# A SEASONAL STUDY OF THE HAEMATOLOGY OF CARP (*CYPRINUS CARPIO*) FROM A LOCALITY IN THE TRANSVAAL, SOUTH AFRICA

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## ABSTRACT

Various haematological parameters of carp blood were examined on a monthly basis from February to November. Seasonal variations were observed in red and white blood cell counts, in haemoglobin and plasma protein concentrations and in mean corpuscular fragilities. Sexual differences were evident in red blood cell counts, haematocrit values and haemoglobin concentrations. Plasma protein electrophoresis showed marked conformational changes during the study period. The results are discussed in relation to previous findings and also in relation to the use of blood as a possible indicator of breeding season in carp.

## INTRODUCTION

Until recently, few papers have been published on the haematology of freshwater fish in South Africa (Hattingh 1973, 1974). The studies mentioned concerned one or more isolated aspects of fish haematology, and were mostly conducted on about 10 specimens of each species. These studies provided very little information about the seasonal fluctuations which are known to occur in fish (Ezzat *et al.* 1973; Young 1949; Snieszko 1960). Moreover, from the research which has been done on carp in other areas of the world, it is known that fish from different localities exhibit different physiological characteristics (Nyman 1965). Apart from the work of Murachi (1959), no seasonal haematological study has been done on carp in South Africa or elsewhere and the present study was planned to provide information on normal values for comparative purposes, as well as to investigate the possible use of blood as an indicator of breeding activity.

## MATERIALS AND METHODS

The fish used in the experiments were seined each month in a farm dam at Honeydew, Transvaal, during the 10-month period from February to November 1974. They were transported to, and maintained in, the laboratory as described previously (Hattingh *et al.* 1975). The animals used were all healthy and mature.

The methods used for anaesthetization, blood sampling, length and mass determinations and the measurement of pH, haematocrit, haemoglobin, plasma protein concentration and electrophoresis have been described in detail elsewhere (Hattingh 1973, 1974). The determination

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of the fragility of carp erythrocytes was done according to the methods of Ezzat *et al.* (1973) using salt solutions ranging from 0,20 per cent to 0,42 per cent NaCl.

## RESULTS

*Seasonal variations in haematology*

The results obtained for the general haematology on a monthly basis for 10 animals have been analysed together for both sexes and are shown in Table 1.

The mean erythrocyte count (RBC) increased steadily from May (autumn in South Africa) to reach a maximum during October (spring, spawning period). During November this parameter decreased to correspond with the values obtained for the period February to April. The mean white blood cell count (WBC) fluctuated during the year but showed a marked decrease during June. Blood pH, as expected, remained reasonably constant throughout the year at 7,25 $\pm$ 0,04. Mean haemoglobin (Hb) and plasma protein (Pp) concentrations indicated maximal peaks during July. The haematocrit (Hc), which is known to be a very variable parameter in fish (Young 1949), showed no seasonal maximum although a tendency to increase during the period July to September was observed. Mean corpuscular volume (MCV), mean corpuscular haemoglobin concentration (MCH) and mean corpuscular haemoglobin concentration per cent (MCHC%) all fluctuated during the year with MCV showing a maximum during November, MCH a minimum during October and MCHC% a minimum during November.

Sexual differences were evident in three of the parameters studied. Male carp exhibited higher RBC counts than females throughout the period of study ($P < 0,001$) but both sexes followed the same pattern. The Hc values and Hb concentrations were also higher for the males throughout ($P < 0,005$). Since the fish were all sexually mature, of the same size, in good health and were on a regular diet, the differences observed between males and females are not attributable to these parameters but are probably genetically determined.

Red blood cell fragilities also showed a seasonal variation. During autumn the mean corpuscular fragility was 0,330 $\pm$ 0,04 per cent NaCl, during winter 0,375 $\pm$ 0,01 per cent NaCl and during spring 0,387 $\pm$ 0,02 per cent NaCl.

*Plasma protein electrophoresis*

The polyacrylamide electrophoretograms showed no differences between the sexes at any stage of the study. Figure 1 shows the results obtained. The full electrophoretogram presented corresponds to the standard pattern obtained during February and indicates that carp plasma separated into 14 fractions under the present experimental conditions (5 per cent acrylamide gels; Tris-glycine buffer, pH 8,5; 5 mA for 35 minutes). The fractions were numerically numbered from the point of application. The plasma protein configuration changed markedly through the year. The changes were in concentration as well as in the number of fractions, and these appeared for a short season. The deviations from the standard February pattern, together with the time of appearance, are shown in Figure 1. The largest change was observed during the spawning months, September to October, when Fractions 9 and 10 increased greatly in concentration and Fraction 10b was prominent. These changes reverted back to normal by the end of November.

TABLE 1

Mean monthly values for carp haematological parameters. Means ± S.D. Values for males and females have been combined.

| *Parameter* | *February* | *March* | *April* | *May* | *June* |
|---|---|---|---|---|---|
| Weight, g | 212,81 ± 42,91 | 214,52 ± 32,55 | 260,40 ± 73,39 | 248,02 ± 50,25 | 269,00 ± 58,99 |
| Length, cm | 23,04 ± 1,46 | 23,49 ± 1,07 | 24,88 ± 1,86 | 24,22 ± 1,81 | 25,15 ± 1,83 |
| pH | 7,33 ± 0,08 | 7,24 ± 0,13 | 7,25 ± 0,14 | 7,23 ± 0,10 | 7,24 ± 0,14 |
| Hc % | 31,42 ± 5,45 | 31,44 ± 6,74 | 30,54 ± 4,35 | 35,43 ± 7,05 | 32,92 ± 5,41 |
| Hb g% | 8,14 ± 2,24 | 8,20 ± 1,37 | 7,89 ± 1,37 | 9,60 ± 2,20 | 9,38 ± 1,28 |
| Pp. mg/ml | 21,75 ± 3,27 | 21,60 ± 2,83 | 21,71 ± 8,96 | 21,75 ± 3,27 | 20,64 ± 2,83 |
| RBC $10^6$/mm³ | 1,46 ± 0,30 | 1,49 ± 0,33 | 1,43 ± 0,47 | 1,67 ± 0,44 | 1,78 ± 0,50 |
| WBC $10^3$/mm³ | | 4,39 ± 0,83 | 5,34 ± 2,15 | 6,62 ± 4,57 | 1,45 ± 0,58 |
| MCV | 232,93 ± 32,71 | 223,36 ± 40,47 | 230,54 ± 71,54 | 207,30 ± 35,28 | 198,46 ± 44,62 |
| MCH ng | 61,00 ± 17,66 | 56,30 ± 9,94 | 59,60 ± 20,14 | 55,06 ± 9,34 | 54,52 ± 12,52 |
| MCHC% | 25,82 ± 17,66 | 26,75 ± 4,83 | 25,84 ± 2,42 | 26,70 ± 2,57 | 27,91 ± 4,10 |

| *Parameter* | *July* | *August* | *September* | *October* | *November* |
|---|---|---|---|---|---|
| Weight, g | 227,81 ± 83,10 | 258,30 ± 44,77 | 267,40 ± 57,47 | 312,40 ± 70,81 | 304,75 ± 49,70 |
| Length, cm | 22,71 ± 3,29 | 25,00 ± 1,00 | 25,20 ± 1,47 | 27,00 ± 2,10 | 26,80 ± 1,79 |
| pH | 7,29 ± 0,07 | 7,19 ± 0,08 | 7,27 ± 0,09 | 7,25 ± 0,07 | 7,25 ± 0,14 |
| Hc % | 37,93 ± 7,65 | 36,35 ± 8,23 | 42,47 ± 5,58 | 34,69 ± 4,13 | 32,60 ± 9,46 |
| Hb g% | 10,00 ± 1,17 | 8,99 ± 2,63 | 9,86 ± 1,40 | 8,74 ± 1,12 | 5,67 ± 1,69 |
| Pp. mg/ml | 36,25 ± 8,10 | 23,45 ± 4,55 | 18,00 ± 5,78 | 23,97 ± 4,87 | 22,08 ± 1,59 |
| RBC $10^6$/mm³ | 1,99 ± 0,83 | 1,83 ± 0,33 | 1,97 ± 0,49 | 2,96 ± 0,87 | 1,43 ± 0,93 |
| WBC $10^3$/mm³ | 5,92 ± 3,00 | 4,97 ± 1,63 | 5,14 ± 1,64 | 4,46 ± 1,89 | 7,84 ± 5,60 |
| MCV | 238,04 ± 121,26 | 194,32 ± 32,72 | 217,34 ± 29,06 | 125,01 ± 36,79 | 301,39 ± 213,51 |
| MCH ng | 61,66 ± 23,26 | 48,94 ± 11,62 | 51,09 ± 5,00 | 32,43 ± 7,47 | 46,95 ± 13,57 |
| MCHC% | 26,98 ± 4,11 | 25,61 ± 6,02 | 23,60 ± 0,96 | 29,09 ± 6,53 | 18,30 ± 6,00 |

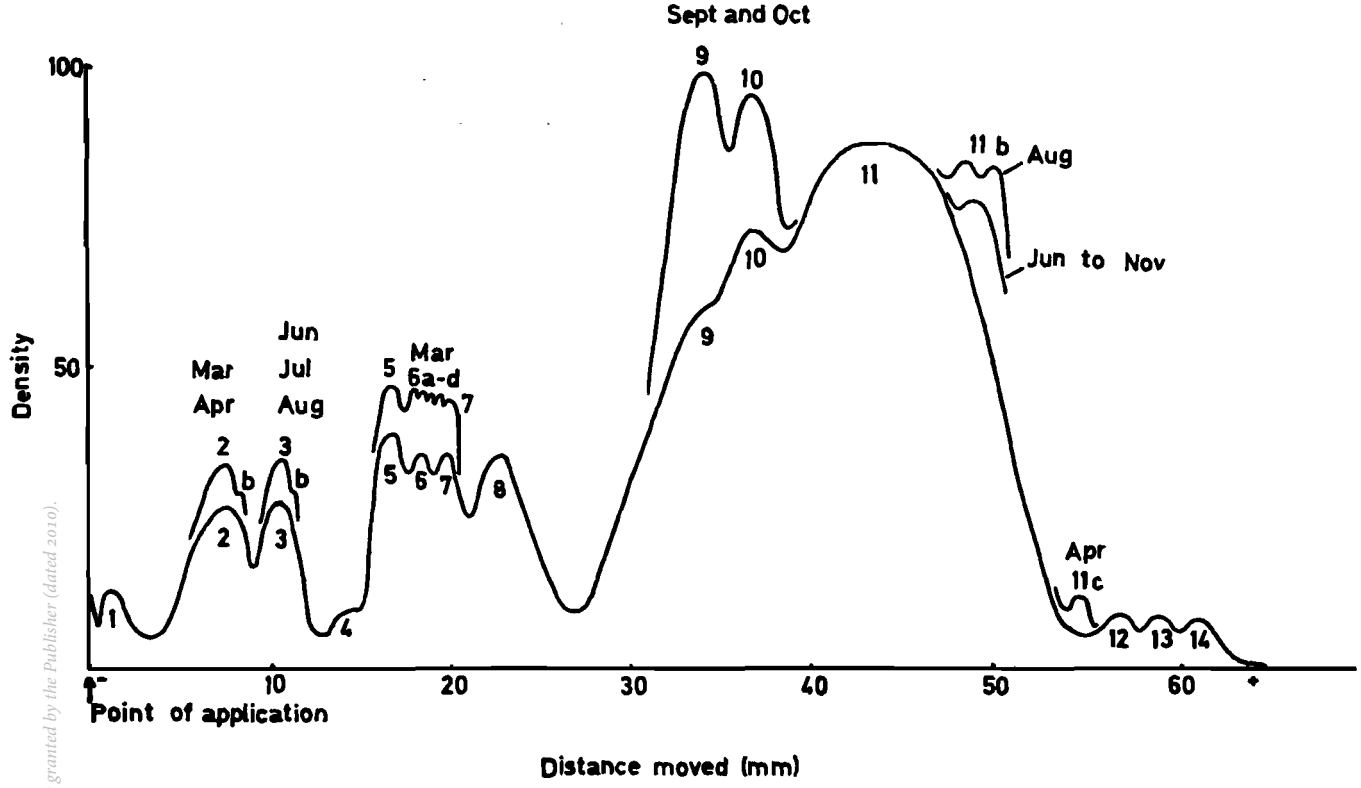


FIGURE 1

Polyacrylamide electrophoretogram of carp plasma. The full pattern is the standard pattern obtained during February. Variations observed during the year in concentration of existing fractions, or new fractions observed, are indicated by month of occurrence.

## DISCUSSION

Published results on the haematology of carp are found in the works of Black (1940), Field *et al.* (1943), Housten & De Wild (1971) and Murachi (1959). These authors reported RBC counts of 1,30–1,56 $\times 10^6$/mm$^3$, haematocrit values of 25,1–29,1 %, haemoglobin concentrations of 6–7,3 g% and plasma protein concentrations of 4,15–5,78 g%. The RBC count, Hc values and Hb concentrations of the present study are on the average higher than these values, and the Pp. concentrations lower. These differences could be attributed to differences in locality which, as has been said before, are known to have a pronounced effect (Nyman 1965).

Ezzat *et al.* (1973) showed a definite seasonal variation in leucocyte counts in *Tilapia zilli* which was also found to be the case in the present investigation. Umminger & Mahoney (1972) found a Hb maximum during summer in *Salmo gairdneri*, but Denton & Yousef (1975) indicated a high Hb concentration in winter in rainbow trout. The last work supports the present results, which are, however, contradictory to the values of Murachi (1959) who found minimum values for Hb and Hc during winter in carp. Robertson *et al.* (1961) described a seasonal variation in plasma protein concentration of salmon, with a peak during winter, again in agreement with the present results. Finally, Ezell *et al.* (1969) found that a negative correlation exists between MCV and erythrocyte fragility; the smaller the cells, the higher the mean corpuscular fragility. The same tendency was again observed in the present study, and a correlation of $r = -0{,}73$ was found between these two variables.

The fact that male fish show higher values than females for some haematological parameters, has been demonstrated by Ezzat *et al.* (1973). The two sexes do, however, follow the same seasonal pattern. The same observations were made in the present study concerning RBC counts, Hc values and Hb concentrations. A tendency for higher RBC counts during the breeding season also corresponds to the results of Ezzat *et al.* (1973) on *Tilapia zilli*. If considered together with the increases in red blood cell fragility and in the concentration of Fractions 9 and 10 of the electrophoretogram, these parameters could be used as indication of breeding season in carp. The fact that these changes correlate with breeding season was confirmed by examining the animals for the presence of ripe sperm or ova in the gonads during this season. The changes in haematology during the breeding season in these animals can thus be used when studying the effect of various hormones and other influences in the artificial stimulation of spawning in the laboratory or elsewhere.

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