International Journal of Zoology and Applied Biosciences **ISSN: 2455-9571**
Volume 1, Issue 4, pp: 198-204, 2016 **http://www.ijzab.com**
https://doi.org/10.5281/zenodo.1310610

Research Article

EFFECT OF FEEDING *SPIRULINA* DIETS ON THE RATES OF METABOLISM AND EXCRETION AND PROXIMATE COMPOSITIONS IN *XIPHOPHORUS HELLERI*

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Article History: Received 14th July 2016; Accepted 28th August 2016; Published 31st August 2016

ABSTRACT

Spirulina diet significantly influenced the rates of metabolic and ammonia excretion and proximate composition in *X. helleri.* The rate of oxygen consumption decreased as the *Spirulina* level increased. A significant ($P < 0.01$) and negative correlation was obtained between rearing period and rate of oxygen consumption while ammonia excretion elicited the reverse trend in *X. helleri* fed all the *Spirulina* diets. The protein content increased with time and *Spirulina* levels in the diet. However, the trend was reversed in lipid and nitrogen free extract. Ovary concentrated more quantum of energy than muscle. The energy content in tissues was gradually increased with time and it elicited the better performance in fish fed with 15% *Spirulina* diet than other diets. Based on the results, the inclusion of *Spirulina* in the diet has beneficial effects on the reduction of metabolic stress and lipid level. The depression of metabolic stress conserves the feed energy and utilization of fat resulting the fish become flabby and attractive.

Keywords : Respiratory metabolism, Ammonia excretion, Organic reserves, *Spirulina, Xiphophorus helleri.*

INTRODUCTION

Studies on metabolic rates of animals, priority has been given to the size and sex of organisms than other factors such as protein diet and density which influence the metabolic activities (Geetha Belliyappa *et al.,* 1983; James *et al.,* 1989). Ammonia is known to be the principal nitrogenous waste product excreted by freshwater fishes (Colt and Tohobanoglous, 1976; James and Sampath, 2003). Ammonia excretion in fish is also influenced by several endogenous factors including body size (Jobling, 1981) and stocking density (James and Sampath, 2003). The organic reserves of fish are of paramount importance in relation to nutritive value and physiological condition (Basade *et al.,* 2000; James and Sampath, 2004). Several authors have separately studied the respiratory metabolism, ammonia excretion and proximate composition in fishes in relation to size, sex, temperature and nutrition (Geetha Belliyappa *et al.,* 1983; Ramasamy and Gopalakrishna Reddy, 1983). However, there is paucity of information on the complete study of respiratory metabolism, ammonia excretion and organic reserves in ornamental fishes. Hence, the present study has been undertaken to assess the effect of *Spirulina* diet on respiratory metabolism, ammonia excretion and organic reserves in *Xiphophorus helleri* as a function of time and body weight.

MATERIALS AND METHODS

Healthy and active juveniles of 45 days old *X. helleri* (300 nos.) were collected from the laboratory bred brooders. They were divided into five groups and offered with five different levels of *Spirulina* diet (0, 5, 10, 15 and 20%). Each group consisting of 20 individuals was reared in circular cement tank (diameter: 58.5 cm; height: 40 cm, 120 l capacity) containing 100 l water. Triplicates were maintained for each group. The experimental tanks were filled with dechlorinated well water (Temperature : 29.0 ± 0.5^0C; pH : 7.71 ± 0.05; Salinity : 0.57 ± 0.01 ppt; Water hardness : 322 mg $CaCO_3 l^{-1}$; DO : 4.17 ± 0.12 ml l^{-1}).

Three individuals were collected from each experimental group and they were separately subjected to estimation of oxygen consumption and ammonia excretion at an interval of 25 days. Oxygen consumption was estimated following Winklers Idometric method. Oxygen consumed by each individual was converted to energy as 14.3 J mg^{-1} O_2 (Lucas, 1996). Ammonia excretion was

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estimated following the phenol-hypochlorite method (Solarzano, 1969). Excretion values were transformed to energy using an energetic equivalent of 20.5 J mg^{-1} N – NH_3 excreted (Lucas, 1996).

After the estimation of oxygen consumption and ammonia excretion, the same fishes were used for the estimation of proximate composition and energy content in muscle and gonad tissues. The muscle and gonad tissues were separately removed from test animals. They were dried in hot air oven at 50^0C for two days and then used for analysis. Protein and lipid contents of experimental animals were estimated following the method of Lowry *et al.* (1951) and Bragdon (1951) respectively. The energy content of test samples was estimated by wet combustion method (Karzinkin and Tarkovskaya, 1964). Regression analysis was carried out based on a least squares method (Zar, 1984). Duncan multiple range test also applied to detect differences between treatments (Sokal and Rolhf, 1973).

RESULTS

Spirulina diet significantly influenced the rates of oxygen consumption and ammonia excretion and proximate composition in *X. helleri*. The rate of oxygen consumption in *X. helleri* decreased as the *Spirulina* levels increased. Duncan multiple range test showed that, fish fed with 15 – 20% *Spirulina* diets did not show significant ($P > 0.05$) difference in oxygen consumption rate. The rate of oxygen consumption of juvenile *X. helleri* was higher in early rearing period (on day 25) and it gradually declined with time in all tested groups. There was a significant ($P < 0.01$) and negative correlation was obtained between rate of oxygen consumption and rearing period in *X. helleri* fed all the diets (Figure 1a). The result obtained for ammonia excretion rate was opposite to that of oxygen consumption rate. The 'b' value obtained for ammonia excretion rate of control *X. helleri* was 0.059 and it increased to 0.064, 0.092, 0.097 and 0.089 in fish received 5, 10, 15 and 20% *Spirulina* diets respectively (Figure 1b). It shows that, fish fed with 15% *Spirulina* diet excreted the more quantum of ammonia than those fed other diets.

The protein content increased in *X. helleri* with time and *Spirulina* levels in the diet. A significant ($P < 0.01$) and positive correlation was obtained between protein contents and time in *X. helleri* fed with *Spirulina* diets (Figure. 2). The decline of lipid content was maximum in fish fed with 15% *Spirulina* diet than those fed other diets. It was confirmed by the 'b' value obtained for lipid content. For instance, the slope values for the reduction of lipid content was -0.002, -0.004, -0.013 and -0.007 in fish fed with 5, 10, 15 and 20% *Spirulina* diets respectively (Figure. 2).

Ovary concentrated more quantum of energy than muscle. Fish fed on 15% *Spirulina* diet registered the more energy than those fed with other diets. However, Duncan multiple range test revealed that higher levels of *Spirulina* diets (15- 20%) did not show the significant ($P > 0.05$) difference in energy content of ovary (Table 1). Also, significant ($P < 0.01$) positive correlation was obtained between energy content in tissues and time (Figure 3).

Table 1. Effect of different levels of *Spirulina* on energy in muscle and ovary (KJ g^{-1} dry matter) of red swordtail, *Xiphophorus helleri*. Each value is the mean ($\bar{x} \pm sd$) of three estimations.

Rearing period (days)	Levels of *Spirulina* (%)				
	0	5	10	15	20
Muscle					
50	0.542 ± 0.04^a	0.636 ± 0.04^b	0.730 ± 0.01^c	1.059 ± 0.08^d	1.013 ± 0.05^d
75	1.295 ± 0.08^a	1.342 ± 0.04^b	1.483 ± 0.04^b	2.124 ± 0.11^d	2.069 ± 0.12^c
100	1.362 ± 0.11^a	1.738 ± 0.10^b	1.948 ± 0.07^c	2.936 ± 0.14^e	2.106 ± 0.08^d
Ovary					
50	ND	0.730 ± 0.02^a	0.824 ± 0.05^b	1.271 ± 0.05^c	1.248 ± 0.04^c
75	1.459 ± 0.05^a	1.766 ± 0.08^b	2.777 ± 0.14^c	3.178 ± 0.04^d	3.157 ± 0.23^d
100	1.954 ± 0.08^a	2.636 ± 0.12^b	3.060 ± 0.11^c	3.978 ± 0.08^d	3.954 ± 0.07^d

ND : Not determined

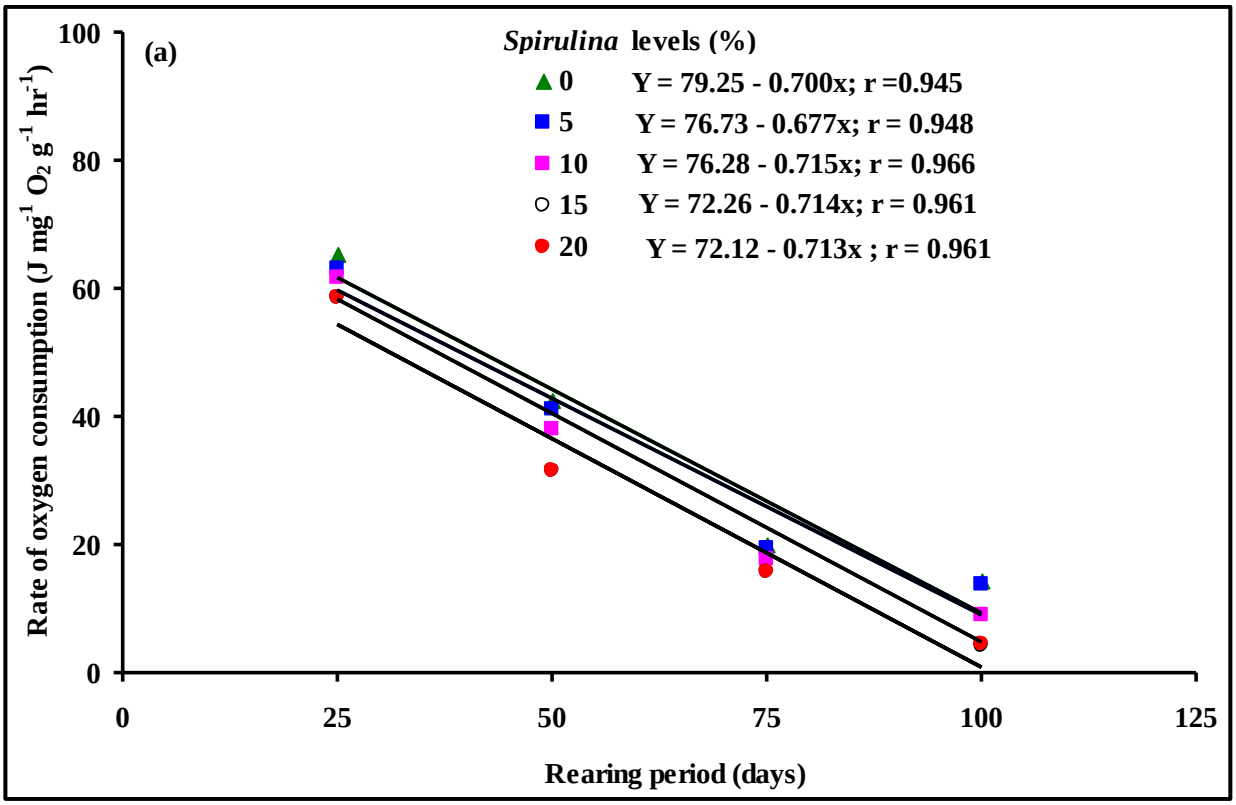
Values (mean ±SD) with different superscripts in the same row are significantly different ($p < 0.05$).

DISCUSSION

The present study revealed that, rate of oxygen consumption gradually decreased with time. A higher rate of oxygen uptake in the juvenile (early rearing period) may be related to the intense growth of the respiratory area and increased metabolic demand (Vasumathi *et al.*, 2001). The oxygen uptake per unit time by a fish is, as in other groups, is not linear but proportional to some fractional exponential function of the animal weight (Ambrose and Venkatachari, 1978). According to Von Bertalanfty (1957), the metabolism can be of three types depending upon the 'b' values (regression co-efficients) as surface proportional ($b = 0.67$), weight proportional ($b = 1.0$) and intermediate between the two ($b = 0.67$ to 1.0). The respiratory metabolism of *X. helleri* is inbetween the surface and body weight proportionality.

Ammonia excretion rate in *X. helleri* was linearly increased with increasing of *Spirulina* levels and time. It may be due

to test animal consumed high protein content of *Spirulina* diets resulting the more quantum of ammonia excretion. Nitrogen excretion rates were reported to be directly influenced by nitrogen consumption in bluegills, *Lepomis macrochirus*, fed high protein mealworms (Savitz, 1971), common carp, *Cyprinus carpio* fed *Chironomus* larva



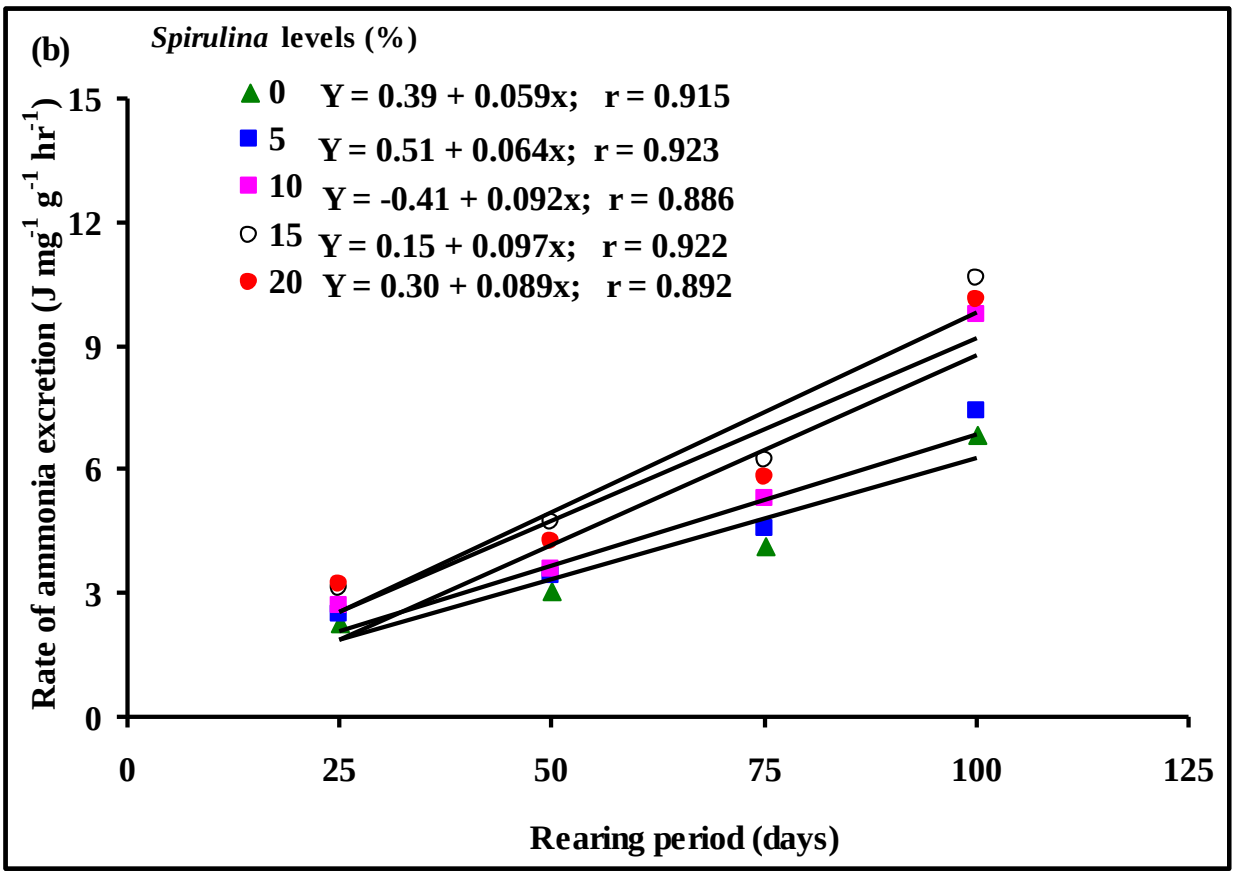


Figure 1. Regression lines for the rates of oxygen consumption (a) and ammonia excretion (b) in *Xiphophorus helleri* as a function of *Spirulina* levels and time.

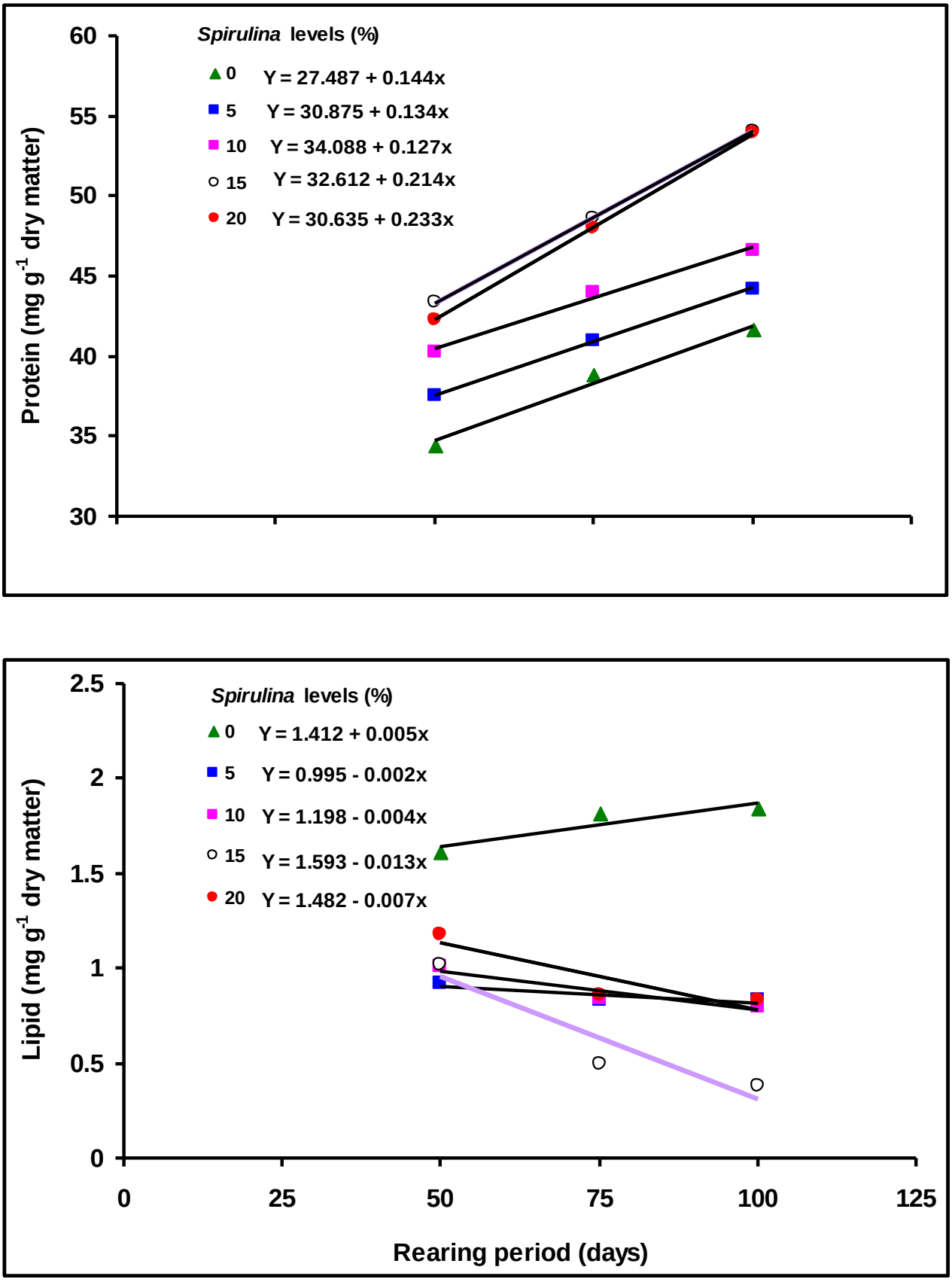


Figure 2. Effect of different levels of *Spirulina* on protein and lipid contents in *Xiphophorus helleri*.

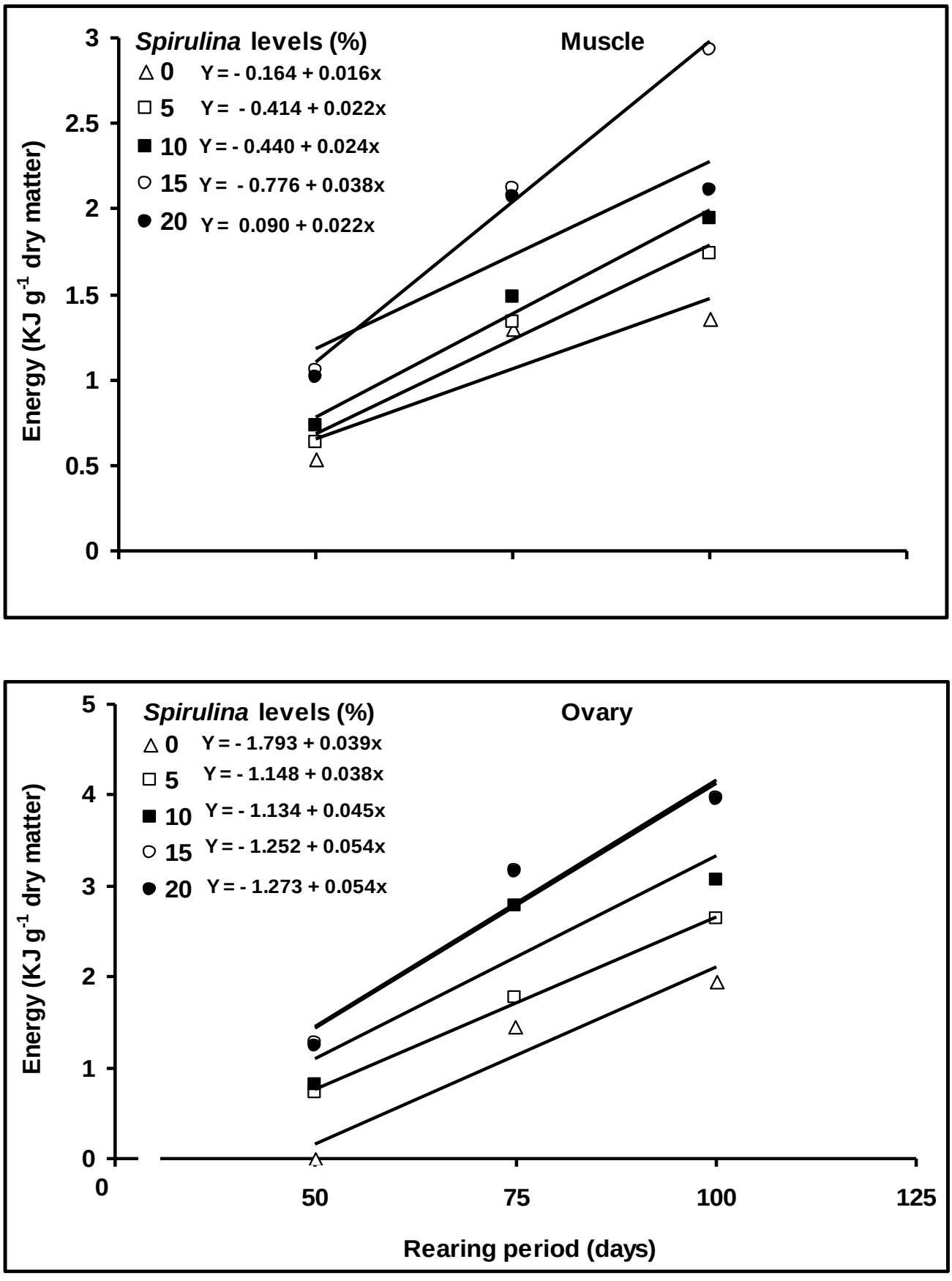


Figure 3. Regression lines for energy contents in tissues of *Xiphophorus helleri* as a function of *Spirulina* levels and time.

(James *et al.*, 1993) and 55% protein diets (Kaushik, 1980) and sea bass, *Dicentrarchus labrax*, fed diets with 40, 44 and 49% protein (Ballestrazzi *et al.*, 1994) supports the present observation. On contrary, ammonia excretion was not linearly increased with increasing dietary protein intake in rainbow trout fed with 35, 40 and 45% dietary protein.

Protein and energy contents of *X. helleri* were increased with increasing of *Spirulina* levels in the diet. It indicates that *Spirulina* contains high amount of protein (50%) which are concentrated in the body of *X. helleri* when consumed *Spirulina* diets. Juveniles grew with time, protein and energy contents were also increased. This indicates the deposition of nutrients in the body with the advent of reproductive period (Lambert and Dehnel, 1974). It has also been shown that the organic materials particularly the protein, carbohydrates and lipid may serve as a reserve food material to be utilized for the formation of gonadal element (Basade *et al.,* 2000; James and Sampath, 2004). James and Sampath (2003) found that allocation of more amount of assimilated feed energy for gonadal and embryo development in *Xiphophorus helleri*, supports the present study.

Fish fed on *Spirulina* diets reduced the lipid level in *X. helleri* as compared to control diet. It suggests that fat utilization for growth and other physiological activities instead of just storing it. Sayed (1994) observed that high amount of *Spirulina* incorporated in fish meal diet reduced the lipid level in silver seabream, *Rhabdosargus sarba.* Watanabe *et al.* (1990) observed a significantly lower lipid level in striped jack (*Pseudocaranx dentex*) fed with 10% *Spirulina* in the diet as compared to control diet, supports the present observation.

CONCLUSION

It concludes that the inclusion of *Spirulina* in the diet has beneficial effects on the reduction of metabolic stress and lipid level. The depression of metabolic stress conserves the feed energy for growth and reproduction and utilization of fat resulting the fish become flabby and attractive.

ACKNOWLEDGEMENT

Authors wish to thank the Department of Zoology, V.O. Chidambaram College for providing facilities to carry out this research work.

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