

**Euro Pathology 2018: The Role of Acute Phase Proteins in Induction of Tension of Nonspecific Resistance System in Various Clinical Phenotypes of Myasthenia - Kalashnikova JV - Institute of General and Urgent Surgery, Kharkov, Ukraine.**

**Kalashnikova JV**

*Institute of General and Urgent Surgery, Kharkov, Ukraine.*

**Introduction:**

Around the world, autoimmune diseases are on the increase. One of the classic examples of these diseases is myasthenia gravis, a progressive autoimmune disease characterized by pathological muscle weakness and increased fatigue. It should be noted that the cause of myasthenia gravis is a dysfunction of neuromuscular transmission.

Myasthenia gravis can be found in 5 to 10 cases per 100,000 people, according to various authors. The opportunity to study and treat myasthenia gravis is related to the social factor, namely: the onset of the disease occurs at a young productive age - the onset can reach 90% before the age of 40; it is more common in women-3: 1, according to data from the Myasthenia Center in Moscow; women get sick earlier than men; the cases of women are more serious than those of men.

In our previous publications, it has been shown that a pathology such as myasthenia gravis of various clinical phenotypes is characterized by a tension of the body's non-specific resistance system. However, the role of acute phase proteins in the formation of the above tension condition as formations produced by the body in the early stages of the immune response is still unknown, as the functional features of acute phase proteins allow to put them in mediators and inhibitors of inflammation.

Proteins of the acute phase (C-reactive protein, haptoglobin and ceruloplasmin) in the literature are considered as indicators of metabolic systems as blood reagents, i.e. groups of plasma proteins, glycoproteins and of their components, the concentration of which increases considerably and not specifically in reaction to various pathological stimuli.

It is very timely to study the influence of proteins of the acute phase as prerequisites for the phenomenon found of the tension of the non-specific resistance system in various clinical phenotypes of myasthenia gravis in order to further develop direct immunocorrection methods.

The objective of this work: to study the influence of acute phase proteins on the phagocytosis process in patients with various clinical phenotypes of myasthenia gravis.

The hypothesis of this article is: the role of proteins of the acute phase in the formation of the stress condition of the non-specific resistance system of patients with different clinical phenotypes of myasthenia gravis is essential and requires further study.

**Materials and Methods:**

A total of 69 patients with myasthenia gravis and morphological and functional changes in the thymus and thymoma hyperplasia were observed, aged 13 to 70 years. All the patients were examined and divided into 3 groups according to the morphological lesions of the thymus: a group of "myasthenia without morphological lesion of the thymus" represented by patients M-30, a group of "myasthenia with hyperplasia of the thymus" represented by MH-23 patients, a group of "myasthenia gravis with thymoma" represented by MT-16 patients, as well as a group of 30 healthy volunteers who constituted a control group.

**Method of separating neutrophil granulocytes:**

The neutrophilic granulocytes were separated from the leukocyte suspension of blood drawn from the ulnar vein. To study the phagocytosis of neutrophilic granulocytes, we use 2.5 ml of blood. From this amount we get 1.3-1.5 ml of the leukocyte suspension. We allocate 0.2-0.3 ml of neutrophilic granulocytes by the

ficoll-verografin density gradient. We use 0.2 ml of neutrophilic granulocytes to study phagocytosis.

Method of determining phagocytic activity of granulocytic neutrophils:

The phagocytic activity of granulocytic neutrophils was determined in a leukocyte suspension obtained from heparinized blood. For research, equal amounts of leukocyte suspension (0.2 ml) and washed *Saccharomyces cerevisiae* (baker's yeast) suspension (0.2 ml) were mixed. The samples obtained were incubated for 60 minutes at 37 ° C and stirred regularly. After incubation, smears were prepared which were dyed with the Sky Romanov dye. The microscopic examination was carried out under an immersion system. The phagocytic index (PI) was calculated as the percentage of cells that joined phagocytosis of the total number. Phagocytic Number (PN) - the average number of yeast cells inside the cell was calculated by dividing the total number of yeast cells absorbed by the number of cells that went into phagocytosis. The two samples (PI and PN) were calculated on smears made after the 30 minute and 90 minute incubation (PI 30, PI 120 and PN 30, PN 120). The phagocytosis completeness index (CIP) was calculated by dividing PN 30 by PN 120.

Method of studying oxygen-dependent metabolism of neutrophil granulocytes (NBT-test):

The oxygen-dependent metabolism of neutrophilic granulocytes was studied in response to spontaneous and stimulated restoration of blue nitro tetrazolium (NBT test) using the A.N. Mayan Sky method. During the reaction, the NBT is restored to an insoluble dipformazane which is deposited in the cells in the form of dark blue granules which can be detected visually by microscopic examination [7]. At each stroke, 100 neutrophils were counted and the percentage of cells containing dipformazane in granular form or in solid calculating positions was calculated. The mean cytochemical coefficients in spontaneous and stimulated tests (ACKspont, ACKstim) were calculated according to the Astaldi-Verg formula. To do this, the cells are divided into 4 groups according to the number of dipformazane granules: (a) - without activity (no granules) - 0; (b) - with a slightly positive reaction (single granules) -1+; (c) - with a positive response

(granules coated at 50% of the surface of the cytoplasm) - 2 ++; (d) - with a strongly positive reaction (more than 50% of the surface occupied by the cytoplasmic granules) - 3 +++. The calculation was performed as follows: + + + + + = 100%, thus, the percentage of positive cells -  $b + c + d = 100\% - a\%$ .  $ACK = (1 \times B + 2 \times c + 3 \times d) / 100$ .

Conclusions: In patients with myasthenia gravis of all clinical phenotypes there is a tension of the body's non-specific resistance system. Its particularity is the activation of the phagocytosis system with the increase in the phagocytosis index and the spontaneous enzymatic production of phagocytes, as well as the decrease in the ability to stimulate in the NBT test.

In patients with myasthenia gravis when a development of a secondary autoimmune process that begins in the peripheral lymphoid tissue, a cascade of inflammatory reactions is induced which is accompanied by a considerable increase in the concentration of reactive protein C in patients of all study groups, particularly evident in the HD group with thymus hyperplasia.

An insignificant increase in the concentration of the complement fragment C3 is observed, which may testify that the complement system is not sufficiently activated in a conventional or alternative manner, which may also be one of the causes of the development of immunocomplex causes. Myasthenia gravis.

#### References

- Carr AS, Cardwell CR, McCarron PO, McConville J (2010) A systematic review of population based epidemiological studies in Myasthenia Gravis. *BMC Neurol* 10: 46.
- Boyko VV, Klimova EM, Kudrevich AN (2008) Treatment of myasthenia gravis taking into account immuno physiological phenotypes. Kharkiv: Publishing Scheinina EV, Kharkiv, Ukraine.
- <http://www.myasthenia.ru/index.php?id=1>
- Klimova EM, Boyko VV, Drozdova LA, Kordon TI, Kostya UP, et al. (2014) Features of the immuno physiological responses in patients with different clinical types of myasthenia gravis. *Kharkiv surgical school* 3: 46-53.

- Jain S, Gautam V, Naseem S (2011) Acute-phase proteins: As diagnostic tool. *J Pharm BioalliedSci* 3: 118-127.