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# STATUS OF Macrobrachium rosenbergii INDUSTRY

# Production, Value and Area

Giant freshwater prawn (*Macrobrachum rosenbergii*) is one of the most important economic species in Thailand as well as in many Southeast Asian countries. It is highly demanded as food for the Thais that lead to its gradual over-catching from the natural waters every year. Thus, domestication experiments on the *Macrobrachium* have been conducted by the Department of Fisheries since 1956 to increase prawn production (Sidthimunka and Bhukaswan, 1982). This resulted in the nationwide extension of its culture technology to a number of commercial giant freshwater prawn culture companies. Now this species has become one of the economic commodities in the country's aquaculture industry. Recently, statistics report showed that the total prawn culture in 2002 increased by 38% from the year 1996, while the total value in the same year increased by 89% from the year 1996. The statistics also showed that in 1996 and 2002, the total production were 7200 and 10,000 mt, respectively, valued at 596.3 and 1,117.6 million Thai Baht, respectively (Department of Fisheries and Suwannatos, 2003). The market price per kg of freshwater prawn, which varies according to the prawn sizes, has been increasing since 1989 (Table 1). In 1997, the large, medium and small sizes increased by 76%, 123% and 81% from those in year 1989, respectively (Table 1).

Table 1. Price of cultured giant freshwater prawn between 1989-1997 (Department of Fisheries, 2001)

	Price (In Thai Baht)								
Size	1989	1990	1991	1992	1993	1994	1995	1996	1997
Large	207.42	233.92	256.01	272.92	278.25	302.83	290.08	325.58	365.92
Medium	93.15	129.17	132.63	124.17	33.24	163.47	170.67	186.17	207.33
Small	49.97	62.51	63.08	66.34	65.62	69.65	72.83	71.03	90.83

The freshwater prawn annual value and the annual production have increased during the past four years as shown in Figure 1. Most of the culture areas are located in the central part of the country. The total area of giant freshwater prawn culture in 2000 was 3926.9 ha, 69 % of which are in six provinces, namely: Ratchaburi (56%), Nakornpathom (13%), Supanburi (18%), Ayuthaya (3%), Karnjanaburi (6%), and Chachanksoa (3%).

# **Commercial Hatcheries**

Domestication of the giant freshwater prawns in Thailand was started in 1966 at the Songkhla and Bangkhen Fisheries Stations while small commercial hatcheries started to operate in 1973 (New, 1982 and Suwannatos, 2003). Giant freshwater prawn hatchery practices have been developed since 1966, when the early researches concentrated on larval rearing until it became a routine, then later on the grow-out culture. Since water quality condition became a critical factor in larval rearing, the optimum levels of this factor were established including salinity from 12 to 15 ppt, temperature from 27 to 31°C, dissolved oxygen from 3 to 5 mg/l, pH from 7 to 8, and ammonia which should not be more than 0.1 mg/l (Colt and Armstrong, 1981 and Suwannatos, 2003). Seed quality in commercial hatcheries also depended on many factors, namely: water quality, parent stock, female parent maturity condition and hatchery management.

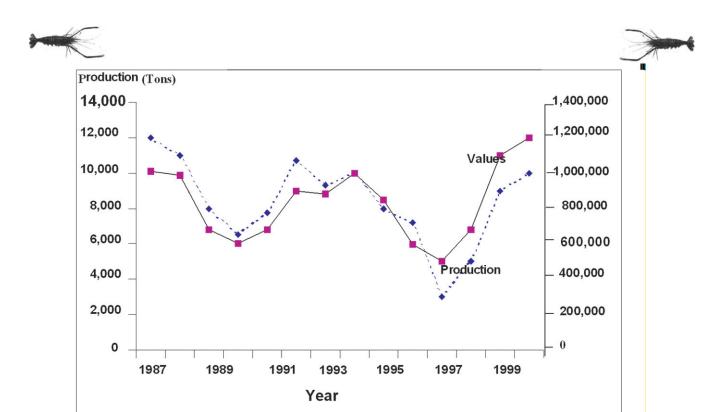


Figure 1. Production and values of cultured *Macrobrachium rosenbergii* in 1987-2000 (Department of Fisheries, 2001)



A typical small-scale freshwater prawn hatchery in Thailand

Suwannatos (2003) reviewed the giant freshwater prawn rearing practices in Thailand, and he concluded that there were four types of larval hatchery management. Each system had different advantages and disadvantages to the farmers. However, a number of researches have been conducted to develop these rearing systems to provide good quality seeds to the farmers. Thus, the farmers have the choice on the type of system they would adopt based on their experience and the hatchery conditions.

#### Clear water system

Simple and less expensive, this system could be applied in a small hatchery such as a backyard hatchery because there is no requirement for ponds. However, the hatchery manager should monitor closely the water quality to make sure that these are above the optimum levels. The pond size should be between 1-3 m<sup>2</sup> and 1.0 m deep.





### Greenwater system

This system was modified from a larval rearing system developed in Hawaii. A large number of quality seeds can be produced using this system by rearing the larvae in greenwater with the phytoplankton, *Chorella* spp. The phytoplankton controls the water quality at an optimum pH and also the disease problem. However, this system requires a number of ponds and the rearing period is longer (more than 30 days) compared with that of the other systems.

### Recirculation system

This system is preferred for hatcheries that are far from the sea because it requires less seawater, and the environmental conditions are easy to control. However, it is more costly than the other systems. Some researches that dealt with the development of this system (Suwannatos, 2003), included the use of water treated with small weed organisms and air circulation, with the water to be continuously reused. Therefore, diseases and use of chemicals could be easily controlled. For this system, the hatchery manager should be well trained on water quality analysis as well as on nutrition and disease control. Rearing period is also longer (more than 30 days) compared with that of the clear water system.

### Earthen pond system

A group of biologists at the Phetchaburi Coastral Aquaculture Station, Department of Fisheries developed a technology for rearing the larvae of giant freshwater prawn in earthen ponds (Tunsutapanich *et.al.*, 1994). In this system, water quality in the earthen pond is maintained to have ecological balance without any water change. The pond is installed with air blower for consistent oxygen supply. Water chemical conditions are observed regularly and adjusted to suitable levels for the balanced ecological system in the pond.

The larvae are fed with natural zooplankton and with *Artemia* nauplii and adult. Although the system gives high survival rate of juveniles, its rearing period is longer than that of the other systems. The total production in this system is 5.1 million juveniles per 0.16 ha with an average survival rate of 88% (Tunsutapanich *et. al.*, 1994). For this system, the hatchery manager should observe the water quality regularly and maintain the ecological balance and food chain within the pond.

# **GOOD QUALITY SEEDS**

Good quality seeds should result in high survival rate and fast growing juvenile prawn. The above hatchery systems would be able to provide good quality seed supply to the *Macrobrachium* industry provided the following measures are followed:

- 1. Suitable socking density for larval rearing should be 20-40 post larvae/l
- 2. Temperature during the rearing period should range from 28 to 30°C
- 3. Female parents should be clean with good maturity condition
- 4. Antibiotics should never be used
- 5. All equipment should be disinfected every after use
- 6. Good quality food such as Artemia, boiled eggs and Pollock, should be used













Larval rearing of giant freshwater prawn in earthenponds developed at the Phetchaburi Coastal Aquaculture Station, Department of Fisheries, Thailand

#### **GROW-OUT CULTURE**

*Macrobrachium* culture in the country is now facing a number of problems such as slow growth, lack of appropriate broodstock management and disease occurrence. To counteract these problems, commercial *Macrobrachium* farms developed a new improved strain with new farming management. The strain has been initially introduced to the farmers, and the private hatcheries have developed their own selective breeding programs for such strain.

*Macrobrachium* farming in Thailand has been classified into two types: the traditional farms growing the local strain, and the *Macrobrachium* farms using a new strain. Traditional farms usually consist of small number of earthen ponds 0.32 –0.96 ha adopting a stocking density of 5 to 20 juveniles/m². The farmers prepare their own feeds. During the first month, the prawns are fed twice a day at a feeding rate of 30-40% of the body weight. During the third month of rearing, the feeding rate is gradually reduced to 5%, and in the following month, to 3%. In the sixth month, large size prawns are seined out for sale while the small ones are left in the culture pond. The market size prawns should be between 50 to 100 g/pc (Tookwinas, 2002).

One farm which uses the new strain of *Macrobrachium* is the Kasetsombuond Farm in Supanburi Province. The owner, Mr. Supon Sovanapreecha, has been operating this farm using the new strain of *Macrobrachium* for three years (interviewed in Thai Fisheries Gazette, 2002). The juveniles are reared in the nursing pond at the stocking density of 75 pc/m² for two months, after which these are stocked in the grow-out pond at 7-15 pc/m². After four and a half months, the female prawns with an average size of 25-33 g are culled for sale. On the sixth month, the large males (100-125 g) are harvested and sold. The total production of the first generation is usually about 3750 kg/ha with a sex ratio of 80% males to 20% females. However, production usually decreases by 10% in the second and third generations (male size: 83 g). Therefore, selective breeding program to improve the growth of the *Macrobracium* domesticated strain has been recognized as an urgent concern for the freshwater prawn industry.





#### **FUTURE PLAN AND PROSPECTS**

Recently, the National Thailand Research Fund in cooperation with the Department of Fisheries conducted a workshop on the "Participation of increased effort in Macrbrachium industry" on 20 August 2003. The main objective of the workshop was to discuss the aspects of increased efficiency in producing *Macrobrachium rosenbergii* as a new premium aquaculture commodity. Problems and plans on the different aspects were discussed including, nursery, grow-out culture, nutrition, diseases and genetics. The Department of Fisheries proposed to develop a selective breeding program to improve the economic traits of the *Macrobrachium rosenbergii* cultured in Thailand. In addition, researches to develop a practical farm management protocol for each part of the country were also planned. As *Macrobrachium rosenbergii* is now becoming a premium aquaculture commodity, good production process from the farm to the table should be established. Therefore, the code of conduct (CoC) is planned to be developed similar to that of the country's marine shrimp culture. The planned guidelines of the CoC for Macrobrachium should include the following 11 aspects (Tookwinas et.al., 2002 and Tookwinas, 2002):

- (1) suitable site selection
- (2) general pond management
- (3) stocking density
- (4) feed management
- (5) prawn health management
- (6) therapeutic agent and other chemicals
- (7) effluents and solid waste management
- (8) harvesting and selling
- (9) social responsibility
- (10) farmer association and education
- (11) data collection.

#### MACROBRACHIUM SPECIES THAT HAVE BEEN IDENTIFIED IN THAILAND

All Macrobrachium species are classified in the Family Palaemonidae Rafinesque, 1815, Subfamily Palaemoninae Rafinesque, 1815. The following nineteen *Macrobrachium* species have been found and identified in Thailand:

Species	Freshwater	Brackishwater
Macrobrachium amplimanus (Cai and Dang, 1999)	Yes	No
Macrobrachium dienbienphuense (Dang and Nguyen, 1972)	Yes	No
Macrobrachium equidens (Dana, 1825)	No	Yes
Macrobrachium eriocheirum (Dai, 1984)	Yes	No
Macrobrachium esculentum (Thallwitz, 1891)	Yes	No
Macrobrachium hirsutimanus (Tiwari, 1952)	Yes	No
Macrobrachium idae (Heller, 1862)	No	Yes
Macrobrachium lanchesteri (De Man, 1911)	Yes	No
Macrobrachium lar (Fabricius, 1793)	No	Yes
Macrobrachium latidactylus (Thallwitz, 1891)	No	Yes
Macrobrachium mirabile (Kemp, 1917)	No	Yes
Macrobrachium mieni (Dang, 19750)	Yes	No
Macrobrachium neglectus (De Man, 1905)	No	Yes
Macrobrachium niphanae (Shokita and Takeda, 19890)	Yes	No
Macrobrachium rosenbergii dacqueti (Sunier, 1925)	Yes	Yes
Macrobrachium sintangense (De Man, 1898)	Yes	Yes
Macrobrachium sirindhorn (Naiyanetr, 2001)	Yes	No
Macrobrachium yui (Holthuis, 1950)	Yes	No
Macrobrachium sp. (Cai, Naiyanetr and Ng, in press)	Yes	No





Of the nineteen species, the following five species are the most economically important species for Thailand:

# 1. Macrobrachium dienbienphuense (Fig. 2 & 3)

*M. dienbienphuense* has been found in northeast of Thailand, distributed in the main rivers of Maekhong, Chi, Moon and their branches. The species has moderate size, the biggest of which is about 6-7 cm. The prawns are being sold in the markets along the Maekhong and Moon rivers, particularly in Ubon Ratchathani Province. There is no developed culture technology for this species yet, may be because the prawn can still be captured in good quantities throughout the year from the natural waters.

# 2. Macrobrachium niphanae (Fig. 4 & 5)

*M. niphanae* has attractive characteristics with red brown spots lining around each of all the five pairs of walking legs. This prawn could pass as an ornamental aquatic species of economic importance. The species could be a subject for study for its biological details in order that an appropriate aquaculture system could be developed. This species is not well known to some people because it is found only in waterfalls and streams and very rarely in other watercourses and rivers.

# 3. Macrobrachium lanchesteri (Fig. 6 & 7)

*M. lanchesteri* is very similar to *M. niphanae* in terms of size and morphology. The species is found in almost every inland waterbody and is very often found together with *M. niphanae*. Because of its abundance, *M. lanchesteri* has become important to the local people in the rural areas. Many people like to eat the prawn even though its size is very small. In some provinces in northeast Thailand, culture of the prawn is usually done with fishes together in the one pond to earn more money.

The following table compares the morphological characteristics of *M. niphanae* and *M. lanchesteri*.

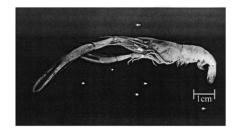
Morphological characteristics	M. niphanae	M. lanchesteri
1. Rostral teeth	2-3 (7-13) / 2-3	1 (7-10) / 3-4
2. Five pairs ofs walking leg	With red brown spots lining around each one of all walking legs	Clear, without red brown spot
3. Rostrum	Longer than antennular peduncle but shorter than scaphocerite	Slender and same length as antennular peduncle
4. Position of hepatic spine	Between the first and second ones of the upper rostral teeth	Behind the first one of the upper rostral teeth
5. The second pair of walking legs	With little short hair appearing on the carpus and merus	With little short hair appearing only on the fingers of mature males

### 4. Macrobrachium sintangense (Fig. 8 & 9)

*M. sintangense* is a species morphologically similar to *M. rosenbergii*. Its size is about the same as the immature *M. rosenbergii*. The biggest size of *M. sintangense* is about 8-9 cm. The shrimp is also popular with the people in the northeastern Thailand mainly because of its moderately large size which is just slightly smaller to *M. rosenbergii*. People in some areas call this prawn "Kung Kam Kram" which is the Thai term for *M. rosenbergii*. The prawn is sold in many provinces along the Maekhong and Moon rivers. Due to the relative economic importance of the species, study of its biology and life cycle should be conducted, so that appropriate aquaculture system could be developed and the species could be introduced to the local fish-farmers.







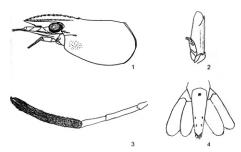


Figure 2. Macrobrachium dienbienphuense (Dang and Nguyen, 1972:)
1. Carapace and rostrum, 2. Scaphocerite
3. The 2<sup>nd</sup> right walking-leg, 4. Telson and

uropod



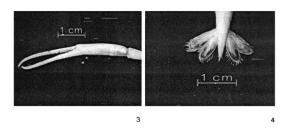


Figure 3. Morphological characters of M. dienbienphuense
1. Rostrum, 2. Scaphocerite
3. The 2<sup>nd</sup> right walking-leg, 4. Telson and uropod

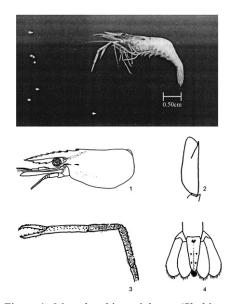


Figure 4. Macrobrachium niphanae (Shokita and Takeda, 1989)
1. Carapace and rostrum, 2. Scaphocerite
3. The 2<sup>nd</sup> right walking-leg, 4. Telson and

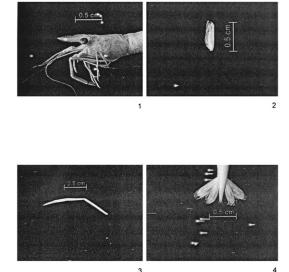
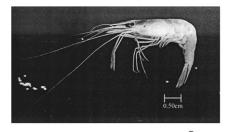


Figure 5. Morphological characters of M. niphanae 1. Rostrum, 2. Scaphocerite 3. The  $2^{\rm nd}$  right walking-leg, 4. Telson and uropod

uropod







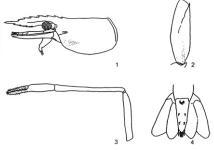
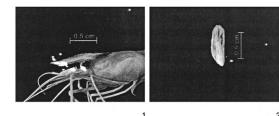


Figure 6. Macrobrachium lanchesteri (DeMan, 1911)

Carapace and rostrum,
 Scaphocerite
 The 2<sup>nd</sup> right walking-leg,
 Telson and uropod



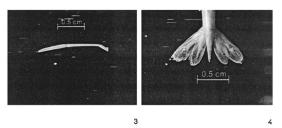
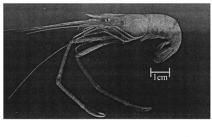


Figure 7. Morphological characters of M. lanchesteri 1.Rostrum, 2. Scaphocerite 3. The 2<sup>nd</sup> right walking-leg, 4. Telson and uropod



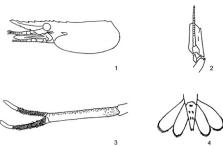
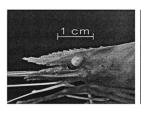


Figure 8. Macrobrachium sintangense (DeMan, 1898) 1. Carapace and rostrum, 2. Scaphocerite

3. The 2<sup>nd</sup> right walking-leg, 4. Telson and uropod





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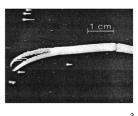




Figure 9. Morphological characters of M. sintangense 1.Rostrum, 2. Scaphocerite 3. The 2<sup>nd</sup> right walking-leg, 4. Telson and uropod





# 5. Macrobrachium rosenbergii dacqueti (Fig. 10 & 11)

*M. rosenbergii* has been famously known as the "giant freshwater prawn" since 1958. It comprises two subspecies identified by the differences in some morphological characteristics and in particular their geographical distribution. One subspecies found in the Papuasia area between Papua New Guinea and Australia, and the areas around the Philippines, is *M. rosenbergii rosenbergii* (De Man, 1879). The other species, distributed in the Indo-west Pacific from Indonesia to India including Thailand, is *M. rosenbergii dacqueti* (Sunier, 1925).

The following table compares the different characteristics of *M. rosenbergii dacqueti* and *M. rosenbergii rosenbergii:* 

_			
	Characteristics	M. rosenbergii dacqueti	M. rosenbergii rosenbergii
1.	Rostrum	The part behind eye is	The part behind eye is straight
		convex and longer than	and as long as or shorter than
		scaphocerite	scaphocerite
2.	The second pair of	With large spines scattering,	With small spines scattering,
	walking legs	the end of the spines being	the end of the spines being
		straight	horn-like
3.	Distribution	India, Burma, Thailand,	Papuasia and the Philippines
		Malaysia and Indonesia	

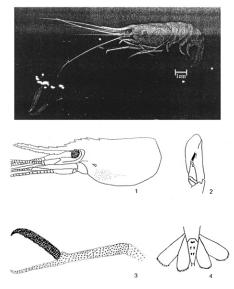


Figure 10. Macrobrachium rosenbergii dacqueti (Sunier, 1925)

- 1. Carapace and rostrum, 2. Scaphocerite
- 3. The  $2^{nd}$  right walking-leg, 4. Telson and uropod









Figure 11. Morphological characters of M. rosenbergii dacqueti

- 1. Rostrum, 2. Scaphocerite
- 3. The 2<sup>nd</sup> right walking-leg, 4. Telson and uropod





In the past, the distribution of *M. rosenbergii* in Thailand was reported only in the brackishwater connected to the sea. But now it has been found in many freshwater bodies of Thailand, especially in places where large amount of the prawn have been released by the DOF (Department of Fisheries of Thailand) over the past decades. The species is highly economically important, particularly the big ones which are very expensive and popular as premium food. Many farms in Thailand, particularly in the central area, have done breeding and culture activities for this species as an industry for quite some time. However, the DOF of Thailand is still trying to improve the prawn quality including seeds, broodstock and products to come up with the best prawn ever produced in the world.

# R & D EFFORTS ON QUALITY IMPROVEMENT OF M. rosenbergii dacqueti

The following gives a summary of the R&D efforts done in Thailand on *M. rosenbergii*:

#### (1) CYTOGENETIC INFORMATION STUDY

Wanitchanon, A. and J. Phetsiri. 2000. A chromosome study of *Macrobrachium rosenbergii* from Thailand. Research Report, Taksin University, Songkhla, Thailand

A chromosome study was conducted on the freshwater prawn, *Macrobrachium rosenbergii* found in Thailand. Fifty specimens of male and female, average 17.00 cm, had a second pair of periopods removed. The specimens were kept in a small aquarium for one week to induce the regeneration blastema. Spreads of metaphase chromosome were prepared from the regenerating tissue. Chromosome number was counted based on 50 cells. The study found that the diploid chromosome number was 118. The karyotype consisted of 45 metacentric and submetacentric pairs and 14 telocentric and acrocentric pairs.

# (2) GENETIC DIFFERENTIATION AND POPULATION STRUCTURE STUDIES

Yaitavorn, P. 1989. Mitochondrial DNA variation in giant freshwater prawn (*Macrobrachium rosenbergii* de Man). M.Sc. Thesis, Chulalongkorn Univ., Bangkok, Thailand

A study on genetic variation in natural population of giant freshwater prawn *Macrobrachium rosenbergii* de Man was undertaken to identify the races of the prawn in different locations of Thailand. The study was based on the analysis of the mitochondrial DNA (mtDNA) variation. The mtDNA was isolated from the hepatopancreas of prawns and cut with restriction enzyme, *Sau3A* I, to generate DNA fragments ranging from 0.2 to 2.0 kb. These DNA fragments were cloned into vector pUC12 at *BamH* I site and transformed into *E. coli* strain JM 107. After colony hybridization and Southern blot hybridization, fifty-one recombinant clones were obtained. These clones were further selected for strongly hybridized signal with a-<sup>32</sup>P dATP labelled mtDNA, and used as probes to analyze mtDNA variation in prawns. The recombinant DNA No.1, which inserted a fragment of about 1.1 kb. and had restriction endonuclease sites for *Rsa* I, *Hha* I, *Hae* III and *BstU* I, was able to distinguish the prawns from the two geograchic populations. It showed significant difference in restriction fragment length polymorphism (RFLP) pattern of mtDNA between prawn from two rivers, Kraburi in the south and Bangpakong in the central, by Southern blot hybridization with the recombinant DNA No.1. The prawns from Bangpakong showed strong discrete band at 1.1 kb. but those from Kraburi showed the band at about 0.7 kb. By using this clone, the RFLP patterns of Bangpakong River and that from Kung Kam Thong Farm, both of which are in central Thailand, were found similar.

Sodsuk, S. and P.K. Sodsuk. 1998. Genetic diversity of giant freshwater prawn from three locations of Thailand. Technical Paper No. 18/1998. National Aquaculture Genetics Research Institute, Department of Fisheries, Thailand

Giant freshwater prawn (*Macrobrachium rosenbergii* de Man) from three locations of Thailand (Chachoengsao, Surat Thani and Songkhla) was identified using allozyme electrophoresis. A total of 24 enzyme loci were detected





from pleopod, muscle and hepatopancreas. Three loci,  $GPI^*$ ,  $MPI^*$  and  $PGM-1^*$ , were found to be polymorphic ( $P_{0.95}=0.125$ ). No significant differences from Hardy-Weinberg equilibrium were observed within any single population. Mean expected heterozygosity ( $H_c=0.031\pm0.018$ ) for the species was found to be relatively low. A number of genetic differences evaluated, inbreeding coefficients (F-statistics), polymorphic loci differences within each pair of populations and among all populations, genetic distances between populations and dendrograms constructed using genetic distances, showed that all three populations studied were the same single population based on the technique at allozyme level.

# Vanavichit, A. et al. (pers. comm.) Agricultural Genetic Engineering and Biotechnology Center, Kasetsart Univ. Research and Development Institute, Kamphaengsaen Campus, Nakhornpathom, Thailand

Genetic differentiation among populations of *Macrobrachium rosenbergii* de Man was carried out. The prawn specimens from six localities used in the study were 68 individuals from Bangpakong River, 83 individuals from Nakhorn Nayok River, 55 individuals from Tapi River, 41 individuals from the upper part of Songkhla Lake, 69 individuals from Kraburi River (a border river between Thailand and Myanmar) and 116 individuals from Yaephew River in Myanmar. The study was based on mtDNA analysis focusing the 12S rDNA and 16S rDNA genes, that had been cut with four restriction enzymes, *Alu* I, *Dra* I, *Hinf* I and *Tru9* I. Based on genetic distances resulting from the study, two major populations of the prawns were obtained. One major population consisted of prawns from all water-bodies in Thailand, the Bangpakong, Nakhorn Nayok, Tapi and Songkhla, and another 63.46% of the prawns from Kraburi together. The other major population included 65.46% of the prawns from the Yaephew in Myanmar. Genetic distance between the two major populations was 0.931. Further to this, the remaining prawns from the Kraburi and Yaephew were mixed together as another intermediate group.

#### (3) SELECTIVE BREEDING PROGRAMS

# Meewan, M. 1991. Morphological inheritability and growth of giant freshwater prawns. M.Sc. Thesis, Asian Institute of Technology (AIT), Thailand

An experiment to estimate the heritability of growth in relation to morphotypic transformation among full and half sib families of the freshwater prawn, Macrobrachium rosenbergii was undertaken. Observations on prawn growth and morphotypic transformation were made over a period of 31 weeks, during which the juvenile prawns were kept in hapa enclosure and the adults in cages. The hapas and cages were suspended in an earthen pond where the water quality was maintained homogeneously under suitable conditions. The heritability estimates were made on the progeny from 32 full and half sib families nested within eight sires with two dams per sire. The broad sense heritability was estimated for carapace length, body length, claw length and total body weight. The heritability estimates on the carapace length based on paternal, maternal and fullsib analyses were found to be the highest  $0.40 \, (\pm 0.22), \, 0.13 \, (\pm 0.07)$  and  $0.26 \, (\pm 0.11)$  at 23 weeks, respectively.

The heritability on morphotypic transformation at 31 weeks from orange claw males (OC) to blue claw males (BC) were  $0 \pm 0.04$ ,  $0.73 \pm 0.08$  and  $0.37 \pm 0.02$  and the morphotypic transformation from small males (SM) after removing bulls were  $0.21 \pm 0.06$ ,  $0.56 \pm 0.05$  and  $0.39 \pm 0.03$  for paternal, maternal and fullsib analyses, respectively. The survival rate in the cage culture of initial stock to 23 weeks ranged from 55% to 96%. The number of females presented in every cage was greater than male and blue claw males dominated among three male morphotypes at 23 weeks. The resulting heritabilities implied the possibility for traits improvement.

# Rattikansukha, C. 1993. Intraspecific hybridization in *Macrobrachium rosenbergii* de Man. M.Sc. Thesis, Chulalongkorn Univ., Bangkok, Thailand

An intraspecific hybridization of two *Macrobrachium rosenbergii* populations, using the reciprocal crosses of prawns from Kraburi and Chao Phraya Rivers was carried out. Results of the study indicated that the postlarvae obtained from the Kraburi parents presented body length significantly larger than body length of those obtained from the Chao Phraya parents, while hybrids of the two populations were not presenting heterosis. The result implied some differences between the economic traits of different populations or strains of this species. Thus, further study on the strain selection and improvement should be carried out.

Uraiwan, S., Sumanojitraporn, S. and K. Ampolsak. 2002. Genetic improvement to increase growth rate of giant freshwater prawn (*Macrobrachium rosenbergii* de Man): heritability estimates and within-family selection. The Proceedings of 40<sup>th</sup> Kasetsart University Annual Conference. pp. 632-640





Sib analysis and selection procedures were respectively designed to estimate heritability and realized heritability on growth rate of *Macrobrachium rosenbergii*. The first procedure was carried out in cages with mixed sex rearing at the Ayuthaya Freshwater Fisheries Center, Ayuthaya Province between 1991 to 1992, and in concrete ponds with separate sex rearing at the Aquatic Animal Genetics Research and Development Institute, Pathumthani Province between 1996 to 1997. Under the cage culture condition, heritabilities were estimated from 16 full-sib and 8 half-sib families using nested analysis of variance model. Heritabilities of length and weight of male and female prawns 5 months old were -0.081±0.014 and 0.122±0.074, and 0.060±0.054 and 0.030±0.041, respectively.

Under pond conditions, 17 full-sib families were designed. Heritabilities of length and weight of male and female prawns 6 months old were  $0.156\pm0.077$  and  $0.142\pm0.096$ , and  $0.254\pm0.080$  and  $0.272\pm0.210$ , respectively. The realized heritabilities were estimated after one generation of within-family selection, the male and female prawns at 6 months old, these were 0.331 and 0.058, and 0.745 and 0.395, respectively. After one generation of selection for large size, female prawns of the selected line 6 months old were significant ( $p \pm 0.01$ ) 6 and 12% larger by length and weight than those of the control line, respectively. Similarly, female prawns at 6 months old of the selected line were significant ( $p \pm 0.01$ ) 5 and 16% larger by length and weight than those of the parental line, respectively. The results illustrated that genetic improvement to increase growth rate of the *Macrobrachium rosenbergii* is possible.

Uraiwan, S., Sumanojitraporn, S., Ampolsak, K. and S. Jeenmik. 2003. Response to within-family selection on growth rate of freshwater prawn (*Macrobrachium rosenbergii* de Man). The Seminar on Fisheries 2003. 7-9<sup>th</sup> July 2003, at the Department of Fisheries, Thailand

Selective breeding program to improve growth rate of giant freshwater prawn ( $Macrobrachium\ rosenbergii$ ) was carried out at the Aquatic Animal Genetics Research and Development Institute from 1998 to 2000. Within family selection procedure was applied on the growth rate of cultured prawns. The experiment consisted of two lines including a high growth selected line and a control line. The selection responses were estimated after one generation of selection. Female prawns of the selected line at 20 weeks old were (p < 0.01) 4 (12%) and 5 (20%) significantly larger by length and weight than those of the control line and their parent generation, respectively.

Similarly, male prawns of the selected line at 20 weeks old were (p < 0.01) 5 (18%) and 7 (14%) significantly larger by length and weight than those of the control line and their parent generation, respectively. The estimated realized heritability at one generation of selection was moderate. The average heritability in length and weight at 20 weeks old were 0.38 and 0.22, respectively. The results of this experiment illustrated that the within-family selection is the efficient procedure to improve growth of the giant freshwater prawn, and this method is being recommended for broodstock management.

### (4) MANAGEMENT PROCEDURE AND SELECTIVE EFFECTS CONTROL

Doyle, R.W., Singholka, S. and M.B. New. 1983. "Indirect selection" for genetic change: a quantitative analysis illustrated with *Macrobarchium rosenbergii*. Aquaculture, 30: 237-247

The term "indirect selection" means selection, which is exerted on a trait by methods other than deliberate artificial selection for the trait itself. It includes selection, which may be an incidental byproduct of harvesting and breeding techniques, as well as correlated selection on a trait caused by artificial selection on another trait.

Expressions are derived for calculating the intensity of indirect selection in aquaculture environments. The calculations are illustrated with growth-rate data on *M. rosenbergii* from prawn ponds in Thailand, and with computer-generated data, which simulate measurements made during a multiple mark-recapture experiment. Indirect selection for growth is probably negligible in Thai prawn farms and hatcheries at present, but small changes in management practice could exert strong indirect selection on growth rate. One very effective change is by collecting the broodstock as early in the production cycle as possible. Control of indirect selection may be useful for the genetic improvement of aquaculture stock, especially in developing countries and other situations where an intensive artificial selection program is not economically or biologically desirable. Like all selection programs, the probability of success is critically dependent on the genetics of the traits being selected.

Doyle, R.W. 1983. An approach to the quantitative analysis of domestication selection in aquaculture. Aquaculture, 33:167-185





Domestication selection is defined as natural selection on traits, which affect survival and reproduction in a human-controlled (domestic) environment. By altering various aspects of the environment, domestication selection can be made either to augment or to oppose artificial selection on traits of commercial importance. An example has been shown on the effects of selection on growth in *Macrobrachium rosenbergii* associated with variable development rate and age-at-harvest. It is concluded that management procedures can have strong selective effects and that genetic changes (for good or ill) may be expected to occur rapidly if the obvious genetic conditions are met.

#### (5) BIOTECHNOLOGICAL APPROACHES TO GENETIC IMPROVEMENT

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All-male production is one method for the aquaculture stock of *Macrobrachium rosenbergii* to be more effective, because normal male prawns (zz) grow much better than the normal females (zw) especially in the first six months of culture. The method for all-male production includes sex-chromosome manipulation based on sex reversal to female using female synthetic hormone. The procedure also needs development of molecular or DNA marker which is a sex-linked marker to be used for selecting the right neofemale prawn, carrying the male zz-chromosomes, to mate with a normal male, also carrying the zz-chromosomes. Consequently, all progenies obtained should be all-male with zz-chromosomes, half of which should be from the mother (neofemale) and father (normal male) each.

#### 1. Sex reversal to female using synthetic hormone

The sex reversal in *Macrobrachium rosenbergii* to female using female synthetic hormone, 17b-estradiol, is based on two treatments: oral administration and immersion in water containing the hormone. Giving hormone begins with the fifteen-days-postlarvae at 0, 50, 100 and 150 mg of 17b-estradiol/g of food for oral administration treatment, and 0, 50, 100, and 150 mg of 17b-estradiol/l of water for the immersion treatment.

Both treatments were for 15, 30 and 45 days. Results in the oral administration treatment indicated that (1) the sex-ratios obtained from every experimental group were not significantly different from the normal 1:1; (2) abnormal males were found in every experimental group; and (3) the best survival rate of 90% was in the group fed with the 100 mg/g for 30 days. Results of the immersion treatment illustrated that the higher dosage and duration of immersion undertaken, the higher female sex ratio is effectively obtained. The highest percent of female, 81.63 was obtained from the group immersed in 150 mg/l for 45 days. The sex ratio of this group was significantly different from the normal 1:1, presenting a 70% survival rate.

# 2. Sex-linked marker development

Searching for sex-linked marker in *Macrobrachium rosenbergii* was based on AFLP (Amplified Fragments Length Polymorphism) technique with the use of 7 primer pairs for PCR-I and 92 primer pairs for PCR-II. Results indicated that only 6 pairs were found to have polymorphic bands between male and female prawns. These DNA bands were sequenced and the DNA pieces being male and female sex-specific were found. Primers to be used for amplifying these pieces of DNA were designed from the sequences at both ends of each DNA piece. The designed primers were tested with prawn samples from male and female populations. The results indicated that the primers were still not good enough to identify the different sex properly and still could not be used for fieldwork.

# Klinbu-nga, S. (pers. comm.). Marine Biotechnology Research Unit, National Center for Genetic Engineering and Biotechnology (BIOTEC), based in Chulalongkorn University campus Bangkok, Thailand

Klinbu-nga (pers. comm.) has also been searching for sex-specific DNA markers in *Macrobrachium rosenbergii* using AFLP technique. He claimed that five male-specific markers and four female-specific markers have been found. Further to this, he has been doing more advanced research by examining the expression of genes at terminal ends of the vas deferens and oviducts of male and female prawns, respectively using the RAP-PCR technique. Markers expressing specifically the small claw males (340 bp) and the females (415 bp) have been found. All of the DNA markers found have been cloned and will be tested for the sex-specificity.

Sagi, A. and E.D. Aflalo. 2003. The androgenic gland and monosex culture in prawns – biotechnological perspective. Dept. of Life Sciences and the Institute for Applied Biosciences, Ben Gurion Univ. of the





#### Negev, Beer Sheva, Israel.

Males of the freshwater prawn, *Macrobrachium rosenbergii*, grow faster and reach a larger size at harvest compared to females, making the culture of monosex all-male population advantageous. Sexual differentiation in crustaceans is regulated by the androgenic gland (AG) found to be exerting morphological, anatomical, physiological and behavioral effects. The AG plays a pivotal role in the regulation of male differentiation and in the inhibition of female differentiation. In *M. rosenbergii*, complete sex reversal was achieved by AG removal in immature males, resulting in female differentiation, including the development of ovaries, oviducts and female gonopores. Similarly, AG implantation into immature females leads to the development of testes, sperm ducts and male gonopores. *M. rosenbergii* that had undergone sex reversal proved to be capable of mating with normal specimens and producing progenies. Early attempts to culture all male populations through manual segregation were reported from Israel and recently from India and other countries. The production of monosex prawn







Harvest and transport of freshwater prawn fry (right) for grow-out culture in earthen ponds (left) following the Code of Conduct Guidelines established by Thailand

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