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GROWTH PROMOTING POTENTIAL OF GARLIC, GINGER, TURMERIC AND FENUGREEK ON THE FRESHWATER PRAWN *MACROBRACHIUM ROSENBERGII*

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ABSTRACT

A 60 day feeding trial was conducted to determine the growth performance of *Macrobrachium rosenbergii* post larvae (PL) on *Allium sativum* (Garlic), *Zingiber officinale* (Ginger), *Curcuma longa* (Turmeric) and *Trigonella foenum-graecum* (Fenugreek) incorporated diets. Survival rate (SR) ranged from 69.0-80.0% in experimental diets fed PL groups were recorded against 66.0% in control. The elevation in SR was in the order of garlic > ginger > turmeric > fenugreek. Similar trend in elevation of FR was recorded, which in turn increases the WG in experimental PL over control. The same patterns of improvements were observed in SGR, FCE, PCE, ME and GE. Concurrently, the values obtained for CF and FCR were in decreasing pattern (garlic < ginger < turmeric < fenugreek). This indicated the fact that the experimental feeds were superior in quality than that of control. Concentrations of total protein, amino acid, RNA, DNA, RNA/DNA and carbohydrate as well were found to be elevated in experimental diets fed PL when compared with control. Instead, herbal dependent utilization of lipid was noted in PL, and thus, the elevation of total lipid was recorded in just the reverse order, garlic < ginger < turmeric < fenugreek. All the data were significant at $P<0.05$. The herbal growth promoter induced transcription rate, which in turn increased RNA and total amino acid, which ultimately enhances proteins synthesis in *M. rosenbergii* PL. This ability was higher with garlic included feed followed by ginger, turmeric and fenugreek. Therefore, these herbs could be taken as supplementary materials in aqua feed formulation.

KEY WORDS: ***M. rosenbergii, Garlic, Ginger, Turmeric, Fenugreek, Survival, Growth, Protein, RNA/DNA.***

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INTRODUCTION

The giant freshwater prawn, *Macrobrachium rosenbergii* has received considerable attention as an aquaculture species because of its nutritious delicacy to mankind (Bhavan *et al*, 2010a). It is a prime inland cultured species, which has recently emerged as an important shellfish species for culture in south Asian countries after significant losses of penaeid shrimp culture in the mid-1990s due to viral diseases (FAO, 2003). In 2004, world total farmed *M. rosenbergii* reached more than 1,94,000 tonnes, with an estimated market value that exceeded US$810 million, of which 99% was produced in Asia (FAO, 2004). It has been reported that the world production of *M. rosenbergii* exceeded 4,00,000 tonnes in 2010, a more than 20-fold increase compared to the early 1990s (FAO, 2010). Diet is a fundamental aspect in culturing decapod crustaceans (Jones *et al*., 1997; Correia *et al*., 2000; Valenti and Daniels, 2000).

The understanding of crustacean nutritional requirements has increased over the last few decades; however, many gaps remain. Shrimp feeds still rely on fish meal as a key protein source despite its recognized problems of price and availability (Forster *et al*., 2003). The search for alternative protein sources of high nutritional quality has been the focus of several segments involved in aquaculture (Tacon and Barg, 1998; Bhavan *et al.,* 2010a,b, 2011a; Rebecca and Bhavan, 2011; Bhavan and Radhakrishnan, 2012). Furthermore, it is necessary to know what ingredients can cover the nutritional requirements at the lowest cost, considering the amount but also the quality of ingredients in formulation of the diet. The nutritive value of feeds depends on the individual components and palatability of the feed formulated. Inclusion of herbal nutrients in the feed will improve the palatability as well as digestibility of the feed (Bhavan *et al.* 2011b; Shanthi *et al.* 2012). Herbal supplements will also aid to improve prawn health and positively reflect on the product quality. Herbs also possess other interesting properties like non-toxic, biodegradable and biocompatible components/ substances (Citarasu *et al.*, 2003). Herbs have also been used for controlling shrimp and fish diseases (Auro de Ocampo and Jimenez, 1993; Dey and Chandra, 1995; Direkbusarakom *et al*., 1996; Dugenci *et al.,* 2003). Additionally herbs have received more attention recently not only for their immune stimulating functions but also for their growth promoting effects. Supplementation of *Gynostemma pentaphyllam* (a traditional Chinese herb) to grass carp feed resulted in increased weight gain, feed conversion efficiency and specific growth rate (Wu *et al.,* 1998; Venkataramani *et al.,* 2002; Anh *et al.,* 2009). Similarly, *Ocimum sanctum*, *Withania somnifera, Andrographis paniculata*, *Cissus quadrangularis* and *Eclipta alba* have produced better growth of *M. rosenbergii* PL (Bhavan *et al.* 2011b; Shanthi *et al.* 2012). In addition diets with high calorific value of protein, carbohydrate and lipid have drawn the attention (Joshi *et al.*, 1979). Proteins are the most prominent biochemical components of crustaceans from eggs to adult and are strikingly dominant in younger phase (Dinakaran and Soundarapandian, 2009).

In the view of the above, the present study dealt with incorporation/ supplementation of herbs, such as *Allium sativum* (garlic) bulb, *Zingiber officinale* (ginger) rhizome, *Curcuma longa Linn* (turmeric) rhizome and *Trigonella foenum-graecum* (fenugreek) seed with diets because they are routinely used in our kitchen. Garlic, ginger, turmeric and fenugreek have culinary and medicinal properties. The derivatives of garlic, such as essential oil and oleoresin possessed antioxidant and antimicrobial properties (Sharma and Prasad, 2001). Ginger increases the pancreatic and intestine lipase (Platel and Srinivasan, 2000). Turmeric is used for wound healing, inflammation and acidity (Jyothi *et al*., 2003; Kumar *et al*., 2006). Fenugreek seed possesses aphrodisiac and digestive effects (Chopra *et al.,* 1982; Fazli and Hardman, 1968). It has high potential to be used as a cheap source of alternative protein supplement (Nasri and Tinay, 2007). The active principles and medicinal properties of these herbs are also given in table-1. As herbs have

characteristic ability to enhance growth rate and immune functions, the present study was conducted to determine the influence of garlic, ginger, turmeric and fenugreek on feed utilization for growth promotion and accumulation of body biochemical constituents, such as protein, amino acid, RNA, DNA, carbohydrate and lipid in *M. rosenbergii* PL.

MATERIALS AND METHODS

M. rosenbergii (PL-5) were procured from Rosen Fisheries (Thrissur, India), transported to laboratory in well-oxygenated plastic bags, stocked in 1000 l tank and acclimated in ground water (pH - 7; temperature - 28° C; total dissolved solids - 1200 mg L^{-1}; dissolved oxygen - 7.2 mg L^{-1}; BOD - 30 mg L^{-1}; COD - 125 mg L^{-1}; ammonia -1.8 mg L^{-1} (APHA, 2005) for two weeks. During acclimatization the PL were feed *ad libitum* with boiled egg albumin, live *Artemia* nauplii, commercial scampi feed alternatively thrice a day.

The herbs, such as garlic (*A. sativum*) bulb, ginger (*Z. officinale*) rhizome, turmeric (*C. longa*) rhizome and fenugreek (*T. foenum-graecum*) seed were purchased from the local market. The fresh herbs were washed thoroughly with tap water, shade-dried at room temperature (25ºC) and powdered. The basal ingredients, such as fishmeal, soybean meal, wheat bran, rice bran and groundnut oil-cake were purchased from local merchants, sun dried adequately and ground separately using a micro pulverizer. Powdered feed ingredients were weighed out to prepare a feed mix based on Pearson square method equated to 40% protein. The feed mix (basal ingredients) was steam-cooked for 5 minutes at 95-100ºC and allowed to cool at room temperature, before pelletization addition of Cod liver oil, binding agents (egg albumin and tapioca flour) and vitamin B-complex with vitamin-C was done and thoroughly mixed. Actually, the basal diet consisted of groundnut oil cake (20g), soy bean meal (20g), fish meal (20g), wheat bran (13g), rice bran (13g), egg albumin (5g), tapioca flour (5g), Cod-liver oil (3 ml) and vitamin B-complex mix (1g). A diet formulated with 100% of these ingredients was served as control. The proximate biochemical compositions of the basal diet (% on dry wt. basis (n=3) are as given below: moisture, 9.24±0.11; crude protein, 37.47±1.42; crude fat, 6.60±0.78; crude fibre, 8.18±0.94; ash, 14.40±1.21; nitrogen free extract, 33.24±4.32 (AOAC, 1995). Each herbal powder (1%) was separately added and mixed with 99% of basal diet and pelletized.

Five groups of PL (mean weight 0.084 ± 0.03 g and mean length 1.92 ± 0.02 cm) in triplicate were randomly allowed to distribute at the rate of 25 each in flat bottom circular tanks of 20 L capacity. PLs were fed with respective control and experimental diets twice daily at 07:00 AM and 07:00 PM for a period of 60 days at the rate of 10% of total body weight. The body weight of the control PL was measured once in 15 days and the diet weight was equated. The uneaten food was collected, dried with absorbent paper and weighed to calculate the feeding rate. Water was renewed daily and continuous aeration was supplied. The nutritional indices were calculated from the morphometric data by adopting the following formulae of Tekinay and Davis (2001).

$$\text{Survival Rate (\%)} = \frac{\text{No. of live prawns}}{\text{No. of prawns introduced}} \times 100$$

$$\text{Feeding rate (g d}^{-1}\text{)} = \frac{\text{Feed eaten}}{\text{Time (days)}}$$

Weight Gain = Final weight (g) – Initial weight (g)

Length gain = Final length (cm) – Initial length (cm)

$$\text{Specific Growth Rate} = \frac{\textit{log}\text{ of Final weight (g)} - \textit{log}\text{ of Initial weight (g)}}{\text{Time (No. of days)}} \times 100$$

$$\text{Condition Factor} = \frac{\text{Final weight (g)}}{\text{Final length}^3\text{ (cm)}} \times 100$$

$$\text{Feed conversion ratio} = \frac{\text{Feed given (dry wt. (g)}}{\text{Body weight gain (wet wt. (g)}}$$

$$\text{Food conversion efficiency} = \frac{\text{Body weight gain (wet wt. (g)}}{\text{Feed given (dry wt. (g)}} \times 100$$

$$\text{Protein conversion efficiency} = \frac{\text{Gain in muscle protein (g)}}{\text{Protein intake (g)}} \times 100$$

Metabolizable energy (kj g^{-1}) = 0.173 x protein (%) + 0.356 x fat (%) + 0.125 x carbohydrates (%)

Gross energy (kj g^{-1}) = 0.2364 x protein (%) + 0.3954 x fat (%) + 0.1715 x carbohydrates (%)

Muscle tissue concentrations of total protein, amino acid, carbohydrate and lipid were estimated on final day of feeding trial. Total protein content was determined by the modified method of Lowry *et al.,* (1951) as modified by Schacterle and Pollack (1973). Total amino acid was estimated by the method of Moore and Stein (1948). The content of total carbohydrate was estimated by the method of Roe (1955). Total lipid content was assayed by the method of Folch *et al.,* (1957). RNA and DNA were separately extracted in triplicate samples. 60 mg fresh weight were weighed in microfuges free of RNase and DNase and homogenized with a Teflon pestle. RNA was processed by the methods of Chomczynski and Sacchi (1987) and Chomczynski (1993). DNA was extracted by following the standard procedures (Sambrook *et al.,* 1989). One unit of optical density (OD) corresponds to approximately 40 µg ml^{-1} of RNA and 50 µg ml^{-1} of DNA was used as respective standard. The ratio between the readings at 260–280 nm provided an estimate of the purity of the nucleic acids (Chomczynski, 1993). The data were subjected to analyses by adopting student t-test through SPSS software, version 11.5. The *P* values less than 0.05 were considered statistically (95%) significant.

RESULTS AND DISCUSSION

Survival and feed utilization on growth

Table 2 depicts the data pertaining to survival and feed utilization on growth of *M. rosenbergii* PL fed with herbals incorporated diets. Survival rate (SR) ranged from 69.0-80.0 % in experimental diets fed PL groups were recorded against 66.0 % in control. These differences were significant at $P<0.05$. However, the difference recorded in SR between control and fenugreek incorporated feed fed PL was found to be statistically not significant. The SR was in the order of garlic > ginger > turmeric > fenugreek. The better SR recorded in experimental diets may be attributed with the feeding rate (FR), which was found to be higher in experimental diets fed PL ($P<0.05$), particularly with garlic and ginger (22.4% and 13.7% respectively) when

compared to control. Therefore, the difference in feeding may have a role in survival (Table 1). Furthermore, the elevation in SR might be due to the active principle (the antimicrobial and anti-stress effects) of these herbs. The elevation recorded in FR resulted in increase of weight gain (WG) in experimental PL over control (P<0.05). This was in the order of garlic > ginger > turmeric > fenugreek (32.3, 28.4, 18.1 and 10.2% respectively). The length gain (LG) attained in experimental diets fed PL groups were also significantly higher (P<0.05) over control in the similar pattern. Therefore, the specific growth rate (SGR) was also significantly higher in these herbs (10.9-3.6%), particularly garlic included diet fed PL (P<0.05). The increase in survival and growth has also been reported in *M. rosenbergii* PL fed with *O. sanctum*, *W. somnifera, A. paniculata*, *C. quadrangularis* and *E. alba* incorporated feeds and such an increase in growth was attributed with increased activity of digestive enzymes, protease, amylase and lipase (Bhavan *et al.,* 2011b; Shanthi *et al.,* 2012). In the present study, the herbal active principles of garlic, ginger, turmeric and fenugreek may have increased the activities of digestive enzymes, therefore may be acted as appetizers as this is the characteristic ability of these herbs (Chopra *et al.,* 1982; Platel and Srinivasan, 2000; Platel *et al*., 2002; Platel and Srinivasan, 2004; Venkataramalingam *et al*., 2007), which in turn enhances the feeding rate and ultimately reflected on growth increment. The growth promoting ability of several herbs on aquatic animals has been tested by many workers (Citarasu *et al*., 2002; Sivaram *et al*., 2004; Vasudeva Rao *et al*., 2006; Immanuel *et al*., 2007, 2009). It has also been reported that herbal active principles promotes utilization of cellular lipid and fatty acid, which in turn resulted in accumulation of protein, which ultimately promotes growth in *Pagrus major* (Ji *et al.,* 2007). The results observed in other parameters, such as condition factor (CF), feed conversion ratio (FCR), conversion efficiency (CE), protein conversion efficiency (PCE), metabolizable energy (ME) and gross energy (GE) are in support to/ or in agreement with above said hypothesis.

Concurrently, the values obtained for CF and FCR were in decreasing pattern (garlic < ginger < turmeric < fenugreek; CF: 26.0, 24.3, 13.8 and 4.0%; FCR: 25.0, 22.0, 14.7 and 8.8%). This indicated the fact the experimental feeds were superior quality than that of control. This was found to be statistically significant (P<0.05) except fenugreek incorporated feed. Therefore, feed incorporated with garlic was superior followed by ginger, turmeric and fenugreek. The similar trend has also been reported in *M. rosenbergii* PL fed with *O. sanctum*, *W. somnifera, A. paniculata*, *C. quadrangularis* and *E. alba* incorporated feeds (Bhavan *et al.,* 2011b; Shanthi *et al.,* 2012), in *P. indicus* fed with *O. sanctum* and *W. somnifera* supplemented diet (Sivaram *et al.,* 2004), in *Penaeus monodon* fed with *Phyllanthus niruri and Aloe vera* included feed (Ahilan *et al.,* 2010).

The food conversion efficiency (FCE) and protein conversion efficiency (PCE) were found to be higher in PL fed with experimental diets (FCE: 31.9, 28.3, 17.7, 9.8%; PCE: 22.5, 19.0, 14.5 and 6.4%) when compared with control (P<0.05). However, the PCE in fenugreek incorporated feed fed PL was not found to be statistically significant. It has been suggested that the increase in protein conversion efficiency (PCE) in prawn fed with stafac-20 could be due to the increased activity of digestive enzymes or increased rate of assimilation (Vatheeswaran and Ali, 1986; Manoj Kumar, 1994; Keshavanath *et al.,* 1999). The metabolizable energy (ME) and gross energy (GE) were significantly higher (P<0.05) in PL fed with experimental diets (ME: 30.0, 23.2, 17.4 and 10.0%; GE: 28.9, 22.5, 17.0 and 9.5%) when compared with control. This indicates the fact that garlic enhances energy level of PL followed by ginger, turmeric and fenugreek. This was attributed with accumulation of biochemical constituents, particularly total protein and amino acid, and thus, corresponding WG and growth promotion was achieved. Elevation of GE in fish, *Oreochromis niloticus* fry fed with radish root extract supplemented formulated diet has been reported (Salama *et al*., 2011).

Biochemical Constituents

Protein was the major biochemical constituent in the PL, followed by carbohydrate and lipid. The concentrations of total protein (22.4, 19.1,

6.2 and 5.3%), amino acid (25.2, 20.4, 16.8 and 14.7%), carbohydrate (38.3, 37.6, 6.8 and 0.89%) and lipid (8.6, 16.1, 20.8 and 38.7%) were found to be elevated in experimental diets fed PL when compared with control (Table 3). These elevations recorded in experimental diets fed PL were found to be statistically significant (P<0.05). The elevation of total protein, amino acid and carbohydrate was in the order of garlic > ginger > turmeric > fenugreek. In the case of total lipid the elevation recorded was just in the reverse trend (garlic < ginger < turmeric < fenugreek). Therefore, garlic has the potency to enhance the utilization of available total lipid in the diet by the PL. This was followed by ginger, turmeric and fenugreek. This suggests that these herbs have protein sparing ability on growth of *M. rosenbergii* PL. The elevation in biochemical constituents has also been reported in *M. rosenbergii* PL fed with *O. sanctum* and *W. somnifera* (Bhavan *et al*., 2011b), *A. paniculata*, *C. quadrangularis* and *E. alba* (Shanthi *et al.,* 2012).

In the present study, the contents of RNA (38.0, 30.8, 19.8 and 19.1%), DNA (31.2, 25.0, 18.7, and 18.7%) and RNA/DNA (4.9, 4.0, 0.6 and 0.4%) were found to be elevated (P<0.05) in experimental diets fed PL groups when compared with respective control. These were also in the order of garlic > ginger > turmeric > fenugreek. Growth in terms of accumulation of protein is always accompanied by high turnover rate of RNA concentration, which is a prime factor of protein synthesis (Wilder and Stanley, 1983). The elevation in DNA level (comparatively lower than RNA) recorded in experimental PL groups indicates increase in packed cell volume in unit area. The RNA/DNA ratio is useful index of growth (Bulow *et al*., 1981; Buckley 1984; Martin *et al.,* 1985). In this study, the recorded elevation in their ratio in herbal incorporated diets fed PL groups indicates there was appreciable quantum of growth. Actually, the herbal growth promoter induces transcription rate, which in turn increases RNA and total amino acid, which ultimately enhances proteins synthesis in the cell. Similar hypothesis has been proposed by Citarasu (2009) to explain the growth promoting potential of certain herbs in aquatic animals.

The present study yields a clear message that garlic incorporation under the test concentration of 1% level promotes growth in *M. rosenbergii* PL by enhancing feeding, and protein synthesis, which was evident through elevated RNA and amino acid levels, followed by ginger, turmeric and fenugreek. However, this study needs further clarification with higher concentrations of these herbs to derive more meaningful conclusion. Anyhow, these herbs can be included in aqua feed formulations for sustainable development of *Macrobrachium* culture.

Table-1

Active principles and medicinal properties of garlic, ginger, turmeric and fenugreek.

Medicinal herbs	Active principle compounds	Medicinal properties	Author
Garlic	Sulfur compounds: alin, **allicin**, ajoene, allylpropyl disulfide, diallyl trisulfide, sallylcysteine, vinyldithines and S-allylmercaptocystein.	Used against bronchitis, respiratory, gastrointestinal and flatulence problem. Also used against high blood pressure, diabetes, high blood cholesterol and cancer. It has antibacterial, antifungal and antioxidant properties. Used externally for warts, corns, arthritis, muscle pain, neuralgia, sciatica, leprosy and menstrual cramps.	Shackebaei *et al*., 2010
Ginger	**Zingerone,** Zingiberene, Sesquiterpenoids (β-sesquiphellandrene, bisabolene and farnesene), Monoterpenoid fraction (β -phelladrene, cineol, and citral), and Non-volatile phenylpropanoid-derivatives (gingerols and shogaols).	Used against dyspepsia, gastroparesis, high cholesterol, high blood pressure, heart disease, hypertension, obesity, hypoglycema, diabetes, tumor, cancer and osteoarthritis. It has antioxidant, antibacterial, anti-inflammatory, antithrombotic and antimicrobial properties.	Rehman *et al*., 2001
Turmeric	**Curcumin**, Arturmerone, Methylcurcumin, Demethoxy –curcumin, Bisdemethoxy-curcumin, and Sodium curcuminate	Used against Alzheimer's disease, arthritis, diabetes, ulcer, cancer, hypertension, and high blood cholesterol. It has anti-inflammatory, anti-mutagenic, antibacterial, antifungal, anti-protozoal, antiviral, anti-fibrotic and anti-venom properties. It has antifertility effect also. It has antioxidant and anticoagulant properties.	Chattopadhyay *et al*., 2004
Fenugreek	**Mucilage,** Coumarin, Alkaloids (choline and trigonelline), and Steroidal aponins (diosgenin).	Used against gastric inflammation, diabetes, digestive disorders, ulcer, cancer, tuberculosis and hyper tension. It is also used in treatment of hypercholesterolemia. It has antiviral, antimicrobial and antioxidant properties.	Kumar *et al*., 2005; Bukhari *et al*., 2008

Compounds in bold letters are major bioactive substances.

Table-2

Survival and feed utilization on growth of M. rosenbergii PL fed with herbs incorporated diets.

Parameters	Control Diet (100% BD)	Experimental Diets (99% BD + 1% Herbal)			
		Garlic	Ginger	Turmeric	Fenugreek
Survival (%)	66.00 ± 6.00 % ↑ t-value/ P<	82.00 ± 7.00 (24.24↑) -27.71/0.001	80.00 ± 4.00 (22.24↑) -12.12/0.007	70.00 ± 5.00 (6.06↑) -6.92/0.020	69.00 ± 3.0^{NS} (4.54↑) -1.73/0.225
FR (g d^{-1})	0.58 ± 0.02 % ↑ t-value/ P<	0.71 ± 0.03 (22.41↑) -22.51/0.002	0.66 ± 0.04 (13.79↑) -6.92/0.020	0.63 ± 0.02 (8.62↑) (*)	0.61 ± 0.02 (5.17↑) (*)
WG (g)	1.76 ± 0.14	2.33 ± 0.21 (32.38↑) -14.10/0.005	2.26 ± 0.17 (28.40↑) -28.86/0.001	2.08 ± 0.15 (18.18↑) -55.42/0.000	1.94 ± 0.17 (10.22↑) -10.39/0.009
LG (cm)	3.40 ± 0.30 % ↑ t-value/ P<	4.43 ± 0.19 (30.29↑) -16.28/0.004	4.39 ± 0.35 (29.11↑) -34.29/0.001	3.96 ± 0.23 (16.47↑) -13.85/0.005	3.65 ± 0.26 (7.35↑) -10.82/0.008
SGR (%)	1.10 ± 0.02 % ↑ t-value/ P<	1.22 ± 0.03 (10.90↑) -20.78/0.002	1.21 ± 0.02 (10.0↑) (*)	1.17 ± 0.02 (6.36↑) (*)	1.14 ± 0.02 (3.63↑) (*)
CF (%)	1.23 ± 0.14 % ↓ t-value/ P<	0.91 ± 0.16 (26.01↓) 27.71/0.001	0.93 ± 0.12 (24.39↓) 25.98/0.001	1.06 ± 0.10 (13.82↓) 7.36/0.018	1.18 ± 0.21^{NS} (4.06↓) 1.23/0.342
FCR (%)	0.68 ± 0.05 % ↓ t-value/ P<	0.51 ± 0.04 (25.0↓) 29.44/0.001	0.53 ± 0.03 (22.05↓) 12.99/0.006	0.58 ± 0.04 (14.70↓) 17.32/0.003	0.62 ± 0.05 (8.82↓) (*)
FCE (%)	146.88 ± 11.91 % ↑ t-value/ P<	193.83 ± 17.63 (31.96↑) -14.217/0.005	188.55 ± 14.17 (28.37↑) -31.93/0.001	173.00 ± 12.58 (17.78↑) -67.52/0.000	161.33 ± 14.45 (9.83↑) -9.85/0.010
PCE (%)	19.96 ± 0.70 % ↑ t-value/ P<	24.47 ± 1.48 (22.59↑) -10.01/0.010	23.77 ± 1.00 (19.08↑) -21.99/0.002	22.87 ± 0.91 (14.57↑) -24.00/0.002	21.24 ± 1.41^{NS} (6.41↑) -3.12/0.089
ME (KJ g^{-1})	22.88 ± 0.38 % ↑ t-value/ P<	29.76 ± 0.62 (30.06↑) -121.82/0.000	28.21 ± 0.74 (23.29↑) -73.75/0.000	26.88 ± 0.52 (17.48↑) -173.20/0.000	25.18 ± 0.40 (10.05↑) -1065.21/0.000
GE (KJ g^{-1})	29.86 ± 0.42 % ↑ t-value/ P<	38.50 ± 0.89 (28.93↑) -31.84/0.001	36.58 ± 0.95 (22.50↑) -21.96/0.002	34.94 ± 0.61 (17.01↑) -46.31/0.000	32.71 ± 0.77 (9.54↑) -14.10/0.005

BD, Basal diet.
FR, Feeding rate; WG, Weight gain; LG, Length gain; SGR, Specific growth rate; CF, Condition factor; FCR, Food conversion ratio; FCE, Conversion efficiency; PCE, Protein conversion efficiency; ME, Metabolizable energy; GE, Gross energy.
Values are expressed as mean ± SD (n=3).
Values in parentheses are % ↑ (or) ↓
^{NS}, not significant statistically.
Significance (P<0.05) of paired samples t-test are also given (* the correlation and t cannot be computed because the SE of the difference is '0').

Table 3

Concentrations of biochemical constituents in M. rosenbergii PL fed with herbs incorporated diets.

Biochemical Constituents	Control Diet (100% BD)	Experimental Diets (99% BD + 1% Herbal)			
		Garlic	Ginger	Turmeric	Fenugreek
Protein (% wet wt.)	7.48 ± 0.26 (% ↑) t-value/ P<	9.16 ± 0.15 (22.4↑) -26.45/0.001	8.91 ± 0.36 (19.1↑) -24.76/0.002	7.95± 0.53^{NS} (6.2↑) -3.01/0.095	7.88 ± 0.81^{NS} (5.3↑) -1.26/0.335
Amino Acids (% wet wt.)	4.40 ± 0.53 (% ↑) t-value/ P<	5.51± 0.13 (25.2↑) -4.80/0.041	5.30 ± 0.35 (20.4↑) -8.66/0.013	5.14 ± 0.64 (16.8↑) -11.65/0.007	5.05 ± 0.42 (14.7↑) -10.23/0.009
Carbohydrate (% wet wt.)	3.35 ± 0.12 (% ↑) t-value/ P<	4.65 ± 0.37 (38.3↑) -9.00/0.012	4.61 ± 0.62 (37.6↑) -4.36/0.049	3.58 ± 0.44^{NS} (6.8↑) -1.24/0.339	3.38 ± 0.93^{NS} (0.8↑) -0.64/0.955
Lipid (% wet wt.)	1.73 ± 0.23 (% ↑) t-value/ P<	1.88 ± 0.26 (8.6↑) -8.66/0.013	2.01 ±0.20 (16.1↑) -16.16/0.004	2.09± 0.22 (20.8↑) -62.35/0.000	2.40 ± 0.21 (38.7↑) -58.02/0.000
RNA (mg/g wet. wt.)	2.92 ± 0.28 (% ↑) t-value/ P<	4.03 ± 0.40 (38.0↑) -16.02/0.004	3.82 ± 0.59 (30.8↑) -5.02/0.03	3.50 ± 0.45 (19.8↑) -1.57/0.02	3.48 ± 0.24 (19.1↑) -25.00/0.002
DNA (mg/g wet. wt.)	0.16 ± 0.01 (% ↑) t-value/ P<	0.21 ± 0.02 (31.2↑) -8.66/0.01	0.20 ± 0.02 (25.0↑) -6.92/0.02	0.19 ± 0.02 (18.7↑) -5.19/0.03	0.19 ± 0.02 (18.7↑) -5.19/0.03
RNA/DNA	18.28 ± 0.70 (% ↑) t-value/ P<	19.18 ± 0.07 (4.9↑) -18.19/0.003	19.02 ± 1.05^{NS} (4.0↑) -3.14/0.08	18.39 ± 0.43^{NS} (0.6↑) -1.56/0.25	18.36 ± 0.67^{NS} (0.4↑) -3.76/0.06

BD, Basal diet.
Values are expressed as mean ± SD (n=3).
Values in parentheses are % ↑.
^{NS}, not significant statistically.
Significance (P<0.05) of paired samples t-test are also given.

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