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ORIGINAL ARTICLE

Bio-pesticides for the Control of Two–spotted Spider Mite, *Tetranychus urticae* Koch, in Strawberry Cultivations

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Abstract

Strawberry is cultivated in polytunnels and open field cultivations in *Nuwara Eliya* district. Recently, a severe infestation of the Two Spotted Spider Mite (TSSM) in strawberry was reported and the farmers observed Department of Agriculture (DOA) recommended insecticide Abamectin was ineffective. Therefore, two Bio-pesticides; M-Impact and Hort-Impact were tested in three locations of Jagro Farms Ltd, *Nuwara Eliya* to control TSSM. Two rates of Bio-pesticides were tested in nursery, vegetative and fruiting stages in a randomized complete block design. Results showed that the average percentage mortality of TSSM at day after each application of Hort-Impact was 95.4 for both concentrations. M-Impact resulted an average mortality of 98.7% and 97.9% for 1.0 mLL^{-1} and 0.5 mLL^{-1}, concentrations respectively during the vegetative stage at *Meepilimana*. However, at *Radella*, during the nursery stage, average mortalities were 98.4% and 98.3% for 1.0 mLL^{-1} concentration and 95.8% and 97.7% for 0.5 mLL^{-1} concentration for Hort-impact and M-impact, respectively. The DOA recommended insecticide Abamectin at 1.0 mLL^{-1} concentration resulted a mean TSSM count >200 at the end of the experiment and was significantly higher than Hort-Impact and M-Impact treatments. The TSSM counts were 52 and 45 and 80 and 36 for concentrations 1.0 mLL^{-1} and 0.5 mLL^{-1} respectively at *Meepilimana* and a similar trend was observed at *Radella*. It is therefore concluded that both Bio-pesticides at both concentrations were effective in controlling TSSM in strawberry. However, M-Impact cannot be used at fruiting stage as it imparts a bitter taste for the fruits at *Welimada* site.

Keywords: *Abamectin, Average mortality, Hort-Impact, M-Impact, Strawberry*

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1. Introduction

Strawberry is a low-growing, herbaceous perennial plant with a fibrous root system and a crown which arise from basal leaves. Considering the time of flower bud initiation and thereafter fruiting, two main types of strawberry plants are there. Short-day types initiate flower buds when days are short (less than 14 h a day). Day-neutral types initiate flowers season-long within certain temperature ranges. They have shallow root systems, which result in sensitivity to deficient or excess water and high salt levels in the soil. According to FAO (2012), the main producers of strawberries in 2012 were the United States, Mexico, Turkey, Spain, Egypt, South Korea, Japan, Russia, Germany, and Poland. In Sri Lanka, few entrepreneurs grow strawberries in the polytunnel and open fields but are restricted to *Nuwara Eliya* district under a cool climate.

Several insect and mite pests can have detrimental effects on Strawberry production (Liburd and Rhodes, 2003; Mossler 2012). Among them leaf pests can be divided into two main groups, sucking pests, and chewing pests. Sucking pests include Two-spotted spider mites (*Tetranichus urticae* Koch) (Plate 1a), cyclamen mites, chilli thrips, aphids, whiteflies, potato leafhoppers and spittlebugs. Injury to leaves (Plate 1b) reduces the plants' ability to photosynthesize, which can reduce the quality and quantity of fruit produced. In perennial production, this injury can affect the yield the following season also (Childers 2003).

Plate 1 (a): Two Spotted Spider Mite

Plate 1(b): Two Spotted Spider Mite damage to Strawberry plants

The *Tetranichus urticae* Koch (TSSM) is a major pest, wherever strawberry plants are grown. Others are major pests in certain regions or under specific production systems (Flint 2012). TSSM are oval-shaped and tiny. Its life cycle progresses through five stages: egg, six-legged larvae, protonymph, deutonymph and adult and takes 19 days to complete. The adults are 0.5 mm in length, which is about the size of a period in 12-point font. They are light greenish yellow in colour with two large dark spots on their abdomens. However, Brown, red, orange, and darker green forms also occur. The eggs are spherical and clear to tan in colour. The eggs and all stages of mite are usually found on the underside of the leaves (Mitchell 2003). Females are larger and rounder than males. The optimum condition for development is found to be high temperatures (Up to 38 °C) and low humidity (Krantz 1978; White and Liburd 2005).

Pesticide resistance is an issue when controlling many agricultural pests with aid of chemicals. The two-spotted spider mite is a notorious example as it has been proven capable of developing resistance to more than 92 active ingredients in pesticides (Neuman 2019). In addition, miticide/acaricide applications might also affect the beneficial arthropods population, TSSM's natural enemies, and therefore "foster" spider mite outbreaks. The modes of action are also very important as spider mites can develop resistance to miticides quickly due to their short life cycle. Abamectin miticide has been the most effective and most used mite control agent since 1992, but it has failed to provide satisfactory mite control on some farms since 1999 (Price et al. 2002). In Sri Lanka Abamectin is the miticide recommended by the Department of Agriculture (DOA) for all types of mites but reported to be ineffective for TSSM even with biweekly application at recommended concentrations (personal communication, with strawberry farmers, *Nuwara Eliya*). In the present study two imported Bio-pesticides; Hort-Impact and M-Impact were evaluated to control TSSM in Strawberries under polytunnel cultivations at Jagro Farms LTD, *Nuwara Eliya*.

2. Materials and methods

Two bio-pesticides namely Hort-Impact and M-Impact along with a department of agriculture recommended acaricide, Abamectin, were evaluated for the control of spider mites in strawberry cultivations in polytunnels of Jagro strawberry farm at *Meepilimana*, *Welimada* and *Radella* in *Nuwara Eliya* District. M-Impact and Hort-Impact are safe for the environment and free of chemical residues. They are effective to control sucking insects like mealybugs and mites. The constituents in the two biopesticides tested were as follows (Table 1).

Table 1: Chemical analysis of the two Bio-pesticides

Ingredient	Percentage composition	
	M-Impact	**Hort-Impact**
Medicinal plant (neem) extract	20	50
Essential oil, (Cinnamon oil + Clove oil)	30	-
Wetting Agent	-	10
Solvent	-	40
Adjuvant	10	-
Filler	40	-

Strawberry plants are planted in compost filled bags of 60 x 30 cm size arranged along the length of the polytunnel and such rows are spaced at 60 cm apart. Strawberries were set about 30 cm apart and four plants were planted per compost bag and each treatment consisted of 12 bags. The recommended dose of NPK fertilizer mixture (150:100:120 kgha^{-1}) was used and applied through fertigation once a week interval throughout the crop growth. Natural infestation of spider mites was allowed and when the leaves showed signs of stress the treatments were imposed.

Plate 2: Strawberry cultivation in Jagro Farm, *Meepilimana*, *Nuwara Eliya*

The crop at *Meepilimana* was at its vegetative growth and *Welimada* the crop was at the fruiting stage, while plants at *Radalla* are at the Nursery stage at the observation of infestations. In all three locations, two concentrations; 1.0 mLL^{-1} and 0.5 mLL^{-1} dissolved in distilled water, each of Hort-Impact and M-Impact, was used and sprayed at two weekly intervals. Department of Agriculture recommended Abamectin at the rate of 1.0 mLL^{-1} was also sprayed at two weekly intervals and a control sprayed with distilled water was also used for contrasting. Experiments were replicated three times at each location. The day before the spraying, the number of adult insects (TSS) were counted on 10 marked leaves on nursery plants and 20 marked leaves on vegetative and bearing plants. On the following day, the counts were taken on surviving mites on same leaves. However, the experiment at *Welimada*, where the crop was at the fruiting stage was abandoned due to the bitter taste of strawberry fruits after application of M-Impact at both concentrations.

Statistical analysis: The design of the experiment was a Randomized Complete Block Design. All treatments except the control gave an average control of TSSM of over 95% (range 95.4–98.6%) (Table 2 and 3) of spider mite in both locations and almost all spraying rounds, hence statistical analysis was not carried out. However, statistical analysis was carried out in experiments at *Radella* and *Meepilimana* on the number of live TSSM in 10 leaves and 20 leaves, respectively at the onset and end of the experiment to find out the most effective treatment.

Results and discussion

Results of the experiments conducted at the Jagro Farms LTD, *Nuwara Eliya* were presented in Tables 1, 2 and 3 and discussed on growth stage basis. The strawberry crop was affected by many pests, globally. In strawberry cultivations, in Sri Lanka, the above pests are not recorded frequently but TSSM is very common. Strand (2008) also indicated that some pests such as the TSSM are major pests, wherever strawberry plants are grown. Butcher et al. 1987 indicated TSSM appears in strawberry crops soon after planting and population, if unchecked, increase for 3–4 months. Then population rapidly declines over 1–2 weeks, such that only a few diapausing females remain. Therefore, the cheapest way to control pests is by monitoring them in the field and applying least harmful insecticides at the earliest need.

Vegetative growth stage of strawberry at Meepilimana

The effect of these two bio-pesticides in strawberry cultivation in the vegetative stage at Jagro Farm *Meepilimana* is presented in Table 1. The results showed that both chemicals at both concentrations are very effective in TSSM control giving more than 95% control though the higher rates (>1.0 mLL^{-1}) were little more effective. Between the two chemicals, M-Impact was more effective (average control of 98%) than Horti-Impact (average control 95%), which compound had only Azadiractin. Even though, there was a high percentage of TSSM control in all the treatments, the TSSM counts were high at day before each spraying in M-impact treatments and was low in Hort-Impact treatments at the start of the experiment (August). Data also showed that the population of mites were increased during the months of September and October in spite of spraying schedule of two-week interval in all four treatments, which could be attributed to low humidity (less than 40%) and high minimum and maximum temperatures of 10 °C and 67 °C respectively experienced at *Meeplimana* (https://weatherspark.com/). Krantz (1978) and White and Liburd (2005) also indicated that optimum conditions required for the development of TSSM in strawberry cultivations are high temperature and low humidity. In global literature, the indications are that survival, developmental time, and reproduction of TSSM are mostly influenced by temperature (Helle and Sabelis 1985; Thomas 2001), under hot, dry (Helle and Sabelis 1985), dusty condition (Guerena and Sullivan 2003) and humidity (Sabelis 1981). White and Liburd (2005) surmised development from egg to mature adult takes an average of 19 days, although this time period can be as short as five (05) days depending on soil moisture and temperature and the optimal conditions for development are high temperatures (up to 38 °C) and low humidity

Further, DOA recommended Abamectin resulted very poor motility and the TSSM populations were gradually increased with the biweekly spraying, probably due to resistance development. Stand (2008) indicated that repeated use of one insecticide develops genetic resistance by the insect population and due to cross-resistance, they will become resistant to other similar insecticides. Childers (2003) indicated that thorough spray coverage and rotating mode of action is very important in spider mite control.

Fruiting stage of strawberry at Welimada

Experiment conducted at Welimada site was at the fruiting stage and a high infestation of TSSM (311 TSSM adults/10 leaves] at the onset of the experiment was observed (data not presented). This could be attributed to generally a higher ambient temperature in Welimada. The Welimada site belongs to the Intermediate zone

(IU_3) whereas Meepilimana site is in the wet Zone (WU_3) of Sri Lanka, where ambient temperatures are generally low. Krantz (1978) and White and Liburd (2005) indicated that optimum condition required for the development of TSSM was high temperature and low humidity. Further, Zou et al (2018) indicated the maximum fecundity (81.5 eggs per female) was at 28 °C and the intrinsic rate of increase (r_m) was highest (0.25) at 32 °C. However, the experiment was forced to abandoned due to M-impact treatments imparting sour taste to strawberry fruits and lost the demand in the markets.

Nursery stage of strawberry at Radella

The nursery stage experiment was conducted at *Radella*, and only seven weeks data (Table 2) was taken as the plants were ready for transplanting. In this experiment, the higher rate of application (1.0 mLL^{-1}) of both bio-pesticides resulted a better control (average mortality 98.3%) of TSSM than the lower concentrations (average mortality 96.7%). However, between bio-pesticides, M-Impact treatments resulted better control (average mortality 98%) than the Hort-Impact treatments (average mortality 97.1%). Data also shows that DOA recommended Abamectin is not effective enough to control TSSM in nursery stage too. However, the TSSM population count gradually increased with two weeks spraying schedule with Abamectin, which could be attributed probably to the development of resistance in TSSM. Stand (2008) indicated that repeated use of one insecticide develop genetic resistance by the insect population and due to cross-resistance, they will become resistant to other similar insecticide. Abamectin has been extensively used for control of TSSM in cotton over the past decade in the Midsouth of Lousiana state in America. Recently, growers have observed field-control failures reduced efficacy and shortened residual control, indicating a possible issue with resistance development to Abamectin (Sebe Brown et al. 2017).

DOA has recommended less harmful Abamectin for the control of mites in general but not specifically recommended for TSSM. Moreover, the strawberries produced are earmarked for the export market and therefore use of strong inorganic insecticides was not recommended because of residual insecticide in fruits reaching higher than the International MRL limits. Therefore M-Impact at the rate of 0.5 mLL^{-1} can be used effectively at the nursery stage for better control (97.7%) though Hort-Impact treatment also gave comparable control (95.8%).

Price et al., (2002) surmised since about 1992, Abamectin miticide has been the most effective and most used mite control agent, but it has failed to provide satisfactory mite control on some farms since 1999. Data from their experiments show a ten-fold resistance to Abamectin exists in spider mites from affected farms, when compared to spider mites taken from strawberry two years earlier and held in a laboratory colony. Strawberry farmers in the region, as a result, are adopting wider use of biological control of spider mite (Price et al., 2002). However, no resistance build up was observed in farms when Abamectin is applied less than six applications per year (Campos et al. 1995).

Further, control treatments in both locations where distilled water was sprayed, the TSSM population was increased tremendously reaching uncountable numbers (>200) in 2–4 weeks time. This showed very high ability to reproduce, if no proper control was practiced and therefore strawberry leaves can get brown in colour and die due to sucking of cell sap.

Data presented in table 3 indicated that DOA recommended Abamectin did not have effect and was comparable to the water sprayed control treatment. Both bio-pesticides on the other hand were effective to control spider mites and significantly low count was observed at the end of the treatment period except Hort-impact at 0.5 mLL^{-1} treatment at *Radella*, where there was no significant difference in insect count. The best control of spider mites was observed in Hort-impact at the rate of 1.0 mLL^{-1} followed by 1.0 mLL^{-1} M-impact at *Meepilimana* and *Radella*. Hort-Impact at 0.5 mLL^{-1} treatment was ineffective in controlling TSSM though M-Impact treatment under same concentration was effective in the control of spider mites at the *Radella* site, which could be attributed to the three chemical ingredients other than azadirachtin which were involved in M-impact bio-pesticide. Even though the pesticide M-Impact was effective at low concentration, it cannot be used at reproductive stage of the crop because of bitter taste it imparts on fruits.

3. Conclusions

The two bio-pesticides, Hort-Impact and M-Impact were equally good at the rate of 1.0 mLL^{-1} treatments for the control of TSSM at both sites though the Hort-Impact was the best. The recommended Abamectin was not effective in controlling TSSM in strawberry cultivations. Further, M-Impact at both concentrations cannot be used in strawberry cultivations at fruiting stage as it imparts a bitter taste for the fruits.

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Conflicts of interest: The authors declare that there are no conflicts of interest regarding the publication of this paper.

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Table 1: Percentage control of TSSM at vegetative satge of strawberry at Meepilimana (Mean n = 3) Effect of two concentrations of two biopesticides on spider mites in the vegetative growth stage of Strawberry at Meepilimana (Average of 3 replicates).

Date of Spraying	**Rate of Application (mLL^{-1})**	**Hort-Impact** Spraying Before	After	**% Control**	**M-Impact** Spraying Before	After	**% Control**	**Abamectin** Spraying Before	After
		Mites	Mites		Mites	Mites		Mites	Mites
12-Aug	1.0	48	2	95.8	133	0	100	96	107
27-Aug		94	3	96.8	150	2	98.7	85	89
6-Sep		159	6	96.2	87	0	100.0	103	105
14-Sep		328	13	96.0	110	1	99.1	92	98
26-Sep		418	13	96.9	323	2	99.4	144	145
8-Oct		228	11	95.2	397	1	99.7	137	143
15-Oct		162	15	90.7	100	2	98.0	>200	>200
24-Oct		130	2	98.5	89	0	100.0	>200	>200
31-Oct		28	0	100.0	41	1	97.6	>200	>200
7-Nov		77	3	96.1	29	2	93.1	>200	>200
	Average			***95.4***			***98.6***	**Control (Distilled water)**	
12-Aug	0.5	108	5	95.4	121	0	100	73	90
27-Aug		31	1	96.8	30	1	96.7	101	127
6-Sep		66	0	100.0	116	1	99.1	157	179
14-Sep		542	18	96.7	229	0	100.0	>200	>200
26-Sep		240	3	98.8	414	12	97.1	>200	>200
8-Oct		289	43	85.1	106	0	100.0	>200	>200
15-Oct		227	10	95.6	196	10	94.9	>200	>200
24-Oct		102	7	93.1	126	3	97.6	>200	>200
31-Oct		27	0	100.0	36	2	94.4	>200	>200
7-Nov		80	4	93.0	86	1	98.8	>200	>200
	Average			***95.4***			***97.9***		

Table 2. Percentage control of TSSM at nursery stage of strawberry at Radella Effect of two concentrations of two bio pesticides on spider mites in nursery stage of Strawberry at Radella (Average of 3 replicates).

Date of Spraying	Rate of Application	Hort-Impact		% Control	M-Impact		% Control	Abamectin	
		Spider mite count			Spider mite count			Spider mites count	
		Before Spraying	After spraying		Before Spraying	After spraying		Before spraying	After spraying
		Mites	Mites (live)		Mites	Mites (live)		Mites	Mites (live)
11-Aug	1.0 ml/1	152	4	97.4	147	2	98.6	96	107
25-Aug		46	0	100.0	103	0	100.0	103	105
8-Sep		21	0	100	59	0	100	123	109
5-Oct		137	1	99.3	13	0	100	144	145
24-Oct		62	1	98.4	69	0	100.0	137	143
10-Nov		86	3	96.5	88	7	92.0	>200	>200
23-Nov		81	2	97.5	83	2	97.6	>200	>200
	Average			***98.4***			***98.3***	**Control (Distilled water)**	
11-Aug	0.5 ml/1	65	4	93.8	99	4	96.0	73	90
25-Aug		214	10	95.3	396	3	99.2	157	179
8-Sep		252	4	98.4	227	3	98.7	>200	>200
5-Oct		26	2	92.3	201	0	100	>200	>200
24-Oct		66	1	98.5	95	1	98.9	>200	>200
10-Nov		81	3	96.3	63	2	96.8	>200	>200
23-Nov		102	4	96.1	69	4	94.2	>200	>200
	Average			***95.8***			***97.7***		

Table 3: Living spider mite counts at the vegetative stage of strawberry plants at Meepilimana and at nursery stage at Radellla (Average count from 10 leaves).

Treatment	**Hort-Impact**		**M - Impact**		**Abamectin**	**Control**
Meepilimana						
Rate of application (mLL^{-1})	1.0	0.5	1.0	0.5	1.0	Unsprayed
Mean No. of living mites (Beginning treatment period)	142 d*	108 c	133 d	121 c	87 b	79 b
Mean No. of living mites (End of treatment period)	52 a	80 b	45 a	36 a	>200 e	>200 e
SED	*23.76*					
Radella						
Rate of application (mLL^{-1})	1.0	0.5	1.0	0.5	1.0	Unsprayed
Mean No. of living mites (Beginning treatment period)	152 c	65 a	147 c	99 b	104 b	73 a
Mean No. of living mites (End of treatment period)	81 a	82 a	83 a	69 a	>200 d	>200 d
SED	*21.15*					

* Mean value denoted by the same letter are not significantly different to each other at 1% probability level