

increase the unspecific fluorescence and does not interfere with the normal immunofluorescent reaction characteristic for amphibian lens proteins. The specific fluorescence was not observed in the sections that were stored for 2 years or more. Thus, antigenic determinants of specific lens proteins of amphibian embryos persist in stained sections at room temperature for 1–1.5 years. These antigen determinants

can be identified using conventional techniques of immunofluorescence.

Attempting to extrapolate this data to other antigenic systems¹⁴ one has to take in account that certain histological stains such as methylene or aniline blue, acidic or basic fuchsin, Congo-red, hematoxyline-eosin, and others^{11–13} lead to a decrease of the immunofluorescence.

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Bovine parathyroid catecholamines: A chemical and histochemical study

D.M. Jacobowitz and E.M. Brown

Laboratory of Clinical Science, Bldg. 10, Rm. 2D46, NIMH, and Metabolic Disease Branch, Bldg. 10, Rm. 9D20, NIAMDD, Bethesda (Maryland 20014, USA), 19 March 1979

Summary. Bovine parathyroid glands contain large amounts of dopamine (3.4–13.9 pg/μg), but very little norepinephrine. Fluorescent histochemistry demonstrates only rare adrenergic nerve terminals on vasculature. Single dopamine-containing cells, most likely mast cells, are scattered in large numbers throughout the connective tissue stroma.

Calcium is generally thought to be the principal physiologic regulator of parathyroid function¹. Recently, however, a number of other agents have been shown to alter parathyroid hormone release in vivo and in vitro. These are: β -adrenergic^{2–4}, α -adrenergic⁵ and dopaminergic⁶ catecholamines, prostaglandin E₂⁷ and secretin⁸. The presence of receptors for catecholamines on dispersed bovine parathyroid cells suggests the possibility of direct innervation of the bovine parathyroid gland. Although nerve endings, apparently terminating on parathyroid cells, have been

demonstrated by light microscopy^{9,10}, electron microscopy¹¹ and fluorescence histochemistry¹², there is relatively little information on the innervation of the bovine parathyroid gland¹³. In the present report, we have directly measured catecholamine levels in bovine parathyroid tissue and employed fluorescence histochemistry to investigate the possibility of catecholamine-containing nerve endings in this species.

Methods. Bovine parathyroid tissue was obtained from a local abattoir within 5–10 min of death. Glands were cut into slices (about 2×4×4 mm) with a scalpel and placed immediately on dry ice for determination of catecholamines or in liquid nitrogen for fluorescence studies. Catecholamines were assayed according to modifications¹⁴ of the method of Coyle and Henry¹⁵. The frozen tissue was processed for fluorescent histochemistry¹⁶. The tissue was then freeze-dried at –30 °C for 4 days and exposed to dry paraformaldehyde gas at 80 °C for 1 h, vacuum embedded in paraffin for 20 min at 60 °C and sectioned at 14 μm.

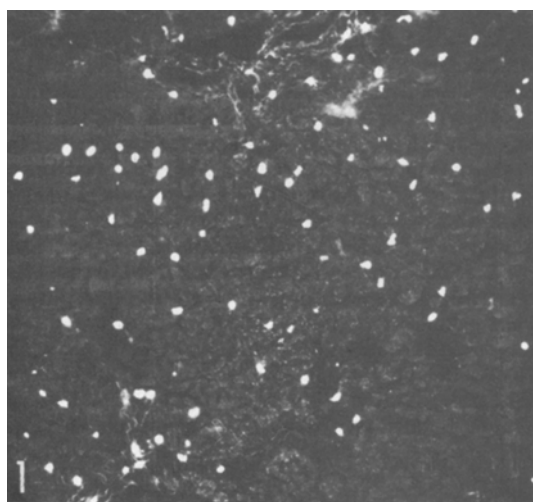


Fig. 1. An abundant number of fluorescent cells scattered throughout the connective tissue stroma of the parathyroid gland. ×100.

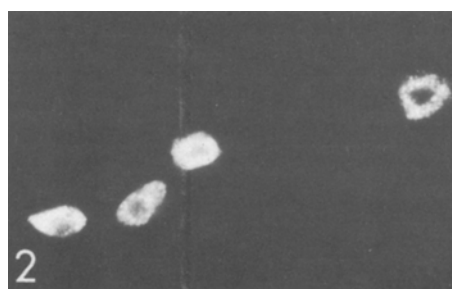


Fig. 2. High power of fluorescent cells, probably mast cells, whose nuclei are unstained. ×655.

Results. Bovine parathyroid catecholamines. Slices of bovine parathyroid tissue contained large amounts of dopamine (3.4–13.9 pg/ μ g). Norepinephrine values were insignificant (less than twice blank). The detection limit for norepinephrine in this assay is 5 pg.

Histofluorescence studies. Only rare green fluorescent norepinephrine-containing nerve endings were observed terminating on parathyroid vasculature. No definite nerve endings were identified on endocrine parathyroid cells per se, which exhibited reddish autofluorescence. Large numbers of single, yellow-green fluorescent cells were observed, scattered diffusely throughout the connective tissue stroma of the gland (figure 1). At higher power, these fluorescent cells could be seen containing greenish-yellow granules, which in some cases appeared to have been released from the cells (figure 2).

Discussion. Unlike the human parathyroid gland¹², we found no evidence of direct catecholamine innervation of parathyroid cells in the bovine parathyroid gland. Sparse norepinephrine-containing nerve endings were observed, but these appeared to terminate exclusively on small blood vessels. This confirms previous electron microscopic evidence of vascular innervation in the other species¹⁷. Thus, if the α -⁵ and β -adrenergic²⁻⁴ receptors on the bovine para-

thyroid cell are of physiologic importance, they must mediate responses to circulating epinephrine. There is, at present, no evidence in this species for a role of circulating catecholamines in modulating parathyroid function in vivo. The presence of large quantities of dopamine in the bovine parathyroid gland is of some interest. By histofluorescent microscopy, dopamine appeared to be localized to single cells scattered diffusely throughout the bovine gland in the connective tissue septa. The bovine parathyroid has been shown previously to contain an abundant number of mast cells¹⁸. Moreover, bovine mast cells contain large amounts of dopamine¹⁹. It is likely, therefore, that dopamine-containing cells within the parathyroid represent mast cells. We have demonstrated that dopamine causes 30- to 40-fold increases in intracellular cAMP in dispersed bovine parathyroid cells with concomitant 2- and 4-fold increases in parathyroid hormone release⁶. It is conceivable, therefore, that release of dopamine from bovine parathyroid mast cells might indirectly stimulate parathyroid hormone secretion. Although various stimuli regulate mast cell release (e.g., immunologic), adrenergic and cholinergic^{20,21} stimuli may modulate the release reaction. A more direct experimental approach will be required to determine if an analogous mechanism is involved in the regulation of parathyroid function.

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Hypophysectomy exerts a radioprotective effect on frog lens¹

J.H. Hayden, H. Rothstein², B.V. Worgul and G.R. Merriam, Jr

Kresge Eye Institute of Wayne State University, Detroit (Michigan 48201, USA), and Department of Ophthalmology, Columbia University College of Physicians and Surgeons, New York (New York 10032, USA), 22 August 1979

Summary. Exposure to X-rays usually causes cataracts in frogs. These cataracts are always preceded by misalignment of the structures called meridional rows (MR). When cell division is completely halted by hypophysectomy, however, irradiation no longer disturbs the orientation of the MR. Since the MR are the structures formed as lens epithelial cells differentiate into lens fibres it is reasonable to propose that radiocataractogenesis depends upon a mitosis-driven formation of pathological fibres from epithelial cells that have been rendered abnormal by exposure to X-rays.

Though he did not choose this interpretation himself, the early work of Chaloupecky^{3,4} raised the possibility that X-rays cause cataracts. The older literature is reviewed in Poppe's dissertation⁵. Worgul and Rothstein recently reemphasized the suggestion that the formation of radiation cataracts depends upon injury to the lens epithelium that eventuates in pathological fibre formation⁶. A strong element in their working hypothesis is that cell proliferation is

required in order to transform the damage sustained by the lens epithelium into abnormal fibres⁷.

Figure 1 shows the important cytoarchitectural features of the frog (*Rana pipiens*) lens. It was noted by Worgul and Rothstein that whenever cataracts developed, the region known as the meridional rows (MR) was disorganized. Where the mitotic index was found to be naturally low (as in a population of animals from South Dakota) or where it