

## Protein Changes in Neurons and Glia in Same Centers of Hypothalamus at Food Motivation

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### Abstract

In this study, the morphological alteration of proteins in neurons and glia at some centers of hypothalamus during the period of food motivation has been analyzed. There is a correlation between morphological changes occurred in the neurons of lateral and ventromedial centers of hypothalamus and their functional properties. There is also a close correlation between physicochemical methods occurred in protein molecules and the function of lateral and ventromedial centers. In our study, we have additionally determined a short-termed correlation between protein molecules and functional system of food.

This shows us that molecular foundation of food motivation is based upon the plasticity of proteins.

**Key words:** Hypothalamus, Protein, Morphological alteration, Functional system of food, Nöro-glia

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### INTRODUCTION

In sophisticated neurophysiologies it is of greater significance to analyze the structures and functions of neurons as well as the mutual relationships of glia cells [1]. Researches have recently shown that glia cells play a prominent role in terms of the morphology and function of the brain. These cells also take part in the tasks, defense and transformation of some substances (especially energy and water-ion transformation) [2,3]. When neurons functions proliferate in number, the amount of glia cells proliferates as well. At the same time, energy changes accelerates. In such a mechanism glia cells are in a prominent activity [4,5]. Galambus [6] and Paez et al., [7] have shown that glia cells accompany in the integration functions of the brain. Cicera and Provine [8], Hyden and Ewen [9] have pointed out that 100-S, a protein responsible for intelligence is synthesized in the cytoplasm and nucleus of glia cells. In addition to such synthesis, glycoproteins such as 10-B, GFA and Alfa-2 are also synthesized in these cells [10,11]. Hyden and Lange [10] and Radic et al., by microchemical methods [12] pointed out that satellite glia form the functional metabolic system together with neurons, respond the requirements of neurons towards biologically active substances and creat some biochemical processes.

This study has been conducted in order to analyze the condition of the proteins in accordance with the data reviewed in this literature in the centers of hypothalamus in the various periods of the state of hunger.

### MATERIALS AND METHODS

We have conducted our study using 50 male rats of same age and weight. Ten of them constituted the control group; these rats were given abundantly of food and water. The rest weres divided into four groups, each consisting of ten rats.

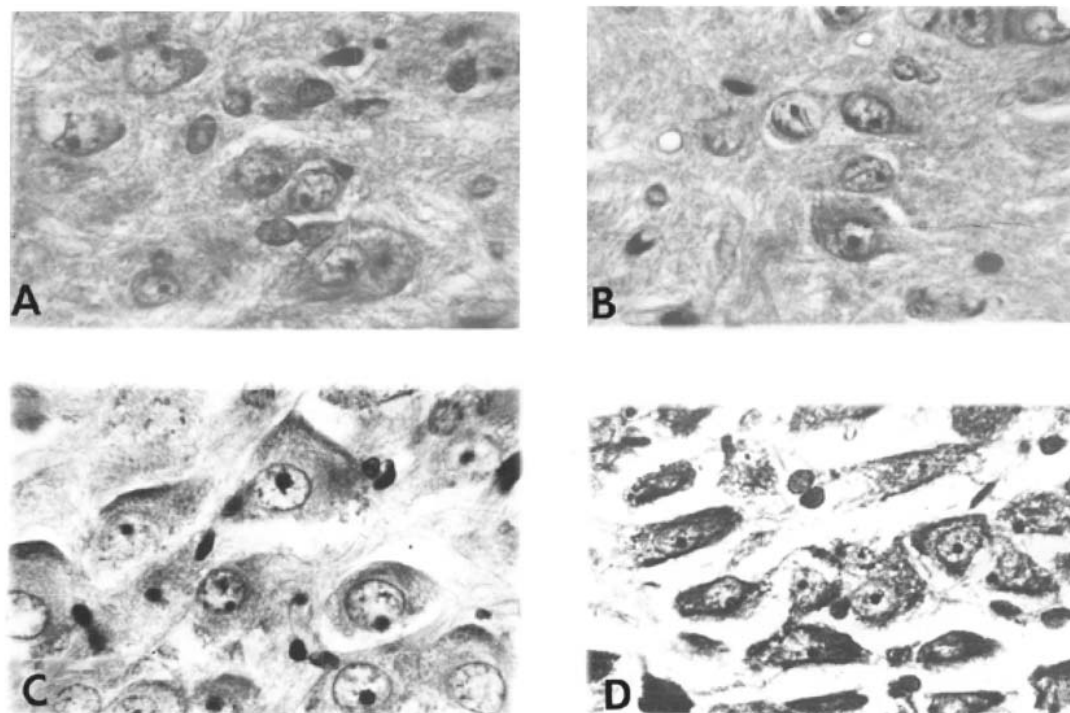
These rats were kept without food for 1-2-3 and 5 days except for water given to them; after this process, the animals were decapitated in the final hours of the day. The brains of these animals were entirely fixed by Carnoy's fixative. After paraffin blocks were prepared and pieces ranging ten microns of scale were taken, these pieces were stained with the modification of Geyer method [13] which is another version of Gerstein and Ball method [14]. This method provides us some means for dying proteins of neural and glia cells in a single pieces at the same time and for discerning of the glia cells on a basis of distinctly dyed cells.

In this study, the Para-ventricular (PV), Supraoptic (SO), Lateral center (LC), Ventromedial (VM), Dorsomedial (DM) and Mamillarmedial (MM) centers of hypothalamus have become the chief focus of our concern.

### RESULTS

While carrying out all these processes we found that neural proteins in the centers of the hypothalamus of the normal animals and the other proteins in the cytoplasm and nucleus of their glia cells had been colored in blue. By the aid of the method mentioned above, the cytoplasm and nuclei of oligodendroglia, a different type of glia cells, were observed to have been dyed in dark red color. The nuclei of astroglia were observed to have been colored in light red. The nucleus chromatin was observed to have been colored slightly in red, microglia were in dark blue, and neuropilles in a reddish yellowish color in all preparations we designed. In the pieces taken from the control group, neural and glia cells in almost all of the centers of hypothalamus were found to have been colored at the same rate. Hyper and hypocrome cells were also observed in all of the phrase (Fig.1).

After a day of deprivation from food, the activity in the reactions in the neural and glia cells was usually observed to



**Figure 1.** Identifying the proteins in neural and glia cells of the hypothalamus of the normal animals.

- a) The periphery of lateral center b) Lateral preoptic center  
c) Supraoptic center d) Paraventricular center X630.

have shown no alteration. Darkly-colored cells were found in ventromedial and dorsomedial centers. But no distinctive changes were observed in glia cells whereas some changes were found in the neurons in lateral centers. A scattering was characterized in the chromatin of the nuclei of these neurons. The nuclei of neurons as well as some protein vacuums characterized in lateral and ventromedial centers were found to be localized in an eccentric way whereas no drastic change was observed in the neurons and the glia cells in supraoptic and paraventricular centers.

On the second day of deprivation, some morph-functional changes were observed in the neurons in the medial centers of hypothalamus (VM and LM). Proteins have been localized at the edges of cytoplasm and have been scattered in different regions of membrane whereas these proteins showed a heterogenic distribution. We have also observed that the membranes of the cells in the lateral, ventromedial, mamillarlateral and mamillarmedial centers of hypothalamus have vanished in an intermittent way and that some presence of hypocrom cells became predominant. But we observed no change in microglia cells, the chromotophile substance in with cells of great magnitudes (SO and PV) shrank to the periphery of the midst of cell. As far the structural elements of the glia cells no change has been recorded in them (Fig. 2).

On the third day of deprivation, further morphological changes were recorded in the neurons of lateral and ventromedial centers of hypothalamus. The basophile substance in the nuclei of the cells was observed to have been coloured much less. Vacuoles of different magnitudes were seen in the cytoplasm. Nuclei of most of the neurons were found to have been localized

in an eccentric way, whereas the nucleoli were found to have been dyed much less.

Proteins were localized at the edges of neurons. The amount of proteins in the bottoms of the neurons in all of the centers of lateral of hypothalamus (anterior, medial, posterior) was observed to have been decreased. The amount of chromatin in the nuclei of glia cells especially in oligodendrocytes was found to have increased. Whereas hyperchromatizm continued to exist at minor neuronal centers of hypothalamus (VM, MM) it become activated in oligodendrocytes to the last degree. All of the neurons in supraoptic and paraventricular centers showed the same degree of reaction. Though the amount of protein decreased in a small number of neurons, this amount did not change in a great number of them. A sort of change resembling a bulging form in the cytoplasm of some of astrocyts at mamillar medial and mamillar lateral (MM, ML) center of hypothalamus was observed. Within one-three days of deprivation from hunger no change was observed in microglia (Fig.3).

In the fifth day of deprivation the amount of protein was observed to have decreased in all of the neurons of the lateral center of hypothalamus. Some hydropic changes occurred in neurons of middle and large scale. In addition to all this changes, the speed of protein reactions in glia cells as well as the amount of protein in the neurons of ventromedial centers also increased. Whereas the amount of protein at supraoptic (SO) and paraventricular (PV) centers of hypothalamus with large neurons was observed to have decreased the amount of nucleoproteins and proteins in the nucleus and nucleolus increased.

## DISCUSSION

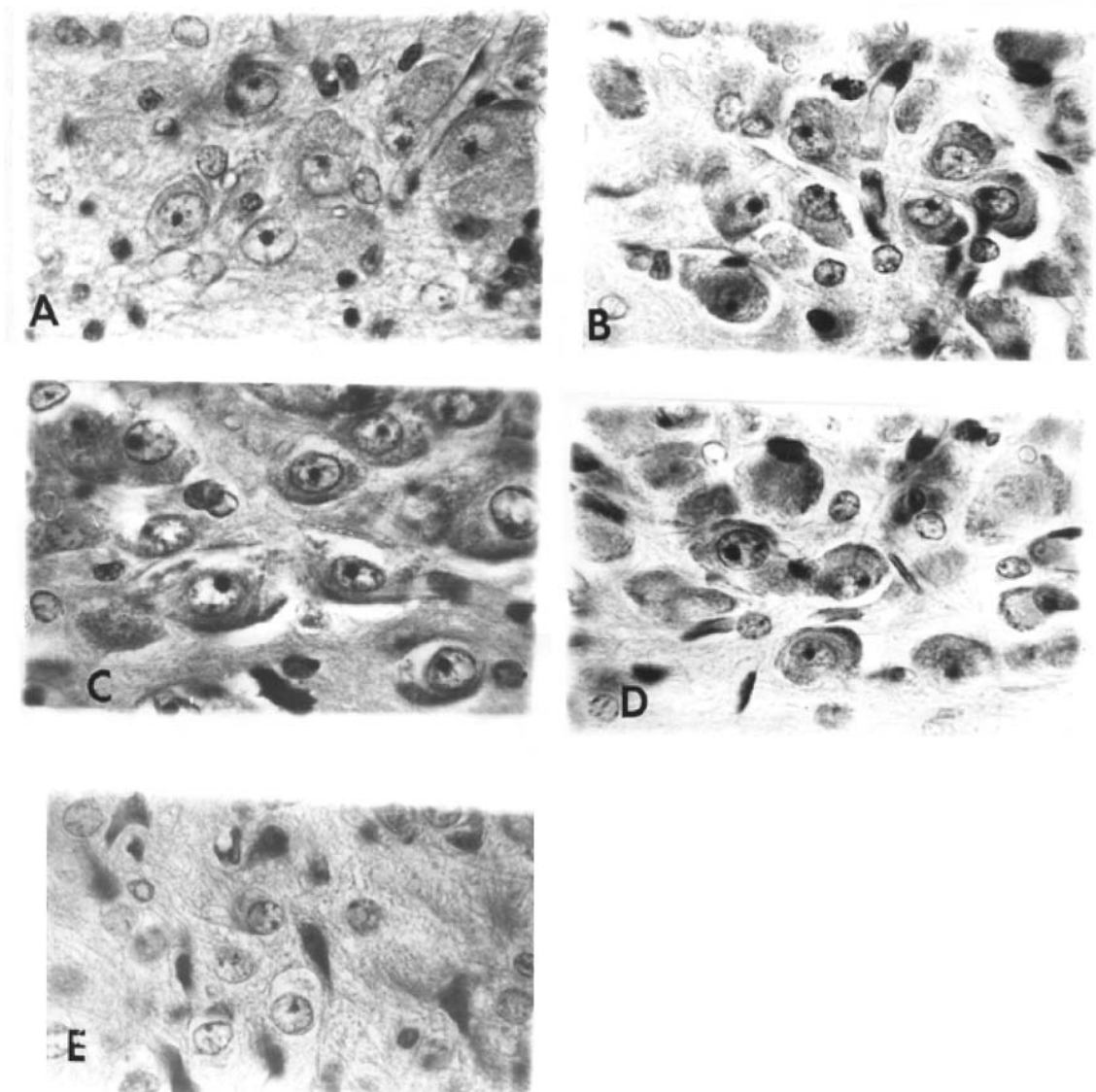
Within the course of our experiment some morphological changes in the aspects of proteins in the nuclei and the cytoplasm of neurons in lateral hypothalamus in the early stage of deprivation were observed. This morphological change suggest that lateral hypothalamus functions as a center of hunger and neurons play an active roll in the functioning of the structure involved in food motivation. In this case, lateral center behaves as a pacemaker, that is to say, impulses passing through pacemaker spread across the hemispheres of cerebral cortex by the aid of reticular formation of the brain.

Anohin et al. [15] and Harel et al. [16] reported that this system constituted the basis of the neuropsychological mechanism of hunger satiety and appetite. In another study conducted by the same researchers, It was argued that the mice

reacted appropriately benefiting from their living experiences and genetic intelligence wrought by afferent synthesis in the circumstances of dominant food motivation [11,17].

Such a food motivation activates same distinct regions of genetic code of neuron cells. And this is believed to accelerate the synthesis and secretion. Thus neuropeptides stimulate the genesis of food motivation as an important factor.

In the first day of deprivation from food an increase was observed in the amount of oligodendroglia and hyperchrom cells in the neurons of ventromedial center. According to the studies conducted about these phenomena such a research prevented the animal from the motives of hunger, accelerating the peptide synthesis involved in satiety of food like a center functioning for this mechanism [18,19]. The increase in the number of oligoastroglial cells in this region is a criterion that neurons are in a functional activity [4,20].



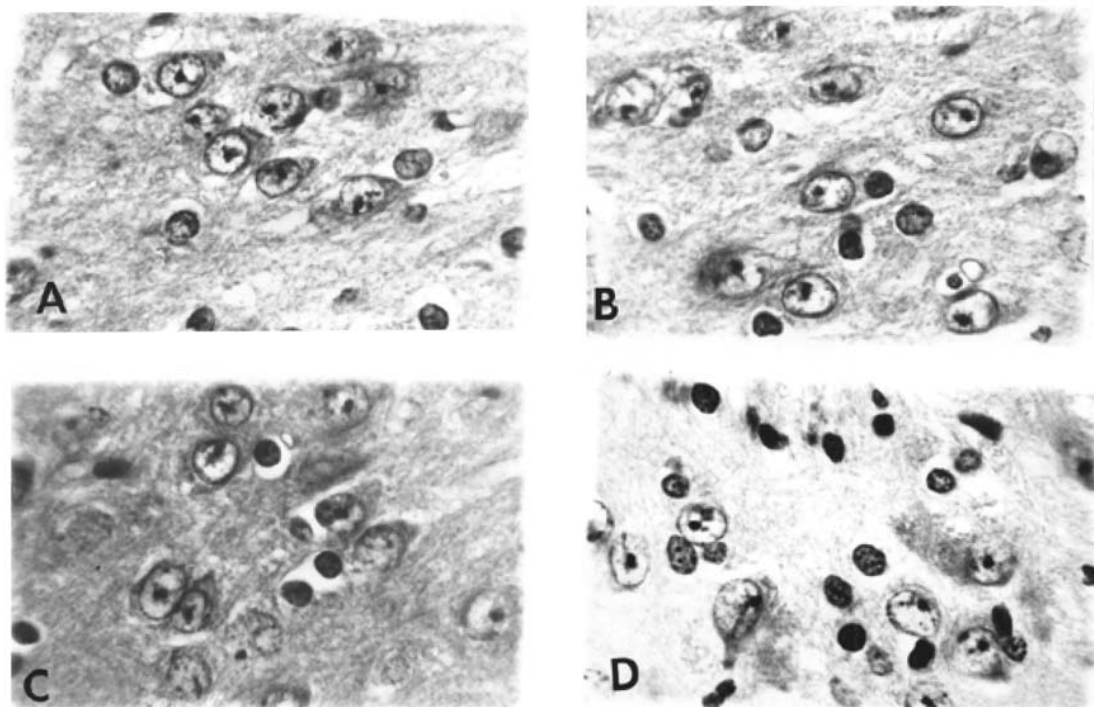
**Figure 2.** The determination of the amounts of proteins in the neural and glia cells of hypothalamus after two days of deprivation from food: a) Lateral outer center b) Paraventricular center c) Supraoptic center d) Mamillar lateral center e) Mamillar medial center X630.

As it became evident in the course of our study, the food's functional system is under the control of afferent synthesis. Dominant food motivation system shapes the behavior of the animal towards the food. The impulse aroused from hunger activates a distinct region of the genum, spreading up to the genetic memory in molecular levels. This impulse also accelerates the synthesis of specific peptides involved in mRNA and satiety of food [21,22].

Ibrahimova reported that the amount of proteins increased in cytoplasm and nuclei of pyramidal neurons of the third layer of the cerebral sensomotor cortex in the first day of hunger in mice. The same researcher also noted that this type of neurons in the brain received impulses coming from the afferent system, providing the associative interaction and forming the afferent synthesis of the functional system involved in feeding [23].

Findings obtained in this study are coherent with those reported in the previous studies based on such phenomena. In the second day of hunger some concrete morphological changes were observed in the neurons of the VM and LM centers of the hypothalamus. Proteins were observed to have localized on the sides of cytoplasm and membrane; they were scattered in the nuclei of the neurons in these centers equally. On the second they such statuses of these proteins show that food motivation is at its peak and the sensitivity of neural receptors at the centers responsible for hunger and satiety increases. Genetic code accelerates the synthesis of specific peptides (gastrin and holocytokinin), providing appropriate behavior designed for a purpose.

The shrinkage of chromotphyle substance to the periphery of cytoplasm of the neurons of SO and PV centers during this



**Figure 3.** Determining proteins in neural and glial cells of hypothalamus after three days of deprivation of hunger. a) Front lateral center b) Medial lateral center c) Outer lateral center X630.

stage of hunger and the increase in the number of glia cells also show that the receptors of all these neurons are in a higher activity [24,25]. It has been reported that all of these neurons respond to hunger with a higher neurosecretional function [26,27]. Such data confirm the results obtained in this study.

In the third day of hunger much more morphological changes were observed in the neurons of lateral VM centers of hypothalamus. The nuclei of neurons were generally observed to have eccentrically localized and proteins were poorly stained.

These findings are indicator of the fact that food motivation is at its peak during the third day of hunger. They also show that proteins in neurons are passed through pericaryon to axodentritic synapses by the mechanism of axoplasmatic current and utilized for establishing short interactions between neurons. Askerov and Alekperova [27] reported that during the

establishment of such mechanism, proteins, which are soluble in water, decreased whereas structural proteins increased in cytoplasm and nucleus.

In the fifth day of deprivation from food the amount of protein in most of the neurons at the lateral center of hypothalamus decreased. But it (protein amount) increased in the neurons at ventromedial, supraoptic and paraventricular centers. At the same time the amount of nucleoproteins in the nuclei of the cells at supraoptic and paraventricular centers decrease. The speed of protein synthesis in glia cells accelerated.

Such findings showed that lateral center is involved in the mechanism related with hunger and thus is stimulated in the prolonged states of hunger. It was seen that the rate of catabolic reaction in these neurons is much higher than anabolic reaction.

The increase of protein in the neurons of ventromedial centers induces an inhibitory phenomenon to block the feeling of hunger on the part of the animal. For this reason, there occurs an acceleration of specific protein synthesis in neurons, especially in their nuclei.

In another study conducted by Askerov [28], in the fifth day of deprivation from food, an increase in the activation of acid and peptide hydrolyses in the homogenate cytosol and mitochondria functions of hypothalamus was observed; in that study of rats, the researchers reported a decrease in the amount of proteins. This researcher also reported that the amount of water-soluble-proteins increased in nucleic fractions.

In this study conducted by us, morphologically remarkable changes in the amount of protein in all of the centers of lateral hypothalamus were observed. In the light of such findings we came to the conclusion that lateral hypothalamus functioned as a center operating processes involved in hunger; and specific changes occurred in the amount of the protein in neurons of lateral hypothalamus in the first phases of hunger. Such changes were also observed in the neurons of ventromedial center in the first stage of hunger. In the first stages of hunger we have come to the conclusion that lateral hypothalamus (LH) functioned as a center of hunger and that specific changes occurred in the amount of proteins in its neurons. Such changes were also observed in the neurons of ventromedial center. In such phases, the changes in the protein synthesis the neurons in hunger and satiety centers and specific peptides (gastrin, holecystocinin) are synthesized in the neurons of the centers of hypothalamus responsible for hunger and satiety of food. These peptides are involved in such process as the establishment of relationships necessary for functional system of food. Due to such phenomena, the amount of the proteins, which are soluble in water, is decreased in cytosol fraction whereas the amount of all of the proteins in nucleus is increased.

In the third day of hunger, the amount of structural proteins in nucleus and cytoplasm was observed to have increased. Such findings suggest that proteins play a role in the occurrence of morph-functional changes in neurons are due to the close interactions between the functional process of these cells and the physico-chemical processes in their protein molecules. For this reason some morphological changes occurred in the neuronal and glia cells of lateral center (responsible for hunger) ventromedial center (responsible for satiety of food) in various stages of food motivation. And this proves that proteins played the most important role in the machinery of food functional system in the stage of hunger.

From this finding, It can be inferred that the integration processes carried out in central nervous system are based on the plasticity of proteins in the neurons of this center.

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### REFERENCES

- [1]. Barak D, Kronman C, Ordentlich A, Ariel N, Bromberg A, Marcus D, Lazar A, Velan B, Shafferman A. 1994. *Journal of Biological Chemistry*. 269: 6296-6305.
- [2]. Glebov RN, Berzucko SM. 1973. Obmennie processi v sisteme neyrogliya pri razlicnih fiziologiceskih i patologiceskih sostoyaniyah nervnoy sistemı. *Journal neuropatologii i psihatrii im 7*: 1085-1095
- [3]. Şelikov VN, Dergacev VV, Poletaev AT, Naumova TS. 1975. O vozmojnoy roli nöroglii v deyatelnosti nervnoy sistemı. *Uspehi fiziologiceskih*. 76: 90-100
- [4]. Aleksandrovskaya MM. 1969. Morfoloiceskiye dannıye o vozmojnoy svyazi mejdu reakciyey neyroglii ivoznkoveniyem medlennih biopotensialov v koze golovnoy mazga jivotnih. V kn: Dlitelniye elektriceskiye potansciali nervnoy sistemı. *Mezniezeba, Tbilisi*, 97-113
- [5]. Hongling Z, Javier C. 2002. Sigma Receptors Inhibit High-Voltage-Activated Calcium Channels in Rat Sympathetic and Parasympathetic Neurons *Journal Neurophysiol* 87: 2867-2879.
- [6]. Galambos RA. 1961. Glia-neuroe theory of brain function. *Proceeding of National Academy of Sciences*. 97: 129-136
- [7]. Paez X., Stanley BG. and Leibowitz SF. 1993. Microdialysis analysis of norepinephrine levels in the paraventricular nucleus in association with food intake at dark onset. *Brain Research.*, 606: 167-170
- [8]. Cicero TJ, Provine RR. 1972. The levels of the brain spesific protein S-100 and 14-3-2 in the devoloping cliek spinal cord. *Brain Research*. 44: 294-298
- [9]. Hyden H, Ewen BA. 1966. Glial protein spesific for the nervous. *Proceeding of National Academy of Sciences*. 55: 354-358
- [10]. Hyden H, Lange PW. 1971. A knetic study of the neuron glia relationship. *Journal Cellule Biology*. 13: 233-237
- [11]. Dahl D, Bigmani A. 1973. Glial fibrillary acidic protein from normal human brain. Purification and properties. *Brain Research*. 57: 343-360
- [12]. Radic Z, Duran R, Vellom DC, Li Y, Cervenansky C, Taylor P. 1994. *Journal of biological Chemistry*. 270: 11233-9.
- [13]. Geyer G. 1960. Lur Eiweibfarburg mit amido schworz 10B. *Acta histochem* 10: 286-292
- [14]. Gerstein LM, Ball TV. 1969. Metodica diferen sirovannogo viyavleniya neyronov i glii na parofinovih srezah. V kn: Sovremenniye metodu morfologiceskih issledovaniy mazga. *Nauka, Moscowa*, pp 11-13
- [15]. Anohin PK, Sudakov KV. 1971. Neyrofiziologi ceskaya teoriya goloda, appetida i nasişeniya. *Uspehi fiziologia*. 2: 3-41

- [16]. Harel M, Schalk I, Ehret-Sabatier L, Bouet F, Goeldner M, Hirth C, Axelsen PH, Silman I, Sussman JL. 1993. Proceeding of National Academy of Sciences. 90: 9015-5.
- [17]. Anohin PK. 1968. *Biologiya i neyrofiziologiya uslavnoga refleksa* Medisina Nauka, Moscowa, 543-547.
- [18]. Gibbs J, Smith GP. 1984. *Frontiers in neuroendocrinology*. Raven press. P. 233.
- [19]. Jhanwar-Uniyal M., Awad, IR., Gearhart GM, Finkelstein JA and Leibowitz SF. 1991. Higher -noradrenergic receptors in paraventricular nucleus of obese Zucker rats: Decline after food deprivation. *Pharmacol. Biochemical Behaviour*. 40: 853-859.
- [20]. Menendez JA, Atrens DM and Leibowitz SF. 1992. Metabolic effects of galanin injections into the paraventricular nucleus of the hypothalamus. *Peptides* 13: 323-327.
- [21]. Sudakov SK. 1988. Neyropeptidi v zentralnih mehanizmah pişevogo povedeniya. *Uspëhi sovremyennoy biologii*. 105: 100-116
- [22]. Leibowitz SF. 1992. Neurochemical-neuroendocrine systems in the brain controlling macronutrient intake and metabolism. *Trends in Neurosciences* 15: 491-497.
- [23]. İbrahimova OS. 1998. Strykturniye belki neyronov i glialnih kletok sensomatornoy i limbiceskoy oblastey kari golovnogo mazga pri razlichnih srokah golodaniya. (Avtoreferat dissertasii) Bakü 21.
- [24]. Yarigin NE. 1973. *Patologiceskie u prispobitelniye irmeneniya neyronov*. Medisina, Moscowa, 190-192.
- [25]. Young KC, Cheng-Shu L, David VS. 2002. Taste Responses of Neurons of the Hamster Solitary Nucleus Are Enhanced by Lateral Hypothalamic Stimulation *Journal Neurophysiol* 87: 1981-1992.
- [26]. Askerov FB, Alekperova SA. 1991. Soderjoniye obşego belka aktivnost neytralnih i kislih peptid-gidrolaz v sybkletocnih fraksiyah gipotalamusa pri razlichnih urovnyah pişeyov motivasii. *Ukrainskiy Biohimiceskiy Jurnal* 63: 38-43
- [27]. Polenov AL. 1968. *Gipotalamiceskaya neyrosekresiya*. Nauka, Leningrad, 158
- [28]. Askerov FB. 1991. Morfohimiceskiye zakonomnosti adaptasionno-kompensatornih reaksiy yader gipotalamusa pri izmeneniy pişeyov i pityevoy motivasii (Avtoreferat dissertasi). Kiev. 50-58