CHAPTER-5

ANALYSIS OF ENERGY UTILIZATION

5.1. INTRODUCTION

Energy is not a nutrient, but released during metabolic oxidation of carbohydrates, lipids and amino acids (Sales, 2009). Energy is derived by animals through the catabolism of dietary carbohydrates, lipids and proteins within the body and is therefore essential for the maintenance of life processes such as cellular metabolism, growth, reproduction, and physical activity. The nutritional value of a feed ingredient is based not solely on its chemical composition, but also on the amount of nutrients and energy that the animal can absorb and use from it. Metabolisable energy is a more exact measure of the energy that becomes available for metabolism by the animal (National Research Council, 1993).

In crustaceans, ingested food energy is primarily channel into growth, metabolic maintenance, ammonia excretion, faeces and moulting (Bhavan et al., 2010). The major food nutrients (ie. carbohydrates, proteins and lipids) are required by animals not only as essential materials for the construction of living tissues, but also as sources of stored chemical energy to fuel these processes as work. The ability of a food to supply energy is therefore of great importance in determining its nutritional value to animals. The chemical energy of food can be measured in heat units by means of a bomb-calorimeter.

Energy metabolism in fish and shrimp is different from that of terrestrial farm animals in two important respects. Firstly, in contrast to warm-blooded animals, fish and shrimp are aquatic
ectotherms and so do not have to expend energy in maintaining a body temperature well above ambient at 37°C. Fish and shrimp therefore have much lower maintenance energy requirements than terrestrial farm animals (Cho and Kaushik, 1985). Secondly, fish and shrimp are able to obtain 10–20% more energy from the catabolism of proteins than terrestrial farm animals, as they do not have to convert ammonia (the end product of protein catabolism) into less toxic substances (ie. urea or uric acid) prior to excretion (Brett and Groves, 1979). The excretion of waste nitrogen therefore requires less energy in these aquatic animals. The energy needs for maintenance and voluntary swimming activity must be satisfied before energy can be made available for growth. Clearly, since fish like other animals, eat primarily to satisfy their energy requirements (Cho and Kaushik, 1985), it is therefore essential that they receive either unrestricted access to food, or a palatable ration of sufficient energy density to meet all their energy requirements. Providing the optimum energy level in diets for shrimp or fish is important because an excess or deficiency of useful energy can result in reduced growth rates (NRC, 1983).

A balanced energy budget is a tool for bioenergetics modeling in aquaculture and fisheries management (Jobling, 1993). Bioenergetics theory deals with energy transformations within and among organism, and has played a prominent role in ecology (Golley, 1993; Benke et al., 1988). A major focus of bioenergy studies has been the flow of energy among species via consumption along food chains. The partition of ingested energy into growth, metabolism, excretion and faeces may vary among fish species depending on factors such as diet composition (Pandian, 1987; Cui et al., 1992), activity level (Perez-Pinzon and Lutz, 1991; Shulman and Love, 1999; Wuenschel et al., 2004) and food ration (Han et al., 2004). In living tissues, the bulk of energy is derived from the oxidation of the three main classes of foodstuffs, namely, carbohydrates, fats and proteins and the energy is utilized as follows. A considerable amount of
energy is converted to heat and is utilized to maintain the body temperature, some portion of energy is utilized for the performance of work like muscular contraction, secretory function and nerve impulse conduction, some amount of energy is stored temporarily in the high-energy phosphate bonds and stored for a longer period in the form of fat and glycogen to provide energy whenever required (Shanmugam, 2004).

Hence in the present study, following bio-energetic parameters (rate of feeding, absorption, conversion, metabolism and ammonia excretion) were calculated to evaluate the energy budget/ utilization by experimental prawns following influence of medicinal herbs.

5.2. MATERIALS AND METHODS

5.2.1. Estimation of the Parameters of Energy Utilization

The prawns were randomly divided into 10 groups with 25 animals in each group. The animals were then given the respective prepared feeds every day at a daily rate of 1% of their live weights. The sampling of animals was done after 90 days of the feeding trial. They were given three concentrations (1%, 3% and 5%) of each herbal supplement. Aeration was provided throughout the experiment. Each diet was fed to triplicate groups and the prawns were fed with the test diet for two days to acclimatize them to the feed. The water medium was renewed daily by siphoning method without severe disturbance to the prawn.

The energy balance was determined in the present study based on the IBP formula of (Petrusewicz and Mac Fadyen, 1970).

The IBP formula is determined as:

\[ C = (P+E) + R + F + U \]

where

- C - is the food consumed
- P - is the growth
- E - is the exuvia
R - is the material lost as heat due to metabolism
F - is the faeces and
U - is the nitrogenous excretory products

a. **Estimation of -C**

Each P.L. was uniformly allowed to feed the respective herbal supplements incorporated diets and control diet (1.0 g wet wt.) for 12 h per day. Every day, the unconsumed food was individually collected into a filter by siphoning method and dried in a hot air oven at 90°C for 48 h and weighed. To estimate the dry weight of food consumed, a sample of food was dried every day and the dry weight of the unconsumed food was subtracted from the dry weight of the food offered.

b. **Estimation of -F**

The faeces of the P.L. being ribbon-like settled at the bottom of the tanks were collected individually every day by siphoning into a blotting silk-filter. This was then dried in a hot air oven at 90°C for 24 h, weighed and stored.

c. **Estimation of -U**

The daily excretion of ammonia by the prawn was estimated in rearing water 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 NH3: 5.9 cal. (Elliot, 1976).

d. **Estimation of -E**

Everyday the moults if any produced by the experimental PLs were collected individually from each tank. After blotting the adhering water, the moults were weighed and dried overnight in a hot air oven (90°C) and the dry weight was then recorded. The number of moults per
individual was also observed. Since, the exuvia (E) constitutes part of the converted energy in crustaceans. In the present study the energy loss through exuvia was considered to be the part of conversion. The production of new tissues was calculated by adding the exuvial weight to the gain in total weight of the prawn and the actual growth was calculated by subtracting the exuvial weight from the gain in total weight.

e. **Estimation of -P**

The term conversion has been used to refer to growth i.e., the P of the IBP terminology. As already mentioned above, prior to the commencement of the experiment, the test PLs were starved for 24 h in order to evacuate the undigested food consumed the previous day. Subsequently, the wet (live) weight of each individual was determined at the commencement of each experiment. To estimate the initial dry weight the ‘sacrifice method’ was adopted (Mayard and Loosli, 1962). A group of twenty prawns of similar live weight and experimental state served as control to determine the initial weight and energy content. These P.L.s were sacrificed and dried in a hot air oven at 90ºC till they attained constant weight. The dry weight and energy content of these prawns were considered to represent those of the test individuals on the commencement of the experiment.

f. **Estimation of -R**

Following the estimation of C, F, U and P metabolism, R (Respiration) was calculated by following.

After 90 days of feeding experiment, the final length and weight of the experimental PLs were measured. For this, the P.L.s were scarified and dried in a hot air oven, as defined above, to estimate the energy content. Of the 100 test prawns in each group, twenty were randomly taken for the analysis of bioenergetics parameters. The energy content of dried food, PL, faecal matter and exuvia were determined by using oxygen bomb calorimeter (Parr 1024), and the energy was
calculated by subtracting F and U from C. Conservation or growth is the sum of energy channeled to somatic growth (P) and exuvia (E). The efficiency of absorption was calculated in percentage form by relating the food absorbed to the food consumed. Feeding rate (FR), absorption rate (AR), conversion rate (CR) and metabolic rate (MR) were calculated by dividing the respective amount of energy by initial live weight of the prawn per unit time in days.

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

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

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

Excretion Rate (ER) = \frac{\text{Mean NH}_3 \text{ Excretion (k.cal/g/day)}}{\text{Final live weight of the prawn (g)}}

Metabolic rate (MR) = \text{Absorption Rate (kg.cal/g/day)} - \text{Conversion Rate (kg.cal/g/day)} + \text{NH}_3 \text{ excretion Rate (kg.cal/g/day)}

5.3. RESULTS AND DISCUSSION

The rate of energy in feed intake, absorption, conversion, ammonia excretion and metabolism of the prawns fed with ten differently prepared diets were presented in the table-5.1. These were found to be significantly higher (P < 0.094) in the groups of prawn fed with 5% *Zingiber officinale*. Groups fed with the control diet showed lesser values compared to the other groups. Groups fed with *Allium sativum* have also shown to increase energy production, which increased with its increased supplementation. However, the production increased initially with the supplementation of *Curcuma longa* and then reduced with its increased supplementation.
Prawns obtain the energy they need by eating protein, lipid and carbohydrate (these nutrients are called the macronutrients). Feeding rate is important for the growth, feed conversion, nutrient retention efficiency and chemical composition of body tissue (Huisman et al., 1976; Henken et al., 1985; Hung and Lutes, 1987). Increased intake of feed, thereby the nutrients present in it have aided for the energy requirements of the prawns. Growth and metabolism are sustained by the energy generated from the catabolism of either dietary protein or non-protein energy sources (Ashraf and Goda, 2007). Without additional alternative energy sources (carbohydrates and lipids) to meet energy demands in the diet, some of the dietary protein consumed will have to be degraded to support the energy demands for tissue synthesis and metabolism (Hawkins and Bayne, 1991). Prawns derive immediate energy from carbohydrates sources. Dietary lipid plays important roles in providing concentrated energy, Essential fatty acids (EFA) and some other non-fat nutrients to organisms (NRC, 1993; Sargent et al., 2002). However, Habashy, (2009) states that, “An excess of carbohydrate energy also proves to be an obstacle to the development of the post larvae when the quantity of the ingested protein is insufficient. High cellulose if present, not only seemed to retard growth and depress locomotive activity”. A similar case being observed in the present study, when the control diet, having a maximum carbohydrate and lipid contents but considerably reduced amount of the protein portion resulted in poor growth of the diet fed groups. Energy ratios of about 125-130 mg protein/K cal are suitable for growth of *M. malcolmsonii* in clear water systems that do not have any supply of natural foods (Watanabe, 1988) also, Shiau and Lan (1996) reported that dietary protein requirements are closely related to dietary energy levels, which clearly states a good proportion of protein, amino acids, carbohydrates and lipids in balanced level in *Z.officinale* supplemented diets have resulted in far better growth of the freshwater prawn, since protein and energy are two “macronutrients” required for animal metabolism and growth. In contrast to micronutrients like vitamins, minerals,
and specific lipids, protein and substrates catabolized for energy must comprise most of the digestible matter in animal diets (Stephen et al., 1995).

In general herbs improves absorption rate (Citarasu et al., 2002). In the present study the digestive absorption of nutrients in the prawns was improved with the supplementation of Z. officinale in the diets. The digestive absorption property of Z. officinale have been already stated by Belewu, (2006); Ghayur and Gilani, (2005). Post-larvae of P. monodon fed with Z. officinalis enriched Artemia, showed increased feed intake, conversion and production efficiencies (Venketramalingam et al., 2007). Part of the food ingested is assimilated in the gut and the remaining fraction is eliminated as faeces. The amount of food assimilated is dependent on the gut content and assimilation efficiency (Franco et al., 2006). The feed conversion rate is one of the important parameters of feed quality. The Conversion rate is expressed as a ratio between foods consumed for increase per unit weight gained by the body discounting the food energy requirement by the prawn for its maintenance and energy requirement (Piska and Naik, 1999). It is evident from the rate of weight gain in the groups fed with medicinal plant supplemented diets (Table-4.1.).

Prawns fed with the control diet suffered due to energy insufficiency as a low calorie diet results in a high feed conversion ratio and hence, in low efficiency, implying that prawns consume more food to overcome energy insufficiency. From this, it appears that control of food consumption through dietary energy density is possible and is economical. Biologists have often observed an inverse relationship between diet energy density and ingestion rate, and generally have concluded that ingestion is regulated to meet the consumers’ need for energy (Rozin, 1961, Harper, 1976).
The present study clearly indicates that the protein requirements of the prawn have been met especially when these are fed with the diet supplemented with *Z. officinale* which could improve feed and nutrients intake as the excess protein in the diet will be metabolized by prawns as a source of energy, and nitrogen will be excreted as ammonia (Lim and Persyn, 1989). However, ammonia excretion rate can serve as a good indicator for the optimum dietary protein content, especially when combined with data on growth rate. This approach looks promising for determining protein requirements, which can reduce dietary costs and minimize the nitrogenous waste output (Li Du and Cui-Juan Niu, 2002). The proportion of ammonia excretion correlates with the proportion of protein in the feed (table-3.1.). Dietary protein will have a marked influence on nitrogenous excretion, with factors such as the protein level and the balance of amino acids in the diet being of particular importance. The excretion rate may be expected to be high when fish are fed feeds rich in protein (Cai *et al.*, 1996; Leung *et al.*, 1999; Engin and Carter, 2001). Thus it is also evident that, groups fed with *Z. officianle* have taken a higher proportion of protein 196.92±1.56 compared to control group 112.37±7.96 (table-4.1.). Moreover, the energy loss through ammonia in fish excretion accounts for 4-15% of ingested energy (Elliott, 1979; Cui and Wootton, 1988) and thus only a minimal fraction of energy is lost through ammonia excretion.

Metabolism is the set of chemical reactions that happens in living organisms to maintain life. These processes allow organisms to grow and reproduce, maintain their structures, and respond to their environments. The increase in metabolism after feeding is called specific dynamic action (Li Du and Cui-Juan Niu, 2002). It is also a major component in the energy budget of fish and has been reported to be dependent on several non-dietary factors, including body weight, density, and water temperature (Cho and Kaushik, 1985; Medland and Beamish, 1985), as a consequence of their influence on the overall metabolism of fish and other aquatic animals.
(Brett and Groves, 1979). From this it is evident that, increased metabolic rates favored better growth of the prawns

5.4. CONCLUSION

The present study further clearly states that the prepared experimental diets are well able to meet the energy requirements of the freshwater prawn *M. rosenbergii* post larvae when compared to the control diet and thus finds suitability for their use in aquafeed.
Table-5.1. Energy budgets of prawns fed with different feeds for a period of 90 days.

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Control Group-1</th>
<th>Experimental</th>
</tr>
</thead>
<tbody>
<tr>
<td>FR (k.cal/day)</td>
<td>0.40±0.04</td>
<td>0.42±0.03</td>
</tr>
<tr>
<td>AR (k.cal/day)</td>
<td>0.32±0.01</td>
<td>0.37±0.05</td>
</tr>
<tr>
<td>CR (k.cal/day)</td>
<td>0.06±0.001</td>
<td>0.08±0.007</td>
</tr>
<tr>
<td>U (k.cal/day)</td>
<td>0.033±0.003</td>
<td>0.037±0.004</td>
</tr>
<tr>
<td>MR (k.cal/day)</td>
<td>0.32±0.01</td>
<td>0.39±0.03</td>
</tr>
</tbody>
</table>

Each value is mean ± SD of triplicate observations. Significance (P<) of paired samples t-test are given in parentheses. FR – Feeding rate, AR- Absorption rate, ECR- Energy Conversion rate, U- Ammonia excretion and MR- Metabolic rate. (* the correlation and t cannot be computed because the SE of the difference is ‘0’).