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Research Article

HEMATOLOGIC PROFILE FOR *CYPRINUS CARPIO***Svetlana D. Chernyavskikh, Do Hyu Kuet, Lyudmila N. Trikola, Lyudmila K. Buslovskaya, Aleksey Y. Kovtunenکو, Yuliya A. Makarova**

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Abstract:

*The hematologic profile of the representatives of the bony fishes inhabiting the Belgorod region - common carp *Cyprinus carpio* is studied. By means a light microscopy method definition of the blood type cells for fishes on the basis of metachromasy of cytoplasm and core, as well as sizes, forms of cells and their cores after fixing and coloring of dabs by Romanovsky method is carried out.*

*It is established that the hemacytrate profile of common carp *Cyprinus carpio* is presented by the erythrocytes prevailing by quantity, platelets and leukocytes. Nongranulocytes include monocytes and lymphocytes, granulocytes - eosinophils, basophiles, neutrophils, as well as other uniform blood cells which are at different development stages: progranulocyte, myelocyte, metamyelocyte, stab cell and segment cell leukocytes. In a leukocyte formula of the *Cyprinus carpio* the most part is occupied by lymphocytes. Among polymorph cell leukocytes neutrophils prevail, quantity of eosinophiles and basophiles at the studied species of bony fishes is rather low.*

*The revealed hematologic profile of the *Cyprinus carpio* can be used for assessment of their functional state, health and extent of adaptation to environment conditions.*

Keywords: *erythrocytes, leukocytes, platelets, common carp, hematologic profile.*

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INTRODUCTION:

For representatives of bony fishes important criterion for evaluation of an organism condition is the hematologic profile. It is known that at the normal state of an organism the quantitative maintenance of the components which are blood particles is regulated strictly and supported in equilibrium state. Due to pathological violations of functions of any bodies and fabrics quite sharp shifts as blood particles are observed [1].

Literature sources concerning a hematologic profile of fishes, in particular granulocytes, are very extensive and contradictory [2-4]. Among materials about morphological features and qualitative structure of final development stages of granulocytes there are judgments that for some species of fish granulocytes are absent.

Proceeding from it, studying the hematologic profile of the bony fish representatives remains urgent so far. Research objective – is to study a hematologic profile of the bony fish representatives of *Cyprinus carpio*.

MATERIAL AND TECHNIQUE:

Researches are executed on cells of peripheral blood of common carp *Cyprinus carpio* (35 individuals). As a research object served the erythrocytes, polymorph cell leukocytes (PMCL) and platelets. Blood sampling was carried out from a tail vein. As

anticoagulant applied heparin, 15 units/ml. For dilution of blood used isotonic solution with concentration of NaCl of 0.75% [5].

Definition of the blood type cells for fish forms of cells and their cores after fixing and coloring of dabs by Romanovsky method were carried out on the basis of metachromasy of cytoplasm and core, as well as by the sizes. Illustrations, described by a number of authors, are also used for comparison of an [5-8]. Images of cells are received by means a light microscopy method [9]. In work we used an optical microscope of AxioStar plus (Carl Zeiss, Germany, 2009) and the Canon Super Short camera (Germany, 2009).

Calculation of blood cells was carried out in Goryaev's camera. Concentration of hemoglobin was measured by the standard method [10].

RESEARCH RESULT:

As a result of the conducted researches it is established that normal mature erythrocytes of common carp have an oval or elliptical form with pale eosinophilic cytoplasm. In the center of cells the dark-violet core which form is similar to a form of a cell is situated (fig. 1a). Young erythrocytes (fig. 1b) more roundish, the ratio of core and cytoplasm at them is higher, than in mature red blood cells.

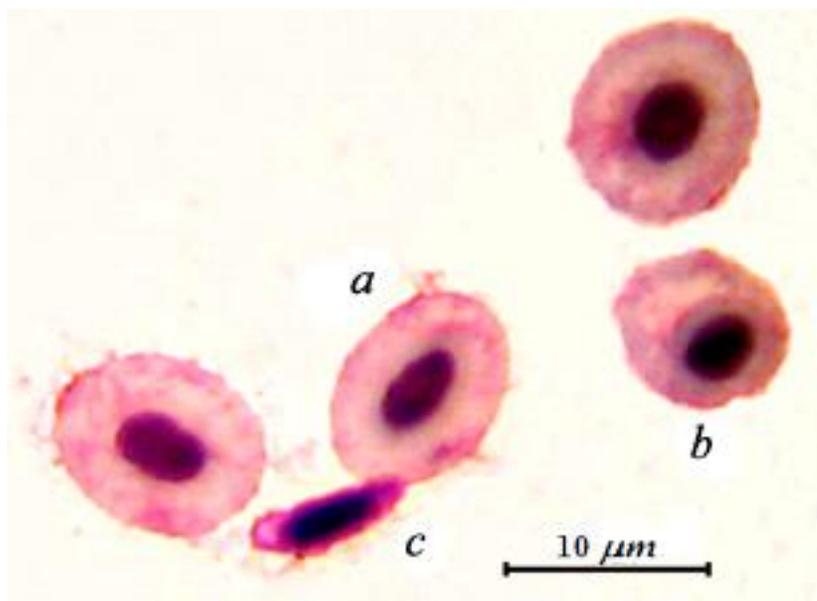


Fig 1: *Cyprinus carpio* blood cells: a - a mature erythrocyte, b - a young erythrocyte, c - a platelet.



Fig 2: A platelet of irregular shape in *Cyprinus carpio* blood (see point mark)

At increase 100× it is visible that edges of erythrocytes are uneven. At calculation it is established that the number of erythrocytes in blood of a common carp is in limits of 1.7 ± 0.2 million mm^3 , the content of hemoglobin - is 104.1 ± 8.1 g/l. Red blood plates of a common carp extended (see fig. 1c) or are irregular shape (fig. 2). The dark extended core of platelets with plentiful hetero-chromatin occupies the most part of a cell. Their cytoplasm is eosinophilic, poorly eosinophilic or basophilic.

Set of factors, including external form, color, type of core and inclusion of cytoplasm was criterion for classification of leukocytes. Under a microscope various forms of leukocytes with the varying sizes are identified. Among them we observed both

nongranulocytes, and granulocytes. Nongranulocytes include monocytes and lymphocytes, granulocytes - eosinophils, basophiles, neutrophils, as well as other uniform blood cells which are at different development stages: progranulocyte, myelocyte, metamyelocyte, stab and segment cell leukocytes. The number of leukocytes in a common carp blood varies within 33.7 ± 7.6 thousand mm^3 .

Monocyte of a common carp – is a cell of the big size, oval, rounded or irregular shape with basophilic or neutral cytoplasm. A half or the most part of cytoplasm are occupied by the wrong core, near it is big vacuoles. The maintenance of monocytes in *Cyprinus carpio* blood makes $2.8 \pm 0.6\%$ of the total amount of leukocytes (fig. 3).

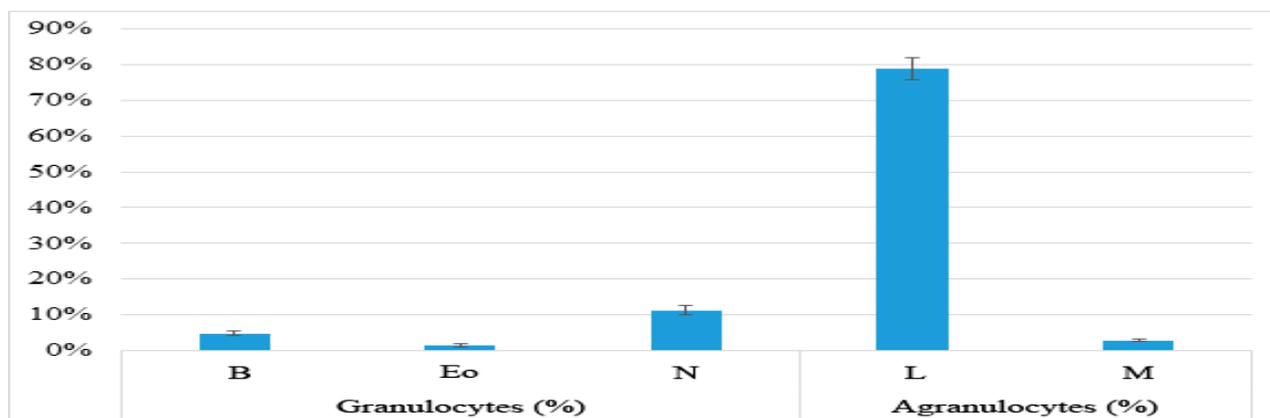
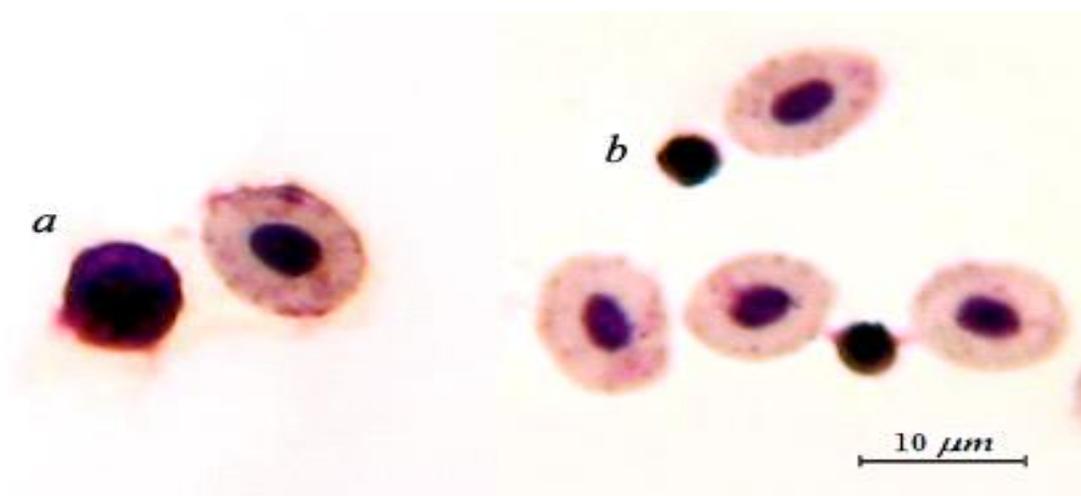


Fig 3: Leukocyte formula *Cyprinus carpio*:

B - basophiles, Eo - eosinophils, N - neutrophils, L - lymphocytes, M - monocytes.



**Fig 4: *Cyprinus carpio* lymphocytes:
a - big lymphocyte, b - small lymphocyte.**

Lymphocytes of common carp rounded shape with a dark basophile cytoplasm. There are identified big (fig. 4a) and small (fig. 4b) lymphocytes. By the size big lymphocytes are 2-3 times larger, than small lymphocytes.

Contents in blood of a common carp $78.8 \pm 6.2\%$ of lymphocytes (see fig. 3).

Depending on color of cells after coloring, under a

microscope we observed different types of granulocytes. Cells of this look make about 20% of total number of leukocytes of a common carp.

Eosinophils have red-violet color (fig. 5), core – is dark-violet, various form, and containing slightly condensed chromatin. In the circulating blood of a common carp are identified eosinophilic myocyte (fig. 5a), eosinophilic metamyelocyte (fig. 5b) and a segment cell eosinophil (fig. 5v).

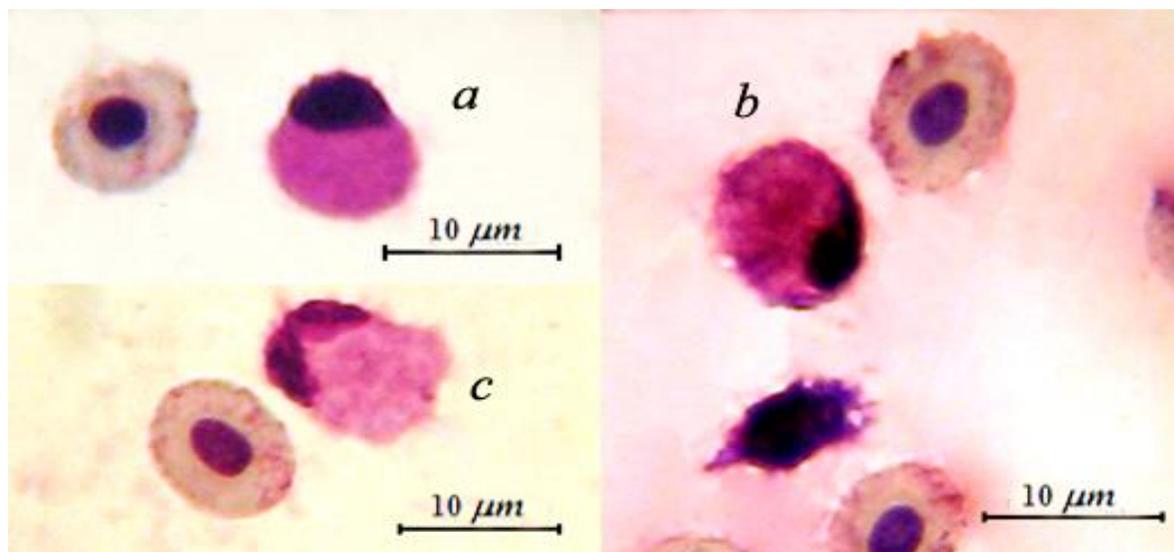


Fig 5: Eosinophils in *Cyprinus carpio* blood:

a - eosinophilic myelocyte, b - eosinophilic metamyelocyte,
c - segment cell eosinophil.

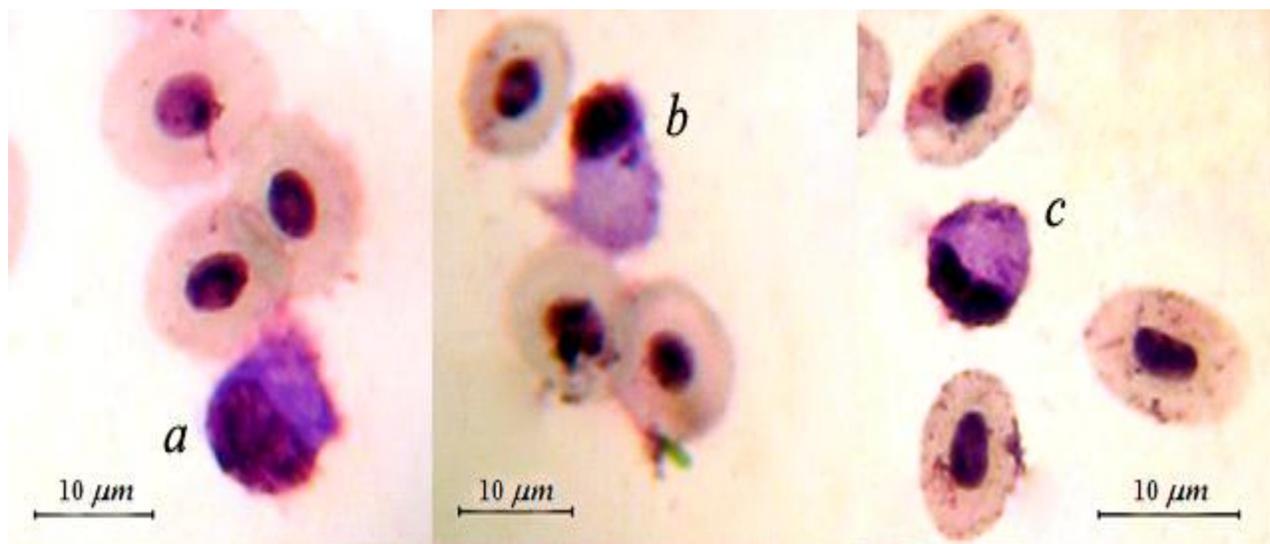


Fig 6: Basophiles in *Cyprinus carpio* blood:

a - basophilic myelocyte, b - basophilic metamyelocyte,
c - stab basophile.

The maintenance of eosinophils in *Cyprinus carpio* – is from 0.6 to 2.4% of total number of leukocytes (see fig. 3).

Cytoplasm of basophiles – is blue or weak-blue color with a dark-violet core. In blood of a common carp basophilic myelocyte (fig. 6a), basophilic metamyelocyte (fig. 6b) and stab basophiles (fig. 6c) are revealed.

Basophiles make from 3.5 to 6.1% of total number of leukocytes of a common carp (see fig. 3).

Neutrophils - cells with cytoplasm of neutral color, their core of violet or dark-violet color of a different form. We identified cells of neutrophils at different development stages: neutrophilic myocyte (fig. 7a), neutrophilic metamyelocyte (fig. 7b), a stab neutrophil (fig. 7c).

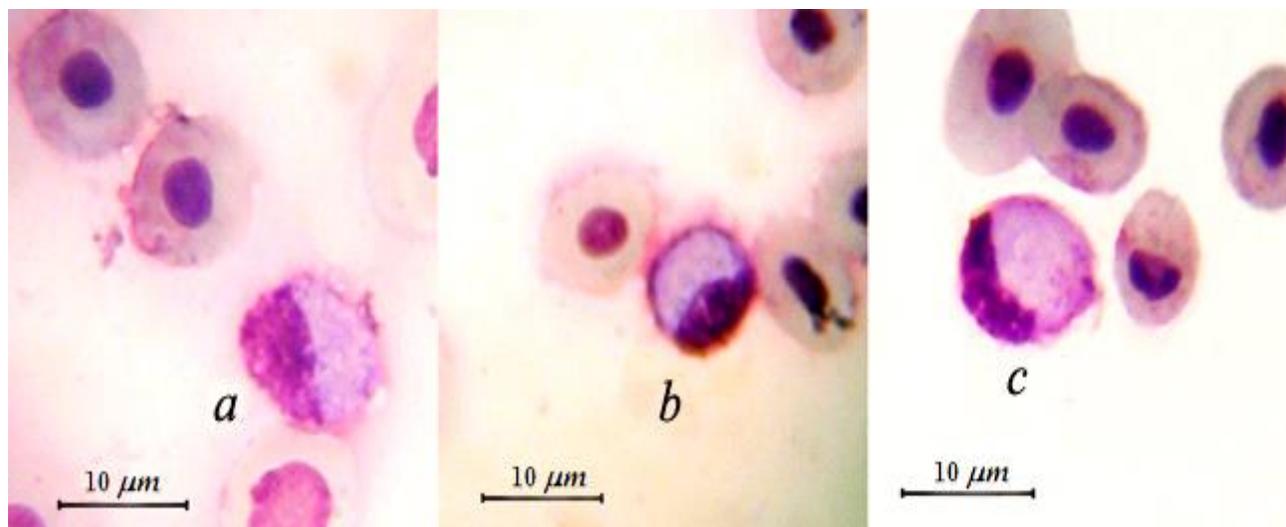


Fig 7: Neutrophils in *Cyprinus carpio* blood:

a - neutrophilic myelocyte , b - neutrophilic metamyelocyte ,
c - stab neutrophil.

The maintenance of neutrophils at a common carp makes $11.3 \pm 2.8\%$ of total number of leukocytes (see fig. 3).

DISCUSSION:

As a result of the conducted researches it is established that according to the morphological characteristic erythrocytes of a common carp are similar to red blood cells of other species of fish [8, 11, 12], at the same time cells of this pool are pre-potent type. As it was told above, the number of erythrocytes at the studied species of fishes makes 1.7 ± 0.2 million in mm^3 blood. The data obtained by us coincide with results of works of a number of authors [5, 7, 13-17].

In blood of the species of fishes studied by us hemoglobin level slightly varied, however was [5, 7, 13-17] within limits of physiological norm. It is known that this indicator depends as on ecological features of an organism of an animal [18], and from their physical [19-20] and a physiological state [21]. Besides, the same species of fishes has non-uniform fractions of hemoglobin differing in the carried-out functions [22, 23].

The lymphocytic profile as in a leukocyte formula of hemocyte lymphocytes occupy the most part is characteristic of the studied species of fishes. Under a microscope of a cell of this look similar to lymphocytes of mammals and birds.

The results on granulocytes received in our work meet expectations which are recognized as most of researchers [5, 7, 24-27] according to which for fishes granulocytes are divided into 3 look: eosinophils, neutrophils and basophiles depending on color of core and cytoplasm. We revealed the granulocytes which are at different development stages: myeloplast, promyelocyte, myelocyte, metamyelocyte, stab, segment cell granulocytes. The most part among polymorph cell leukocytes neutrophils, quantity of eosinophils and basophiles occupy rather low the studied species of bony fishes.

SUMMARY:

Thus, in the conducted research the hematologic profile of the representatives of the fresh-water fishes inhabiting the Belgorod region - common carp *Cyprinus carpio* is studied. On the basis of cytoplasm and core metachromasy, the sizes, a form of cells and their cores after fixing, as well as colourings of dabs by Romanovsky method with use a light microscopy method in peripheral blood of common carp *Cyprinus carpio* are revealed erythrocytes, polymorph cell leukocytes and platelets. It is established that in a hematologic profile of a common carp red blood cells dominate. The most part in a leukocyte formula is occupied by lymphocytes. Among granulocytes neutrophils prevail.

The revealed hematologic profile of the bony fish representatives can be used for assessment of their

functional state, health and extent of adaptation to environment conditions.

CONCLUSIONS:

1. It agrees to morphological and hematologic analyses, blood cells of common carp *Cyprinus carpio* are in limits of borders of normal amounts.

2. The hemacytrate profile of blood of a common carp is presented by the erythrocytes prevailing by quantity (1.7 ± 0.2 million/ mm^3), platelets and leukocytes among which the greatest percent is made by lymphocytes ($78.8 \pm 6.2\%$).

3. Granulocytic forms of leukocytes *Cyprinus carpio* are presented by neutrophils ($11.3 \pm 2.8\%$), basophiles (from 3.5 to 6.1%) and eosinophils (from 0.6 to 2.4%).

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