International Journal of Fisheries and Aquaculture Sciences.
ISSN 2248-9975 Volume 3, Number 1 (2013), pp. 63-70

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Seasonal variations in haematological parameters of hill stream fish, *Garra gotyla gotyla* from Jhajjar stream of Jammu region, India

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ABSTRACT

Seasonal variation in total erythrocyte count (TEC), Haemoglobin level (Hb), Haematocrit (Hct), Differential leucocyte count (DLC), Total leucocyte count (TLC), Mean corpuscular volume (MCV), Mean corpuscular haemoglobin (MCH) and Mean corpuscular haemoglobin concentration (MCHC) were determined in hill stream fish, *Garra gotyla gotyla* from Jhajjjar stream of Jammu region, J&K India. Presently RBC dependent parameters viz. TEC, Hb and Hct exhibit a marked increase ($P<0.01$) during spring and summer season. TLC in tune with RBC dependent parameters also depicted a significant increase ($P<0.01$) during the spring and summer season. Whereas an increase in the calculated values of RBC, MCV and MCH has been observed from monsoon to winter season. MCHC showed an increase during the hot period of the year.
Key Words: Seasonal variation, Hill stream, *Garra gotyla gotyla*, Jhajjar stream.

Introduction

Since hematological parameters reflect the poor condition of fish more quickly than other commonly measured parameters, and since they respond quickly to changes in environmental conditions (Alkinson and Judd, 1978), they have been widely used for the description of healthy fish (Blaxhall, 1972), for monitoring stress responses (Soivio and Oikavi, 1976 and Kocabatmaz andEkingen, 1984) and for predicting systematic relationships and the physiological adaptations of animals. Haematological analysis will enhance fish cultivation by facilitating early detection of situations of stress and or diseases that could affect production performance (Rehulka et al., 2004; Tavares-Dias et al., 2005). Likewise the available information also indicates that haematological parameters of fishes vary with age and development (Syrove, 1970), starvation (Kawatsu, 1974 and Mahajan and Dheer, 1983), temperature and season (Joshi et al., 1980), sex (Rukusa and Zukowski, 1980; Sharma, 1984; Pickering, 1986 and Al-Hassan et al., 1993), salinity (Dheer et al., 1986) activity of fish and disease etc. Therefore, it is useful to build up a bank of information by generating basic data of haematological variation in different fish species in relation to ecological and environmental conditions so that later comparison can be easily made from this information for detecting deviation from the normal under different stocking, feeding and water condition for proper health management.

Presently an attempt has been made to generate data on the hematological parameters of hill stream fish *Garra gotyla gotyla* during the different seasons of the year.

Table 1: Seasonal variations in hematological parameters of Garra-gotyla gotyla in relation to water temperature and Dissolved oxygen during Nov. 2009- to Oct. 2010

Month	Water Tempt.	DO	TEC ($\times 10^6$/cmm)	TLC ($\times 10^3$/cmm)	Hb (gm%)	Hct (%)	MCV (fl)	MCH (pg)	MCHC (%)
November	18.2	8.0	2.27±0.19	16.95±0.24	7.8±0.22	39.8±0.53	175.4±1.25	34.2±0.32	19.5±0.23
December	15.5	8.3	2.16±0.45	14.62±0.52	7.6±0.63	39.2±0.43	181.3±1.33	35.0±0.56	19.2±0.05
January	13.6	9.1	2.05±0.94	14.29`±0.72	7.4±0.15	38.8±0.18	189.0±2.10	35.8±0.88	19.0±0.62
February	14.5	8.5	2.36±0.31	14.92±0.07	7.8±0.42	39.6±0.96	167.7±1.84	33.0±0.73	19.7±0.83
March	17.6	7.4	2.56±0.62	15.46±0.51	8.3±0.98	41.1±0.37	160.4±1.67	32.3±0.21	20.1±0.42
April	22.8	7.2	2.78±0.04	16.05±0.38	8.8±0.61	41.9±0.07	150.9±2.34	31.7±0.19	21.0±0.31
May	26.3	6.6	3.02±0.15	16.74±0.95	9.3±0.74	43.0±0.14	142.6±2.69	30.7±0.03	21.5±0.14
June	30.5	6.2	3.03±0.49	17.21±0.34	9.5±0.31	44.2±0.62	141.1±2.47	30.9±0.46	21.5±0.76
July	28.2	6.4	2.87±0.84	16.63±0.19	9.1±0.12	42.8±0.68	148.9±3.20	31.6±0.75	21.2±0.43
August	24.7	6.7	2.65±0.23	16.02±0.46	8.5±0.51	41.3±0.52	155.8±1.63	32.0±0.32	20.6±0.92
September	21.5	7.2	2.52±0.98	15.85±0.83	8.2±0.49	40.8±0.33	162.0±2.94	32.5±0.51	20.1±0.26
October	20.2	7.8	2.41±0.65	15.29±0.55	8.0±0.05	40.2±0.46	166.8±2.44	33.1±0.69	19.8±0.59

Table 2: Seasonal variations in the differential leucocyte count (DLC) of *Garra gotyla gotyla* in Relation to water temperature and Dissolved oxygen during Nov. 2009- to Oct. 2010

Month	Water Tempt.	DO	Lymphocyte %	Neutrophil %	Monocyte %	Eosinophil %	Basophil %	Thrombocyte %
Nov.	18.2	8.0	39.4±0.42	17.5±0.12	2.3±0.26	0.7±0.25	0.8±0.74	39.3±0.26
Dec.	15.5	8.3	37.5±0.62	16.4±0.53	2.0±0.39	0.9±0.63	0.7±0.32	42.5±0.18
Jan.	13.6	9.1	39.0±0.88	18.2±0.41	2.2±0.14	0.6±0.44	0.9±0.67	39.1±0.64
Feb.	14.5	8.5	42.5±0.25	19.9±0.36	2.5±0.62	0.4±0.52	1.1±0.43	33.6±0.76
Mar.	17.6	7.4	45.8±0.04	22.5±0.48	2.4±0.81	1.5±0.06	1.3±0.02	26.5±0.41
Apr.	22.8	7.2	48.6±0.36	23.7±0.05	3.3±0.52	1.3±0.17	1.5±0.15	21.6±0.36
May	26.3	6.6	51.5±0.64	28.7±0.62	4.0±0.43	1.8±0.43	1.5±0.64	12.5±0.06
Jun.	30.5	6.2	53.9±0.23	30.1±0.85	4.3±0.19	2.1±0.72	1.9±0.39	7.7±0.43
Jul.	28.2	6.4	50.5±0.14	25.4±0.40	3.9±0.69	1.3±0.91	1.0±0.94	17.9±0.82
Aug.	24.7	6.7	48.2±0.72	21.4±0.96	3.5±0.22	1.1±0.34	0.5±0.40	25.3±0.43
Sept.	21.5	7.2	46.3±0.61	19.5±0.16	2.7±0.31	1.0±0.11	0.3±0.18	30.2±0.88
Oct.	20.2	7.8	43.5±0.33	18.5±0.56	2.5±0.41	0.8±0.84	0.3±0.54	34.4±0.16

Material and Methods

Live specimens of fish *Garra gotyla gotyla* were netted out from Jhajjar stream of river Tawi of Jammu region during the entire year by using cast nets. Blood samples were drawn by cardiac puncture and carefully opening the thoracic cavity by a longitudinal incision medially between the pectoral fins. The needle was inserted into the heart and blood was drawn. The blood was then gently expressed into vials containing EDTA as anticoagulants. Erythrocyte and leucocyte counts were determined by using Neubauer's improves Haemocytometer (Shaw, 1930) using Haem and Turk solution respectively. Haematocrit was measured by Wintrobes tubes after filling with anticoagulant and spinning them at 3000 rpm for 30 minutes. Haemoglobin concentration of blood was evaluated by Sahil's haemometer (Dethloff et al., 1999). Differential leucocyte count was done by using technique of Mahajan & Dheer (1979). Erythrocyte indices the so called absolute/calculated values viz. mean corpuscular volume (MCV), mean corpuscular haemoglobin concentration (MCHC) and mean corpuscular (MCH) were calculated mathematically from the haematocrit, haemoglobin concentration and erythrocyte count. The haematological parameters were determined in relation to water temperature and dissolved oxygen in different seasons spring (February-March), summer (April-June), monsoon (July-August), autumn (September- October) and winter (November- January).

Results and Discussion

The results (Table 1 and 2) obtained clearly show that haematological parameters of fish, *Garra gotyla gotyla* exhibit mark alterations in response to changing levels of water temperature and dissolved oxygen. The haematological parameters have a direct relation with water temperature and an inverse relation with dissolved oxygen. This has earlier been reviewed by Aldrin et al., 1982; Azizoglu and Cengizlu, 1996; Denton and Yousef, 1975; Hickey 1982; Houston and Koss, 1984; Jewet et al., 1991 and Martinez et al., 1994.

The data obtained comprehensively indicate a significant ($P<0.01$) increase in the RBC dependent parameters viz. TEC, Hb and Hct during spring and summer season and exhibit maxima during summer season (June). The rise observed in the RBC dependent parameters viz. TEC, Hb and Hct during spring and summer season has been attributed to an adaptation on the part of fish to cope with natural stress caused by an increase in water temperature and low dissolved oxygen content (environmental factor) in aquatic ecosystem (Preston,1960; Joshi et al., 1980; Wells, 1999; Jawad et al., 2004 and De Pedro et al., 2005) and moreover to prepare itself for breeding period which requires greater physical activity and high energy demand (physiological factor) (Eisler, 1965; Joshi and Tandon, 1977 and Khan,1977).

After exhibiting an increase during spring and summer season, RBC dependent parameters viz. TEC, Hb and Hct tend to fall during monsoon till winter and minima has been recorded during winter (January). The possible reason for this decline is metabolically inactiveness of fish due to less availability of food during this period in spite of the fact that DO is available in plenty. Franzoni et al. (1977) also reported marked numerical decline in circulatory erythrocytes and hence related Hb and Hct during the period of low temperature in cyprinid, *Phracticthys andruzii*. Present author feels that reduction in TEC has directly influenced the Hb and Hct values where the declining number of RBCs in the circulation led to haemodilution of blood thereby decrementing the Hb and Hct (Preston, 1960).

RBC calculated indices; MCV and MCH exhibit a significant increase from summer onwards till winter whereas MCHC showed a relation parallel to RBC dependent parameters viz. TEC, Hb and Hct. Both MCV and MCH exhibit a negative correlation with TEC and temperature therefore as the temperature raises both MCV and MCH values decline and vice versa. Likewise after summer onwards as the temperature showed a marked decline the MCV and MCH showed a significant increase in their values. Decrease in the values of MCV and MCH with an increase in water temperature finds a relation with the process of erythropoises through which young erythrocytes (corpuscles) were added to blood stream of fish. These young erythrocytes being smaller in size have low volume and possess low haemoglobin content (Blaxhell and Daisley, 1973) which result in decline in the values of MCV and MCH. The increase in the values of MCHC during spring and summer has been an outcome of greater physical and higher metabolic activity of fish and a decline during autumn and winter season (cold period) to low oxygen demand of fish during this period.

Like RBC dependent parameters total leucocyte count (TLC) also depict a marked increase during the spring and summer season. During this period increased water temperature acts as a natural stressor and creates pathogenic condition in fish. To counter such condition fish respond by either increasing leucocyte count or their products e.g. lysozymes, peroxides, interferons, lysins etc., an immunostimulatory response and thus become immunologically strong enough to face any type of stress (natural and anthropogenic) in their habitat. To support present view findings of Adewoye (2010) are taken in to consideration who also reported an immunostimulatory response by fish during hot period of the year. Moreover Smirnova (1962) and Ibrahim et al. (2003) also observed a positive correlation between TLC and feeding status of fish, thus an increase has been observed during the hot period (summer season) of the year. However low temperature during the winter season usually limit the prevalence of pathogenic events in the aquatic ecosystem and therefore results in immunodepressive state in fish. Hence a decline in leucocyte count has been observed during this period. It is further revealed from the table (2) that increase in TLC during spring and summer finds a direct relation with corresponding increase in both agranulocytes (lymphocyte and monocyte) and granulocytes (neutrophils, eosinophils and basophils). Moreover major contribution toward TLC has been found to because of lymphocytes. Thrombocytes which play an important role in blood clotting exhibit marked increase during the colder period of year.

Conclusion

Thus, the present study has demonstrated a significant difference in the haematological parameters of hill stream fish *Garra gotyla gotyla* during the different seasons of the year with corresponding variation in temperature and DO on one hand and feeding intensity and reproduction on other.

Acknowledgement

The authors wish to thank Ms Madhu Sudan for advice with the statistical analyses.

References

[1] Adewoye, S.O. (2010). Haematological and biochemical changes in *Clarias gariepinus* exposed to *Trephosia vogelli* extract. Advances in Applied Science Research, 1(1): 74-79.

[2] Aldrin, J.F., Messager, J.L. & Saleun, S. (1982). Analyses sanguineous de turbots d'eleuages immature.(Scophtalmus maximus l.) Aquaculture,40 : 17-25.

[3] Al-Hassan, L.A., Ahmed, H.K. & Majeed, S.A. (1993). Some haematological parameters in relation to the biology of the fish Acanthopagrus latus. J.Environ.Sci.Health.Part A,Environ Sci.Eng.,28 (7) : 1599-1611.

[4] Alkinson, J & Judd, F.W. (1978). Comparative haematology of *Lepomis microlophus* and *Chiclasoma cyanoguttarum*. Copeia 12: 230-237.

[5] Azizoglu, A. & Cengizler, I. (1996). Saglikli Oreochromis niloticus (L.) bireylerinder bazi haematologik parametrelerin saptanmasi uzerine bir araptirma. Turkish J.Vet.Ani.Sci, 2: 425-431.

[6] Blaxhall, P.C. (1972). The haematological assessment of the freshwater fish. A review of selected literature. Journal of Fish Biology 5(6): 771-781.

[7] Blaxhall, P.C. & Daisley, K.W. (1973). Routine haematological methods for use with fish blood. J. Fish Biol.5: 771-781.

[8] Denton, J.E. & Yousef, M.K. (1975). Seasonal changes in the haematology of raibow trout,Salmo gairdneri . Comp.Biochem.Physiol.,51: 151-153.

[9] De Pedro, N., Guijarro, A.C., Lopez Patino, M.A., Martinez-Alvarez,R. & Delgodo, M. (2005). Daily and seasonal variations in haematology and blood biochemical parameters in tench *Tinca tinca.*

[10] Dethloff, G.M., Schlenk, D., Khan, S. & Bailey, H.C. (1999). The effect of copper on blood and biochemical parameters of rainbow trout *(Oncorhynchus mykiss*). Hakis Environ. Contam.Toxicol.,36: 415-423.

[11] Dheer, J.M.S., Dheer, T.R. & Mahajan, C.L. (1986). Haematological and haemopoietic response to sodium chloride stress in a fresh water air-breathing fish *Channa punctatus (Bloch.). J.Fish Biol.,28 : 119-128.*

[12] Eisler, R. (1965). Erythrocyte count and Haemoglobin content in nine species of Marine Teleost, *Chesapeake*. Sci. 6: 116-120.

[13] Franjoni, G., Berli, R & Borgioli, G. (1997). Hepatic respiratory compensation and haematological changes in cave cyprinid, *Phrealichthys andruzii*.J. Comp. Physol.,167: 461-467.

[14] Hickey, C.R.Jr. (1982). Comparative haematology of wild and captive canners.Trans. Am.Fish.Soc.,111: 242-249

[15] Houston, A.H. & koss, T.F.1984. Plasma and red cell ionic composition in rainbow trout exposed to progressive temperature increase. J.Exp.Biol.;110: 53-63.

[16] Ibrahim, S.A., Tayel, S.I., Mahmoud, S.A & El-Kashief, M.A. (2003). Impact of the carbamate pesticide sevin on haematology and histology of teleost fish (*Oreochromis niloticus*). Global Veterinaria,3(3): 196-203.

[17] Jawad, L.A., Almukhtar, M.A., Ahmed, H.K. (2004). The relationship between haematocrit and some biological parameters of Indian Shad, *Teniualosa ilisha* (family clupidae). Anim Biodivers Conserv 27 :478-483.

[18] Jewet, M.G., Behmer, D.J. & Johnson, G.H. (1991). Effects of hyperoxic rearing water on blood haemoglobin and haematocrit levels of rainbow trout. Journal of Aquatic Health,3(3): 153-160.

[19] Joshi, B.D. & Tandon, R.S. (1977). Seasonal variation in some haematological values of a hill stream fish *Noemacheilus rupicola*.Him.J.Env.Zool.,5: 103-108.

[20] Joshi, B.D., Chaturvedi, L.D. & Debral, R. (1980). Some haematological values of *Clarias batrachu*s L.following its sudden transfer to varying temperatures.Indian J.Exp.Biol.,18(1): 76-77.

[21] Kawatsu, H. (1974). Studies on the anemia of fish.VI. Further note on anemia caused by starvation in rainbow trout. Bull.Fresh water fish. Res. Lab. Tokyo, 24: 89-99.

[22] Khan, S.A. (1977). Studies on haematology of fresh water catfish, *Clarias Batrachus* (I.) I. Seasonal variations in erythrocyte and leucocyte count..Comp.Physiol.Ecol.,22: 88-92.

[23] Kocabatmaz, M. & Ekingen, G. (1984). DeÛißik tŸr balÝklarda kan šrneÛialÝnmasÝ ve hematolojik metotlarÝn standardizasyonu. DoÛa Bilim Dergisi D1, 8: 149-159**.**

[24] Mahajan, C.L. & Dheer, J.M.S. (1979) . Cell types in the peripheral blood of an air breathing fish, *Channa punctatus*. Jounal of Fish Biology,14: 281-287

[25] Mahajan, C.L. & Dheer, T.R. (1983). Haematological and haematopoietic responses to starvation in an air breathing fish *Channa punctatus* (Bloch.).J. Fish Biol., 22(1): 111-123.

[26] Martinez, F.J., Garcia-Riera, M.P., Canteras, DeCosta, J.& Zamora, S. (1994). Blood parameters in rainbow trout *Onchorhynchus mykiss* :simultaneous influence of various factors .Comp.Biochem.Physiol.,101: 95-100.

[27] Pickering, A.D. (1986) .Changes in the blood cells composition of the brown trout *Salmo trutta* L.during season. J. Fish Biol., 29: 35-347.

[28] Preston, A. (1960). Red blood values in the plaice (*Pleuronectes platersa*,L.) .J.Mar.Biol.Ass.U.K.,39: 681-687.

[29] Rehulka, J., Minarik, B. & Rehulkov, A.E. (2004). Red blood cell indices ofrainbow trout, Onchorhynchus mykiss (Walbaum) in aquaculture. Aquac. Res. 35: 529 - 546.

[30] Rukusa, S. & Zukowski, C. (1980). Blood of antartic fishes,Notothenia rossimarmorata (Fischer) and Notothenia nybelin. Pol. Polar Res., 1(1): 103-108.

[31] Sharma, T. (1984). Haematological studies on some hillstream fishes.Ph.D.Thesis,Kumaon University.Almora.U.P.

[32] Shaw, A.F. (1930). A direct method for counting the leucocytes ,thrombocytes and erythrocytes of bird’s blood.J.Path.Bact.,33: 833-838.

[33] Soivio, A. & Oikari, A. (1976). Haematological effects of stress on a teleost (*Esox lucius* L.). J. Fish Biol. 8: 397-411**.**

[34] Smirnova, L.I. (1962). Vop. Ikhtiol., 2: 677.
[35] Syrove, V.S. (1970). Seasonal and age related changes in morphological composition of the blood in grass carp and silver carp. Rubn. Khoz Repuls. Mexhvedon Temet Nouch, 98-103.
[36] Tavares-Dias, M. & Barcellos, J.F.M. (2005). Peripheral blood cells of the armoredcatfish *Hoplosternum littorale* Hancock, 1828: a morphological and cytochemical study. Braz. J. Morphol. Sci. 22: 215 - 220.
[37] Wells, R.M.G. (1999). Haemoglobin function in aquatic animals:molecular adaptations to environmental challenge.Mar.Freshwater Res.,50: 933-939.