3. Genetic Variation in *Nautilus pompilius*

by

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Among four (SAUNDERS, 1981 a, b) or six (JECOLN, 1980) currently recognized *Nautilus* species, *N. pompilius* shows the widest geographical distribution in the waters of Southwest Pacific (HAMADA, 1977; SAUNDERS, 1981 b). Because of its wide distribution, *N. pompilius* shows a fairly large intraspecific variation for the total live weight and shell size in the mature stage (HAYASAKA *et al.*, 1982).

Recently, we had a chance to obtain samples of *N. pompilius* from the Philippines and Fiji, the opposite extremities of its distribution area. The present study of electrophoretically detectable genetic variation in *N. pompilius* was undertaken to investigate the degree of genetic differentiation between the two samples.

Materials and Methods

Seventeen specimens captured in 1981 at Tañon Strait, the Philippines and 36 specimens captured in 1982 off Suva Barrier Reef, Viti Levu Island, Fiji were examined. After capture, small pieces of mantle and mid-gut gland were dissected from the specimens and were frozen immediately, then transported to the Laboratory of Fisheries Resources, Kagoshima University, where they were stored at −30°C until used.

Samples of mantle and mid-gut gland were homogenized in an equal volume of distilled water and the homogenates were electrophoresed horizontally for approximately 4 hours at 5°C. Starch gels were prepared using 12.5% electrostarch in citrate-N-(3-aminopropyl)-diethanolamine buffer, pH 7.0 (CLAYTON and TRETIAK, 1972). The enzymes assayed and staining procedures used are presented in Table 1.

Table 1. Enzymes assayed, and tissues and staining procedures used.

Enzyme	Abbreviation	Tissue	Reference for staining procedure
Fumarase	*Fum*	Mantle	Shaw and Prasad(1970)*
Glutamate-oxaloacetate transaminase	*Got*	Mantle	Taniguchi and Numachi(1978)
Glucosephosphate isomerase	*Gpi*	Mid-gut gland	Shaw and Prasad(1970)
Malate dehydrogenase	*Mdh*	Mantle	Numachi(1970)
Tetrazolium oxidase	*To*	Mid-gut gland	Numachi(1972)

* Modification: 0.1M Tris-HCl buffer, pH8.7

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Results and Discussion

A. Electrophoretic variation

Nine gene loci coding for 5 enzymes were scored. Electrophoretic patterns and allele frequencies for each locus are given in Fig. 1 and Table 2, respectively. The results for each enzyme scored are described below:

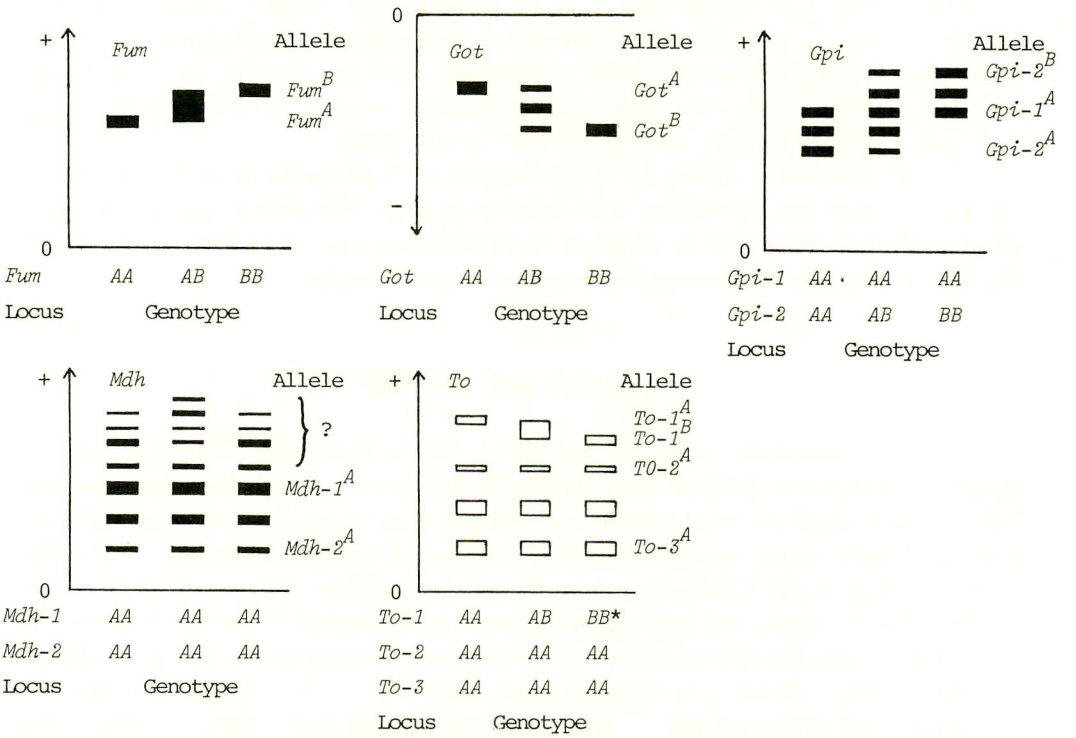

Fig. 1. Electrophoretic patterns observed in five enzymes of *N. pompilius*.
*Postulated genotype

(1) Fumarase (*Fum*)

One locus with two alleles was scored for *Fum*. The sample from the Philippines was fixed for Fum^A allele and the sample from Fiji had Fum^A and Fum^B alleles.

(2) Glutamate-oxaloacetate transaminase (*Got*)

One locus with two alleles was scored for *Got*, but the two samples had very different allele frequencies, with Got^A in high frequency in the sample from the Philippines and Got^B in high frequency in the sample from Fiji.

(3) Glucosephosphate isomerase (*Gpi-1* and *Gpi-2*)

Two *Gpi* loci were observed in the sample from the Philippines, but not observed in the sample from Fiji because of the deactivation of this enzyme during transportation. In the sample from the Philippines, *Gpi-1* was fixed for $Gpi\text{-}1^A$ allele and *Gpi-2* was polymorphic for two alleles.

Table 2. Allele frequencies at 9 loci in *N. pompilius*. N is the number of individuals sampled.

Locus	Allele	Locality	
		Philippines	Fiji
Fum	Fum^A	1.00	0.70
	Fum^B		0.30
	(N)	(17)	(30)
Got	Got^A	0.74	0.19
	Got^B	0.26	0.81
	(N)	(17)	(8)
Gpi-1	$Gpi\text{-}1^A$	1.00	——
	(N)	(17)	——
Gpi-2	$Gpi\text{-}2^A$	0.82	——
	$Gpi\text{-}2^B$	0.18	——
	(N)	(17)	——
Mdh-1	$Mdh\text{-}1^A$	1.00	1.00
	(N)	(17)	(36)
Mdh-2	$Mdh\text{-}2^A$	1.00	1.00
	(N)	(17)	(36)
To-1	$To\text{-}1^A$	1.00	0.99
	$To\text{-}1^B$		0.01
	(N)	(17)	(35)
To-2	$To\text{-}2^A$	1.00	1.00
	(N)	(17)	(35)
To-3	$To\text{-}3^A$	1.00	1.00
	(N)	(17)	(35)

(4) Malate dehydrogenase (*Mdh-1* and *Mdh-2*)

Three major zones of *Mdh* activity were observed in starch gels. We have scored them as two loci (*Mdh-1* and *Mdh-2*) and the intermediate heteropolymer zone *Mdh-1/2*. *Mdh-1* and *Mdh-2* were fixed for $Mdh\text{-}1^A$ and $Mdh\text{-}2^A$ alleles in all specimens, respectively.

(5) Tetrazolium oxidase (*To-1*, *To-2* and *To-3*)

Four zones of *To* activity were observed in starch gels which we have scored as three loci (*To-1*, *To-2* and *To-3*) and the intermediate heteropolymer zone *To-2/3*. At *To-1* locus, the two samples had the same allele either fixed or in high frequency. *To-2* and *To-3* were fixed for $To\text{-}2^A$ and $To\text{-}3^A$ alleles in all specimens, respectively.

B. Heterogeneity tests between the two samples collected from the Philippines and Fiji

Statistically, the two samples collected from the Philippines and Fiji showed significant differences in allele frequencies at *Fum* and *Got* loci (*Fum* : $X^2=12.7$, $P<0.001$; *Got* : $X^2=13.4$, $P<0.001$). These results suggest that there may be genetic differentiation and little gene flow between the two localities.

To clarify the genetic population structure of *N. pompilius*, extensive sampling and analysis of other enzymes are required.

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