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Impact of *Tulsi* Ingredients on Gonadal Development of Common Carp, *Cyprinus Carpio* under Tarai Region of Uttarakhand

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ABSTRACT

The present study was conducted for comparing the effect of *Tulsi* ingredients, Eugenol and Ursolic acid on the gonadal of common carp (*Cyprinus carpio*) under Tarai condition of Uttarakhand. The results revealed that role of Ursolic acid in enhancing gonadal development and effect of Eugenol in decreasing gonadal development in *C. carpio*. Indicators of reproductive profile (GSI, fecundity and sperm count) exhibited more potency of Ursolic acid and less potency of Eugenol in gonadal differentiation and development. These observations indicate that Eugenol and Ursolic acid can also be used for selective enhancement of gonadal development depending upon the requirement.

1. Introduction

The carp (*Cyprinus carpio*), is one of the most extensively translocated and domesticated fish species in the world. Carp originated in central Asia and spread east and west to China and the Danube (Balon, 1974). The species was successfully spread throughout Asia and Europe, was domesticated as an ornamental and aquaculture species. Demand for fish exceeds production and per capita consumption is steadily increasing day-by-day. To enhance fisheries production it has become a necessity to increase fisheries production by developing effective technologies for ensuring maximum output from minimum input and to get maximum profit. Strategies for increasing fish production include quality stocking material, water quality management, adequate balanced diet and incorporation of various additives. There are large number of feed additives available to improve fish growth performance some of these additives used in feed mill are chemical products especially hormones and antibiotics may cause unfavorable side effects (Bello *et al.*, 2012). Basil *(Ocimum basilicum* L.) is one of the most famous, annual or perennial herb belonging to the family Lamiaceae.

Eugenol and Ursolic acids are the two ingredients of tulsi plant responsible for growth of fishes (Prakash and Gupta, 2004). Eugenol is a phenolic compound and major consistuent of the essential oils extracted from different parts of Tulsi plant (Boyer *et al*., 2008). Ursolic acid is a triterpenoid compound which exists widely in natural plants in the form of free acid or aglycones for triterpenoid saponins (Pemminati *et al*., 2011). Previous researches have confirmed that tulsi is cheap and easily available medicinal plant and have effect on fertility of fishes. The present study have to aim to observe the effects of the active ingredients of tulsi, Eugenol and Ursolic acid, on the gonadal development of the fish *Cyprinus carpio.*

2. Materials and Methods

Hundred specimens of common carp, obtained from the instructional fish farm of College of Fisheries, Pantnagar. Ten aquaria 4′x1′x1′ were divided into five groups having one replicate for each group. Six specimens were put from the acclimatized fish after recording length and body weight. Diet was prepared by mixing Eugenol and Ursolic acid with the conventional diet in different concentrations. Feeding was done @ 5% body weight twice daily.

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Short-term treatment (7days) was carried out to observe the acceptability of diet supplemented with Eugenol and Ursolic acid and its effect on survival and behaviour of fish. Long-term treatment (60days) was recorded in response to feeding of different concentrations of Eugenol and Ursolic acid for sixty days. The length-weight data of each specimen from all experimental groups was recorded separately at the time of first sampling. All other parameters were recorded for each group of fish separately after sixty days at the time of termination of the experiment. Paired't' test was used for comparing length and weight of common carp while Statgraphics statistical package was used for analyzing GSI, HSI, Fecundity, sperm count, motile and non- motile sperm.

3. Results and Discussion

3.1 Gonadosomatic index (GSI)

The GSI was shown in table 1. Among the feeding groups, maximum GSI was obtained in T_2 (male-27.22 and female-27.67) group followed by T_1 (male-22.4 and female-23.7) group while minimum GSI in T_3 (male-4.55 and female-4.17) group followed by T_4 (male-3.08 and female-3.44) along with control (male-8.79 and female-12.00) in both sexes. However, the differences among the treatments was significant.

3.2 Hepatosomatic index (HSI)

The HSI was shown in table 1. Comparison among the feeding groups exhibited maximum HSI for T_1 (male-0.82 and female-0.69) group followed by T_2 group (male-0.64 and female-0.44) while minimum in T_3 (0.21) followed by T_4 (0.16) group in male and T_4 (0.18) followed by T_3 (0.16) group in female. The difference among the treatments was significant.

3.3 Fecundity

The fecundity was shown in table 1. Relative fecundity ranged from 2966 to 5018, with maximum (5018) in T_2 and minimum (995) in T_4. The effect of different diets on fecundity was significant.

3.4 Sperm count

The sperm count was shown in table 1. Sperm count ranged from 62.30 to 11.58, maximum (62.30) in T_2 and minimum (11.58) in T_4. The effect of different diets on sperm count was significant.

3.5 Motile Sperm

The motile sperm was shown in table 1. Motile sperm ranged between 46.83 to 3.58, with maximum (46.83) in T_2 and minimum (3.58) in T_4. The effect of different diets was significant.

3.6 Non motile sperm

The non motile sperm was shown in table 1. Non motile sperm ranged from 23.22 to 8.89, with maximum (23.22) in T_0 and minimum (8.89) in T_4. The effect of different diets was significant. Comparison between GSI, HSI, fecundity, sperm count, motile and non-motile sperm indicated that significant gain in Ursolic acid fed fishes. However, these parameters decrease in Eugenol fed fishes. Indicators of reproductive profile exhibited more potency of Ursolic acid fed fishes and less potency of Eugenol fed fishes in gonadal development. Maximum GSI, fecundity and sperm count were registered higher in Ursolic acid fed fish and minimum values recorded in Eugenol fed fish. These parameters are indicator of gonadal development of fish. The findings are in line with those of Boyer *et al*. (2008) who reported that Eugenol decrease growth and occurrence of bleaching in three species of corals. Prakash *et al*. (2004) reported that Eugenol possesses properties of anti-fertility effects and Ursolic acid increases spermatogenesis and sperm count because of its estrogenic effects. In this study, diet T_2 supports higher gonadal development (GSI, fecundity and sperm count) as compared to other diets. So, Ursolic acid fed fish are hence also expected to grow bigger in the subsequent years and support higher GSI, fecundity and egg size as compared to fish fed on other diets. Since the larger eggs carry more yolk which usually result in larger larvae with a capacity to thrive longer under unfavorable condition of food and physical environment. Hence Ursolic acid appears to be best in terms of both fecundity and sperm count, which are the basic parameters in quality brood stock production. Eugenol can also be used for selective enhancement of growth or gonadal development depending upon the requirement.

Table 1. Status of Gonadal development during rearing period in long term treatment with common carp

Parameters	Treatments									
	T_0		T_1		T_2		T_3		T_4	
	Female	Male	Female	Male	Female	Male	Female	Male	Female	male
Hepatosomatic Index	0.48^{a} ±0.08	0.21^{b} ±0.02	0.69^{a} ± 0.07	0.82^{a} ± 0.05	0.44^{ab} ± 0.86	0.64^{a} ± 0.76	0.16^{c} ± 0.02	0.21^{b} ± 0.02	0.18^{bc} ± 0.03	0.16^{c} ± 0.01
Gonadosomatic index	12.00^{b} ±1.15	8.79^{b} ±0.18	23.70^{a}± 1.75	22.45^{a} ± 0.60	27.67^{a} ± 2.68	27.22^{a} ±4.22	24.17^{c} ±0.60	4.55^{b} ±0.11	3.44^{c} ±0.16	3.08^{b} ±0.30
Fecundity (per g of ovary)	2966.60^{b} ± 220.96		4672.90^{a} ± 151.20		5018.60^{a} ± 100.22		1525.89^{c} ± 189.3		995.20^{c} ± 34.30	
Sperm count(per ml of milt)	-	33.18^{c} ± 1.77	-	48.59^{b} ± 0.97	-	62.30^{a} ± 1.26	-	15.15^{d} ± 1.44	-	11.58^{d} ±0.40
Motile sperm (per ml of milt)	-	9.95^{c} ± 0.76	-	32.96^{b} ± 1.18	-	46.83^{a} ± 1.75	-	5.40^{cd} ± 0.86	-	3.58^{d} ±0.45
Non motile sperm (per ml of milt)	-	23.22^{a} ± 1.86	-	16.46^{b} ± 1.21	-	14.76^{bc} ± 1.03	-	9.76^{cd} ± 0.81	-	8.89^{d} ±1.03

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