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Influence of shrimp head meal incorporated diet on growth and maturation of goldfish *Carassius auratus*

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Abstract

The present study was carried out to find the influence of shrimp head meal on the growth and gonad maturation of goldfish, *Carassius auratus*. The experiment was carried out with gold fish juveniles (Average Body Weight - 5.98 ± 0.65 g) stocked in troughs and fed with shrimp head meal supplemented diet at two concentrations T₁ (10%) and T₂ (15%) with control diet for a period of 60 days. Sampling on the growth and histological observation was done within 10th and 30th days respectively. T₂ fishes had the highest growth rate and weight (75.03 g) and the control group had immature ova. Generally, both T₁ and T₂ group fishes had well developed ovary with eggs in Tertiary Vitellogenesis (TVO) stage on the 30th day. Therefore, shrimp head meal incorporated at (15%) diet can give better biological parameters of growth and gonad maturation in goldfishes.

Keywords: goldfish, growth, gonad development, shrimp head meal

1. Introduction

Fish meal is the major source of protein in formulated aqua feeds throughout the world^[11]. The availability of fish meal is in critical situation due to stagnation in capture fisheries and increased price^[11]. It has been felt necessary to identify an alternative protein source for preparing feeds has been felt and research is being focused on this line^[17]. There are two types of protein sources, viz., plant origin and animal origin. Plant protein sources such as cotton seed meal, soybean meal, duckweed, maize bran, rice bran, palm kernel cake, ground nut oil cake, brewers waste, pulses, legumes, etc. are commonly used^[35]. Presence of growth inhibitors such as alkaloids, glycosides, oxalic acids, phytates, protease inhibitors, haematoglutinin, momosine, cyanoglycosides and linamarin limit their use in feeds^[35]. There are many animal protein sources such as silkworm, maggot, termite, grub, earthworm, snail, tadpoles, etc that are being used in aqua feeds. The protein content of animal based products can be 54-70% while the plant based product it varied from 15 to 50%^[6]. Therefore, it is obvious that animal protein can have a better influence on growth and reproduction capacity in farmed animals^[27].

In shrimp processing plants, shrimp head is considered as a waste. But it is as an excellent animal protein additive in aquaculture due to rich protein, cholesterol, free fatty acids and mineral contents present in it^[3, 10] used to produce shrimp head meal. It is a by-product defined as an undecomposed, ground, dried waste of shrimp^[14] and used as a feed ingredient which can be used as an alternative protein source in aquafeeds^[3]. Harnessing of this waste into fish feed production will not only reduce the unit cost of fish production, but also would serve as an excellent means of preventing environment pollution. Various benefits of shrimp head meal as a nutrient source and palatable feed ingredient have been described by several authors^[6, 37, 38, 29, 33, 9, 21].

In India, there are 30-35 major freshwater ornamental fish species available for trades^[23]. Goldfish is one of the most important ornamental fishes for trade in international market^[22] and it is one among the popular ornamental fishes with high economic value in Japan^[25]. It enjoys wide distribution and demands high price.

Goldfishes are hardy in nature as they can survive in poor water quality also^[19]. It is an omnivore, which feeds on wide variety of live and artificial feeds. The natural diet consisted of animal matter like crustaceans, insects and their larvae, detritus and various plant matters which meet their protein requirement^[28]. Shrimp head meal has been recommended using in

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the diet of gold fishes as they could observe positive responses in terms of growth [7, 13]. Shrimp head meal has nutritional additives such protein and fatty acids, which can enhance gonad of cultured animals [1]. The above foregoing suggest possible use of shrimp head meal as an additive in fish feeds enhance growth and gonad development. The present study was used to find the influence of shrimp head meal on the growth and gonad maturation of goldfish at different concentration.

2. Materials and Methods

The experiment was carried out with replicates in the wet lab of Fisheries College and Research Institute, Thoothukudi TNFU. Experimental fishes were procured from Dev's Aqua Farm, Keeranoor in Thoothukudi District. The fishes were with the bodyweight of 3-5g and maintained in FRP (Fibre Reinforced Plastic) tanks. They were acclimatized for a period of 30 days under controlled condition with control feed. After acclimatization, goldfishes were separated for the feeding experiment. The initial body weight of the fishes stocked in the experiment was 5.79 ± 0.91 g. They were of 4 numbers per trough.

Shrimp head wastes were collected from processing plant and dried under sun after thorough washing. The unwanted shrimp appendages and detritus were removed and the dried shrimp head wastes were then ground in a dry grinder to make the powder and the powder was sieved to get uniform sized particles for homogeneous mixing [18]. All the feed items were sieved using a metal sieve of 200 μ size. The ingredients used

in control and experimental feeds and their inclusion levels are presented in Table 1. Rice bran was used as filler in the experimental feed.

The ingredients at the specified level were weighed in an electronic balance taken and mixed well. Except Vitamin E, fish oil and feed additives, all other ingredients were mixed well with water and made into dough. The dough was divided into 3 equal parts based on the weight. One part was kept as control feed, where no experimental feed additive was added. Other two parts were used for mixing shrimp head meal at 10% and 15% as given in Table 1. The feed balls were cooked in a pressure cooker for 15 minutes and air cooled. Fish oil and Vitamin E at the specified levels were mixed and the dough was kneaded thoroughly for evenly distribution of feed ingredients and other additives. Each feed ball was extruded separately using the manual pelletizer. The pelletizer was cleaned well after every individual feed and then the next feed ball was pelletized. The extruded pellets were then sundried and stored in a labeled airtight container [8].

The feed pellets were analyzed for their quality and proximate composition. Parameters such as moisture, crude protein, crude fibre, ether extract, total ash content and gross energy of experimental diets were determined by using standard analytical methods [2] in Animal Feed Analytical and Quality Assurance Laboratory, Namakkal. The proximate composition of experimental feeds is given in Table 2. The feeds were isoproteinecious ($32.19 \pm 0.25\%$) and isocaloric (3944 ± 59.54 kcal/kg).

Table 1: Feed ingredients (%) of inclusion in different treatments

Sl. No	Ingredients	Control (CF)	Shrimp Head Meal T ₁ SHM 10% (T ₁₁)	Shrimp Head Meal T ₂ SHM 15% SHM 15%
1	Ground nut oil cake	20	20	20
2	Rice bran	50	40	35
3	Wheat bran	5	5	5
4	Maize flour	7.5	7.5	7.5
5	Salt	0.1	0.1	0.1
6	Vitamin & mineral mix	0.1	0.1	0.1
7	Fish meal	15	15	15
8	Fish oil	2	2	2
9	Yeast	0.1	0.1	0.1
10	Vitamin E	0.2	0.2	0.2
11	Shrimp Head Meal	0	10	15
	Total	100	100	100

Bio-growth parameters such as growth, survival, feed consumption rate, weight gain (g), weight gain percentage and specific growth rate were estimated based on the body weight [38]. Sample fishes were sacrificed to observe Gonado Somatic Index before starting the experiment. Further by development of gonad was observed at 30th and 60th day of the experiment scarifying the animal. The GSI was calculated using the

formula given by Billard *et al.* (1993). The fishes were dissected and gonads were taken out with the help of forceps [16]. The collected gonads were preserved in 10% formalin [15]. The preserved gonads were sliced and then mounted in slides. The histological sections of gonad were observed under microscope to study the development of gonads at 30 days interval period during the experiment.

Table 2: Proximate composition of experimental feeds

Sl. No	Proximate composition	T ₁	T ₂	CF	Mean \pm SD
1	Crude protein (%)	32.22	31.93	32.43	32.19 ± 0.25
2	Crude fibre (%)	11.26	9.99	6.28	9.17 ± 2.58
3	Ether Extract (%)	6.48	5.16	4.32	5.32 ± 1.08
4	Gross energy (kcal/kg)	4011	3924	3897	3944 ± 59.54

The collected data and the estimated parameters were subjected to 'F' test for finding out statistical significance. The growth parameters were analyses using Regression analysis and compared using ANOVA.

3. Results and Discussion

The feed utilization pattern for the feeds used in the experiment is presented in Fig 1. Higher feed consumption was observed in fishes fed with diet containing shrimp head

meal at 15% (T₂). This is statistically significant at 99% level when compared to control (C) and T₁ group fishes. As it is seen in fig 1, feeding pattern was highly irregular in treatments as well as control fishes, which is believed to have been due to the experimental condition and regular cleaning of tanks. After 30 days of experiment, there was a phenomenal increase in the feed utilization in all the groups. Fishes fed with diets containing 15% shrimp head meal utilized highest amount of feed (92.60%) as can be seen in Table 3 and depicted in Fig 1. The control group fishes showed lowest feed utilization (67.88%) which is in accordance with the report that the same effect was found in juveniles of *Litopenaeus schmitti* when it was fed with 15%

shrimp head meal containing diet [4].

The feed conversion efficiency of fishes followed the same order as that of feed utilization. However, the feed conversion ability does not to be proportional to the quantum of feed that is unutilized by the fish. The flesh conversion was the highest (81.02%) for shrimp head meal (15%) (T₂) incorporated diet (Table 3 and Fig 3). The percentage of feed converted into flesh in respect of *C. auratus* fed with control diet was 12.83% (Fig 3), while rest of the feed utilized is believed to have been spent for metabolic activities of fishes [26]. The same favorable effect of shrimp head meal in diet has been reported by researchers, who tried shrimp head meal at low levels [41].

Table 3: Feed consumption and conversion details of *C. auratus* fed with feeds containing shrimp head meal at two inclusion levels

Treatment	Total Initial Biomass (g)	Total Final Biomass (g)	Total Biomass produced (g)	Total feed given as dry matter (g)	Total fecal excretion as dry matter (g)	Total Feed Utilized (g)	Feed converted into flesh (%)
CF	45.69	54.4	8.71	108.38	40.90	67.88	12.83
T ₁	45.69	68.48	22.79	125.34	44.02	85.14	26.76
T ₂	45.69	120.72	75.03	147.26	48.20	92.6	81.02

Table 4: Bio- growth parameters of *C. auratus* fed with feeds containing shrimp head meal at two inclusion levels

Treatment	Mean initial weight (g)	Mean final weight (g)	Weight gain (g)	Weight Gain (%)	SGR (%)
CF	5.98±0.65	8.05±0.213	2.07±0.079	34.261±1.97	0.495±0.179
T ₁	5.98±0.65	8.56±0.39	2.58±0.385	43.13±0.86	0.598±0.268
T ₂	5.98±0.65	15.09±2.31	9.11±1.42	152.34±5.226	1.543±0.422

High inclusion level of shrimp head meal in the fish feed was found to reduce the digestibility of feed in fishes [29]. In the present study, shrimp head meal incorporated at 15% level gave higher feed consumption and utilization than the other groups indicating that inclusions levels higher than 15% might be of negative due to the presence of exoskeleton,

chitin and ash content [29]. The same result was observed in the study reported that inclusion of shrimp head meal at 17% level in the feed produced higher feed consumption and reduced digestibility [40] due to the presence of attractants such as amino acids, peptides, nucleotides and chitin in shrimp head meal could promote the feed consumption in fishes [26].

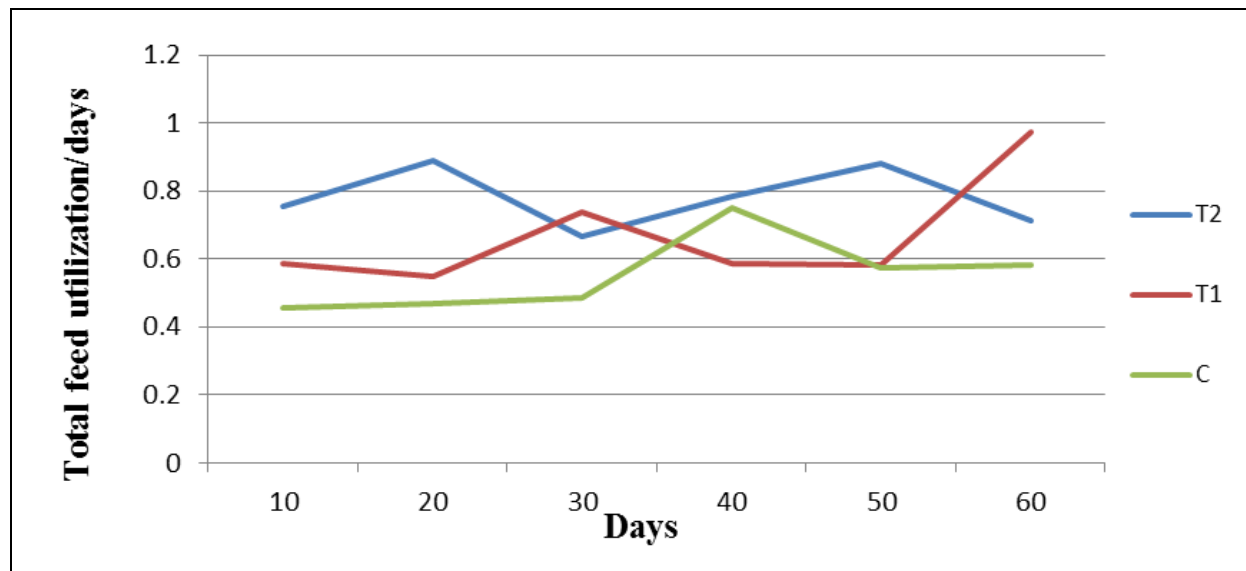


Fig 1: Feed utilization pattern in *C. auratus* fed with feed containing shrimp head meal at two inclusion levels

3.1 Effect of shrimp head meal on the growth of *C. auratus*

The weight gain percentage estimated for goldfishes fed with diet containing shrimp head meal at 15% (T₂) concentration positively affected the growth performance leading to highest weight gain percentage (152.34±5.22%). This is in accordance with the study in Tilapia when fed with feed containing shrimp head silage at 15% level [32]. The same

result also reported in African catfish when fed with diet included the fish meal was replaced with 20% of shrimp head meal which showed significantly higher growth than 30% and 40% replaced diet fed fishes [30]. They also reported that certain amount of shrimp head meal in the diet can improve the growth performance.

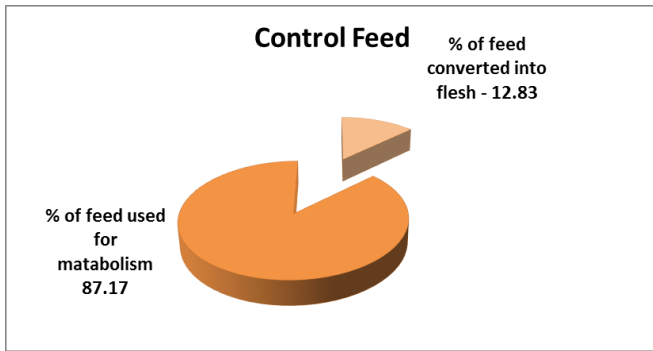


Fig 2: Feed conversion in *C. auratus* fed with control feed

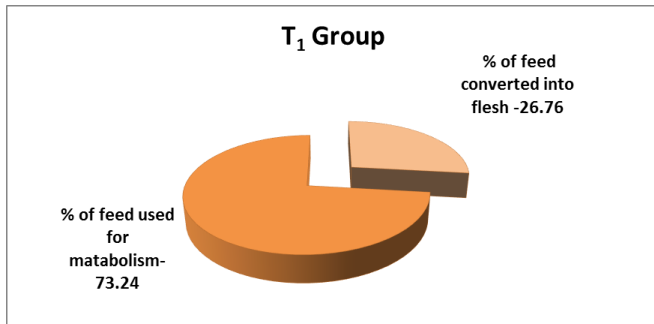


Fig 3: Feed conversion in *C. auratus* fed with diet containing shrimp head meal at 10% level

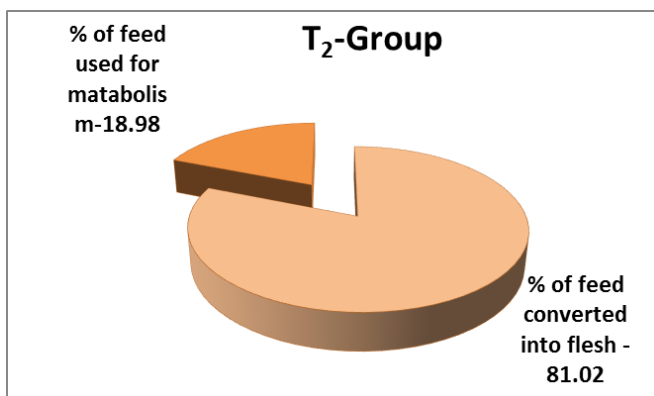


Fig 4: Feed conversion in *C. auratus* fed with diet containing shrimp head meal at 15% level

The specific growth rate was also of the same order as that of weight gain percentage. The higher quantities of lysine and arginine in the muscular protein of shrimp waste could improve the growth rate of fishes as observed in many fishes. This result was reported in shrimp that the lesser level of inclusion of shrimp head meal than 15% in the diet reduced the protein availability consequently it reduced the growth of fishes [1]. These findings are found true in the present study.

3.2 Influence of different diets on the development of ovary in *C. auratus*

At the commencement of the feeding experiment all the fishes had lowest GSI (2.68%) mostly immature eggs were noticed in the ovary (Plate I-a). In Control group, there was less development (GSI 2.11%) as evident from the same Chromatin Nucleolus (CNS) stage of the ova much similar to that of day 1 and 30 (Plate-I a).

The weight of the gonad and GSI observed on 30th day of experimental fishes are given in Table 5. As it is evident, the highest gonad weight and GSI (11.20%) were observed in fishes fed with 15% shrimp head meal containing diet (T₂).

The histological observation of the female gonad from T₂ diet fed fishes (Plate-I b-ii) had ova partially at Primary Vitellogenesis (PVO) stage characterized by the presence of lipid vesicles spread throughout the ova with cytoplasm in the periphery without yolk granules and remaining at Chromatin Nucleolus Stage (CNS) characterized by presence of ova small in size with large nucleus.

Table 5: Observations on the weight of the gonads of *C. auratus* on day 30 when fed with feeds containing Shrimp head meal at two inclusion levels

Treatment	Weight of the fish (g)	Weight of the gonad (g)	GSI (%)
CF	7.74	0.164	2.11
T ₁	8.50	0.291	3.42
T ₂	8.79	0.985	11.20

Fishes fed with diet contained shrimp head meal at 10% had lower GSI (3.42%) than T₂ group fishes but had more distinct development in their ova. The ovary contained ova in different stages of maturation but the majority in Tertiary Vitellogenesis stage (lipid vesicles are pushed to a thin layer in the periphery with yolk granules occurring 80% of the ova space) (Plate-I b-i). It was explained by the statement that goldfish had an asynchronous type of maturation in which ovary contains ova at various stages of development [24]. In the present study, highly developed ova with low GSI in T₁ (3.42%) group fishes and T₂ group fishes had medium developed ova with highest GSI (11.20%) due to the presence of nutritional additives such as vitamin, minerals and fatty acids [1] for aquatic organisms which can enhance gonad in present study.

There is no report on early maturation of fishes through feeding with shrimp head meal at any specific concentration. In the present study it was observed that shrimp head meal contained diet at 10% level had enough nutrients to induce maturing of goldfishes within 30 days of feeding. The presence of exoskeleton and chitin content through cooking or fermentation can increase the protein level with excellent amino acid profile which can increase the maturation in fishes [12]. It is also reported that higher dosage of shrimp head meal in the diet increased high level of chitin and ash, which were considered as limits the nutrient for the maturation in fishes [26].

On the 60th day of the experiment, the control group had less development in the ovaries, which were small in size with large nucleus and undifferentiated cytoplasm. There were no yolk granules or lipid vesicles visible in the cells under microscope indicating the Chromatin Nucleolus (CNS) stage or early immature stage of the ova. This indicates that there was no marked development of eggs in the ovary. This shows that feed additives had stimulants which helped to mature earlier in fishes [20, 34].

Table 6: Observations on the weight of the gonads of *C. auratus* on day 60 when fed with feeds containing shrimp head meal at two inclusion levels

Treatment	Weight of the fish (g)	Weight of the gonad (g)	GSI (%)
CF	9.805	0.415	4.23
T ₁	8.12	0.810	9.97
T ₂	14.21	1.140	8.02

The gonad weight and GSI of experimental fishes observed on 60th day of experiment are presented in Table 6. As it can

be seen, the highest GSI was observed in fishes fed with diet containing Shrimp head meal at 10% (T₁) level followed by diet containing Shrimp head meal at 15% level (T₂). In contrast, except control all the fishes in groups (T₁ & T₂) had exhibited marked development in their ovaries (Plate I c). There was more number of ova at this stage indicating the fully ripped ovary in the case of goldfishes fed with diets containing shrimp head meal at two different inclusion levels. Nucleus in the centre with spherical shape indicates that eggs

are in further developing stage to Tertiary Vitellogenesis (TVO) stage. Disappeared lipid vesicles also further vouch for the stage due to the presence of Chitin (N-acetyl-D-glucosamine) which is known to enhance the gonad maturation [1]. It is explained in the study reported that fishes fed special feed with continue rearing period also increased nutrient availability. This also believed to enhance the maturation in fishes [39].

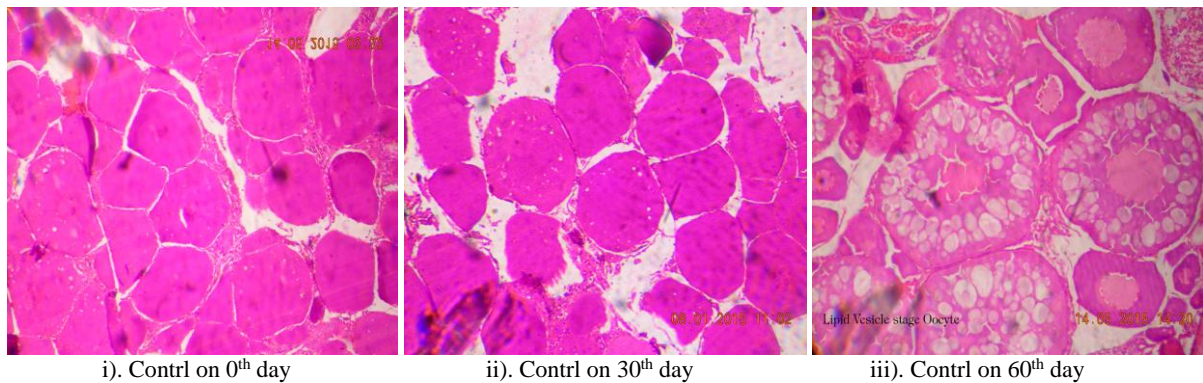


Plate I: a) Control group

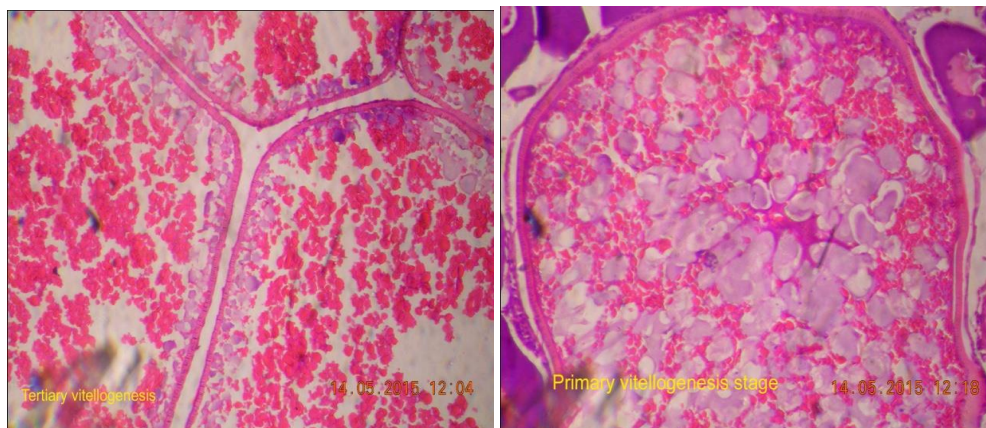


Plate I: b) shrimp head meal on 30th day

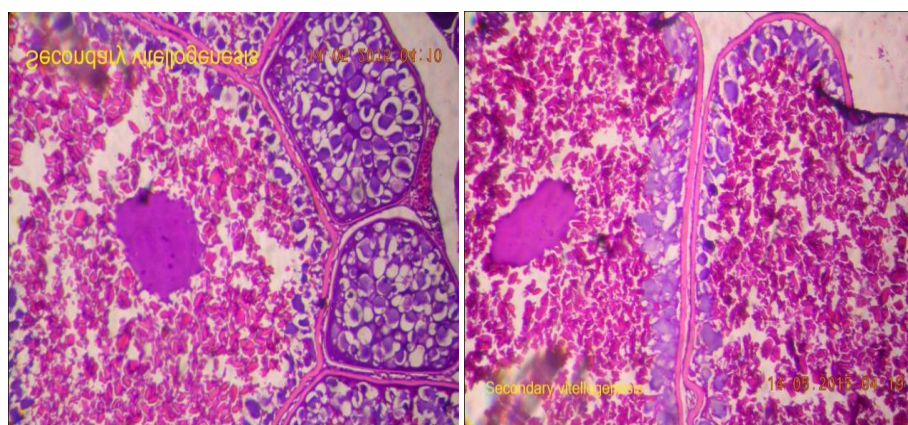


Plate I: c). Shrimp head meal on 60th day

4. Conclusion

From the present study, it can be concluded that shrimp head meal included diet at 10% and 15% level can improve the performance of fish in terms of growth and gonad maturation respectively at 30th day of experiment when compared to Control. Hence, shrimp head meal incorporated feed can be

used for growth and earlier maturation of fishes on 30th day. There is no need to fishes fed with shrimp head meal at 10% level. Reduced inclusion level of shrimp head meal at less than 10% also give better result after 30th day. It can reduce feed cost also.

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