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PITUITARY - LOBULATION AND SEASONAL CHANGES OF THE BASOPHIL PITUITARY IN CYPRINIDS

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Abstract

The pituitary gland is noted through distinct particularities to those of the other vertebrates. Within this class there are morphological variations in form, the size and lobulation of the pituitary gland. Thus, in some species, adenohypophysis is not anatomically divided but it is consisting of a single lobe. In many species of Teleostei adenohypophysis it consists of two lobes, one anterior or distally subdivided into two zones and the other intermediate. The present paper aims to highlight the structure of the pituitary gland and cell types present in cyprinids lobes. The studies were performed on microscopic preparations containing sagittal sections through carp pituitary. The sagittal histological sections made of 5µm reveal the presence of two distinct parts: neurohypophyses of nervous origin, and the adenohypophysis of epithelial origin. Considering the scientific importance and the practice of pituitary hormones in stimulating the maturation of gonads in fish also followed the changes of the pituitary structure, especially of the middle gonadotropic lobe. The histological study of the pituitary gland in the H. molitrix species at different times of the year has given us the possibility to track the seasonal variations of the basophil gonadotropic lobe structure. In the present study demonstrated that during the annual cycle, GTH cells go through different stages of activity that are not synchronous in all cells GTH. We were able to draw conclusions regarding the best period for harvesting pituitary glands used for pituitary injections under artificial reproduction conditions.

Key words*: adenohypophysis, basophil gonadotropic, basophilic cells GTH, lobe, neurohypophysis, pituitary.*

INTRODUCTION

Given the problematic complexity and difficulties of approaching a fish endocrine mechanism, we need a claim on this topic. We are just trying to highlight the structure of the pituitary gland, aspects of hormonal regulation, related to a form of reproduction of a certain cyprinid.

The pituitary gland is located at the base of the brain, which is in close connection with the hypothalamus (Grasse, 1970). It is a small, soft, whitish body whose size and shape vary with species (Shanthanagouda et al., 2018).

The pituitary, considered a master gland of the endocrine system, consists of two lobes, the anterior pituitary or adenohypophysis, the glandular part of the pituitary consisting of cells which secrete pituitary hormones, and the posterior pituitary or neurohypophysis, containing nerve bundles originating from the hypothalamus as well as other parts of the brain (Ooi et al., 2004; Zohar et al., 2010).

Anterior pituitary is derived from the embryonic pouch, Rathke's pouch arising from the roof of the buccal cavity as an outward evagination and the posterior pituitary which originates from the downward evagination from floor to third ventricle (Yadav, 2009; Ball et al., 1969).

The adenohypophysis is subdivided into three parts, histologically, and in the nomenclature of the adenohypophysis fish is presented differently by the authors:

- after Pickfort and Atz, in the year 1957:

Adenohypophysis: pro-adenohypophysis, meso-adenohypophysis, meta-adenohypophysis;
Neurohypophysis;

- after Gorbman, in the year 1965:

Adenohypophysis: rostral pars distalis, pars distalis proximal, pars intermedia;
Neurohypophysis.

Most current authors have adopted Gorbman's nomenclature (Ekici et al., 2013; Evans, 2003).

MATERIALS AND METHODS

For histological research, pituitary samples were taken from adult females of *H. molitrix*, aged 4-5 years, with an average weight of 5-6 kg, from fish farm Cârja 1 - Vaslui county.

Sampling periods coincided with:

- vitellogenetic growth period (November 2015-April 2016);
- maturation period, after the end of vitellogenesis (April-May 2016);
- ovulation period, which took place in June 2016.

The biological material taken for the histological examination was processed by classical methods. Fixation was done in Bouin and Formalin, and after inclusion in paraffin, the pieces were cut at 5 μm by microtome Slee. Sections were stained with hemalaun-eosin (HE).

RESULTS AND DISCUSSIONS

Distribution of cells in the lobes of the adenohypophysis and neurohypophysis

In each of these parts the cells can be identified both by the arrangement and morphology and by the characteristic staining.

The types of cells contained in the adenohypophysis are: chromophobic, which does not participate directly in the secretion process and chromophilic, which in turn are of two types: acidophilic and basophilic.

The anterior lobe (rostral pars distalis) consists of fine-grained acidophilic cells and chromophobic cells mixed with small basophilic cells: prolactin cells, ACTH cells, TSH cells (Figure 1).

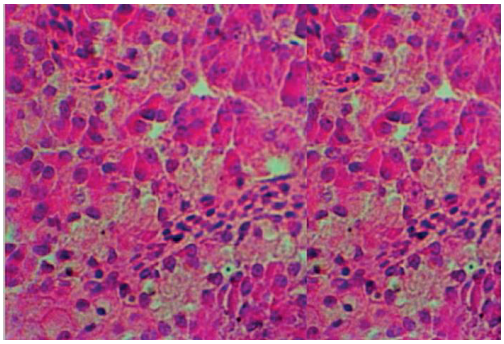

Figure 1. Anterior lobe of the pituitary gland. Acidophilic cells that produce prolactin predominate, HE, 40x

The middle lobe (proximal pars distalis) contains two types of cells, the cells pass successively from the basophilic type to the chromophobic type, then to the acidophilic type: STH cells, GTH cells. STH cells secrete pituitary somatotropic hormone that stimulates growth, and GTH cells secrete gonadotropic hormone that intervenes in the maturation of the gonads (Figure 2).

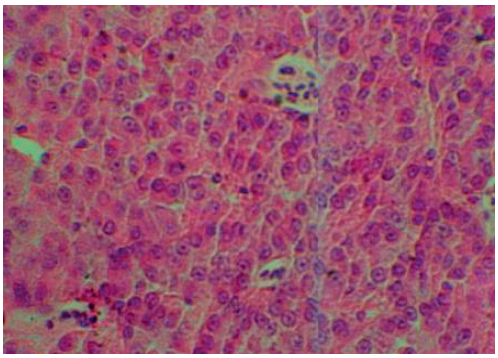

Figure 2. Gonadotropic middle lobe. Basophilic cells (GTH) are well represented. There are also sinusoidal capillaries that take over the secretion product, HE, 20x

The intermediate lobe (pars intermedia) is made up of a single type of cell that secretes a stimulating melanophore hormone, MSH (Takaski, 1982).

In the histological sections made by us, the cellular elements characteristic of the adenohypophysis lobes is highlighted (Figure 3).

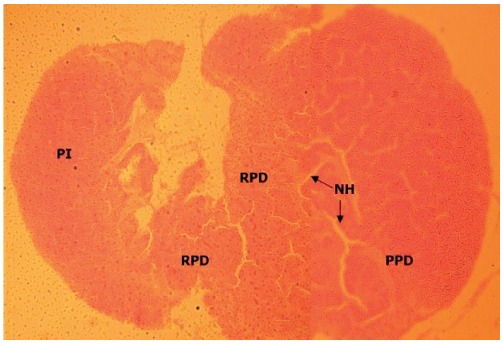

Figure 3. The pituitary gland, sagittal section, shows the adenohypophysis with the three lobes (RPD - rostral pars distalis, PPD - proximal pars distalis and PI - pars intermedia) and the neurohypophysis (NH), HE, 5x (original photo)

The neurohypophysis, like all teleosthenes, branches deeply to the middle of the adenohypophysis in a very particular multiple arborization (Figure 4).

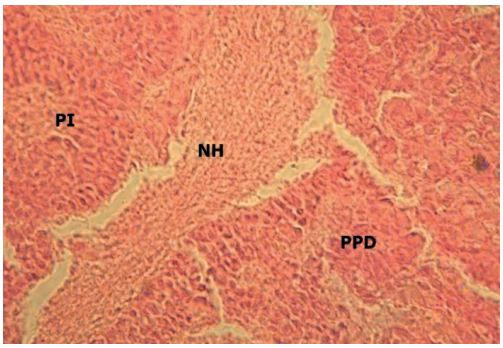

Figure 4. Arborizations of the neurohypophysis inside the adenohypophysis (PPD - proximal pars distalis and PI - pars intermedia), HE, 6x (original photo)

The neurohypophysis has a fibrous consistency being made up of numerous neuroglial fibers, but especially the neurosecretors that originate mostly in the preoptic nucleus of the hypothalamus and glial cells called pituitary.
These cells have an irregular shape and have short branched extensions.
According to some authors, through the hypothalamic - pituitary fibers mentioned above, a functional connection is established between the gonadotropic lobe in the adenohypophysis and the neurohypophysis (Figure 5).

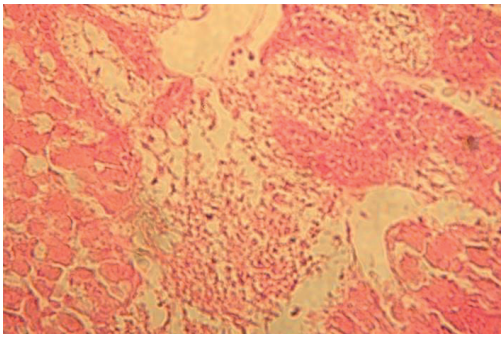

Figure 5. Neurohypophysis, amyelin nerve fibers and pituitary cells are observed, HE, 40x (original photo)

These anatomical connections also demonstrate the existence of a hypothalamic neurosecretory control over pituitary function that occurs in teleost fish as well as in mammals.
The two types of octopeptide hormones produced by the neurohypophysis, arginine vasotocin and isotocin are produced by neurosecretory cells of the preoptic nucleus transported along the axon and released into the capillaries of the neurohypophysis (Figure 6).

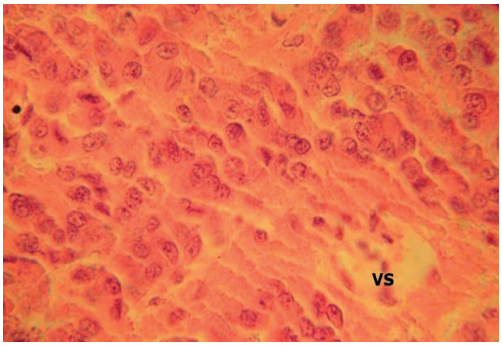

Figure 6. Neurohypophysis and PI - pars intermedia covering blood vessels (VS) for secretion, HE, 90x (original photo)

Seasonal changes in the basophilic pituitary gland in *Hypophthalmichthys molitrix*

The histological study of the pituitary gland in *H. molitrix* species at different times of the year, gave us the possibility to follow the seasonal variations of the gonadotropic lobe structure.

Vitellogenetic period

During this period on the histological sections performed on pituitary glands taken from mature females of *H. molitrix* from November to April in pars distalis proximalis (PPD), the middle lobe or meso-adenohypophysis predominates glycoprotein basophilic gonadotropic cells.
They have an eccentric nucleus, and the cytoplasm is loaded with intensely colored granules, which suggests a hormonal storage (Figures 7, 8, 9 and 10). There is a gradual loading of cells with granules of hormone secretion during this winter, which suggests a high secretory activity of the pituitary gland.
In pars distalis proximalis (meso-adenohypophysis) acidophilic somatotropic cells (STH) can also be observed which are smaller and with fine granulations.

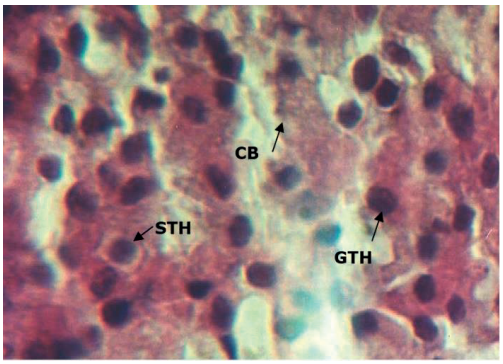

Figure 7. Proximal pars distalis - PPD: among corded GTH cells, rare STH cells and chromophobic (CB) cells, HE, 90x (original photo)

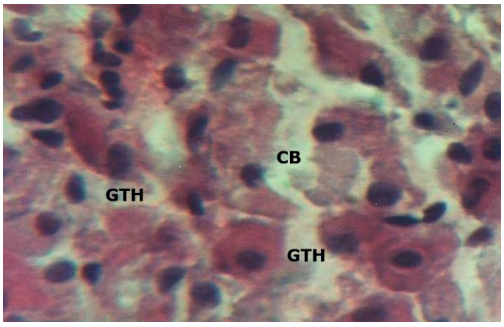

Figure 8. Gonadotropic lobe (PPD): GTH cells elongated with an eccentric or round nucleus, including chromophobic (CB) cells whose cytoplasm does not stain, HE, 90x (original photo)

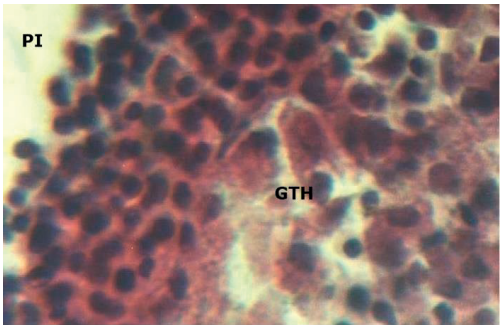

Figure 9. GTH cells at the limit of pars intermedia - PI, HE, 90x (original photo)

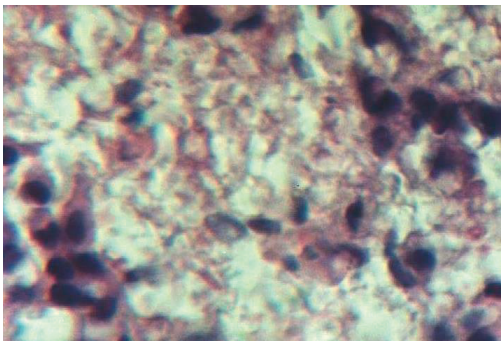

Figure 10. GTH cells ending on nerve fibers in this area their number is smaller, HE, 90x (original photo)

Maturation period

At maturity, the arrangement of highly granulated gonadotropic cells is observed near the blood capillaries where it secretes its secretion product (Figure 11).

The gonadotropic area is distinguished by an increase in the number and size of cells.

After the eggs are laid, the cells lose their polarity to the blood vessels.

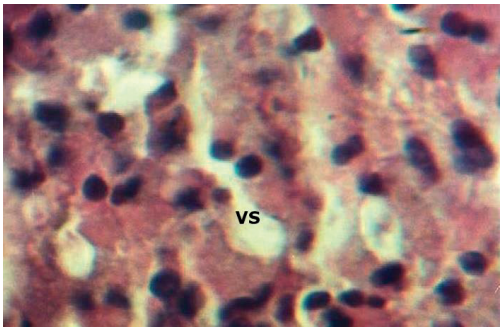

Figure 11. Proximal pars distalis - PPD: GTH cells arranged in cords, facing the blood vessels, HE, 90x (original photo)

Ovulation period

During the spawning season, the gonadotropic area changes considerably. Progressive degranulation and internal vacuolation are observed that occur in most cells. In some areas the cells appear hypertrophied, degranulated and vacuolated almost completely (Figures 12, 13 and 14).

Some gonadotropic cells are located in the dorsal part of the PPD among the branches of the neurohypophysis and are considered to produce GtH-I (FSH), and others located ventrally release the hormone GtH-II (LH) (Evans, 2003).
Gonadotropic cells therefore have a cycle of secretory activity correlated with the evolution of oocytes, having a role in vitellogenesis, maturation and ovulation.

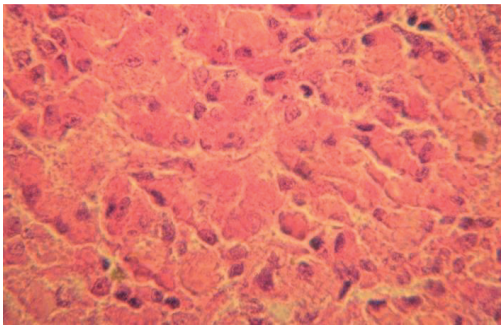

Figure 12. Partially degranulated GTH cells, pycnotic nuclei, HE, 40x (original photo)

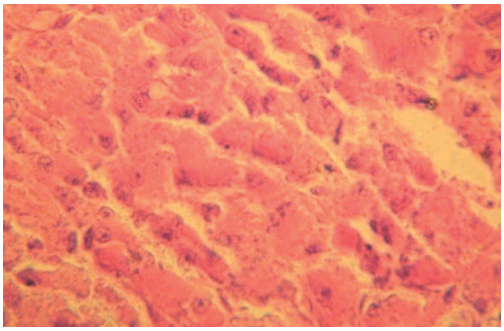

Figure 13. GTH cells, partially degranulated, pycnotic nuclei, HE, ob. 40x (original photo)

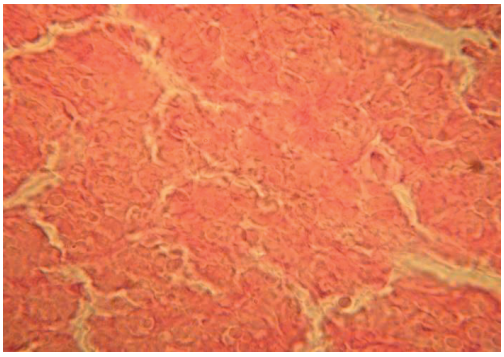

Figure 14. Hypertrophied and vacuolated GTH cells during egg laying, HE, 40x (original photo)

CONCLUSIONS

- From a structural point of view, the adenohypophysis of cyprinids has a glandular structure.
- From a histological point of view, the carp adenohypophysis consists of three distinct lobes: the anterior lobe, the middle lobe and the intermediate lobe, with all the variations of the pituitary gland from one species to another. The technique used highlighted the types of chromophobic cells that do not participate directly in the secretion process and chromophilic cells that participate directly in the hormone secretion process.
- This leads to the important conclusion for the practice of hormone induction in aquaculture, to take pituitary glands from sexually mature females, for pituitary injections, in the season preceding reproduction (December-April). This being the period in which the largest amount of gonadotropic hormones accumulates in the pituitary gland, which it releases in the spring during the maturation period of the oocytes.

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REFERENCES

Ball, J.N., Baker I. Bridget (1969). The Pituitary Gland: Anatomy and Histophysiology, *Fish Physiology*, 2, 1–10.

Ekici, A., Timur, M. (2013). An anatomical and histochemical examination of the pituitary gland of carp (*Cyprinus carpio*). *Turkish Journal of Veterinary and Animal Sciences*, 37, 399–403.

Evans, D. H. (2003). *The physiology of fishes*, second edition, CRC Press LLC.

Grasse, P. P. (1970). *Traite de Zoologie*, vol. XIII, Paris, F: Masson Publishing House.

Yadav, M. (2008). *Fish Endocrinology*. New Delhi, India: Discovery Publishing House, 296.

Ooi, G.T., Tawadros. N., Escalona, R.M. (2004). Pituitary cell lines and their endocrine applications. *Mol. Cell Endocrinol.*, 228, 1–21.

Shanthanagouda, A.H., Sachin O. Khairnar (2018). Breeding and spawning of fishes: Role of endocrine

gland, *International Journal of Fisheries and Aquatic Studies*, 6(4), 472–478.

Takaski, H. (1982). *Fish Histology*, Stuttgart, GE: Gustav Fischer Verlag. Publishing House.

Zohar, Y., Muñoz-Cueto, J.A., Elizur, A., Kah, O. (2010). Neuroendocrinology of reproduction in teleost fish. *Gen. Comp. Endocrinol.,* 165, 438–455.