid cell	Histiocyte	Fibrohistiocytoid cell	
				Fibroblast-like cell	Histiocyte-like cell
Cell body	large, elongated	short-spindle, elliptical, angular	elliptical, irregular	short-spindle, elliptical	large, rounded
Nucleus	oval, elliptical	oval, elliptical, irregular	reniform	elliptical, elongated, irregular	oval, elliptical, polygonal
Nucleolus	one or two oval, large	one or two oval, large	one or a few small	one or two large, prominent	one or two small ~ large
rER	++	+ ~ ++ branching, fragmented	+	++ fragmented, often dilated	+ branching, fragmented
Mitochondria	+	+, small	+	+	+, small
Free ribosome	++	++, dispersed	+	++	++
Golgi complex	++	++	++	+	++
Primary lysosome	+	+ ~ ++	++	+	++
Secondary lysosome	−	−/+	+ ~ ++	−/+	+
Microfilament	++	++	−/+	++	−/+
Pseudopodium	−	−	++	−	−
Erythrophagocytosis	−	−/+	+ ~ ++	−	−/+

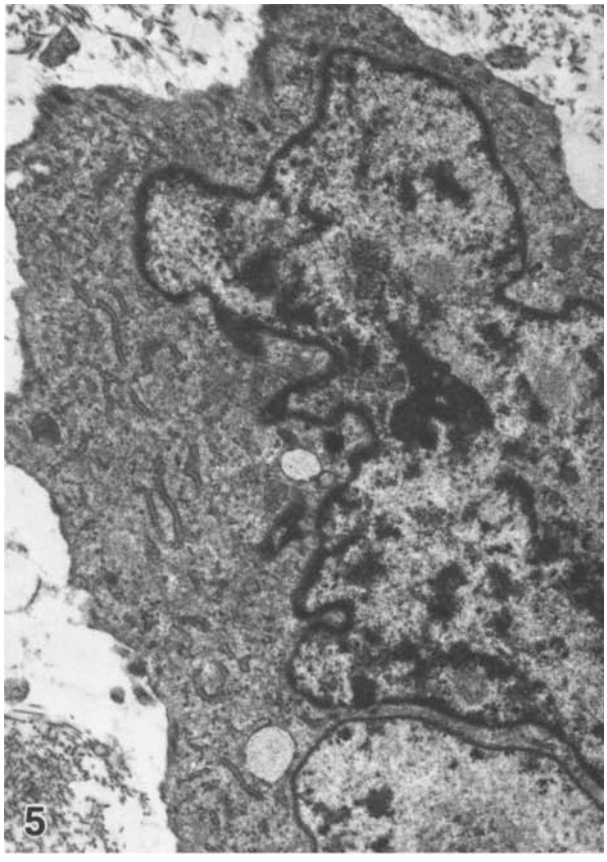
−: none −/+: often appeared +: moderate in degree ++: marked in degree + ~ ++: a variety of positivity

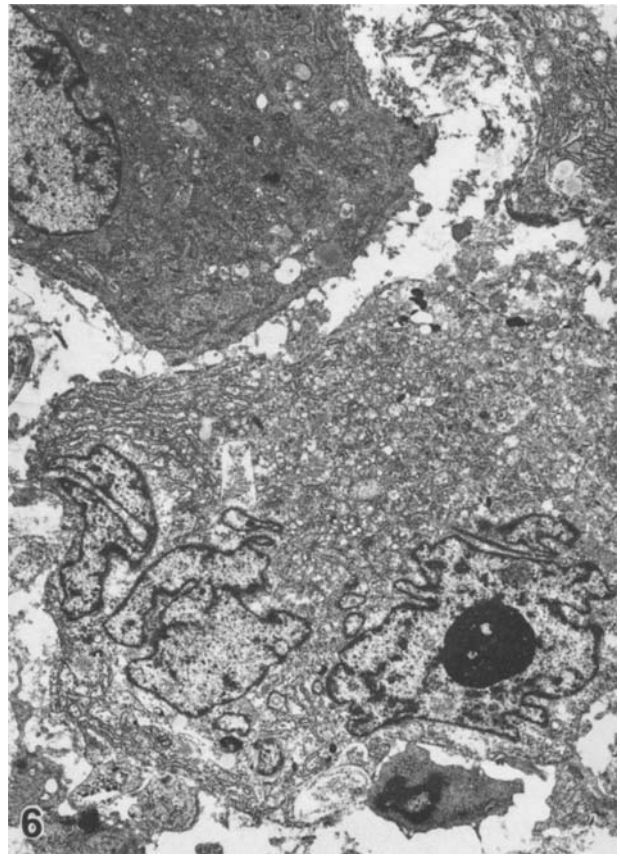
Results

The histological appearances of MFH have been well described in several reports (Fu et al. 1975; Tsuneyoshi et al. 1981; Enzinger and Weiss 1988). Light microscopy of our cases revealed two major cell types: fibroblast-like cells and histiocyte-like cells (Fig. 1). The fibroblast-like cells were spindle shaped and had an elliptical or irregularly shaped nucleus with one or two large nucleoli. One of the pathognomonic features of MFH was a "striform pattern" composed primarily of this cell type. The histiocyte-like cells were larger, with a bizarre polygonal nucleus surrounded by a vacuolated, drab, eosinophilic cytoplasm. Occasionally, Touton-type multinucleated giant cells and foamy xanthomatous cells were observed.

Electron microscopy revealed a clearly identifiable variety of cell types including both fibroblast-like cells and histiocyte-like cells (Table 3, Figs. 2–5), small undifferentiated cells, xanthomatous cells and multinucleated giant cells (Figs. 6, 7). The fibroblast-like cells had a large, elliptical or somewhat irregularly shaped hyperchromatic nucleus with one or two prominent nucleoli. The envelope of the irregularly shaped nuclei had complicated folds or invaginations. The cytoplasm contained well-developed, fragmented and often dilated rough endoplasmic reticula (rER), one or more well-differentiated Golgi complexes, a few mitochondria, numerous free ribosomes, microfilaments and occasional lipid droplets. In addition, nuclear bodies were often found.

Histiocyte-like cells had an oval or elliptical nucleus, one or two small, and often large nucleoli, and a cytoplasm which contained fragmented, branching rER, many free ribosomes, small mitochondria and a few dense bodies which resembled lysosomes. Although this cell type often demonstrated erythrophagocytosis, fusion of each phagosome and secondary lysosome was observed less frequently. In some cases, intracytoplasmic filaments were found focally near the cell membrane, in both fibroblast-like cells and histiocyte-like cells as seen in fibroblasts. The cell bodies were smooth, without prominent pseudopodia. Typical histiocytes were occasionally found, with multivesicular bodies, phagocytotic vacuoles and pseudopodial





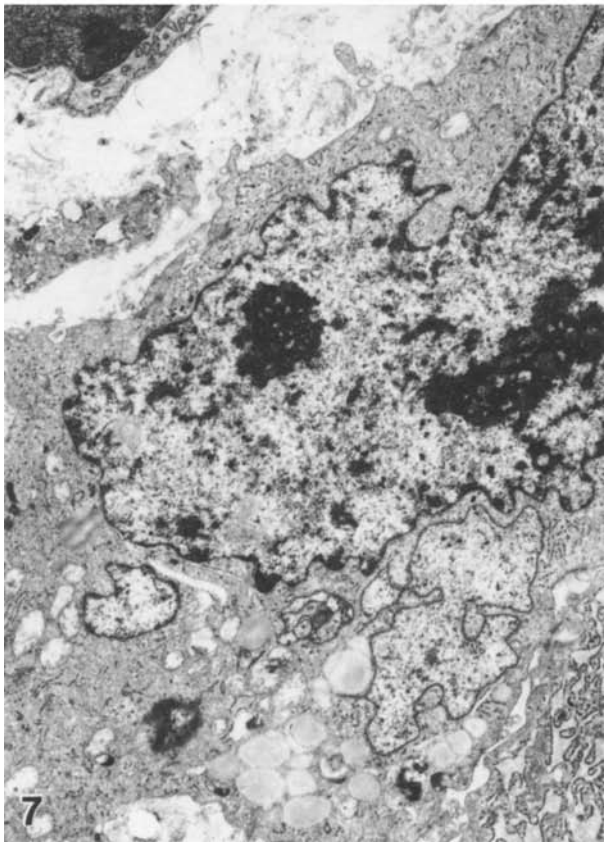


Fig. 5. A fibroblast-like cell has a lobulated nucleus or multinucleation. Note the fragmented, branching rough endoplasmic reticula. × 7000

Fig. 6. Shows two large tumour cells. Their cytoplasms contain abundant, fragmented, branching rough endoplasmic reticula and a few dense bodies. × 4000

Fig. 7. A high-power view of a multinucleated giant cell. Note the fragmented rough endoplasmic reticula, a few dense bodies and lipid droplets. × 7000

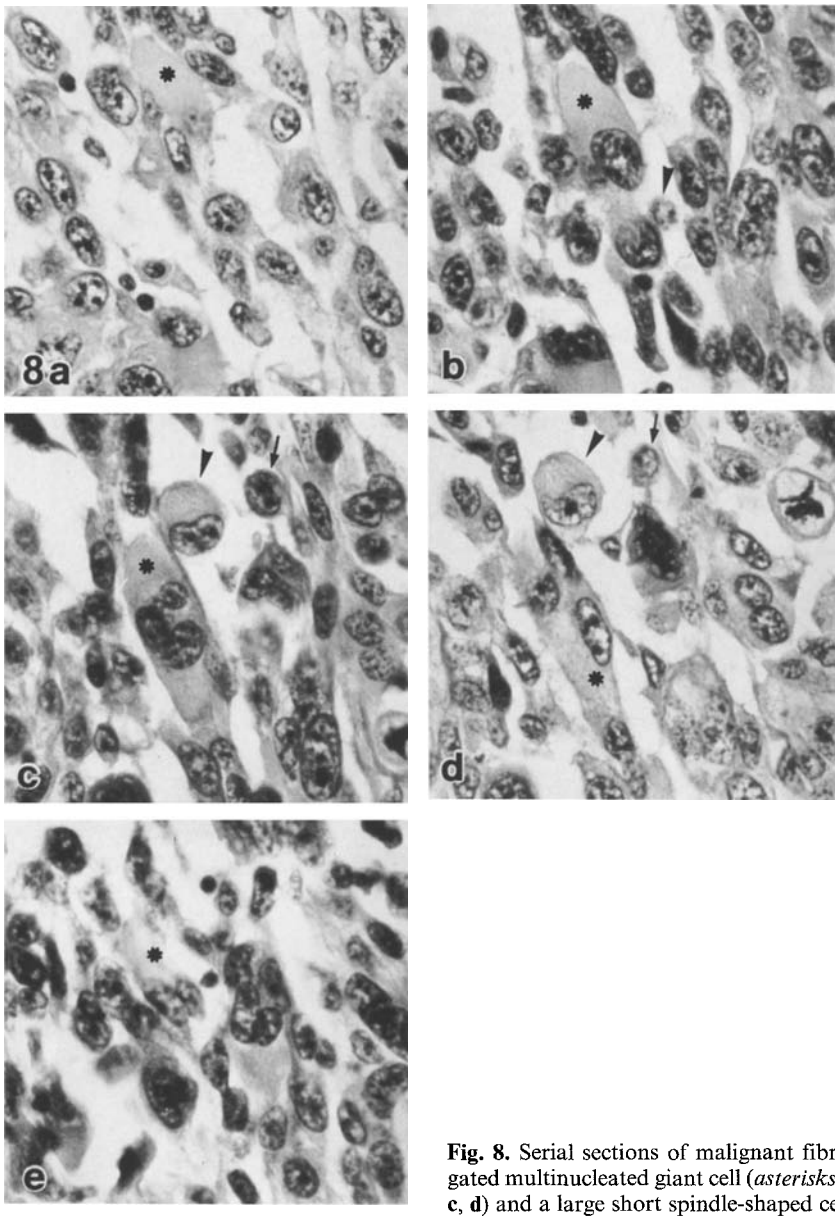


Fig. 8. Serial sections of malignant fibrous histiocytoma. Note not only an elongated multinucleated giant cell (*asterisks*, **a–e**) but also a small elliptical cell (*arrows*, **c**, **d**) and a large short spindle-shaped cell (*arrowheads*, **b–d**). HE stain, ×680

cell processes. However, they did not display well-developed free ribosomes or the large nucleoli which are found in typical neoplastic cells. A few small, round undifferentiated cells were found, exhibiting smooth cell bodies, a round nucleus, a thin rim of cytoplasm containing either free and/or rosettes of ribosomes, few mitochondria, and occasional filamentous material. Only rarely were clusters of rER or distinct Golgi complexes found in these cells.

When an elongated cell is cut transversely, it may appear to be a short-spindle or elliptical cell. To resolve this problem, serial sectioning of paraffin-embedded tissue was performed. Although a part of the short-spindle or elliptical cells in a tissue section were shown to be elongated cells in serial section, other cells with similar initial appeareance were shown to be short-spindle or elliptical cells in serial section (Fig. 8).

Xanthomatous cells contained numerous lipid droplets which appeared either as empty spaces or as electron-dense homogenous material. Laminated myelin figures were seen frequently. The cytoplasmic membrane had coarse pseudopodia in contrast with the delicate filopodia of the histiocyte-like cells. The occasional giant cell contained two or more nuclei in an abundant cytoplasm. As in fibroblasts, the cytoplasms contained numerous mitochondria and rER.

The results of enzyme histochemical and immu-

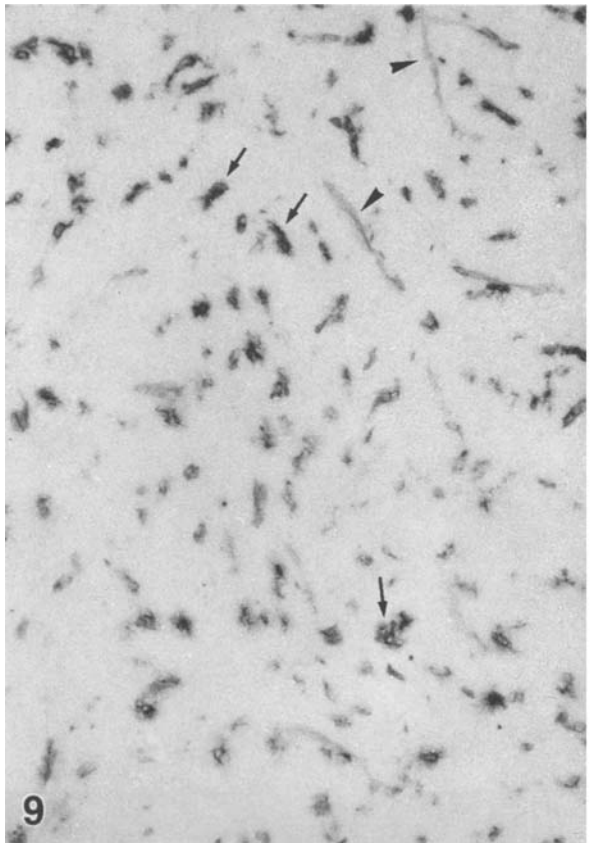
Table 4. Results of enzyme histochemical and immunohistochemical studies of granulation tissues and malignant fibrous histiocytomas

Reagent	Granulation Tissue			Malignant Fibrous Histiocytoma							
	FB	FH cell	MΦ	FH cell						Pleomorphic type	Storiform type
				Fib	His	Und	Xan	Gia	Fibrohist		
AcP	−/+	+	+	+	+	−	+	+	+ Ψ	++	++
AlP	−	+	++	−	−/+	−	−/+	−/+	−/+ V	−/+	−
ANBE	+	++	++	++	++	−	++	+	− V	++	+
PO	−	−	++	−	−	−	−/+	−		−	−
Ferritin	−/+	+	++	+	+	−	++	++		++	++
Lysozyme	−	−/+	++	−/+	−/+	−	−/+	−/+	−/+ Γ	−/+	−/+
AT	−	−/+	+	−/+	+	−	+	+	−/+ Γ	++	+
ACT	−	−/+	+	−/+	+	−	+	+	−/+ Ψ	++	+
Fibronectin	+	+	+	+	+	−/+	++	−/+		++	++
Leu M1	−	−	−/+	−	−	−	−/+	−	− Ψ	−	−
Leu M3	−	−	−/+	−	−	−	−/+	−	− V	−	−
Leu M5	−	−/+	++	−/+	−/+	−	++	−/+	− Π	−/+	−/+
Mo 1	−	−	++	−	−	−	−/+	−		−	−
Mo 2	−	−	++	−	−	−	−/+	−	− Π	−	−
Macrophage	−	−	++	−	−	−	+	−		−	−
CR 1	−	−/+	++	−	−/+	−	+	−/+	− Π	−	−
CR 2	−	−	++	−	−	−	+	−/+		−	−
CR 3	−	−	+	−	−	−	−/+	−	− Π	−	−
HLA-A,B,C	+	+	+	+	+	−/+	+	+	+ V	+	+
HLA-DR	−	++	++	++	++	−	++	−/+	− Π, −/+ Σ	++	++
HLA-DP	−	++	++	++	++	−	++	−/+		++	++
Leu 10	−	++	++	++	++	−	++	−/+		++	++
OKT 9	−/+	++	++	++	++	−	++	−/+		++	++

FB: Fibroblast FH cell: Fibrohistiocytoid cell MΦ: Macrophage Fib: Fibroblast-like cell His: Histiocyte-like cell Und: Undifferentiated cell Xan: Xanthomatous cell Gia: Multinucleated giant cell Fibrohist: Fibrohistiocyte −: negative −/+: often positive +: moderate in degree ++: marked in degree V, Γ, Σ, Ψ and Π: referred to Wood et al. 1986; Roholl et al. 1985a, b, c; and Brecher and Franklin 1986, respectively

nohistochemical studies are summarized in Table 4. MFH cells reacted to a differing degree depending on the histological subtypes. FH cells, both the fibroblast-like and histiocyte-like cells, reacted to AcP and ANBE on enzyme histochemistry and to ferritin (Fer), α1-antitrypsin (AT), α1-antichymotrypsin (ACT), fibronectin (FN), HLA-A,B,C, HLA-DR (Fig. 9), HLA-DP, Leu 10 and OKT 9 (Fig. 10) in immunohistochemical studies. Occasional cells which reacted to lysozyme (Lys), Leu M5 and ALP were found. No marked reactions to monocyte/macrophage markers were found, with the exception of Leu M5. Of special interest was the diverse immunostain with Fer, HLA-DR, HLA-DP, Leu 10 and OKT 9, and the prominence of these reactions in areas which displayed a well developed "striform pattern". The small, undifferentiated cells were only occasionally stained with FN and HLA-A,B,C. The reactions of xanthomatous cells were similar to that of benign-appearing histiocytes that were small in comparison with the pleomorphic tumour cells, and lacked both mitotic figures and a high nuclear/cytoplasm ratio. Giant cells were stained in a pattern similar to histiocyte-like cells. Mouse monoclonal antibody against human macrophages reacted only with non-neoplastic histiocytes.

Two separate types of granulation tissues were identified in the dermal and subcutaneous lesions: fibrous and cellular. In the former, bundle-like collagen fibers were apparent, and the number of fibroblasts was increased. A slight perivascular cellular infiltrate was found, composed of lymphocytes, plasma cells, histiocytes and neutrophilic leukocytes. In areas with marked collagenization, the scanty cellular component was composed of fibroblasts primarily. In addition to the typically elongated fibroblast, FH cells (short spindle-shaped, or elliptical cells) were found. The cellular granulation tissue contained abundant neutrophilic leukocytes as well as mononuclear cells. Occasional fibroblasts had pyknotic nuclei and were degenerate. While the number of long, spindle-shaped cells was decreased, the number of the short, spindle-shaped and elliptical cells was increased, with slightly increased collagen fibers. In



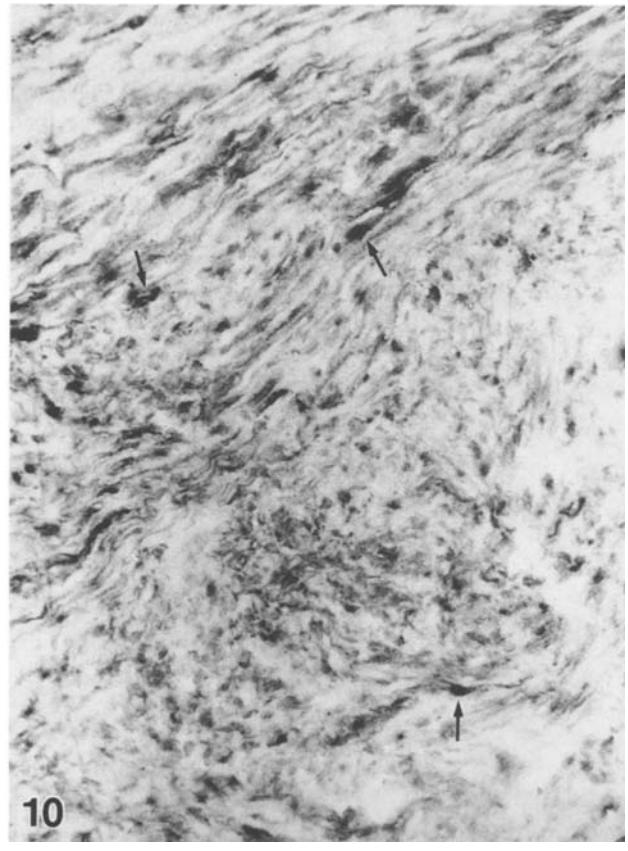


Fig. 9. HLA-DR immunostain in malignant fibrous histiocytoma. The marked reactions are found in many tumour cells (*arrows*), endothelial cells (*arrowheads*). Counterstain with methyl green, ×40

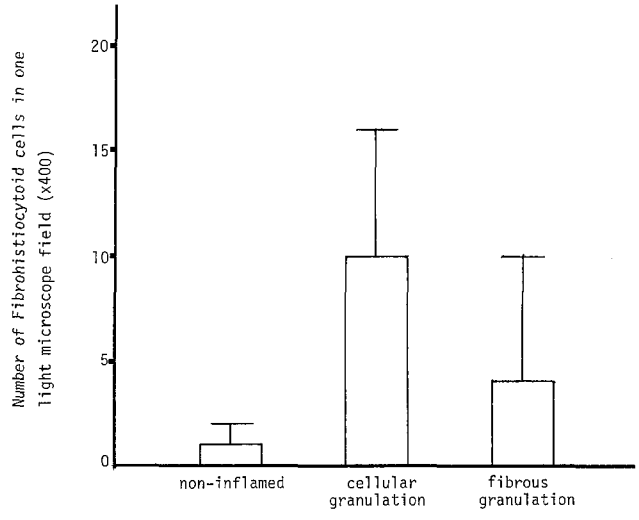
Fig. 10. OKT 9 immunostain in a area showing a storiform pattern in malignant fibrous histiocytoma. A majority of tumour cells react, including large elliptical cells (*arrows*). It does not appear that non-tumour cells react. Counterstain with methyl green. ×40

the granulation tissue as well as the MFH, serial sectioning was performed, demonstrating both short-spindle and the elliptical cells (Fig. 11).

Table 5 reveals the frequency of FH cells in tissue from human sacral decubitus ulcers. The number of FH cells in one ×400 light microscope field was calculated in areas remote from the inflammation, in areas with cellular granulation and in areas with fibrous granulation. The mean number of FH cells per field was evaluated after observation ten fields on each investigated slide. Few FH cells were found in non-inflamed areas, and only a few were found in areas with fibrous granulation. Increasing numbers of FH cells were found in areas with cellular granulation. An average of five fibroblasts and one FH cell per ×400 field were found in areas with fibrous granulation and one fibroblast and one FH cell were found per ×400 field in areas with cellular granulation.

The ultrastructural findings of granulation tis-

Table 5. Frequency of Fibrohistiocytoid cell appearance in tissues from human sacral decubitus ulscers



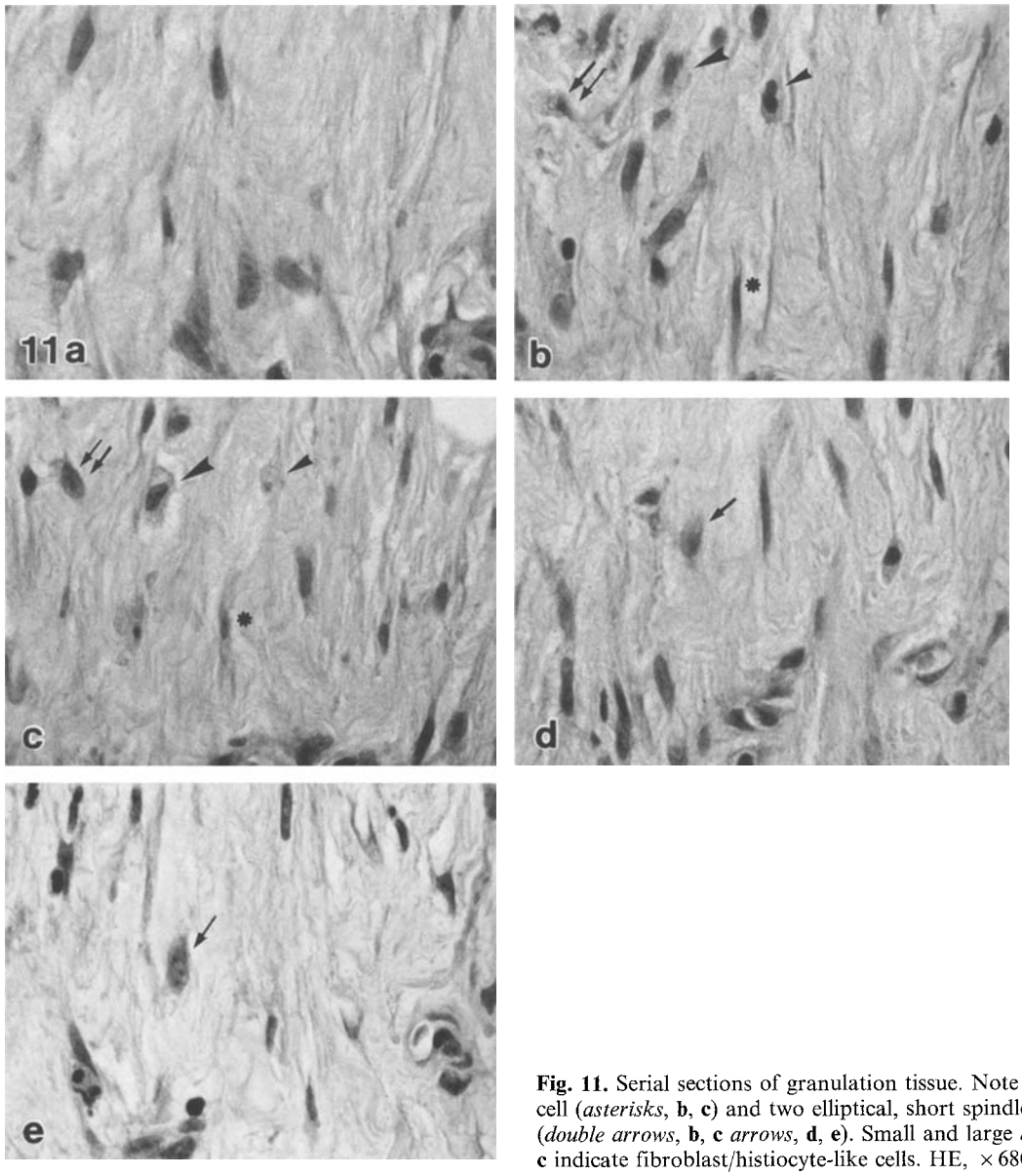
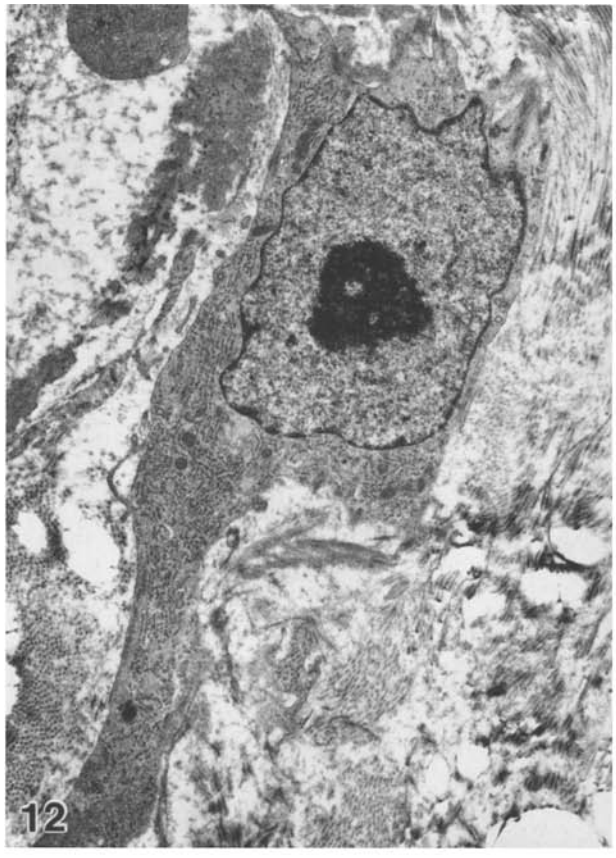


Fig. 11. Serial sections of granulation tissue. Note a small elongated cell (*asterisks*, **b**, **c**) and two elliptical, short spindle-shaped cells (*double arrows*, **b**, **c** *arrows*, **d**, **e**). Small and large *arrowheads* in **b** and **c** indicate fibroblast/histiocyte-like cells. HE, ×680

sue are summarized in Table 3. Electron-microscopic findings of the long, spindle-shaped fibroblasts were similar to findings of typical fibroblasts (Copenhaver 1964) (Fig. 12). In contrast, FH cells (the short, spindle-shaped cells and elliptical cells) generally had cell bodies which were larger than histiocytes, and displayed rather smooth cytoplasmic extensions without diverse pseudopodial processes (Figs. 13–17). The large elliptical or irregularly-shaped nucleus was centrally located in the cell body. The chromatin was clumped in the periphery of the nucleus and there was a prominent, centrally-located large nucleolus. The cytoplasm contained few Golgi complexes, rather small mitochondria, and a large amount of branching rER. The amount of rER, however, was decreased in comparison to that seen in a typical fibroblast. Abundant free ribosomes were distributed throughout the cytoplasm. In general, both the short, spindle-shaped cells and the elliptical fibroblasts had fewer phagosomes compared to histiocytes. Both cells had occasional dense bodies without either distinct fusions or well-developed multivesicular bodies. While histiocytes demonstrated erythrophagocytosis as well as vigorous phagocytosis, neither type of fibroblast exhibited erythrophagocytosis with any frequency. Both types of fibroblasts occasionally had bundles of filamentous material in the periphery of the cytoplasm, and were surrounded by abundant collagen fibers.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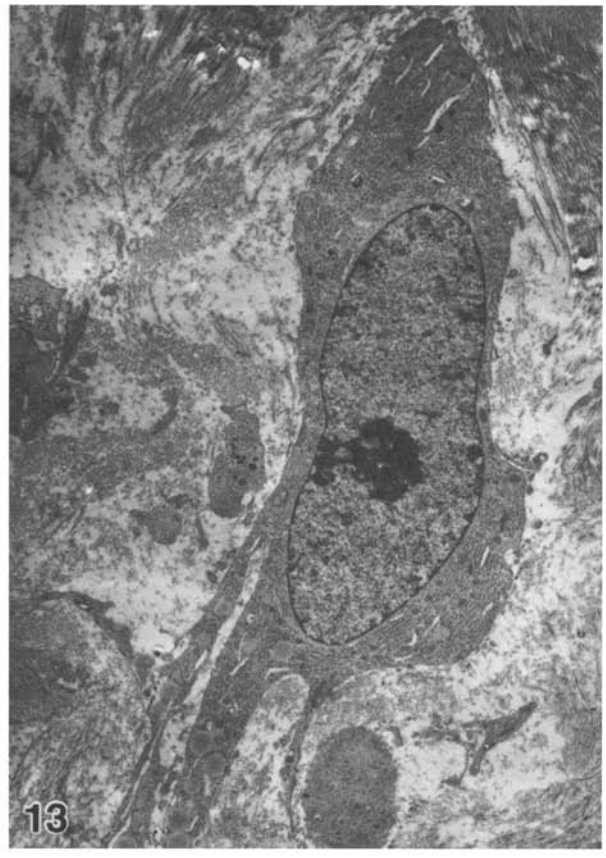
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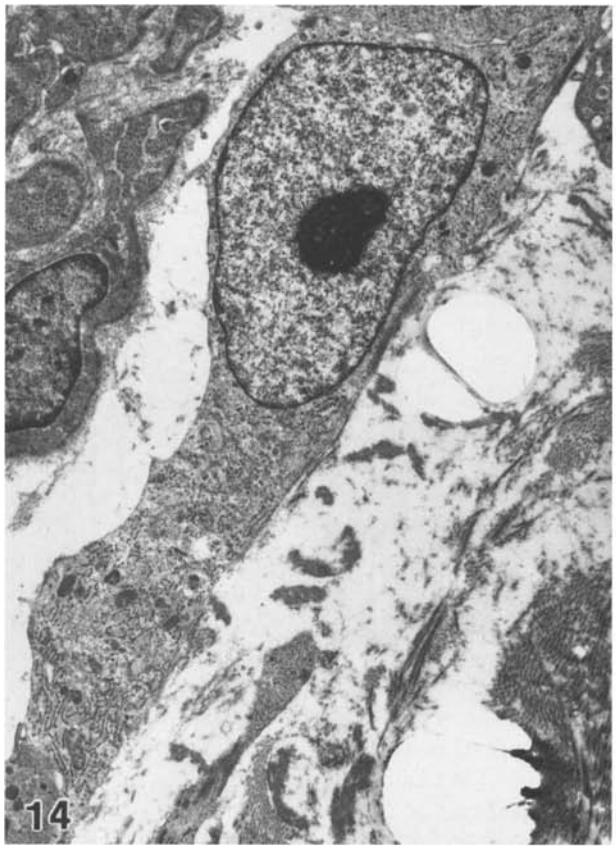
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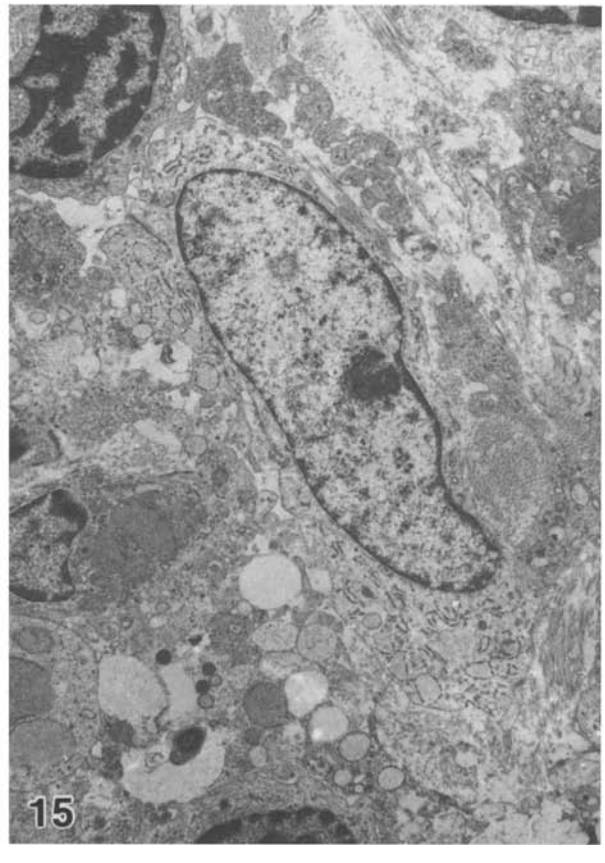
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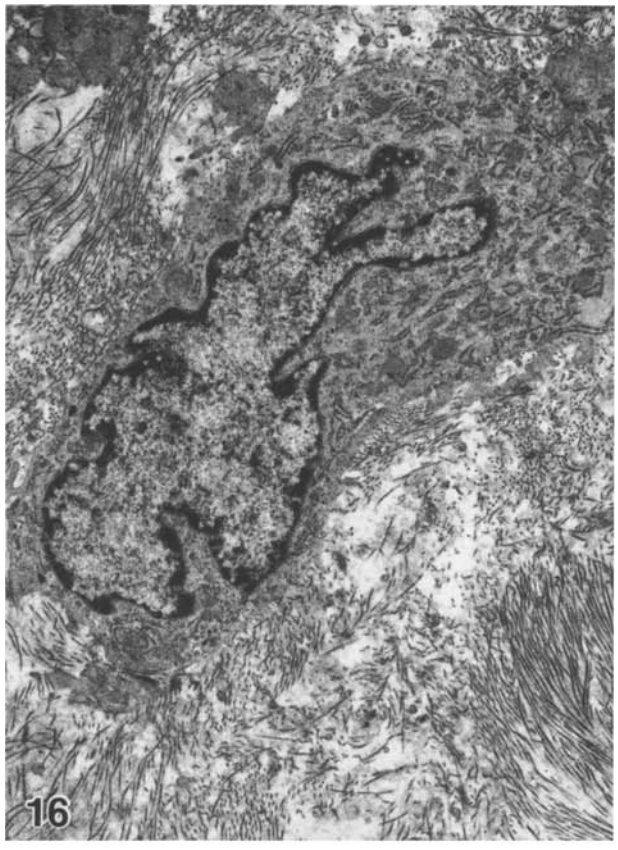

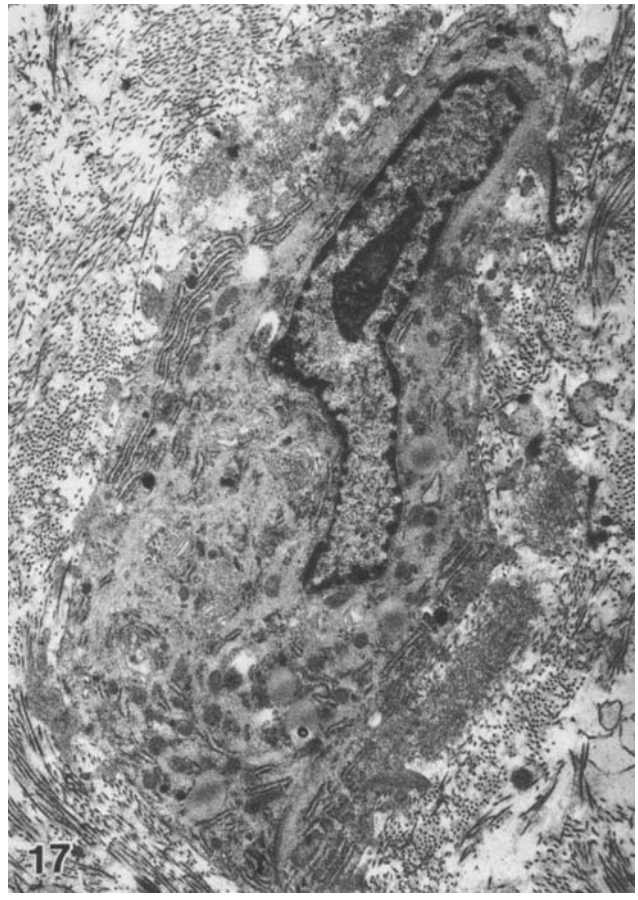

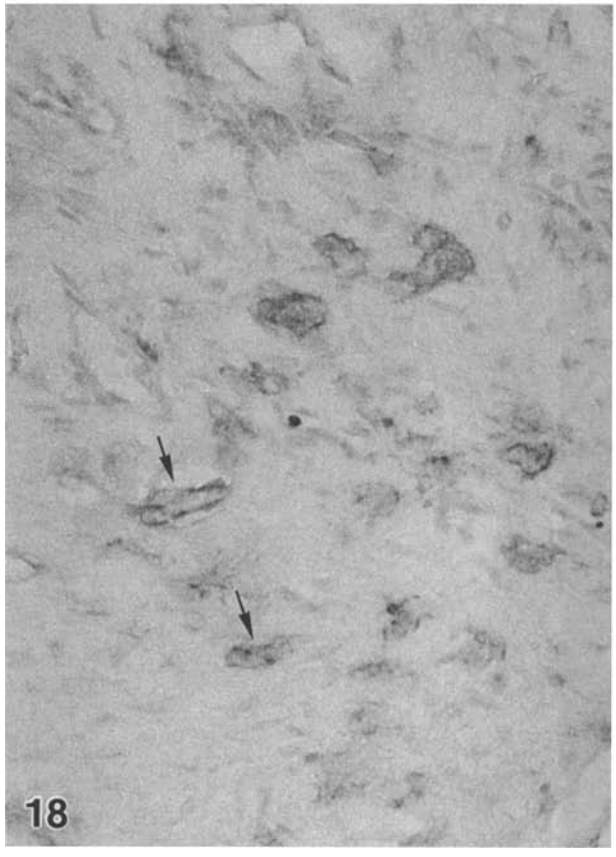

Fig. 12. An electron micrograph of a long spindle-shaped fibroblast in granulation tissue. × 5000

Figs. 13–17. Electron micrographs show a variety of "Fibrohistiocytoid (FH) cells" in granulation tissue. The "FH cells" include a series of cell types from a metamorphosized fibroblast (**13, 14, 15**) to a certain cell type which resembles a histiocytic fibroblast (**16, 17**). Note that the "FH cells" have an elliptical, angular large nucleoli, many fragmented, branching rough endoplasmic reticula, dispersed free ribosomes, small mitochondria and a few dense bodies. The nuclear envelope has complicated folds (**16**). × 5000

Fig. 18. Leu 10 immunostain in granulation tissue. Many positive cells, including fibroblastic cells (*arrows*) are found. Counterstain with methyl green. × 100

The filamentous bundles were fewer than those seen in the long, spindle-shaped fibroblasts.

The results of enzyme histochemical and immunohistochemical studies in granulation tissue are summarized in Table 4. The long, spindle-shaped fibroblasts reacted only to FN, HLA-A,B,C and occasionally to Fer. The short, spindle-shaped cells and the elliptical cells reacted to AcP, ANBE, HLA-DR, HLA-DP, Leu 10 (Fig. 18), OKT 9 and, occasionally, to Lys, AT, ACT, Leu M5 and ALP as well as FN, Fer and HLA-A,B,C. Histiocytes were, in addition, positive to monocyte/macrophage markers and complement receptors. The long spindle-shaped fibroblasts, the short, spindle-shaped cells and the elliptical cells demonstrated a diffuse cytoplasmic stain to Fer while histiocytes demonstrated a granular stain to Fer.

Discussion

Examination of granulation tissues showed, in addition to the typical elongated fibroblasts, morphologically differentiated short spindle-shaped cells and more elliptical cells. Their rER was decreased, fragmented and distributed in a branching pattern. Free ribosomes were dispersed. The mitochondria were increased in number and small. The cells had a few dense bodies resembling lysosomes, but not diverse fused lysosomes. There were poorly developed multivesicular bodies and no distinct pseudopodial cytoplasmic extensions. They had a round or elliptical nucleus, occasionally with prominent nuclear indentations, and one or two prominent nucleoli. Their nuclei and cytoplasm showed some similarities to both fibroblasts and histiocytes. In addition, enzyme histochemical and immunohistochemical studies of the short, spindle-shaped cells and the elliptical cells revealed a staining pattern intermediate between fibroblasts and histiocytes. These cells reacted to Fer, Lys, AT, ACT, FN, HLA-DR, HLA-DP, Leu 10, OKT 9, AcP, ANBE and ALP, but not to monocyte/macrophage markers and C3d- and C3bi-receptors. A diversity in the reaction pattern to Fer was noted: both the short, spindle-shaped cells and the elliptical cells and fibroblasts demonstrated a diffuse cytoplasmic stain while histiocytes were stained in a granular pattern. In addition, the short, spindle-shaped cells and the elliptical cells demonstrated no erythrophagocytosis, although they occasionally had a few secondary lysosomes. We proposed the term "fibrohistiocytoid (FH) cells" for both the short, spindle-shaped cells and the elliptical cells (Imai et al. 1987). This family includes a series of cell types ranging from a metamorphosized fibroblast (the short, spindle-shaped cell) which resembles a typical fibroblast, to a cell type which resembles a histiocytic fibroblast (elliptical cell), a cell which appears to be different from a histiocyte.

Hoffman and Dickersin (1983) have reported a cell type in the MFH called a "fibrohistiocyte" which demonstrated morphological characteristics both of a fibroblast and a histiocyte. This elongated cell had well-developed, dilated rER and, occasionally, a few microfibrils, observed as osmiophilic material deposited in the periphery of the cytoplasm. The morphology of this "fibrohistiocyte" is similar to the morphology of the short, spindle-shaped cell in our FH cell series, although the enzyme histochemical and immunohistochemical findings of these "fibrohistiocytes" are more similar to a fibroblast than to a histiocyte (Table 4). However, as illustrated in Figs. 16 and 17, we observed an additional cell type in our FH cell series which resembled a histiocytic fibroblast. In contrast to the "fibrohistiocyte", this elliptical cell demonstrated fragmented rER distributed in a branching pattern without marked dilatation, and the number of rER was decreased although the number of primary and secondary lysosomes was increased. The existence of cell types such as the FH cells in non-tumour tissues has been reported only rarely.

Although some morphological differences in the nucleoli and free ribosomes are found, the morphological findings of FH cells (the fibroblast-like and the histiocyte-like cells) in MFH are similar to cells in the FH cell series in granulation tissue (Table 3). Fibroblast-like cells are similar to the short, spindle-shaped cells and histiocyte-like cells are similar to the elliptical cells. In addition, a variety of cell types including the long, spindle-shaped, short, spindle-shaped, and elliptical cells in the tissues of the MFH, were observed and morphological cellular transitions from one to another seem likely.

Erythrophagocytosis, believed to be a feature of histiocytes and histiocyte-like cells in MFH, often demonstrate this phenomenon. One may not conclude from this finding that the histiocyte-like cell in the MFH is of histiocyte lineage. If MFH tumour cells originate from histiocyte lineage, more frequent erythrophagocytosis should be observed, and not only solitary but also compound erythrophagocytosis should be present. In addition, quite a few cell types (including synovial intimal cells, bladder epithelial cells and thyroid cells) other than histiocytes can demonstrate limited erythrophagocytosis (Ghadially 1982).

In the enzyme histochemical and immunohisto-

chemical studies, the fibroblast-like and histiocyte-like cells in the MFH reacted in a fashion similar to the FH series cells (Table 4). From morphological observations, Cozzutto et al. (1981), and Hoffman and Dickersin (1983) have concluded that MFH tumour cells are derived from fibroblasts, and Roholl et al. (1985b) have reported that MFH tumour cells react to monoclonal antibodies against human fibroblasts. Our results support a similarity between a majority of MFH tumour cells and FH cells, but not histiocytes.

Morphological evidence alone is insufficient to resolve the question of a possible independent cell lineage for the fibroblast-like and histiocyte-like cells in the MFH – as Fu et al. (1975) admit. However, if any morphological differences exist between both these cell types, as shown in Table 4, enzyme histochemical and immunohistochemical evidence demonstrates similarity. Furthermore, malignant FH cells (fibroblast-like cells and histiocyte-like cells) in the MFH were negative for peroxidase, monocyte/macrophage markers, and C3d- and C3bi-receptors, suggesting a dissimilarity in the enzyme histochemical and immunohistochemical phenotypes of malignant FH cells in the MFH from benign histiocytes in the granulation tissue. Rather, these malignant FH cell phenotypes appeared similar to benign FH cells in granulation tissue. As far as we know, no published data has revealed a distinct immunophenotypic difference between the fibroblast-like cells and the histiocyte-like cells in the MFH. Finally, an interpretation which may be more logically appealing postulates a common lineage for both cell types.

However, light and electron microscopy has little value in establishing the benign or malignant nature of a given FH cell. Our morphological study of the granulation tissue and MFH revealed that malignant FH cells in the MFH had hyperchromatic, enlarged nuclei with complex nuclear projection, prominent nuclear bodies, a large nucleolus, frequent atypical mitoses and abundant free ribosomes, and often had higher nucleus/cytoplasm ratios than benign FH cells in the granulation tissue. These findings, however, cannot distinguish between benign and malignant FH cells. The enzyme histochemical and immunohistochemical stainings did not demonstrated a distinct difference between either cell type. This suggests that, as in granulation tissue, benign fibroblasts and FH cells exist in the MFH as stromal cells in addition to the malignant FH cells, the fibroblast-like and histiocyte-like cells. In fact, small numbers of benign FH cells was found in the MFH, but morphological observation alone was insufficient to distinguish malignant from benign FH cells (data not shown).

Xanthomatous cells in the MFH are considered to be a special form of histiocyte because cellular atypism is less frequently observed. However, it is difficult to evaluate whether multinucleated giant cells are malignant. From our results, it appears that the morphological, enzyme histochemical and immunohistochemical features of this cell type are similar to either the histiocyte-like cell or a mixture of both the histiocyte-like and the fibroblast-like cell, suggesting that multinucleated giant cells are probably malignant.

Fumarola et al. (1982) demonstrated that low concentrations of lipopolysaccharide in culture transformed fibroblasts into histiocyte-like cells. We have also observed the transformation in culture of normal human dermal fibroblasts into histiocyte-like cells (which may be benign FH cells) but not into typical histiocytes (Takagi et al. 1988). As Wood et al. (1986) mentioned, fibroblasts are typically negative for ALP, while granulation tissue fibroblasts are positive for ALP. As far as we know, no published reports have revealed the transformation of fibroblasts into true histiocytes, except in a case of corneal fibroblasts (Kaye and Pappas 1962). Our results in this study suggest that normal fibroblasts and histiocytes are independent cell lines.

After treatment with IFN-γ, fibroblasts express HLA-DR antigen but not HLA-DQ (Geppert and Lipsky 1985). In our experiment, FH cells expressed both antigens while fibroblasts expressed neither. HLA-DR-expressing cells are considered to be antigen-presenting cells, and FH cells, as well as lymphocytes and macrophages may play a role in the immune reaction of chronic inflammation. However, the meaning of the Ia-like antigen expression of a majority of MFH tumour cells is still unknown. FH cells and major MFH tumour cells also react to OKT 9 (transferrin receptor), which is considered to be closely related to the rate of cell growth.

FH cells are found in various sites including bone marrow, peritoneum, skin, and lymph nodes under chronic inflammatory conditions. These sites also have a predilection for the development of MFH. This suggests that the similarity between FH cells and MFH tumour cells may be an important finding in the evaluation of MFH histogenesis.

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