Effect of ~~feeding~~ abamectin 1.9 % EC sprayed mulberry leaves on growth and survival of parental breeds of silkworm, *Bombyx mori* L.

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Abstract

~~Chemical toxicity that could be through the food,~~ mulberry leaf makes the sole food for silkworm, while mulberry is highly prone for infestation by sap feeders majorly being thrips and mites, it is therefore important to observe the suitability of the chemicals used for managing these pests. An experiment to evaluate the suitable chemical, abamectin being a chemical with novel mode of action and an acaricide as well as insecticide was sprayed on mulberry foliage at 25 and 30 days after pruning (DAP) and fed to the silkworms at 15 and 20 days after spray on 3^{rd} instar onwards. There was no mortality of silkworm was observed in the treatment group during two rearing seasons. The larval duration was shortest (24.08 days), the larval weight (31.54 g/10 larvae), effective rate of rearing (ERR) (98.17 %) and cocoon weight (1.59 g) found significantly maximum in the treatment abamectin 1.9 % EC at 20 days after spray.

Keywords: Abamectin; mulberry; silkworm; parental breeds.

1. Introduction

Mulberry, an economically important tree, is being cultivated in China, India, Thailand, Brazil, Uzbekistan and other Countries across the globe, for its foliage to feed the monophagous silkworm, *Bombyx mori* L. The sustainability of silk industry is therefore directly correlated with the production and continuous supply of quality mulberry leaves (Sarkar et al., 2017). Which is influenced by a variety of factors *viz*., the variety, agronomic practices and both biotic and abiotic components (Krishnaswami et al., 1970). Despite adopting recommended agronomic practices, the nutritional quality of mulberry leaves often deteriorates due to various biotic factors, such as pests and diseases. Because mulberry plants are highly nutritious having lush green foliage for extended periods, they are attractive to numerous pests.

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Both insect and non-insect pests affect the growth and productivity of mulberry causing considerable yield loss and hence need effective management. Pesticides are the synthetic chemicals that interfere in the physiology of these pests and protect the host from their damage. The insecticides applied for the control of mulberry pests have a greater impact on silkworm, which is also an insect. Apart from managing the pest population, pesticides will leave residues on the host and exposure to such insecticide residues in the mulberry leaves could affect the growth and reproduction, economic characteristics of cocoons, moth eclosion and fecundity in *B. mori* (Bhosale & Kallapur, 1988). Hence, a safe concentration of suitable pesticide and post spray waiting period should be followed for the quality and sustained productivity of cocoon crop (Yokoyama, 1962).

The pesticide use pattern in the present-day situation has led to accumulation of residues in the harvestable produce, resistance build-up by pests and eradication of non-target organisms, which demands newer and safer pesticides with different modes of action. Abamectin, an avermectin derivative is one such molecule found effective against phytophagous mites and insect pests on several agricultural and horticultural crops worldwide (Lasota & Dybas, 1990). Avermectins bind to GABA (gamma aminobuytric) gated chloride channel and opens the channel for chloride influx. Abamectin is found to block the neurotransmitter, GABA (gamma aminobutyric acid) at the neuromuscular junction in insects and mites and bring about paralysis finally leading to death of the pests. In the light of the above, a study was conducted to know the effect of feeding abamectin 1.9 % EC @ 0.75 ml/l sprayed mulberry leaves on growth and survival of parental breeds of silkworm, *B. mori*.

2. Material and methods

The experiment was conducted during the year 2023-2024, at the Department of Sericulture, UAS, GKVK, Bengaluru with well-established mulberry garden of V1 variety. The performance of parental breeds namely PM, CSR2, FC1 and FC2 were reared to assess the impact of chemical used in mulberry for management of thrips and mites.

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The entire rearing room and appliances were disinfected by following standard procedure (Dandin & Giridhar, 2014). The rearing room was kept air tight for 24 hours and then the room was kept open and used for rearing. The chawki silkworms were reared on the leaves harvested from control plots while the third instar onwards the larvae were fed with mulberry leaves of treatment plots harvested at 15 and 20 DAS of the chemical post spray. A total of 150 larvae were transferred to each experimental tray in three replications after 30 minutes of initial feeding along with the mulberry leaves. In order to assess extent of toxicity of pesticide to silkworm and to determine the safe period of the chemical post spray. The impact of feeding chemical sprayed leaf on the larval growth and productivity such as larval duration (days), larval weight (g/10 larvae), larval mortality (%), ERR (%), cocoon weight (g) and defective cocoons (%) of the parental breeds was observed for recorded the data that were analysed using Factorial- CRD for testing of significance by Fisher's method of analysis of variance as outlined by (Sundararaj et al., 1972). The level of significance used in the F-test was P = 0.05. The critical difference (CD) values were computed to compare significance of the treatments.

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Table 1: Treatment details

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Treatments	Description
T$_1$	PM (abamectin 1.9 % EC @ 0.75 ml/l at 15 DAS)
T$_2$	CSR2 (abamectin 1.9 % EC @ 0.75 ml/l at 15 DAS)
T$_3$	FC1 (abamectin 1.9 % EC @ 0.75 ml/l g/l at 15 DAS)
T$_4$	FC2(abamectin 1.9 % EC @ 0.75 ml/l at 15 DAS)
T$_5$	PM (abamectin 1.9 % EC @ 0.75 ml/l at 20 DAS)
T$_6$	CSR2(abamectin 1.9 % EC @ 0.75 ml/l at 20 DAS)
T$_7$	FC1(abamectin 1.9 % EC @ 0.75 ml/l at 20 DAS)
T$_8$	FC2(abamectin 1.9 % EC @ 0.75 ml/l at 20 DAS)
T$_9$	PM (control)
T$_{10}$	CSR2 (control)
T$_{11}$	FC1 (control)
T$_{12}$	FC2 (control)

DAS: Days after spray; * No spray was used in the control treatment plots of mulberry for management of thrips and mites.

3. Results and Discussion:

Feeding silkworms with the chemical sprayed leaves was initiated from second feed of third instar onwards and the observation recorded on parameters are as follows was observed

3.1 Larval duration (Days)

A significant difference with respect to larval duration during third, fourth and fifth instars upon feeding silkworms with the leaves of abamectin 1.9 % EC sprayed mulberry plants harvested at 15 and 20th day post spray. Among the parental breeds, the bivoltine pure breed, CSR2 exhibited shortest larval duration (3.30, 4.43 and 7.72 days) and FC2 (3.27, 4.45 and 7.74 days, respectively) during 3rd, 4th and 5th instar, respectively. While it was longest in multivoltine pure breed PM (4.26, 5.29 and 8.34 days) during 3rd, 4th and 5th instar, respectively. Feeding on leaves harvested from the control plot resulted in the shortest larval durations during the 3rd, 4th and 5th instars (3.29, 4.40 and 7.63 days) followed by the safety period of 20 DAS (3.56,4.69 and 7.86 days, respectively during 3rd, 4th and 5th instar. Feeding silkworms on the leaves harvested from the treatment plot at 15 days after post spray prolonged the larval duration among all the breeds during different instars. Indicating a uniformity in the impact of chemical toxicity across the silkworm breeds as reflected by non-significant interaction (Table 2).

3.2 Total larval duration (Days)

The total larval duration was significantly shorter in bivoltine breed, CSR2 (23.56 days) and FC2 (23.47 days), while the longer larval duration was recorded by the multivoltine pure breed, PM (25.89 days). Irrespective of the parental breeds, the total larval duration extended significantly when the silkworms were fed with abamectin 1.9 % EC @ 0.75 ml/l sprayed mulberry leaves harvested at 15 DAS (25.06 days) compared to 20 DAS (24.21 days) and absolute control (23.33 days), respectively. The same trend was followed among all the breeds tested in the study at different durations safety periods that was clearly reflected in terms of non-significant interactions between the breeds and the spray schedule (Table 2).

The variation in the larval duration of different parental breeds are attributed to the breed character (Ashoka et al., 2016). However, the chemicals used for management of pests in mulberry may alter the length of larval duration. The high residual content of insecticide molecules in the initial days after spray might result in alternations in the normal physiological processes of silkworm and this residual toxicity may diminish over time with an increased safety period (Kumutha et al., 2013). The insecticides interfere with the release of hormones necessary for metamorphosis leading to a delay in instar duration (Tiwari et al., 2006) The altered physiology might result in extended larval duration (Singh et al., 2008). (Yeshika et al., 2019) reported a significant reduction in the fifth larval duration in the cross breed, PM ×CSR2 when the leaves harvested from dinetofuran 20 SG (@ 0.25 g/l) sprayed treatment plot at different durations after spray (8.23, 8.12, 8.05 and 7.97 days, respectively at 10, 20, 30 and 40 DAS). (Kalpana et al., 2022) recorded a significant reduction in the total larval duration when leaves harvested from abamectin 1.9 % EC sprayed plot (23.72 and 22.50 days, respectively at 15 and 20 DAS) (PM ×CSR2). The earlier studies conducted by (Kenchappa et al., 2024) also confirmed the present investigation with respect to total larval duration that recorded (22.30 and 23.22 days) when the silkworms were fed with the leaves harvested from abamectin 1.9 % EC sprayed treatment plot of mulberry at 15 and 20 DAS, respectively in the bivoltine double hybrid.

Table 2: Larval duration of parental breeds of silkworm, *B. mori* as influenced by feeding mulberry leaves treated with abamectin 1.9 % EC at different days after spray

Treatments	3rd instar (Days)	4th instar (Days)	5th instar (Days)	Total larval duration (Days)
Breeds (B)				
B_1: PM	4.26	5.29	8.34	25.89
B_2: CSR2	3.30	4.43	7.72	23.46
B_3: FC1	3.39	4.53	7.87	23.80
B_4: FC2	3.27	4.45	7.74	23.47
F-test	*	*	*	*
S.Em±	0.070	0.030	0.039	0.102
$CD_{0.05}$	0.204	0.087	0.114	0.300
Safety period (S)				
S_1: 15 DAS	3.81	4.98	8.26	25.06
S_2: 20 DAS	3.56	4.65	7.86	24.08
S_3: Control	3.29	4.40	7.63	23.33
F-test	*	*	*	*
S.Em±	0.061	0.026	0.034	0.089
$CD_{0.05}$	0.177	0.076	0.099	0.539
Interaction (B×S)				
B_1S_1	4.44	5.52	8.68	26.64
B_1S_2	4.28	5.31	8.27	25.87
B_1S_3	4.06	5.04	8.07	25.17
B_2S_1	3.58	4.70	8.08	24.37
B_2S_2	3.29	4.41	7.66	23.36
B_2S_3	3.03	4.18	7.41	22.63
B_3S_1	3.72	4.93	8.22	24.87
B_3S_2	3.40	4.47	7.82	23.70
B_3S_3	3.05	4.20	7.59	22.84
B_4S_1	3.52	4.76	8.10	24.38
B_4S_2	3.27	4.41	7.69	23.37
B_4S3	3.03	4.19	7.44	22.67
F-test	NS	NS	NS	NS
S.Em±	-	-	-	-
$CD_{0.05}$	-	-	-	-

*Significant at 0.05; NS: Non-significant; DAS- Days after spray; The mentioned values represent the average of two rearing.

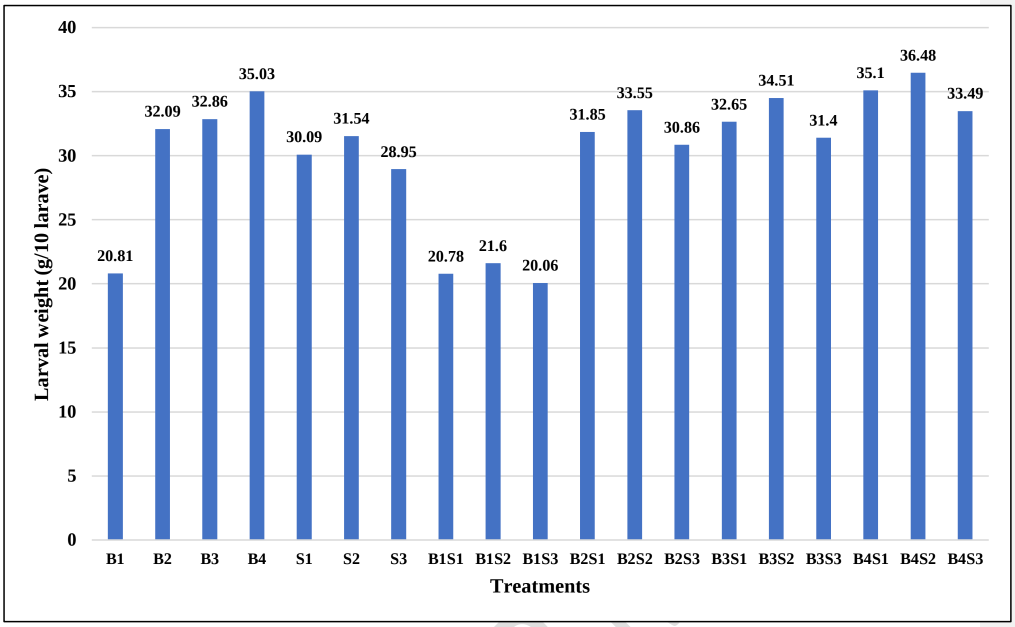

Fig. 2. Larval weight of parental breeds of silkworm, *B. mori* as influenced by feeding mulberry leaves treated with abamectin 1.9 % EC at different days after spray

B1: PM, **B2:** CSR2, **B3:** FC1, **B4:** FC2

S1: 15 DAS, **S2:** 20 DAS, **S3:** control

B1S1: PM (15 DAS), **B1S2:** PM (20 DAS), **B1S3:** PM (control)

B2S1: CSR2 (15 DAS), **B2S2:** CSR2 (20 DAS), **B2S3:** CSR2 (control)

B3S1: FC1 (15 DAS), **B3S2:** FC1 (20 DAS), **B3S3:** FC1 (control)

B4S1: FC2 (15 DAS), **B4S2:** FC2 (20 DAS), **B4S3:** FC2 (control)

DAS: Days after spray

The mentioned values represent the average of two rearing.

3.3 Larval weight (g/10 larvae)

Among parental breeds, the larvae weighed significantly heavier in the bivoltine hybrid FC2 (35.03 g/10 larvae) followed by FC1 (32.86 g/10 larvae). The multivoltine breed, PM exhibited the lowest larval weight of 20.81 g/10 larvae. Regardless of the parental breeds, the heavier larval weight was observed at 20 DAS (31.54 g/10 larvae) followed by 15 DAS (30.09 g/10 larvae), while it was least in absolute control group, where no spraying was taken (28.95 g/10 larvae). There was no significant difference in the interaction between parental breeds and duration of spray concerning the larval weight (Fig. 1).

Mites feeding on mulberry leaves deplete the primary nutrients from leaves and enhance the quantity of secondary chemicals (phenols, prolines) and the activity of peroxidases and phenylalanine ammonia lyase associated with production of secondary metabolites, adversely affecting the nutritional quality of leaves. Both these consequences of mite feeding might interfere with the leaf consumption by the silkworms leading to reduced larval weight in untreated control group (Dar et al., 2011). Abamectin 1.9 % EC when sprayed @ 0.75 ml/l was found effective against pests like mites and thrips thereby encouraging the growth and quality of mulberry plants and provide essential nutrients to the larvae, potentially leading to increased larval weight when sprayed leaves were fed to the worms at 15 and 20 DAS. These results are in accordance with the findings of (Muthuswami et al., 2010) who reported a significant improvement in larval weight and cocoon parameters upon feeding the cross-breed silkworms with the mulberry leaves treated with a neonicotinoid, thiamethoxam at 20 DAS. Molecules like dimethoate showed maximum larval weight if the leaves were used for rearing after specific safe period (Misra et al., 2003). The worms fed with abamectin 1.9 % EC @ 0.75 ml/l sprayed mulberry leaves at 15 and 20 DAS recorded significantly maximum larval weight in the silkworm hybrid, PM × CSR2 (24.46 and 26.44 g/10 larvae, respectively) as reported by (Kalpana et al., 2022 & Kenchappa et al., 2024) noticed that the maximum larval weight of 4.29 and 4.08 g in silkworm double hybrid, FC1× FC2 when the worms fed with abamectin 1.9 % EC sprayed mulberry leaves harvested at 15 and 20 DAS, respectively.

3.4 Larval mortality (%)

The molecule, abamectin 1.9 % EC was sprayed on the mulberry plants @ 0.75 ml/l and the leaves were fed to the worms at 15 and 20 days after spray. The treatment-wise larval mortality was recorded across the four parental breeds (PM, CSR2, FC1 and FC2). There was no report of larval mortality across the breeds in both the safety durations after spray of the chemical similar to the control treatment and also the interaction between breeds and the safety period, which is documented in the Table 3.

Silkworm, *B. mori* is a beneficial insect reared for cocoon production. Consequently, any chemical used to control pests in mulberry plants can influence the growth and productivity of silkworms. Toxic residues from insecticides may damage the integument, lead to a complete cessation of feeding, cause body stiffness resulting in 'S' or 'C' shaped larvae, induce vomiting, and hinder proper ecdysis (Jyothi et al., 2013). Therefore, a specific waiting period is recommended for the safe application of these chemicals (Yokoyoma, 1962). Insecticides like pyrethroid, lambda-cyhalothrin and macrocyclic lactone, emamectin benzoate are highly toxic to silkworms with 100 per cent mortality even at 30 DAS that might be due to decreased levels of inorganic elements in the haemolymph. However, the Pyrrole group of insecticide *viz.,* chlorfenapyr @ 1.5 ml/l recorded lesser percentage of mortality (12.09 %) in silkworm (PMxCSR2) at 10 DAS (Sunil Kumar & Naika, 2019). Further, (Satish et al., 2014) opined that chlorfenapyr is safer for chawki worms at lesser concentration that becomes toxic with an increase in the concentration. The present study is in accordance with (Kalpana et al., 2022 & Kenchappa et al., 2024) who reported the safety of abamectin 1.9 % EC (@ 0.75 ml/l) and diafenthiuron 50 % WP (@ 1 g/l) to the silkworms of both cross breed and bivoltine double hybrid when fed with the sprayed mulberry leaves at 15 and 20 DAS as evidenced by zero per cent mortality at third, fourth and fifth instars.

3.5 Effective Rate of Rearing (ERR %)

Several factors influence the effective rate of rearing in silkworm rearing that include optimal feeding practices, proper pest management and maintaining ideal environmental conditions. Sericulturists can enhance the health and productivity of silkworms by focusing on these parameters and achieve sustained productivity of superior quality cocoons and the silk.

Table 3: Larval mortality and effective rate of rearing (ERR %) of parental breeds of silkworm, *B. mori* as influenced by feeding mulberry leaves treated with abamectin 1.9 % EC at different days after spray

Treatments	Larval mortality (%)	ERR (%)
Breeds (B)		
B_1: PM	0.00	99.22
B_2: CSR2	0.00	95.77
B_3: FC1	0.00	97.56
B_4: FC2	0.00	97.22
F-test	NS	*
S.Em±	-	0.475
CD $_{0.05}$	-	1.394
Safety period (S)		
S_1: 15 DAS	0.00	95.91
S_2: 20 DAS	0.00	97.83
S_3: Control	0.00	98.58
F-test	NS	*
S.Em±	-	0.411
CD $_{0.05}$	-	1.207
Interaction (B×S)		
B_1S_1	0.00	98.33
B_1S_2	0.00	99.33
B_1S_3	0.00	100.00
B_2S_1	0.00	94.00
B_2S_2	0.00	96.33
B_2S_3	0.00	97.00
B_3S_1	0.00	95.66
B_3S_2	0.00	98.00
B_3S_3	0.00	99.00
B_4S_1	0.00	95.66
B_4S_2	0.00	97.66
B_4S3	0.00	98.33
F-test	NS	NS
S.Em±	-	-
CD $_{0.05}$		

*Significant at 0.05; NS: Non-significant; DAS- Days after spray; The mentioned values represent the average of two rearing.

The ERR showed a significant difference among the parental breeds of silkworm, *B. mori*. It was recorded maximum in the multivoltine pure breed, PM (99.22 %) followed by bivoltine hybrid FC1 (97.56 %) and the lowest ERR was observed in bivoltine pure breed, CSR2 (95.77 %). A significant influence of safety period was also observed on the ERR when the silkworms were fed with the mulberry leaves harvested at different durations after spray of the chemical, abamectin 1.9 % EC @ 0.75 ml/l. The maximum ERR was recorded in the absolute control group where no spraying was carried out (98.58 %) followed by 20 DAS (97.83 %). The minimum ERR was observed at 15 DAS (95.91 %). Similar trend was followed among all the breeds during different safety periods of various treatments indicated by non-significant interaction effects (Table 3).

An ERR of 97.77, 97.77 and 98.87 per cent was observed in silkworm hybrid (PM ×CSR2) when the larvae were fed with a neonicotinoid, dinotefuron 20 SG @ 0.25 g/l treated mulberry leaves at 10, 20 and 30 DAS, respectively (Yeshika et al., 2019). Propargite 57 EC when sprayed @ 1.5 ml/l was found effective in reducing mites population (*Polyphagotarsonemus latus*) in mulberry and resulting in an ERR of 96.66 per cent when the leaves were fed to the silkworms of multixbivoltine hybrid, PM ×CSR2 16 DAS (Sharath et al., 2022). Among the different chemicals used to control the thrips and mites in mulberry spraying abamectin 1.9 % EC @ 0.75ml/l and diafenthiuron 50% WP @ 1 g/l recorded significantly maximum ERR of 97.78, 97.78 % and 96.65, 96.65 %, respectively at 15 and 21 DAS, in the bivoltine double hybrid (Kenchappa et al., 2024) which is in accordance with the current study.

3.6 Cocoon weight (g)

A significant difference was noticed for cocoon weight amongst the parental breeds reared in the experiment. The highest cocoon weight was recorded in bivoltine hybrid, FC2 (1.73 g), while the lowest cocoon weight was observed in multivoltine breed, PM (1.24 g), which is the breed character. The duration of spray also found to have profound influence on cocoon weight when the silkworms were fed with abamectin 1.9 % EC @ 0.75 ml/l treated mulberry leaves harvested at different durations after spray. The highest cocoon weight was observed at 20 DAS (1.59 g), while the absolute control group showed least cocoon weight of 1.45 g. The interaction between parental breeds and timing of spray on cocoon weight showed no significant difference indicating the consistency of residual toxicity of the chemical across the parental breeds of *B. mori* silkworm with respect to cocoon weight (Table 4).

The silkworm breeds with different voltinism exhibit a noticeable variation in the cocoon weight, which is an inherent characteristic of the breed (Ashoka et al., 2016) and the same has been reflected in the present study. The multivoltine though less productive, are known to have high intensity of sturdiness and can withstand the adverse rearing conditions compared to bivoltine (Kumaresan et al., 2012). The same is true even in the present study where the residual toxicity of the chemical had less effect on multivoltine, PM than the bivoltine breeds. Further, minimal impact of longer safety period may be attributed to gradual degradation of the molecules after certain period after spray while reducing the pest population and subsequent increase in the quality and productivity of mulberry plant and the silkworm. Hence, following a safe waiting period strictly before feeding the silkworms with mulberry leaves from chemical sprayed gardens is imminent as observed in the study conducted by (Kariappa & Narasimhanna, 1978). in their study that documented a significant improvement in larval and cocoon weight upon feeding silkworms on mulberry leaves treated with dimethoate @ 0.2 %.

Table 4: Cocoon weight (g) and defective cocoons (%) of parental breeds of silkworm, *B. mori* as influenced by feeding mulberry leaves treated with abamectin 1.9 % EC at different days after spray

Treatments	Cocoon weight (g)	Defective cocoons (%)
Breeds (B)		
B_1: PM	1.24	1.34
B_2: CSR2	1.49	3.39
B_3: FC1	1.59	2.18
B_4: FC2	1.73	2.76
F-test	*	*
S.Em±	0.008	0.343
CD $_{0.05}$	0.024	1.008
Safety period (S)		
S_1: 15 DAS	1.49	4.02
S_2: 20 DAS	1.59	1.78
S_3: Control	1.45	1.45
F-test	*	*
S.Em±	0.007	0.297
CD $_{0.05}$	0.021	0.873
Interaction (B×S)		
B_1S_1	1.21	2.03
B_1S_2	1.32	1.34
B_1S_3	1.19	0.66
B_2S_1	1.46	6.03
B_2S_2	1.57	2.06
B_2S_3	1.43	2.06
B_3S_1	1.57	3.84
B_3S_2	1.67	1.36
B_3S_3	1.52	1.34
B_4S_1	1.71	4.18
B_4S_2	1.79	2.38
B_4S3	1.68	1.70
F-test	NS	NS
S.Em±	-	-
CD $_{0.05}$	-	-

*Significant at 0.05; NS: Non-significant; DAS- Days after spray; The mentioned values represent the average of two rearing.

3.7 Defective Cocoons (%)

Among the parental breeds used in the present experiment a significant difference was observed regarding percentage of defective cocoons which includes flimsy, double and thin shelled cocoons. The bivoltine breed, CSR2 shows highest percentage of defective cocoons (3.39 %) followed by bivoltine hybrid, FC2 (2.76 %), while PM had the lowest percentage of defective cocoons (1.34 %). The spray schedule found to significantly affect the defective cocooning percentage when the silkworms were fed with abamectin 1.9 % EC sprayed mulberry leaves. The highest percentage of defective cocoons was recorded in 15 DAS schedule (4.02 %), that was least in 20 DAS (1.78 %) on par with absolute control (1.45 %). The interaction between breeds and duration of the spray showed no significant difference concerned to percentage of defective cocoons, indicating that the effect of the spray duration on per cent good cocoon was consistent across all breeds (Table 4).

4. Conclusion:

Chemical sprays have become an integral part of pest management in the present day agriculture system. Silkworms are sensitive for chemicals so chain of experiments has been carried out to find the newer chemical that could be safer to silkworm. However, a safe usage of the chemical is utmost important for effective management of pests in cropping systems without compromising the economic product. The present investigations have indicated the safety of abamectin 1.9 % EC to the silkworm, *B. mori* when the molecule was employed for management of thrips and mites in mulberry, the sole food plant of the larva. The silkworms showed zero mortality, shortest larval duration, maximum larval weight and ERR, cocoon weight and least defective cocoon percentage when fed with abamectin 1.9 % EC sprayed mulberry leaves harvested after 20 days of post spray. The chemical take 20 days to degrade that its toxicity level had no influence on silkworm health and productivity.

DISCLAIMER (ARTIFICIAL INTELLIGENCE):

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of manuscripts.

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