PRELIMINARY RESULTS OF ELECTROPHORETIC STUDIES ON MARINE PRAWNS

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ABSTRACT

•pleetroptooreitiic separation of proteins of abdominal muscle tissue of four commercfelHy important specie® of peroaeid[1] prawns, viz., *Penaeus indicus, Metapenaeus dobsoni, M. monoceros* and *M. affinis* ww drae by polyacrylanride gel electrophoresis. The number of bam* representing *h« protein fractions varied from severi to thirteen', indCcating the distinctive nature of the specieis investigated.

Electrophoretic separation of tissue proteins of fish and prawns has been generally acclaimed as a promising technique for the detection of population units (Johnson et al 1974). This technique has been found useful in answering questions on systematics or population genetics in penaeid prawns of the Gulf of Mexico (Lester 1979). As pointed out by Lester (1980), it is important to study the genetic nature of the species of prawns involved in both offshore and in nursery areas and the number of separate populations within each species before attempting the population dynamics of shrimp fishery. The utility of this method in estimating the genetic variation in a stock has been emphasised by Lester (1979a) with special reference to shrimp culture.

At present, very little information is available on population genetics of Indian prawns of commercial importance. Therefore, studies on population genetics of marine prawns have been initiated in the Crustacean Fisheries Division of the Central Marine Fisheries Research Institute, at Cochin. As part of these investigations, an attempt was made to study the differences in the electrophoretic patterns of muscle proteins of four of the important species of prawns of the southwest coast of India, viz., *Penaeus indicus, Metapenaeus dobsoni, M. monoceros* and *M. affinis.*

Juvenile prawns were collected from Cochin backwaters and kept in pools containing fresh brackish-water. The feshly killed prawns from these stocks were shelled and 1 g of the muscle tissue from the first abdominal segment was taken and homogenized with 2 ml of distilled water, keeping the tissue cool with ice pack. The extract was centrifuged at 3500 rpm for 10 minutes and the supernatant containing proteins was used as sample for electrophoresis.

Three-layer-disc electrophoresis of Davis (1964) using 7% acrylamide for separating gels was adopted with certain modifications. Ammonium persulphate was used as chemical catalyst in separating gel while riboflavin was used as catalyst in spacer and sample gels. Tris-glycine buffer of pH 8.4 was used as electrode buffer in both chambers. A current of 5mA per tube was applied using a D.C. power pack. Bromophenol blue was used as the marking dye. The run took about an hour for completion.

Staining for general protein was done with 2% Coomassie Brilliant Blue solution in acetic acid, methanol, water in the ratio 1:2:5, after fixing the gels in 10% trichloroacetic acid (TCA) for 30 minutes. Destaining was done, first with one change with same acetic acid-methanol-water solution and then with 7% acetic acid in water, till the gels were clear with well-differentiated bands. The relative mobolites of the different fractions of proteins were estimated from the distance travelled by jthe marking dye from the origin and the distance travelled by each of the fractions from the origin. The percentage transmission of the stained gel was determined, using a red filter of 640 mu for every millimetre with a densitometer.

P. indicus exhibited thirteen protein fractions while the number of fractions varied from seven in *M. momceros* to eleven in *M. affinis,* with *M. dobsoni* having eight fractions (Fijg. 1). There was considerable variation in the relative mobilities of the various fractions of proteins in these species. In fact, there

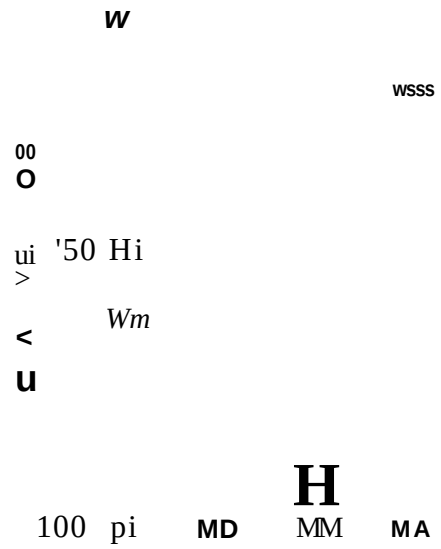

FIG. 1. Diagrammatic ^presentation of the proteins bands obtained in podyacryilamiidie gel electrophoresis: PI f= *P. indicus;* IMD = *M. dobsoni;* MM — *M. monoceros* and MA = *M. affinis.*

were less number of slow moving fraction viz., fractions 1 to 6 in all species. The number of these slow fractions .was more in *P. indicus* (3) and nil in *M. monoceros* while in *M. dobsoni* and *M. affinis* their numbers were 1 and 2 respectively. Protein fractions of medium mobility, viz., 7 to 13 were least in number, namely in *M. dobsoni,* while this number varied from 3 in *M. monoceros* to 4 in *M. affinis* and to 5 in *P. indicus.* The fast moving fractions were more in number than the other fractions. In *M. monoceros* there were only 4 of these fractions while there were 5 in *M. affinis* and *P. indicus* and 6 in *M. dobsoni.*

The relative mobilities (Rm) exhibited by the various fractions of tissue proteins also differed in the different species. Thus, the light band nearest to the origin in *P. indicus* was broad with relative mobilities ranging from 4.5 to 19.0. The other light band was far away from this band (Rm = 62.0-65.5). The fastest moving fraction exhibited a relative mobility of 93.5-95.5. In *M. dobsoni,* although the light band nearest to the origin was wide (Rm = 24.5-30.0), the next band which was deeply stained and broad was far away (Rm = 65.0-69.0). The dark band, farthest from the origin in this species was with relative mobility 90.0-91.0. The relative mobility of the band nearest to the origin was 50.0-51.0, in *M. monoceros,* while that of the third band which was poorly stained was 56.5-60.0. The most deeply stained fraction having relative mobility of 75.5-77.0. In the case of *M. affinis,* the bands were more or less uniformly distributed, except for a gap between the second and third bands. The first band (Rm = 21.0-23.5). The second, light band (Rm = 68.0-72.0) was broader than the others. The fastest fraction had a relative mobility of 92.0-93.0.

The densitometric anlysis of these stained gels exhibited well defined peaks representing the clear dark bands and less prominent peaks showing less stained fractions. Marked differences were noticed in the curve patterns of the four different species of prawns in accordance with the intensity and width of the various protein bands, representing the fractions.

The results of these preliminary experiments clearly indicate that there are differences in the protein structure of the muscles of the different species of prawns with reference to the protein fractions and their relative mobilities. Further detailed studies on the individual species based on samples collected from geographically isolated places may help in delineating different stocks or races, if any. Investigations on these lines are being taken up to evaluate the genetic resources of various commercial species of prawns.

The author is thankful to Dr. E. G. Silas, Director, Central Marine Fisheries Research Institute, for his encouragements and to Dr. M. J. George, for critically going through the manuscript and, suggesting improvements.

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