DETECTION OF SMOOTH MUSCLE MYOSIN IN THE THECA CELLS OF THE OVARIES

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Smooth muscle myosin was detected by the indirect Coons' method, using an appropriate monospecific immune serum, in the theca cells of the ovaries in rodents.

KEY WORDS: ovary; theca cells; smooth muscle myosin; immunofluorescence.

The first communications on smooth muscle cells in the membrane of vertebrate ovarian follicles were written last century [6]. Since that time many researches have been published both confirming this fact [8, 12, 14, 17-19] and disputing it [10, 11, 16, 20]. The contradictory results can probably be explained by the morphological and functional properties of the ovarian stromal cells, which differ from those of ordinary connective tissue [2, 21]. Having noted this difference, Glazunov [1] proposed the name "cells of the theca tissue" for them, since the tumors arising from stromal cells differ both from ordinary fibromas and from leiomyomas and they can be distinguished as a special group – the thecomas [1, 13].

Electron-microscopic investigation revealed three groups of cells in the ovarian stroma of vertebrates, including man: 1) typical fibroblasts, 2) typical smooth-muscle cells; 3) fibroblast-like or undifferentiated smooth-muscle cells [8, 18]. The inclusion of the cells in a particular group was based chiefly on the presence or absence of filaments connected with the dense bodies. However, as Lazarides and Weber [15] showed

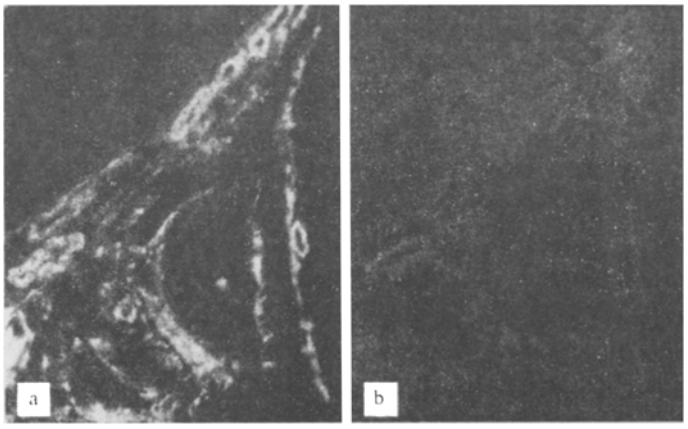

Fig. 1. Section through rat ovary: a) fluorescence of muscles in blood vessels and theca cells after treatment with antiserum against smooth muscle myosin by the indirect Coons' method. In the center, an atretic follicle; on the right and left, membranes of corpora lutea with adjacent blood vessels, 60 ×; b) control section treated with nonimmune serum by indirect Coons' method. Weak nonspecific fluorescence, 60 ×.

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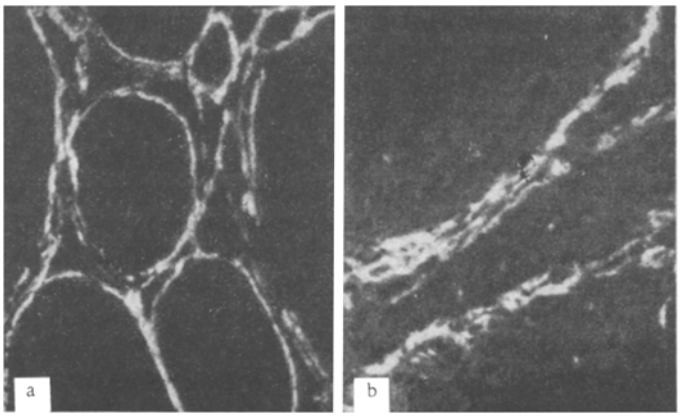

Fig. 2. Section through mouse ovary. Treated with antiserum against smooth muscle myosin by the indirect Coons' method: a) fluorescence of membranes of follicles at different stages of development. On right and left, segments of corpora lutea, 30 ×; b) the same section under higher power (120 ×). Fluorescent fusiform cells in membrances of ripe follicles.

recently, by the use of antiserum reacting specifically with the actin of skeletal myoblasts, fibroblasts may also contain contractile fibrils corresponding to actin.

This fact, like the existence of intermediate cells between fibroblasts and smooth muscle cells, probably makes it harder to identify stromal cells purely on the basis of electron-microscopic investigations and may explain the lack of unanimity about the localization of smooth muscle in the ovary [9, 12, 17-19].

Discovery of smooth muscle myosin in the cells would be direct proof of their smooth-muscle nature. The absence of such evidence for the theca cells of the ovary was the justification for the present investigation.

EXPERIMENTAL METHOD

Ovaries of guinea pigs, rats, and mice were studied. Serial cryostat sections 6 μ in thickness were fixed for 10 min in 96° ethanol and treated by the indirect Coons' method with rabbit antiserum against human smooth muscle myosin [3], using pure donkey antibodies against rabbit immunoglobulin G [7], labeled with fluorescein isothiocyanate, eluted from an immunosorbent. Control sections were incubated with nonimmune sera.

EXPERIMENTAL RESULTS

In all the ovaries studied fluorescence of the muscular layer of the blood vessels and theca cells was found in sections treated with antiserum against smooth muscle myosin. In control sections incubated with nonimmune serum no fluorescence was present (Fig. 1a, b). Fluorescent fusiform cells (Fig. 2b) formed a network of 1-3 layers ensheathing the corpora lutea and secondary follicles at different stages of development, including atretic follicles (Figs. 1a and 2a). Bands and single cells also were found in the medullary layer and in the hilus among the numerous blood vessels there. The cortical zone was ill defined in these rodents because of the abundance of follicles and corpora lutea and it was therefore difficult to decide what was the typical distribution of smooth muscle in them. Usually individual cells or groups of a few cells among other cortical elements were found in the sections.

Although the smooth-muscular layer around the corpora lutea was more strongly developed than around the follicles, its fluorescence was appreciably weaker.

The fluorescence of all cells (of both the muscular layer of the vessels and the theca cells) in the sections of the guinea pig ovary also was much weaker than in the ovaries of the rats and mice; this evidently points to a very small number of common antigenic determinants in human and guinea pig myosin.

Since the monospecificity of the antiserum used and its ability to give crossed reactions with smooth muscle myosin of animals were verified in previous investigations [3-5], the results now obtained reliably

confirmed the presence of smooth muscle myosin in the theca cells of the ovary, i.e., the smooth-muscle nature of some of the stromal cells.

No definite conclusion can be drawn from these observations regarding the function of these cells, but considering the results of physiological studies [14] it can be postulated that they participate in the process of ovulation and, in particular, in the collapse of the ruptured follicle.

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