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Haematological parameters of Lake Baikal oilfish (golomyanka) (*Comephorus dybowskii* and *Comephorus baicalensis*)

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Abstract

Two endemic Baikal species of oilfish, *Comephorus dybowskii* ("small golomyanka") and *Comephorou baicalensis* ("big golomyanka") live in the pelagic zone down to the bottom (1600 m). Both have numerous adaptations to the pelagic habitat. In this regard, the paper gives results of studies of blood under various natural conditions. The cell composition of peripheral blood does not differ between the species. Light and electron microscopy showed absence of granulation in polymorphic nuclear leukocytes. Significant differences in shape (number eccentricity) and sizes of mature erythrocytes were apparent. Parameters of red blood and cytometric indices do not change significantly within each species. However, high values of variation coefficient in the parameters of white blood were observed in both. Small golomyanka in comparison with big golomyanka have a higher percentage of erythroblasts, haemoglobin concentration and oxygen capacity of blood (OCB), while the percentage of polymorphic nuclear leukocytes and mature erythrocytes is less. Both species differ only insignificantly in number of erythrocytes, leukocytes, haemoglobin concentration in erythrocytes, index of leukocyte abundance, and thrombocytes per unit of blood volume. Small golomyanka caught at different depths do not differ much in most parameters of blood, but our analysis shows a high positive correlation between habitat depth, haemoglobin concentration and erythrocyte number per unit of blood volume. Blood characteristics indicate that small golomyanka is the more metabolically active species.

Introduction

The multiple functions and high mobility of blood allow the determination of the physiological state of fish. Haematology may also be of assistance in understanding adaptation mechanisms and evolutionary processes.

Two endemic Baikal species of oilfish – big golomyanka (*Comephorus baicalensis* Pallas) and small golomyanka (*C. dybowskii* Korotneff) (*Scorpeniformes, Comephoridae*) inhabit the pelagic zone between the surface and bottom (about 1600 m). Both have numerous morphological, physiological and biochemical adaptations (Taliev & Koryakov, 1947; Taliev, 1955; Koryakov, 1972; Jakubovsky, 1995; Ju et al., 1997; Eshenroder et al., 1999). However, the blood of Baikal fish has not been investigated except for blood cell morphology of some species (Yakhnenko, 1984).

This research is aimed at determining the ranges and dynamics of changes in parameters of peripheral blood of big and small golomyanka under various conditions of their natural habitat.

Materials and methods

Small and big golomyanka were caught with a trap (Mamontov, 1999) between March and April 2001 at a station in Southern Baikal 1400 m deep near

Listvyanka. Big golomyanka were represented by 25 specimens of immature males (mean length 180 mm and mean weight 172 g). Small golomyanka were females with embryos (30 specimens) and mature males (19 specimens) of mean length 124 mm and 117 mm and weight of 107 g and 87 g, respectively. Haematological analysis was carried out with standard techniques (Kalashnikova, 1981). Morphology of blood cells including cytometric parameters was studied with light (LM) and scanning microscopy (SM). Reliability of differences in haematological parameters was checked with Student's criteria (Urbakh, 1964; Zhivotovsky, 1991).

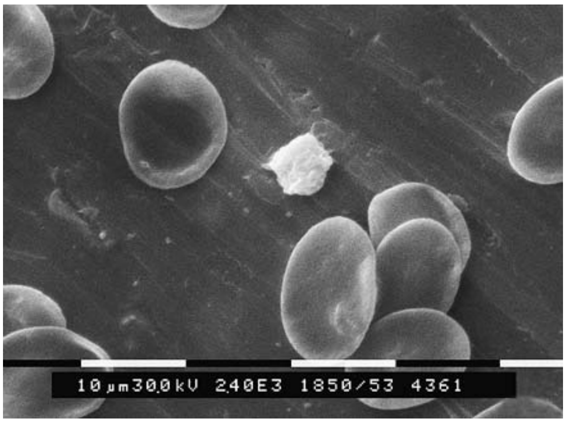

Figure 2. Structure of blood cells of big golomyanka. In the centre there is lymphocyte.

Results

Cell morphology of peripheral blood of golomyanka

Cell composition of peripheral blood does not differ between the two species of golomyanka (Figs. 1 and 2). It consists of erythrocytes and their immature forms, as well as leukocytes and thrombocytes. Due to intravascular haemopoiesis and poikilothermism of these fish, there are immature cells in peripheral red blood, the number of which corresponds to the activity of haematogenous organs and metabolism. All cells of the erythroid series (basophilic, polychromatophilic and oxyphilic erythroblasts), except haemocytoblasts, were found in the peripheral blood. Leukocytes are represented by agranulocytes (monocytes, macrophages and lymphocytes) and pseudogranulocytes (polymorphonuclear leukocytes) (Figs. 1–3). Macrophages occur in vascular blood of small golomyanka in small amounts.

Mature erythrocytes of both species differ significantly – in shape and size. Erythrocytes of small golomyanka are ellipsoidal. In big golomyanka, they are round and have a larger "useful" volume (cell volume without nucleus volume) (Figs. 1 and 2, Table 1).

Variability of blood parameters of golomyanka

Cytometric and red blood parameters within each species did not differ significantly. However, both

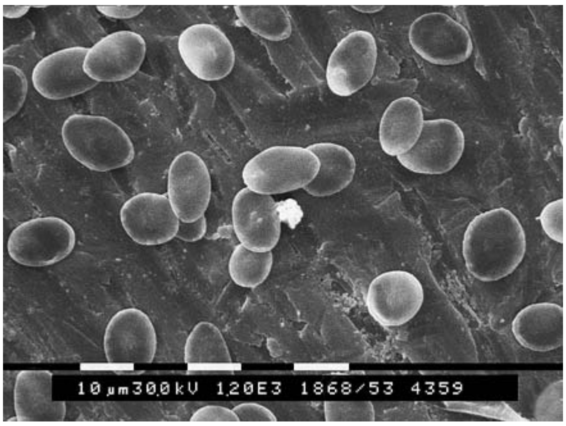

Figure 1. Structure of blood cells of small golomyanka. In the centre there is lymphocyte.

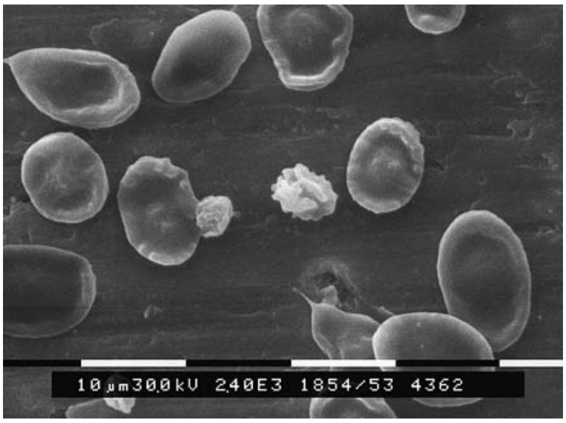

Figure 3. Structure of blood cells of small golomyanka. In the centre there is a polymorphonuclear leucocyte and thrombocyte near an erythrocyte.

Table 1. Haematological parameters of oilfishes.

Haematological parameters	Comephorus dybowskii			C. baicalensis			Distinction coefficient
	Average	± m	*C*	Average	± m	*C*	
Mature erythrocytes, %	76.01 (49)	1.47	10.1	82.31(25)	1.67	7.87	2.82*
Erythroblasts, %*	23.99 (49)	1.99	31.9	17.69 (25)	1.67	36.6	2.22*
Polymorphonuclear leukocytes, %	20.21 (49)	2.75	73.4	29.64 (24)	2.55	32.2	2.54*
Monocytes, %	2.39 (49)	0.54	122.0	3.8 (24)	0.78	76.6	1.47
Lymphocytes, %	73.81 (49)	3.42	24.9	66.48 (24)	3.04	17.1	1.6
Macrophages, %	4.04 (49)	2.12	282.0	0 (24)	0	0	1.9
Erythrocytes, mln/mm^3	0.98 (43)	0.046	26.6	0.93 (19)	0.16	53.8	0.28
Haemoglobin, g%	7.08 (43)	0.29	27.7	6.07 (16)	0.31	12.4	2.4*
OCB	11.68 (43)	0.28	11.73	10.60 (16)	0.32	7.36	2.51*
Haemoglobin in erythrocyte, μg	73.40 (43)	5.55	33.0	61.95 (16)	6.41	25.3	1.36
Abundance of leukocytes	0.19 (47)	0.02	73.0	0.32 (22)	0.13	138.4	1.0
Leukocytes, thousand/mm^3	2.73 (39)	0.44	69.06	6.51 (19)	1.56	67.1	2.16
Thrombocytes, thousand/mm^3	10.29 (39)	1.59	67.1	13.16 (19)	4.03	92.4	0.66
D_1, μm	11.54 (572)	0.04	8.23	11.28 (383)	0.05	10.1	3.44***
D_2, μm	8.01 (572)	0.03	10.1	8.18 (383)	0.03	7.82	4.33***
S, μm^2	72.62 (572)	0.40	13.26	72.43 (383)	0.44	11.8	1.58
Numerical eccentricity(D_2/D_1)	0.699 (572)	0.004	12.87	0.73 (383)	0.008	20.5	3.93***
V, μm^3	748.9 (572)	163.2	13.26	759.37 (383)	136.5	11.8	0.062
V_u, μm^3	726.8 (572)	176.2	13.26	740.6 (383)	135.7	11.8	0.049

Abbreviations: ± m – average error; C – coefficient variation; * – total erythroblasts; in brackets – number of studied objects; D_1 – big diameter of erythrocyte cell; D_2 – small diameter of erythrocyte cell; S – area of erythrocyte cell; V – volume of erythrocyte; V_u – useful volume of erythrocyte; reliability of differences: *$p < 0.05$, $p < 0.01$, $p < 0.001$.

species had high variation coefficients in the parameters of white blood (Table 1).

The percentage concentrations of erythroblasts, haemoglobin and oxygen capacity of blood (OCB) are much higher in small than in big golomyanka, while the percentage of polymorphonuclear leukocytes and mature erythrocytes is lower than in big golomyanka.

Both species differ slightly in number of erythrocytes, leukocytes and thrombocytes per unit of blood volume, as well as in haemoglobin concentration in erythrocytes and in index of leukocyte abundance (Table 1).

Clear differences occur in number and 'useful' volume of erythrocytes (Table 1, Figs. 1 and 2).

Habitat depth and blood parameters

The fishing gear used was appropriate to catch these fish concentrated at their normal habitat depth. Analysis of depth distribution indicated their constant presence there. Hence, it may reasonably be assumed that when blood samples were taken the fish were adapted to conditions in their habitat.

Analysis of variation of haematological parameters in small golomyanka caught at different depths did not reveal significant differences in most of the blood parameters. However, there was a high positive correlation of haemoglobin concentration with catch depth and with erythrocyte abundance per unit volume of blood (Table 2).

Discussion

Historical evolution of golomyanka from their divergence from a general ancestor till the formation of modern species, has been indicated by morphological and ecological data to have been about 2.0 million years (Eshenroder et al., 1999), and from genetic data about 0.6 million years (Slobodyanyuk et al., 1995). During their evolution, golomyanka became adapted to occupy the entire pelagic zone of Lake Baikal. Adaptations

Table 2. Variabilty of blood parameters with depth in small golomyanka.

Parameters	Depth, m				
	50–400	400–750	750–1000	1000–1400	Correlation coefficient
Number of erythrocytes, mln/mm^3	0.91 ± 0.12	0.98 ± 0.13	0.92 ± 0.11	1.06 ± 0.04	0.67
Haemoglobin concentration, g %	5.32 ± 0.33	6.43 ± 0.93	7.05 ± 0.35	7.45 ± 0.48	0.87

included means of maintaining the stability of the internal environment. Blood plays a part in this.

High values of variation coefficient between different populations of leukocytes (Table 1) reflect the different functions they perform (Parnes, 1977) and the individual peculiarities of organisms (Likhachyova et al., 2000).

This research supports previous conclusions by Yakhnenko (1984) on the absence of real granulocytes in golomyanka's blood. Granulocytes of fish are known to undergo morphological and functional changes during process of formation. Their presence in blood is associated with qualitative and quantitative changes of cells and granulation – an immune reaction (Mikryakov, 2001). The adaptive nature of immune reactions to different types of effect is indicated by numerous studies (Pulsford et al., 1994; Serpunin & Likhachyova, 1998; Mikryakov, 2001). Absence of granulation containing material necessary for the function of leukocytes (Parnes, 1977) in polymorphonuclear leukocytes, suggests that this mechanism of immunity has not yet evolved in golomyanka.

The different physiological states that these species undergo indicates a high level of variability in the parameters of white blood. A higher level of erytropoiesis and input of tissue macrophages and lymphocytes into peripheral blood is most likely connected with the spawning phase in small golomyanka.

Parameters of red blood characterizing respiratory features of golomyanka have intermediate values compared with corresponding parameters in benthic and littoral spawning species of sculpins (Yakhnenko, 1984).

Among pelagic sculpins golomyanka have a relatively low oxygen demand, which may result from a less active way of life (Taliev, 1955). Evidence of this are the small dimensions of the gill respiratory area (Jakubovsky, 1995) and infrequent respiratory movements (Taliev & Koryakov, 1947). Parameters of red blood in small golomyanka, however, are higher than those in big golomyanka, which may relate to a more active way of life (Eshenroder et al., 1999). Functional differences in both species affect morphological characteristics, including size and shape of erythrocytes which are smaller and ellipsoidal in small golomyanka. Low intraspecific variability in sizes of mature erythrocytes, and sharp interspecific morphometric differences between erythrocytes suggest that these are fixed species characteristics.

Our results showed that in small golomyanka changes of blood composition with depth are in general insignificant. It is known that pressure changes do not significantly influence the blood parameters of aquatic organisms without respiratory cavities (Itazawa, 2001).

Hence, during the period when golomyanka are at considerable depths in winter–spring (Mamontov et al., 2002), their blood composition is correspondingly adapted, and vertical migrations are probably accomplished in phases which allow pauses for depth adaptation. Reduction of water oxygen concentration is the factor that causes some rise in haemoglobin concentration and number of erythrocytes in small golomyanka at great depths. Anoxia is known to intensify the activity of haemopoietic organs and as a result erythrocytes are discharged into the blood and haemoglobin concentration increases (Serpunin & Likhachyova, 1998; Likhakhyova et al., 2000).

Conclusion

Granulation of leukocytes has not evolved in golomyanka. Their rather sluggish way of life probably accounts for low oxygen demand and low parameters of red blood. Small golomyanka are the more active of the two species and their red blood cells are smaller and ellipsoidal. Morphometric parameters of erythrocytes are fixed species characteristics.

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