

## HAEMATOLOGICAL VALUES IN COD (*GADUS MORHUA*)

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### ABSTRACT

Blood samples from cod, fed for 20 months at constant water temperature and photoperiod, were collected eight times during the feeding period.

The range of haematology values of samples means over collection times were: Haematocrit (Hct): 23–33%, haemoglobin (Hb): 5.0–7.4 g 100mL<sup>-1</sup>, red blood cell count (RBC): 1.24–1.56 10<sup>12</sup>L<sup>-1</sup>, MCV: 212–252 10<sup>-15</sup>L, MCH: 43–53 10<sup>-6</sup>g and MCHC: 19–23 g 100mL<sup>-1</sup>.

The Hct values were significantly higher in males than in females, but neither Hb nor Hct varied with the stages of gonadal development.

Significant correlations were found between Hct and RBC; Hb and RBC; and between Hct and Hb.

### INTRODUCTION

Haematological test are, due to their availability and sensitivity, a basis for appraising the status of an organism, although they are not very specific (Aldrin et al., 1982).

Seasonal changes in the haematology of fish populations have been demonstrated due to natural light periods (Sandnes et al., 1988) and water temperature (Härdig and Høglund, 1983). Such changes make it difficult to define normal values independent of variations in the environment.

Several authors have reported haematological values in various fish species, but data from Atlantic cod are scarce.

The present study reports values for haematocrit (Hct), haemoglobin (Hb), red blood cell count (RBC) and the derived blood variables mean cell volume (MCV), mean cell haemoglobin (MCH) and mean cell haemoglobin concentration (MCHC) in a population of Atlantic cod fed at constant water temperature and light regime for 20 months.

## MATERIALS AND METHODS

### *Fish and diets*

140 cod hatched and reared at the Aquaculture Station Austevoll with an initial weight of  $235 \pm 69$  g were set up in a sheltered  $25 \text{ m}^3$  tank supplied with running sea water at  $8 \pm 1^\circ \text{C}$  and  $35 \pm 2\%$  salinity. The photoperiod was constant at 12 hrs. dark and 12 hrs. light.

The fish were fed *ad lib.* three times weekly. The diet was whole capelin (44%), fish meal (40%), capelin oil (6%) and cooked starch (10%) supplied with vitamin and mineral mixtures according to Lie et al. (1986).

### *Sampling*

The experiment lasted from July 1984 to March 1986. Blood samples were collected eight times: 3, 4, 5, 6, 17, 18, 19, and 20 months after the start. At each sampling 12 fish were collected randomly, all together 41 males and 55 females. The fish were killed by a sharp blow on the head and the blood was drawn from *ductus cuvieri* as described by Lied et al. (1975).

### *Analytical methods*

Haematocrit (Hct), haemoglobin (Hb) and red cell count (RBC) was determined according to Sandnes et. al. 1988.

### *Statistics*

Correlations between the parameters determined were carried out according to Bailey (1981) and statistical evaluation of the data was carried out using the Mann-Whitney U-test (Wonnacott and Wonnacott, 1977).

## RESULTS AND DISCUSSION

The fish grew from 235 g to about 3500 g (Table 1) during the experiment. No signs of disease or nutritional imbalance were observed during this time. The mortality was very low and only four fish of a total of 140 died in the course of the experiment.

Gonad weight as percentage of body weight was used as a general indicator of the stage of sexual development. Juvenile gonads in cod have a fairly constant relative weight, below 0.5% of the body weight. The relative weight increases to above 10% during the gonad ripening. The highest value observed was from a female with a gonadosomatic index of 30%.

The haematological values are given as mean values ( $n = 12$ ) from each sampling in Table 1.

Table 1. Haematological values in cod.

Month	3	4	5	6	17	18	19	20	Significantly correlated to	
									weight	gonad weight
Weight, g .....	414	583	684	931	2758	3004	3478	2837		
SD .....	114	133	172	142	359	516	574	586		
Gonad index, % .....	0.6	0.5	0.5	1.1	3.7	2.3	2.8	4.9		
SD .....	0.3	0.4	0.3	1.7	4.9	2.7	3.2	8.0		
Haematocrit, % .....	23	30	28	29	31	33	32	32	***	
SD .....	2	2	5	2	4	4	3	5		
Haemoglobin, g 100mL <sup>-1</sup> .....	5.0	6.0	6.1	5.7	6.6	6.7	7.4	6.4	***	
SD .....	0.6	0.7	0.7	0.5	0.8	0.7	0.7	0.9		
RBC, *10 <sup>12</sup> L <sup>-1</sup> .....	-	-	-	-	1.24	1.56	1.50	1.34	*	
SD .....					0.14	0.15	0.13	0.15		
MCV, 10 <sup>-15</sup> L .....	-	-	-	-	252	212	215	237	*	*
SD .....					21	15	15	18		
MCH. 10 <sup>-6</sup> g .....	-	-	-	-	53	43	49	48		
SD .....					5	4	4	3		
MCHC, g 100mL <sup>-1</sup> .....	21	20	21	19	21	20	23	20	*	
SD .....	2	2	3	2	2	1	2	1		

\*: 0.01 < p < 0.05, \*\*: 0.001 < p < 0.01, \*\*\*: p < 0.001

The mean Hct values and the Hb concentrations (table 1) correspond well with values (Hct =  $32.0 \pm 5.2$  and Hb =  $7.4 \pm 0.6$ ) reported from cod by Larsson et al. (1976). Addison and Ackmann (1971) found Hct in the range of 30–40% in cod weighing between 5 and 6 kg. Larsson et al. (1976) reported a wide interspecies variation in Hct values, and showed mean values in teleosts ranging from 17.2% in the angler fish *Lophius piscatorius* to 52.5% in the mackerel (*Scomber scombrus*) with corresponding Hb values of 3.2 and 12.7 g/100mL. These variations are probably due to evolutionary physiological adaptation to the mode of life and ecological habitat. Mean values of RBC between  $1.24$  and  $1.56 \cdot 10^{12} \text{L}^{-1}$  were found. A mean RBC value of  $1.04 \cdot 10^{12} \text{L}^{-1}$  was found in cod of mean weight 270 g fed at 8° C (Lie et al., 1989).

Fish bone does not contain marrow for haemopoiesis, the haemopoietic sites are primarily the kidney and the spleen (Satchell, 1971). Fish erythrocytes are nucleated as in other nonmammalian vertebrates, and show a wide range of sizes among different species.

There are few data for MCV available from cod. The mean values found in this study (212–252) were about half the values reported for Atlantic salmon (Conroy, 1972; Sandnes et al., 1988).

The values of the index MCHC were in accordance with the values from cod reported by Larsson et al. (1976).

Many factors influence fish haematology. Among these are nutritional status (Barnhart, 1969; Spannhof et al., 1979), infectious diseases (Amend and Smith, 1975; Barham et al., 1980; Iwama et al., 1986), environment (Goel et al. 1981; Giles et al., 1984; Munkittrick and Leatherland, 1983 and stress (Yamamoto et al., 1980; Lowe-Jinde and Nimi, 1983; Wells et al., 1984; Ellsaesser and Clem, 1986). According to Hårdig and Høglund (1983) blood variables undergo seasonal variations in a fish population, concomitant with climatic changes in light and water temperature, but are to a lesser extent influenced by age. Conroy (1972) on the other hand, reported variations in haematological values within fish species due to age. In our experiment in which photoperiods and water temperature were kept constant, both Hct and Hb were significantly correlated ( $p < 0.001$ ,  $r = 0.502$  and  $r = 0.643$ , respectively,  $n = 89$  for both) to fish weight. Hårdig and Høglund did not find any correlation between blood variables and fish weight in a group of Baltic salmon (*Salmo salar* L.) with a mean weight of  $41.0 \pm 8.1$  g.

RBC was significantly correlated ( $0.01 < p < 0.05$ ,  $r = 0.350$ ,  $n = 47$ ) to weight in the present study. The derived blood variable MCV was significantly ( $0.01 < p < 0.05$ ,  $r = 0.313$  and  $r = 0.348$ , respectively,  $n = 47$  for both) correlated to weight as well to gonad weight. MCHC was correlated ( $0.01 < p < 0.05$ ,  $r = 0.244$ ,  $n = 89$ ) to weight.

Differences in the values for Hct and Hb related to sex were observed, the mean Hct values for females ( $n = 52$ ) and males ( $n = 39$ ) were  $29 \pm 4$  and  $31 \pm 4\%$ , respectively, with corresponding Hb values of  $6.2 \pm 1.0$  and  $6.4 \pm 0.9$  g/100mL. The difference in Hct values was significant ( $0.01 < p < 0.05$ , Mann-Whitney U test). Similar differences were observed in rainbow trout by Lane (1979), whereas Hårdig and Høglund (1983) found no correlation between blood variables and sex in a group of ten immature Baltic salmon (*Salmo salar* L). In cod Hct and Hb does not vary with the stages of gonadal development, as no correlation was observed between Hct and the gonadosomatic index nor between Hb and the gonadosomatic index.

The mean RBC values of females and males were not significantly different,  $1.36 \cdot 10^{12} \text{L}^{-1}$  and  $1.46 \cdot 10^{12} \text{L}^{-1}$ , respectively.

No difference in MCV and MCHC related to sex was observed in cod in contrast to results from rainbow trout (Lane 1979). Such a difference ( $0.01 < p < 0.05$ , Mann-Whitney U test) was observed in the MCH values, as the female cod had the highest mean value of  $49.7 \pm 4.1$  and the male a mean value of  $46.9 \pm 6.0$ . Lane (1979) reported significant sex related differences in MCH in rainbow trout, but in that report the male had the highest value.

Consistent with the findings of Lane, (1979) we found good correlations ( $p < 0.001$ ) between Hct and RBC ( $r = 0.711$ ,  $n = 47$ ) and Hb and RBC ( $r = 0.637$ ,  $n = 47$ ). A correlation between Hct and RBC was not observed in Baltic salmon (Hårdig and Høglund, 1983). According to Hårdig and Høglund, immature (immRBC) and mature (matRBC) erythrocytes are not of equal size. Eisler (1965) reported positive correlation between Hb and RBC in several fish species, whereas Hårdig and Høglund (1983) observed no such correlation. However, Hårdig and Høglund (1983) did find a positive correlation between Hb and the proportion of matRBC. This observation and the finding of a negative correlation between the proportion of immature RBC and MCH and a negative correlation between immRBC and MCV implies that the erythrocytes increase in size during maturation as they become capable of haemoglobin synthesis.

In the present study total RBC was negatively correlated ( $p < 0.001$ ) to both MCV ( $r = -0.468$ ,  $n = 47$ ) and MCH ( $r = -0.532$ ,  $n = 47$ ) but not to MCHC ( $r = -0.145$ ,  $n = 47$ ). Thus with increasing number of erythrocytes in the circulation the cells get smaller, thereby containing less haemoglobin but with the same haemoglobin concentration.

A high correlation ( $p < 0.001$ ) was found between Hct and Hb ( $r = 0.821$ ,  $n = 89$ ). According to Hårdig and Høglund (1983) this implies a constant Hb concentration in the RBC pool, emphasized by a small variation in MCHC as also observed in this population. Larsson et al. (1976) reported

that mean Hb values from 27 different fish species showed a positive correlation ( $p < 0.001$ ) with the corresponding mean Hct values.

#### ACKNOWLEDGEMENTS

The work was supported by a grant from the Norwegian Fisheries Research Council project. The skilled technical assistance of Mrs Betty Irgens is gratefully acknowledged.

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