

HAEMOGLOBIN POLYMORPHISM IN A CAT FISH *TACHYSURUS CAELATUS* (VALENCIENES)

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Haemoglobin polymorphism is a useful tool for the identification of fish populations. Sick (1961) by using electrophoresis identified two populations of cod, *Gadus morhua*. Differences in the electrophoretic fractions of the haemoglobin of the larval form of *Petromyzon planeri* and the foetal form of elasmobranch *Squalus suckleyi* were described respectively by Adinolfi et al. (1958, 1959) and Menwell (1963). Moller and Naevdal (1968) described haemoglobin polymorphism in a number of species of the gadoid family. Haemoglobin polymorphism in *Tinca tinca* consists of single haemoglobin and was split into two polypeptide chains. The polypeptides have been known to cause haemoglobin polymorphism in their presence or absence (Callegrini and Cuochi, 1963).

The cat fish *Tachysurus caelatus* is abundant in coastal waters of Goa and the estuary of Mandovi and constitutes a small fishery. Haematology and populations of the species have not been studied so far. Therefore, haemoglobin phenotypes have been studied by electrophoresis. Specimens of *T. caelatus* of both sexes and of different sizes and maturity have been studied. Blood was obtained by severing caudal peduncle in tubes previously rinsed with sodium citrate. The erythrocytes were separated by centrifugation at 3000

rpm for twenty minutes and were washed four times with cold 0.9% sodium chloride and lysed by adding two volumes of cold distilled water. The lysate was again centrifuged and the clear supernatant haemoglobin solution was used for electrophoresis.

The electrophoresis was carried out on cellulose acetate strips (Shandon) by modifying slightly the method described by Du Toit et al. (1973). The cellulose acetate strip size 25x160 mm, barbitone acetate buffer (oxid) pH 8.6, 0.06 ionic strength, were used and a current of 250 volts was applied for one hour. The proteins were stained with Amido Schwartz 10B. Identification of all the haemoglobin fractions were confirmed by the benzidine test.

The two electrophoretic patterns of the haemoglobin of *T. caelatus* show resolution of all the haemoglobin fractions towards anode only (Table 1). The repeated experiments produced the same consistent pattern and these did not show any variation either due to sex or size. Two patterns of electrophorograms were obtained consistently. Pattern one consists of three bands: two major and one minor. Two major bands show maximum mobility whereas minor band has minimum mobility. The second pattern has two bands and the

Table 1. Electrophoretic data for haemoglobin of *T. caelatus*.

No. of individuals	Electrophoresis cellulose acetate	No. of haemoglobin fractions	Protein percentage	Distance (mm) from the point of application	Relative mobility (%)
49	Pattern I	1	11.2	0.70	53.9
		2	48.8	1.00	76.9
		3	4.0	1.30	100.0
12	Pattern II	1	78.4	1.20	85.7
		2	21.6	1.40	100.0

position of the lighter band is also different. The lighter band has maximum mobility and major band has slower mobility.

The number of haemoglobin fractions, relative proportion of each fraction and the relative mobilities are shown in Table 1.

Variation in number and proportions of haemoglobin components have been shown in Japanese eel (*Anquilla japonica*) by Yamanka et al. (1965). Riggs (1970) pointed out that the number of fractions obtained in electrophoresis depends on the details of the separation procedure and one also on the binding of other molecules with haemoglobin. The results obtained show intraspecific variations but in both the patterns, the binding of other ions cannot be excluded. The appearance of such intraspecific variation of haemoglobin fractions of varying electrophoretic mobility is due to the substitution of an amino acid in each of the sub-molecules that constitutes the globin (Ingram 1967). This substitution alters the physico-chemical properties of the haemoglobin completely and causes the differences in electrophoretic mobility.

The differences in the electrophoretic pattern of the haemoglobin of *T. aculeatus* suggest the presence of two populations of these fishes in the Mandovi estuary. Further studies to assess the amino acid compo-

sition of each haemoglobin fraction of these two populations are in progress.

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REFERENCES

- Adinolfi, M. *Nature* 1958, 182, 730.
 Adinolfi, M., Chiolfi, G. and Sirtacaka, M. *Nature* 1959, 184, 1326-1329.
 Callegarini, C. and Cuschi, C. *Biochem. Biophys. Acta* 1968, 10:294-296.
 Du Toit, D., Hattingh, J. and Scheepers, J. C. *J. Fish. Biol.*, 1973, 5: 525-530.
 Ingram, V. J. *Nature* 1957, 180, 326-332.
 Minwall, C. *Arch. Biochem. Biophys.*, 1963, 101, 804-809.
 Motter, D. and Naeyna, G. *Oceanogr. Mar. Biol.* 1964, 7, 400-533. Harold Barnes (ed.) George Allen & Unwin Ltd., London.
 Riggs, A. in *Fish Physiology* 4, 209-252 W. S. Hoar and D. J. Randall (eds.), Academic Press, New York, London, 1970.
 Sisk, K. *Nature* 1961, 193, 894-895.
 Yamanka, H., Yamaguchi, K. and Matsumura, F. *Bull. Jap. Soc. Scient. Fish.* 1965, 31, 827-832.