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Direct and indirect effects of leaf extracts of *Vitex* spp. on okra red spider mite *Tetranychus macfarlanei* Baker and Pritchard (Acari: Tetranychidae)

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Abstract

Laboratory study was carried out in the Acarology section of Dept. of Agril. Entomology, University of Agril. Sciences, Bengaluru during the period 2015-2016 to determine direct (mortality) and indirect (repellence and oviposition deterrence) effects of various organic solvent (such as petroleum ether, diethyl ether, ethyl acetate and methanol) extracts from the leaves of *Vitex* spp. namely, *V. altissima*, *V. negundo* (white type), *V. peduncularis* and *V. trifolia* on okra red spider mite, *Tetranychus macfarlanei*. Among the different solvent extracts of *Vitex* spp., methanol extract showed comparatively more acaricidal potential followed by ethyl acetate, diethyl ether and petroleum ether extracts. Repellence caused by methanol leaf extract of *V. trifolia* was highest *i.e.* 76%, while it was moderate (48-49%) with *V. altissima* & *V. negundo* extracts and was lowest (37%) with *V. peduncularis* extract. Maximum mortality of okra spider mites observed was 79% with methanol leaf extract of *Vitex peduncularis* compared to other species of *Vitex* (71 to 77% mortality). On methanol extract treated leaf discs female mites laid relatively less number of eggs in three days period (0.40 to 1.89 eggs) compared to that on water treated control leaf discs (1.83 to 4.64 eggs).

Keywords: *Vitex*, okra mite, repellence, oviposition deterrence, mortality

1. Introduction

Terrestrial plants provide potential alternatives to currently used insect control tools because they constitute a rich source of bioactive chemicals. These plant derived compounds are active against a limited number of species and are potentially suitable for use in integrated pest management. There has been a long history of use of botanical preparations for the protection of field crops and stored products (Berger, 1994) ^[1]. In recent times, focus on plant research for diverse benefits has increased all over the world and evidences have been collected to show the immense potential of medicinal plants used in traditional systems (Meena *et al.*, 2010) ^[7]. Botanical insecticides are extracted from various plant parts (stems, seeds, roots, leaves and flower heads) of different plant species. These insecticides are known for having a broad spectrum of activity, being easy to process and use/apply, having a short residual activity and for not accumulating in the environment or in the fatty tissues of warm blooded animals in view of their innate biodegradability (Philip and Robert, 1998) ^[11]. Botanical pesticides, if sufficiently exploited, can surely play a greater role in reducing pollution, health risks *etc.*, apart from reducing crop losses due to pests.

The botanical genus, *Vitex* of family Lamiaceae comprises of approximately 270 known species of shrubs and trees within tropical and sub-tropical regions, although few species may be found in temperate zones (<http://en.wikipedia.org/wiki/vitex>). Several *Vitex* species are folk remedies to treat diarrhoea, gastrointestinal infections; with antimalarial, antimicrobial and antifungal properties have been reported for *V. gaumeri*, *V. agnus-castus* and *V. negundo*, respectively (Zamani *et al.*, 2012; Keerti & Kumar Padma, 2012) ^[17, 6]. Thus, *Vitex* is an important medicinal plant found throughout India and all parts of this plant are used in Ayurvedic and Unani systems of medicine, extracts from leaves and roots are more important in the field of medicine and drugs. Leaves are widely used for rheumatism & inflammations of joints and also reported to have insecticidal properties (Sumathi, 2011; Tandon *et al.*, 2008) ^[15, 16]. Species of *Vitex* such as *Vitex altissima*, *Vitex negundo*, *Vitex peduncularis* and *Vitex trifolia* known to possess prime insecticidal properties were investigated for their direct

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(mortality) and indirect (repellence and oviposition deterrence) toxic effects on major red spider mite pest *Tetranychus macfarlanei*, which damages okra crop more severely throughout India.

2. Material and Methods

Laboratory study was carried out in the Acarology section of Dept. of Agril. Entomology, University of Agril. Sciences, Bengaluru during 2015-2016.

2.1 Mass culturing of okra red spider mite, *Tetranychus macfarlanei*: Okra leaves infested by red spider mites in the field were collected and further reared on mulberry leaves kept on moist cotton wads in plastic trays in the laboratory. Cultured mites were further used for bioassay studies with treated okra leaves.

2.2 Preparation of Extracts: Four organic solvents with ascending order of polarity index viz. petroleum ether (0.1), diethyl ether (4), ethyl acetate (4.4) and methanol (5.1) were used for extraction from leaves of four species of *Vitex* viz., *Vitex altissima* L., *Vitex negundo* L. (white), *Vitex peduncularis* L. and *Vitex trifolia* L.

Fresh leaves of *Vitex* spp. were collected from the Medicinal and Aromatic plants section of the Division of Horticulture and Botanical Garden of University of Agril. Sciences, Bangalore. Leaves were shade dried and powdered separately using a Waring blender. A two fifty gram of leaf powder was used for hot extraction with respective organic solvent using Soxhlet apparatus for 8-10 hours. The extracts were then concentrated to remove the solvent using Vacuum Flash Evaporator. Thick semi-solid extract thus obtained was stored at 4°C in a refrigerator and used to investigate on acaricidal activity on okra red spider mite under laboratory conditions. After the preliminary assay with reasonable mortality records, relative toxicity of different solvent extracts at 5 to 6 concentrations (2%, 4%, 6/8%, 10%, 12%/14%/15%) was determined.

2.3 Leaf spraying method: Fresh okra leaves were sprayed uniformly with the desired concentration of the extract and were air dried under a ceiling fan. Leaf bits measuring 2.5cm X 2.5cm were prepared from the treated leaves and kept on moist cotton wad in Petri plates. Thirty adult female mites released on each of these leaf bits served as one replication and three such replications were maintained for each concentration of the extract. Leaf bits treated with water (or teepol @ 0.1%, when the extract was not easily miscible with water) served as control.

To ascertain the walk-off response or repellency, the number of individuals which were found drowned in water or got stuck to moist cotton wad was recorded at 3, 6, 9 and 12 hours after release and percent walk-off was computed (Penman *et al.*, 1986) [10]. At each observation, adult mites which were found drowned/entangled, but alive, were again released back onto the respective leaf bit carefully using a fine camel hair brush and the observations were continued.

To determine the mortality, the number of female adults found dead was recorded after 72 hours and percent mortality was computed treatment-wise. Individuals, which were unable to move (moribund) were considered as dead or killed.

Effect on egg-laying, if any on the surviving females in 72 h period was also recorded and computed as the mean number of eggs laid by each female in 3 days period.

The data in percentages were subjected to angular transformation, while the numerical data were subjected to $\sqrt{X+0.5}$ transformations and analysed statistically following the Analysis of Variance technique for Completely Randomized Design and the results were interpreted at five percent level of significance.

3. Results and Discussion

Data corresponding to direct (mortality) and indirect effect (repellence and ovipositional deterrence) of different organic solvent extracts of leaves of different *Vitex* spp. are presented in Table 1-4. Across different organic solvents, as the concentration of the leaf extract increased *i.e.*, 2 to 12% with *Vitex negundo*, 2 to 14% with *Vitex peduncularis* and 2 to 15% with *Vitex trifolia*, gradual increase in both direct (mortality) and indirect (repellence and ovipositional deterrence) effects on okra red spider mite was more evident.

3.1 Walk-off/ Repellence

In 12 hours period, walk-off or repellence of adult mites due to petroleum ether leaf extract of *Vitex* spp. was fairly low, as it ranged from 6 to 16 percent only. Repellence was modest with ethyl acetate extract (20 to 29% walk-off) and diethyl ether extract (31-38% walk-off), while it was appreciable with methanol extract (73% walk-off) at the test concentration of 12%. Thus organic solvent methanol might be more efficient in extracting active principles with repellent activity against okra spider mite (Table 4). Among different *Vitex* spp., methanol extract from *V. peduncularis* recorded the lowest repellence of 37 percent, while repellence of *V. altissima* and *V. negundo* extracts was moderate (with 48-49% repellence) and repellence caused by *V. trifolia* was the highest *i.e.*, 73 percent. Hence repellent activity of *V. trifolia* (methanol leaf extract) against okra red spider mite was more evident compared to other species of *Vitex*. Srinivasa and Sugeetha (1999) [13] reported 56 percent walk-off of *Tetranychus macfarlanei* adults from *V. negundo* methanol leaf extract @ 6% concentration treated okra leaf discs and it is comparable to 48 percent walk-off observed in the present study, when the extract was used at the higher concentration of 10%. But Gopal (2000) [5] reported 50 percent walk-off of *Tetranychus urticae* adults on rose leaves treated with 10% extract concentration. However, repellence of *Oligonychus indicus* on treated sorghum leaves was as high as 69 percent reported by him may be attributed to difference in the mite species. Mehlhorn *et al.*, (2005) [8] also appreciated the repellent effect of crude leaf extract of *V. agnus-castus* for at least 6 hours, when applied on animals and human beings against blood sucking ticks, *Ixodes ricinus* and *Rhipicephalus sanguineus*.

3.2 Mortality

Maximum mortality of okra spider mites observed due to petroleum ether leaf extract of *Vitex* spp. ranged from 9-18% while mortality caused by diethyl ether and ethyl acetate extracts was moderate and more or less similar *i.e.* 36-41 percent and 28-37 percent, respectively. But spider mite mortality due to methanol extract was a maximum of 71.79 percent across different species of *Vitex* (Table 4). But for *Vitex negundo*, no information is available on the biological activity (toxicity) of other species of *Vitex* against insect or mite pests.

According to Nandini (2016) [9], based on median lethal concentration (LC₅₀) values, the descending order of direct toxicity of methanol extracts of *Vitex* spp. to okra spider mite

was; *Vitex altissima* (3.14%) > *Vitex negundo* (6.5%) > *Vitex peduncularis* (7.79%) > *Vitex trifolia* (10.91%). Differential toxicity of methanol extract of *Vitex negundo* across different species of mites, their host plants and doses or concentrations is well documented. 78-80% mortality of *Tetranychus urticae* on French bean leaves @ 5-6% treatment concentration was reported by Chandrashekar (1997) [2] and this is comparable to 77 percent mortality of *T. macfarlanei* adults on treated okra leaf discs in the present study (Table 4). Owing to difference in spider mite species and their host plants, Gopal (2000) [5] recorded 40-58 percent mortality of *T. urticae* on treated rose leaves and 60-100 percent mortality of *Oligonychus indicus* on treated sorghum leaves at 6-10% test concentration. Ranilalitha *et al.* (2015) [12] attributed the higher nymphal mortality of red cotton bug *Dysdercus cingulatus* caused by methanol extract of *Vitex negundo*, due to higher concentration of biologically active principles like alkaloids, terpenoids and phenolic compounds in the extracts and toxicity of these active principles to sap feeding spider mite pest on okra crop in our study was also apparent.

3.3 Ovipositional Deterrence

Another indirect effect *i.e.*, ovipositional deterrence was recorded upto 3 days after treatment. On solvent extract treated leaf discs, female mites laid relatively less number of

eggs *i.e.*, 1.06 to 1.82 eggs. 0.84 to 1.88 eggs, 1.00 to 1.40 eggs and 0.40 to 1.89 eggs in petroleum ether, diethyl ether, ethyl acetate and methanol extract treatments, respectively (Table 5). On water treated or control leaf discs number of eggs laid in 3 day period was comparatively more *i.e.*, 1.8 to 4.6 eggs. Across different species of *Vitex*, oviposition deterrent principle might be present to a similar extent and also all the four solvents used in the study were equally efficient in extracting the deterrent chemical from the leaves. Srinivasa and Gopal (2001) [13] reported oviposition deterrent index (ODI) value of 78 (equivalent to 78% inhibition in egg laying) for *Tetranychus urticae* on rose leaf discs and ODI of 79 (equivalent to 79% inhibition in egg laying) for *Oligonychus indicus* on sorghum leaf discs treated with *Vitex negundo* methanol extract at 8- 10% treatment concentration. The present study ascertained the overall potential biological activity of leaf extracts of *Vitex* spp. against a major spider mite pest *Tetranychus macfarlanei* on okra crop. Potential use of *Vitex negundo* methanol leaf extract (in view of its direct toxic effect) against biting and chewing insect pest, *Spodoptera litura* (Deepthy *et al.*, 2010) [4], sucking insect pest, *Dysdercus cingulatus* (Ranilalitha *et al.*, 2015) [12] and stored product insect pest, *Tribolium castaneum* (Chowdhury *et al.*, 2009) [3] has been documented.

Table 1: Effect of petroleum ether extract of *Vitex* spp. on okra red spider mite, *Tetranychus macfarlanei*

<i>Vitex altissima</i>				<i>Vitex negundo</i> (white)				<i>Vitex peduncularis</i>				<i>Vitex trifolia</i>			
Conc.	Walk-off (%)	Mortality (%)	OD	Conc.	Walk-off (%)	Mortality (%)	OD	Conc.	Walk-off (%)	Mortality (%)	OD	Conc.	Walk-off (%)	Mortality (%)	OD
2%	4.44 (12.00)	4.44 (12.00)	1.79 (1.51)	2%	5.56 (11.13)	11.11 (19.16)	1.59 (1.46)	2%	2.22 (4.99)	3.33 (8.49)	2.43 (1.71)	2%	1.11 (3.51)	4.44 (12.00)	1.49 (1.41)
4%	3.33 (8.49)	5.56 (13.48)	1.52 (1.42)	4%	5.56 (13.48)	8.89 (16.79)	1.90 (1.55)	4%	6.67 (12.13)	10.00 (18.27)	2.47 (1.71)	4%	4.44 (12.00)	7.78 (16.12)	2.36 (1.69)
8%	4.44 (9.98)	10.00 (18.27)	1.98 (1.57)	6%	6.67 (14.96)	8.89 (17.11)	1.29 (1.34)	6%	7.78 (16.12)	11.11 (19.27)	2.00 (1.57)	6%	4.44 (12.00)	7.78 (16.12)	2.10 (1.61)
10%	6.67 (14.64)	12.22 (20.32)	1.71 (1.48)	8%	2.22 (7.01)	7.78 (16.12)	2.14 (1.63)	8%	11.11 (19.43)	12.22 (20.42)	2.10 (1.61)	10%	4.44 (12.00)	8.89 (17.11)	2.22 (1.65)
12%	7.78 (15.80)	12.22 (20.16)	1.06 (1.25)	10%	4.44 (9.65)	8.89 (17.11)	1.99 (1.58)	10%	13.33 (21.32)	17.78 (24.92)	2.51 (1.73)	12%	4.44 (9.65)	6.67 (12.29)	1.65 (1.47)
15%	15.56 (22.70)	17.78 (24.85)	2.22 (1.65)	12%	6.67 (14.64)	11.11 (19.16)	1.80 (1.52)	14%	10.00 (18.01)	15.56 (23.03)	1.82 (1.52)	15%	5.56 (13.48)	8.89 (17.28)	1.46 (1.40)
Control (Water)	0.00 (0.00)	0.00 (0.00)	3.06 (1.88)	Control (Water)	0.00 (0.00)	0.00 (0.00)	2.60 (1.76)	Control (Water)	0.00 (0.00)	0.00 (0.00)	2.32 (1.68)	Control (Water)	0.00 (0.00)	0.00 (0.00)	2.32 (1.68)
CD at P=0.05	(13.8)	(5.43)	(0.05)	CD at P=0.05	(10.43)	(6.92)	(0.09)	CD at P=0.05	(10.38)	(6.91)	(0.10)	CD at P=0.05	(8.20)	(8.12)	(0.06)

Figures in parentheses are angular transformation of walk-off and mortality data; $\sqrt{x+0.5}$ transformation of Oviposition Deterrence (OD) data (Mean number of eggs per female in 3 days period).

Table 2: Effect of diethyl ether extract of *Vitex* spp. on okra red spider mite, *Tetranychus macfarlanei*

<i>Vitex altissima</i>				<i>Vitex negundo</i> (white)				<i>Vitex peduncularis</i>				<i>Vitex trifolia</i>			
Conc.	Walk-off (%)	Mortality (%)	OD	Conc.	Walk-off (%)	Mortality (%)	OD	Conc.	Walk-off (%)	Mortality (%)	OD	Conc.	Walk-off (%)	Mortality (%)	OD
2%	7.78 (16.19)	8.89 (16.79)	1.34 (1.36)	2%	3.33 (10.52)	5.56 (13.48)	1.60 (1.45)	2%	7.78 (16.12)	10.00 (18.27)	1.75 (1.50)	2%	6.67 (14.64)	7.78 (15.80)	2.59 (1.74)
4%	20.00 (26.57)	23.33 (28.64)	2.49 (1.74)	4%	10.00 (17.68)	14.44 (21.17)	2.22 (1.66)	4%	12.22 (20.16)	17.78 (24.80)	1.36 (1.37)	4%	8.89 (17.28)	16.67 (23.86)	1.88 (1.54)
8%	22.22 (28.13)	30.00 (33.19)	1.76 (1.50)	6%	13.33 (21.32)	18.89 (25.62)	2.23 (1.65)	6%	12.22 (19.57)	15.56 (21.99)	0.89 (1.19)	6%	7.78 (15.63)	18.89 (25.04)	2.10 (1.62)
10%	31.11 (33.90)	36.67 (37.22)	1.96 (1.58)	8%	36.67 (37.25)	41.11 (39.86)	2.66 (1.76)	8%	21.11 (27.34)	25.56 (30.29)	1.10 (1.27)	10%	20.00 (26.36)	30.00 (33.12)	2.59 (1.75)
12%	28.89 (32.51)	34.44 (35.90)	1.27 (1.33)	10%	27.78 (31.64)	31.11 (33.78)	2.06 (1.60)	10%	20.00 (26.43)	22.22 (28.02)	0.84 (1.16)	12%	21.11 (27.13)	28.89 (32.33)	2.31 (1.67)
15%	33.33 (35.26)	37.78 (37.91)	1.50 (1.41)	12%	37.78 (37.90)	41.11 (39.86)	2.62 (1.76)	14%	31.11 (33.80)	36.67 (37.14)	1.40 (1.38)	15%	33.33 (34.79)	35.56 (36.11)	2.03 (1.61)
Control (water)	0.00 (0.00)	0.00 (0.00)	1.83 (1.53)	Control (water)	0.00 (0.00)	0.00 (0.00)	1.83 (1.53)	Control (water)	0.00 (0.00)	0.00 (0.00)	1.83 (1.53)	Control (water)	0.00 (0.00)	0.00 (0.00)	1.83 (1.53)
CD at P=0.05	(9.06)	(6.76)	(0.08)	CD at P=0.05	(6.60)	(8.41)	(0.08)	CD at P=0.05	(7.60)	(9.40)	(0.06)	CD at P=0.05	(7.57)	(11.52)	(0.10)

Figures in parentheses are angular transformation of walk-off and mortality data; $\sqrt{x+0.5}$ transformation of Oviposition Deterrence (OD) data (Mean number of eggs per female in 3 days period.).

Table 3: Effect of ethyl acetate extract of *Vitex* spp. on okra red spider mite, *Tetranychus macfarlanei*

<i>Vitex altissima</i>				<i>Vitex negundo</i> (white)				<i>Vitex peduncularis</i>				<i>Vitex trifolia</i>			
Conc.	Walk-off (%)	Mortality (%)	OD	Conc.	Walk-off (%)	Mortality (%)	OD	Conc.	Walk-off (%)	Mortality (%)	OD	Conc.	Walk-off (%)	Mortality (%)	OD
2%	12.22 (19.99)	15.56 (23.13)	1.33 (1.35)	2%	16.67 (22.73)	20.00 (25.71)	1.97 (1.40)	2%	4.44 (7.14)	13.33 (20.76)	2.32 (1.67)	2%	4.44 (12.00)	10.00 (15.46)	2.90 (1.84)
4%	18.89 (25.74)	17.78 (24.80)	1.47 (1.40)	4%	21.11 (26.37)	22.22 (27.12)	1.47 (1.51)	4%	20.00 (26.03)	27.78 (31.47)	3.48 (1.99)	4%	8.89 (17.28)	16.67 (21.55)	2.07 (1.60)
8%	15.56 (21.41)	27.78 (31.68)	1.78 (1.52)	6%	16.67 (23.20)	21.11 (26.43)	1.76 (1.27)	6%	11.11 (19.27)	15.56 (23.03)	1.66 (1.45)	6%	6.67 (12.13)	13.33 (18.62)	1.44 (1.39)
10%	24.44 (29.47)	33.33 (35.25)	1.52 (1.42)	8%	18.89 (24.21)	27.78 (30.40)	1.09 (1.22)	8%	13.33 (21.41)	16.67 (24.03)	1.29 (1.33)	10%	26.67 (29.83)	33.33 (37.91)	1.75 (1.51)
12%	25.56 (30.29)	35.56 (36.59)	1.57 (1.43)	10%	17.78 (24.09)	26.67 (29.77)	1.00 (1.24)	10%	8.89 (17.28)	17.78 (24.92)	1.11 (1.27)	12%	28.89 (30.06)	36.67 (40.87)	2.84 (1.89)
15%	27.78 (31.64)	34.44 (35.84)	1.22 (1.30)	12%	25.56 (28.92)	36.67 (35.11)	0.98 (1.70)	14%	17.78 (23.88)	23.33 (27.69)	1.94 (1.27)	15%	23.33 (28.42)	31.11 (32.12)	1.60 (1.46)
Control (water)	0.00 (0.00)	0.00 (0.00)	2.21 (1.65)	Control (water)	0.00 (0.00)	0.00 (0.00)	2.41 (1.64)	Control (water)	0.00 (0.00)	0.00 (0.00)	2.21 (1.59)	Control (water)	0.00 (0.00)	0.00 (0.00)	2.21 (1.65)
CD at P=0.05	(10.85)	(6.07)	(0.08)	CD at P=0.05	(19.14)	(12.67)	(0.09)	CD at P=0.05	(12.69)	(11.34)	(0.07)	CD at P=0.05	(21.38)	(13.48)	(0.10)

Figures in parentheses are angular transformation of walk-off and mortality data; $\sqrt{x+0.5}$ transformation of Oviposition Deterrence (OD) data (Mean number of eggs per female in 3 days period.)

Table 4: Effect of methanol extract of *Vitex* spp. on okra red spider mite, *Tetranychus macfarlanei*

<i>Vitex altissima</i>				<i>Vitex negundo</i> (white)				<i>Vitex peduncularis</i>				<i>Vitex trifolia</i>			
Conc.	Walk-off (%)	Mortality (%)	OD	Conc.	Walk-off (%)	Mortality (%)	OD	Conc.	Walk-off (%)	Mortality (%)	OD	Conc.	Walk-off (%)	Mortality (%)	OD
2%	20.00 (26.03)	34.44 (35.75)	1.73 (1.45)	2%	6.67 (14.64)	24.44 (29.62)	1.66 (1.46)	2%	0 (0.00)	10.00 (17.68)	2.63 (1.76)	2%	2.22 (4.99)	13.33 (21.14)	4.74 (2.28)
4%	48.89 (44.34)	62.22 (52.17)	0.44 (0.93)	4%	15.56 (22.65)	21.11 (27.29)	0.69 (1.09)	4%	4.44 (9.65)	22.22 (27.96)	2.38 (1.63)	4%	3.33 (8.49)	16.67 (23.91)	2.93 (1.85)
8%	47.78 (43.74)	71.11 (57.64)	1.00 (1.22)	6%	16.67 (23.91)	38.89 (38.55)	0.62 (1.06)	6%	6.67 (14.96)	25.56 (30.17)	2.69 (1.78)	6%	26.67 (31.01)	45.56 (42.45)	4.55 (2.20)
10%	40.00 (38.86)	68.89 (56.52)	1.07 (1.29)	8%	37.78 (37.32)	57.78 (50.05)	0.79 (1.21)	8%	36.67 (36.73)	48.89 (44.33)	2.50 (1.77)	10%	33.33 (33.71)	37.78 (37.90)	2.06 (1.45)
12%	38.89 (38.55)	58.89 (50.14)	0.89 (1.19)	10%	47.78 (43.70)	66.67 (54.87)	0.58 (1.05)	10%	13.33 (21.05)	66.67 (54.80)	2.07 (1.61)	12%	44.44 (41.75)	74.44 (59.63)	1.78 (1.33)
15%	45.56 (42.42)	53.33 (46.91)	1.75 (1.50)	12%	38.89 (38.51)	76.67 (62.19)	0.40 (1.06)	14%	20.00 (26.36)	78.89 (63.90)	1.89 (1.21)	15%	73.33 (59.35)	75.56 (60.37)	2.00 (1.58)
Control (water)	0.00 (0.00)	0.00 (0.00)	4.17 (2.16)	Control (water)	0.00 (0.00)	0.00 (0.00)	2.80 (1.81)	Control (water)	0.00 (0.00)	0.00 (0.00)	2.28 (1.67)	Control (water)	0.00 (0.00)	0.00 (0.00)	4.64 (2.26)
CD at P=0.05	(12.66)	(8.51)	(0.09)	CD at P=0.05	(14.10)	(10.72)	(0.13)	CD at P=0.05	(11.45)	(10.37)	(0.20)	CD at P=0.05	(7.62)	(7.62)	(0.25)

Figures in parentheses are angular transformation of walk-off and mortality data; $\sqrt{x+0.5}$ transformation of Oviposition Deterrence (OD) data (Mean number of eggs per female in 3 days period.)

Table 5: Summarized data indicating direct and indirect effects of *Vitex* on okra red spider mite

Indirect effect (% Repellence)				
Solvents	Petroleum ether	Diethyl ether	Ethyl acetate	Methanol
<i>Vitex altissima</i>	16	33	28	49
<i>Vitex negundo</i> (white)	7	38	26	48
<i>Vitex peduncularis</i>	13	31	20	37
<i>Vitex trifolia</i>	6	33	29	76
Direct effect (% Mortality)				
Solvents	Petroleum ether	Diethyl ether	Ethyl acetate	Methanol
<i>Vitex altissima</i>	18	38	36	71
<i>Vitex negundo</i> (white)	11	41	37	77
<i>Vitex peduncularis</i>	18	37	28	79
<i>Vitex trifolia</i>	9	36	37	76
Indirect effect Oviposition Deterrence (mean number of eggs per female in 3 days period)				
Solvents	Petroleum ether	Diethyl ether	Ethyl acetate	Methanol
<i>Vitex altissima</i>	1.06	1.27	1.2	0.44
<i>Vitex negundo</i> (white)	1.29	1.60	1.0	0.4
<i>Vitex peduncularis</i>	1.82	0.84	1.1	1.89
<i>Vitex trifolia</i>	1.50	1.88	1.4	1.78
Control (water)	2.32-3.06	1.83	2.21-2.41	2.28-4.64

4. Conclusion

Present study revealed direct (mortality) and indirect (repellence and oviposition deterrence) effects of organic solvent leaf extract of *Vitex* spp. against okra red spider mite, *Tetranychus macfarlanei*. Among four different organic solvents, methanol leaf extract was more acaricidal with significant repellent, oviposition deterrent and killing effects. Methanol leaf extract of *V. trifolia* resulted in maximum repellency of 76%, while maximum mortality of 79% was caused by the extract from *V. peduncularis*. Methanol leaf extract of *V. negundo* inhibited spider mite's egg-laying to the maximum extent of 66%. Separation and identification of promising active principle(s) present in *Vitex* extract would enable the development of an acaricide formulation for further use under field conditions.

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6. Reference

1. Berger A. Using natural Pesticides: Current and future perspectives. A report for the Plant Protection Improvement Programme in Swedish University of Agricultural Sciences, Uppasala, 1994.
2. Chandrashekar K. Toxicological investigations on the spider mite, *T. urticae* (Acari: Tetranychidae) infesting French bean using synthetic & botanical pesticides. M.Sc. (Agri.) thesis, University of Agricultural Sciences, Bengaluru, 1997, 89.
3. Chowdhury NY, Islam W, Khalequzzaman M. Insecticidal activities of the leaves of nishinda (*Vitex negundo*: Verbenaceae) against *Tribolium castaneum*. Pakistan Entomology. 2009; 31(1):120-122.
4. Deepthy KB, Sheela MK, Jacob SS, Estelitta. Thomas J. Insecticidal and growth inhibitory action of *Vitex negundo* L. against Asian army worm, *Spodoptera litura* Fab. Journal of Biopesticide. 2010; 3(1):289-295.
5. Gopal TS. Evaluation of certain botanicals against spider mite *Tetranychus urticae* Koch and *Oligonychus indicus* Hirst (Acari: Tetranychidae) and their effects on mite predator, *Amblyseius longispinosus* (Acari: Phytoseiidae). M. Sc. (Agri.) thesis. University of Agricultural Sciences Bengaluru, 2000, 100.
6. Keerti G, Kumar Padma. Evaluation of phytochemical and antimicrobial study of extracts of *Vitex negundo* Linn. Drug Development and Research. 2012; 4(4):192-199.
7. Meena AK, Singh U, Yadav AK, Singh B, Rao MM. Pharmacological and phytochemical evidences for the extracts from plants of the genus *Vitex*: A Review. International Journal of Pharmaceutical and Clinical Research. 2010; 2(1):1-9.
8. Mehlhorn H, Schmahl G, Aschmidt J. Extract of the seeds of the plant *Vitex agnus castus* proven to be highly efficacious as a repellent against ticks, fleas, mosquitoes and biting flies. Parasitology Research. 2005; 95:363-365.
9. Nandini KN. Biological activity of organic solvent extracts of *Vitex* species on red spider mite *Tetranychus macfarlanei* Baker and Pritchard. M.Sc. (Agri.).Thesis, University of Agricultural Sciences, Bengaluru, 2016.
10. Penman DR, Chapman RB, Bowie MK. Direct toxicity

and repellent activity of pyrethroids against *Tetranychus urticae* (Acari: Tetranychidae).Journal of Economic Entomology.1986; 79:1183-1186.

11. Philip GK. Robert AB. Florida Pest Control Association, 1998.
12. Ranilalitha PM, Sukumaran S, Raveendran, Mirthanayagi KA. Evaluation of nymphicidal effect of two indigenous plant extracts on cotton pest, *Dysdercus cingulatus* (Fab.).International Journal of Pure and Applied Zoology. 2015; 3(1):24-30.
13. Srinivasa N, Sugeetha G. Bioeffectiveness of certain botanicals and synthetic pesticides against okra spider mite *Tetranychus macfarlanei*. Journal of Acarology. 1999; 15:1-5.
14. Srinivasa N, Gopal TS. Evaluation of certain botanicals on phytoseiid mite predator *Amblyseius longispinosus* (Evans) (Acari: Phytoseiidae) (Abstract). Proceedings of Symposium on biocontrol based pest management for quality crop protection in the current millennium, Ludhiana, 2001.
15. Sumathi P. Smoke toxic effect of *Vitex negundo* for the control of Malarial vector, *Anopheles stephensi* Liston. Journal of Natural remedies. 2011; 11(2):177-181.
16. Tandon S, Ashutosh K, Mittal AK. Insect growth regulatory activity of *Vitex trifolia* and *Vitex agnus-castus* essential oils against *Spilosoma obliqua*. Plant Fitoterapia. 2008; 79:283-286.
17. Zamani M, Neghab N, Torabian S. Therapeutic effect of *Vitex agnus-castus* in patients with premenstrual syndrome. Acta Medica, Iran. 2012; 50(2):101-106.