Autoradiographic Study of Parasympathetic Paragangliomas (Chemodectomas) of the Neck

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Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 137, No. 4, pp. 447-450, April, 2004
Original article submitted January 19, 2004

Study of the morphofunctional characteristics of chemodectomas of the neck by photoautoradiography (semithin sections) showed the absence of protein-producing and proliferative activity in light and dark tumor cells against the background of strained biosynthetic processes in stromal cells. Invasion of capillary walls by tumor cells with their release into the capillary lumen is a prerequisite for metastases of these tumors.

Key Words: *chemodectomas; proliferation; protein producing activity; autoradiography*

Publications on endocrine tumors, specifically, on parasympathetic paragangliomas (chemodectomas) of the neck indicate great interest of specialists to the problem of diagnosis and treatment of these tumors [2,4]. However, the majority of publications are addressed to clinicians, while morphological reports are scanty [3].

Our previous histological study of these tumors [1] showed that they retain the main morphological features of initial chemoreceptor tissue (neuroendocrine structure). In most cases these tumors retain their alveolar structure, but solid, fibrous, and angiomatous variants were also observed.

Our aim was more detailed study of the morphology of chemodectomas and their morphofunctional features by autoradiography.

MATERIALS AND METHODS

Specimens of tumor tissue resected in 7 patients aged 40-50 years were examined.

The material was placed into warm medium 199. Fragments (1 mm^3) were incubated in a medium containing 100 μCi/ml ^{3}H-uridine (specific activity 26.0 Ci/mM) or 20 μCi/ml ^{3}H-thymidine (specific activity 21.6 Ci/mM) for 1.5 h at 37°C. Autoradiographs were prepared as described previously [2], examined and photographed under a Leitz light microscope.

RESULTS

The tumors had primarily alveolar and/or solid structure. The alveoles were usually of different size, some of them had no clear-cut interface because of changes in tumor cells. Alveoles contained light and dark cells (Fig. 1, *a*). The interface between the cells was poorly not discernible; they differed from each other by the structure of the nucleus. Light cells had large polymorphic clarified nuclei with low chromatin content usually finely distributed along the nuclear membrane; nucleoli were sometimes seen. Dark cell nuclei were usually smaller and hyperchromatic. Tumor cell nuclei did not incorporate labeled precursors (^{3}H-uridine or ^{3}H-thymidine), which attested to the absence of protein-producing and proliferative activities by the moment of analysis.

Tumor tissue contained abundant vessels of different size, some of them had sinusoidal structure. Some vessels retained their structure and functions, others demonstrated destructive changes (Fig. 1, *b*). Endotheliocytes of preserved vessels intensively incorporated labeled uridine, *i. e.* synthesized RNA, which attested to their high functional activity. Some cells retained their functional activity even after vessel destruction: they produced RNA, which was seen from

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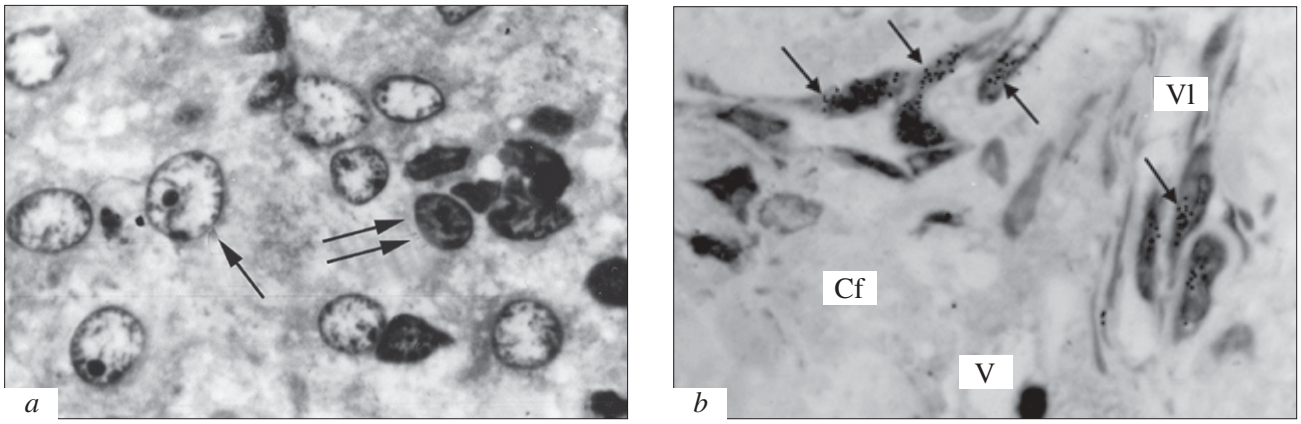

Fig. 1. Tumor tissue. *a*) two types of cells: light (arrow) and dark (double arrow). Polymorphism and chaotic disposition of cell nuclei; *b*) microvessels; left: vessel with intact endothelial wall, RNA produced in endotheliocyte nuclei (arrows), right: vessel in the process of destruction (arrow); Cf: degeneration of collagen fibers; Vl: vascular lumen; V: vacuole. Here and in Figs 2, 3: semithin sections. Toluidine blue staining, ×1000.

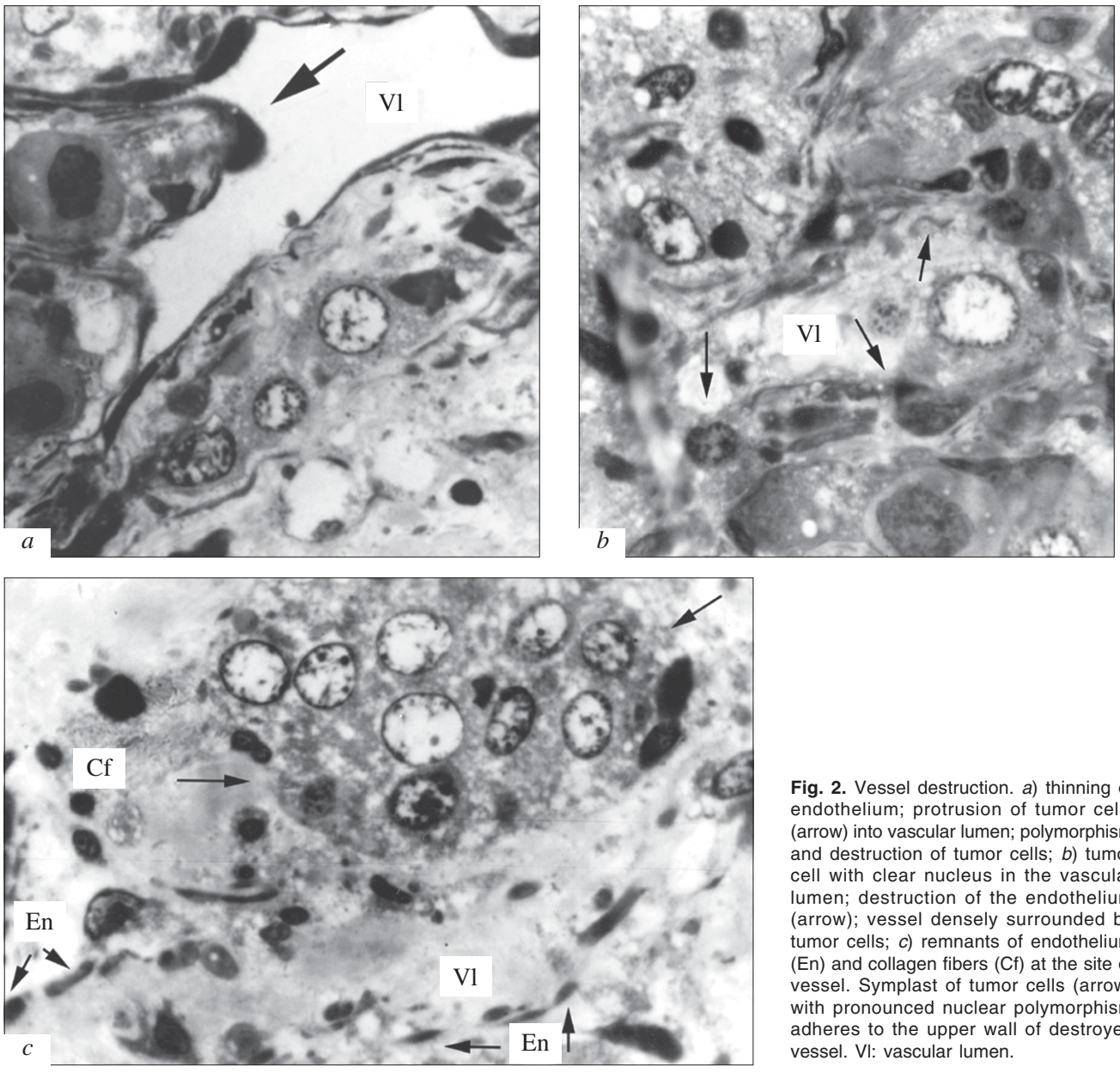

Fig. 2. Vessel destruction. *a*) thinning of endothelium; protrusion of tumor cells (arrow) into vascular lumen; polymorphism and destruction of tumor cells; *b*) tumor cell with clear nucleus in the vascular lumen; destruction of the endothelium (arrow); vessel densely surrounded by tumor cells; *c*) remnants of endothelium (En) and collagen fibers (Cf) at the site of vessel. Symplast of tumor cells (arrow) with pronounced nuclear polymorphism adheres to the upper wall of destroyed vessel. Vl: vascular lumen.

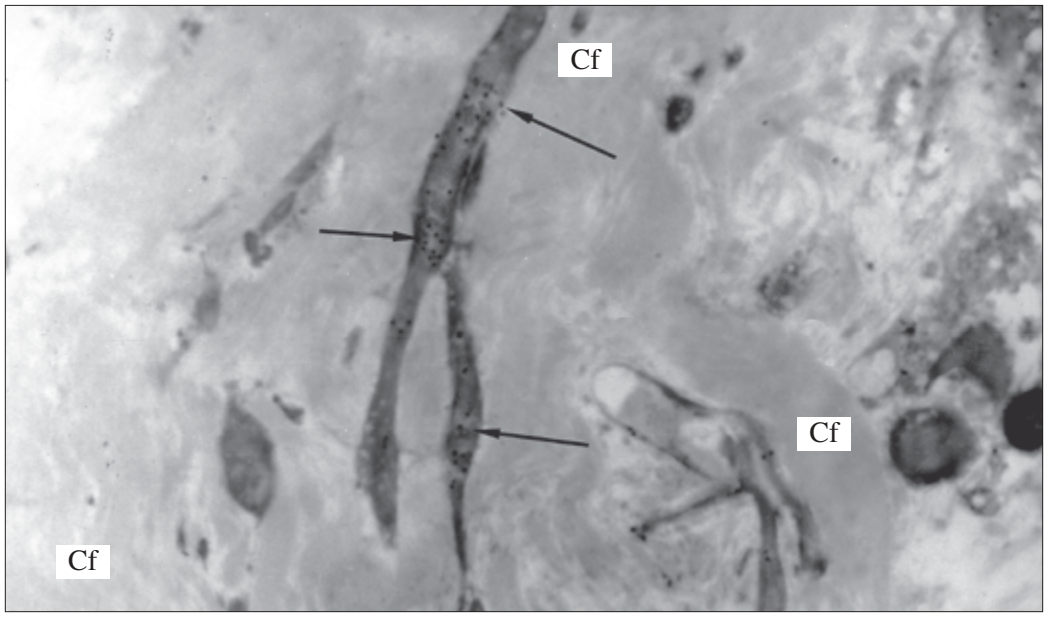

Fig. 3. Giant fibroblasts (arrows) with intense RNA synthesis (dark silver grains) in tumor stroma. Amorphous collagen fibers (Cf).

^{3}H-uridine incorporation into cell nuclei. DNA synthesis was rarely observed: in some cells of vascular walls (endotheliocytes, pericytes) or in the immediate vicinity of them. Capillaries were best of all preserved among all microvessels. Sinusoidal vessels were as a rule destroyed, the greater part of endothelial lining was destroyed and remaining endotheliocytes were thinned or swollen, often vacuolated, with pyknotic nuclei. However, active destruction of vessels was more often observed as a result of their invasion with tumor cells. Different stages of this process were observed, up to tumor cell penetration into vascular lumen. At the first stage the vessel was densely surrounded by tumor cells, the endothelium was thinned, destroyed, and somewhere protruded into the vascular lumen (Fig. 2, *a*). The second stage was characterized by almost complete destruction of the endothelium and its replacement with tumor cells, some cells penetrated into the lumen, the lumen of destroyed vessels was filled with tumor cells or detritus (Fig. 2, *b*). After destruction of the vessel, which was in this case presented by endotheliocyte fragments, collagen fibers grew into its lumen (Fig. 2, *c*).

Giant fibroblasts were seen, their nuclei intensely synthesized RNA (^{3}H-uridine incorporation; Fig. 3). These cells were usually surrounded by collagen fibers with signs of degeneration (loss of fibrillar structure, amorphous structures). The appearance of these hypertrophic cells in the stroma is probably a reaction to progressive hypoxia in the tumor tissue resulting from vessel compression and destruction.

Thus, protein synthesis and proliferative activity were absent in light and dark tumor cells. This can be due to the absence of cell activity at this stage and to the fact that these cells rarely proliferate, because these tumors, though malignant, grow slowly, and we failed to detect DNA synthesis in them. We can rule out the error of the method in this case with great confidence, because we constantly observed unlabeled tumor cells beside intensely labeled cells of the vascular walls and fibroblasts, which attested to different functional status of these cells.

Functional activity of cells in the vascular wall (endotheliocytes, pericytes) and fibroblasts (intense incorporation of ^{3}H-uridine) attests to a strain of biosynthetic processes in the stroma of tumor tissue.

Invasion of the vascular wall by tumor cells and its destruction with the release of these cells into the vascular lumen is a prerequisite for tumor metastasizing, which, along with other morphological signs (for example, pronounced polymorphism) confirms malignancy of these tumors.

The appearance of hypertrophic giant cells in the stroma can be a result of augmenting hypoxia in tumor tissue resultant from vessel compression and destruction.

REFERENCES

1. V. N. Dan, E. G. Matyakin, L. S. Kokov, *et al.*, *Angiol. Sosud. Khir.*, No. 2, 109-115 (2000).
2. A. A. Pal'tsyn, *Microscopic Methods* [in Russian], Moscow (1996), pp. 253-283.
3. N. T. Raikhlin, E. A. Smirnova, Kh. V. Badalyan, *et al.*, *Arkh. Patol.*, No. 5, 18-23 (2000).
4. E. Solcia, G. Kloppel, L. H. Sobin, and E. D. Williams, *Histological Typing of Endocrine Tumors*, Berlin, New York (2000), pp. 39-44.