

## Soluble Lens Protein Polymorphism in Flying-Fishes from the Central Arabian Sea

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Soluble eye lens nuclei proteins of flying-fishes were studied by cellogel electrophoresis. Three distinct patterns characterized by the number of bands, mobility and staining intensity were observed. Morphological studies of these fishes showed that they belonged to species *Exocoetus volitans* Linn. and *Cypsilurus speculiger* (Cuv. & Val.). The eye lens proteins of *C. speculiger* showed 2 distinct electrophoretic patterns suggesting the occurrence of 2 populations of *C. speculiger* which can be differentiated externally by the length of the pectoral fin and the depth of the body.

The flying-fishes (family Exocoetidae) are widely distributed<sup>1</sup>. During the 3rd Cruise of *ORV Sagar Kanya* in the Arabian Sea in Oct. 1983, a number of flying-fishes were collected by simple dip-nets at the 3 sampling stations. In the present study, they were taxonomically differentiated using electrophoresis of eye lens proteins.

Eye lenses were removed from the fish and freed from the aqueous and vitreous humor, pieces of retina, capsule and cortex. The lens nuclei were weighed and placed in a dry tube. To extract the lens proteins, 0.018g (100 ml)<sup>-1</sup> saline solution, equal to 7 times the wet weight (g) of the lens tissue, was added to each tube<sup>2</sup>. After the addition of the fluid, the lens tissue was macerated vigorously and kept for overnight extraction at 4°C. It was then centrifuged at 3,000 rpm for 1 min. Electrophoresis (Systronics Electrophoresis Migration Cell) was carried out on cellogel strips of 2.5 × 14 cm size. Veronal buffer (pH 8.6) of ionic strength 0.05 was used in the electrode and in the buffer compartments and also for soaking the strips. About 3 µl of the sample was applied to the strip on the cathode side (about 2 cm from the bridge) in a straight continuous line. Following the electrophoresis, the

strips were stained with ponceau S stain (0.2% in 3% trichloroacetic acid) by immersing them in the stain for about 5 min. After staining, the strips were washed in 5% glacial acetic acid until the background became perfectly white. The strips were scanned using a photoelectric densitometer. The mobility of the band was calculated as percentage of the ratio of the distance travelled by the band to that of the distance travelled by the fastest moving band. The percentage of protein in each band was calculated from the densitometric readings.

Eye lens nuclei of 30 flying-fishes were screened individually at each sampling station. Three distinct electrophoretic patterns were seen at all stations (Table 1). Morphological examination of the fishes showed that they belonged to the species *Exocoetus volitans* Linn. and *Cypsilurus speculiger* (Cuv. & Val.). The morphological characters of the fishes were as follows:

*E. volitans*—Total length 13.0-16.3 cm, Dorsal 14, Anal 13, Pectoral 1.13, Ventral 1.5. Pectorals reaching to base of caudal. Origin of dorsal and anal opposite. Ventrals small, their origin much nearer to the tip of snout than to the base of caudal.

*C. speculiger* (type I) —Total length 16.4-19.1 cm,

Table 1—Electrophoretic Analysis of Eye Lens Proteins

(Relative mobility as percentage along with the percentage of protein in each fraction is given in parenthesis)

Species	Fractions (in order from the slowest to the fastest)							No. of samples		
	1	2	3	4	5	6	7	St 1	St 2	St 3
<i>E. volitans</i>	-36.26 (8.09)	-9.09 (29.03)	+9.09 (20.95)	45.45 (13.24)	100 (28.68)			17	20	19
<i>C. speculiger</i> (type I)	-22.73 (9.87)	0 (26.75)	13.64 (28.4)	45.45 (8.23)	68.18 (7.41)	100 (19.34)		5	5	7
<i>C. speculiger</i> (type II)	-26.32 (9.55)	-5.26 (12.86)	10.53 (12.86)	34.21 (7.47)	57.89 (12.45)	78.95 (14.52)	100 (30.29)	8	5	4

Station locations: st 1 Lat. 16°N, Long. 62°E; st 2 Lat. 16°15'N, Long. 60°03.43'E; and st 3 Lat. 18°N, Long. 62°E

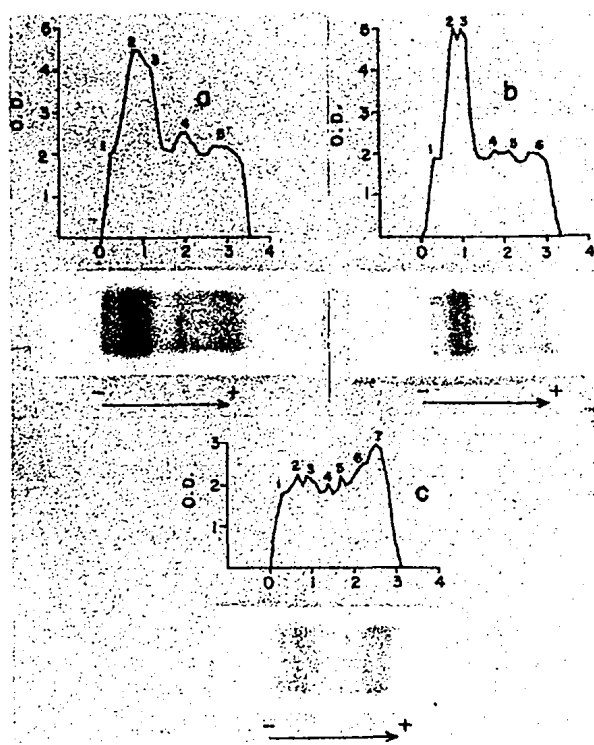


Fig. 1—Electrophoretic patterns of eye lens protein along with the densitometric tracings of *E. volitans* (a); *C. speculiger*, type I (b); *C. speculiger*, type II (c)

Dorsal 10, Anal 10, Ventral 6, Pectoral 1.16. Pectoral fin reaches to the end of dorsal fin. Origin of dorsal opposite to that of anal. Ventrals longer than head reaching to about the middle of anal, their origin nearer to caudal than the eye. Pectorals with a reddish patch and a white hind margin.

*C. speculiger* (type II)—Total length 18.0-19.5 cm. Dorsal 10, Anal 10-11, Pectoral 1.16, Ventral 6. Fishes somewhat broader than type I. The pectoral fin reaches only up to middle of dorsal fin. Other characters similar to type I.

Clear differences are seen in the electrophoretic patterns also (Table 1). The eye lens proteins of *E.*

*volitans* shows 5 bands. The 2nd and 5th bands are major protein bands. In *C. speculiger* (type I), the eye lens proteins show 6 bands. The 2nd and 3rd bands are the major protein bands. The eye lens proteins of *C. speculiger* (type II) shows 7 bands. The 7th band is the major protein band. The patterns along with the densitometric tracings showing the intensity of proteins in each band are given in Fig. 1. The fractions have been assigned numbers in the order of increasing mobility.

Electrophoretic patterns of eye lens proteins were constant with respect to number, mobility and intensity of the bands within each species/group. They did not show any variation either due to sex or size. The available evidence indicates that the variations in the nuclear eye lens proteins have a genetic basis<sup>3-5</sup>.

Weber and Beaufort<sup>1</sup> have described *C. speculiger* as consisting of pectorals reaching to the end of dorsal or not so far. From the present study, it is seen that these narrow bodied and slightly broad forms with pectorals reaching to the end of dorsal and not so far respectively, show clear differences in the eye lens nuclear proteins. *C. speculiger* (type I) showed 6 bands and *C. speculiger* (type II) 7 bands. They further differed in the amounts of protein in the various bands. In view of the above differences noticed, it may be concluded that these two forms of *C. speculiger* belong genetically to 2 different populations.

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