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Original Research Paper

Alterations in the Nutritional Parameters of Silkworm *Bombyx mori* L. on Exposure to Selenium

A. Vijaya Bhaskara Rao and S. Smitha*
Department of Ecology and Environmental Sciences, Pondicherry University, Puducherry-605 014, India
*STSN Govt. Degree College, Kadiri, A.P., India

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ABSTRACT

The silkworm, *Bombyx mori* L. feeds on mulberry leaves from which it ingests various nutrients to support physiological activities. Nutritional parameters like food consumption, excretion, assimilation, food combustion and total food converted under lethal and sub lethal doses of selenium at 3, 4, 5, and 6 days of V instar silkworm were studied. The selenium at lethal doses could decrease the parameters like food consumption assimilation efficiency and food conversion rate. However, the sub-lethal doses of selenium, particularly at 6 days of exposure, could exhibit significantly increased levels of the above parameters in the silkworm leading to its better survival.

INTRODUCTION

Study of various nutritional parameters such as food consumption, excretion, food assimilation and oxidization give an idea regarding growth and energy of an organism. A deficiency of certain nutrients such as phosphorus or Vitamin-B exerts an indirect influence on increase in size by decreasing appetite, as well as causing direct physiological effects. Restriction in diet up to 800 days of age in rats resulted in much leaner animals with somewhat less skeletal size, but improved health, female fertility and longevity and delayed the onset of degenerative disease (Benjamin 1960, Benjamin & Simms 1960). Tonge & McCance (1965) exhibited that growth retardation is due to food restriction in pigs. It was reported that the appetizing factors, biting factors, swallowing factors and repellent substances play a key role in accepting the diet (Hanamura et al. 1962). Food ingestion by the larvae of the silkworm *Bombyx mori* L. varies during its different instars. The intake of mulberry leaves into oral cavity is ingestion. Active feeding occurs only during IV and V instars and 97% accounts for total ingestion. The larval feeding is discontinuous, and initial feeding time is different among various instars. This phenomenon of feeding helps the newly moulted larvae in hardening of new cuticle and the continuous development of internal organs (Naik 1985, Radhakrishna 1989). The duration of feeding is only about 27 percent of the larval feeding period. Each time of feeding lasts for 12-16 minutes. Delvi (1972) described the cessation of appetite considerably 20-30 hrs prior to moult in many insects as pre-moult starvation period. The complex organic nutrients in mulberry leaves are covered by cell wall of cellulose hence are insoluble, impermeable and not utilized by the silkworm directly. Silkworm needs to convert the macro, complex insoluble and impermeable substances into simple, permeable products by the process of digestion.

Food in the buccal cavity is digested primarily by the saliva and lubricated and expelled to midgut, which is the main region for digestion and assimilation. The goblet cells of midgut play a key role in digestion by secreting digestive juices and absorb nutrients into the haemolymph through cylindrical cells of the midgut. The remains are combined with the secretions by the Malpighian tubules, the mixtures are pressed by the colon into the hexagonal excrements and then expelled into rectum. Water in the excrements is reabsorbed and the faecal matter is pressed further and excreted as the undigested matter in the form of solid faecal pellets. Even though the literature available on selenium nutritional aspects of vertebrates is abundant, a little is known about lower organisms such as arthropods. Hence, the authors have made an attempt to study various nutritional parameters on exposure to lethal and sub-lethal doses of selenium.

MATERIALS AND METHODS

In the present investigation the cross breed of V Instar silkworm, *Bombyx mori* L., larvae from the parentage of $PMXNB_4D_2$ used as test insects. They were obtained from the Government Grainage, Anantapur district, A. P. Since sodium selenite is readily soluble in water, a pure salt with

molecular formula of Na_2SeO_3 with molecular weight of 172.94 was selected. Food budget involves the various nutritional parameters such as food ingestion, faeces defaecated, total food assimilated, total food oxidised and total food converted. These nutritional parameters were estimated by the methods of Waldbauer (1964) and Delvi & Pandian (1972). Consumption was determined by subtracting the dry weight of uneaten food from the dry weight of the food provided (Waldbauer 1964). All faeces were separated daily at 6 a.m. from the rearing tray prior to first feeding and its dry weight was taken as measurement for excretion. Dry food assimilated by the test individuals during the final instars was calculated subtracting the dry weight of the faeces produced from the dry food consumed. Assimilation of food was calculated by the method followed by Delvi & Pandian (1972). The total amount of food converted into body substance was calculated by subtracting the dry weight of the individual before the experiment from the dry weight of the individual after the experiment. Food oxidised was calculated by subtracting the food converted from the food assimilated. Food utilization budget of silkworms was studied using IBP terminology (Petrusewicz & MacFayden 1970). The data obtained for each parameter were analysed for their significance by Duncan's multiple range test (Duncan 1955). The significance was calculated at 5% level ($P < 0.05$).

RESULTS

The data on various nutritional parameters of V instar silkworm *Bombyx mori* (groups 2, 5, 8 and 11) exposed to lethal and groups 3, 6, 9 and 12 to sub-lethal doses of selenium at 3, 4, 5 and 6 days of exposure period along with controls are presented in Table 1. For comparative assessment, the differences obtained in relation to controls in nutritional parameters at the said exposure periods of lethal and sub-lethal doses were converted as percentage of the corresponding controls (groups 1, 4, 7, and 10) and these percent change values are also given in Table 1 and plotted against exposure periods in Fig. 1. It is observed that relative to controls, the total amount of food consumed at 3, 4, 5 and 6 days exposure of V instar silkworms to lethal dose of selenium, was significantly ($P < 0.05$) decreased. In the sub-lethal dose groups, however, the amount of food consumed gradually increased in all days of the exposure periods studied and this increase was significant ($P < 0.05$). The percent increase was high at day-6 when compared to that of day-3. Corresponding to the decrease in food consumption in lethal dose of selenium treated groups, the excretion of faecal matter also decreased significantly ($P < 0.05$) in relation to controls groups on all exposure periods. Even in sub-lethal dose (groups 3, 6, 9 and 12), a significant decrease was observed at 3, 4, 5 and 6 days of exposure periods in faecal output corresponding to the decrease in food consumption. Faecal output of the V instar silkworm exposed to lethal dose of selenium and the other nutritional parameters such as food assimilation, conversion and oxidation registered a significant ($P < 0.05$) decrease at 3, 4, 5 and 6 days of exposure periods studied. In sub-lethal dose of selenium treated groups all the nutritional parameters registered an elevation at 3, 4, 5 and 6 days of exposure.

DISCUSSION

The parameters like food consumption, digestion and utilization in insects are of immense importance to understand the nutritional aspects in insects (Waldbauer 1964). The results presented on food consumption, digestion, utilization and excretion of silkworm bring out many significant and interesting aspects with regard to the lethal and sub-lethal

Table 1: Estimation of nutritional parameters (mg/kg body weight/day/larva) at 3, 4, 5 and 6 days in V instar of silkworm fed on mulberry leaves treated with lethal and sub-lethal doses of selenium.

Days exposure	Dosage	Consumption	Excretion	Assimilation	Conversion	Oxidization
3 days	Control	510 b	254 c	256 b	21.24 b	234.76 b
	Lethal	400 a (-21.56)	216 a (-14.9)	184 a (-28.1)	16.0 a (-24.6)	168 a (-28.4)
	Sub-lethal	560 c (+9.8)	221.2 b (-12.9)	338.8 c (+32.3)	29.47 c (+38.7)	309.33 c (+31.7)
4 days	Control	605 b	302 c	303 b	24.24 b	278.75 b
	Lethal	470 a (-22.31)	253.8 a (-16.2)	216.2 a (-28.7)	18.7 a (-22.8)	197.5 a (-29.14)
	Sub-lethal	685 c (+13.2)	270.5 b (-10.4)	414.5 c (36.6)	36.0 c (+48.5)	378.5 c (+35.7)
5 days	Control	720 b	359 c	361 b	29.7 b	331 b
	Lethal	515 a (- 28.47)	278 a (-22.5)	237 a (-34.3)	20.61 a (-30.6)	216.3 a (-34.6)
	Sub-lethal	810 c (+12.5)	319.95 b (-10.8)	490.05 c (+35.7)	42.6 c (+43.4)	447.45 c (+35.1)
6 days	Control	940 b	469 c	471 b	38.8 b	432.18 b
	Lethal	600 a (-36.17)	324 a (-30.9)	276 a (-41)	24.01 a (-38.14)	251.9 a (-41.7)
	Sub-lethal	1115.0 c (+18.6)	440 b (-6.2)	675 c (+43.3)	58.7 c (+51.2)	616.3 c (+42.6)

Each value is a mean of eight estimates; Percent decrease/increase over control is given in parentheses; Mean within a column followed by the same alphabet are not significantly different ($P > 0.05$) from each other according to Duncan's Multiple Range Test.

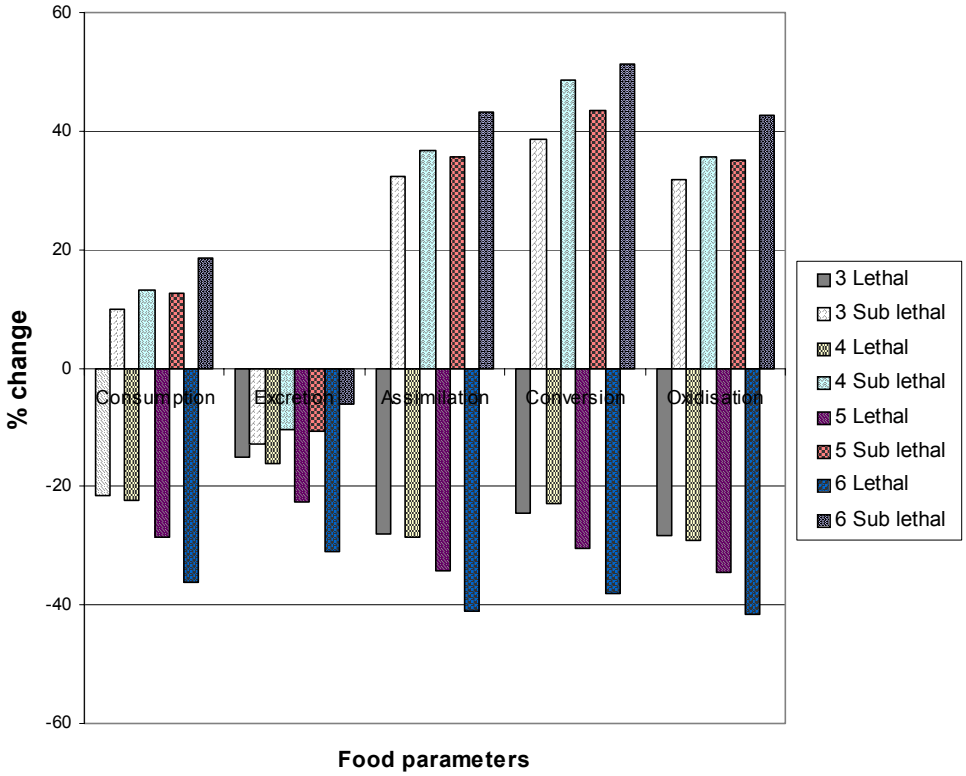

Fig. 1: Estimation of nutritional parameters (mg/kg body weight/day/ larva) at 3, 4, 5 and 6 days in V instar of silkworm fed on mulberry leaves treated with lethal and sub-lethal doses of selenium.

doses of selenium at different days. The literature on food utilization budgets related to selenium effects on animals is very scanty (Harrison & Conrad 1984). The food intake in selenium treated silkworms was significantly reduced at lethal intoxication, whereas sub-lethal dose enhanced the food intake significantly when compared to the controls. Here, the lethal dose acted as an inhibitor and sub-lethal dose acted as an attractant. However, these changes in the food intake may be attributed to food assimilation efficiency, when fed with selenium, may not be entirely due to the decrease in the consumption, but it may also be due to the action of selenium, which enhances the digestibility by increase in enzyme secretion rate/activity at least in the sub-lethal selenium exposed silkworm. It has been suggested that selenium acts as an oxidant functioning as the metal co-factor for important enzymatic activity requiring glutathione peroxidase (Mayland 1994). Lethal dose of an insecticide when fed to *Philosamia ricini* resulted in inhibition of acetyl choline activity and the inhibited esterase may induce the toxicity that results in the lack of appetite in silkworm (Pant et al. 1982). The decrease in food assimilation at lethal intoxication is reportedly due to lack of enzymatic mechanism, due to breakdown of proteins into amino acids and peptones and also may be due to the malfunction of mid intestine for transport of amino acids and peptones to respective organs. The results coincide with the reports of Lemly (1998), who observed that high concentration of selenium that substituted for sulphur in sulphur-to-sulphur linkages of proteins. This results in its inability to form a helical structure leading to non-functioning of malformed proteins. Such an inhibitory activity in the digestive process might also have brought in the silkworm of the present study during the lethal dose exposure of selenium. The total food assimilation and oxidation in silkworm treated with lethal selenium dose are suppressed and silkworm treated with sub-lethal dose of selenium are enhanced. The results are in agreement with Deka et al. (1999) as in non-mulberry silkworms. The enhanced assimilation and oxidation of food is probably due to the better consumption of food and enhanced transaminase activity of the intestine and the haemolymph of the silkworm as suggested by Shyamala et al. (1956) with reference to toxicity of chloromycetin. The lepidopteran larvae are able to adopt and express in different ways in various environmental conditions by altering one or more parameters of food utilization. The efficiency increases during toxicant supplement is in agreement with the homolygosis hypothesis, which

credits that sub-lethal dose of any stress agent may be stimulatory to the organism by providing it increased sensitivity or respond to change in its environment and increased efficiency for coping with suboptimum environment (Lucky 1968). Cholinesterase inhibition may induce the toxicity by killing the larva reflecting on their lack of appetite and under nourishment. This further leads to the lysis of all nutrients such as carbohydrates, glycogen, proteins and lipids, and also an increase in lipolytic, proteolytic, phosphorylase and aminotransferase activities (Pant et al. 1982, Pant & Katiyar 1983). From the present study, it is evident that the findings of the present study are in agreement with earlier reports.

In the present study, the selenium at lethal doses could decrease the parameters like food consumption assimilation efficiency and food conversion rate. However, at the sub-lethal dose of selenium particularly at 6-day of exposure, silkworm could exhibit significant increased levels of these parameters leading to better survival and normal life by suppressing toxic action by detoxification mechanisms.

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