A QUANTITATIVE STUDY OF FOOD PREFERENCES IN PENAEID SHRIMP

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Marine Biology

Submitted in Partial Fulfillment of the Requirements
of the University Undergraduate Fellows Program

1983-1984

Approved by:

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April 1984

ABSTRACT

A laboratory approach to quantify food preferences in penaeid shrimp is developed in this study. Direct visual observations from beneath the experimental tanks provided accurate data with which to assess relative attractivity of one artificial and three natural foods.

The method revealed that *Penaeus setiferus* has no significant preferenceamong the natural foods tested, which were polychaetes (*Nereis* sp.), and the muscle tissue of fish (*Archosargus probatocephalus*), shrimp (*Penaeus setiferus*), and squid (*Loligo* sp.). There was, however, a significant preference for any of the natural foods over the artificial feed "K" (Fenucci, Zein-Eldin and Lawrence, 1980).

Further research is needed to isolate and identify attractants in preferred natural foods. These compounds can then be supplemented in prepared shrimp diets to make them more attractive. The method developed in the present work can serve as a useful tool in this important aspect of diet development for penaeid shrimp culture.

ACKNOWLEDGEMENTS

I would like to thank my advisor, Dr. David V. Aldrich, for all the good advice and time he spent with me on this study. I also thank Dr. Zoula P. Zein-Eldin for providing the laboratory facilities and for her help and ideas.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

INTRODUCTION

Commercial shrimp farming in the United States is still considered a high risk investment. The high land, labor and feed costs can approach the expected value of crop yields. It is therefore, important to design an artificial feed that promotes maximum shrimp growth. This could increase crop yields per unit time, producing higher earnings to cover operating costs. As a result, most research efforts have been centered on the nutritional requirements of shrimp (New, 1976; Kanazawa, 1982).

Nutrition is, of course, an important factor to be considered when designing an artificial feed. However, the nutritional value of any diet is zero unless the shrimp is attracted to the feed and accepts it. Since attraction and feeding are stimulated by certain chemical substances, their supplementation in an artificial feed could enhance ingestion and growth (Heinen, 1980). Determining what natural foods shrimp prefer to eat would enable researchers to better isolate and identify what the chemical attractants are.

The conventional method to determine food preferences in most animals is a stomach content analysis. However, this approach produces inaccurate results with shrimp due to their modified stomach, the gastric mill (Maguire, 1980). Food that is ingested is reduced to a semi-fluid state by the combination of mechanical and enzyme activity (Gibson, 1981). Identification of the stomach contents would, therefore, only reveal harder substances whereas the soft, juicy items, perhaps the most preferred and nutritious would be unrecognizable. In addition,

--- format based on Journal of the World Mariculture Society the omnivorous feeding habits of shrimp results in a wide range of accepted foods making it even more difficult to establish whether one is preferred (Wickens, 1976).

The alternative and best method of approach is through laboratory observations. As well as determining whether the shrimp prefer any of the natural foods tested, this study was designed to structure a method that can evaluate any potential diet. Direct visual observation of shrimp feeding behavior offers an effective data collection technique. In addition, the data can be quickly obtained instead of committing resources to large, long, expensive experiments such as pond studies.

MATERIALS AND METHODS

Rationale

The approach used to study preferences is based on a previous experiment by H.Y. Chen (1983). His work quantified preferences of post-larval Penaeus vannamei and P. stylirostris for several different artificial feeds. The present study modifies his technique in several ways. First, the design was changed to accommodatelarger animals and to compare natural foods as well as artificial feeds. Second, and most importantly, I modified the method of observations. Chen viewed his shrimp from the top of each tank assuming that if at least the shrimp's cephalothorax was observed inside a food dish, that particular food was eliciting a positive response. However, since shrimp have ventrally located mouthparts, this method does not quantify food preferences adequately. In this study, observations were made from beneath, through the bottom of the tank, so that a positive response (food in shrimp's mouth) could be clearly recognized and accurately counted.

Experimental Design

Shrimp used in this experiment were Penaeus setiferus (size 8-11 cm) obtained at a local live bait camp. The animals were kept in 3 fiber-glass holding tanks (1 m x 0.5 m) initially stocked at 45-55 shrimp/tank. At the start of testing however, (3 days after initial stocking), the number was reduced to 35-40 shrimp/tank due to the effects of handling during the events prior to purchase. This period of time was sufficient for the shrimp to adjust, since the mortality rate was virtually zero. Natural filtered seawater pumped from Galveston beach (20 C, 22-23 ppt) was always kept well aerated and clean (3/4 of sea-water changed daily) in the holding tanks. The water temperature was raised to 24-26 C at least 12-24 hours before shrimp were to be used in an experiment. An extra holding tank filled only with seawater was kept at 24-26 C as well to fill experimental tanks. The shrimp were starved 12-39 hours prior to conducting the experiments. Two experiments were conducted in the morning and four in the evening.

Each trial was conducted in triplicate (Figure 1) using three 75 l (20 gal) rectangular glass tanks. Each tank was supported on 5 cm x 10 cm (2" x 4") beams resting on cinder blocks arranged to permit observations from beneath. This experiment was designed to test three different food items at once. The food dishes (diameter 14 cm, height 2 cm) were arranged in the tanks as in Figure 2, so that a particular food would appear once in each of the three possible

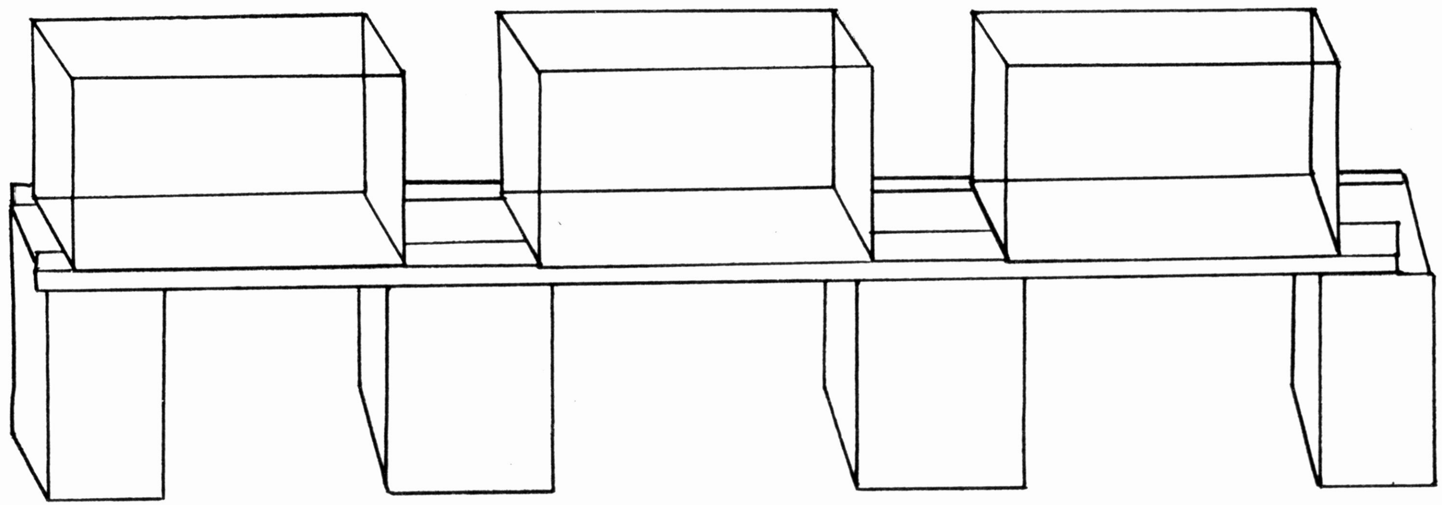

Figure 1: Arrangement of experimental tanks.

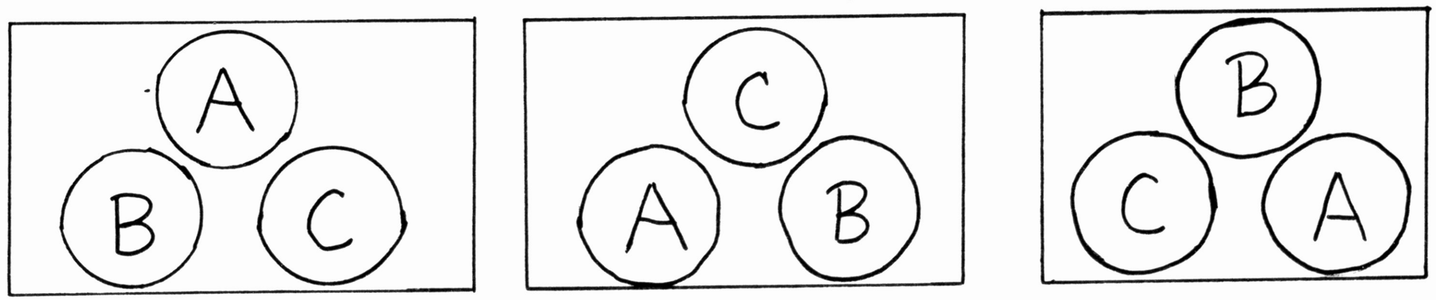

Figure 2: Arrangement of food items in each tank (vertical view).

positions. This was done to control location as a reason for the positive responses.

Large polychaete worms (Nereis sp.) and muscle tissue of squid (Loligo sp.), fish (Archosargus probatocephalus) and shrimp (Penaeus setiferus) were the natural foods compared in this study. In addition an artificial feed "K" (Fenucci and Zein-Eldin, 1976) was also tested. Since the squid, shrimp and fish all have similar colors, different identifiable shapes of the food pieces were used to permit subsequent recognition by the observer. The shrimp was cut into small pieces (1 cm x 0.5 cm, 0.3 cm thick), squid was cut into long strips (2.5 cm long, 0.5 cm wide), and fish was shaped into larger blocks (2 cm x 1 cm, 0.5 cm thick). Worm pieces (1.5 cm long, 0.5 cm diameter) were easily identified by color. Artificial feed "K" was in string pellet form (0.2 cm diameter, 1.0-2.5 cm long).

Since different food shapes and sizes introduce additional variables that might influence the responses, the following experiments were conducted. First, to determine whether large or small blocks were preferred, the same food (fish) was cut into the approximate dimensions of the shrimp and fish pieces used. These two sizes were then tested using the same procedure as if comparing the different food items. The three shapes, long strips, large and small blocks, were also compared in the same manner.

In order to decrease the amount of disturbance to the shrimp during testing periods, the sides of each tank were covered with light brown paper towels and the top was covered with a green plastic sheet. Both materials were translucent enough to allow for clear observations from beneath the aquaria. Each tank was filled to approximately one half its volume so that vertical movement of the shrimp was somewhat restricted. This tended to facilitate quantitative identification of mouth contents. The water was well aerated before each experiment, but during the experiments, the air supply was stopped so that water currents would not influence the shrimp's behavior.

Experimental Procedure

After half-filling the three experimental tanks with 24-26 C seawater and providing aeration, I arranged three empty glass food dishes in each tank. The foods to be compared were then cut into pieces (8 pieces food/food dish). After the food pieces were prepared, twelve shrimp were netted from the holding tank and placed in a plastic mesh cylinder (14 cm diameter, 20-30 cm high) within each tank. Air was still provided while the shrimp were allowed to settle (about 10 minutes). During this period, each food item was placed in its proper food dish. After all the food was arranged, aeration was stopped. The shrimp in tank 2 were released first; then at 1 minute intervals, the shrimp in the remaining two tanks were released. Observations for each tank began about ten minutes after the release of the shrimp and followed the same sequence as the shrimp release. An automobile mechanic's creeper covered with a piece of foam was used to lie on and allowed the observer to move under each tank easily. For each observation the positive responses were counted and categorized according to food type. This procedure was repeated every five minutes for 1½ hours.

The results were statistically analyzed using the Analysis of Variance and Duncan's Multiple Range test (Steel and Torrie, 1960).

RESULTS

There was no particular preference among the natural foods tested (Table 1). Analysis of variance showed no significant differences among them (Table 2). The analysis of variance for the results of experiments 5 and 6, however, indicate a significant preference among the foods tested. Duncan's multiple range test for these two experiments revealed the difference to be between the artificial feed and the natural foods (Table 3).

Table 1: Food preferences of *P. setiferus*. Mean positive responses ± 1 standard deviation for each food item for 3 tanks

Experiment	Starting time	Fasting period (hrs)	Worm	Squid	Shrimp	Fish	K	Significance
1	1030	18	11.3±1.5	6.0±7.1	12.3±5.5	---	---	NS
2	2050	27	25.3±9.9	17.7±6.1	24.0±6.2	---	---	NS
3	0950	39	---	19.0±8.2	25.0±3.0	13.3±2.0	---	NS
4	2010	12	---	32.0±8.0	27.7±7.4	24.0±5.7	---	NS
5	2115	32	---	62.0±8.6	---	41.3±6.6	5.0±4.1	$p<0.01$
6	1800	24	---	54.0±15.5	59.3±7.4	---	16.7±13.7	$p<0.05$

Table 2: Analysis of variance for data of Table 1.

Experiment	Source of variation	df	SS	MS	F	Significance
1 (W-Sq-Sh)*	treatment	2	70	35	0.84	NS
	error	6	243	40.5		
2 (W-Sq-Sh)	treatment	2	101	50.5	0.58	NS
	error	6	519	86.5		
3 (Sq-Sh-F)	treatment	2	204	102	2.55	NS
	error	6	239	40		
4 (Sq-Sh-F)	treatment	2	96	48	0.63	NS
	error	6	457	76		
5 (Sq-F-K)	treatment	2	4996	2498	37.3	$p<.005$
6 (Sq-Sh-K)	treatment	2	3248	1624	6.7	$p<.05$
	error	6	1449	241.5		

* W=worm; Sq = squid; Sh = shrimp; F = fish

Table 3: Duncan's multiple range test for experiments 5 and 6.

Experiment	Probability	K	F	Sq	Sh
5	0.01	5.0	41.3	62.0	--
6	0.05	16.7	--	54.0	59.3

The variability of the mean positive responses between experiments (Table 1), appears to be due to the time of day and period of starvation of each experiment. Further testing of these factors is needed. Such testing should lead to improved standardization of methods and a reduction of variation in responses between experiments.

The mean responses according to location are given in Table 4. The analysis of variance shows no significant difference among the locations except for experiment 4 (Table 5). Duncan's multiple range test indicates the difference to be between location C and A (Table 6). However, this can be considered an accident since all the remaining experiments reveal very nonsignificant results.

The results of the size and shape control experiments (Table 7) also shows that none was significantly preferred (Table 8).

Table 4: Test for location preference of *P. setiferus*. Mean positive responses ± 1 standard deviation for each location for 3 tanks.

Experiment	A	B	C	Significance
1	7.7±4.9	10.3±7.8	11.7±3.2	NS
2	24.7±10.1	21.0±9.0	21.3±4.0	NS
3	19.3±5.4	17.7±7.8	20.3±7.5	NS
4	26.7±5.9	36.3±4.1	20.7±2.6	$P<0.05$
5	33.7±25.3	42.7±28.2	32.0±17.2	NS
6	45.7±11.2	43.3±28.5	41.0±24.3	NS

Table 5: Analysis of variance for data of Table 4.

Experiment	Source of variation	df	SS	MS	F	Significance
1	treatment	2	24	12	0.25	NS
	error	6	289	48		
2	treatment	2	24	12	0.12	NS
	error	6	596	99		
3	treatment	2	10	5	0.06	NS
	error	6	433	72		
4	treatment	2	374	187	6.23	$p<.05$
	error	6	179	30		
5	treatment	2	197	99	0.11	NS
	error	6	5172	862		
6	treatment	2	32	16	0.003	NS
	error	6	4660	4654		

Table 6: Duncan's multiple range test for experiment 4 of Table 5.

Experiment	Probability	C	A	B
4	0.05	21	27	36

Table 7: Test for food size and shape preference of *P. setiferus*. Mean positive response ± 1 standard deviation for 3 tanks.

Experiment	Large	Small	Strip	Significance
7	33.3±7.4	29.7±0.9	35.0±5.9	NS
8	16.7±1.8	16.7±5.7	---	NS

Table 8: Analysis of variance for data of Table 7.

Experiment	Source of variation	df	SS	MS	F	Significance
7	treatment	2	44	22	0.49	NS
	error	6	270	45		
8	treatment	1	0	0	0.0	NS
	error	4	123	31		

DISCUSSION

The method used in this study provides researchers with an easy, efficient approach to compare shrimp diets. The data obtained was sufficient to indicate whether any foods were preferred.

The design of the experiment is not limited to comparing only natural foods. It can be used as a screening method to indicate which artificial feeds are most acceptable to shrimp. Being able to "weed" out the diets that are less attractive to shrimp will save researchers time and money that would otherwise be spent on needless growth studies for these poorer diets. Since artificial feeds often have similar colors and shapes, the pellets can be dyed different colors for easy recognition. A control experiment for color would then be required to test for color preferences.

The results of this study confirms that natural foods are significantly preferred over artificial feeds. This finding parallels the results of several others working with various different crustaceans reviewed by Heinen (1980). This strongly suggests that natural foods contain chemical substances attractive to shrimp.

To determine what these attractants are, the natural foods need to be chemically analyzed. Cowey and Tacon (1982) analyzed taste attractants in preferred natural foods of fish by fractionating and then bioassaying each fraction for attractant activity. Active fractions were then analyzed, and the active components were identified by making synthetic mixtures and running omission tests with bioassay. The authors suggest that quantitative analyses are then needed to indicate the stimulatory capacity of the feed. The procedure developed in this study could easily be used for this type of research. However, further growth studies would be required to assess the final value of the artificial feed.

It should be noted that the artificial feed tested in this study has supported better growth than the most widely used commercial shrimp diet (Fenucci, Zein-Eldin and Lawrence, 1980), yet in my work "K" was far less acceptable than natural foods to the shrimp. How much better results would artificial diets give if the proper stimulatory substances were added? It is interesting that Deshimaru (1982) incorporated a short-neck clam, *Venerupis philippinarum*, extract in a diet and obtained shrimp (*P. japonicus*) growth results comparable to those obtained when live short-neck clams were fed to shrimp.

In conclusion, the method developed in the present research provides a rapid quantitative method for the evaluation of diet acceptability to shrimp. It can serve as a useful tool in the important field of diet development for penaeid shrimp culture.

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