Indian Journal of Entomology 84(2): 466-469 (2022) DoI No.: 10.55446/IJE.2021.257

MANAGEMENT OF BORER AND FLUBENDIAMIDE RESIDUE IN OKRA USING MILK-MADE BIOAGENT AND CALCIUM HYPOCHLORITE

K Elakkiya*, P Yasodha[1] and C Gailce Leo Justin[1]

Department of Agricultural Entomology, Tamil Nadu Agricultural University (TNAU), Coimbatore 641003, Tamil Nadu, India
[1]Department of Plant Protection, Anbil Dharmalingam Agricultural College and Research Institute (TNAU), Tiruchirappalli 620027, Tamil Nadu, India
*Email: elakkiyaagri96@gmail.com (corresponding author)

ABSTRACT

Okra is an important vegetable, and it is subject to heavy yield losses by insect pests, of which the shoot and fruit borer *Earias* spp. is serious. Insecticides are used against this pest, leading to insecticide residues. This study evaluates Milkoid, an LAB formulation spray in field by spraying in tandem with flubendiamide and bleaching powder (calcium hypochlorite). The results indicate that the population of epiphytic LAB density was maximum on plants sprayed with Milkoid with or without flubendiamide. The pest damage was significantly reduced after spraying flubendiamide with or without bleaching powder (3.76- 5.30%). Milkoid accelerated the degradation of flubendiamide with the dissipation rate of 70.07% on 15th day after spray. The efficacy of Milkoid in reducing the toxicity and the potential of calcium hypochlorite as an antimicrobial agent and the shoot and fruit borer management are discussed.

Key words: Okra, shoot and fruit borer, *Earias* spp., imidacloprid, milkoid, bleaching powder, lactic acid bacterial formulation

Okra is an important vegetable rich in nutrients (Gemede et al., 2015), and its productivity in India and Tamil Nadu is 11.97 mt/ ha and 10.56 mt/ ha, respectively. The productivity is less due to insect pests, of which the shoot and fruit borer *Earias* spp. (Lepidoptera: Noctuidae) accounts for 69% yield loss. To avoid this insecticides are used leading to residues (Aktar et al., 2009). Flubendiamide is widely used insecticide to manage *Earias* spp. in okra (Srinivasnaik et al., 2015). Because of indiscriminate use of insecticides market samples are found to be contaminated with insecticide residues**.** Despite regulations and monitoring, pesticide residues are found in vegetables and fruits (Farag et al., 2011; Trivedi et al., 2014). Dordevic et al. (2013) observed *Lactobacillus plantarum* to degrade 81% of pirimiphos methyl residues in wheat. These have also been exploited in crop production as biofertilizers, biocontrol agents and biostimulants promoting plant growth (Lamont et al., 2017). Milk is a rich source of probiotic LAB, especially *Lactobacillus acidophilus* (Wang et al., 2016). Thus, LAB formulations might help reduce the risks from pesticides if sprayed on crops as an adjuvant that performs specific functions including wetting, spreading, sticking and spray drifting (Green, 2000). Milkoid is a milk-made formulation (David et al., 2018). However, how these influence pest infestations is not known**.** This study assesses the status of borer pests and insecticide residue in okra after spraying flubendiamide and milk- made formulation (LAB) in comparison with bleaching powder (calcium hypochlorite), a commercially available chlorine compound with antimicrobial properties.

MATERIALS AND METHODS

Field experiment was conducted at the Anbil Dharmalingam Agricultural College and Research Institute, Tiruchirappalli, Tamil Nadu, during 2018-19. Okra crop (hybrid Jaani) was ised in 5x 4 m^2 plots with a spacing of 45x 30 cm. The seven treatments were evaluated in a randomized block design (RBD) with three replications. At reproductive stage, sprays were made with the flubendiamide 39.35SC (Bayers Crop Science) at 1.2 ml/ 10 l. The lactic acid bacterial formulation, referred to as Milkoid, was prepared as per Elakkiya et al. (2019). There were three controls- Bleaching powder (calcium hypochlorite) was purchased from the local market and sprayed at 1.0% as the antimicrobial control while water-spray and no-spray were the other two controls. Two rounds of sprays were made at fortnightly interval early in the morning using a 1l capacity hand-operated sprayer. Separate sprayers were used for each treatment to avoid contamination. Observation was made at weekly intervals on borer infestation in % from the fruits harvested. The population density of the LAB present on the fruit samples (2 cm length) was assessed at weekly interval as per Elakkiya et al. (2019). The residue of flubendiamide was analysed using HPLC. The fruit samples were collected from treated plots at 1, 3, 5, 7, 9, 11, 13 and 15 days after spraying and sample was done using the QuEchERs method before HPLC instrumentation. HPLC analysis: using HPLC (HPLC-SPD- M2OA) with the Photo Diode Array Detector, C-18 coloumn and acetonitrile as a carrier. The volume of sample injected was 20 µl and the wavelength fixed was 254 λ with the solvent ratio of 50:50 (acetonitrile: water). The insecticide residue recovery was calculated as per Hamid and Hamid (2015). The dissipation was calculated by subtracting residue on first day from residue on present day divided by residue on first day multiplied by 100.

RESULTS AND DISCUSSION

In the field experiment, in the first week incidence of borer was significantly less when the plants were treated with flubendiamide 39.35% SC 0.12 ml/ l; in tandem with bleaching powder 1.0% (4.07%) was also on par with flubendiamide alone (6.75%) (Table 1); it was of a moderate level after flubendiamide / milkoid 2.0% spray (12.86%), and bleaching powder (10.99%). In the second week, the infestation was significantly less with flubendiamide/ bleaching powder (7.16%), on par with flubendiamide (7.65%) and flubendiamide/ milkoid (11.37%); flubendiamide/ milkoid was on par with bleaching powder (13.36%). The incidence was moderate with milkoid, and on par with bleaching powder spray in both weeks. Pooled data indicated significantly less damage after spraying flubendiamide/ bleaching powder (3.76%), on par with flubendiamide alone (5.30%), followed by flubendiamide/ milkoid and bleaching powder (10.01- 10.10%); milkoid 2.0% also caused significant reduction (16.51%). Though not an insecticide, milkoid was reduced the incidence of borer. The LAB density was significantly maximum on okra leaves after spraying flubendiamide / milkoid (38.89- 37.22) and milkoid 2% (35.39- 34.11), in both the weeks (Table 1). The density was second highest on fruits after spraying flubendiamide (26.72– 23.11) as on control plants. Bleaching powder 1.0% with or without flubendiamide (19.78-12.61), suppressed the LAB density significantly the most. The pooled data showed that LAB density was significantly maximum when Milkoid was sprayed either alone or in combination with flubendiamide (35.67- 37.14 CFU/ cm^2), and

Table 1. Fruit borer incidence after two sprays of flubendiamide, milkoid and bleaching powder in okra

Treatments	Damaged fruits (%) At first spray	Damaged fruits (%) At second spray	Pooled mean (%)	LAB density (CFU/ 2 cm^2) At first spray	LAB density (CFU/ 2 cm^2) At second spray	Pooled mean (CFU/ 2 cm^2)
Flubendiamide 39.35% SC 0.12 ml/ l	6.75 (14.91)^{ab}	7.65 (16.06)^{a}	5.30 (13.31)^{a}	23.11 (1.36)^{b}	26.72 (1.36)^{b}	24.92 (1.40)^{b}
Flubendiamide 39.35% SC 0.12 ml/ l + milkoid 2.0 %	12.86 (20.79)^{bc}	11.37 (19.71)^{ab}	10.01 (18.44)^{b}	38.89 (1.59)^{a}	37.22 (1.59)^{a}	37.14 (1.57)^{a}
Flubendiamide 39.35% SC 0.12 ml/ l + bleaching powder 1.0 %	4.07 (9.37)^{a}	7.16 (15.52)^{a}	3.76 (11.18)^{a}	12.61 (1.06)^{c}	19.78 (1.30)^{c}	16.19 (1.21)^{c}
Milkoid 2.0 %	18.81 (25.38)^{c}	18.17 (25.23)^{c}	16.51 (23.98)^{c}	34.11 (1.53)^{a}	35.39 (1.55)^{a}	35.67 (1.55)^{a}
Bleaching powder 1.0 %	10.99 (18.96)^{bc}	13.36 (21.44)^{bc}	10.10 (18.53)^{b}	14.00 (1.13)^{c}	18.17 (1.26)^{c}	16.08 (1.21)^{c}
Water spray	47.15 (43.37)^{d}	37.02 (37.48)^{d}	42.43 (40.65)^{d}	20.83 (1.32)^{b}	26.94 (1.43)^{b}	23.89 (1.38)^{b}
Untreated control	46.48 (42.95)^{d}	39.91 (39.18)^{d}	43.61 (41.33)^{d}	22.28 (1.35)^{b}	25.39 (1.40)^{b}	25.83 (1.38)^{b}
Mean	21.02 (27.29)	16.62 (24.95)	18.82 (25.71)	23.69 (1.37)	27.09 (1.43)	25.39 (1.40)
CD (p =0.05)	6.50	4.68	2.08	0.05	0.05	0.05
SEd	3.16	2.34	1.04	0.03	0.02	0.03

Mean of 3 replications; Figures in parentheses arc sin transformed and are log transformed values for borer LAB respectively; Means followed by the same letter are not significantly different, LAB, lactic acid bacteria; CFU, colony forming units significantly lowest after spraying bleaching powder either alone or in combination with flubendiamide (16.08- 16.19 CFU / cm^2).

The level of flubendiamide decreased from 1.39 ppm on the 3rd day to 0.73 ppm on 15th day (Table 2); in combination of flubendiamide and Milkoid it decreased from 1.37 ppm on 3rd day to 0.41 ppm on 15th day with highest dissipation ranging up to 70.07% on 15th day while flubendiamide in combination with bleaching powder had dissipated up to 1.00 ppm on 15th day with dissipation rate of 1.42% on 5th day to 52.38% on the 15th day (Fig. 1). Milk is a very good source of heat-tolerant LAB (Sugimoto et al., 2008). As pure cultures, several bacterial strains degrade pesticides in the presence of an additional carbon source (Hussain et al., 2016). The present study revealed that epiphytic LAB can modulate pest incidence and accelerate dissipation of pesticide residues. The density of LAB increased on plants sprayed with flubendiamide/ milkoid 60% and the residue dissipation was also more with the plants sprayed with flubendiamide in tandem with milkoid, demonstrating that LAB in milkoid can influence residues. The LAB densities are significantly lower whenever bleaching powder was used, either alone or with pesticide, and is a pointer that the latter had inhibitory effects on microbes, especially LAB because chlorine compounds have sporicidal actions (Russell, 1990). When sprayed in tandem with bleaching powder, flubendiamide was the most effective treatments, suppressing *Earias* incidence by 91.38%. Though not an insecticide, Milkoid was also able to reduce pest incidence in okra to various levels, either alone or with pesticides, and flubendiamide was significantly less effective or less hazardous, probably through biodegradation- there are reports that LAB degrade pesticides not only in products like kimchi (Cho et al., 2009) and skimmed milk (Zhou and Zhao, 2014) but also on plants as epiphytic (Islam, 2010).

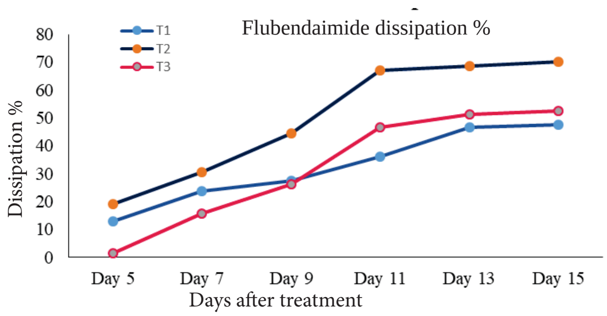

Fig. 1. Dissipation of Flubendaimide 39.35% SC (T1, Flubendiamide 39.35% SC 0.12 ml/l; T2, Flubendiamide 39.35% SC 0.12 ml/l + Milkoid 2.0 %; T3, Flubendiamide 39.35% SC 0.12 ml/l + Bleaching powder 1.0 %)

In conclusion, the milk-made sugar-preserved LAB formulation Milkoid could be used as an adjuvant bio-remedy to limit the toxicity of pesticides. Whether it could be mixed directly with insecticide-mixed spray fluids needs further study. It is also worth exploring the potential of combination of calcium hypochlorite and Milkoid in tandem with flubendiamide in crop protection as the former suppressed the population density and the later reduced the residue level of flubendiamide.

Table 2. Residue of flubendiamide 39.35% SC

Days after spray	Treatments	Residues (ppm)	Days after spray	Treatments	Residues (ppm)
	T_1	1.39	11	T_1	0.89
3	T_2	1.37		T_2	0.45
	T_3	2.10		T_3	1.12
	T_1	1.21	13	T_1	0.74
5	T_2	1.11		T_2	0.43
	T_3	2.07		T_3	1.02
	T_1	1.06	15	T_1	0.73
7	T_2	0.95		T_2	0.41
	T_3	1.77		T_3	1.00
	T_1	1.01			
9	T_2	0.76			
	T_3	1.55			

T1, Flubendiamide 39.35% SC 0.12 ml/ l; T2, Flubendiamide 39.35% SC 0.12 ml/ l / Milkoid 2.0%; T3, Flubendiamide 39.35% SC 0.12 ml/ l / Bleaching powder 1%.

REFERENCES

Aktar W, Sengupta D, Chowdhury A. 2009. Impact of pesticides use in agriculture: their benefits and hazards. Interdisciplinary Toxicology 2(1): 1-12.

Cho K M, Math R K, Islam S M A, Lim W J, Hong S Y, Kim J M, Yun M G, Cho J J, Yun H D. 2009. Biodegradation of chlorpyrifos by lactic acid bacteria during kimchi fermentation. Journal of Agricultural and Food Chemistry 57(5): 1882-1889.

David P M M, Mathialagan M, Prabina J J, Sabarinathan K G, Pushpam A K, Elanchezhyan K, Edward Y S J T, Mohankumar S. 2018. Honey-simulated discriminate fermentation process for preserving perishable juices as probiotics without chemical preservatives. Patent Application 201841016405 dated 1.5.2018 (Trade name: TNAU Winectar)

Elakkiya K, Yasodha P, Justin C G L, Ejilane J, Somasundaram S, David P M M. 2019. Suppression of sucking insects in okra after spraying imidacloprid and milk-made lactic acid bacterial formulation. The Pharma Innovation Journal 8(6): 592-598.

Farag R S, Latif A, El-Gawad A E A, Dogheim S M. 2011. Monitoring of pesticide residues in some Egyptian herbs, fruits and vegetables. International Food Research Journal 18(2).

Gemede H F, Ratta N, Haki G D, Woldegiorgis W Z, Beyene F. 2015. Nutritional quality and health benefits of okra (*Abelmoschus esculentus*): a review. Journal of Food Processing Technology 25(1): 16-25.

Green J. 2000. Adjuvant outlook for pesticides. Pesticide Outlook 11(5): 196-199.

Hamid F, Hamid F H. 2015. Manual of methods of analysis of foods. Food safety and standards authority of India. 256 pp.

Hussain S, Hartley C J, Shettigar M, Pandey G. 2016. Bacterial biodegradation of neonicotinoid pesticides in soil and water systems. Microbiology Letters 363(23).

Islam S, Asraful M, Math R K, Cho K M, Lim W J, Hong S Y, Kim J M, Yun M G, Cho J J, Yun H D. 2010. Organophosphorus hydrolase (OpdB) of *Lactobacillus brevis* WCP902 from kimchi is able to degrade organophosphorus pesticides. Journal of Agricultural and Food Chemistry 58(9): 5380-5386.

Lamont J R, Wilkins O, Bywater-Ekegard M, Smith D L. 2017. From yogurt to yield: Potential applications of lactic acid bacteria in plant production. Soil Biology and Biochemistry 111: 1-9.

Russell A D. 1990. Bacterial spores and chemical sporicidal agents. Clinical microbiology reviews 3(2): 99-119.

Srinivasnaik S, Kuttalam S, Philip H, Bhuvaneswari K. 2015. Study on pesticide usage pattern by the farmers in major okra growing areas of Coimbatore district in Tamil Nadu. International Conference on innovative insect management approaches for sustainable agro ecosystem. Madurai, Tamil Nadu.

Sugimoto S, Sonomoto V. 2008. Molecular chaperones in lactic acid bacteria: physiological consequences and biochemical properties. Journal of Bioscience and Bioengineering 106(4): 324-336.

Trivedi P, Sharma V P, Srivastava L P, Malik S. 2014. Multiresidue analysis of organophosphorus pesticides in fruits and vegetables by GC-NPD. International Journal of Advanced Research 2 (10): 600-606.

Wang D, Wenjun L, Yan R, Liangliang D, Donglei Z, Yanrong Y, Qiuhua B, Heping Z, Bilige M. 2016. Isolation and identification of lactic acid bacteria from traditional dairy products in Baotou and Bayannur of midwestern Inner Mongolia and q-PCR analysis of predominant species. Korean Journal for Food Science of Animal Resources 36(4): 499.

Zhou X, Zhao X. 2015. Susceptibility of nine organophosphorus pesticides in skimmed milk towards inoculated lactic acid bacteria and yogurt starters. Journal of the Science of Food and Agriculture 95(2): 260-266.

(Manuscript Received: November, 2020; Revised: January, 2021;
Accepted: January, 2021; Online Published: May, 2021)
Online published (Preview) in www.entosocindia.org Ref. No. e20281