

RESEARCH ARTICLE

Impact of newer insecticidal residue on mulberry silkworm, *Bombyx mori* L.

Vasanth V¹, K Senguttuvan^{1*}, P Priyadharsini¹, Ponnuvel K M², Ranjith Kumar S³, Arasakumar E⁴, Swathiga G⁵ & Prety Rekha Narzary¹

¹Department of Sericulture, Forest College and Research Institute, Tamil Nadu Agricultural University, Mettupalayam 641 301, Tamil Nadu, India

²National Silkworm Seed Organisation, Central Silk Board, Bengaluru 560 068, Karnataka, India.

³Regional Sericultural Research Station, Central Silk Board, Koraput 764 021, Odisha, India.

⁴Regional Sericultural Research Station, Central Silk Board, Sahaspur 248 197, Uttarkhand, India.

⁵Department of Silviculture and Natural Resource Management, Forest College and Research Institute, Tamil Nadu Agricultural University, Mettupalayam 641 301, Tamil Nadu, India

*Email: senguttuvan.k@tnau.ac.in

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Abstract

The present study evaluated the impact of three insecticides, Chlorantraniliprole 18.5% SC at a dosage of 0.02%, Imidacloprid 17.80% SL at 0.05% and Fipronil 5% SC at 0.2%, on silkworm development, cocoon production and silk quality. Cross-breed silkworms (PM x CSR2) were reared on insecticide-treated mulberry leaves and parameters such as larval mortality, instar duration, cocoon yield and silk denier were assessed. The results indicated that all insecticides reduced disease incidence and boosted cocoon yield but adversely affected silk quality. Chlorantraniliprole exhibited a slow yet steady impact, reducing the third instar duration from 88.7 hr at 10 days after spraying (DAS) to 83.54 hr at 25 DAS. On the other hand, Imidacloprid showed a faster effect, decreasing the third instar duration from 84.57 hr at 5 DAS to 72.45 hr at 25 DAS. Fipronil accelerated development, with the shortest instar duration among all treatments. Despite the increase in cocoon yield, the control groups (water spray and untreated) outperformed the insecticide-treated groups in silk quality, with the highest cocoon yield (150.98 g/100 cocoons) and silk denier (2.5) observed in the untreated group. The findings emphasized that although insecticides improve productivity and pest control, natural or minimally treated conditions favour high-quality silk production, advocating for a balanced approach to pest management in sericulture.

Keywords

Bombyx mori; economic traits; insecticides; mulberry; silkworm larvae

Introduction

Sericulture is essential to India's rural economy, as the country ranks second only to China in terms of silk production. The mulberry leaf contributes greatly, accounting for 38.2% of effective sericulture. Mulberry silkworms feed exclusively on mulberry leaves, ensuring quantitative and qualitative aspects of foliar feeding that are crucial for high-quality cocoons production (1). However, the production of high-quality leaves is affected by a number of factors, including diseases (24%), pests (8%), weeds (7%) and other factors (51%). Mulberry, the sole food source of the silk moth *Bombyx mori*, is susceptible to various pests that hamper plant growth and reduce yield (2).

Mulberry plants are subject to the presence of approximately 300 species of pests, both insect and non-insect, of which more than 100 have been recorded in India (3). Among these pests, certain sap suckers and defoliators can cause significant damage, resulting in production losses of 12-25%. This loss can be attributed to factors such as reduced nutritional value or large-scale leaf fall due to pests (4). The silk moth, *Bombyx mori* is a commercially important insect species that is used in sericulture for silk production. However, the silk industry faces significant challenges as resistance to various synthetic insecticides develops in silkworm populations. The insecticides used to control these pests often leave residues on mulberry leaves, which can harm delicate silkworms. To prevent this, it is important to maintain a safe waiting period before harvesting leaves (5). Field studies conducted in India have shown that feeding silkworm insecticide-treated leaves results in decreased cocoon yield (6). Pesticide residues on mulberry leaves negatively affect the growth and economic properties of silkworm cocoons. The silk moth, *Bombyx mori*, is particularly sensitive to new insecticides, such as chlorantraniliprole (Coragen), which belongs to the class of anthranilic diamides, as well as the neonicotinoid imidacloprid (Confidor) and phenylpyrazole fipronil (IXUS), which are targeted against butterflies. Although anthranilic acid diamides are not commonly used in mulberry cultivation, they can still cause larval death and reduce cocoon production in nearby sericulture farms (7). Given the potential risks, it is important to investigate the toxic effects of these insecticides on *B. mori* performance, which motivated this study.

Materials and Methods

Bioassay on silkworm

The cross-breed silkworms (Kolar Gold) were obtained from the Sanjay Chawki Rearing Center in Gobichettipalayam and reared using G4 mulberry leaves. The larvae were fed mulberry leaves in required quantities. After an initial 30 minutes of feeding, 30 larvae were transferred to each experimental tray, with four replications and provided with mulberry leaves sprayed with different pesticides. The treatments included Chlorantraniliprole 18.5% SC at a dosage of 0.02% (T1), Imidacloprid 17.80% SL at 0.05%, sprayed at intervals of 5, 10, 15, 20 and 25 days, standard check treatment of Fipronil 5% SC at 0.2% (T3) sprayed at 5 days intervals, control with only water spray (T4) and untreated control leaves (T5). The silkworms were fed from the third instar onward. During the rearing process, the following observations were obtained.

Rearing parameters

The total larval durations and moulting durations (hr) was recorded by summing the durations of the third, fourth and fifth instars. Mature larval weight (g) was determined by weighing individually the 10 randomly chosen silkworms from the fifth day of the fifth instar in each replication and their average weight was calculated. Disease incidence observed by visual observation. Cocoon

weight (g) is determined by calculating the mean weight from the individual weight of 10 randomly picked cocoons on the fifth day of mounting (7).

Cocoon parameters

Cocoon yield (kg) was calculated based on the yield per 100 worms reared in each replication. Shell weight (g) was measured by cutting open 10 randomly selected cocoons and the average was calculated to determine the mean shell weight. The shell ratio (%) was calculated using the formula: (shell weight / whole cocoon weight) × 100. For silk filament length (m), five cocoons per replication were reeled using an Euprovette and filament length was recorded. The formula used to calculate the length was $L = R \times 1.125$, where L is the filament length and R is the reading from the reeling device. Fisher's method of analysis of variance was used to statistically analyze the data for the significance test. 5 % was the F-test's level of significance. Critical difference (CD) values were used to understand the data (8).

Results and Discussion

Insecticidal residue in mulberry on silkworm larval parameters

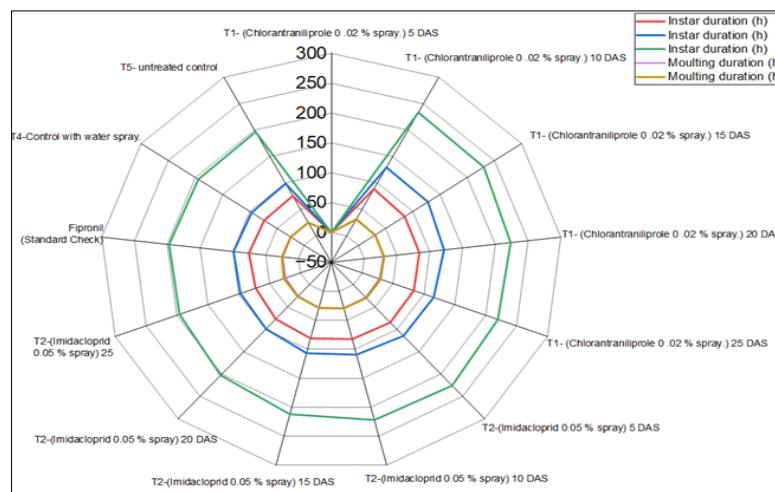
The impact of various insecticide treatments on the instar and moulting durations of *B. mori* demonstrated distinct developmental patterns across treatments, with each exerting a unique influence. T1 initially exhibited no discernible impact at 5 DAS, from 10 to 25 DAS, both the instar and moulting durations steadily decreased. The third instar duration declined from 88.7 hr at 10 DAS to 83.54 hr at 25 DAS, signifying a delayed yet consistent physiological effect. Previous studies have similarly noted the gradual action of Chlorantraniliprole on silkworms, leading to eventual developmental disruptions (8, 9). T2 showed a more rapid reduction in developmental duration compared to T1. The third instar of T2 duration decreased from 84.57 hr at 5 DAS to 72.45 hr at 25 DAS, with a significant decrease in moulting time, reflecting Imidacloprid's neurotoxic effects on the pest's development. This insecticide's efficiency in disrupting the pest's lifecycle has been documented in prior studies, reinforcing its role in pest management (10,11). T3, standard check, resulted in shorter instar and moulting durations than T1 and T2, with the third instar lasting 75.67 hr and moulting durations recorded at 24.5 and 25.68 hr for the 3rd and 4th moults, respectively. Fipronil's efficacy in accelerating developmental stages is well-documented and these results further validate its application as a standard treatment in silkworm rearing (12). Interestingly, the control groups T4 and T5 exhibited the fastest development rates, with T4 having the shortest instar durations (73.45 hr for the third instar and 96.54 hr for the fourth instar). Among the untreated groups, T5, showed slightly longer moulting times than T4, which may be attributed to natural variability in development, but overall trends indicate faster development in untreated conditions (13, 14). The results are shown in Table 1 and Fig. 1.

Table 1. Effects of insecticidal residue in mulberry on silkworm larval parameters

Treatments	Instar duration (h)			Moult duration (h)	
	3 rd instar	4 th instar	5 th instar	3 rd moult	4 th moult
T1- (Chlorantraniliprole 0.02 % spray.)					
5 DAS	0	0	0	0	0
10 DAS	88.7	129.6	233.53	31.23	31.65
15 DAS	85.08	127.89	230.54	30.89	30.23
20 DAS	84.23	121.98	223.54	29.8	30.7
25 DAS	83.54	115.67	219.78	28.3	29.97
T2-(Imidacloprid 0.05 % spray)					
5 DAS	84.57	114.76	225.89	28.9	29.98
10 DAS	82.5	109.25	221.98	29	29.56
15 DAS	81.23	106.78	211.98	28.95	28.56
20 DAS	76.98	99	202.78	25.67	26.78
25 DAS	72.45	97	195.67	24.7	27.11
T3-Fipronil (Standard check)	75.67	99.23	197.34	24.5	25.68
T4-Control with Distilled water spray.	73.45	96.54	194.56	24.32	24.98
T5- untreated control	75.67	98.75	196.73	24.98	25.23
SE(d)	6.09	8.68	16.18	2.14	2.18
CD (0.05)	12.79**	18.23**	33.98**	4.50**	4.58**

**Highly significant.

h-hours

SE (Standard Error) represents variability in the dataset.**CD (Critical Difference)** indicates significant treatment effects at a 5% probability level ($p \leq 0.05$).**Fig. 1.** Effects of insecticidal residue on larval parameters.

Insecticidal residue in mulberry on silkworm economic traits

In terms of production, the treatments showed significant differences in disease incidence (%), cocoon yield (g/100 cocoons), shell weight (g) and shell ratio (%). T1 demonstrated a steady reduction in disease incidence from 7.57% at 10 DAS to 5.87% at 25 DAS, accompanied by an increase in cocoon yield from 22.85 g/100 cocoons to 69.85 g/100 cocoons, increase in shell weight from 4.11 g to 19.56 g, gradual increase in shell ratio from 18% to 28% and an improvement in silk denier from 0.7 to 1.54.

These positive trends suggested that Chlorantraniliprole enhances productivity and silk quality over time while minimizing disease incidence (8). T2 was more effective, compared to T1, with a rise in cocoon yield from 34.57 g/100 cocoons to 139.98 g/100 cocoons, increase in shell weight from 7.26 g to 43.39 g, gradual increase in shell ratio from 21% to 31% and drop in disease incidence from 6.78% at 10 DAS to 5.02% at 30 DAS. The silk denier also improved from 1.98 to 2.26, indicating better silk quality. These findings align with the documented efficiency of Imidacloprid in reducing pest burdens and enhancing crop health (15, 16). T3 also demonstrated robust results,

with a cocoon yield of 142 g/100 cocoons, shell weight of 69.58 g, shell ratio of 49%, disease incidence of 6.5% and a silk denier of 2.33. These outcomes are consistent with Fipronil's established role in promoting productivity and silk quality in silkworm farming (12). Remarkably, the control groups, T4 and T5, outperformed the insecticide-treated groups in cocoon yield and silk quality. T4 achieved the highest cocoon yield at 150.98 g/100 cocoons, shell weight of 78.51g, shell ratio of 52% and the best silk quality with a denier of 2.45. Similarly, T5 recorded a cocoon yield of 147.89 g/100 cocoons, a shell weight of 79.86 g, a shell ratio of 54% and the highest silk denier of 2.5, indicating superior silk quality. These findings suggest that natural rearing conditions, devoid of chemical interference, can yield higher silk quality, though developmental rates may be slower compared to insecticide-treated groups (17, 18). The current results support the negative effects of insecticides on silkworm physiology and productivity, which is consistent with previous research (19, 20). This highlights the need for careful evaluation and control of pesticide applications in sericulture (21, 22). Furthermore, concerns raised in previous research regarding the ecological impacts of pesticide use on non-target organisms such as silkworms and are supported by the observed toxicity levels (23-25).

The results of this study not only expand our knowledge of the toxicological effects of insecticides on silkworms but also serve as an important reminder of the need for sustainable pest management techniques to protect the health of silkworms and the general ecosystem. The results are shown in Table. 2 and Fig. 2.

Conclusion

This study investigated the effects of pesticide treatments on the development, productivity and silk quality of *Bombyx mori*. 25 after spraying of chlorantraniliprole and imidacloprid, a significant reduction in disease incidence was observed, accompanied by increased cocoon output and higher silk denier. Fipronil acts as a standard check. However, the untreated groups produced the highest quality silk. The study suggests that while pesticides effectively manage pests and enhance yield, untreated or minimally treated conditions are more favorable for superior silk quality. These findings underscore the need for sericulture strategies that balance pest management with the production of high-quality silk.

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Authors' contribution

VV involved in conducting experiments related to the rearing efficiency of silkworms and data collection. SK was responsible for overseeing the experimental design and coordinating research activities. PP contributed to the analysis of data and interpretation of the ecotoxicological effects of insecticide. PKM did statistical analysis and preparation of graphical representations for the study. RKS focused on the silkworm physiology aspects, providing expertise in *Bombyx mori* biology. AE contributed to the manuscript writing. SG did critical revision of the paper and PRN gave a final approval for publication.

Table 2. Effects of insecticidal residue in mulberry on silkworm economic traits

Treatments	Disease incidence (%)	Cocoon yield (g/100 cocoon)	Shell Weight (g)	Shell Ratio (%)	Denier
T1- (Chlorantraniliprole 0.02% spray.)					
5 DAS	0	0	0	0	0
10 DAS	7.57	22.85	4.11	18	0.7
15 DAS	6.98	36.78	7.72	21	0.85
20 DAS	6	48.96	10.77	22	1.23
25 DAS	5.87	69.85	19.56	28	1.54
T2-(Imidacloprid 0.05 % spray)					
10 DAS	6.78	34.57	7.26	21	1.98
15 DAS	6.78	46.78	11.70	25	1.99
20 DAS	6.12	71.23	18.52	26	2.08
25 DAS	5.08	136.78	41.03	30	2.23
30 DAS	5.02	139.98	43.39	31	2.26
T3-Fipronil (Standard check)	6.5	142	69.58	49	2.33
T4-Control with water spray.	6.09	150.98	78.51	52	2.45
T5- untreated control	5.97	147.89	79.86	54	2.5
SE(d)	0.50	15.21	8.10	4.55	0.21
CD (0.05)	1.05**	31.93**	17.00**	9.56**	0.45**

**Highly significant.

g-grams, %- percentage

SE (Standard Error) represents variability in the dataset.

CD (Critical Difference) indicates significant treatment effects at a 5% probability level ($p \leq 0.05$).

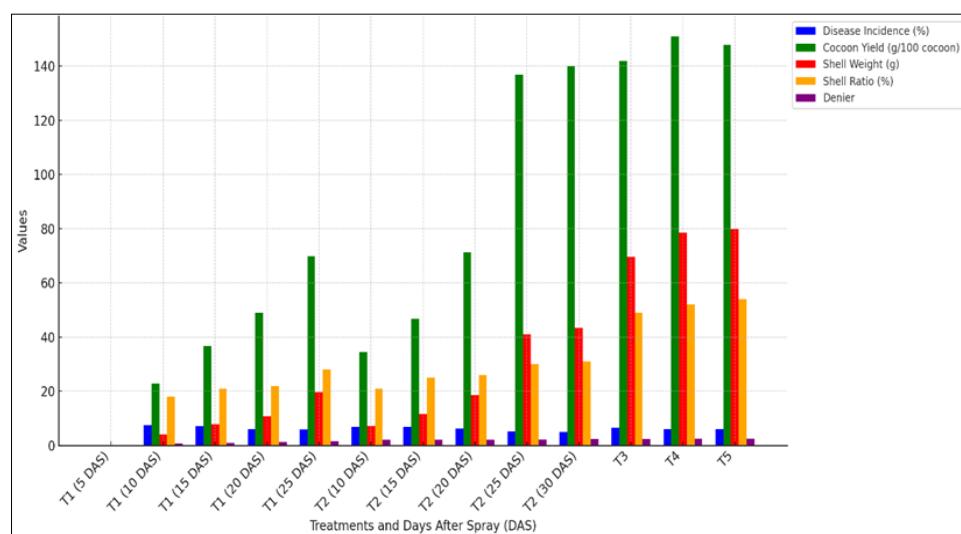


Fig. 2. Impact on insecticidal residue in silkworm economic parameters.

Compliance with ethical standards

Conflict of interest: The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Ethical issues: None

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