Virchows Arch [Pathol Anat] (1983) 402:175–183

Virchows Archiv A

Granular cell tumor arising in myelinated peripheral nerves

Light and electron microscopy and immunoperoxidase study

Carlos D. Bedetti[1], A. Julio Martinez[2], N.S. Beckford[3], and Mark May[3]

[1] Pathology Department, Veterans Administration Hospital, University Drive C, Pittsburgh, PA 15240
[2] Pathology Department, Presbyterian-University Hospital, University of Pittsburgh School of Medicine, Pittsburgh, PA 15213
[3] ENT Department, Eye & Ear Hospital of Pittsburgh, University of Pittsburgh School of Medicine, Pittsburgh, PA 15213 USA

Summary. The histologic, immunohistochemical and ultrastructural characteristics of two granular cell tumors arising from the right recurrent laryngeal and left facial nerves are described. S-100 protein was detected both in the nuclei and cytoplasm of the granular cells using the peroxidase-anti-peroxidase method. The ultrastructural findings in both cases support a Schwann cell derivation of the granular cells. It is suggested that the granularity of cells of granular cell tumor may represent a lysosomal disorder affecting most frequently neoplastic and nonneoplastic Schwann cells and occasionally other cells.

Key words: Granular cell myoblastoma – Granular cell tumor – Peripheral nerve – Immunoperoxidase – Electron microscopy, Schwann cell

Introduction

The granular cell tumor, also known as granular cell myoblastoma, is an uncommon histopathological entity of unknown etiology and uncertain histogenesis. Among different theories on the histogenesis, the neurogenic theory is currently favored by several lines of evidence (Bangle 1952; Berman et al. 1978; Budzilovich 1968; Finkel and Lane 1982; Fisher and Wechsler 1962; Garancis et al. 1970; Nakazato et al. 1982; Sobel et al. 1971; Weisman et al. 1980). Its occurrence in a wide variety of organs including tongue, skin, respiratory tract, breast, gallbladder, neurohypophysis and other cutaneous and visceral sites has been well documented (Bangle 1952; Burston et al. 1962; Garancis et al. 1970; Strong et al. 1970). However, this lesion is rarely seen arising in major peripheral nerves (Budzilovich 1968; Weisman et al. 1980). The purpose of this report is to present two cases of granular cell tumors involving myelinated peripheral nerves: right recurrent laryngeal

Offprint requests to: C.D. Bedetti at the above address

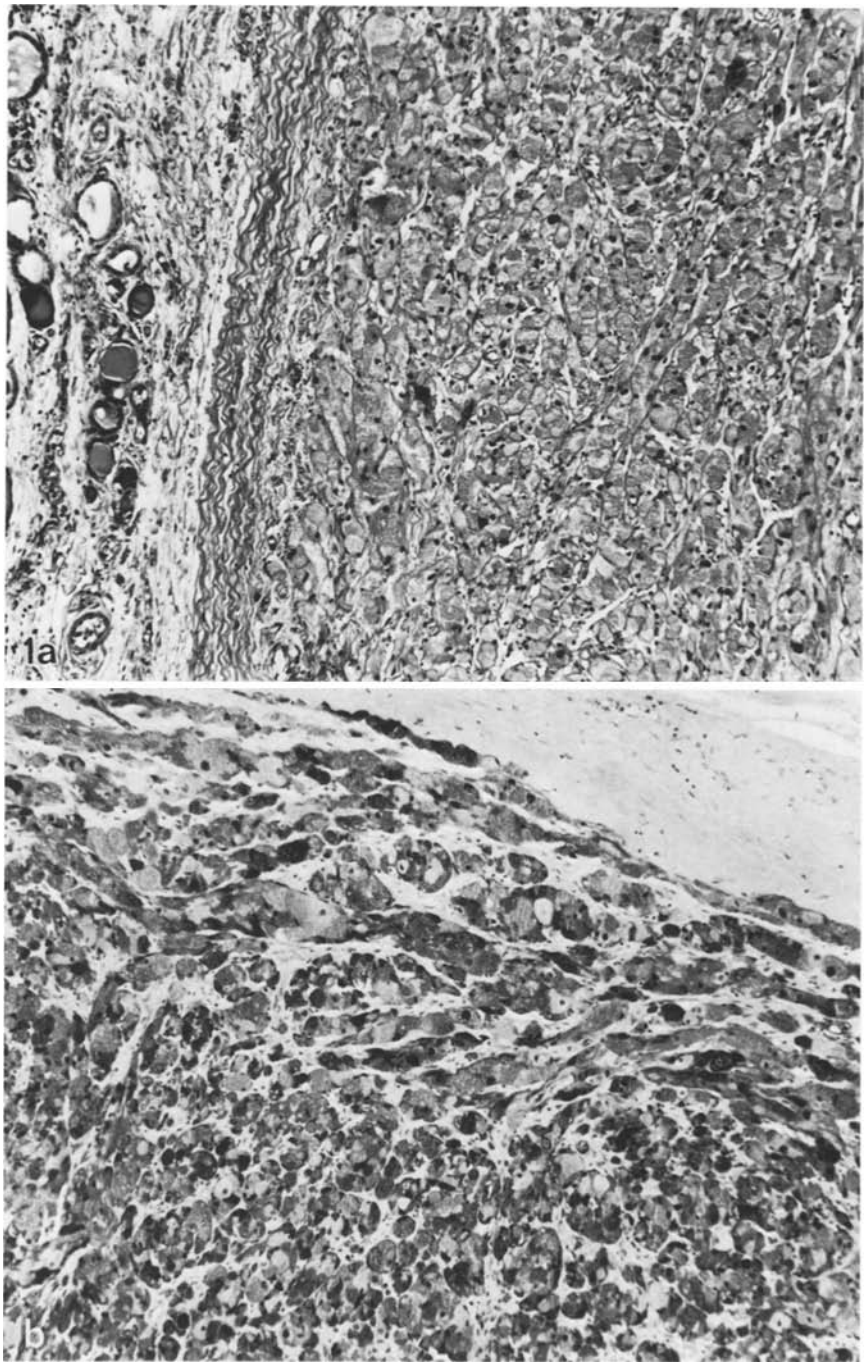

Fig. 1. A Granular cell tumor of the right recurrent laryngeal nerve. The tumor is composed of large, polyhedral cells with abundant granular cytoplasm and small dark stained nuclei. A fibrous capsule separates the tumor from the surrounding thyroid acini (Hematoxylin and eosin $\times 100$). **B** Immunoperoxidase method for S-100 protein shows uneven intense staining of the granular cells. Note the lack of staining of the fibrous capsule (right upper corner) and the connective tissue stroma ($\times 100$)

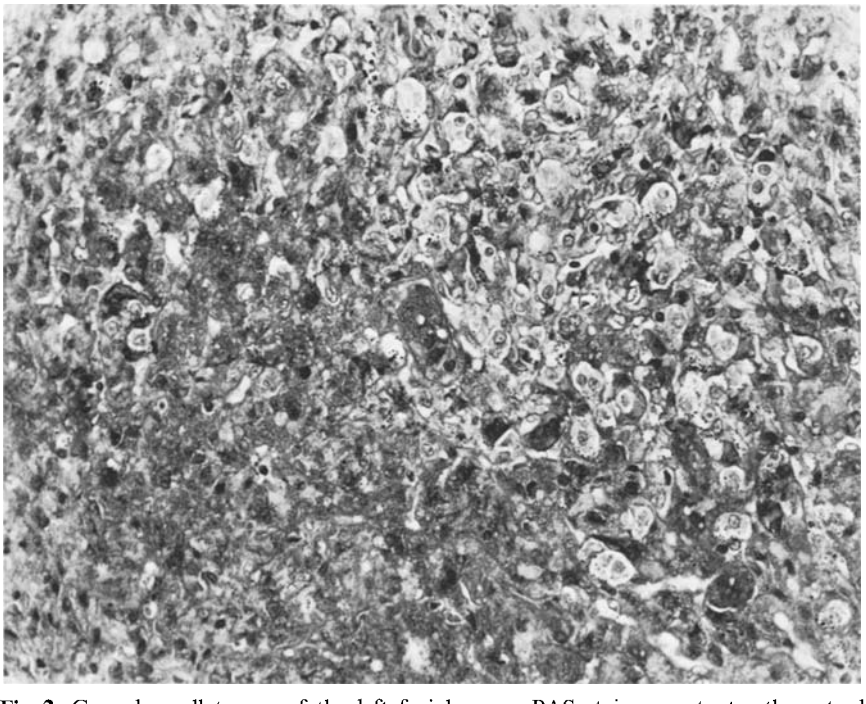

Fig. 2. Granular cell tumor of the left facial nerve. PAS stain accentuates the cytoplasmic granulation of the tumor cells. There is mild nuclear pleomorphism (× 250)

and left facial nerves. The ultrastructural findings in our cases and the detection of S-100 protein in the tumor cells by an immunoperoxidase method, further supports previous observations of the Schwann cell derivation of the granular cells.

Case report

Case 1. A 51 year-old black woman complained of a "lump on her throat" that had been present for the past six months. Physical examination revealed a firm solitary mass in the right lobe of the thyroid. The vocal cords moved normally and findings from the remainder of her physical examination were unremarkable. A thyroid scan revealed a cold nodule on the right lower lobe. Complete blood count, chest x-ray, urinalysis, and other laboratory tests were within normal limits. The right side of the neck was subsequently explored through an extended thyroidectomy incision. There was a 3.0 cm round, firm, white mass along the posterior and medial aspect of the right lobe of the thyroid. The mass completely surrounded the right recurrent laryngeal nerve as it became associated with the gland. A right thyroid lobectomy was performed and the right recurrent laryngeal nerve was sacrificed. The patient has been without evidence of recurrent disease for 22 months.

Case 2. A six year-old white girl, one month status post chicken pox, was admitted to Eye & Ear Hospital of Pittsburgh with a 14 day history of left facial nerve paralysis. Total paralysis progressed over four days and she was initially treated with steroids to no avail. Topagnostic testing showed intact hearing and tearing with loss of stapes reflex. A left transmastoid explora-

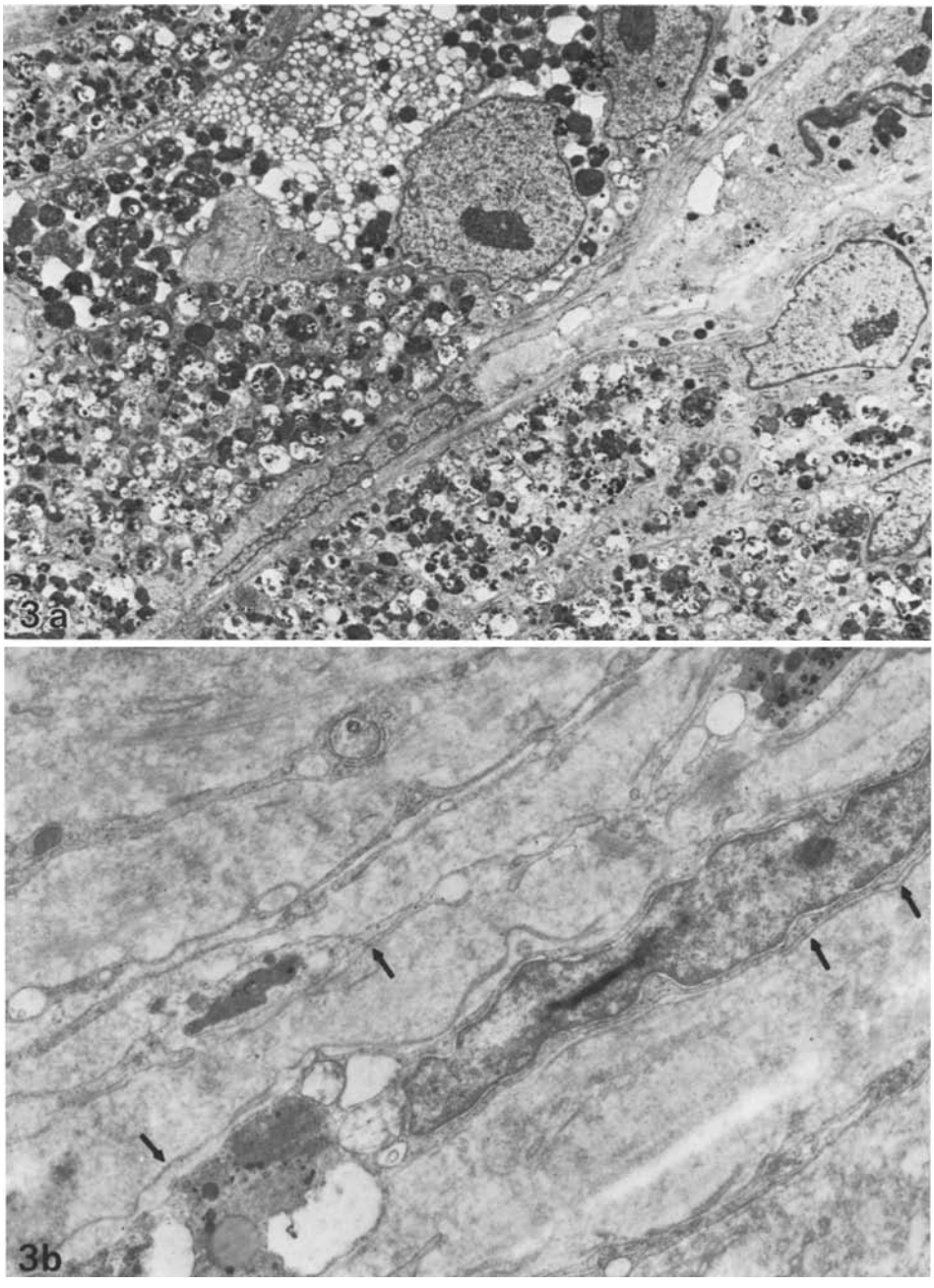

Fig. 3. A Electron micrograph of case #1 showing that the tumor cells contain an abundant cytoplasm filled with pleomorphic, electron dense, membrane-bound granules consistent with lysosomes. × 3,600. **B** Other areas of the same lesion are composed of elongated cells with few pleomorphic granules. Note that the cells and the thin cytoplasmic processes are incompletely enveloped by a well defined basement lamina (*arrows*). × 8,000

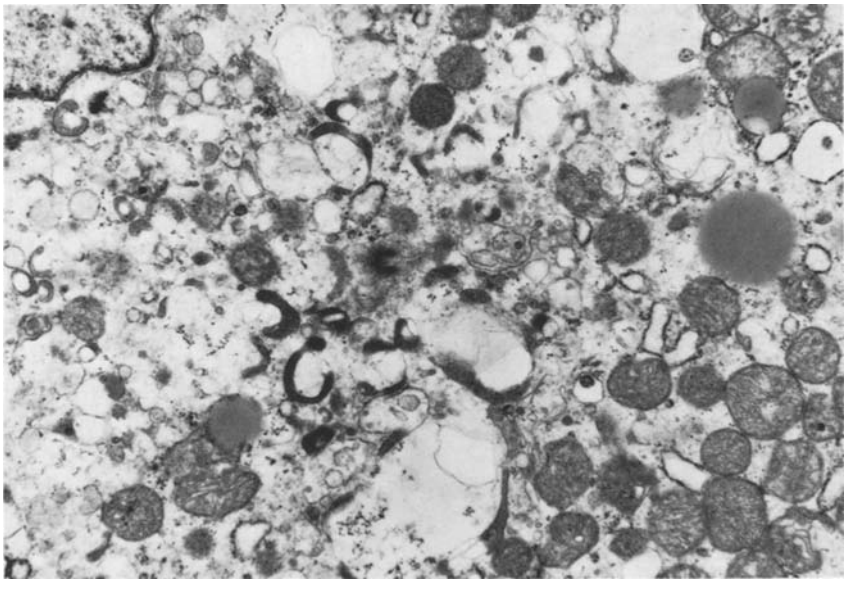

Fig. 4. Electron micrograph of case #2. The cytoplasm of this cell contains scattered mitochondria, lipid droplets, clear vesicles and some irregular dense bodies of probable lysosomal nature. These features are consistent with an early granular cell. ×10,000

tion revealed a 1.2 cm fusiform, pink, medium, firm tumor involving the left facial nerve from the pyramidal process to the stylomastoid foramen. This tumor was resected with placement of a greater auricular nerve graft. She is well and apparently free of disease ten months after surgery.

Methods

The resected specimens were processed in the following manner: Sections for light microscopy were fixed in 10% buffered formalin. They were embedded in paraffin, cut at 6 μm thickness and stained with hematoxylin and eosin (H&E), periodic acid Schiff reagent (PAS) with and without diastase and with Masson's Trichrome.

Representative formalin fixed paraffin embedded sections were immunostained using a standard peroxidase anti-peroxidase (PAP) technique with antibodies against S-100 protein (Sternberger 1979).

The primary antibody was prepared by Dr. H. Brent Clark from the Department of Pathology, Washington University in St. Louis, Missouri and it was used at 1:2000 dilution (Clark and Hartman 1981). Substitution of the primary antibody by normal rabbit serum and Schwann cells of myelinated nerves present in the tissue sections were simultaneously used as controls.

Tissues for electron microscopy were diced into small fragments and were fixed in 2% glutaraldehyde, washed thoroughly in Sorensen's phosphate buffer and post fixed in 1% solution of osmium tetroxide. After alcohol dehydration, they were embedded in Epon-Araldite®. Thin sections were stained with lead citrate and uranyl acetate and examined and photographed with a Philips 200 electron microscope.

Results

Light microscopy

Both lesions exhibited the typical histologic features of granular cell myoblastomas. The tumors were fairly well circumscribed but lacked a complete fibrous capsule. They were composed of clusters and sheets of large polyhedral to somewhat spindle cells with abundant eosinophilic granular cytoplasm (Fig. 1 A). The nuclei were round to oval and they showed mild to moderate pleomorphism. Mitoses were absent. The granules in the tumor cells were better seen with the periodic acid-schiff reaction (PAS) (Fig. 2). In case 1, the lesion was partially surrounded by thyroid tissue and myelinated axons were seen in the proximity of the granular cells. The myelinated fibers of the facial nerve (case 2) were partially replaced and infiltrated by granular cells and the tumor was confined within the perineurium.

Electron microscopy

The lesion involving the recurrent laryngeal nerve (case 1) had the typical ultrastructure of granular cell myoblastoma as has been described by others (Fisher and Wechsler 1962; Garancis et al. 1970; Sobel et al. 1971 and 1973a). Briefly, the cytoplasm of the tumor cells was filled with numerous membrane bound vacuoles of different size and content (Fig. 3 A). These vacuoles were morphologically consistent with lysosomes. The remainder of the cytoplasm contained very few mitochondria, and small fragments of endoplasmic reticulum. The cells were surrounded individually and in small groups by a well defined lamina. In addition, thin elongated cytoplasmic processes enveloped by basement lamina characteristic of Schwann cells, were often seen in the tumor cells (Fig. 3 B). "Myelin" figures and whorls of membranes were present within the cytoplasm. Interstitital cells near blood vessels contained large angulated bodies composed of stacks of fibrils.

The lesion involving the facial nerve (case 2) had similar ultrastructural features but the cells contained many mitochondria, a well developed endoplasmic reticulum, numerous empty vacuoles and few dense granules. These findings were consistent with the early myoblastoma cell described by Sobel et al. (1971 and 1973b).

Immunoperoxidase

Immunoperoxidase reaction for S-100 protein was intensely positive in Case #1 (Fig. 1 B). Both nuclei and cytoplasm of the granular cells were stained, however in Case #2 the reaction was weak with focally positive cells. The latter result was probably due to additional manipulation of the tissue during its use for frozen section diagnosis. Another possibility is that perhaps some cells had lost their capacity to synthesize S-100 protein. The Schwann cells of the adjacent myelinated nerve stained intensely with a dark-brown color and served as an internal positive control for the immunoperoxidase reaction.

Discussion

The light microscopic and fine structural features of the granular cells in the present tumors coresponded to that of granular cell tumors found elsewhere in the body. The occurrence of these tumors in major peripheral nerves has been rarely documented (Budzilovich 1968; Weisman et al. 1980). Only one previous report of a granular cell tumor involving the recurrent laryngeal nerve, similar to our first case, was found (Weisman et al. 1980). The involvement of the facial nerve by this type of lesion, our second case, has not been previously reported. The fact that this lesion can be associated with peripheral nerves further supports the concept of the neurogenic origin of granular cell tumor. In addition, the ultrastructural findings in both cases and the presence of S-100 protein in the tumor cells particularly in case 1, are further evidence of a Schwann cell derivation of the granular cells. The S-100 protein is a highly acidic nervous tissue specific protein which appears to be a useful marker for identifying neoplasms derived from Schwann cells and melanocytes (Nakajima et al. 1982; Nakazato et al. 1982; Stefansson et al. 1982; Armin et al. 1983).

The role and function of Schwann cells is not only to produce and maintain myelin, but they are considered pluripotential primitive cells which can multiply and proliferate like fibroblasts and may assume phagocytic properties (Church et al. 1973; Cravioto 1965; Martin and Webster 1973). They can also, under certain circumstances, produce collagen and have ciliated appearance (Bunge et al. 1967; Church et al. 1973). These features of Schwann cells indicate the limitations in the morphological interpretations and pitfalls that may occur when trying to identify the cell of origin and the histogenesis of granular cell tumors. Therefore, it is not necessary to postulate an undifferentiated fibroblast-like cell as the cell of origin of both granular cell myoblastoma and schwannomas as has been suggested by Sobel et al. (1973a and 1973b). The neurogenic origin of granular cell tumors is also supported by the presence of granular cells in suprasellar lesions and in the pituitary stalk resembling the same tumors found in other parts of the body (Burston et al. 1962). In addition, granular cell tumors have been recently described in association with neurofibromatosis, some of them having malignant transformation (Finkel and Lane 1982). On the other hand, changes histochemically and ultrastructurally indistinguishable from those seen in typical granular cell tumors have been documented in tumors of ameloblasts and smooth muscle cells (Christ and Ozzelo 1971; Navarrete and Smith 1971). The common denominator in all these lesions has been the eosinophilic granular appearance of the cytoplasm of the cells due to the accumulation of large number of lysosomes. On this basis, it is suggested that the granularity of cells of granular cell tumor may represent a lysosomal disorder affecting most frequently nonneoplastic and neoplastic Schwann cells and occasionally other cell lines such as ameloblasts and other mesenchymal cells. Most authors consider granular cell tumor to be a true neoplasm, but some have suggested that it may represent a degenerative change, a reactive process or a metabolic disorder (Azzopardi

1956; Garancis et al. 1970). It is of interest that in our second case, the facial nerve paralysis occurred approximately two weeks after the onset of chicken pox. The underlying granular cell tumor of the facial nerve exhibited ultrastructural features consistent with the early myoblastoma cell described by Sobel et al. (1971 and 1973b). It is tentative to speculate that the viral infection may have played an etiologic role in the development of the granular cell tumor in this young girl. However, unlike Fisher et al. (1962) we were unable to find virus like particles in the granular cells by electron microscopy. Although an attractive possibility, the role of a virus in the etiology of granular cell tumor remains to be elucidated. In summary, we describe two cases of granular cell tumors arising in major peripheral nerves. The anatomic site of these tumors, the ultrastructural findings and the presence of S-100 protein in the tumor cells strongly suggest the neural derivation of these lesions and their histogenesis from Schwann cells.

Acknowledgments. The authors are indebted to Drs. Daniel Santa-Cruz and H. Brent Clark from the Barnes Hospital, Division of Surgical Pathology and Department of Pathology and Washington University in St. Louis, Missouri for performing the immunoperoxidase technique. We also thank Dr. Victor Schramm for permission to use his case, and for supplying follow-up information in case #1.

References

Armin A, Connelly EM, Rowden G (1983) An immunoperoxidase investigation of S-100 protein in granular cell myoblastomas: Evidence for Schwann cell derivation. Am J Clin Pathol 79:37–44

Azzopardi JG (1956) Histogenesis of the granular-cell "myoblastoma". J Pathol Bacteriol 71:85–94

Bangle R (1952) A morphological and histochemical study of the granular-cell myoblastoma. Cancer 5:950–965

Berman JJ, Rice JM, Strandberg J (1978) Granular cell variants in a rat schwannoma. Evidence of neurogenic origin of granular cell tumor (Myoblastoma). Vet Pathol 15:725–731

Budzilovich G (1968) Granular cell "Myoblastoma" of vagus nerve. Acta Neuropathol (Berl) 10:162–165

Bunge MB, Bunge RP, Peterson ER, Murray MR (1967) A light and electron microscope study of long-term organized cultures of rat dorsal root ganglia. J Cell Biol 32:439–466

Burston J, John R, Spencer H (1962) "Myoblastoma" of the neurohypophysis. J Pathol Bacteriol 83:455–461

Christ ML, Ozzelo L (1971) Myogenous origin of a granular cell tumor of the urinary bladder. Am J Clin Pathol 56:736–749

Church RL, Tanzer ML, Pfeiffer SE (1973) Collagen and procollagen production by a clonal line of Schwann cells. Proc Nat Acad Sci USA 70:1943–1946

Clark HB, Hartman BK (1981) S-100 protein as an immunohistochemical marker for neoplasms of glial and Schwann cell origin. (Abstract). J Neuropathol Exp Neurol 40:335

Cravioto H (1965) The role of Schwann cells in the development of human peripheral nerves. J Ultrastruct Res 12:634–651

Finkel G, Lane B (1982) Granular cell variant of neurofibromatosis: Ultrastructure of benign and malignant tumors. Hum Pathol 13:959–963

Fisher ER, Wechsler H (1962) Granular cell myoblastoma – A misnomer. Electron microscopic and histochemical evidence concerning its Schwann cell derivation and nature (Granular Cell Schwannoma). Cancer 15:936–954

Garancis JC, Komorowski RA, Kuzma JF (1970) Granular cell myoblastoma. Cancer 25:542–550

Martin JR, Webster HdeF (1973) Mitotic Schwann cells in developing nerve: Their changes in shape, fine structure, and axon relationships. Dev Biol 32:417–431

Nakajima T, Watanabe S, Sato Y, Kameya T, Shimosato Y, Ishihara K (1982) Immunohistochemical demonstration of S-100 protein in malignant melanoma and pigmented nevus, and its diagnostic application. Cancer 50:912–918

Nakazato Y, Ishizeki J, Takahashi K, Yamaguchi H (1982) Immunohistochemical localization of S-100 protein in granular cell myoblastoma. Cancer 49:1624–1628

Navarrete AR, Smith M (1971) Ultrastructure of granular cell ameloblastoma. Cancer 27:948–955

Sobel HJ, Marquet E, Avrin E, Schwarz R (1971) Granular cell myoblastoma. An electron microscopic and cytochemical study illustrating the genesis of granules and aging of myoblastoma cells. Am J Pathol 65:59–78

Sobel HJ, Marquet E, Schwarz R (1973a) Is Schwannoma related to granular cell myoblastoma? Arch Pathol 95:396–401

Sobel HJ, Schwarz R, Marquet E (1973b) Light and electron-microscope study of the origin of granular-cell myoblastoma. J Pathol 109:101–111

Stefansson K, Wollmann R, Jerkovic M (1982) S-100 protein in soft-tissue tumors derived from Schwann cells and melanocytes. Am J Pathol 106:261–268

Sternberger LA (1979) Immunocytochemistry, 2nd edition. Wiley & Sons, New York, Chichester, Brisbane, Toronto

Strong EW, McDivitt RW, Brasfield RD (1970) Granular cell myoblastoma. Cancer 25:415–421

Weisman RA, Konrad HR, Canalis RF (1980) Granular cell myoblastoma involving the recurrent laryngeal nerve. Arch Otolaryngol 106:294–297

Accepted September 26, 1983