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GROWTH PERFORMANCE OF THE FRESHWATER PRAWN *MACROBRACHIUM ROSENBERGII* POST LARVAE FED WITH *OCIMUM SANCTUM* (TULSI) AND *WITHANIA SOMNIFERA* (ASHWAGANDHA) INCORPORATED FEEDS

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ABSTRACT

The feasibility of uses of *Ocimum sanctum* and *Withania somnifera* as herbal nutrients to promote the growth and survival of the freshwater prawn, *Macrobrachium rosenbergii* post larvae (PL) was evaluated in a sustainable manner. Feeds were formulated with fishmeal, soya bean meal, groundnut oilcake, coconut oilcake, and green gram as basal ingredients. Tapioca flour and egg albumin were used as binding agents. Cod liver oil was added as lipid sources. Vitamin B-complex and a pinch of salt were also mixed. Diets with incorporation of *O. sanctum* and *W. somnifera* separately were served as experimental feeds, and diet without herbal incorporation was served as control. These feeds were fed to the *M. rosenbergii* PL in a triplicate feeding trial conducted for a period of 45 days (PL15-65) under laboratory condition. The growth performance in terms of nutritional indices (weight gain, specific growth rate, condition factor and survival rate), energy utilization (feeding, absorption, conversion and metabolism) and concentrations of biochemical constituents

(total protein, amino acid, carbohydrate and lipid, and profiles of individual amino acids) were found to be significantly ($P<0.05$) changed in PL fed with herbal incorporated feeds when compared to that of control. Among the two herbs used, the PL fed with *W. somnifera* incorporated feed has showed better growth performance followed by *O. sanctum*. Therefore, *W. somnifera* incorporated feed proved to be more beneficial than *O. sanctum* for the overall growth and maintenance of *M. rosenbergii* PL.

KEY WORDS: *Macrobrachium rosenbergii*, *Ocimum sanctum, Withania somnifera*, Growth, Biochemical constituents

1. INTRODUCTION

In India, aquaculture of freshwater prawns, particularly *Macrobrachium rosenbergii*, *Macrobrachium malcolmsonii* and *Macrobrachium gangeticum* are in practice. These prawns have good demand in both domestic and export markets, and are fetched for high price (Radheyshyam, 2009). However, *M. rosenbergii* has become the main candidate species for aqua farming due to its fast growing capacity, large size, better meat quality and omnivorous feeding habit. Diet usually plays an important role in all stages of rearing. It affects survival and growth of prawns. To attain good growth, first the food should be perceived, captured, accepted and efficiently ingested. A palatable and nutritious diet would always preferred by prawns. Feed formulations to *Macrobrachium* with locally available low cost commodities have been reported (Bhavan *et al*., 2010a, b; Bhavan *et al*., 2011; Rebecca and Bhavan, 2011). Incorporation of herbal products have been reported to promote various activities, like anti-stress, appetite stimulation, tonic, aphrodisiac and anti-microbial properties in shrimp and prawns due to their active principles, such as antioxidants, alkaloids, flavonoids, pigments, phenolics, terpenoids, steroids and essential oils (Prasad and Padhyoy, 1993; Nadkarni, 1995; Citarasu *et al*., 2001, 2002; Sivaram *et al*., 2004). Herbal active principle in the diet induces the secretion of the digestive

enzymes to stimulate the appetite and increases food consumption efficiencies. Several herbal principles have been reported for their growth-promoting activity in aquatic animals (Jayaprakas and Eupharsia 1996; Citarasu *et al*., 2002). Medicinal plants are highly promised in the aquaculture industry to increase feed consumption and improve digestion, which in turn leads to better survival, growth and production (Venkataramalingam *et al*., 2007). Herbals are the store houses and sources of safer and cheaper chemicals. Applications of the traditional medicines in aquaculture to overcome the drawbacks in the usage of chemical therapeutics is relatively new venture and the potential of the herbals with multifunctional active principles are promising (Jayaprakas and Euphrasia, 1996; Sambhu and Jayaprakas, 2001; Citarasu *et al*., 2002; Jian and Wu, 2003; Immanuel *et al*., 2003; Sivaram *et al*., 2004; Babu *et al*., 2008).

In the view of the above, two herbs, *Ocimum sanctum* (Tulsi or 'Holy Basil') and *Withania somnifera* (Ashwagandha or winter cherry) have been selected and incorporated with formulated feeds. *O. sanctum* is a small annual herb rich in essential oil 'Eugenol'. In addition, the essential oil from the leaves of *O. sanctum* has exhibited anti-bacterial and anti-fungal activity and it has rich anti-oxidant content (Grover and Rao, 1977). Recent studies suggest that tulsi may be a COX-2 inhibitor, like many modern painkillers, due to its high concentration of eugenol, 1-hydroxy-2-methoxy-4-allylbenzene (Singh, 1999). Tulsi has beneficial effect on blood glucose levels due to its antioxidant properties (Sethi *et al*., 2004). The fixed oil has demonstrated anti-hyperlipidemic and cardioprotective effects in rats fed a high fat diet (Suanarunsawat, 2010). Some of the main chemical constituents of tulsi are: Oleanolic acid, Ursolic acid, Rosmarinic acid, Eugenol, Carvacrol, Linalool, β-caryophyllene (Kuhn, 2007), β-elemene (c.11.0%), β-caryophyllene (Circa 8%) and germacrene D (c.2%) (Padalia and Verma, 2011).

The root of *W. somnifera*, the 'Indian ginseng' is also known as winter cherry has been an important herb in the ayurvedic, siddha and unani medical

systems for over 3000 years in the treatment of various physiological disorders. The plant was traditionally used to promote youthful vigor, endurance, strength, and health by increasing the production of vital fluids, muscle fat, blood, lymph, semen and cells (Singh and Kumar, 1998). The main active constituents in *W. somnifera* are alkaloids and steroidal lactones. These include tropine and cuscohygrine. The leaves contain the steroidal lactones, withanolides, notably withaferin-A. Withanolides have been found to have regenerative properties on brain-cell synapses in mice and human cell line. Withaferin A inhibits notch-1 signaling and down regulates pro survival pathways, such as Akt/NF-κB/Bcl-2, in three colon cancer cell lines, HCT-116, SW-480, and SW-620 (Koduru *et al*., 2010). Recent research in mice suggests that withaferin A may have anti metastatic activity (Thaiparambil *et al*., 2011).

In order to promote the growth and survival of *M. rosenbergii* PL in a sustainable manner, the present study was conducted with an objective to identify whether *O. sanctum* and *W. somnifera* are suitable for incorporation in artificial feed formulation. This was revealed through analyses of nutritional indices, energy utilization, concentrations of biochemical constituents and profiles of individual amino acids in *M. rosenbergii* PL fed with these herbs incorporated feeds.

2. MATERIALS AND METHODS

The Post Larvae of *M. rosenbergii* (PL-5) were purchased from Happy Bay Aqua Nova Hatchery at Kancheepuram District. They were safely brought to the laboratory in well-oxygenated polythene bags. They were stocked in a large cement tank (1000 L capacity) and acclimatized for 10 days in ground water (pH, 7; total dissolved solids, 0.9 g L^{-1}; dissolved oxygen, 7.2 mg L^{-1}; BOD, 30.0 mg L^{-1}; COD, 125.0 mg L^{--1}; ammonia, 0.028 mg L^{-1} (APHA, 2005), during which they were fed with boiled egg albumin, *Artemia* nauplii and commercially available scampi feed alternatively thrice a day, and the medium

was adequately aerated. On daily basis three fourth of the water was renewed by siphoning method causing minimum disturbance to the prawns. The unfed feed if any, the excreta and exuviae were all removed.

2.1 Feed formulation and proximate composition of biochemical constituents

In this study, all the ingredients used for feed formulation were purchased from local merchants and were of good quality. Table 1 depicts proportions of ingredients used to formulate artificial feeds. The basal ingredients used were fishmeal, groundnut oilcake, coconut oilcake, soya bean meal, and green gram. A mixture of these basal ingredients in the mentioned proportions for each feed was steam cooked for 15 minutes. Egg albumen and tapioca flour were used as binding agents. 2% (w/v) of Cod liver oil was added as lipid source. Vitamin B-complex (1%) and a pinch of salt were also added. To this, at room temperature, *O. sanctum* (2.5g) and *W. somnifera* (2.5g) were separately incorporated and made into thick dough which was immediately squeezed out manually into pellets using a hand pelletizer of 3 mm pore-diameter. Pellets were sun dried for about three days and were stored in air tight containers to attain maximum shelf-life. The feed formulated without any herbal incorporation was served as control. The proximate composition of biochemical constituents of the formulated feeds were also given in table 1. The concentration of total protein was determined following the method of Lowry *et al.*, (1951). 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). Actually, concentrations of total protein, amino acid, carbohydrates and lipid were found to be similar in proportion in both control and experimental feeds. However, the content of total carbohydrate was found to be slightly higher in *O. sanctum* incorporated feed (table 1).

Table 1 : Proportion of basal ingredients and proximate composition of biochemical constituents in formulated feeds

Parameters		Control	Experimental	
		Feed-1 (BI only)	Feed-2 (BI+OS)	Feed-3 (BI+WS)
Ingredients (%)	Fish meal	20.0	20.5	20.5
	Ground nut oil cake	25.0	25.7	25.7
	Coconut oil cake	10.0	10.3	10.3
	Soya bean meal	25.0	25.7	25.7
	Green gram	10.0	10.3	10.3
	Tapioca	3.0	3.0	3.0
	Egg albumin	4.0	4.0	4.0
	Cod liver oil	2.0	2.0	2.0
	*Vitamin mix	1.0	1.0	1.0
	Leaf powder of *O. sanctum*	-	2.5	-
	Root powder of *W. somnifera*	-	-	2.5
Biochemical composition (%)	Protein	42.0	42.0	42.0
	Amino acids	30.0	30.0	30.0
	Carbohydrates	20.0	21.0	20.0
	Lipids	14.0	14.0	14.0

BI, Basal ingredients; OS, *O. sanctum*; WS, *W. somnifera*.

2.2 Nutritional indices and concentrations of biochemical constituents

Three groups of 30 individuals (PL-15; length: 1.59 ±0.02 cm; body mass: 0.053±0.006 g) each in triplicate were taken. Each group was housed in an aquarium of 25 L capacity. From each aquarium 10 PL (i.e. 30 PL from each group) were randomly taken and introduced in a separate aquarium. From this 10 PL were taken for measurement of initial morphometric data. Therefore,

30 individuals were separately measured (3x10=30). The resulted mean values (each taken from 10 PL) were pooled to calculate an average mean. After measurement these PL were re-introduced into the respective separate aquaria. These PL (30 PL from each group) were immediately taken for analyses of concentrations of initial basic biochemical constituents, such as total protein (Lowry *et al.*, 1951), total amino acid (Moore and Stein, 1948), total carbohydrate (Roe, 1955) and total lipid (Folch *et al.*, 1957), and analysis of profiles of initial amino acids through HPTLC method (Hess and Sherma, 2004). For initial basic biochemical analyses, tissues from 20 PL were pooled in triplicate and the remaining 10 PL in triplicate were used for analysis of initial amino acid profile. Thus, each initial parameter was measured in triplicate.

The remaining 20 PL in each aquarium (20x3=60 PL in each group) were subjected to feeding trials for a period of 45 days (up to PL65). One group served as control and the other two groups were fed with formulated feeds, one with *O. sanctum* (2.5g) incorporated feed and another with *W. somnifera* (2.5g) incorporated feed. On 45^{th} day, the final morphometry, analyses of final biochemical constituents and final profiles of amino acids were done followed by above said standard methods. 10 PL from each aquarium were measured for final morphometric data and three such measurements were made for each group and the resulted mean values were taken for calculating the parameters of nutritional indices, such as survival rate, percentage weight gain, specific growth rate and condition factor (Tekinay and Davis, 2001). Among the available 20 PL in triplicate per group, 15 PL were utilized for estimation of final levels of biochemical constituents, and the remaining 5 PL were used for the analysis of final amino acid profile. Actually, a separate aquarium for each group was maintained exactly under respective experimental condition. These PL were utilized to replace the mortality of PL occurred in respective original aquarium in order to maintain the required number of individuals in each original experimental group.

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

$$\text{Weight Gain (WG)} = \text{Final weight (g)} - \text{Initial weight (g)}$$

$$\text{Biomass Index (BI)} = \frac{\text{Final weight (g)} - \text{Initial weight (g)}}{\text{Initial weight (g)}} \times 100$$

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

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

2.3 Profiles of amino acids

Prawns were subjected to feeding trial in an experimental setup essentially similar to that described previously. 45 individuals per group (15 individuals per aquarium) were taken for analyses of amino acid profiles. The profile of amino acids was done following high performance thin layer chromatographic (HPTLC) method of Hess and Sherma (2004). The prawns were dried (80^{o}C for 3 hrs), digested with 6 M aqueous hydrochloric acid and dried under vacuum. The powdered sample was dissolved in distilled water and 5 µl of sample was loaded on 8 mm thick pre-coated Silica gel 60F254 TLC plate (20 cm x 15 cm) and processed in CAMAG- LINOMAT 5 instrument. The plate was developed in butane-Ammonia-Pyridine-Water (3.9:1:3.4:2.6) mobile phase. The plate was sprayed with ninhydrin reagent prepared in propan-2-ol and dried. The developed plate was documented using photo- documentation chamber (CAMAG-REPROSTAR 3) at UV 254 nm and UV366 nm lights. Finally, the plate was scanned at 500 nm using CAMAG-TLC SCANNER 3. The peak area of the sample was compared with standard amino acids and quantified. Four

groups of standard amino acids were also run in parallel. Group-I: Proline, Serine, Asparagine, Glutamine and Metheonine; Group-II: Aspartic acid, Glutamic acid, Alanine, Valine and Phenyl alanine; Group-III: Lycine, Glycine, Threonine, Isoleucine and Tyrosine; Group-IV: Arginine, Cystine, Histidine, Leucine and Tryptophan.

2.4 Energy utilization

Three groups of 15 individuals (PL-15; length: 1.59 ±0.02 cm; body mass: 0.053±0.006 g) each in triplicate were taken, housed in an aquarium with 12.5 L ground water and were subjected to feeding trial exactly as described previously. The parameters of energy utilization, such as feeding rate, mean absorption, mean conversion and metabolic rate were calculated. The energy content of whole prawn, feeds, faeces and exuvia was measured using Parr 1281 Oxygen Bomb Calorimeter. The energy budget was calculated using the equation (C = (P+E) + R + F +U) derived by Petrusewicz & Macfadyen (1970); where, C is the energy consumed in food; P, is the conversion or growth; R, the material lost as heat due to metabolism; F, the energy lost through faeces; U, the energy lost in ammonia excretion and; E, the energy lost through exuvia. The daily excretion of ammonia by the prawn was estimated after feeding as per the phenol hypochloride method of Solorzano (1969). The energy loss occurring by ammonia excretion was calculated using the ammonia calorific quotient, 1 mg NH_3: 5.9 cal. (Elliot, 1976). The food energy consumed was measured as the difference between the energy content of food offered and that of the uneaten food. The quantity of absorbed food energy was calculated by subtracting F from C. Conversion or growth is the sum of energy channelled to somatic growth (P) and exuvia (E). Following the estimations of C, F, U, and P, the metabolism (R=Respiration) was calculated by dividing the respective amount of energy by initial live weight of the prawn per unit time in days.

$$\text{Feeding Rate} = \frac{\text{Mean Food Consumption (k.cal/day)}}{\text{Initial live weight of the prawn (g)}}$$

Mean Absorption = Mean Food Consumption (k.cal/day) – Mean Food Excreted as Faeces (k.cal/day)

$$\text{Absorption Rate} = \frac{\text{Mean Absorption (k.cal/day)}}{\text{Initial live weight of the prawn (g)}}$$

Mean Conversion = Mean weight gain (k.cal/day) + Mean exuvial weight (k.cal/day)

$$\text{Conversion rate, P} = \frac{\text{Mean Conversion (k.cal/day)}}{\text{Initial live weight of the prawn (g)}}$$

$$\text{NH3 Excretion rate, U} = \frac{\text{Mean NH3 Excretion (k.cal/day)}}{\text{Initial live weight of the prawn (g)}}$$

Metabolic Rate, R = Absorption rate (k.cal./g/day – Conversion rate (k.cal/g/day) + NH3 excretion rate (k.cal/g/day)

The data obtained in this study were subjected to statistical analysis by adopting Student `t` test (Zar, 1984).

3. RESULTS

3.1 Nutritional indices

The initial average length and weight of *M. rosenbergii* PL are presented in table 2. Similarly, the final morphometric data obtained after 45 days of feeding trial are given in Table 2. The comparison of initial and final morphometric data showed statistically significant growth differences ($P<0.01$). Similarly, the

differences observed between control and experimental groups were also found to be statistically significant (P<0.05). Among the two herbs, *W. somnifera* incorporated feed was produced higher growth when compared with *O. sanctum* (Table 2). Therefore, the weight gain, biomass index and specific growth rate were found to be in the following order: *W. somnifera* > *O. sanctum* > control. The condition factor observed was just the reverse in order. It indicates *W. somnifera* incorporated feed was the supreme over *O. sanctum* and the feed without herbal incorporation (Table 1). Moreover, the survival rate observed was the best in *W. somnifera* incorporated feed followed by *O. sanctum* and control (Table 2).

Table 2: Morphometric data, nutritional indices and energy utilization of *M. rosenbergii* PL fed with the formulated feeds

Parameters		Control	Experimental	
		Feed-1 (BI only)	**Feed-2 (BI+OS)**	**Feed-3 (BI+WS)**
Morphometric data	Initial length (cm)	1.59 ±0.02	1.59 ±0.02	1.59 ±0.02
	Final length (cm)	3.13 ±0.05	3.64 ±0.08	4.36 ±0.12
	Initial weight (g)	0.053 ±0.006	0.053 ±0.006	0.053 ±0.006
	Final weight (g)	0.262 ±0.010	0.373 ±0.015	0.465 ±0.016
	Weight gain (WG)	0.216 ±0.010	0.322 ±0.015	0.412 ±0.019
Nutritional indices	Biomass index (BI)	413.7 ±19.6	625.3 ±31.1	809.7 ±34.3
	Specific growth rate (SGR) %	1.18 ±0.03	1.43 ±0.02	1.59 ±0.02
	Condition factor (CF)	0.853 ±0.008	0.773 ±0.020	0.569±0.017
	Survival rate (SR %)	84 ±2	89 ±3	94 ±3
Energy utilization	Feeding Rate (FR)	0.85 ± 0.05	0.95 ± 0.06	1.15 ± 0.05
	Absorption Rate	0.70 ± 0.06	0. 80 ± 0.05	1.0 ± 0.05

(k.cal./g/day)	(AR)			
	Conversion Rate (CR)	0.38 ± 0.05	0.44 ± 0.06^{NS}	0.48 ± 0.06
	Ammonia excretion rate (U)	0.11 ± 0.003	0.13 ± 0.005	0.14± 0.004
	Metabolic rate (MR)	0.46 ± 0.05	0. 57 ± 0.06	0.65 ± 0.06

Each value is mean ± SD of triplicate observations.

BI, Basal ingredients; OS, *O. sanctum*; WS, *W. somnifera*.

^{NS}, not significant; *All other values are significant at P<0.05 when compared with control.

3.2 Energy utilization

Generally, the parameters of energy utilization, such as the rate of feeding, absorption, conversion, ammonia excretion and metabolism were found to be higher in experimental groups when compared with control group (Table 2). Among the two experimental groups, the elevation in these parameters, except ammonia excretion were found to be statistically significantly (P<0.05), particularly in PL fed with *W. somnifera* incorporated feed when compared with control. In the case of *O. sanctum* incorporated feed fed PL, statistically significant elevation was recorded in absorption and metabolism alone (Table 2).

3.3 Biochemical constituents

The contents of total protein, amino acid, carbohydrate and lipid between initial and final day PL were found to be statistically significant (P<0.01). Similarly, concentrations of these biochemical constituents were found to be significantly higher (P<0.05) in PL fed with experimental feeds when compared with control feed (Table 3). Among the two herbs used, *W. somnifera* incorporated feed fed PL showed significantly higher levels of biochemical

constituents (P<0.05) than that of *O. sanctum* incorporated feed fed PL (Table 3).

Table 3 : Proximate composition of biochemical constituents and profiles of amino acids in *M. rosenbergii* PL fed with formulated feeds

Parameters		Initial	Final		
			Control	Experimental	
			Feed-1 (BI only)	Feed-2 (BI+OS)	Feed-3 (BI+WS)
Biochemical constituents (mg/g wet wt.)	Total Protein	15.00 ±0.50	22.60 ±2.00	26.60 ±1.27	31.50 ±1.50
	Total Amino acids	10.60 ±0.79	17.56 ±1.28	21.93 ±2.00	26.40 ±1.56
	Total Carbohydrates	10.16 ±0.65	20.56 ±1.28	24.50 ±1.50	23.63 ±1.26
	Total Lipids	5.43 ±0.51	10.43 ±1.00	15.33 ±0.76	16.40 ±1.50
Profiles of amino acids (% dry wt.)	Lycine*	1.8 ± 0.16	2.0 ± 0.15	2.1 ± 0.20^{NS}	2.1 ± 0.20^{NS}
	Arginine*	1.2 ± 0.15	1.4 ± 0.13	1.6 ± 0.14	1.7 ± 0.10
	Aspartic acid	1.6 ± 0.18	1.9 ± 0.15	2.3 ± 0.23	2.5 ± 0.30
	Proline + glutamic acid	2.3 ± 0.22	2.4 ± 0.24^{NS}	2.7 ± 0.26	2.7 ± 0.25
	Glycine	0.6 ± 0.06	0.7 ± 0.05^{NS}	1.0 ± 0.09	1.0 ± 0.10
	Serine + Alanine + Cystine	2.2 ± 0.23	2.4 ± 0.24	2.8 ± 0.24	2.7 ± 0.22
	Asparagine + Threonine* + Histidine*	0.9 ± 0.12	1.2 ± 0.12	1.4 ± 0.13	1.3 ± 0.12^{NS}
	Glutamine + Valine*	2.7 ± 0.24	2.9 ± 0.25	3.3 ± 0.24	3.4 ± 0.30
	Metheonine* + Isoleucine* + Tyrosine* + Leucine*	2.9 ± 0.26	3.2 ± 0.25	3.5 ± 0.26	3.5 ± 0.30
	Phenyl alanine* + Tryptophan*	1.0 ± 0.12	1.2 ± 0.12	1.6 ± 0.13	1.6 ± 0.14

Each value is mean ± SD of triplicate observations.

BI, Basal ingredients; OS, *O. sanctum*; WS, *W. somnifera*.

Biochemical values are significant between initial and final (P<0.01), and between control and experimental groups (P<0.05)

*Essential amino acids.

^{NS}, not significant; Individual amino acid values between initial and final, and between control and experimental groups are significant at P<0.1.

3.4 Profiles of amino acids

On final day, concentrations of individual amino acids were found to be significantly higher in PL fed with control feed when compared with initial levels of these amino acids (P<0.1). Similarly, levels of these amino acids were found to be significantly higher in experimental groups when compared with control (P<0.1). Among the experimental groups, the values of arginine; aspartic acid; glutamine + valine were found to be significantly higher (P<0.1) in *W. somnifera* incorporated feed fed PL when compared with *O. sanctum* (table 3). Whereas, the combined value of serine + alanine + cystine; asparagine + threonine + histidine were found to be significantly higher (P<0.1) in *O. sanctum* incorporated feed fed PL when compared with *W. somnifera* (table 3). However, the values for lycine; proline + glutamic acid; glycine; metheonine + isoleucine + tyrosine + leucine; phenyl alanine + tryptophan were same in two different experimental feeds fed PL (Table 3).

4. CONCLUSIONS

In the present study, *M. rosenbergii* PL responded well to the formulated feeds. The overall response was the best in the feed incorporated with *W. somnifera*. This is followed by the feed incorporated with *O. sanctum* and the feed without any herbs. Efficiency and suitability of a feed depends on number of factors, such as its composition, palatability, conversion efficiency and water stability. Generally, the ingredients present in the formulated feeds significantly influenced the performance of the animal feeds upon it. A well-balanced amino acid profile is recommended in the diet for the good growth of the prawn (Millamena *et al*., 1997). In the present study, as far as dietary nutritional quantity is concerned all three formulated feeds possessed almost similar proportion (Table 1). However, the incorporated herbal nutrients have slightly altered the quality of these feeds, which reflected on the growth and biochemical constituents of *M. rosenbergii* PL (tables 2 and 3). Small differences of certain amino acid levels in the whole body among the dietary treatments (table35) may

be due to the differences in free amino acids at the tissue levels (Kaushik and Luquet, 1980). However, in the present study, *W. somnifera* incorporated feed was much influential than that of *O. sanctum*, and produced marked elevation in concentrations of total protein, amino acid, carbohydrate and lipid, and thereby produced better growth in *M. rosenbergii* PL (tables 2 and 3).

Growth in terms of biomass is a better measurement of the nutritional value of diets (Kuban *et al*., 1985). In this study, the experimental feeds prepared with incorporation of herbals acts as appetizers, thereby increased feed intake (increased intake of proteins and amino acids) was resulted (table 2). This may be due to increased secretion of digestive enzymes, which facilitated the breakdown of food materials and availability of nutrients for absorption, which in turn ultimately promotes the growth, particularly in PL fed with *W. somnifera* incorporated feed (table 2). Similar observations has been reported by Das *et al*., (1987) in mullet, *Liza parasia*, Citarasu *et al*., (2002) in the shrimp, *P. monodon* fed with *W. somnifera* and *E. tauvina*, and by Citarasu (1998) in *P. indicus* PL fed with stresstol. The feed responsible for good growth rate also experiences good survival rate (Jones *et al*., 1997a). Higher rates of survival and growth recoded in the experimental groups over control indicate the fact that the quality of PL was improved through herbal incorporated feeds (tables 2 and 3). It has also been reported that Ashwagandha enriched *artemia* has improve the larval quality of *P. monodon* (Babu *et al*., 2008).

In conclusion, both *O. sanctum* and *W. somnifera* incorporation have proved to induce optimistic changes in terms of concentrations of biochemical constituents and growth with verified survival capability. Therefore, these herbs can be utilized for sustainable production of good quality seeds of *M. rosenbergii*. However, performance wise, *W. somnifera* has produced better result than that of *O. sanctum* under the established study concentration of 2.5% in the laboratory. Further studies are needed with various concentrations of these herbs on various parameters to clarify this point.

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