

Allozyme-based Genetic Variation in Crossbreds Produced from Three Thai *Macrobrachium rosenbergii* Stocks⁸

Panom K. Sodsuk, Supattra Uraiwan, Srirat Sodsuk

Aquatic Animal Genetics Research and Development Institute, Department of Fisheries,
Khong Ha, Khlong Luang, Pathumthani 12120, THAILAND.

Introduction

Molecular technology at enzyme/protein level known as “allozyme marker” is a widely accepted powerful technique to study genetic variation (Ward and Grewe, 1995) as well as intraspecific population studies (Sodsuk, 1996; Sodsuk and Sodsuk, 1998a & 1998b; Sodsuk *et al.*, 2001). Since the allozyme technique can be readily applied, it has become a basic tool for the evaluation of genetic variation in aquaculture stocks. This study aims to: (1) evaluate genetic variation (measured as per locus averages of observable heterozygosities and number of alleles) of nine crosses from three *Macrobrachium rosenbergii* stocks (genetically improved AAGRDI, WILD, FARM); (2) apply polymorphic allozyme markers in the evaluation; (3) compare genetic variation among the nine crosses to determine genetic stock differences; and (4) use genetic variability and performance evaluation information in choosing the best cross for the conduct of a selective breeding program in specific farm environments.

Materials and Methods

Sample Analysis

About 40-60 individuals from both sexes of each of three stocks (the AAGRDI, wild, and private farm) and each progeny population of nine crosses were sampled. Pleopods were collected and placed in separate microtubes. The samples were kept in a -70 °C freezer prior to allozyme marker analysis. The preserved samples were electrophoretically analysed at 19-25 allozyme loci (Sodsuk *et al.*, 2005) following the protocol described by Sodsuk and Sodsuk (1998b).

Data Analysis

All allozyme data from the laboratory analyses were collected and calculated as per locus averages of heterozygosities (H) and number of alleles (NoA) for the evaluation of genetic variation. Data were analysed using BIOSYS release 1.7 of Swofford and Selander (1989). Genetic variation in the nine crosses, measured as per locus averages of heterozygosities and number of alleles (see Tables in appendix) were statistically compared following the methods of Sokal and Rohlf (1981) and Ward *et al.* (1994). This procedure was done using a statistical software known as SYSTAT of Wilkinson *et al.* (1992).

Results and Discussion

Genetic variation data (evaluated from per locus averages of heterozygosities and number of alleles) of the three stocks used in the parental crosses and all the nine crossbred stocks, are shown respectively in Tables 1 and 2. There were no significant differences among the three stocks as well as the nine crossbred stocks, based on heterozygosities and number of alleles. The heterozygosity levels and number of alleles both in the three stocks (H = 0.023-0.043, NoA = 1.20-1.44) and in the nine crosses (H = 0.010-0.042, NoA = 1.11-1.53) were similar to the natural stocks (H = 0.027-0.036, NoA = 1.29-1.33) earlier studied by Sodsuk and Sodsuk (1998b).

⁸based on the paper presented during the 3rd Roundtable Discussion



Table 1. Per locus averages of heterozygosities (H) and number of alleles (NoA) of the three initial stocks

Stock	H	NoA
AAGRDI	0.043 (± 0.018) ^A	1.36 (± 0.11) ^a
Wild	0.023 (± 0.014) ^A	1.20 (± 0.10) ^a
Farm	0.036 (± 0.016) ^A	1.44 (± 0.13) ^a

Values in parentheses are standard errors (\pm S.E.)

Same superscripts in the same column means no significant differences ($p > 0.05$)

Table 2. Per locus averages of heterozygosities (H) and number of alleles (NoA) in all nine crosses

	Cross (male x female)	H (Average \pm S.E.)	A (Average \pm S.E.)
T1	(Wild x AAGRDI)	0.011 (± 0.008) ^A	1.11 (± 0.07) ^a
T2	(AAGRDI x Wild)	0.042 (± 0.027) ^A	1.26 (± 0.10) ^a
T3	(AAGRDI x Farm)	0.010 (± 0.007) ^A	1.16 (± 0.09) ^a
T4	(Farm x AAGRDI)	0.016 (± 0.007) ^A	1.32 (± 0.13) ^a
T5	(Wild x Farm)	0.030 (± 0.010) ^A	1.53 (± 0.14) ^a
T6	(Farm x Wild)	0.026 (± 0.013) ^A	1.26 (± 0.13) ^a
T7	(Farm x Farm)	0.024 (± 0.010) ^A	1.37 (± 0.11) ^a
T8	(Wild x Wild)	0.018 (± 0.009) ^A	1.21 (± 0.10) ^a
T9	(AAGRDI x AAGRDI)	0.015 (± 0.009) ^A	1.16 (± 0.09) ^a

Same superscripts in the same column means no significant differences ($p > 0.05$)

Table 3 shows the genetic information of the resulting heterozygosities and number of alleles, together with those resulting from growth performance (Uraiwan *et al.*, 2005). This genetically informative table is very helpful for choosing the best breeding-pair for further selection program in an appropriate area.

Conclusion

Genetic variation (measured as per locus averages of heterozygosities and the number of alleles), in the three initial stocks and all nine crosses showed no significant differences among the stocks. Genetic variability information generated by this study and the growth performance data from the study of Uraiwan *et al.* (2005), would help in choosing the best cross for selective breeding in each environment.



Table 3. Genetic variability (expressed as heterozygosities H, number of alleles A), growth performance indicators and % heterosis of all crosses in four different areas

Environment (months)	Mated Pair	Cross	Sodsuk (<i>et al.</i>) 2005		Uraivan <i>et al.</i> (2005)			
			H	A	Performances		% heterosis	
					Length	Weight	Length	Weight
Uttaradit (5)	AAGRDI x Wild	T1	0.011 ^A	1.11 ^a	12.982	24.354	2.28	15.47
		T2	0.042 ^A	1.26 ^a	12.671	24.449		
	AAGRDI x Farm	T3*	0.010 ^A	1.16 ^a	13.140	23.977*	20.28*	9.16*
		T4*	0.016 ^A	1.32 ^a	13.822*	22.083		
	Wild x Farm	T5	0.030 ^A	1.53 ^a	12.500	22.908	1.01	3.13
		T6	0.026 ^A	1.26 ^a	12.002	20.681		
	Farm x Farm	T7	0.024 ^A	1.37 ^a	12.212	21.965	-	-
		Wild x Wild	T8	0.018 ^A	1.21 ^a	12.044	22.035	-
	AAGRDI x AAGRDI	T9	0.015 ^A	1.16 ^a	12.267	20.230	-	-
Buriram (4)	AAGRDI x Wild	T1	0.011 ^A	1.11 ^a	10.430	17.220	0.61	0.48
		T2	0.042 ^A	1.26 ^a	10.783	16.140		
	AAGRDI x Farm	T3*	0.010 ^A	1.16 ^a	11.061*	20.709*	1.58*	19.86*
		T4	0.016 ^A	1.32 ^a	10.447	16.710		
	Wild x Farm	T5	0.030 ^A	1.53 ^a	10.618	17.740	-2.85	-0.30
		T6	0.026 ^A	1.26 ^a	10.049	15.040		
	Farm x Farm	T7	0.024 ^A	1.37 ^a	10.687	15.450	-	-
		Wild x Wild	T8	0.018 ^A	1.21 ^a	10.589	17.430	-
	AAGRDI x AAGRDI	T9	0.015 ^A	1.16 ^a	10.496	15.770	-	-
Pathumtani (2)	AAGRDI x Wild	T1	0.011 ^A	1.11 ^a	7.516	3.905	-6.67	-23.61
		T2	0.042 ^A	1.26 ^a	7.244	3.588		
	AAGRDI x Farm	T3	0.010 ^A	1.16 ^a	7.922	4.963	1.97	18.39
		T4	0.016 ^A	1.32 ^a	6.706	3.156		
	Wild x Farm	T5	0.030 ^A	1.53 ^a	7.628	4.546	1.66	6.96
		T6*	0.026 ^A	1.26 ^a	8.329*	5.244*		
	Farm x farm	T7	0.024 ^A	1.37 ^a	7.113	3.299	-	-
		Wild x Wild	T8*	0.018 ^A	1.21 ^a	8.583*	5.854*	-
	AAGRDI x AAGRDI	T9	0.015 ^A	1.16 ^a	7.232	3.559	-	-
Chumphon (2)	AAGRDI x Wild	T1*	0.011 ^A	1.11 ^a	8.122*	4.681*	4.54*	14.48*
		T2	0.042 ^A	1.26 ^a	7.576	4.036		
	AAGRDI x Farm	T3	0.010 ^A	1.16 ^a	7.30	3.69	2.74	13.21
		T4	0.016 ^A	1.32 ^a	7.506	4.102		
	Wild x Farm	T5	0.030 ^A	1.53 ^a	7.456	3.634	-1.35	-1.98
		T6	0.026 ^A	1.26 ^a	7.210	3.502		
	Farm x Farm	T7	0.024 ^A	1.37 ^a	7.131	3.274	-	-
		Wild x Wild	T8	0.018 ^A	1.21 ^a	7.736	4.006	-
	AAGRDI x AAGRDI	T9	0.015 ^A	1.16 ^a	7.280	3.609	-	-

Asterisks (*) refer to the best crosses with good genetic attributes for rearing in specific environments



References

- Sodsuk PK, S Leesa-nga, S Sodsuk, P Tevaratmaneeikul, K Komenpririn. 2001. Genetic diversity of a freshwater badgrid catfish (*Hemibagrus nemurus*) present in Thailand. Technical Paper No. 4/2001
- National Aquaculture Genetics Research Institute, Department of Fisheries, Ministry of Agriculture and Cooperatives. 28 pp
- Sodsuk PK, S Uraiwan, S Sodsuk. 2005. Allozyme marker based comparison on genetic variation among *Macrobrachium rosenbergii* populations produced from a cross-breeding system of three different stocks in Thailand. A paper presented during the "3rd Round Table Discussion on the Development of Genetically Improved Strain of *Macrobrachium*," 3-4 December 2005. Bangkok, Thailand. SEAFDEC Aquaculture Department
- Sodsuk S. 1996. Genetic differentiation and population structure of *Penaeus monodon* in Thailand. Technical Paper No. 12. National Aquaculture Genetics Research Institute, Department of Fisheries, Ministry of Agriculture and Cooperatives. 19 pp
- Sodsuk S, PK Sodsuk. 1998a. Genetic diversity of banana shrimp from three locations in Thailand. Technical Paper No. 17/1998. National Aquaculture Genetics Research Institute, Department of Fisheries, Ministry of Agriculture and Cooperatives. 45 pp
- Sodsuk S, PK Sodsuk. 1998b. Genetic diversity of giant freshwater prawn from three locations in Thailand. Technical Paper No. 18/1998. National Aquaculture Genetics Research Institute, Department of Fisheries, Ministry of Agriculture and Cooperatives. 40 pp
- Sokal RR, FJ Rohlf. 1981. Biometry, 2nd ed. WH. Freeman, San Francisco. 859 p
- Swofford DL, RB Selander. 1989. BIOSYS-1: A Computer Program for the Analysis of Allelic Variation in Population Genetics and Biochemical Systematics. DL Swofford, Illinois Natural History Survey. 43 p
- Uraiwan S, *et al.* 2005. Progress report of the project "Selective Breeding for Genetic Improvement of *Macrobrachium rosenbergii*" presented during "the 3rd Round Table Discussion on the Development of Genetically Improved Strain of *Macrobrachium*," 3-4 December 2005. Bangkok, Thailand
- Ward RD, PM Grewe. 1995. Appraisal of molecular genetic techniques in fisheries. In GR Carvalho, TJ Pitcher (eds). pp. 29-54. Molecular Genetics in Fisheries. Chapman and Hall, London
- Ward RD, M Woodwark, DOF, Skibinski. 1994. A comparison of genetic diversity levels in marine, freshwater, and anadromous fishes. *Journal of Fish Biology* 44: 213-232
- Wilkinson L, M Hill, JP Welna, GK Birkenbeuel. 1992. SYSTAT for Windows Statistics, Version 5 Edition. Evanston IL: SYSTAT, Inc