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EFFECTS OF FOUR STOCKING DENSITIES AND THREE DIETS ON GROWTH AND SURVIVAL OF POSTLARVAL *Macrobrachium rosenbergii* and *M. acanthurus*[1]

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ABSTRACT

Effects of stocking density and three diets on growth and survival of 1 week post-metamorphic *Macrobrachium rosenbergii* and *M. acanthurus* were studied. Experiments were conducted on shrimp held for 2 months in laboratory tanks under a controlled temperature, light, and feeding regime.

Initial stocking densities were 215, 430, 645, and 860 shrimp/m^2 tank bottom. Shrimp were fed Purina 20% protein marine ration at 5% body weight daily. Habitat was gravel bottom. *M. rosenbergii* survival was highest at 215 shrimp/m^2, but overall growth was best at 430 shrimp/m^2. *M. acanthurus* survival and growth were very erratic.

Two commercially available diets (Purina 25% marine ration, and Purina No. 1 trout chow) and a "natural" diet composed of chopped fish, shrimp, earthworm, rice, and mung beans were selected for dietary studies Shrimp were stocked at 107.5 shrimp/m^2 tank bottom. A three-tiered habi-

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tat was added to reduce aggression and food competition. Shrimp were fed at 20% body weight daily for the first six weeks and 12% daily for the final two weeks. Trout chow produced the most rapid growth; maximum average increase in M. rosenbergii was from 0.03 g initially to 1.20 g after 2 months.

INTRODUCTION

Larviculture of freshwater shrimp (Macrobrachium spp.) has outgrown its infancy in the last few years due to improvements in rearing techniques. Changes in larval rearing procedures now allow 40-50% average survival through metamorphosis (Smith et al., 1974; Dugan et al., 1975; Hagood and Willis, 1976). Several private Macrobrachium farms are currently producing shrimp for sale to the general public, while numerous other research organizations are currently involved in optimization of various aspects of shrimp culture (Goodwin and Hanson, 1975). Commercial production is currently feasible and the number of private organizations interested in Macrobrachium culture is rapidly increasing. Improvements are still needed in such areas as pond management technique, feeding strategy, harvest methods, processing procedures, and determination of the market potential of Macrobrachium.

The possibility of producing a larger, hardier juvenile shrimp prior to pond stocking has received little attention. Early studies on postlarval growth in warm, tropical environments (Ling, 1967) indicate growth to an improved stocking size of 25 mm is possible in holding tanks in 30 days with a stocking density of 200 shrimp/m^2.

This project was designed to study growth of postlarval M. rosenbergii and M. acanthurus in indoor tanks under controlled temperature, light, water, and feeding conditions (i.e. "nursery" growth) for a two-month period. Nursery-type growouts in outdoor tanks (Fujimura, 1966, 1970; Balazs et al., 1973, in press) have shown that under known feeding regimes with maintenance of good water quality, survival of postlarval shrimp is high. Fujimura (1970) stocked postlarval M. rosenbergii at 750 shrimp/m^2 with 89.1% survival after 62 days. It is currently standard procedure to stock ponds with newly metamorphosed postlarval shrimp with an anticipated loss due to weak postlarvae and predation by insect larvae (Jim Williams, Caribe King Shrimp, 1975, pers. comm.). Ling (1967) suggested that stocking ponds with 25 mm juvenile shrimp would reduce initial losses and allow a more predictable pond survival rate and, therefore, more accurate estimates of proper feeding rate, harvest size, and restocking rates.

Several other benefits would also accrue from a nursery growout. Such a procedure would allow for elimination prior to pond stocking of the inherently weak, slow-growing shrimp normally present in every hatch (Ling, 1962; Sandifer and Smith, in press). Sizing shrimp from a nursery growout would provide initial uniformity in size of pond-stocked shrimp. Uniformity of size has been found to reduce aggression among post-juvenile M. rosenbergii (Segal and Roe, 1974), resulting in fewer losses due to cannibalism. Dobkins (1974) found that early postlarval male M. acanthurus are generally larger than females.

It is generally agreed that M. rosenbergii has the most commercial potential of all freshwater shrimp species (Goodwin and Hanson, 1975). Rapid growth rates (Sandifer and Smith, in press; Fujimura, 1970) and market potential (Fujimura, 1974; Shang, 1972; J. Parker in Goodwin and Hanson, 1975) make M. rosenbergii a popular species for culture. Several other species with commercial potential successfully reared through metamorphosis include M. carcinus (Choudhury, 1971) and M. acanthurus (Choudhury, 1970); however, aggression and long larval period make M. carcinus less desirable for commercial rearing (Goodwin and Hanson, 1975). M. acanthurus was selected for our experiments because of its cold tolerance and commercial potential as bait (Goodwin and Hanson, 1975). Dobkins (1973) found reduced survival and slow growth for M. acanthurus during autumn-winter, although 20% survived water temperatures as low as 15 C. M. acanthurus was found to be relatively expensive to rear through metamorphosis (Hagood and Willis, 1976), with postlarval production cost of $13.42/1000 compared to $3.56/1000 for postlarval M. rosenbergii.

METHODS AND MATERIALS

Macrobrachium spp. used in the experiments were reared through larval stages using the two-phase rearing method outlined by Hagood and Willis (1976). Newly metamorphosed shrimp were slowly acclimated to reduce salinities (4-5 ppt) 4-7 days prior to experiments. Deionized water and Instant Ocean artificial sea salts were used to create low salinities.

Shrimp were transferred to a 6.0 x 9.7 m temperature controlled building for the studies. Two sets of experimental tanks were set up, each with a 1.0 x 1.9 m biological filter containing crushed shell and coral 20 cm deep. Each system was cross-linked, insuring uniform water quality in all tanks. This system consisted of four tanks each with a 2.090 m^2 bottom area (636 liter) and six tanks 1.035 m^2 (318 liter) apiece. Four 1/4 hp pumps circulated the water in the system and two electric heating elements, 7.5 KW and 5.0 KW, maintained constant temperature (28-30 C). Water quality, temperature, salinity, pH, and nitrite/nitrogen were monitored daily. Lighting over experimental tanks was kept at 14 hr per day with full spectrum fluorescent bulbs delivering 25-30 foot candle intensity at the waters' surface.

During the stocking density study, the four larger tanks were divided down the center with epoxy-coated boards, creating eight 1.045 m^2 areas stocked with 215, 430, 645, and 860 shrimp/m^2 of each species. Ling (pers. comm., 1974) suggested that 322-420 shrimp/m^2 is optimal for postlarval growout of M. rosenbergii under laboratory-nursery tank conditions. Habitat consisted of sand gravel substrate. Shrimp were fed Purina 20% protein marine ration at 5% body weight daily. Feeding amounts were adjusted weekly based on growth data from Fujimura (1966) postlarval M. rosenbergii. Feeding was also adjusted for mortality after one month.

During the diet experiment all ten tanks were used. The epoxy-coated dividers were removed and three-tiered platforms made of 1.27 cm plastic fluorescent light louver material was added to all tanks to reduce aggression and cannibalism. Stocking density was reduced to 107.5 shrimp/m^2.

Diets tested were Purina #1 trout chow, Purina 25% protein marine ration, and a "natural" diet based upon food habits found in the wild by Ling (1961, 1967). The natural diet consisted of 25% chopped fish, 25% chopped live earthworms, 25% chopped shrimp, 12.5% chopped sprouted mung beans, and 12.5% chopped long grain wild rice. Balazs et al. (in press) showed that high protein levels (15–35%) produce superior growth for M. rosenbergii with 35% yielding best growth after 244 days.

Each diet was administered to two 1.045 m^2 tanks and a 2.090 m^2 tank. The feeding rate during dietary testing was 20% body weight daily for the first 6 weeks and 12% for the last 2 weeks. Length, weight, and survival data were gathered at 1 and 2 months, with feeding amounts adjusted for actual weight and shrimp number.

At 1 month, tanks were drained and all detrital debris removed; prior to refilling, tank walls were flushed with fresh water but not cleaned. At 1 and 2 months, all shrimp were counted for survival. An aliquot sample of 50 shrimp was removed randomly from each tank, with exception of an attempt to obtain the largest and smallest shrimp from each group to determine size range. Each of the 50 shrimp were measured (total length, rostrum to telson) and the entire aliquot weighed to obtain average weight per shrimp.

RESULTS AND DISCUSSION

Stocking Density Study

An unusually large number of mortalities were observed on day 7 with a sharp increase on day 13. From day 13 through day 29 mortalities were observed; thereafter, deaths tapered off and were nominal for the balance of the study. Death counts were made difficult by presence of the habitat, rapid decomposition of the dead shrimp, and cannibalism. Dead shrimp observed were representative of the entire size range in the tank. Population densities at one month had reduced to such an extent that cannibalism was minimal and the stronger shrimp remained. Original densities of 215, 430, 645, and 860 shrimp/m^2 had decreased to 113, 69, 45, and 45 shrimp/m^2 for M. rosenbergii and 115, 21, 217, and 306 shrimp/m^2 for M. acanthurus. Histological examination showed no evidence of major pathogens, however, gills appeared to be clogged with the filamentous bacterium Leucothrix mucor. Many shrimp were observed to have damaged or lost appendages and lacerations in their exoskeletons. L. mucor was possibly a secondary infection brought on by the numerous injuries. Observing the same bacterium in larval and postlarval systems, Sandifer and Smith (1975) suggested various methods of control, but found best success by simply reducing the density of shrimp. We observed that shrimp near death displayed white abdominal musculature characteristic of stressed shrimp (Sinderman, 1974). Death occurred slowly, with shrimp first appearing lethargic and therefore subject to cannibalism.

M. rosenbergii survival was inversely proportional to stocking density, however, weight gain showed little relation to stocking density (Table 1). Unexpectedly, shrimp stocked at the greatest density (860/m^2) showed best weight gains (Table 1). Sandifer and Smith (in press) found both survival and growth to be inversely proportional to stocking density, with densities of 10-500 juvenile M. rosenbergii/m^2. During that study, he had very high survival (90%) for the first 90 days at a density of 200 shrimp/m^2 feeding 10% body weight daily.

M. acanthurus exhibited very erratic growth and survival rates at the different stocking levels (Table 1) possibly due to its more aggressive nature and early sexual dimorphism. M. acanthurus becomes sexually mature as early as 2-1/2 months post-metamorphosis (Willis, unpublished data, 1974).

Active aggression and cannibalism with resultant poor survival indicate that the initial feeding rate was insufficient. The unexpected weight gain in M. rosenbergii stocked at the greatest density reflected excess food availability since the feeding rate was not adjusted for the high mortality until survival was accurately determined at 1 month. The gravel bottom provided little habitat for shrimp. High survival during the dietary study utilizing a three-tiered habitat, indicated that habitat complexity is required for post-metamorphic, high density growout of Macrobrachium spp.

Dietary Study

Good growth and survival rates were found during the dietary studies of M. rosenbergii and M. acanthurus. A Bartletts chi-squared test (Sokal and Rohlf, 1969) indicated that large variations in weight gains between sampling periods yielded data not normally distributed until mean weights were changed to natural log values. A nested ANOVA test for variance indicated no significant difference in mean weight gain between species but significant variation between diets ($P<0.05$). A Student-Newman-Keuls test (Sokal and Rohlf, 1969) used to determine the actual source of dietary variation indicated no significant difference ($P<0.05$) in weight gain between trout chow and the natural diet fed M. rosenbergii. Weight gains by M. rosenbergii fed trout chow and the natural diet were significantly greater than gains by M. rosenbergii fed marine ration and all M. acanthurus diets. This indicates that M. rosenbergii did indeed have a growth rate superior to M. acanthurus, with the exception of M. rosenbergii fed marine ration. This group did poorly and exhibited no significant species growth rate differential. Although high protein diets are generally considered uneconomical for rearing shrimp to market size (Fujimura, 1966; Goodwin and Hanson, 1975), we found trout chow (40% protein) to produce excellent growth and survival. Balazs et al. (in press) also found excellent growth and survival with M. rosenbergii fed a 35% protein diet for 244 days. Trout chow sells for $.62/k retail in small quantities, but with an average conversion ratio of 1.31:1 for month one with M. rosenbergii (Table 2), food costs were negligible.

The natural diet produced growth rates comparable to commercial diets, particularly during the first month of experimentation (Tables 2 and 3). A natural diet is uneconomical because of lengthy preparation required for chopping to appropriate particle size, high cost of ingredients, and short storage life. This type of diet may also allow introduction of pathogens or bacteria into the systems via spoiled, old, or contaminated food particles. Of the different portions of the diet, the order of acceptance was observed to be: worm, fish, shrimp, mung bean, and rice. All portions were readily eaten, although rice often had to soften before it was consumed.

Examination of causes for poor growth and survival of M. rosenbergii fed marine ration revealed that the food had been exposed to high temperatures prior to shipping, causing fat content to be slightly rancid. Since this may have been somewhat responsible for poor results, a new batch of 25% protein marine ration was used for the M. acanthurus dietary study. Survival was good for M. acanthurus with the new batch, but growth remained poor (Table 3).

Size range in stocking density and dietary studies was similar for M. acanthurus and M. rosenbergii (Tables 1, 2, and 3). Range of size became more pronounced during the second month for both species (Tables 2 and 3). Each treatment had two to four males with large chelae which dominated the tank particularly during feeding periods. M. acanthurus tended to be evenly distributed throughout the entire size range. Each group of M. rosenbergii produced a few dominant males, a small number (approximately 5%) exhibiting little or no growth, and the majority of shrimp growing at nearly the same rate. Such constancy of size provides predictability of growth rate and could also allow separation of the fast growers for rapid growth to market size and elimination of the inherently weak, slow-growing shrimp.

SUMMARY

1) Postlarval M. rosenbergii and M. acanthurus fed high protein (40%) trout chow exhibit rapid growth rates under controlled temperature, light, and water conditions.

2) Feeding rates must be high early after metamorphosis. Good survival and growth was obtained by feeding 20% body weight daily.

3) Nursery growout of Macrobrachium to an ideal pond-stocking size of 25 mm can be accomplished in less than 1 month. Postlarval M. rosenbergii (12.98 mm) fed trout chow averaged 39.09 mm after 1 month.

4) Habitat complexity is required for all post-metamorphic growth of Macrobrachium in intensive culture systems. Our stocking density data are inconclusive but suggest that 430 shrimp/m^2 is optimal.

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Table 1. Two month stocking density study with *M. rosenbergii* and *M. acanthurus* post larvae

Macrobrachium rosenbergii

Initial Stocking Density		$215/M^2$			$430/M^2$			$645/M^2$			$860/M^2$	
Sampling Period	Initial	One Month	Two Month	Initial	One Month	Two Month	Initial	One Month	Two Month	Initial	One Month	Two Month
Number Shrimp	225	131	118	450	80	72	675	77	47	900	150	47
Percent Survival	-	58.2	52.4	-	17.8	16.0	-	11.4	6.9	-	16.6	5.2
Average Weight (g)	0.032	0.046	0.140	0.032	0.080	0.335	0.032	0.054	0.308	0.032	0.102	0.444
Average Length (mm)	13.22	13.54	24.32	13.22	18.14	29.12	13.22	17.26	31.45	13.22	19.98	32.04
Range Length (mm)	8-17	9-34	18-52	8-17	11-36	16-52	8-17	13-37	21-60	8-17	11-42	19-71

Macrobrachium acanthurus

Initial Stocking Density		$215/M^2$			$430/M^2$			$645/M^2$			$860/M^2$	
Sampling Period	Initial	One Month	Two Month	Initial	One Month	Two Month	Initial	One Month	Two Month	Initial	One Month	Two Month
Number Shrimp	225	131	120	450	29	22	675	371	227	900	430	320
Percent Survival	-	58.2	53.3	-	6.4	4.8	-	54.9	33.6	-	47.7	35.5
Average Weight (g)	0.029	0.085	0.258	0.029	0.092	0.363	0.029	0.061	0.296	0.029	0.043	0.172
Average Length (mm)	13.07	19.48	26.30	13.07	18.76	34.15	13.07	15.80	26.54	13.07	13.16	23.68
Range Length (mm)	7-21	12-40	18-62	7-21	11-38	24-62	7-21	10-47	17-73	7-21	8-40	15-58

Table 2. Macrobrachium rosenbergii two month dietary study

Natural Diet

Tank Number	1			4			7		
Sampling Period	Initial	One Month	Two Month	Initial	One Month	Two Month	Initial	One Month	Two Month
Number Shrimp	112	112	98	112	106	106	224	206	177
Percent Survival	-	100	87.5	-	94.6	94.6	-	91.9	79.0
Average Weight (g)	0.030	0.268	0.760	0.030	0.314	0.802	0.030	0.312	0.859
Total Weight Gain (g)	-	26.66	71.12	-	29.97	81.65	-	57.47	145.32
Total Amount Fed (g)	-	62.09	231.70	-	62.09	250.25	-	124.18	487.41
Total Food Conv. Ratio	-	2.33:1	3.26:1	-	2.07:1	3.06:1	-	2.16:1	3.35:1
Average Length (mm)	12.98	32.65	42.72	12.98	32.18	43.60	12.98	29.92	37.31
Range Length (mm)	7.5-22	22-50	23-70	7.5-22	20-46	27-62	7.5-22	19-47	28-75

Marine Ration

Tank Number	2			5			8		
Sampling Period	Initial	One Month	Two Month	Initial	One Month	Two Month	Initial	One Month	Two Month
Number Shrimp	112	29	24	112	65	39	224	84	78
Percent Survival	-	25.8	21.4	-	58.0	34.8	-	37.5	34.8
Average Weight (g)	0.030	0.087	0.361	0.030	0.167	0.527	0.030	0.131	0.521
Total Weight Gain (g)	-	Neg.	5.30	-	7.50	17.19	-	4.27	33.92
Total Amount Fed (g)	-	62.09	75.67	-	62.09	123.48	-	124.18	186.41
Total Food Conv. Ratio	-	Neg.	14.28:1	-	8.28:1	7.18:1	-	29.08:1	5.50:1
Average Length (mm)	12.98	22.83	35.86	12.98	26.68	36.68	12.98	22.26	39.45
Range Length (mm)	7.5-22	16-32	27-50	7.5-22	17-38	25-73	7.5-22	14-39	25-65

Trout Chow

Tank Number	3			6			9		
Sampling Period	Initial	One Month	Two Month	Initial	One Month	Two Month	Initial	One Month	Two Month
Number Shrimp	112	105	61	112	104	104	224	206	185
Percent Survival	-	93.7	54.5	-	92.8	92.8	-	91.9	82.6
Average Weight (g)	0.030	0.426	1.080	0.030	0.517	1.195	0.030	0.533	0.977
Total Weight Gain (g)	-	41.39	62.52	-	50.43	120.92	-	103.08	174.08
Total Amount Fed (g)	-	62.09	371.63	-	62.09	366.03	-	124.18	745.01
Total Food Conv. Ratio	-	1.50:1	5.94:1	-	1.23:1	3.03:1	-	1.20:1	4.28:1
Average Length (mm)	12.98	37.98	48.22	12.98	39.57	48.08	12.98	39.73	47.66
Range Length (mm)	7.5-22	21-52	27-76	7.5-22	22-56	22-79	7.5-22	25-55	25-69

Natural Diet

Tank Number	1			4			7		
Sampling Period	Initial	One Month	Two Month	Initial	One Month	Two Month	Initial	One Month	Two Month
Number Shrimp	112	100	91	112	110	96	224	196	143
Percent Survival	-	89.3	81.3	-	98.2	85.7	-	87.5	63.8
Average Weight (g)	0.018	0.143	0.292	0.018	0.140	0.294	0.018	0.111	0.271
Total Weight Gain (g)	-	12.32	24.55	-	13.34	26.20	-	17.65	34.72
Total Amount Fed (g)	-	32.97	127.61	-	32.97	134.54	-	65.94	209.58
Total Food Conv. Ratio	-	2.68:1	5.20:1	-	2.47:1	5.13:1	-	3.74:1	6.04:1
Average Length (mm)	12.09	20.87	26.92	12.09	20.44	26.55	12.09	20.59	25.52
Range Length (mm)	9-19	13-52	18-63	9-19	16-55	18-86	9-19	12-35	18-65

Marine Ration

Tank Number	2			5			8		
Sampling Period	Initial	One Month	Two Month	Initial	One Month	Two Month	Initial	One Month	Two Month
Number Shrimp	112	112	105	112	110	107	224	223	176
Percent Survival	-	100	93.8	-	98.2	95.5	-	99.6	78.6
Average Weight (g)	0.018	0.107	0.360	0.018	0.086	0.247	0.018	0.118	0.232
Total Weight Gain (g)	-	10.01	35.78	-	7.46	24.41	-	22.37	36.80
Total Amount Fed (g)	-	32.97	107.73	-	32.97	95.83	-	65.94	240.59
Total Food Conv. Ratio	-	3.29:1	3.01:1	-	4.42:1	3.93:1	-	2.95:1	6.54:1
Average Length (mm)	12.09	20.64	26.86	12.09	18.80	25.65	12.09	19.54	25.10
Range Length (mm)	9-19	14-44	17-62	9-19	14-28	19-40	9-19	13-34	18-43

Trout Chow

Tank Number	3			6			9		
Sampling Period	Initial	One Month	Two Month	Initial	One Month	Two Month	Initial	One Month	Two Month
Number Shrimp	112	101	91	112	112	105	224	211	202
Percent Survival	-	90.2	81.3	-	100	93.8	-	94.2	90.2
Average Weight (g)	0.018	0.114	0.446	0.018	0.163	0.517	0.018	0.102	0.345
Total Weight Gain (g)	-	9.51	38.57	-	16.28	52.27	-	17.58	65.66
Total Food Conv. Ratio	-	32.97	109.41	-	32.97	154.07	-	65.94	208.81
Total Food Conv. Ratio	-	3.47:1	2.84:1	-	2.03:1	2.95:1	-	3.75:1	3.18:1
Average Length (mm)	12.09	20.81	31.64	12.09	22.17	31.50	12.09	19.34	26.94
Range Length (mm)	9-19	16-28	23-60	9-19	11-49	18-65	9-19	14-31	18-53