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NUTRITIONAL EFFICACY OF *CENTELLA ASIATICA* SUPPLEMENTED DIET ON *MACROBRACHIUM ROSENBERGII*

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Abstract: Aquaculture is under the threat of diseases outbreaks due to intensive culture. Various biotic and abiotic factors within intensive culture systems of *M. rosenbergii* challenge the health of cultured populations. Failure to control them leads to feeble even disastrous production losses. Nutritive feed along with antimicrobial property is effective against microbial diseases. Numerous medicinal plants are currently being used as feed additive in aquaculture. The present study was undertaken to assess the nutritional efficacy of medicinal plant *Centella asiatica* as a aquafeed supplementation . The effect of feed supplementation -250mg, 500mg, 750mg of methanolic extracts of medicinal plants were incorporated per kilogram of basal feed and was assessed at two levels of feeding schedule. One set of prawns were fed medicinal extract supplemented diet daily. The second sets of prawns were administrated supplemented diet twice a week. Each experimental set had 20 prawns /1000L tanks. The experiment continued for 60 days. Samples are collected for processing on 30th and 60th days. Daily removal of 50% of water and fecaces were done.The parameters such as % survival, weight gain(g), length gain(cm), feed conversion ratio(FCR), specific feed consumption(SFC)and protein efficiency ratio(PER) were checked for each treatment. In addition to that microbial load at non-specific immune sites such as gut, exoskeleton and gill were also checked. Results of 500mg/kg (daily administration) treated prawns revealed a notable improvement comparable to controls. The parameters assessed for 250mg/kg (daily administration) feed schedule expressed better feed utilization and growth compared to 250mg/kg(twice a week administration).the antimicrobial efficacy of the feed was evaluated to substantiate the nutritional status in terms of growth efficacy of the experimental prawns. The microbial load at exoskeleton, gut and gill was recorded as cfu/gm tissue. The antimicrobial data was better for the daily treated prawns compared to twice a week treatments. The observations have been statistically analyzed and discussed in the view of available current literature.

Key words: *M. rosenbergii, Centella asiatica*, Feed conversion ratio, Specific feed consumption, Protein efficiency ratio, Medicinal plant

INTRODUCTION

Aquaculture is the most progressive food sector in the world and it is the major source of income and dietary protein in most of the developed countries, especially Asian countries. Unscientific and irregular intensive culture will leads to the production loss at high level. Disease progresses more rapidly and results in higher cumulative mortality, in particular at early stages of development and especially shellfish (Tommy *et al.,* 2013).

Freshwater prawn *Macrobrachium rosenbergii* is one of the best candidates of cultural importance. Vietanam, China, Taiwan, Bangladesh, Thailand and India are the major countries of prawn farming. Fresh water prawn is also good for polyculture, along with other farmed fishes. *Macrobrachium rosenbergii* lives in freshwater, that are influenced by nearby brackish water, because it's larval life needs it . *Salmonella sp., Shigella sp, Staphylococcus auerus, Vibrio sp, Flavobacterium sp.* are the main microbes found in the penaeids (Abu Hena *et al*., 2008). Antibiotic cannot be used as a long term remedy. Drug resistance is an off shoot of antibiotic application. Oxytetracycline Hcl, Sulfamerazine, Sulfadimethozine Ormetoprim combination, Tricaine Methanesulfonate (Ms-222) are some of the antibiotics commonly used in aquaculture (Charles, 2002). Biomagnification of antibiotics is dangerous to human also.

Certain ayurvedic herbal compounds are having potent effect on growth and survival as well as antimicrobial properties in aquatic organisms. Numerous herbal products are added as feed additives in aquaculture

traditionally. *Withania somnifera, Mucuna pruriens,Myristica malabarica, Mimosa pudica, Ipomea digitata, Asparagus racemosa Leaves, Hygrophila spinosa, Phasedus roxburghii Moringa tinctoria and Hemidermus indicus* are some of the tradionally used plants in aquaculture (Sankar Murthy and Kiran, 2013)In the present study *Macrobrachium rosenbergii* is supplemented with diet containing *Centella asiatica* at selected concentrations.

MATERIALS AND METHODS

Experimental lay out

Macrobrahium rosenbergii of size (12.4 ± 0.8 mg) were purchased from ADAK (Agency for Development of Aquaculture (ADAK), Varkala, kerala**.** Acclimated in lab under optimum salinity, pH and temperature.

Experimental tank were set with proper aeration and water quality (Trivedi and Goel, 1984). Each experiment was done in triplicates. Experiment was conducted for 60 days. Feeding scheduled twice a day.

Feed preparation

Feed was prepared by using the ingredients listed in the Table 1. The methanol extract of *Centella aisatica* was obtained by extracting 50g of dried plant powder in 100ml 80% methanol and kept on a rotary shaker for 48 hour. The extract pooled after 24hrs and 48 hrs as vacuum filtered, and dried in an oven at 40 °C and was stored at 4 °C in airtight bottles till further use. (Deshmukhe and Borle. 1975).

Table 2. Rearing conditions and water quality parameters of the experimental setup

Parameters	mean ±SD
Stocking density	20 (per tank)
Tank capacity	1000 L
Feeding level	10–15% body weight
Feeding frequency	Twice daily
Feeding period	60 days
Temperature	28 ± 2°C
pH	7.05 ± 0.4
Dissolved Oxygen	4 mg/l

1. Growth parameters and Feed utilization parameters

   Growth parameters and Feed utilization parameters were evaluated as outlined in one –of- our previous works by (Jasmine *et al*., 2011).

2. Microbial load

   Microbial loads at non- specific immune sites such as exoskeleton, gut and gill were processed and analysed as per the protocol described by (Kannan, 2003). The bacterial loads are reported as cfu/gm tissue.

3. Statistical analysis

   Data were statistically analysed ,mean and standard deviation.level of significance were also found.

Table 1. Ingredients of experimental feed

Ingredients (gm/kg)	Control	Test diet 1	Test diet 3	Test diet 5
• Fish Meal	300	300	300	300
• Prawn Head	50	50	50	50
• Squid Meal waste	50	50	50	50
• Squilla	50	50	50	50
• Soyabean Meal	250	250	250	250
• Wheat Flour	250	250	250	250
• Fish Oil	30	30	30	30
• Vitamin Mineral Mixture	20	20	20	20
• *Centell asiatica* extract	-	250	500	750

Test diet 2,Test diet 4 and Test diet 6 have same concentrations of *Centella asiatica* as Test diet 1,Test diet 2 and Test diet 5 respectively.only the feeding schedule is different. Test diet 2,Test diet 4 and Test diet 6 have twice a week administration of experimental feed.

RESULTS AND DISCUSSION

Table 3 showing the growth parameters of the experimental prawns. Test diet 3 have high survival rate.it is daily administration of 500mg/kg of *Curcuma longa* followed by Test diet 2. The least survival rate is found in Control diet. Weight gain also high in Test diet 3 follwed by Test diet 1 and then Test diet 2. Least were found in Control diet. In the case of length gain is high in Test diet 1, it is a daily administration of 250 mg/kg of *Curcuma longa.* Least length gain is found in Control diet. These values are significant at ($p>0.01$)

Table 4 shows the feed utilization parameters of experimental prawns. FCR is high Test diet 2 followed by Test diet 4 and then least FCR were found in Test diet 1. Test diet 3 have better SFC. Least SFC value were found in Control diet. PER is high in Test diet 3, followed by Test diet 5. Least PER were found in Control diet. These values are significant at ($p>0.05$)

Table 5 and 6 showed the microbial load at certain non-specific immune sites such as exoskeleton, gut and gill of *Macrobrachium rosenbergii*. Here the microbial load is high in Test diet 3 followed by Test diet 1 and then test diet 3. More microbial load was found in daily medication. Twice a week feeding showed less microbial reduction. Microbial population at non-specific immune sites showed marginal varations. Here least reduction is found in the test diet 4. All of these values showed that daily medication of *Centella asiatica* is better. Test diet 3 with 500mg/kg of *Centella asiatica* showed good effects as compared to other test diets.

Centella asiatica is traditional medicinal plants. Which is indigenous to most of the Asian countries. They numerous properties such as antimicrobial, antifungal, antioxidant and anti-inflammatory properties. Now a day's application of herbal medicines is high as feed additives. These will reduce the application of antibiotics against microbial diseases. Herbal extracts can be used in fish culture as alternatives to vaccines, antibiotics or chemotherapeutic agents. They work together with nutrients and dietary fibre to protect the body against diseases(Pepsi *et al.*, 2012).

Table 3. Growth parameters of *Macrobrachium rosenbergii* suppelemented with *centella asiatica*

Sample	% Survival		Weight gain(g)		Length gain(cm)	
	30^{th}	60^{th}	30^{th}	60^{th}	30^{th}	60^{th}
Control diet	75±0.10	30±1.00	0.26±0.02	0.47±0.01	0.28±0.01	1.93±0.04
Test diet 1	88±0.58	80±1.00	1.00±0.03	2.12±0.02	2.10±0.01	3.60±0.02
Test diet 2	84±1.52	82±1.53	1.10±0.01	2.01±0.01	2.49±0.01	3.01±0.01
Test diet 3	87±1.00	85±1.00	2.00±0.02	2.80±0.01	2.85±0.02	3.08±0.02
Test diet 4	87±2.31	71±1.00	1.09±0.02	1.91±0.02	2.32±0.28	2.8±0.02
Test diet 5	82±1.00	78±1.53	1.08±0.02	2.09±0.02	2.29±0.01	3.00±0.02
Test diet 6	87±1.53	80±2.00	0.40±0.02	2.01±0.01	0.98±0.02	2.06±0.02

Table 4. Feed utilization parameters *Macrobrachium rosenbergii* suppelemented with *centella asiatica*

Sample	Feed Conversion Ratio(FCR)		Specific Feed Consumption(SFC)		Protein Efficiency Ratio(PER)	
	30^{th}	60^{th}	30^{th}	60^{th}	30^{th}	60^{th}
Control diet	2.27±0.25	7.01±0.01	1.07±0.01	0.50±0.10	0.13±0.06	0.38±0.01
Test diet 1	2.56±0.05	5.97±0.15	2.45±0.03	1.01±0.01	1.02±0.01	3.04±0.05
Test diet 2	3.80±0.02	9.00±0.02	2.03±0.05	1.78±0.03	1.38±0.00	3.15±0.01
Test diet 3	3.00±0.02	3.09±0.02	1.99±0.02	2.10±0.02	3.03±0.00	4.02±0.00
Test diet 4	3.43±0.16	7.63±0.15	1.65±0.01	0.91±0.01	1.02±0.00	2.01±0.00
Test diet 5	7.00±1.00	5.67±1.52	0.21±0.01	0.80±0.02	3.01±0.00	3.82±0.00
Test diet 6	6.67±1.53	7.00±1.73	0.20±0.03	0.81±0.01	0.89±0.01	1.02±0.00

Table 5. Microbial load at certain non-specific immune sites such as exoskeleton,gill and gut of *Macrobrachium rosenbergii* reared on daily supplementation of *Centella asiatica*.

	Control diet			Test diet 1			Test diet 3			Test diet 5		
	Exo*	Gut	Gill	Exo*	Gut	Gill	Exo*	Gut	Gill	Exo*	Gut	Gill
1st	$120x10^5$	$112x10^5$	$118x10^5$	$150x10^5$	$120x10^5$	$128x10^5$	120x105	100x105	118x105	$120x10^5$	$110x10^5$	$130x10^5$
30th	$115x10^5$	$124x10^5$	$120x10^5$	$99x10^3$	$84x10^3$	$78x10^3$	$88x10^2$	$72x10^2$	$60x10^2$	$100x10^4$	$80x10^3$	$90x10^3$
60th	$170x10^5$	$140x10^5$	$148x10^5$	180	185	138	170	160	156	200	194	306

Table 6. Microbial load at certain non-specific immune sites such as exoskeleton,gill and gut of *Macrobrachium rosenbergii* reared on twice a week supplementation of *Centella asiatica*.

	Control diet			Test diet 2			Test diet 4			Test diet 6		
	Exo*	Gut	Gill	Exo*	Gut	Gill	Exo*	Gut	Gill	Exo*	Gut	Gill
1st	$120x10^5$	$112x10^5$	$118x10^5$	$150x10^5$	$120x10^5$	$138x10^5$	$108x10^5$	$99x10^5$	$120x10^5$	$128x10^5$	$110x10^5$	$120x10^5$
30th	$115x10^5$	$124x10^5$	$120x10^5$	$100x10^3$	$89x10^3$	$28x10^3$	$98x10^4$	$88x10^4$	$38x10^4$	$99x10^3$	$89x10^3$	$82x10^3$
60th	$170x10^5$	$140x10^5$	$148x10^5$	190	195	148	$20x10^2$	380	$18x10^2$	184	450	$12x10^2$

Jagtap *et al.* (2009) deals with the evaluation of extract of plant of *Centella asiatica* for antimicrobial activity. As per data obtained the growth of inhibition of four bacterial strains and three fungal strains varied largely. *Centella asiatica* is traditionally used as antibacterial, antifungal and antioxidant properties. A work by Debajit and Jagat (2012) revealed the ethnomedicinal, antibacterial and Antifungal Potentiality of *Centella asiatica*. *Zingiber officinalis* and *Cyanodon dactylon* showed growth promotion in *macrobrachium rosenbergii (Hiam*El-Desouky *et al.*, 2012*)*.

CONCLUSIONS

From the present work it is evident that *Centella asiatica* have better antibacterial effect also can be used as a growth promoter. Daily administration of *centella asiatica* is more effective than twice a week application. In the daily administration 500 mg/kg of *Centella asiatica i*s more better which have more growth promotion, feed utilization and antibacterial properties compared with other experimental diets.

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