

[Note]

## Efficacy of Selenium on Silkworm (*Bombyx mori* L.) Cocoon Characters

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**As the Selenium is known to be growth promoter in sheep and cattle, the efficacy of selenium has been tested in silkworm *Bombyx mori* L. The V instar larvae were fed with lethal and sub-lethal doses of selenium treated leaves. The larvae fed with lethal dose showed a significant decrease in growth and Cocoon commercial characters. The treatment with sub lethal dose exhibited a significant increase in the growth of the silkworm. Correspondingly, the cocoon commercial characters on exposure to the lethal dose showed significant decrease and sub lethal dose showed a significant increase. In the light of similar findings reported earlier in other cocoon crops and vertebrates, it can be inferred that selenium at lower doses acts as a growth stimulator, resulting in the higher yield of cocoon crop.**

**Key words:** Selenium, *Bombyx mori*, LD<sub>50</sub>, Cocoon characters.

### Introduction

The quality of feed is one of the important parameters in determining the silkworm cocoon yield. There were many studies to improve quality and quantity of cocoon by supplementing nutrients such as sugars, proteins, lipids, vitamins, trace elements etc. (Nagarajan and Radha, 1990; Masilamani *et al.*, 1991; Radhakrishnaiah and Chamundeswari 1994). It is known that trace elements play a key role in the metabolism of animals. Among them, selenium occupies a special place due to the small margin between the toxic and physiological dose response. Selenium is required in trace quantities in diets of most ani-

mals (Flow *et al.*, 1979). It is an essential nutrient in the diets of mature poultry and it is known that sodium selenite is an effective selenium supplement (Ort and Latshaw, 1977). Metabolic disorders resulting from selenium inadequacy have been recognized practically in all the major livestock producing countries of the world. Levander and Beck (1977) reported the association of Keshan disease with selenium deficiency and it is also responsible for an enlarged heart and poor heart function. Studies on selenium deficiency proved that the active synthesis of thyroid hormone was associated with the selenium and its deficiency lead to abnormality of thyroid functioning (Arthur, 1991). It is evident that the involvement of selenium in maintaining structure and functional efficacy of mitochondria (Rani and Lalitha, 1996). Selenium supplements were investigated in terms of their effect on live weight and wool production in sheep and it had increased the wool length and fiber diameter (Langlands *et al.*, 1991). The known function of selenium is that it is an important part of antioxidant enzymes that protects cells against the effects of free radicals that are produced during normal oxygen metabolism which would otherwise can damage cells and contribute to the development of some chronic diseases (Combs *et al.*, 1997). Bansal and Parmindar Kaur (2005) showed that the absence of selenoprotein of low molecular weight which can be compared to cytochrome, caused muscular dystrophy in selenium deficient sheep. Although, much information is available on the effects of selenium on mammals, a little information has been reported in insects (Deka *et al.*, 1999) and they demonstrated that selenium could increase the economic characters of Eri silk cocoon. Supplementation of the diet with selenium enhances the *Dosophila* life span by a process that may involve the newly identified proteins (Clyburn *et al.*, 2001). Since studies pertaining to growth and cocoon commercial characters of mulberry silkworm *B. mori* exposed to selenium are scanty, the present investigation was under taken to study the efficacy of selenium under lethal and sub lethal doses of selenium exposure.

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## Materials and Methods

The silkworm hybrid PM×NB<sub>4</sub>D<sub>2</sub> was selected for the study. The disease free layings of this cross breed were incubated and reared as per the rearing technology as advocated by Krishna Swamy (1986). The chemical Sodium selenite was procured from S.D. Fine Chem. Ltd., Mumbai, India is used in the present study. Evaluation of 24 hour LD<sub>50</sub> was done through graphical plots of with percent mortality as well as probit mortality verses log doses of selenium concentration suggested by Finney (1971). For subsequent verification of the selenium LD<sub>50</sub> obtained by graphical methods Dragsted-Behrens was employed as given by Carpenter (1975). After IV moult, the larvae of same size and age were collected from the rearing house and divided into batches and each batch consists of 50 larvae. They were maintained at 23.5±1°C. Fresh leaves of *Morus alba* variety Victory-1 collected from the garden and are sprayed with lethal and sub-lethal doses selenium, prepared for the experimentation separately, and dried under fan at room temperature. A batch of silkworms were fed without selenium as control. As it is an established fact that at 120 hrs after resumption from the IV moult, the silkworm larvae of V instar are very active, at 144 hrs after IV moult for V instar were fed with selenium treated leaves at different intervals of 6.00, 10.00, 16.00 and 22.00 hrs in a day. Afterwards, Cocoon commercial characters were analysed. Variability in cocoon commercial characters was determined using DMR (Duncan Multiple Range) test (Megharaj *et al.*, 1999).

## Results and Discussion

In the present study the 24 hour LD<sub>50</sub> obtained for V instar

of silkworm *B. mori* is 32.29 µg/kg body weight. LD<sub>50</sub>/24 of selenium was taken as lethal dose and about one fifth of the 24 hour LD<sub>50</sub> i.e., 6.47 µg/kg body weight for silkworm *B. mori* was taken as the sub-lethal dose of selenium for further studies.

Data on the effect of selenium in the lethal and sub-lethal doses to the silkworm on growth and cocoon commercial characters were presented in the Table 1. The results indicated that the cocoon commercial characters such as cocoon length, cocoon width, single cocoon weight, , shell ratio, filament length and filament weight of silkworm, *Bombyx mori* L. are significantly decreased when exposed to selenium in lethal dose (32.29 µg/kg b.w.). Significant decrease was observed in these parameters at lethal dose of selenium, it may be attributed to the affected silk protein synthetic path way and due to some physiological alterations. Many researchers, Wyatt *et al.*, 1957; Srikanth *et al.*, 1988; reported that reduced intake of food ingesta causing severe fall in the total carbohydrate level, which ultimately lead to low energy and affected the protein synthetic mechanism. It is known that under stress conditions, decreased proteins, amino acids in the haemolymph and fat bodies were observed (Von Brand 1979; Venkatarami Reddy *et al.*, 1992). However, cocoon commercial traits were significantly improved on exposure to sub-lethal dose (6.47 µg/kg b.w.) of selenium. Significant increase in the commercial characters at lower dose of selenium might be due to enhanced silk gland protein metabolism. The improvement in commercial characters of cocoon, coincides with the work done by Deka *et al.* (1999), on the effect of sodium selenite on Eri silk production. These results suggest that selenium in minute quantities trigger the protein synthesis in the silkworm. The results were also in conformity with significant growth of silkworm on exposure to sub-lethal dose of

**Table 1.** Cocoon characters of silkworm *Bombyx mori* L. fed with Mulberry leaves supplement with selenium

S. No.	Parameter	Control	Lethal	Sub-Lethal
1.	Cocoon Length (cm)	32.92 <sup>b</sup>	31.32 <sup>a</sup> (-4.86)	38.51 <sup>c</sup> (16.98)
2.	Cocoon width (cm)	18.02 <sup>b</sup>	16.67 <sup>a</sup> (-10.87)	21.83 <sup>c</sup> (21.7)
3.	Cocoon Weight (gms)	1.481 <sup>b</sup>	1.1949 <sup>a</sup> (-19.31)	1.921 <sup>c</sup> (29.70)
4.	Shell Weight (mg)	0.260 <sup>b</sup>	0.185 <sup>a</sup> (-28.84)	0.368 <sup>c</sup> (41.53)
5.	Cocoon Shell Ratio (%)	17.09 <sup>b</sup>	12.86 <sup>a</sup> (-24.75)	21.16 <sup>c</sup> (23.81)
6.	Filament length (mts)	540.02 <sup>b</sup>	364.59 <sup>a</sup> (-32.50)	764.32 <sup>c</sup> (41.48)
7.	Filament Weight (mgs)	220.31 <sup>b</sup>	201.22 <sup>a</sup> (-19.2)	253.83 <sup>c</sup> (15.16)

\*Each Value is a mean of eight estimations.

\*\*Percent decrease over control is given in parenthesis

\*\*\*Means within a column followed by the same letter are not significantly different (P>0.5) from each other according to Duncan's Multiple Range Test.

selenium (6.47 µg/kg b.w.). Mayland (1994) reported that selenium deficiency produce marked decrease in growth and greatly interfere with efficiency of food utilization by animals. Hence it can be recognized that the micro quantities of selenium has improved the food utilization and growth and increased energy status, which could be utilized for efficient protein synthesis. In the present investigation, selenium profoundly influenced the biochemical pathways to improve the important economic traits of cocoon of silkworm *Bombyx mori* L. It can be assumed that the harmful microbial flora of silkworm might have also been eliminated in the sub lethal doses without affecting the silkworm which ultimately improved general health of the larvae leading to the better expression of cocoon commercial characters. In the light of similar findings, it was reported earlier in sheep as increased wool production, body weight gain and productivity rate with selenium supplementation. (Wilkins *et al.*, 1982; Langlands *et al.*, 1991; Whelan *et al.*, 1994).

Thus the results on the whole, indicate that Selenium at sub-lethal dose when supplemented with Mulberry V-1 variety leaves acts as a growth stimulant resulting in the higher production of cocoon crops and promotes the silk production in *B. mori* L.

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### References

- Arthur, T. R. (1991) The role of selenium in thyroid hormones metabolism. *Can. J. Physiol. Pharmacol.* **69**, 1648-52.
- Burk, R. F. and Hill K. E. (1993) Regulation of Seleno proteins. *Ann. Rev. Nutrition.* **13**, 65-81.
- Carpenter, P. L. (1975) *In: Immunology and serology* III edition, W.B. Saunder Company, Philadelphia, London, Toronto, pp. 254.
- Clyburn, B. S., Richards son, C. R., Montgomery, J. L., Pollard, G. V., Hearing, A. D. and Miller, M. F. (2001) Effect of selenium source and Vitamin E level on performance and meat quality of seed lot steers *In: Science & Technology in the feed industry.* (T. P. Lyons and K. A. Jacques, eds.) pp. 377-392 Nottingham University Press, Nottingham, U.K.
- Combs, G. F., J.r., Clark, L. C. and Turn Bull, B. W. (1997) Reduction of cancer risk with an oral supplement of selenium. *Bio. Med. Environ. Sciences* **10**, 227-234.
- Deka, J., M. R. H. Azad, S. Saharia, D. K. Sharma and A. Borkotoki (1999) Effect of sodium selenite on nutritional indices, cocoon characters, silk gland, protein and productivity of silk yarn of ERI silkworm, (*Samia Cynthia*, *S. Ricini*). Proceedings of NSTS, 3, Department of Sericulture, University of Agric Sciences, GKVK, Bangalore.
- Finney, D. T. (1971) Probit Analysis. 3<sup>rd</sup> Edition, Cambridge University Press. London and New York, pp. 336.
- Flow, L., W. A. Gunzler and G. Loschen (1979) *In Trace elements in Human health and disease* (Knarash, N., ed.), Academic Press, New York. pp. 263-286.
- Horie, Y. (1980) Quantitative requirements of nutrients for growth of the silkworm, *Bombyx mori* L., *JARQ*, **12**, 210-213.
- Krishna Swamy, B. (1986) New Technology of silkworm rearing, Bulletin 6, C.S.R. & T.I., Mysore, pp. 16-20.
- Langlands, J. P., Donald, G. E., Bowels, J. E. and Smith, A. J. (1991) Sub clinical selenium insufficiency. 1. Selenium status and the response in live weight with selenium. *Aus. J. Expt. Agric.* **31**, 25-31.
- Levander, O. A. and M. A. Beck (1997) Interacting nutritional and infectious etiologies of Keshan diseases. Insights from coxsackie virus B-induced myocarditis in milk deficient of selenium or Vitamin-E. *Biol. Trace Chem. Res.* **56**, 5-21.
- Masilamani, S. Subramniyam, R. K., Chikkanna and Noamani, M. K. R. (1991) The role of secondary metabolites of mulberry leaf in silkworm feeding. *Indian Silk*, **30**(1), 44-46.
- Mayland, H. F. (1994) Selenium in plant and animal nutrition. *In: Frenxberger*, (Ed. W.J. Jr., and S. Besen). Selenium in the environment, New York. Marcel Dekker, pp. 29-45.
- Megharaj, M., K. Kookana and S. Singleton (1999) Activities of fenamiphos on native algae population and some enzyme activities in soil. *Soil Biol. Biochem.* **49**, 1549-1553.
- Bansal, M. P. and Parmindar Kaur (2005) Selenium, a versatile craze element: current research implications. *Ind. J. Experi. Biol.* **43**, 1119-1129.
- Nagarajan, P. and Radha, N. V. (1990) Supplementation of amino acids through mulberry leaf for increased silk production. *Indian Silk*, **29**(4), 21-22.
- Ort, J. F. and J. D. Latshaw (1977) The toxic level of sodium selenite in the diet of laying chickens. Journal Article, 164, 77 of the Ohio Agri. Research and Development Centre, Wooster, Ohio.
- Radhakrishnaiah, K. and Chamundeswari, P. (1994) Effect of zinc and nickel on the larval and cocoon characters of the silkworm, *Bombyx mori* L. *Sericologia*, **34**(2), 327-332.
- Rani, P. and Lalitha, K. (1996) *In: Evidence for altered structure and impaired mitochondrial electron transport function in selenium deficiency.* *Biological Trace Elements. Research*, **51**(3), 225-234.
- Srikanth, J., Basappa, H. and Lingappa, S. (1988) Effect of parasitisation on food consumption and growth of the mulberry silkworm, *Bombyx mori*. *Insect Sci. Appl.* **9**, 373-379.

- Venkatarami Reddy, K., Benchamin, K. V. and Rama Devi, O. K. (1992) Metabolic profiles of Haemolymph and fat body of the silkworm, *Bombyx mori* in response to parasitisation by Uzi fly, *Exorista sorbillans* during the final instar. *Serico-logia*, **32**, 227-233.
- Von Brand, T. (1979) Patho physiology of the host in Bio chemistry and physiology of endo parasites. Elsevier North Holland Bio medical press. Amsterdam, pp. 321-390.
- Whelan, B. R., Barrow, N. J. and Peter, D. W. (1994) Selenium fertilizers for pastures grazed by sheep wool and live weight responses to selenium. *Aus. J. Agri. Res.* **45**, 877-887.
- Wilkins, J. F., Kilgour, R. J., Gleeson, A. C., Cox, R. J., Geddes, S. K. and Simpson, I. H. (1982) Production responses in selenium supplemented sheep in northern New South Wales 2. Live weight gain wool production and reproductive performance in young Merino ewes given selenium and copper supplements. *Aus. J. Exper. Agric. Anim. Husban.* **22**, 24-28.
- Wilkinson, L. 1990. 'SYSTAT: The System for Statistics. (SYSTAT, Inc.: Evanston, Illinois). 48.
- Wyatt, G. R. and Kalf, G. F. (1957) The Chemistry of Insect haemolymph Trehalose and other carbohydrates. *J. Gen. Physiol.* **40**, 833-847.