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RESEARCH NOTE

HAEMATOLOGICAL STUDIES ON SOME FRESHWATER TELEOSTS

Receipt of MS 13-11-1978

G.L. Smit* and J. Hattingh**

**Department of Zoology, Rand Afrikaans University and*

***Department of General Physiology, University of the Witwatersrand, Johannesburg, 2001*

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The diagnostic value of hameatological indices for fish blood has been discussed by Hesser (1960) and Blaxhall and Daisley (1973). Their results signify the importance of such basic information when investigating the ecophysiology of the various freshwater fish species in South Africa where fish farms are being established in order to provide an economical source of high protein diets. When studying the effects of diseases as well as the pathophysiology of various freshwater fish species, it is therefore of the utmost importance to establish base line haematological values. In his study on the effects of bacterial infection on the haematology of rainbow trout, Barham (1978) found for instance, raised ESR values and reduced haematocrit and total protein values. Apparently healthy stock was found to be severely infected and antibiotic treatment improved the general haematological profile as well as the general production ability of the stock. Consequently, outbreaks of diseases, which can be disastrous to the fish farmer, could be diagnosed by simple haematological techniques, resulting in their timeous prevention or control. The aim of the present study was to investigate the haematology of some of the fish species in our local freshwaters under standardised laboratory conditions and to compare these results mutually and with existing information. Simultaneously, it was hoped to relate the haematology of the fish species investigated to their physiological activity and/or taxonomic value as well as gaining insight into some of the factors influencing fish haematology.

Ten adult and healthy specimens of both sexes of the common carp *Cyprinus carpio* L. and the endemic freshwater bream, *Sarotherodon mossambicus* (Peters) were obtained from the Fisheries Research Station at Marble Hall, Transvaal. Ten rainbow trout, *Salmo gairdneri* (Richardson), of both sexes, were obtained from a local fish farmer. (These species are all currently being investigated as to their suitability for local fish farming). The acclimatory procedure for the fish species investigated during the late summer/early autumn of 1978 has been described elsewhere (Smit & Hattingh, 1979) and consisted of laboratory acclimatization for two months prior to use followed by transfer to individual aquaria 24 hours before experimentation. Blood was sampled by cardiac puncture (Smit *et al.* 1977a) from stunned fish of each species (Hattingh *et al.* 1975). Thereafter, the blood was immediately analysed for the parameters shown in Table 1 utilizing the methods described by Barham (1978). In the case of blood glucose, lactic acid, ATP and total protein concentrations, commercial biochemical test combinations (Boehringer, Mannheim) were used. Plasma osmolality was measured with a Wescor Model 51008 digital osmometer. Analyses were preformed on plasma (Hattingh, 1972) except in the case of pH, pCO_2, pO_2, haemoglobin concentrations (Hb), red blood cell count (RBCC), and glucose. In Table 1 Ht refers to haematrocrit, ESR to erythrocyte sedimentation rate and TPP to total plasma protein concentration. The fish were acclimatized at 18 ± 1°C at a pO_2 of 126,9 ± 1,1 mm Hg, a pCO_2^2 of 0,5 ± 0,01 mm Hg and a pH of 7,71 ± 0,02. Blood samples were obtained from fasting animals at a standardized time of day. Mean mass for *Cyprinus carpio* was 395,11 ± 34,95 g, for *S. mossambicus* 188,66 ± 37,07 g and for *S. gairdneri* 174,00 ± 10,55 g. Statistical significance of differences between means was tested according to an unpaired Student's t-test.

The results obtained are shown in Table 1. An asterisk next to the mean value for any given haematological parameter of *Salmo gairdneri* indicates that the value differs significantly from the values of both *Cyprinus carpio* and *Sarotherodon mossambicus* with $P < 0{,}001$. Similarly, an asterisk next to the mean value for any given haematological parameter of *Sarotherodon mossambicus* indicates that the value differs significantly from both the mean value of *Cyprinus carpio* and *Salmo gairdneri* with $P < 0{,}001$. The present results do not correspond with those reported by Hattingh (1972) and Fourie (1974) for common carp in their natural habitat. They do however agree with results obtained for trout (Barham, 1978) and *Sarotherodon mossambicus* (Hattingh, 1972). These intraspecies differences may be attributed to a variety of factors, the most important of which seems to be water quality (Van Vuren & Hattingh, 1978). Hughes (1973) drew attention to the fact that haematological parameters of fish show acclimatization to the acquatic environment. It is, however, clear that different fish species acclimated to identical laboratory conditions, still show considerable differences in their haematology. This could possibly be ascribed to species specific metabolic characteristics which will be reflected

Table 1

Haematological values for C. carpio, S. mossambicus *and* S. gairdneri *obtained under laboratory conditions (n = 10)*

Parameter	*Cyprinus carpio*				*Sarotherodon mossambicus*				*Salmo gairdneri*			
	Mean	S.D.	Span	C.V.	Mean	S.D.	Span	C.V.	Mean	S.D.	Span	C.V.
pH	7,77	0,04	7.70– 7,81	0,51	7,70	0,04	7,65– 7,78	0,51	*7,40	0,01	7,37– 7,43	0,13
pCO_2 mm Hg	4,72	0,71	4,00– 6,00	15,04	4,33	0,66	3,00– 5,00	15,24	*3,50	0,35	3,00– 4,00	10,00
pO_2 mm Hg	4,55	0,68	4,00– 6,00	14,94	*14,00	1,22	12,00– 15,00	8,71	*8,38	0,70	8,00– 10,00	8,40
Ht %	20,44	1,94	18,00– 23,00	9,49	20,22	2,81	16,00– 23,00	13,83	*35,44	5,72	27,00– 49,00	16,13
Hb g %	4,26	0,61	3,48– 5,13	14,31	5,16	0,56	3,97– 5,68	10,85	*6,72	0,91	5,87– 8,99	13,15
RBCC x $10^6/mm^3$	0,60	0,05	0,50– 0,66	10,00	*1,19	0,13	0,96– 1.32	10,92	*1,01	0,19	0,82– 1,51	18,81
MCV μm^3	338,10	11,06	322,03–360,00	3,27	*169,05	8,15	149,53–178,29	4,82	349,06	13,56	324,50–361,70	3,80
MCH pg	69,31	5,49	61,05– 76,56	7,92	*43,30	1,24	41,35– 45,00	2,86	66,44	3,21	59,53– 70,09	4,83
MCHC %	20,77	1,09	18,31– 22,30	5,24	*25,67	1,63	24,81– 29,81	6,34	19,05	1,09	18,32– 21,24	5,72
Glucose mg %	49,71	8,47	40,00– 63,33	17,03	42,91	3,49	37,93– 48,38	8,13	47,12	2,98	41,38– 51,72	6,32
Lactic acid mg %	9,50	0,65	8,20– 10,00	6,84	*4,28	0,65	2,70– 4,60	15,42	*20,18	1,89	17,30– 23,70	9,36
ATP mg %	72,54	8,33	60,40– 80,34	11,51	*21,94	11,66	8,80– 37,20	53,14	*52,56	5,73	43,80– 61,32	10,90
ESR mm/hr	3,30	0,82	2,00– 4,00	24,62	*31,00	4,15	27,00– 38,00	13,38	*1,11	0,22	1,00– 1,50	19,81
TPP g %	2,66	0,07	2,53– 2,81	2,63	2,72	0,14	2,52– 2,90	5,14	*3,55	0,15	3,39– 3,87	4,22
Na mEq/ℓ	237,77	7,12	230,00–250,00	2,99	*194,44	8,07	180,00–210,00	4,15	230,00	0	–	0
K mEq/ℓ	3,16	0,50	2,50– 3,50	15,82	3,05	0,52	2,50– 3,50	17,04	3,00	0	–	0
Ca mg %	14,39	0,39	13,57– 15,00	2,71	*10,07	0,33	9,76– 10,71	3,27	*11,45	0,45	10,71– 12,14	3,93
Cl mEq/ℓ	120,22	1,56	118,00–122,00	1,29	*149,88	3,21	142,00–156,00	2,14	*134,44	1,01	133,00–136,00	0,74
Osmolality mOs/kg	259,11	13,33	227,00–277,00	5,14	*301,66	6,65	287,00–312,00	2,20	264,00	3,70	256,00–268,00	1,40

S.D. – Standard deviation C.V. – Coefficient of variation

in blood acid – base balance and ATP, lactic acid and plasma iron concentrations, etc. These on their part probably reflect the different mechanisms in the way fish adapt to their environment.

The taxonomic value of most haematological parameters reported on in this study is also of little importance. The range within a given specie and the overlapping of different species values are such that any comparison is of taxonomic insignificance. Those parameters which could possibly be of taxonomic value can however be influenced by so many internal and external factors that a physiological consideration would be more meaningful. A close relationship between Ht, Hb and RBCC was observed in all species with significant interspecies differences. Other parameters are also involved. It is suggested that the interspecies haematological values, of possible taxonomic significance, should rather be considered as indicating compensatory mechanisms for adaptation to different aquatic environments.

Another factor of importance when considering the differences in blood values reported for our South African freshwater fish is the use of tricaine methane sulphonate (MS 222–Sandoz) as an anaesthetic for obtaining blood samples. This substance was routinely used in the past for anaesthetization of larger fish in order to simplify the procedure of blood sampling (Fourie, 1974). Smit *et al.* (1977b) have indicated that MS 222 produces severe changes in pH, pCO_2 and alkalinity of freshwater with different mineral contents. It is possible that such changes may be reflected in the haematology of freshwater fish and Soivio *et al.* (1977) reported on such effects of MS 222 in rainbow trout. However, associated with this problem is also the possible incidence of capture stress. It is known that stress results in undesireable effects on the haematology of freshwater fish (Casillas & Smith, 1977; Hattingh, 1977) and the possibility that MS 222 anaesthesia indirectly creates a stress situation through its effects on water quality, requires further investigation. In the present study no anaesthetic was employed which eliminated this possibility. Furthermore, results obtained here also provide evidence for species specific reactions to capture stress. In *S. gairdneri*, a high blood lactic acid level was recorded, which may eventually result in a lactic acid shock mortality as suggested by Caillouet (1971). In addition, the differences observed in haematocrit and RBCC values probably indicate different magnitudes of the increase in RBCC which is known to be mediated by the central nervous system (Kirk, 1974) and the increase in plasma volume (Hattingh *et al.* 1975). This latter effect, however, seems to be least in trout which compares well with the results obtained by Barham (1978).

From the present results, it is suggested that the aquatic environment probably plays a minor role in determining interspecies haematological values but that the metabolic scope and activity of a fish species should rather be considered as the primary factors involved. Intraspecies differences are however strongly influenced by water quality (Van Vuren & Hattingh, 1978). Further studies on the haematology of these species under unusual environmental stress would therefore be of great value.

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References

BARHAM, W.T., 1978. An investigation of certain haematological parameters of the rainbow trout *Salmo gairdneri* Richardson with reference to the possible effects of bacterial infection. M.Sc. Dissertation, RAU.

BLAXHALL, P.C. & DAISLEY, K.W., 1973. Routine haematological methods for use with fish blood. *J. Fish Biol.* 5, 771.

CAILLOUET, C.W., 1971. Lactate acidosis as a cause of mortality in captured sharks: A hypothesis. *Trans. Amer. Fish Soc.* 100, 139.

CASILLAS, E. & SMITH, L.S., 1977. Effects of stress on blood coagulation and haematology in rainbow trout *(Salmo gairdneri). J. Fish Biol.* 10, 481.

FOURIE, F. LE R.,.1974. 'n Studie van die algemene bloedfisiologie van die tiervis *(Hydrocynus vittatus)* en die karp *(Cyprinus carpio).* M.Sc. Dissertation, RAU.

HATTINGH, J., 1972. Observations on the blood physiology of five South African freshwater fish. *J. Fish Biol.* 4, 555.

HATTINGH, J., FOURIE, E. LE R. & VAN VUREN, J.H.J., 1975. The transport of freshwater fish. *J. Fish Biol.* 7, 447.

HATTINGH, J., 1977. Bloodsugar as an indicator of stress in the freshwater fish *Labeo capensis* (Smith). *J. Fish Biol.* 10, 191.

HESSER, E.R., 1960. Methods for routine fish haematology. *Progve. Fish Cult.* 22, 164.

HUGHES, G.M., 1973. Respiratory responses to hypoxia in fish. *Amer. Zool.* 13, 475.

KIRK, W.I., 1974. The effects of hypoxia on certain blood and tissue electrolytes of channel catfish, *Ictolurus punctatus* (Rafinesque). *Trans. Amer. Fish Soc.* 3, 593.

SMIT, G.L., HATTINGH, J. & SCHOONBEE, H.J., 1977a. Observations on some effects of EDTA and heparin on fish blood. *Comp. Biochem. Physiol.* 57C, 35.

SMIT, G.L., SCHOONBEE, H.J. & BARHAM, W.T., 1977b. Some effects of the anaesthetic MS 222 on freshwater quality. *S.A. Journal Sci.* 73, 351.

SMIT, G.L. & HATTINGH, J., 1979. Anaesthetic potency of MS 222 and neutralized MS 222 as studied in three freshwater fish species. *Comp. Biochem. Physiol.* (In Press).

SOIVIO, A. NYHOLM, K. & HUHTI, M., 1977. Effects of anaesthesia with MS 222, neutralized MS 222 and bezocaine on the blood constituents on rainbow trout, *Salmo gairdneri. J. Fish Biol.* 10, 91.

VAN VUREN, J.H.J. & HATTINGH, J., 1978. A seasonal study of the haematology of wild freshwater fish. *J. Fish Biol.* 13, 305.