RESEARCH ARTICLE

EFFECT OF SPINACIA OLERACEA (SPINACH) ON MORPHOMETRIC PARAMETERS OF SILK WORM BOMBYX MORI

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ABSTRACT

Sericulture is both an art and science of raising silkworms for silk production. India’s traditional and culture bound domestic market and an amazing diversity of silk garments that reflect ‘geographic specificity’ gave India the status of being the second largest producer of raw silk after China. Nutrition plays an important role in improving the growth and development of the silkworm, Bombyx mori. L. and the silk production is dependent on the larvae nutrition and nutritive value of mulberry leaves and finally in producing good quality cocoons. The cocoon spinning activity is an important phase in the silkworm which produces the cocoons, the final product of the insect. This activity which lasts for 5 to 6 days requires continuous function of the nervous system and the muscular system where the Cholinergic and Glutamergic neurotransmitters play key roles. Since, the larval stage is the only feeding stage in silkworm development, intake of balanced diet is very essential for silk production. By supplementing the diet with minerals, vitamins and trace elements, the various functions of the hormonal system, neuromuscular system, reproductive system etc. can be modulated effectively. The current investigation highlights the effect of different concentrations of spinacia oleracea on the morphometric parameters viz. larval length, width and weight of the silk worm Bombyx mori.

INTRODUCTION

The silkworm Bombyx mori rearing is a traditional industry in Asia and the life of many people is depended on it. Increase of larval growth and cocoons quality and quantity would result better economics for this industry and meet the production needs. Consequently, the enrichment of mulberry leaves by supplementary compounds with the aim of increasing the production of cocoons is a very important aspect. Many investigations have been done on this topic and various reports have been published (Etebari, 2002; Etebari et al., 2004; Islam et al., 2004). Fortification of mulberry leaves with complementary compounds was found to increase the larval growth and post cocoon characteristics (Etebari, 2002; Etebari and Fazilati, 2003). Mulberry leaf is the sole food and source of nutrition for the silkworm, Bombyx mori. L. due to the presence of morin (Tribhuwan, et al., 1989). The growth and development of larva and subsequent cocoon production are greatly influenced by nutritional quality of mulberry leaves. Supplement in silkworm nutrition like protein substitute fortified with food stuff are needed for nutritional requirement among several insects (House, 1996). In recent years, attempts have been made in sericulture with nutrients such as proteins, carbohydrates, amino acids, vitamins, sterols, hormones, antibiotics etc. for better performance and get higher yield, quantity and quality cocoons (Sannappa, 2002). Various researches have been carried out on the dietary supplementation of mulberry leaves fed to silkworms. These supplementations include vitamins such as ascorbic acid, thiamine, niacin, folic acid and multivitamins (Etebari et al., 2004). Its nutrients are very easy to digest protein (biliprotein), carbohydrates (mucopolysaccharides, rhamnose and glycogen), 50 different minerals and trace minerals, beta-carotene, chlorophyll, GLA omega3 fatty acid, and many other nutrients. The presence of vitamins is appropriate for growth of larvae and the reproduction in many insects (Ishii, 1971; Yazgan,1972; Baker, 1975; Ritter and Johnson, 1991; Ozalp and Emre, 1992; Chang and Li, 2004). The elimination of niacin (nicotinic acid) from Ceratitis capitata diet causes the increased mortality of larvae and decrease in the proportion of pupal to adult emergence. Deficiency of vitamin B3 in the diet structure does not improve by adding tryptophan, but nicotinamide, NAD and NADP are proper substitutes for resumption of normal larval growth. The dietary supplements like protein, vitamins, lipids etc. evincing their specificity at specific dose for various metabolic activities of silkworm (Horie, 1980). Amino acid such as aspartic acid and glutamic acid are considered to be essential for silkworm growth (Ito and Inokuchi, 1981). Nutritional study on silkworm is an essential prerequisite for its proper commercial exploitation. Nutrition of silkworm is sole factor which almost individually augment quality and quantity of silk (Laskar and Datta, 2000). The current investigation highlights the effect of different concentrations of spinacia oleracea on the morphometric parameters viz. larval length, width and weight of the silk worm Bombyx mori.
MATERIALS AND METHODS

Rearing

The present study was conducted in the year 2010 at the experimental lab of Zoology Department, Annamalai University, Annamalai Nagar. The first day of III instar of popular Indian Bivoltine hybrid (CSRxCSCR) were collected from farmers training centre at Jayankondapattinam, Tamilnadu, India. The larvae were placed at ambient temperature of 25± 2°C and relative humidity of 70 to 80%. Further, these larvae were divided into 5 experimental groups including control (distilled water control), each group consisting of 10 larvae. The larvae were reared in card board boxes measuring 22×15×5cm³ covered with polythene sheet and placed in an iron stand with ant wells. The larvae were subjected to the following treatments. Spinacia oleracea was purchased from local market in Chidambaram, Tamilnadu, India, identified and authenticated from the department of Botany, Annamalai University. Shade dried and powdered using mortar and pestle. Finely powdered Spinacia oleracea was diluted in distilled water to 25%, 50%, 75% and 100% concentrations. Fresh mulberry leaves were soaked in each concentration and then dried in air for 10 minutes. The supplementary leaves were fed to silkworms, five feedings / day. Group I larvae received mulberry leaves sprayed with distilled water and served as control, group 2 larvae received 25% of Spinacia oleracea treated mulberry leaves, group 3 larvae received 50% Spinacia oleracea treated mulberry leaves, group 4 larvae received 75% Spinacia oleracea treated mulberry leaves, group 5 larvae received 100% Spinacia oleracea treated mulberry leaves, respectively and they were maintained up to cocoon. III, IV and V instar larval length, width and weight were determined for all groups.

Mulberry (M. alba) MR2 leaves treated with spinach oleracea

It was diluted to 25%, 50%, 75% and 100% (without dilution) concentrations. Fresh mulberry leaves were soaked in each concentration for 15 minutes and then were dried in air for 10 minutes. The treated leaves were used for feeding the III, IV and V instar larvae of silkworm B. mori.

Statistical analysis

Data were analyzed by one way analysis of variance (ANOVA) followed by Duncan’s multiple range test (DMRT) using a commercially available statistics software package (SPSS® for Windows, V. 16.0, Chicago, USA). Results were presented as means ± SD. P values <0.05 were regarded as statistically significant.

RESULTS AND DISCUSSION

The results of the present work, the effect of supplement Spinacia oleracea with MR2 mulberry leaves on biological parameters such as nutritional and commercial parameters in LBN, D2 (Local Bivoltine) are summarized in Table 1, 2, and 3 shows that the Morphometric data of length, width and weight of larval parameters of B. mori. Table 1 shows the mean length, width and weight of III instar larvae of group 1 were (1.6828±0.14682cm, 0.3490±0.05481 cm and 0.1115±0.00742gm), respectively. The mean length, width and weight of III instar larvae of group 2 were (1.9331±0.16239cm, 0.3828±0.04079cm, 0.1228±0.01029gm), respectively. The mean length, width and weight of III instar larvae of group 3 were (1.8490±0.10398cm, 0.3490±0.05398cm, 0.1096±0.00794gm), respectively. The mean length, width and weight of III instar larvae of group 4 were (1.8821±0.14270cm, 0.3495±0.05472cm, 0.1148±0.01047gm), respectively. The mean length, width and weight of III instar larvae of group 5 were (1.8678±0.13562cm, 0.3662±0.05064cm, 0.1128±0.00776gm), respectively. In these five observations, 25% Spinacia oleracea treated III instar larvae length, width and weight were significantly increased than the other four groups (1,3,4 and 5).

Table 2 shows that the Morphometric data of length, width and weight of larvae of B. mori fed with control MR2 leaves and Spinacia oleracea treated MR2 leaves in IV instar larvae of B. mori. The mean length, weight and weight of IV instar larvae of group 1 were (5.7232±0.23122cm, 0.5331±0.08065cm, 0.4231±0.03440gm), respectively. The mean length, width and weight of IV instar larvae of group 2 were (6.0158±0.14680cm, 0.6152±0.07128cm, 0.5348±0.03936gm), respectively. The mean length, width and weight of IV instar larvae of group 3 were (5.440±0.18699cm, 0.6000±0.8741cm, 0.4340±0.03814gm), respectively. The mean length, width and weight of IV instar larvae of group 4 were (5.4300±0.20689cm, 0.6490a±0.05271cm, 0.4148±0.02637gm), respectively. The mean length, width and weight of IV instar larvae of group 5 were (5.3900±0.20896cm, 0.6490±0.05398cm, 0.4148±0.02398gm), respectively. In these five observations, 25% Spinacia oleracea treated IV instar larval length, width and weight were significantly increased than the other four groups (1,3,4 and 5). The mean length, width and weight of V instar larvae of group 1 were (6.7067±0.24813 cm, 1.0310±1.2108cm, 2.8320± 0.84080gm), respectively. The mean length, width and weight of V instar larvae of group 2 were (7.2490±0.18694 cm, 1.1328±0.07675 cm, and 3.5578± 0.23675gm), respectively. The mean length, width and weight of V instar larvae of group 3 were (7.000±0.17789 cm, 1.0000±0.13162cm, 3.0948±0.59698gm) respectively.

Table 1 Morphometric data (biological traits) of III instar larvae of Bombyx mori fed with control and different concentrations of Spinacia oleracea treated MR2 mulberry leaves

<table>
<thead>
<tr>
<th>Experimental Group/Concentration</th>
<th>Larvae length(cm)</th>
<th>Larvae width(cm)</th>
<th>Larvae weight(gm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>1.6828±0.14682</td>
<td>0.3490±0.05481</td>
<td>0.1115±0.00742</td>
</tr>
<tr>
<td>MR2+25% Spinach</td>
<td>1.9331±0.16239</td>
<td>0.3828±0.04079</td>
<td>0.1228±0.01029</td>
</tr>
<tr>
<td>MR2+50% Spinach</td>
<td>1.8490±0.10398</td>
<td>0.3490±0.05472</td>
<td>0.1096±0.00794</td>
</tr>
<tr>
<td>MR2+75% Spinach</td>
<td>1.8821±0.14270</td>
<td>0.3495±0.05472</td>
<td>0.1148±0.01047</td>
</tr>
<tr>
<td>MR2+100% Spinach</td>
<td>1.8678±0.13562</td>
<td>0.3661±0.05064</td>
<td>0.1128±0.00776</td>
</tr>
</tbody>
</table>

Values are mean ± standard deviation of ten observations. Values in the same column with different superscript letters (a, b, & c) differs significantly at p < 0.05 (DMRT)
Table 2 Morphometric data (biological traits) of IV instar larvae of *Bombyx mori* fed with control and different concentrations of *Spinacia oleracea* treated MR2 mulberry leaves

<table>
<thead>
<tr>
<th>Experimental Group/Concentration</th>
<th>Larvae length (cm)</th>
<th>Larvae width (cm)</th>
<th>Larvae weight (gm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>5.723±0.2312</td>
<td>0.533±0.0865</td>
<td>0.423±0.03440</td>
</tr>
<tr>
<td>MR 2+25% Spinach</td>
<td>6.015±0.14680</td>
<td>0.615±0.0712</td>
<td>0.534±0.03903</td>
</tr>
<tr>
<td>MR 2+50% Spinach</td>
<td>5.440±0.18698</td>
<td>0.600±0.18741</td>
<td>0.434±0.03814</td>
</tr>
<tr>
<td>MR 2+75% Spinach</td>
<td>5.430±0.20689</td>
<td>0.649±0.05271</td>
<td>0.414±0.02637</td>
</tr>
<tr>
<td>MR 2+100% Spinach</td>
<td>5.390±0.20896</td>
<td>0.649±0.05398</td>
<td>0.414±0.02398</td>
</tr>
</tbody>
</table>

Values are mean ± standard deviation of ten observations. Values in the same column with different superscript letters (a, b, & c) differs significantly at p < 0.05 (DMRT).

The mean length, width and weight of V instar larvae of group 4 were (6.8931±146990cm, 1.0000±0.6235cm, 3.2248±0.35219gm) respectively. The mean length, width and weight of V instar larvae of group 5 were (6.7497±38242cm, 0.9567±0.13562cm, 2.4538±0.35787gm) respectively. In these five observations, 25% *Spinacia oleracea* treated V instar larvae length, width and weight were significantly increased than the other four groups (1,3,4 and 5).

Similarly, the growth of silk worm larvae improved significantly upon feeding them with mulberry leaves supplemented with different nutrients (Sarker, 1993). Subburathanam and Krishnan,(1998) have noticed soya bean meal to accelerate the larval growth significantly at certain level when supplemented along with food to the silkworm *B. mori*. In the present study, the treatment of *Spinacia oleracea* at the concentration of 25% may have beneficial effects on the growth of the silkworm larval and pupal length, width and weight and also increased the quantity of silk production by enhancing the feed efficacy than control. So, this supplementation could be prescribed to the farmers to get more quantity of silk.

**DISCUSSION**

The overall observations from the present study have reported that the 25 % concentration of *Spinacia oleracea* treated MR2 mulberry leaves with the silk worm *B. mori* shows significantly increased morphometric parameters such as larval length, width and weight. Nutrition plays an important role in improving the growth and development of *B. mori* (Kanafi et al., 2007). Since most of this multi-vitamin compound is composed of ascorbic acid, it could be thought that the increase of larval weight is due to an enhancement of feeding activity in treated larvae although the vitamins as cofactors can facilitate the metabolic pathway. Similar findings have also been observed in the present study that *Spinacia oleracea* act as vitamin to stimulate the feeding activity in the silkworms. Therefore, *Spinacia oleracea* can improve the food digestibility and increase the larvae length, width and weight. This work is corroborate with Nirwani and Kalilw, (1996), suggested that this enhancement in larval and cocoon length, width and weight related to phagostimulation of folic acid. Several authors also have reported these effects about ascorbic acid (Dobzhenshon, 1974; Ito, 1978; Singh and Reddy, 1981; Kl-Karkasy and Idriss, 1990). In this study, cocoon parameters changed in different treatments. Previously, it was reported that enrichment of mulberry leaves by some vitamins could increase the cocoon yield. Balasundaram et al.,(2008) have reported that the larval length,width and weight have been improved when the larvae fed with the supplementation of vitamin C treated mulberry leaves. Nirwani and Kalilw, (1996) have determined that folic acid causes a significant increase in economical parameters such as female and male cocoon weight. Evangelista et al., (1997) have also reported that the larval and cocoon length, width and weight increase under multi-vitamin treatment.

**Table 3 Morphometric data (biological traits) of V instar larvae of *Bombyx mori* fed with control and different concentrations of *Spinacia oleracea* treated MR2 mulberry leaves**

<table>
<thead>
<tr>
<th>Experimental Group/Concentration</th>
<th>Larvae length (cm)</th>
<th>Larvae width (cm)</th>
<th>Larvae weight (gm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>6.7067±0.24813</td>
<td>1.0310±1.210848</td>
<td>2.8320±0.8480</td>
</tr>
<tr>
<td>MR 2+25% Spinach</td>
<td>7.2490±0.18694</td>
<td>1.1328±0.07675</td>
<td>3.5578±0.23687</td>
</tr>
<tr>
<td>MR 2+50% Spinach</td>
<td>7.0000±1.77898</td>
<td>1.0000±0.13162</td>
<td>3.0948±0.56968</td>
</tr>
<tr>
<td>MR 2+75% Spinach</td>
<td>6.8931±1.46999</td>
<td>1.0000±0.62355</td>
<td>3.2248±0.35219</td>
</tr>
<tr>
<td>MR 2+100% Spinach</td>
<td>6.7497±38242</td>
<td>0.9567±0.13562</td>
<td>2.4538±0.35787</td>
</tr>
</tbody>
</table>

Values are mean ± standard deviation of ten observations. Values in the same column with different superscript letters (a, b, & c) differs significantly at p < 0.05 (DMRT).

**REFERENCE**


