VGF-like immunoreactivity in amphibian hypothalamo-hypophysial system.

VGF-like Immunoreactivity in Amphibian Hypothalamo-Hypophysial System

SILVIA DE MARCHIS,[a] MARJORIE LECOMTE[b], MARIA-FOSCA FRANZONI,[a]
PAOLA MARITANO,[a] EZIO CAMPANTICO,[a] GIAN-LUCA FERRI,[c]
AND ALDO FASOLO[a]
[a]Department of Animal Biology, University of Torino, Turin, Italy
[b]Laboratory of Molecular and Cellular Neuroendocrinology, University of Rouen, Rouen,
France
[c]Laboratory of Cytomorphology, University of Cagliari, Cagliari, Italy

VGF is a neurotrophin-inducible early-delayed gene that encodes a 617-amino-acid polypeptide–VGF protein–bearing no significant homology with known sequences. The VGF protein is selectively expressed in specific groups of cells in the mammalian central nervous system, particularly in the hypothalamus (e.g., suprachiasmatic, paraventricular, and supraoptic nuclei)[1] and in endocrine cells such as adenohypophysial and adrenal medullary cells.[2] VGF is stored and transported in secretory granules and processed to intermediate to small molecular weight products, which are preferentially released.[3]
In the present immunohistochemical study, the distribution of VGF protein was examined in the hypothalamo-hypophysial complex of two anuran species: *Xenopus laevis* and *Rana esculenta*. The two antibodies a-VGF[d] used were: anti-VGF_{215}, raised against the C-terminus, and anti-VGF_{216}, against a potential cleavage product of VGF. Double labellings were performed in a simultaneous procedure using a-VGF and a-LH[e] (luteinizing hormone) antibodies.

VGF NEUROENDOCRINE LOCALIZATION

In *Xenopus laevis*, the hypothalamus is a major site of VGF localization: many VGF-immunoreactive (-ir) fibers and scattered bipolar neurons were detected in the anterior and supra/retrochiasmatic areas. The most conspicuous VGF-ir cell group (liquor-contacting neurons) was observed in the paraventricular organ (PVO) (FIG. 1a). Immunostained beaded fibers were also observed along the preoptic-hypophysial tract, and a strong VGF-immunoreactivity was found in the fibrous layer of the median eminence and in the pars nervosa (FIG. 1b) of the pituitary (in perivascular terminals and Herring bodies). Small clusters of VGF-ir cells were seen in the pars distalis, while no reaction was detected in the pars intermedia of adenohypophysis.
In *Rana esculenta*, only after a-VGF_{216} incubation, a strong reaction was observed in clusters of liquor-contacting neurons in the lateral infundibular recesses of the posterior hypothalamus. No positivity was detected in other hypothalamic areas and in pituitary pars nervosa. In contrast, numerous adenohypophysial cells were immunopositive in the pars distalis, where, after double immunostaining, some cellular colocalizations of VGF- and LH-immunorectivity were found. VGF-ir fibers were also seen, after a-VGF_{216} incubation, in the pars intermedia of the gland.

[d]From G.L. Ferrri, university of Calgliari (Italy).
[e]From Affinity Research Products (Ilkeston, United Kingdom).

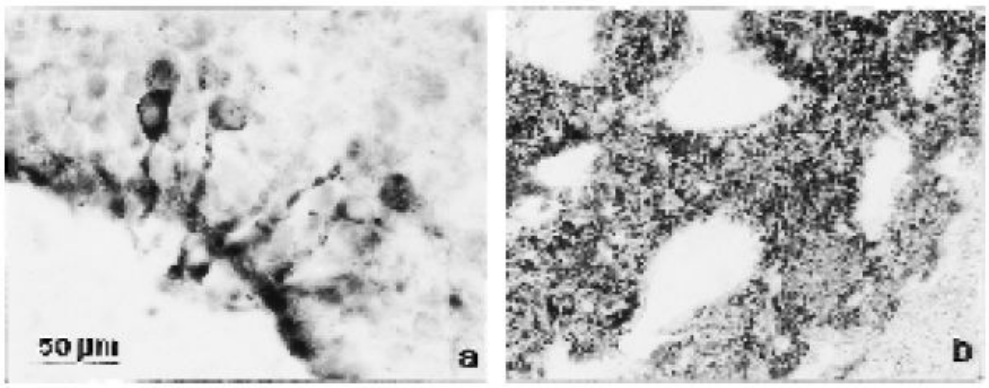

FIGURE 1. Sections of PVO (a), and pituitary pars nervosa (b) of *Xenopus laevis* immunostained with a-VGF antisera.

The present data show the occurrence of a VGF-like antigen in the CNS of amphibians. The observed VGF-immunoreactivity in various mammalian species,[1,2] as well as in amphibians (present study), indicates a phylogenetic conservation of this protein, although the different distribution patterns observed with the two antibodies (VGF_{215} and VGF_{216}) might reflect species-specific processing of the molecule.

It has been suggested that in mammals VGF may represent a precursor of bioactive peptides and control pituitary secretion. Given the abundant VGF-immunoreactivity in the hypothalamo-hypophysial complex, we propose that a similar role could be active also in the neuroendocrine regulation mechanisms of amphibians.

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