Original Research Article

Efficacy of Some Biorational Approaches Against Two-Spotted Spider Mite (*Tetranychus urticae* Koch) in Bitter Gourd (*Momordica charantia* L.)

ABSTRACT

Aims: The aim of the study was to test the effectiveness of several biorational techniques against two-spotted spider mite, *Tetranychus urticae* Koch. in bitter gourd.

Study design: The study used a single component Randomized Complete Block Design (RCBD) with three replications.

Place and Duration of Study: The experiment was conducted at Sher-e-Bangla Agricultural University's central farm in Dhaka, Bangladesh from March 2021 to July 2021.

Methodology: The study included seven treatments. T_1= Spinomax (0.4ml/L water); T_2= Neem Oil (5ml/L water); T_3= Biomax (1.2ml/L water); T_4= Ripcord (1ml/L water); T_5= *Trichoderma* spp (200mg/L water); T_6= Mehogany Leaf Extract (200mg/L water); T_7= Absolute (Control). All the treatments were applied at 15 days interval.

Results: Treatments for mite infestations showed varying responses in experiments with various biorational control techniques. *Trichoderma* spp. had the highest results, followed by neem oil, which was effective against mite pests infesting bitter gourd. The results showed that when *Trichoderma* spp. was used to treat leaf infestation, the total number of fruit set, % healthy fruit, and % infested fruit were all greater. Ripcord had the lowest outcomes against mite pests in most circumstances, with the exception of control treatment.

Conclusion: It can be concluded that *Trichoderma* spp. @200mg/L of water can be used to treat two spotted spider mites in an environmentally acceptable manner.

Keywords: Biorational approach; Tetranychus urticae; Trichoderma spp.; Botanicals; Biopesticides.

1. INTRODUCTION

With a spectacular growth rate of 1.37 %, Bangladesh has one of the world's densest populations (1125 people per square kilometer). Despite Bangladesh's goal of becoming a middle-income country by 2021, agriculture remains the country's major employer, employing 40.6 % of the population [1]. Agriculture has been a driving force behind Bangladesh's economic prosperity and stability [2]. Bangladesh cultivates a diverse array of vegetables. It is an important aspect of Bangladesh's crop agriculture, contributing $718 million (3.2%) to the country's agricultural GDP in 2018 [3]. Because vegetables are a good source of micronutrients, they are the most commonly consumed group of plants. Bangladesh grows brinjals, tomatoes, cauliflower, bitter gourd, ridge gourd, beans, carrots, spinach, and other key vegetables.

Bitter gourd (*Momordica charantia* L.) is a popular summer vegetable in Bangladesh and throughout Asia [4]. It's high in ascorbic acid, iron, vitamins A, B, and C, as well as carbs [5]. It is a fast-growing cucurbitaceous vegetable in Bangladesh, with a total yield of 59,371 metric tons on 27,484 acres [1]. Bitter gourd production is 24-27 t ha^{-1} [6]. It is thought to have medicinal benefits, and a chemical called 'Charantin' found in bitter gourds can help diabetic people lower their blood sugar levels [7]. Due to substantial damage caused by different insects, pests, and illnesses, bitter gourd growers frequently fail to achieve the desired output. As a result, farmers sprayed pesticides often since pests alone resulted in a 25 % annual output loss for vegetable crops every year [8].

Mites are the most diverse members of the phylum Arthropoda, belonging to the subphylum Chelicerata and subclass Acari. The only arachnid species that consumes plants is the acari. Plant feeding mites are a type of agricultural pest that feeds on fruits, vegetables, forage crops, ornamentals, and other plants. Tetranychidae, Tenuipalpidae, Eriophyidae, Tarsonemidae, and Tuckerillidae are the five families of phytophagous mites comprising 7000 mite species that have been recognized globally [9]. The Tetranychidae family, popularly known as spider mites, is a huge family with approximately 1,200 species distributed across 70 genera. *Tetranychus urticae* Koch (Acari: Tetranychidae), a two-spotted spider mite that affects over 1000 plant species from over 140 plant families, is one of the most devastating polyphagous pest species [10,11]. Koch originally characterized it in 1836, and it is assumed to have originated in temperate areas [12]. It feeds mostly on the midrib and plant veins, resulting in 50 to 100 % yield reductions [13]. Mites are treated with a variety of systemic chemical pesticides, including synthetic pesticides, albeit the mite has quickly developed resistance to these chemicals. The rapid evolution of pesticide resistance has been facilitated by mites' quick growth and development, high fecundity, and haplo-diploid sex determination [14]. Pesticide residues in vegetables are causing a drop in vegetable exports, owing to substantial concerns among importing countries [15]. Furthermore, the usage of synthetic chemicals has caused major environmental issues and posed a threat to human [16]. Alternative techniques, such as the use of various bio-control agents, essential plant oils, and bio-pesticides, should be investigated for their acaricidal activity against mites in order to reduce the usage of chemical acaricides and develop integrated management tactics.

Plant-based pesticides are environmentally friendly and non-toxic to humans, fish, and other wildlife. Many essential oils extracted from plants had insecticidal and acaricidal activities against a variety of soft-bodied arthropod pests [17]. They have a variety of useful qualities, including repellence, anti-feedant activity, growth regulation, and toxicity to a variety of insect and mite pests [18]. Today's vegetable growers in Bangladesh and other nations utilize a variety of synthetic chemical pesticides, including organophosphates, organocarbamates, pyrethroids, and nicotinoids [19]. Bangladeshi farmers rely heavily on the use of hazardous pesticides, with dangerous pesticides accounting for up to 25% of the cost of agriculture [20].

There are findings available in Bangladesh on the efficiency of different chemical pesticides [21] and predators [22] in suppressing mites. However, there was no indication that entomopathogens, plant oils, and bio-pesticides were effective against mite populations. As a result, the current study was developed to develop an eco-friendly management strategy for mite populations in Bangladesh employing *Trichoderma* spp., bio-pesticide, plant oils, plant extracts, and insecticide.

2. MATERIALS METHODS

From March 2021 to July 2021, the research was conducted at Sher-e-Bangla Agricultural University's central farm at Sher-e-Bangla Nagar, Dhaka-1207.

2.1 Geographical location and climate

The site is located in latitude 23077'N and longitude 90033'E, with an elevation of 8.2 meters above sea level. The experimental site is located in a subtropical environment, and its climatic circumstances were characterized by significant sparse rainfall throughout the rabi season. The soil was from AEZ-28, "The Modhupur Tract." The test area was flat, with an irrigation and drainage system in place, and was above flood level.

2.2 Planting materials

BARI Korola 1 was used as the test crop in this experiment. Seeds were collected from BARI (Bangladesh Agricultural Research Institute), Gazipur, Bangladesh.

2.3 Treatments

Table 1. Treatments used in order to check efficacy of miticide with bio- pesticides in controlling mites

Treatment No.	Name	Dose/ha
T_1	Spinomax	0.4 ml/L of water at 15 days interval
T_2	Neem oil	5 ml/L of water at 15 days interval
T_3	Biomax	1.2 ml/L of water at 15 days interval
T_4	Ripcord	1 ml/L of water at 15 days interval
T_5	*Trichoderma sp.*	200 mg/L of water at 15 days interval
T_6	Mehogany leaf extract	200 mg/L of water at 15 days interval
T_7	Control	No pesticide applied

2.4 Experimental design

To eliminate soil heterogeneity, the experiment was set up in a single factor randomized complete block design (RCBD) with three replications, with the experimental area divided into three blocks representing the replications. Each block was separated into seven-unit plots, with elevated bunds separating the treatments. As a result, there were 7X3=21 plots in total. The unit plot was 3.6 m×1.6 m in size. Two blocks and two plots were kept at a distance of 0.5 m and 0.5 m, respectively.

2.5 Land preparation and intercultural operation

On March 2021, seeds of the bitter gourd variety were sown in the seedbed. With the help of the farm department, the plot chosen for the experiment was opened in the first week of April 2021and kept exposed to the sun for a week. After one week, the soil was harrowed, ploughed, and cross-ploughed numerous times, followed by laddering to guarantee good tilth. Each unit site's soil was amended with organic and inorganic manures. On April 15, 2021, seedlings were transplanted. When irrigation and drainage were required, they were provided. Weeding was carried out to keep the plots free of weeds, allowing for greater growth and development.

2.6 Manuring and fertilizer application

Fertilizers N, P, K in the form of Urea, TSP, MOP, and S, Zn, and B in the form of gypsum, zinc sulphate, and borax were supplied, as recommended by the Bangladesh Agricultural Research Institute [23].

2.7 Data recording

2.7.1 Data recording on plant height affected by mite infestation and treatments

The height of the bitter gourd plant was measured three times in the field using a meter scale. At 20 DAP, 40 DAP, and 60 DAP, the heights of the plants were measured.

2.7.2 Data recording on infested leaves affected by mite infestation and treatments

During the early morning hours, when the pest was less active, the number of mite nymphs and adults was counted on six leaves (each from two upper, middle, and lower leaves per plant). Mites on the upper surface of the leaves were counted first, followed by a careful tilt of the leaf to count the population on the lower surface.

2.7.3 Data recording on infested fruits affected by mite infestation and treatments

On 2, 7, and 15 days after spraying, live mite populations were observed before and after treatment. Individual plot fruit yields were recorded independently at each harvest, and total yield was calculated on a hectare basis.

2.8 Statistical analysis

The collected data was entered into an MS Excel spreadsheet and collated. Data was processed using STATISTIX 10 software for analysis of variance later on. The t variance test was used to create an ANOVA, and mean value comparisons were done by Least Significant Difference (LSD) test.

3. RESULTS AND DISCUSSION

3.1 Position of mite species

Mite is a minute pest of vegetables which suck the plant sap from the lower part of the leaves. It causes viral disease as secondary pest.

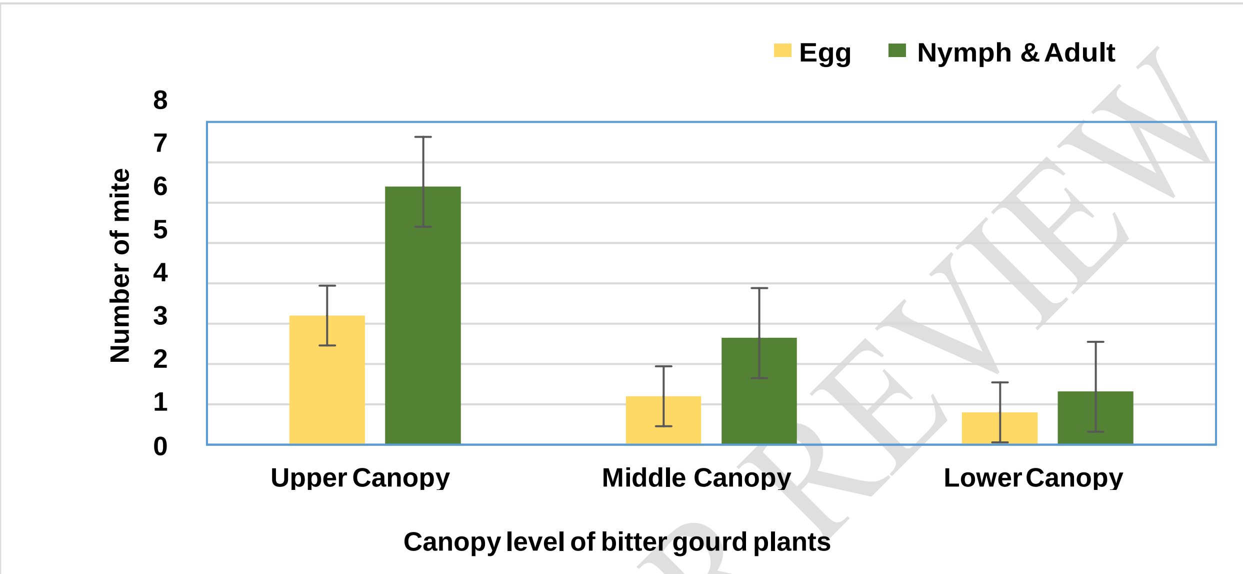

Fig. 1. Population of *Tetranychus urticae* Koch found in different canopy level of bitter gourd

From the figure 1, it is found that the highest number of mites was obtained from the upper canopy (3.2 eggs and 6.4 nymphs and adults respectively). Subsequently, from the middle canopy the number of mite population was 1.2 and 2.65 eggs and active stages of mite respectively. Furthermore, the lowest number (0.8 eggs and 1.32 nymphs and adults respectively) of mite populations were obtained from the lower canopy of sampled bitter gourd plants. [24] conducted an experiment on population abundance of red spider mite, *T. urticae* Koch in cucumber, ribbed gourd, bitter gourd, snake gourd, aroids and teasel gourd as well as in brinjal. They found highest number of mite pests in upper canopy of bitter gourd field. Thus, findings of present study show conformity with the study of [24]. Along with their presence, mites affect the plants in many ways. The first sight of infestation by red spider mite was usually chlorotic, stippled appearance on the leaves.

3.2 Effect of treatment on the bitter gourd leaf infestation of mite pests during the study period

It is evident that there was varied abundance and population density of mite in the bitter gourd field during the study period after the application of treatments. The plots were sprayed thrice at 15 days interval. The counting was made after each spray. From the table 2, it is evident that the best control was obtained after the first spray followed by second and third spray. In case of the first spray, the lowest population was found from *Trichoderma* (1.3 per leaf) which is significantly different from any other treatments of the experiment. After *Trichoderma,* the lowest mite population was found from neem oil (2.67 per leaf) followed by Spinomax (4.0 per leaf), Biomax (4.34 per leaf), mehogany leaf extract (5.34 per leaf) and Ripcord (6 per leaf). Though numerically differed, there was no significant difference between T_3 and T_6, T_1 and T_3 (Table 2). However, the highest mite population was found in control which was significantly different from other treatments.

In case of second spray, the lowest population was found from *Trichoderma* (2.54 per leaf) which was significantly similar with neem oil (3.0 per leaf). After *Trichoderma* and neem oil, the lowest mite population was found from followed by Biomax (5.0 per leaf), Spinomax (5.34 per leaf), mehogany leaf extract (6.14 per leaf) and Ripcord (7.33 per leaf). There was no significant difference between T_1, T_3 and T_6, T_4 and T_6, T_4 and T_7. However, the highest (8.67 per leaf) mite population was found in control.

In case of third spray, the lowest population was found from *Trichoderma* (4.2 per leaf) which was significantly similar with neem oil (4.31 per leaf). After *Trichoderma* and neem oil, the lowest mite population was found from followed by Biomax (6.67 per leaf), Spinomax (7.0 per leaf), Mehogany leaf extract (8.33 per leaf) and Ripcord (8.62 per leaf) (Table 2). There was no significant difference between T_1 and T_3 and T_1 and T_6, T_4 and T_7. However, the highest (9.0 per leaf) mite population was found in control.

Overuse of synthetic insecticide such as Ripcord might lose its effectiveness against mite pest. Since the agrochemical industry began, insecticides and acaricides have played an increasingly important role in crop protection. However, many species of insects and mites have shown an ability to develop resistance to the pesticides used against them. In our study the lowest effect from treatments except control was found from ripcord that we can relate with the trend of resistance development by mite against synthetic pesticide. Our study shows conformity with [25]. They also found ripcord resistance in whitefly (*Bamisia tabaci*). In their experiment, they found that all the *B. tabaci* field strains exhibited resistance to cypermethrin and susceptibility to chlorpyriphos and endosulfan.

Table 2. Effect of treatment on the bitter gourd leaf infested by mite pests during the study period

Treatment	**No. of Mites (Nymph and adult/ Leaf)**		
	First Spray	**Second Spray**	**Third Spray**
Spinomax (T_1)	4.0 d	5.34 c	7.0 bc
Neem oil (T_2)	2.67 e	3.0 d	4.31 d
Biomax (T_3)	4.34 cd	5.0 c	6.67 c
Ripcord (T_4)	6.0 b	7.33 ab	8.62 a
Trichoderma (T_5)	1.3 f	2.54 d	4.2 d
Mehogany Leaf extract (T_6)	5.34 bc	6.10 bc	8.33 ab
Control (T_7)	7.34 a	8.67 a	9.0 a
CV (%)	**15.05**	**20.68**	**12.95**
Standard Error (SE)	**0.38**	**0.64**	**0.51**

[In a column, means having a similar letter(s) are statistically identical at 0.05 level of probability]

3.3 Effect of treatments on the total number of bitter gourd fruit during the study period

It has been seen that maximum number (29.33) of bitter gourd fruits was found from *Trichoderma* (T_5) which was statistically significant from any other treatments of the study. After *Trichoderma,* the maximum number (27.66) of fruit was obtained from neem oil (T_2) followed by Biomax (T_3) from which we obtained 26 fruits per plant, Spinomax (T_1) from which we obtained 25.33 fruits per plant, Ripcord (T_4) from which we obtained 24.3 fruits per plant and Mehogany Leaf extract (T_6) from which we obtained 23.67 fruits per plant (Table 3). However, the lowest number (20.67) of fruits obtained from Control (T_7) which was significantly different from any other treatments. Furthermore, there were no statistical variations between T_4 and T_6, T_4 and T_1, T_1 and T_3. *Trichoderma* found to be effective against mite pests. Overall, fungal disease in insects is common and widespread. There are more than 700 species of entomopathogenic fungi currently known. Entomopathogenic fungi infect their victims through the cuticle, enter them, and spread throughout the body. After the fungus has killed the host, it can grow out of the cadaver and generate new spores, increasing the likelihood of others becoming infected. *Trichoderma* spp. have long been employed as antifungal agents against a variety of pests and as plant growth promoters. Biocontrol effects of these fungi include mycoparasitism, spatial and nutritional competition, antibiosis by enzymes and secondary metabolites, and stimulation of the plant defense system [26]. The biochemical investigation of the *Trichoderma* treated adult cotton aphid indicated different quantitative changes in the total soluble protein, transaminase enzymes, and carbohydrates hydrolyzing enzymes relative activities as compared to the untreated ones, according to [27]. In addition, histological and ultrastructural examinations revealed a variety of changes and malformations in the treated adult *A. gossypii* body and tissue, as well as the establishment and colonization of *Trichoderma* hamatum within the insect tissues.

Table 3. Effect of treatments on the total number of bitter gourd fruit during the study period

Treatment	**Total number of fruits per plant**	**Increase over control (%)**
Spinomax (T_1)	25.33 cd	22.00
Neem oil (T_2)	27.66 b	33.81
Biomax (T_3)	26.0 c	25.78
Ripcord (T_4)	24.3 de	17.56
Trichoderma (T_5)	29.33 a	41.89
Mehogany Leaf extract (T_6)	23.67 e	14.51
Control (T_7)	20.67 f	-
CV (%)	**2.28**	-
Standard Error (SE)	**0.47**	-

[In a column, means having a similar letter(s) are statistically identical at 0.05 level of probability]

3.4 Effect of treatments on the healthy and infested bitter gourd fruit during the study period

Table 4 shows that there was significant variation between the different treatments. The highest number of healthy fruits (27.66 fruits per plant) was obtained from *Trichoderma* (T_5) which was statistically different from any other treatments and *Trichoderma* (T_5) shows 72.87% increase in healthy fruit yield compared to control. The number of healthy bitter gourd fruits (25.33 fruits per plant) by neem oil (T_2) which was also statistically different from other treatments and showed 58.31% increase in healthy fruit yield compared to control. However, there was no statistically significant variation between Spinomax (T_1) and Biomax (T_3) but numerically better yield was obtained from Biomax (23.66 fruits per plant) which showed 47.87% higher result than control where Spinomax (22.67 fruits per plant) showed 41.68% increase over control (Table 4). Similarly, there was no statistical variation between Ripcord (T_4) and mehogany leaf extract (T_6) rather numerically differed from each other. Ripcord (21.33 fruits per plant) showed 33.31% increase over control treatment whereas mehogany leaf extract (20.33 fruits per plant) showed 27.06% increase over control treatment. However, the lowest healthy fruit yield (16 fruits per plant) was obtained from the control treatment where no insecticide was applied.

On the other hand, there was also significant variation between the obtained infested bitter gourd fruits from different treatments. The lowest number of infested fruits (1.67 fruits per plant) was obtained from *Trichoderma* (T_5) which showed 71.45% decrease over control. Further, the number of infested fruits was observed in following order, neem oil (T_2), Biomax (T_3), Spinomax (T_1), Ripcord (T_4), mehogany leaf extract (T_6) and untreated. No statistical variations found among the T_2, T_3 and T_5 as well as T_1, T_2, T_3 and T_4. Numerically, neem oil and Biomax showed 50% decrease over untreated field followed by Spinomax, Ripcord, mehogany leaf extract which recorded 42.91%, 35.62% and 28.54% decrease in infested fruit number over control. However, the highest infested bitter gourd fruits (4.66 fruits per plant) were obtained from control which was statistically different from all other treatments of the current experiment. Overall, bitter gourd plots exhibited lower level of infestation. This might due to the fact that generally bitter gourd plants are less susceptible to mite pests. [28] reported that bitter (pentanorcucurbitacin A and B, momordicine II and IV) in bitter gourd make the crop somewhat resistance against insect pests. Our findings show conformity with their finding.

Table 4. Effect of biorational approaches on the number of healthy bitter gourd fruit during the study period

Treatment	Number of healthy fruits per plant	Increase over control (%)	Number of infested fruits per plant	Decrease over control (%)
Spinomax (T_1)	22.67 c	41.68	2.66 bc	42.91
Neem oil (T_2)	25.33 b	58.31	2.33 cd	50.00
Biomax (T_3)	23.66 c	47.87	2.33 cd	50.00
Ripcord (T_4)	21.33 d	33.31	3.0 bc	35.62
Trichoderma (T_5)	27.66 a	72.87	1.67 d	71.45
Mehogany Leaf extract (T_6)	20.33 d	27.06	3.33 b	28.54
Control (T_7)	16.0 e	-	4.66 a	-
CV (%)	**3.08**		**19.47**	
Standard Error (SE)	**0.39**		**0.32**	

[In a column, means having a similar letter(s) are statistically identical at 0.05 level of probability]

3.5 Effect of treatments on the yield of bitter gourd fruit during the study period

The highest yield (28.833 t/ha) was obtained from T_5 (*Trichoderma)* that was significant from any other treatments of the present study and obtained 31.04% more yield than control treatment. The yield was then followed by neem oil (T_2) and Biomax (T_3) which observed 27.333 and 26.333 t/ha respectively and no significant variation was observed between these two treatments (Table 5). The lowest yield (22 t/ha) comes from the control treatment which was statistically similar with the result obtained from mehogany leaf extract (T_6). Mehogany leaf extract showed only 4.5% increase in yield over control. Overall, the *Trichoderma* (T_5) showed the best effect in every case. This may be attributed to holistic approach of eco-friendly management tactics.

Table 5. Effect of biorational approaches on the yield of bitter gourd fruit during the study period

Treatment	Yield (t/ha)	Increase Over Control (%)
Spinomax (T_1)	25.000 c	13.63
Neem oil (T_2)	27.333 b	24.22
Biomax (T_3)	26.333 b	19.68
Ripcord (T_4)	23.557 d	7.04
Trichoderma (T_5)	28.833 a	31.04
Mehogany Leaf extract (T_6)	22.993 de	4.50
Control (T_7)	22.000 e	-
CV (%)	**2.81**	-
Standard Error (SE)	**0.41**	-

[In a column, means having a similar letter(s) are statistically identical at 0.05 level of probability]

4. CONCLUSION

The effectiveness of several biorational techniques against the two-spotted spider mite *T. urticae*, which infested bitter gourd, was tested in the field. *Trichoderma* sp. at 200 mg/L of water was shown to be superior to the other treatments in terms of mite population reduction, although it was followed with neem oil at 5 ml/L of water at a 15-days interval. In bitter gourd, where the pest population was significantly larger, the treatment with Ripcord at 1 ml/L of water was the least effective in lowering the mite population. *Trichoderma* spp. at 200 mg/L of water and neem oil at 5 ml/L of water at 15-days intervals were found to be efficient against *T. urticae* in the bitter gourd field, out of various biorational techniques examined.

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