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In-Vitro and *In-Vivo* Evaluation of *Aloe vera* Gel Extract on Inhibition Zone and Larval Parameters of Silkworm *B. Mori* L. Administered with *Bacillus* Sp.

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Abstract

The experiment on *Aloe vera* gel extract on larval parameters of silkworm *B. Mori* L. administered with *Bacillus* sp. was conducted during 2008-09. The results showed complete inhibition at 100 and 75% leaf extract. But partial and no inhibition zone was recorded at 50, 25% and control lot, respectively. Whereas in sterilized gel batches no inhibition zone was noticed at 25, 50, 75, 100% and control lots. *In-vivo* evaluation showed significant improvement. When silkworms were fed with mulberry leaves smeared with 100% *Aloe vera* gel extract recorded least disease incidence (9.20%) and maximum larval weight (3.39 g) at 10^{-3} bacterial spore dilution followed by 75, 50 and 25% concentrations compared to control.

Key words : *Aloe vera,* Gel extract concentrations, Disease incidence, Mulberry, *Bombyx mori* L.

Even though India ranks second in silk production, several factors have direct influence on the production of good quality cocoon crops. Mulberry alone contributes 38.2% towards the harvest of a successful cocoon crop, together with climatic factors which constitute 75.2%. Mulberry being only food of mulberry silkworm, its nutrient management should aim at fulfilling nutritional requirements of silkworm like carbohydrates, proteins, minerals and moisture content of mulberry leaves. *Aloe vera* gel is composed mainly of water (99%) and mono and polysaccharides (25% of the dry weight of the gel), it also contains lignin, salicylic acid, saponins, sterols and triterpenoids. The fresh gel contains the proteolytic enzyme carboxypeptidase (which breaks down bradykinin), glutathione peroxidase and several isozymes of superoxide dismutase (1, 2). The *Aloe vera* gel also contains vitamins A, C, E, B_{12},thiamine, niacin and folic acid, and minerals sodium, potassium, calcium, magnesium, manganese, copper, zinc, chromium and iron (3, 4). Anthraquinones, which are present in *Aloe vera* gel, have direct viricidal effects. The anthraquinone present in *Aloe vera* gel was shown to inactivate various enveloped viruses at low concentrations (5) and exhibit significant antiviral activity.

Methods

The required quantity of fesh leaves of *Aloe vera* were collected and surface sterilized with 70% ethyl alcohol and then washed with sterile distilled water and slit open longitudinally, the gel was scooped with sterile stainless steel knife and homogenized in a domestic mixi and filtered through a sterile stainless steel tea strainer. The extract was filtered through double layered musline cloth, and extract was maintained as stock solution (100% gel) from which different concentrations were made by using distilled water to arrive at 75, 50 and 25% concentrations. Different concentrations of *Aloe vera* gel extracts and the spore dilution of 10^{-1},10^{-2},10^{-3} dilutions of *Bacillus* sp., were smeared on mulberry leaves and air dried for 5—10 minutes, then fed to the silkworms ($PM \times CSR_2$) once a day during fourth instar first day. The remaining feeds were provided with normal mulberry leaf. The control lot was also maintained without any treatment. Hundred uniform worms were maintained in each treatment and each replication. The package of practices for silkworm

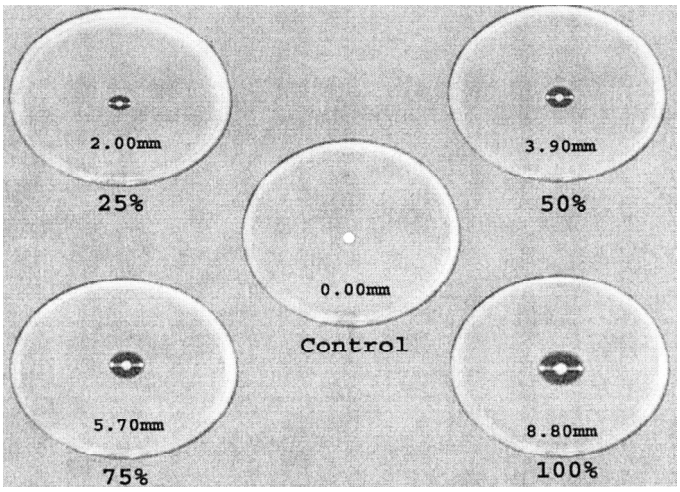

Figure 1. Effect of *Aloe vera* gel on inhibition zone of *Bacillus* sp.

rearing was carried out based recommendation (6).

Results and Discussion

Fifth Instar Larval Weight (g)

Silkworm fed with mulberry leaves smeared with *Aloe vera* extract at different bacterial spore dilutions showed a positive effect with respect to fifth instar larval weight. Significant difference was noticed in fifth instar larval weight, when larvae were fed with 100% *Aloe vera* gel extract smeared mulberry leaves which recorded higher (3.29g) larval weight followed by 75, 50 and 25% concentrations (3.21, 3.16 and 3.09 g) respectively. Least larval weight was observed in control (3.02 g).

Marked differences were observed among different bacterial spore dilutions with respect to fifth instar larval weight. Significantly higher (3.24g) larval weight was noticed when silkworms were fed with *Aloe vera* gel extract smeared mulberry leaves at 10^{-3} bacterial spore dilution followed by 10^{-2} (3.17 g) and lower larval weight at 10^{-1} spore dilution (3.06 g). The interaction effect of *Aloe vera* extract and bacterial spore dilutions had a significant effect on fifth instar larval weight. Higher larval weight was recorded, when silkworms were fed with mulberry leaves smeared with 100 per cent *Aloe vera* gel extract (3.39 g) followed by 75 per cent concentration (3.32 g) at 10^{-3} bacterial spore dilution. While low larval weight noticed in control (3.02 g) (Table 1).

The present results are supported by the findings of Manimegalai and Chandramohan (2005) who reported that, higher weight at higher concentrations of *Bacillus thuriengiensis* combined with *Thuja orientalis* at 10,000 ppm was found to be highly effective showing higher larval weight of 3.46g with higher dose of *T. orientalis i.e.,* 20,000 ppm extracted a negative influence and recorded lesser values of larval weight (2.95g). Manjunath (7) also confirmed that application of plant extracts to PM × CSR_2 worms coupled with 10^{-1}, 10^{-2}, 10^{-3} *Bacillus* spore suspension resulted in significant effect on third and fourth instar larval weight. The minimum and maximum larval weight of 1.19 and 1.24 g was noticed at 1 : 1 proportions of *Phyllanthus niruri* and *Adathoda vasica* compared to1: 3 (1.15 to 1.19 g). Further, Priyadharshini (8) reported that amla treated larvae recorded significantly higher larval weight (3.91 g) followed by boerhavia (3.70 g),asparagus (3.63g) and bael (3.52 g) against *Bacillus* sp., while control (2.60 g) recorded significantly lower larval weight.

Disease Incidence (%)

The disease incidence in PM × CSR_2 hybrid rearing showed significant differences due to administration of *Aloe vera* extract a different bacterial sore dilutions.

However, disease incidence was high in control (13.50%) whereas, least incidence of disease was

Table 1. *In-vivo* effect of *Aloe vera* (L.) N. Burman gel on fifth instar larval weight and disease incidence of *Bombyx mori* L. (PM × CSR_2) administered with *Bacillus* sp. * = Significant at 5%; NS = Non — significant.

Aloe vera gel / control	Larval weight (g) Bacterial spore dilution 10^{-1}	10^{-2}	10^{-3}	Mean	Disease incidence (%) Bacterial spore dilution 10^{-1}	10^{-2}	10^{-3}	Mean
T_1 25% *Aloe vera*	2.99	3.09	3.19	3.09	13.87	12.12	11.00	12.33
T_2 50% *Aloe vera*	3.04	3.16	3.28	3.16	11.87	11.37	10.25	11.16
T_3 75% *Aloe vera*	3.09	3.22	3.32	3.21	9.37	7.75	6.62	7.91
T_4 100% *Aloe vera*	3.16	3.33	3.39	3.29	6.62	5.25	4.62	5.50
T_5 Control	3.02	3.02	3.02	3.02	13.50	13.50	13.50	13.50
Mean	3.06	3.17	3.24	–	11.05	10.00	9.20	–

Test of significance	Larval weight (g) *F* test	SE ±	CD at 5%	Disease incidence (%) *F* test	SE ±	CD at 5%
Bacterial spore dilutions	*	0.011	0.032	*	0.648	1.846
Aloe vera gel	*	0.014	0.042	*	0.836	2.383
Interactions	*	0.025	0.073	NS	1.449	4.128

noticed in 100 per cent *Aloe vera* extract (5.50 %) followed by 75 per cent (7.91%) and 50 per cent (11.16%) when smeared on mulberry leaves and fed to silkworms.

Silkworm hybrid PM × CSR_2 fed with mulberry leaves smeared with *Aloe vera* extracts at three different bacterial spore dilutions of *Bacillus* sp. caused significant difference in disease incidence. The minimum incidence of disease was noticed at 10^{-3} dilution (9.20%) than that of 10^{-2} (10.00%) and 10^{-1} (11.05%) bacterial spore dilutions. Non-significant difference in disease incidence of silkworm was noticed in interaction of *Aloe vera* extract with bacterial spore dilutions (Table 2).

These results are comparable to the findings of Manimegalai and Chandramohan (9) who reported that, per cent mortality was least in *B. thuriengiensis* + *T. orientalis*, 15000 ppm (28.93%) however, it did not significantly differ with *B. thuriengiensis* + *T. orientalis,* 10000 ppm (30.03%) in case of 01-TAD-01 isolate. Further, Priyadharshini (2006) reported that, amla (16.6%) recorded significantly lower mortality followed by boerhavia (17.70%), asparagus (20.0%) and bael (22.2%) against *Bacillus* sp. The mortality recorded was significantly higher in control (26.86%).

Manjunath (7) reported that larval mortality of PM × CSR_2 was greatly influenced by the application of different medicinal plant extracts. The larval mortality was found maximum in case of *P. niruri* (5.77 and 8.00%) and minimum mortality of 2.22 and 3.11 per cent followed by *Aegle marmelose* (2.66 and 4.22%) and *Ocimum sanctum* (5.33 and 7.11%) as compared to control (9.77 and 12.44%).

Table 2. *In-vitro* effect of *Aloe vera* gel on zone of inhibition (mm) of *Bacillus* sp. + : Inhibition, – : No Inhibition, ± : Partial Inhibition.

Botanical	Treatments	Zone of inhibition (mm) 25%	50%	75%	100%	Control
Aloe vera leaf gel	Raw gel	± 2.00	± 3.90	+ 5.70	+ 8.80	– 0.00
	Sterilized gel	–	–	–	–	–

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