Cercetări Agronomice în Moldova Anul XXXX, Vol. 4 (132) / 2007

THE NUCLEI OF THE HYPOTHALAMUS IN COW

C. COTEA*, O.Z. OPREAN, P. BOIŞTEANU, Carmen SOLCAN, I. COTEA

University of Agricultural Sciences and Veterinary Medicine of Iași

Received February 12, 2007

ABSTRACT - As a research material we have used the hypothalamo-hypophyseal complexes of 21 cows in the phases of their sexual cycle. From the 21 samples, we have obtained only ten hypothalamico-hypophyseal complexes; in 11 females, the hypothalamus has been obtained separately from the hypophysis, because of the difficulty in getting the complex of the whole. The samples have been fixed in Orth, Carnoy and Helly, led up to paraffin and serially sectioned at 5 µm. We have finally obtained 550 serial sections, stained through the methods Novelli, PAS, Fontana, Steedman-Mowry, Mikami, Bielschowsky, Bodian, Holmes and Nissl. In the cow hypothalamus, we have found the following groups of neurons: anterior (nucleus supraopticus – NSO; nucleus paraventricularis – NPV), middle (nucleus infundibularis), lateral (nucleus hypothalamicus dorsomedialis; nucleus hypotalamicus ventromedialis) and posterior (nucleus periventricularis caudalis; nucleus premamilaris; nucleus corporis mamilaris). The name of the groups was given according to their site, compared to the hypothalamo-neurohypophyseal tract. In the walls of the third hypothalamus ventricle in cow the neurons were localized, which elaborated the releasing hormones (RH) for the gonadotrophic hormones. These neurons have structured the nucleus infundibularis of the cow hypothalamus. The neurons were multipolar, with an average size of 10-12 μm, a 5 μm large vesiculous nucleus and nucleolated. In the neuroplasm of the axon hillock, we have found the numerous PAS-positive granules. These granules were positive in Mikami staining, got stained in blue, which demonstrated that they were of polypeptidic nature. FSH-RH and LH-RH are polypeptids and stimulated the secretions of the gonadotrophic hormones in the β -FSH and the y-LH cells from the cow adenohypophysis.

Key Words: cow, hypothalamus, nuclei, releasing hormones (RH)

REZUMAT - Nucleii hipotalamici la vacă. Ca material de cercetat au servit complexele hipotalamo-hipofizare de la 21 vaci în fazele ciclului sexual. De la cele 21 cazuri, s-au obținut numai 10 complexe hipotalamo-hipofizare; la 11 femele, hipotalamusul a fost obținut separat de hipofiză, datorită dificultății recoltării în totalitate a acestui complex. Probele au fost fixate în Orth, Carnoy și Helly, conduse la parafină și secționate seriat la 5 μm. În final, au fost

^{*} E-mail: cvcotea@univagro-iasi.ro

obținute 550 secțiuni seriate, colorate prin metodele Novelli, PAS, Fontana, Steedman-Mowry, Mikami, Bielschowsky, Bodian, Holmes și Nissl. În hipotalamusul de vacă au fost evidențiate următoarele grupuri de neuroni: anterior (nucleus supraopticus — NSO; nucleus paraventricularis — NPV), mijlociu (nucleus infundibularis), lateral (nucleus hypothalamicus dorsomedialis; nucleus hypotalamicus ventromedialis) și posterior (nucleus periventricularis caudalis; nucleus premamilaris; nucleus corporis mamilaris). Denumirea grupurilor a fost dată în funcție de poziția lor față de fasciculul hipotalamo-neurohipofizar. În pereții ventriculului al III-lea al hipotalamusului la vacă sunt localizați neuronii care elaborează hormonii de eliberare (RH) pentru hormonii gonadotropi. Acești neuroni structurează nucleus infundibularis al hipotalamusului de vacă. Neuronii sunt multipolari cu dimensiunea medie de $10-12~\mu$ m, cu nucleul veziculos de $5~\mu$ m și nucleolat. În neuroplasma conului de emergență axonală au fost evidențiate numeroase granule PAS-pozitive. Aceste granule sunt pozitive în colorația Mikami, fiind cu nuanța de albastru, ceea ce demonstrează că ele sunt de natură polipeptidică. FSH-RH și LH-RH sunt hormoni polipeptidici și stimulează secreția de hormoni gonadotropi în celulele β -FSH și γ -LH din adenohipofiză la vacă.

Cuvinte cheie: hipotalam, vacă, nuclei, hormoni de eliberare (RH)

INTRODUCTION

Hypothalamus was studied in sow and ewe (Cotea, Arseni, 1994; Cotea, Arseni, 1999; Cotea et al., 2002; Cotea, 2003; Runceanu, Cotea, 2001), pointing out the polypeptide secretory neurons FSH-RH and LH-RH. The groups of neurons from the hypothalamus of these species have different positions towards the hypothalamo-neurohypophyseal tract, the following nuclei being noticed: nucleus supraopticus — NSO; nucleus paraventricularis — NPV, nucleus infundibularis, nucleus hypothalamicus dorsomedialis, nucleus hypotalamicus ventromedialis, nucleus periventricularis caudalis, nucleus premamilaris and nucleus corporis mamilaris (Cotea, Arseni, 1994; Cotea, Arseni, 1999; Cotea et al., 2002). For the nuclei from cow hypothalamus, we have extrapolated the research from studied species (swine and sheep). For completing the studies in this species, too, by mentioning the groups of neurons involved in cow reproduction, we carried out this scientific paper.

MATERIALS AND METHODS

As a research material, we have used the hypothalamus and hypophysis from 21 cows in the phase of their sexual cycle, sampled immediately after their slaughtering. The samples have been fixed in Orth, Carnoy and Helly, led up to paraffin and serially sectioned at 5 μ m. The hypothalamo-hypophyseal complex was obtained only in 11 females; in the other females, the hypothalamus has been obtained separately from the hypophysis, because of the difficulty in getting the complex on the whole. From the 11 samples obtained, we have carried out for each case, 240 serial sections, for observing the hypothalamic neurons, according to the hypothalamo-hypophyseal axis and to the third

ventricle and its infundibulum. The used staining was Novelli, PAS, Fontana, Steedman-Mowry, Mikami, Bielschowsky, Bodian, Holmes and Nissl.

RESULTS AND DISCUSSION

In the cow hypothalamus, more neuron regions and areas were delimited. These regions, known as hypothalamic nuclei (Cotea, Arseni, 1994; Cotea, Arseni, 1999; Cotea et al., 2002; Cotea, 2003) and structured from secretory neurons, have a great importance in the endocrine functional system. In the hypothalamus, we have found the following groups of neurons: anterior, middle and posterior ones. The name of the neuron groups was given according to the position of each, compared to the hypothalamo-hypophyseal tract, to the third ventricle and its infundibulum (*Plate 1*).

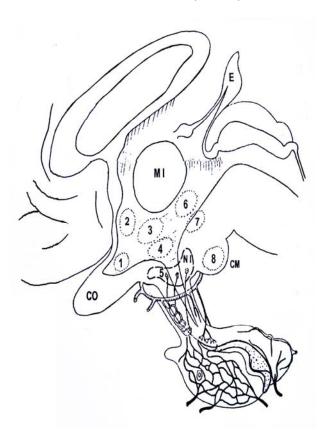


Plate 1 -The hypothalamohypophyseal and epythalamo- epiphyseal system in cow: Anterior group

- 1. Nucleus supraopticus:
- 2. Nucleus paraventricularis);

Lateral group

- 3. Nucleus hypotalamicus dorsomedialis:
- 4. Nucleus hypotalamicus ventromedialis);

Middle group

5. Nucleus infundibularis;

Posterior group

- 6. Nucleus periventricularis caudalis:
- 7. Nucleus premamilaris;
- 8. Nucleus corporis mamilaris
- E epiphysis;
- MI intermediary mass;
- CO optic chiasma;
- CM– mammilar body
- *3, 4, 5, 6, 7 form the
- hypophysothrope region of cow hypothalamus, with multipolar, RH and IH secretory neurons

In the anterior group, we have found *nucleus supraopticus* – NSO and *nucleus paraventricularis* – NPV (*Figures 1, 2, 3, 4, 5 and 6*). NSO is found over the optic chiasma, while NPV is in the anterior third ventricle.

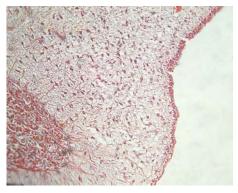


Fig. 1 - Hypothalamo-neurohypophyseal complex in cow. Col. Novelli; x 100



Fig. 2 – Infundibulum of the third ventricle in cow. Technical silver impregnation Holmes; x 80



Fig. 3 - Nucleus supraopticus (NSO) from cow hypothalamus. Technical silver impregnation Holmes; x 200

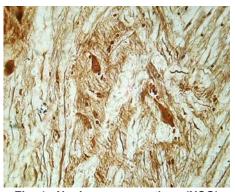


Fig. 4 - Nucleus supraopticus (NSO) from cow hypothalamus. Technical silver impregnation Holmes; x 400



Fig. 5 - Nucleus paraventricularis (NPV) from cow hypothalamus. Technical silver impregnation Holmes; x 200

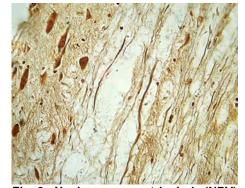


Fig. 6 - Nucleus paraventricularis (NPV) from cow hypothalamus. Technical silver impregnation Holmes; x 200

The neurons from NSO and NPV were of great size (24-31 µm), multipolar, with vesiculous nucleus, eccentrically located and nucleolate. The axons of these neurons could be seen in the microscopic field, at distances of hundreds of micrometers. Across these axons, there were ranges of different size PAS- positive granules. These granules were pointed out in Herring bodies of the neurohypophysis. In the interstitial substance of the posterior lobe, we have noticed across the nervous fibres, round or unregulated ovalary formations, of variable size, which are called the Herring bodies. These are the stocking form of the secretion of hypothalamic neurons in the posterior lobe, because, after sectioning the hypothalamo-hypophyseal tract (the totality of axons from supraopticus and paraventricularis nuclei of the hypothalamus), the Herring substance was accumulated at the proximal extremity of the sectioned neuron (Runceanu, Cotea, 2001). The nervous fibres were mostly myellinic and represented the axons of secretory neurons from the nuclei of the hypothalamus: supraopticus - NSO and paraventricularis nuclei- NPV. These fibres are disposed parallel in the hypophyseal tract, have an irregular disposition in the posterior lobe and they end by dilations under the shape of knob around the sanguine capillary and of pituicytes. The neurons from *nucleus supraopticus* have produced by secretion the ADH- Antidiuretic Hormone or vasopressin with antidiuretic action, by water resorption at the level of distal contort and collector tubes, thus diminishing diuresis. In exchange, the nucleus paraventricularis has produced by secretion OXT-Ocitocin, which stimulated the uterine contractions for the beginning of travail during parturition and the contraction of myoepithelial cells of galactophore channels, thus determining the milk secretion. The secretory neurons of the two nuclei from hypothalamus have a rich cytoplasm in Nissl bodies, a great nucleolate nucleus, short dendrites and a unique extension – axon. All the axons of these neurons from the two nuclei constitute the hypothalamo-neuro-hypophyseal tract. Neurons have produced the secretion of the two hormones bound by a specific protein (neurophysine) and were transported across the axons and stored at the posterior lobe of hypophysis, in the Herring bodies. The neurosecretion also appeared as granular thin material, agglomerated around the capillaries and terminal nervous fibres. This mode of secretion along the nervous fibres is called *neurocrvne*. Morphologically, these were pointed out by the secretion granulations in the perikaryon of secretory neurons, across nervous fibres (the axons of these neurons), which cross the neurons, through the neurohypophyseal tract, in the posterior lobe of hypophysis and at the nerve terminations around capillaries. The special staining has also pointed out proteoglycans and L-ascorbic acid in these Herring bodies. The hormone release from Herring bodies was done by pituicytes, which separated them from neurophysine and sent them to blood.

The lateral groups are found in relation with medio-sagital axis of the hypothalamus, one side and another of the third ventricle. Within the structure of

a lateral group, there are two nuclei: **dorso-medial hypothalamic nucleus** (*Figures 7, 8*) and **ventro-medial hypothalamic nucleus.** These nuclei are different only by their position, while as concerns their structure, they have neurons with similar morphology. The neurons have the size of 14-15 μ m, are multipolar and contain PAS-positive granules in the neuroplasm.

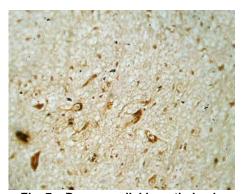
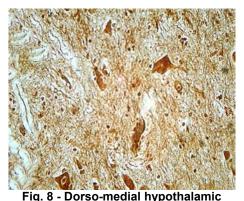


Fig. 7 – Dorso-medial hypothalamic nucleus from cow hypothalamus.
Technical silver impregnation Holmes; x 200



nucleus from cow hypothalamus.
Technical silver impregnation Holmes; x 200

The middle group is found around the origin of infundibulum in the third ventricle (*nucleus infundibularis*) (*Figures 9, 10, 11, 12*), which is structured from multipolar neurons, with the size of $10-12~\mu m$, and many granules in the cytoplasm.

The posterior group includes the neurons located posterior to the third ventricle. Some neurons having the size of 10 μ m are disposed next to the ependimal epithelium of the third ventricle, structuring the caudal periventricular nucleus, while other neurons are located anterior to mammilar body, with the size of 12 μ m, forming the premammilar nucleus (Figures 13, 14). In the mammilar body, we found out multipolar neurons with the size of 16-18 μ m, structuring the mammilar nucleus (Figures 15, 16).

The interrelation systems between hypothalamus and hypophysis are hypothalamo-adenohypophyseal and hypothalamo-neurohypophyseal systems.

The hypothalamo-neurohypophyseal system is represented by nuclei from the anterior hypothalamic zone - *supraopticus* and *paraventricularis nuclei*. The axons form the hypothalamo-neurohypophyseal fascicle. The *supraopticus* and *paraventricularis nuclei* produce the secretion of **vasopressin (ADH)** and **ocitocine (OXT).** These two hormones are bound to a specific protein – **neurophisine** – being transported across the axons in neurohypophysis.



Fig. 9 - Nucleus infundibulum from cow hypothalamus. Technical silver impregnation Holmes; x 80

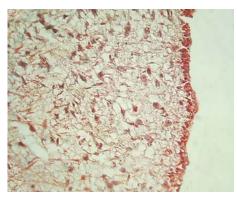


Fig. 10 - Nucleus infundibulum from cow hypothalamus. Holmes; x 200

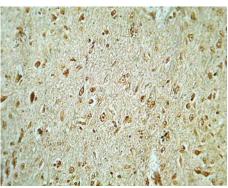


Fig. 11- Nucleus infundibulum from cow hypothalamus. Technical silver impregnation Holmes; x 200

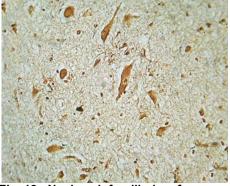


Fig. 12 - Nucleus infundibulum from cow hypothalamus. Technical silver impregnation Holmes; x 400

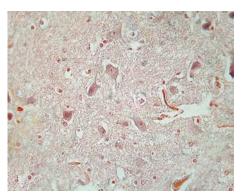


Fig. 13 – Premammilary nucleus from cow hypothalamus. Technical silver impregnation Holmes; x 200

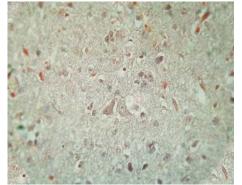


Fig. 14 - Premammilary nucleus from cow hypothalamus. Technical silver impregnation Holmes; x 200

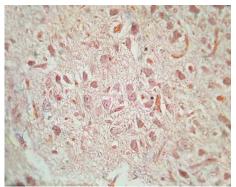


Fig. 15 - Mammilary nucleus from cow hypothalamus. Technical silver impregnation Holmes; x 200

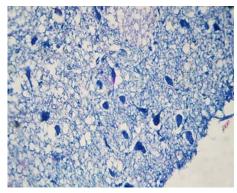


Fig. 16 - Mammilary nucleus from cow hypothalamus Col. Nissl; x 200

The hypothalamo - adenohypophyseal system was characterized by the presence of secretory neurons from the hypophysothrope zone of the hypothalamus, which synthesized the hypophysothrope releasing-hormones – RH and inhibiting hormones – IH of adenohypophyseal secretions.

In the neurons of dorso-medial, ventro-medial, infundibulum, caudal and premammilar periventricular, PAS-positive granules were found, showing a polypeptidic nature-secretion in these neurons. They structured the hypophysothrope area within the cow hypothalamus, because they demonstrated that RH and IH are polypeptide from chemical point of view. The axons of these neurons were shown around the sanguine capillaries from the walls of the infundibulum of the third ventricle, where they released the specific secretions for stimulating or inhibiting the secretory cells from adenohypophysis.

CONCLUSIONS

In cow hypothalamus, the following groups of neurons are pointed out: anterior, lateral, middle and posterior ones.

The anterior group is made of *supraopticus* – NSO and *paraventricularis*– NPV *nuclei*, with the size of perikaryon of 24 - $31~\mu m$.

The lateral groups are structured from dorso-medial and ventro-medial hypothalamic nuclei, with neurons which perikaryon is of $14-15~\mu m$.

The middle group (*nucleus infundibularis*) is structured from multipolar neurons, having the size of 10-12 μ m, located around the infundibulum of the third ventricle, and is secretory of FSH-RH and LH-RH.

The posterior group includes the $10-12~\mu m$ sized-neurons, located posterior to the third ventricle and systematized in caudal periventricular and premammilar nuclei.

REFERENCES

- **Cotea C., Arseni O.V., 1994** *Hypothalamus in swine.* Scientific works, University of Agricultural Sciences and Veterinary Medicine Iasi, vol. 37, pp. 25 27
- Cotea C., Solcan Carmen, Arseni O.V., 1997 Follicular structures in the anterior lobe of hypophysis in sows from Landrace breed. Scientific works, University of Agricultural Sciences and Veterinary Medicine Iaşi, vol. 40, pp. 20 22
- Cotea C., Arseni O.V., 1998 Folliculogenesis in ovary of swine. Scientific works, University of Agricultural Sciences and Veterinary Medicine Iaşi, vol. 41, pp 13 23
- Cotea C., Arseni O.V., Solcan Carmen, 1998 Ovarian tecal gland in swine. Scientific works, University of Agricultural Sciences and Veterinary Medicine Iaşi, vol. 41, pp 6 12
- Cotea C., Arseni O.V., 1999 Hypophysis in swine. Scientific works, University of Agricultural Sciences and Veterinary Medicine Iaşi, vol. 1 (42), p. 24 31
- Cotea C., 2001 Cell biology, general embryology, general histology. Tehnopress, laşi
- Cotea C., Runceanu L., Solcan Carmen, Cotea I., 2002 Neurosecretion of hypothalamic nuclei in sheep. Scientific works, University of Agricultural Sciences and Veterinary Medicine Iaşi, vol. 45 (4), pp. 16 22
- Cotea C., 2003 Special histology. Tehnopress, Iaşi
- Runceanu L., Cotea C., 2001 Reproduction, veterinary obstetrics and gynecology.

 University of Agricultural Sciences and Veterinary Medicine, "Ion Ionescu de la Brad" Publishing House, Iaşi