**The Effects of Different Diets on Ovary Maturation and Spawning of Pond-reared White Indian Shrimp (*Fenneropenaeus indicus*)**

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**Abstract:** Current investigation is conducted to determine effect of four different diets on ovary maturation and spawning of the pond-reared White Indian Shrimps for a time span of 60 days. One group of shrimps (control group) was fed with concentrate food of broodstocks. The second, third, and forth groups were fed by squid meat, Solen vagina meat, and Polychaete worms from Nereididae family, collected along the intertidal zone in Bandar Abbas, respectively. For each experimental group, three iterations were considered. Results of experiments indicate that the maximum amount of growth and maturation of ovary and spawning was for the fourth group (fed by Polychaete worms); the mentioned group had a significant difference with other groups (P<0.05). The second and the third groups (fed by squid and solen vagina) showed lower significant difference. The first group did not have a significant difference. Based on results and data, it can be concluded that three major factors of environmental conditions, endocrine hormones, and the diet type are important factors that have an impact on maturation of ovary and spawning of reared White Indian Shrimps.

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**1. Introduction**

White IndianShrimp (*Fenneropenaeus indicus*) is a species that have the highest amount of application among penaeidae family in aquiculture industry of Hormozgan Province. According to development of shrimp reproduction and breeding industry in south of the country, demands for post larvae are increasing year after year and providing broodstocks with high quality is major problem for producing high quality ovule and sperm. Most of shrimp hatcheries are dependent to wild reserves of shrimp, which have variable quality and supply. On the other hand, providing wild broodstocks has attracted most of the shrimp reproduction workshops, due to its low price. However, providing broodstocks by this method is only possible in a seasonal and limited manner. In addition, providing broodstocks by wild hunting causes many costs of a reproduction workshop, due to variation price of broodstock in each year. If the broodstocks can be reared in ponds, this would avoid hunting of wild reserves and reduces costs of egg production; broodstock is provided to the reproduction section with the same quality during the season, and since there are various diseases in this industry, if all the sanitary concerns on broodstocks maintenance are considered, diseases would not be transferred to reproduction and breeding section.

Stimulating sexual maturation and spawning in closed conditions is dependent to maintenance conditions. If the maintained shrimps are fed well with the live food, they would have better conditions. In current study, it was tried to feed the reared shrimps with 4 types of diet and the ovary maturation and spawning amount of these groups were compared.

# Material and Method:

Figure 1: white Indian Shrimps

White IndianShrimps (fig. 1) that were employed in this study were provided from ponds of north Tiab region of Minab town. Afterward, to conduct experiments, they were brought to shrimp reproduction workshop in Jask port in east of the Hormozgan province. This workshop was selected since it has appropriate condition of reservoirs, reproduction hall, and thermostat to adjust water temperature. After transferring shrimps to the workshop, they were maintained in initial conditions to be adapted to new conditions. Then, they were ranked based on their size and they were categorized for start of the experiment.

Twelve black fiberglass reservoirs [7] with 2 meters diameter, 1.5 meters height, 1.2 meters water depth, and 3.7 cubic meters volume were employed for experiments. Samples were divided into four groups and were kept in maturation reservoirs, with density of 5 samples in each square meter, including 8 female shrimp and 8 male shrimps with sexual ratio of 1:1 (sexual ratio was set according to Lawrence and Barry suggested method) [1]. Initial and final average weights of broodstocks in different treatments are depicted in Table 1. Samples were exposed to natural photoperiod for about 12 hours lighting and 12 hours of darkness. A green florescent lamp was employed for lighting [6]. In each day, water was replaced from the middle of reservoirs, passing through sandy filter with mild speed and with daily amount of 200%. Four air stones were placed on each reservoir. Reservoirs are covered by black covering to eliminate stress and disturbance of shrimps. Water physical and chemical indicators, such as temperature, salinity, pH, and dissolved oxygen were measured regularly. These amounts were obtained as 29-30 °C, 32 ppt, 7-8, and 4-5 mg/l, respectively.

Experiments on samples were conducted in four diet treatments and with three iterations; as in the first treatment, there were shrimps that were fed by special concentrate for broodstocks (Howrash Co. with 42% protein). Second treatment of shrimps was fed by squid meat (Loligo duvauceli), the third one was fed by Solen vagina, and the fourth treatment was fed by Polychaete worms, collected along the intertidal zone in BandarAbbas. Diets were consisted of four meals each day with amount of 10% of the body weight (based on the wet weight of body) with different intervals (8 AM, 1 PM, 6 PM, and 11 PM). Amounts of food was 30% at 8 AM, 40% at 1 PM, 20% at 6 PM, and 10% at 11 PM. Water replacement procedure was stopped during feeding for one hour, in order to avoid dispersal and loss of the food. Unused foods and offal were eliminated from the reservoir each morning. Fresh foods (squid and Solen vagina) were washed with clean sea water; then, they were cut to 1 to 1.5 centimeter pieces, are weighted, marked and kept in the freezer for using in other meals [14]. Squid was provided by hunters of Jask zone and Solen vagina was collected from a distance of 12 km from west of the workshop in Jask. Polychaete worms that were collected along the intertidal zone in BandarAbbas were washed and dried. They were also weighted and kept in the refrigerator, as other two samples.

When the mentioned shrimps reached to weight of 35 g, one ablation of the female shrimp was cut. To do so, the ablation was cut with surgical string, and then frontal section of the ablation was cut by a surgical scissor. Afterward, ovary maturation conditions were investigated by a stick, equipped by a small underwater lamp, daily between 6 and 7 PM. Prolific females were identified by the dark base in the abdominal section. Each prolific female was transferred to the 250 liters spawning reservoirs with the same physical and chemical conditions of the producing reservoirs with the filtered sea water depth of 60 cm, equipped with two air stones that were disinfected with 100 PPM formalin. Other females in the other maturation stages were kept in the producing reservoirs. Each morning, females are investigated to control spawning and detect released yellow protein uncus released in spawning period.

Females that had released the eggs completely were returned to the producing reservoir. Females that had released eggs partially or had not spawned were kept in the spawning reservoir for one or two more nights. They were returned to the producing reservoir after that. Producer female shrimps that were kept in the spawning reservoir spawned around 5 AM to 8 AM of the next morning, after transferring to the spawning reservoir. Their spawning indicator was observing protein remains of ovary tissue that is floating on the water surface. At this time, broodstock shrimp is returned to the producing reservoir, the amount of aeration is increased to help mixing of the water and eggs and hatching of eggs. If the temperature is as mentioned above, hatching would occur around 10 to 11 hours later. To count the number of a broodstock eggs after mixing of eggs and water in spawning reservoir, three 100 cc samples were taken from the spawning reservoir and the eggs were counted by the counting lam, which is usually used in plankton biology or microbiology lab, with help of 40 Nikon Microscope. Afterward, the obtained average of these three samples was multiplied by the volume of the reservoir water and the amount of eggs in each reservoir for each broodstock was calculated.

2 or 3 hours after hatching of the eggs, Napelioses counting was started. In this stage, after mixing water and napelioses, three 100 cc samples were taken from the reservoir. Counting and averaging the mentioned samples and multiplying this number by the reservoir water volume, the number of napelioses was calculated. Dividing this number to all the produced eggs of a broodstock and multiplying it by 100, hatch of the eggs was obtained.

Furthermore, shrimps of each treatment group were weighted for estimation of food commutation rate (FCR), food commutation efficiency (FCE), body specific growth (BSG), specific growth rate (SGR), and daily growth rate (DGR) (milligrams in each day). Results of the experiments were analyzed by unilateral variance analysis method of SPSS software and mean comparison of Danken method.

# Results:

Obtained results for growth and maturation stages of White IndianShrimps for all four kinds of diets are summarized in Table 2. Body weight was increased in all treatments, but there is a significant difference between groups. The most weight increment was observed in the group that was fed by Polychaete worms. These samples had a higher BSG (77.99%) and SGR (0.97%) and lower amount of FCR (2.85). The second feeding treatments (fed by squid) and the third one (fed by Solen vagina) showed better growth after the fourth treatment (fed by Polychaete worms). Statistically, there was not any significant growth difference between second and third treatments (P>0.05). The shrimps fed by concentrate food of broodstocks showed a slower growth compared to other three treatments. Also, the fourth treatment showed a better breeding efficiency and had the highest amount of growth, reproducibility, and hatching percent, among all experimented groups (P<0.05). In this group, the first symptoms of maturation were observed 20 days after drain of the ablation and in some samples, mature gonads were observed. Figure 2 illustrates an White IndianShrimp with cut ablation of the fourth stage.

Table 1: average initial and final weight in each experimented treatment

|  |  |  |
| --- | --- | --- |
| treatment | Initial weigh of shrimps (grams) | Final weight of shrimps (grams) |
| female | male | female | male |
| 1 (M1) | 25.50 | 18.14 | 37.98 | 27.14 |
| 2 (M2) | 25.41 | 18.20 | 39.14 | 29 |
| 3 (M3) | 25.52 | 18.15 | 39.25 | 29.85 |
| 4 (M4) | 25.50 | 18.12 | 44.22 | 33.42 |

Table 2: data obtained from different treatments

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Indicators | M1 | M2 | M3 | M4 |
| Average initial weight | 21.823.68 | 21.813.68 | 21.843.68 | 21.813.69 |
| Average final weight | 32.525.42b | 34.075.07b | 34.554.7b | 38.825.4a |
| FCR | 4.05n.s | 3.65b | 3.55b | 2.85a |
| FCE (%) | 24.7n.s | 27.4b | 28.2b | 35.1a |
| BSG (%) | 49.22n.s | 56.21b | 58.20b | 77.99a |
| SGR (%) | 0.67n.s | 0.75b | 0.77b | 0.97a |
| DGR (mg/day/indivi) | 179n.s | 204.3b | 211.8b | 283.5a |
| Maturation rate (ratio of matured female to all broodstocks) | 3.16 | 6.16 | 6.16 | 8.16 |
| Average number of each broodstock eggs | 62000n.s | 75000b | 82000b | 100000a |
| Average percentage of egg hatching (%) | 50n.s | 72b | 75b | 90a |

Figure 2: sample of White Indian Shrimp cut ablation fourth stage

# Discussion:

Nutrition is an important factor in growth and breeding of Penoied shrimps. Knowing shrimp nutrition requirements is essential for successful management of broodstocks. Live foods, such as squid, mollusks or Polychaete worms have been investigated in broodstocks nutrition of some shrimp species like monodon or vanami [1, 3, 12, and 19]; however, in Iran, there has not been such investigation to determine effect amount of live food on productivity of reared shrimps. Nowadays, it is well known that appropriate nutrition of shrimps increase their productivity. Therefore, diets containing materials with gonad objective are more important in managing broodstocks and can reduce investigation period and hatching functional cost.

Significant differences in FCR of White Indian Shrimps in different diets indicate essential differences in efficiency of food and growth of species; observations show that FCR values are variable with shrimp growth stages. Generally, used food in adults has a higher amount, while their FCR value is lower for them compared to their pre-maturation stage. Small shrimps showed a fast growth pattern; while larger shrimps had a smaller growth with the same diet [15]. Cheo [4] pointed out that in monodon shrimp, growth decreases with size and age increment. Viben et al. [17] reported that growth and nutrition have a direct relation with temperature and an inverse relation with size in vanami species. This shows that in pre-maturation stage, shrimps absorb nutrients and present a higher efficiency compared to elders, which indicates their lower FCR. Based on the information obtained in this study, it is obvious that for determining economic aspects of a diet, several factors, such as FCR, growth stage, and price of all daily diets should be taken into account. As a whole, diet type, physiologic efficiency, and diet usage have an effect on shrimp FCR.

Mature White IndianShrimps growth rate that were fed by fresh food (squid, Solen vagina, and Polychaete worms) was higher that shrimps that were fed by artificial food. However, in artificial food or concentrate, shrimp growth rate depends on protein amount of the diet. For example, Sudaryono et al. [15] stated that when monodon juvenile shrimps are fed by a diet based on lupin and soya, they grow for about 83.5 mg each day. Also, Cruz-Suarez et al. [4] showed that adding 10% squid flour to diet of monodon juveniles would improve growth of shrimps reared in pond and tank. Data obtained in this study reveals that White Indian Shrimp is not ideally fed by squid meat and these shrimps are more intended to be fed by mollusks meat. This may be due to meat tissue of these groups. Higher growth percent of these two treatments is due to nutrients of diets.

Higher growth rate and FCR of the fourth treatment group (fed by Polychaete worms) can be related to the mentioned worms. Polychaete worms, especially Nereididae family, are natural organisms that support maturation of penoied shrimps, due to HUFA, which help gonad maturation. Higher growth rate of shrimps fed by Polychaete worms is due to HUFA and nutrients that are essential for tissue making. Also, diets that lack essential fatty acids delay shrimp growth [11, 15].

It has been proved that fat source affects shrimp growth rate, as well as the whole fat amount [5, 13]. Although in fishes, it is possible that with reduction of diet protein and maintaining its high calorie value by fat growth procedure continues [18], not such investigations are reported for shrimps.

According to spawning, the first spawning occurred 20 days after cutting the ablation in most of the samples fed by the fourth diet. Furthermore, the amount of produced eggs and eggs hatching percent was higher than other treatments. This was certainly due to providing broodstocks with HUFA that affected gonads growth. Without a natural biosynthesis, these compositions cannot be used by broodstock shrimps. This important duty is performed by Polychaete worms. These results are in agreement with other researchers’ investigations [3, 8, and 9].

# Conclusion:

Sexual growth and maturation of White Indian Shrimps are affected by nutrients. Live food, such as Polychaete worms are effective in nutrition of shrimp broodstocks and improve better growth of ovary. In addition, Polychaete worms may cause better absorption of chemical compositions like cholesterol and long chain unsaturated fatty acids (HUFA), which are effective for growth of ovary and production of eggs with high quality. It should be mentioned that for better perception of this mechanism, it is required to conduct more physiologic studies in this field. Results of this study show that diet type has an important impact on ovary maturation and spawning of reared White Indian Shrimp, as well as environmental conditions and endocrine hormones.

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