International Journal of Biology Research
ISSN: 2455-6548
Impact Factor: RJIF 5.22
www.biologyjournal.in
Volume 2; Issue 4; October 2017; Page No. 97-100

Larvicidal and Anti-feedant Activity of *Phyllanthus emblica and Syzygium cumini* extracts on the Diamondback Moth: *Plutella xylostella*

Neeta Minj, * Calistus Jude AL, Sowmya SRI N
Department of Life Sciences, Kristu Jayanti College, Autonomous, K. Narayanapura, Kothanur P.O., Bangalore, Karnataka, India

Abstract
Cruciferous vegetables are one of the dominant food crops worldwide. *Plutella xylostella*, is one of the most important pests of the cruciferous crops. Larvae damage leaves, buds, flowers, and seed-buds of cultivated cruciferous plants. In the present study *Phyllanthus emblica* and *Syzygium cumini*, are checked for their antifeedant and larvicidal potential against the *Plutella xylostella*. Different parts of these plants like leaf, seed and bark extracts were used after soaking in methanol, ethanol and ethyl acetate separately. Varying concentrations of thefiltrateranging from 1-3% were tested and observed for 48hrs of exposure. Different concentrations of all the three extracts in control showed minimal antifeedant activity. 24.64% of leaf area was consumed in methanolic leaf extract of *Phyllanthus emblica* at 3% concentration and 48hrs of exposure, while *Syzygium cumini*, ethanolic leaf extract showed 39.26% of leaf consumed after 48hrs of exposure. The larvicidal activity was recorded by observing the mortality rate, of the insect after feeding on these leaves. Almost 90% mortality rate was recorded at 3% methanolic leaf concentration of *Phyllanthus emblica* after 48hrs of exposure, whereas *Syzygium cumini*, showed around 50% mortality at higher concentration and maximum exposure.No mortality rate was recorded in the control even after 48hr exposure. *Phyllanthusembilica*methanolic leaf extractshowed maximum effect on the activity of the*Plutella xylostella* compared to *Syzygium cumini*.

Keywords: cruciferous crops, *plutella xylostella, phyllanthus embilica, syzygium cumini, anti-feedant, larvicidal*

1. Introduction
Agriculture is the main source of livelihood of many peopleand is the backbone of the economic system in India. Agriculture not only provides food and raw material but also provides employment opportunities to a very large proportion of population. In a tropical country like India, owing to climatic conditions and its particular environment, agriculture suffers severe losses due to pests[1].In agriculture, insects affect directly the growing part of the crop and cause severe damage, resulting in revenue loss. Crop loss due to insect pests is estimated between ten to thirty per cent for major crops.

Cruciferous vegetables are one of the dominant food crops worldwide, Cruciferae or alternately Brassicaceae. They are high in vitamin C and soluble fiber and contain multiple nutrients. Many commonly consumed cruciferous vegetables come from the *Brassica* genus, including broccoli, Brussels sprouts, cabbage, cauliflower, collard greens, kale, kohlrabi, mustard, rutabaga, turnips, bokchoy, and Chinese cabbage. Arugula, horse radish, radish, wasabi, and watercress are also cruciferous vegetables.

One of the major constraints in the production of cruciferous crops throughout the world is infestation by the Diamondback moth (*Plutella xylostella*), sometimes called cabbage moth. The larvae are small, pale green caterpillars with tapered ends. The adult is a small, slender, grayish-brown moth with pronounced antennae. It is about 6 mm long, and marked with a broad cream or light brown band along the back. They feed on the upper and lower leaf surfaces. This pest has been reported to cause more than 90% crop loss in the area of their outbreaks [2, 3]. The economic loss due to this pest has been estimated worldwide to be US$ 4-5 billion[4]and $16 million annually in India [3, 5]. Because of concentrated utilization of insecticides, this pest has created imperviousness to about all classes of insecticides [6, 7] and furthermore issues like resurgence of pests, elimination of natural enemies, dangerous buildups in food, water, air and soil which influence human wellbeing and upset the biological community is a concern. Their continued use may further harm the environment. Under such alarming situations, potential alternative for the sustainable management of insect pest might be common plants and plant inferred items, which have been effectively utilized for quite a long time [8] over manufactured pesticides as control specialists for the nuisances of agriculture. The crude plant extract consists of complex mixtures of active compounds. The complex mixtures act synergistically and show greater overall bioactivity compared to the individual components [9, 10]. Also, there is less likelihood for the insect pest to develop resistance against such mixtures [3, 11, 12].

Phyllanthus emblica also known as Indian gooseberry or amla [13] is a deciduous tree of the family *Phyllanthaceae*. It is known for its edible fruit. All parts of the plantincluding the fruit, seed, leaves, root, bark and flowers are used in various ayurvedic herbal preparations. *Syzygium cumini*, known as jambul or jamun, is an evergreen tropical tree in the flowering plant family *Myrtaceae*. It is native to the Indian subcontinentand adjoining regions of Southeast Asia. The seed of the fruit is used in various alternative healing systems like Ayurveda to control diabetes, and in Unani and Chinese medicine for digestive ailments. Wine and vinegar are also

made from the fruit. It has a high source in vitamin A and vitamin C [14].

The potential antifeedent and insecticidal activity of the leaf, bark and seed extracts of *P.emblica and S.cumini* are analysed in the present study against the diamondback moth.

2. Materials and Methods

2.1 Collection of material

The experimental materials *P.emblica and S.cumini*were collected from K.Narayanapura and Kormangala areas of Bangalore. Different parts of these plants like leaf, bark and seeds were used in the present study.Fresh cabbage was collected from nearby shop the leaf was cut into 36cm each, for the study.

2.2 Collection of Diamondback moth

3rdinstar larvae of *P.xylostella* were collected from the mass production unit of the National Bureau of Agricultural Insect Resources (NBAII), Bangalore (AccessionNo-NBAII-MP-PLU-0), Bangalore, Karnataka.

2.3 Preparation of extract

Three different solvents Methanol, Ethanol and Ethyl Acetate were used in the extraction of bioactive compounds.

Simple percolation method was performed at room temperature where the plant material was washed thoroughly with distilled water to remove the soil particles,then it was shade dried, powdered with electric blender and used. 10g of different parts like leaf,bark and seed of each plant were soaked in 50ml of different solvents like methanol, ethanol and ethyl acetate separately for 48hours and filtered using whatmannno 1 filter paper. Then the filtrate was placed on the water bath to get a concentrated crude extract. The filtrate obtained was stored in stopper bottles in refrigerator and used for antifeedant and larvicidal assays.

Analysis of Antifeedant and Larvicial Activity

Third instar larvae of *P.xylostella* were used in the present study. Fresh cabbage leaves were washed and cut into 36cm each, for the test. Three different concentrations (1%, 2% and 3%) of different extracts (methanol, ethanol and ethyl acetate) of the two plants were sprayed separately on the leaves. The treated leaves were placed in a container and 10 third instar larvae were introducedinto each of these containers and observed for 24hours and 48hours. Moist cotton balls were placed at the bottom of the container to maintain moisture.10 larvae introduced into containers with cabbage leaves not treated with any extract served as control. The antifeedant activity and larvicidal effect was recorded at the end of the experiment.

Mortality rate and antifeedant activity was calculated using the following formula.

$$\text{Mortality (\%)} = \frac{\text{Number of dead larvae}}{\text{Total number of treated larvae}} \times 100$$

$$\text{Antifeedant activity (\%)} = \frac{\text{Area given for feeding- Area left after feeding}}{\text{Area given for feeding}} \times 100$$

3. Results and Discussion

In the present study, *P.emblica* and *S.cumini* were screened against 3rd instar larvae of *P.xylostella* for their possible potential antifeedant and larvicidal properties.

Table 1 depicts the percent of leaf consumed by the larvae of *P.xylostella* when allowed to feed on cabbage leaves treated with different concentrations of methanolic, ethanolic and ethyl acetate extracts of *P.emblica* and *S.cumini*for 24 and 48 hours.

In the present study different concentrations of all the three extracts in control showed minimal antifeedant activity. The larvae were fed on treated leaves with three different crude extracts at 1%, 2% and 3% concentration and observed for 24hrs and 48 hrs exposure time. The antifeedant effect was estimated by comparing the averages of consumed leaf area in the treated leaves against the control. Methanolic leaf extract of *P.emblica* showed 24.61% leaf consumed at 3% concentration and 48hrs of exposure. While 45.15% leaf was consumed when the larvae was treated with 3% ethanolic bark extract of *P.embilica*. Earlier study byMalliga*et.al,* 2015 [15] revealed that the petroleum ether leaf extract of *P.emblica* has potential antibacterial and antifungal activity. In a previous study by Xiaoli Liu *et al*., 2008 [16] methanolic extracts of *P.emblica* fruit from some selected regions exhibited stronger antioxidant activities.

Both methanolic and ethanolic, leaf and bark extract of *S.cumini*, showed approximately 40-50% of anti-feedant acitivity, when treated against 3% concentration and 48hrs of exposure. Whereas seed extract of ethyl acetate showed approximately 55% anti-feedant activity in both plants. Previous studies by Veeramuthu Duraipandiyan *et.al.* (2006) [17] showed the presence of flavonoids, in *S.cumini* seeds. Whereas the leaves were found to be rich in acylated flavonol glycosides, tannins, etc. In this study the control which was not treated with any of the extracts, showed approximately 64-93% of leaf area consumed, after 48hrs by both the plants.

Table 1: Antifeedant effect of different extracts (Leaf, Bark, and Seeds) of *P.emblica* and *S.cumini* on larvae of *P.xylostella*

Type of extracts			*P. emblica*						*S. cumini*					
			Consumed area (%) at 24hr exposure			Consumed area (%) at 48hr exposure			Consumed area (%) at 24hr exposure			Consumed area (%) at 48hr exposure		
			a	b	c	a	b	c	a	b	c	a	b	c
Methanol	Leaf	Control	83.26	78.07	71.15	88.46	83.19	84.65	82.26	74.07	76.15	81.46	84.19	89.65
		Experimental	55.12	52.61	50.84	53.84	39.42	24.61	72.11	59.61	52	47.96	46.15	40
	Bark	Control	80.26	75.07	72.15	83.46	85.19	88.65	74.96	73.23	85.38	75	86.88	92.73
		Experimental	41.5	56.5	60	44.61	52.69	48.07	75.46	66.53	59.61	47.5	41.5	56.5
	Seed	Control	82.26	74.07	76.15	81.46	84.19	89.65	71.80	72.07	84.65	76.75	77.38	88.84
		Experimental	70	71.15	59.5	66.53	59.61	58	75.96	71	65.38	67.65	63.07	63.19

Ethanol	Leaf	Control	80.26	75.07	72.15	83.46	85.19	88.65	81.26	74.07	76.15	82.46	84.19	89.65
		Experimental	56.15	56	62.30	51.11	43.38	38.46	76.92	72.92	58.38	65.38	53.84	39.26
	Bark	Control	82.26	74.07	76.15	81.46	84.19	89.65	73.23	75.99	85.38	74.96	76.92	88.46
		Experimental	57.61	61	66.15	41.53	54.26	45.15	57.61	61	66.15	41.53	54.26	45.15
	Seed	Control	83.06	78.07	71.15	88.46	83.19	84.65	82.26	74.07	76.15	81.46	84.19	82.46
		Experimental	68.84	70	48	65.53	49.23	46.30	70	71.15	56.5	66.53	59.61	48
Ethyl acetate	Leaf	Control	73.23	75	85.38	74.96	76.92	88.46	76.92	85.38	74.96	77.69	88.46	92.73
		Experimental	67.69	71.15	49.11	50.38	48.73	45.07	78.84	72.92	58.38	65.38	53.84	58.26
	Bark	Control	71.80	72.07	84.65	76.75	77.38	88.84	83.06	78.07	71.15	88.46	83.19	84.65
		Experimental	76.92	72.92	58.38	65.38	53.84	58.26	83.84	66.53	59.61	47.5	41.5	56.5
	Seed	Control	74.96	73.23	85.38	75	86.88	92.73	82.26	74.07	85.38	81.46	84.19	89.65
		Experimental	78.80	77.69	73.23	76.15	63.07	55	85.39	71	65.38	67.65	63.07	63.19

a = 1% concentration; b = 2% concentration; c = 3% concentration

The larvicidal effect of different extracts (Leaf, Bark and Seeds) of *P.emblica*and *S. cumini* on larvae of *P. xylostella*is shown in table 2.

In the present study, the mortality percentage of *P. emblica* with methanol, ethanol, and ethyl acetate extracts on larvae of *P. xylostella* is comparable to the toxicity observed for *S. cumini* extracts for the same solvent. When treated with methanolic leaf extract of *P. emblica* 90% mortality was observed at 48hrs of exposure. Whereas bark extract showed highest mortality rate at 24hr exposure, i.e., 50% in 3% concentration, while at 1% concentration there was no mortality observed. At 48 hr exposure, 80% mortality was recorded at 3% concentration of the extract. None of the larvae maintained as control died. Exposure to ethanolic bark extract. The ethyl acetate seed extract showed minimum mortality at 24hr and 48hr. In a previous study by Alagarmalai Jeyasankar, *et.al*, (2012) [18] *P.emblica* showed susceptibility against the larvae of *Cx. quinquefasciatus* with a LC50 value of 78.89 ppm.

When treated with methanolic leaf extract of *S.cumini*, 50% mortality was observed at 48hrs of exposure. Whereas bark extract showed minimum mortality rate of 10% at 24hr exposure and at 1% concentration there was no mortality observed. At 48 hr exposure, 40% mortality was recorded at 3% concentration of the extract. No mortality was observed in Control. Exposure to ethanolic bark extract of *S.cumini* for 24hr resulted in mortality percentage of 20 and 30, at 2% and 3% whereas no larvae died at 1% concentration. At 48hr exposure time and 40% mortality was observed at 3% against 0% mortality in Control. The ethyl acetate seed extract showed least mortality at 24hr and 48hr. A study by Ravi Shankara Birur Eshwarappa, *et.al* 2014 [19] showed leaf galls extracts of *S.cumini* possesses high antioxidant activity. Previous studies by Ruan ZP *et.al* 2008 [20] showed that the ethyl acetate fraction of *S. cumini* extracts had stronger antioxidant activity.

Table 2: Larvicidaleffect of different extracts (Leaf, Bark, and Seeds) of *Phyllanthus emblica* and *Syzygium cumini* on larvae of *Plutella xylostella*

Type of extract			***P. emblica***						***S. cumini***					
			Mortality (%) at 24hr exposure			**Mortality (%) at 48hr exposure**			**Mortality (%) at 24hr exposure**			**Mortality (%) at 48hr exposure**		
			a	**b**	**c**	**a**	**b**	**c**	**A**	**b**	**c**	**a**	**b**	**c**
Methanol	Leaf	Control	0	0	0	0	0	0	0	0	0	0	0	0
		Experimental	10	40	60	30	80	90	0	20	30	10	30	50
	Bark	Control	0	0	0	0	0	0	0	0	0	0	0	0
		Experimental	0	20	50	20	70	80	0	10	10	10	20	40
	Seed	Control	0	0	0	0	0	0	0	0	0	0	0	0
		Experimental	0	20	40	20	40	50	0	0	10	10	10	20
Ethanol	Leaf	Control	0	0	0	0	0	0	0	0	0	0	0	0
		Experimental	30	40	50	40	50	60	10	20	30	20	20	50
	Bark	Control	0	0	0	0	0	0	0	0	0	0	0	0
		Experimental	0	10	40	10	30	60	0	20	30	30	30	40
	Seed	Control	0	0	0	0	0	0	0	0	0	0	0	0
		Experimental	0	10	20	10	10	30	0	0	10	10	20	20
Ethyl acetate	Leaf	Control	0	0	0	0	0	0	0	0	0	0	0	0
		Experimental	0	10	30	10	20	40	0	10	20	10	20	30
	Bark	Control	0	0	0	0	0	0	0	0	0	0	0	0
		Experimental	0	10	20	10	20	30	10	10	20	20	20	30
	Seed	Control	0	0	0	0	0	0	0	0	0	0	0	0
		Experimental	0	0	10	10	10	20	0	0	10	10	20	20

a = 1% concentration; b = 2% concentration; c = 3% concentration

Of *P.emblica* for 24hr resulted in mortality percentage of 10 and 40, at 2% and 3% whereas no larvae died at 1% concentration. Thus it can be inferred from the above results that the methanolic leaf extract of *P.emblica* and ethanolic leaf

extract of *S.cumini* possess the highest larvicidal effect at all the three concentrations when compared to bark and seed extracts. Dose dependant mortality was observed throughout the study.

3. Conclusion

Brassica vegetables are regarded highly for their nutritional value. These crops are often affected by *P.xylostella*. In the present study, root, leaf, bark extracts of *P.emblica* and *S.cumini* treated with 3 different solvents against *P.xylostella.*It is evident from the tabulated results that different extracts of *P.emblica*had a better effect when compared to extracts of *S.cumini.* It was observed that increase in concentration and exposure period, increased the rate of mortality and decrease leaf area consumed.

4. References

1. Arivoli S. Samuel Tennyson. Antifeedant activity, developmental indices and morphogenetic variations of plant extracts against Spodopteralitura (Fab) (Lepidoptera: Noctuidae). Journal of Entomology and Zoology Studies. 2013; 1(4):87-96.
2. Robert HJ, Verkerkand Denis J, Wright. Multitrophic interactions and management of the diamondback moth: a review. Bulletin of Entomological Research. 1996; 86(3):205-216.
3. Sharma S, Senrungand A, Singh AK. Toxic effect of neem, Azadirachtaindica(A. Juss) foliage extracts against diamondback moth (DBM), Plutella xylostella (L.) (Lepidoptera, Plutellidae). Journal of Biopesticides. 2014; 7:99-105
4. Zalucki MP, Shabbir A, Silva R, Adamson D, Shu-Sheng L, Furlong MJ. Estimating the economic cost of one of the world's major insects pests, *Plutella xylostella* (Lepidoptera: Plutellidae): just how long is a piece of string?. Journal of Economic Entomology. 2012; 105(4):1115-1129.
5. Mohan M, Gujar GT. Local variation in susceptibility of the diamondback moth *Plutella xylostella* (Linnaeus) to insecticides androle of detoxification enzymes. Crop Protection. 2003; 22(3):495-504.
6. Shelton AM, Robertson JL, Tang JD, Cperez C, Eigenbrode SD, Preisler HK, *et.al.* Resistance Of diamondback moth (Lepidoptera: Plutellidae) To Bacillus Thurillgiensis subspecies in the field. Journal of economic entomology. 1993; 86(3):697-705.
7. Lingathurai S, Ezhil Vendan S, Gabriel Paulraj M, Ignacimuthu S. Antifeedant and larvicidal activities of *Acalyphafruticosa* Forssk. (Euphorbiaceae) against *Plutella xylostella* L. (Lepidoptera: Yponomeutidae) larvae. Journal of King Saud University – Science. 2011; 23(1):11-16.
8. Crosby DG. Minor Insecticides of Plant Origin. In: Naturally Occurring, Insecticide (Jacobson, M. and Crosby, D.G eds.). Marcel Dekker Inc., New York. 1971, 117-239.
9. Berenbaum M. Brementownrevisted. Interactions among allelochemicals in plants. Rec. Adv. Phytochemicals. 1985; 19:139-169.
10. Chen W, Isman MB, Chiu SF. Antifeedant and growth inhibitory effects of the limonoidtoosendanin and Meliatoosendan extracts on the variegated cutworm, Peridromasaucia (Lepidoptera: Noctuidae). Journal of Applied Entomology. 1995; 119(1-5):367-370.
11. Feng R, Isman MB. Selection for resistance to azadirachtin in the green peach aphid, My zuspersicae. Experimentia. 1995; 51(8):831-833.
12. Shukla A, Toke NR. Plant products as a potential stored product insect management agents. Indian Journal of Research. 2013; 2(2):4-6.
13. https://en.wikipedia.org/wiki/Phyllanthus_emblica
14. https://en.wikipedia.org/wiki/Syzygium_cumini
15. Malliga Elangovan N, Dhanarajan MS, Elangovan I. Evaluation of Antibacterial and Antifungal Activity of Phyllanthus emblica Leaf Extract. International Research Journal of Pharmaceutical and Biosciences. 2015; 2(2):59-66.
16. Xiaoli Liu, Mouming Zhao, Jinshui Wang, Bao Yang, Yueming Jiang. Antioxidant activity of methanolic extract of emblica fruit (*Phyllanthus emblica* L.) from six regions in China. Journal of Food Composition and Analysis. 2008; 21(3):219-228
17. Veeramuthu Duraipandiyan, Muniappan Ayyanar, Savarimuthu Ignacimuthu. Antimicrobial activity of some ethnomedicinal plants used by Paliyar tribe from Tamil Nadu, India. Bio Med Central. 2006; 6:35.
18. Alagarmalai Jeyasankar, Premalatha and Kuppusamy Elumalai. Larvicidal activity of Phyllanthus emblica Linn. (Euphorbiaceae) leaf extracts against important human vector mosquitoes (Diptera: Culicidae). Asian Pacific Journal of Tropical Disease. 2012; 2(1):S399-S403.
19. Ravi Shankara Birur Eshwarappa, Raman ShanthiIyer, Sundara Rajan Subbaramaiah, Austin Richard Sand Bhadrapura Lakkappa Dhananjaya. Antioxidant activity of *Syzygium cumini* leaf gall extracts. Bioimpacts. 2014; 4(2):101-107.
20. Ruan ZP, Zhang LL, Lin YM, Ruan ZP, Zhang LL, Lin YM. Evaluation of the antioxidant activity of *Syzygium cumini* leaves. Molecules. 2008; 13(10):2545-2556.