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Short Communication

Toxicity of natural and organic insecticidal products to greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood) (Aleyrodidae: Hemiptera)

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Abstract

Lethal toxicity of six natural and organic insecticidal products namely, *Darekastra*, *Dashparni*, fermented buttermilk, fermented cow urine, *Tamarlassi* and vermiwash to second instar nymphs of greenhouse whitefly, *Trialeurodes vaporariorum* was evaluated using leaf dip method of bioassay. The insecticidal products evaluated at six concentrations ranging from 1.25 to 40.00 %proved to be progressively more lethal to second instar nymphs with an increase in concentration resulting in mortality varying from 1.67 to 7.33 % and 43.33 to 62.96 at concentrations of 1.25 and 40.00 %, respectively. *Tamarlassi* proved most toxic against the nymphs. The order of toxicity was Tamarlassi = fermented buttermilk≥ fermented cow urine ≥ Dashparni≥ Darekastra≥ vermiwash.

Key words: Greenhouse whitefly, toxicity, natural products, organic insecticidal products

Greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood) (Hemiptera:Aleyrodidae) is a serious pest under protected environment in temperate regions and in field crops where the summers are warm enough (Byrne and Bellows 1991; Sood and David 2012). It is highly polyphagous and is widely distributed throughout the world. Currently, greenhouse whitefly is known to exist on 859 plant species belonging to 469 genera and 121 families (CABI 2021). The nymphs and adults of *T. vaporariorum* suck the phloem cell sap thereby affecting the plant vigour. Additionally, they secrete honeydew, which facilitates the development of sooty mould interfering with photosynthetic activity. Apart from the whitefly *T. vaporariorum*, aphid *Myzus persicae* (Sulzer), two spotted spider mite *Tetranychus urticae* Koch, russet mite *Aculops lycopersici* (Massee) are of importance under protected environment in Himachal Pradesh (Kashyap *et al*. 2015; Ghongade and Sood 2019; Ghongade and Sood 2021; Sharma *et al*. 2021).

Trialeurodes vaporariorum breeds throughout the year and completes 13 generations under protected environment inflicting losses to the extent of 50 % in different crops (Sood *et al.*2014; Sood *et al.* 2018).The greenhouse whitefly feeds on the abaxial surface of the leaves and remains sessile during immature stages that minimize its contact with the insecticides thereby resulting in frequent insecticidal applications, which leads to pesticide residues on crop produce, killing of non-target organisms and development of resistance in *T. vaporariorum*(Van Lenteren 2000; Sood *et al.*2006; Pilkington *et al.*2010; Pappas *et al.*2013). Nowadays more emphasis is being laid on the use of natural products in plant protection. Natural products are effective in enhancing crop productivity, suppressing growth of insects,are inexpensive,easily biodegradable, have different modes of action and are less harmful to non-target organisms and the environment (Lengai *et al.* 2020). However, research on evaluation of natural and organic insecticidal products is lacking against greenhouse whitefly. Therefore, considering the economic importance of greenhouse whitefly as well as that of natural products (cattle and plant-based) against insect pests, studies were made on evaluating toxicity of natural insecticidal products against second instar nymphs of greenhouse whitefly under laboratory conditions.

Raising stock culture of greenhouse whitefly

Trialeurodes vaporariorum was reared on potted Frenchbean plants raised in a soil-less medium comprising cocopeat, perlite, vermiculite and vermicompost(3:1:1:1). Field-collected population of greenhouse whitefly was used to raise the stock culture under laboratory conditions. The adults were collected with the help of an aspirator and were released in large numbers on 15-day-old Frenchbean plants kept in screened cages (45×45×55 cm) for 24 hours for egg deposition. Thereafter the adults were removed and the plants were shifted to rearing cages for raising to desired developmental stage.

Natural and organic insecticidal products evaluated: Six natural and organic products namely, *Darekastra*, *Dashparni*, fermented buttermilk, fermented cow urine, *Tamarlassi* and vermiwash were evaluated for their lethal toxic effects against 2nd instar nymphs of *T. vaporariorum*. The products were prepared following the standardized methodologies of Department of Organic Agriculture and Natural Farming (Rameshwar *et al.* 2014). The composition of products is being given in Table 1

Each insecticidal product was regarded as 100% stock solution and the test concentrations were further prepared by serial dilution method.

Method of Bioassay

Leaf dip method of bioassay was used for evaluating toxicity of natural and organic insecticidal products against second instar nymphs. For obtaining the cohort of second instar nymphs, Frenchbean plants were exposed to adult whiteflies for 24 hours for egg-laying. The eggs were allowed to develop to 2nd instar nymphs. After 13-14 days, the eggs develop into 2nd instar nymphs. The leaves containing the nymphs were detached and examined under a microscope to validate the stage. The leaves containing the second instar nymphs ($n \geq 30$) were excised into discs (2.5 cm diameter). These leaf discs were dipped in test concentrations of different products ranging at six concentrations of 1.25, 2.50, 5.00, 10.00, 20.00 and 40.00 % for 30 seconds. An untreated check (UTC) was maintained by dipping the leaf discs in water. The discs were air-dried at room temperature and placed on an agar-agar bed (2.00%) in petri plates (5 cm dia.) with abaxial surface facing upwards. Each experiment was replicated three times. These petri plates were placed under controlled conditions at 25±1^{0}C temperature and 70±5 % relative humidity. Observations on mortality of nymphs were recorded upto 72 hours after treatment. The nymphs that shrunk, dried and turned brown after treatment were considered dead. Mortality data recorded upto 72 hours after treatment was taken into consideration for working out per cent mortality. The per cent mortality was corrected as suggested by Abbott (1925) using the following formula:

$$\text{Corrected Mortality (\%)} = \frac{\textit{Mortality in Treatment}\ (\%) - \textit{Mortality in UTC}\ (\%)}{100 - \textit{Mortality in UTC}\ (\%)} \times 100$$

Natural and organic products resulted in mortality of second instar nymphs of *T. vaporariorum* ranging from 2.08 to 47.78, 3.33 to 50.07, 6.00 to 61.11, 5.00 to 54.07, 7.33 to 62.96 and 1.67 to 43.33 % for

Table 1.Natural and organic products evaluated for insecticidal activity

Natural/Organic Product	Composition
Darekastra	*Melia* leaves (5 kg), cow urine (5 L), cow dung (2 kg) and water (20 L)
Dashparni	*Melia* leaves (5 kg), *Ipomea* leaves (2 kg), *Polygonum* leaves (2 kg), *Juglans* leaves (2 kg), *Lantana* leaves (2 kg), *Datura* leaves (2 kg), *Vitex* leaves (2 kg), *Psidium* leaves (2 kg), *Eupatorium* leaves (2 kg), wild *Tagetes* leaves (2 kg), garlic paste (500 gm), red chilli paste (500 gm), turmeric powder (500 gm), dry ginger powder (200 gm), cow urine (10 L), cow dung (2 kg) and water (10 L)
Fermented buttermilk	Prepared from 15-20 days old cultured milk
Fermented cow urine	15 days old cow urine of Indian cow
Tamarlassi	Prepared by keeping the fermented buttermilk in copper pot for 10 to 15 days
Vermiwash	Cow dung (4 kg), biomass (1 kg) and adult earthworms (200-300)

Darekastra, *Dashparni*, fermented buttermilk, fermented cow urine, *Tamarlassi* and vermiwash, respectively,in the concentration range of 1.25 to 40.00 % (Table 2). Nymphal mortality increased significantly with an increase in evaluated concentrations. Also, a relationship was established by working out correlation (r) between concentration and nymphal mortality which resulted in the corresponding 'r' values 0.96, 0.95, 0.96, 0.96, 0.96 and 0.96, respectively, indicating a positive and significant (P=0.05) effect of concentration on bringing about the kill. Based on the mean mortality observed in different products, *Tamarlassi* resulted in maximum mortality and was at par to fermented buttermilk. It was followed by fermented cow urine. Vermiwash resulted in minimum mortality among the second instar nymphs and was preceded by *Darekastra* and *Dashparni*, all being at par to each other. However, the interaction effect among the concentration and natural and organic products was found to be non-significant.

Although the literature on the bioactivity of natural and organic products against insects is limited, there are a few references available on their potential bioactive components responsible for their activity against various insect pests. The insecticidal activity of *Darekastra* comprising *Melia* leaves can be attributed to the biologically active compounds, triterpenoids in *Melia azedarach.* The insecticidal properties of the triterpenoids have been demonstrated on coleopteran, dipteran, and lepidopteran pests under laboratory conditions. (Carpinella *et al.* 2003). Ayil-Gutierrez *et al.* (2018) demonstrated that secondary metabolites from *M.azedarach*, such as coumarin, sterols, and terpenes, reduced pupal viability, resulting in adult deformities in *Spodoptera litura*. Thakur and Sood (2022) found *Darekastra* to be the best-performing bioacaricide for maintaining spider mite population at lower levels when used both as preventive and curative spray under greenhouse conditions. *Dashparni* ark has been known to contain phytochemicals such as saponin, proteins, phenol, steroid and phytosterol having protection activity (Kasarkar *et al*. 2021). Chandel *et al.* (2011) observed that the extracts of *Azadirachta indica* and *Vitex negundo* were more effective against *Bagrada hilaris,* with mortality rates of 80.9 % in nymphs and 74.9 % in adults, respectively. Joshi *et al.* (2021) observed *Dashparni* ark to the be most effective bioformulation against *Agrotis ipsilon* causing maximum mortality among different larval instars.

Bioactive constituents in cow urine affect insect development and survival (Gahukar 2013). Cow urine contains high content of urea which is toxic to most insect pests and can be considered a good means for insect pest management (Kumawat *et al.* 2014).

Table 2. Toxicity of natural and organic products to second instar nymphs of *Trialeurodes vaporariorum*

Concentration (%)	Corrected mortality (%) 72 hours after treatment						Mean
	Darekastra	*Dashparni*	Fermented buttermilk	Fermented cow urine	*Tamarlassi*	Vermiwash	
1.25	2.08(4.82)	3.33(8.61)	6.00(14.09)	5.00(12.92)	7.33(15.36)	1.67(4.31)	4.24(10.02)
2.50	5.59(13.52)	6.67(14.79)	9.33(17.70)	8.00(16.07)	10.67(18.98)	4.89(12.68)	7.52(15.62)
5.00	11.11(19.16)	12.22(20.41)	15.56(23.19)	14.44(22.30)	17.14(24.35)	10.00(18.43)	13.41(21.31)
10.00	21.33(27.48)	24.67(29.69)	29.33(32.77)	25.33(30.19)	32.00(34.41)	20.05(26.51)	25.45(30.18)
20.00	36.67(37.24)	40.00(39.21)	46.67(43.07)	42.22(40.48)	48.89(44.35)	32.22(34.53)	41.11(39.81)
40.00	47.78(43.71)	50.07(45.02)	61.11(51.40)	54.07(47.32)	62.96(52.55)	43.33(41.15)	53.22(46.86)
Mean	20.76(24.32)	22.83(26.29)	28.00(30.37)	24.85(28.21)	29.83(31.67)	18.69(22.93)	

Figures in parentheses are the arc sine transformed values
CD (P=0.05)
Concentration (A) = (2.19)
Product (B) = (2.19)
A × B = NS

Onunkun (2014) reported cow urine + water(1:1) to be the most effective insecticidal solution for the management of brown stink bug, *Aspavia armigera*, green stink bug, *Nezara virudula* and the coreid bug, *Clavigralla tomentosiccolis* in *Amaranthus*. Enrichment of cow urine with various botanicals enhanced the insecticidal property of different plant extracts against sucking pests of cotton (Patel *et al.* 2003) and sesame (Ahirwar *et al.* 2010). Cow urine, *Darekastra* and fermented buttermilk proved to be potent acaricides with deterrent properties against *Tetranychus urticae* (Thakur and Sood 2021).

Cow milk-based fermented products like fermented buttermilk have acidic properties (Gebreselassie *et al.* 2016) and *Tamarlassi* contains acidic buttermilk enriched with copper (Sudeshna 2017). Karthikeyan *et al.* (2005) found buttermilk to cause a 60 % reduction in whitefly infestation in bhendi. Fermented buttermilk and *Tamarlassi* have also been reported to possess acaricidal activity against mites (Thakur and Sood 2019). The coelomic fluid of earthworms in vermiwash is reported to possess insecticidal, antifungal and pesticidal bioactive compounds (Nandana *et al.* 2020). Vermiwash is toxic to the survival of insects (Sayyad 2017). It possesses excellent biopesticidal properties (Nayak *et al.* 2019) and is effective against aphids (Venkateswarlu *et al.* 2011).

Conclusion

The study concludes that the natural and organic insecticidal products assessed had toxic effects against the second instar nymphs of *Trialeurodes vaporariorum*. The order of toxicity was Tamarlassi = fermented buttermilk $\geq$ fermented cow urine $\geq$ Dashparni $\geq$ Darekastra $\geq$ vermiwash. Hence, *Tamarlassi* and vermiwash resulted in the maximum and the minimum mortality of the second instar nymphs, respectively.

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References

Abbott WS. 1925. Method for computing the effectiveness of an insecticide. Journal of Economic Entomology **18**: 265-267.

Ahirwar RK, Gupta MP and Banerjee S. 2010. Field efficacy of natural and indigenous products on sucking pests of sesame. Indian Journal of Natural Products and Resources **1** (2): 221-226.

Ayil-Gutierrez BA, Sanchez-Teyer LF, Vazquez-Flota F, Monforte-Gonzalez M, Tamayo-Ordonez Y, Tamayo-Ordonez MC and Rivera G. 2018. Biological effects of natural products against *Spodoptera* spp. Crop Protection **114:** 195-207.

Byrne DN and Bellows TS 1991. Whitefly biology. Annual Review of Entomology **36**: 431-57.

CABI. 2021. Invasive Species Compendium. *Trialeurodes vaporariorum* (whitefly, greenhouse). *http://www.cabi.org/isc/datasheet/54660* [25th July 2021].

Carpinella C, Defago M, Valladares and Palacios SM. 2003. Antifeedant and insecticide properties of a limonoid from *Melia azedarach* (Meliaceae) with potential use for pest management. Journal of Agricultural and Food Chemistry **51** (2): 369-374.

Chandel BS, Vajpai S and Singh V. 2011. Bioefficacy of plant products against painted bug, *Bagrada cruciferarum* Kirk. (Hemiptera : Pentatomidae). Indian Journal of Entomology **73** (3): 230-233.

Gahukar RT. 2013. Cow urine: a potential biopesticide. Indian Journal of Entomology**75**(3): 212-216.

Gebreselassie N, Abrahamsen RK, Beyene F, Abay F and Narvhus JA. 2016. Chemical composition of naturally fermented buttermilk. International Journal of Dairy Technology **69** (2):200-208.

Ghongade DS and Sood AK. 2019. Population dynamics of two spotted spider mite (*Tetranychus urticae* Koch) on cucumber in relation to weather parameters under protected environment. Journal of Agrometeorology **21**: 204-210.

Ghongade DS and Sood AK. 2021. Economic injury level for *Tetranychus urticae* Koch on parthenocarpic cucumber under protected environment in north-western Indian Himalayas. Phytoparasitica **49** (5): 893-905.

Joshi M, Verma KS, Chandel RS, Inamdar AG, Kaushal S and Rana A. 2021. Efficacy of bioformulations against cutworm *Agrotis ipsilon* Hufnagel in potato. Indian Journal of Entomology **83** (3): 454-458.

Karthikeyan C, Veeraragavathatham D, Karpagam D and Firdouse SA. 2005. Cow based indigenous technologies in dry farming. Indian Journal of Traditional Knowledge

5 (1): 47-50.

Kasarkar AR, Kulkarni DK and Salokhe SP. 2021. Phytochemicals investigated in Dashparni ark. International Journal of Agriculture and Plant Science **3** (1): 48-51.

Kashyap L, Sharma DC and Sood AK. 2015. Infestation and management of russet mite, *Aculops lycopersici* in tomato, *Solanum lycopersicum* under protected environment in North-Western India. Environment & Ecology **33** (1): 87-90.

Kumawat N, Shekhawat PS, Kumar R and Sanwal RC. 2014. Formulation of biopesticides for insect pests and diseases management in organic farming. Popular Kheti **2** **(**2): 237-242.

Lengai GMW, Muthomi JW and Mbega ER. 2020. Phytochemical activity and role of botanical pesticides in pest management for sustainable agricultural crop production. Scientific African **7** (3): 1-13.

Nadana GRV, Rajesh C, Kavitha A, Sivakumar P, Sridevi G and Palanichelvam K. 2020. Induction of growth and defense mechanism in rice plants towards fungal pathogen by eco-friendly coelomic fluid of earthworm. Environmental Technology and Innovation **19**: 101011(1-10).

Nayak H, Rai S, Mahto R, Rani P, Yadav S, Prasad SK and Singh RK. 2019. Vermiwash: A potential tool for sustainable agriculture. Journal of Pharmacognosy and Phytochemistry **5**: 308-312.

Onunkun O. 2014. Field trials using cow urine and dung as biopesticides against sucking bugs of *Amaranthus cruentus*. International Journal of Research in Agricultural Sciences **1** (4): 167-171.

Pappas ML, Migkou F and Broufas GD. 2013. Incidence of resistance to neonicotinoid insecticides in greenhouse populations of the whitefly, *Trialeurodes vaporariorum* (Hemiptera: Aleyrodidae) from Greece. Applied Entomology and Zoology **48** (3): 373-378.

Patel PS, Shukla NP and Patel GM. 2003. Enhancing insecticide properties of cow urine against sucking pests of cotton. In: Proceedings of National Symposium on Frontier Areas of Entomological Research pp-227.

Pilkington LJ, Messelink G, Van Lenteren JC and Mottee KL. 2010. Protected biological control: biological pest management in the greenhouse industry. Biological Control **52**: 216–220.

Rameshwar, Saini JP, Chadha S and Sharma S. 2014. Organic Farming. Department of Organic Agriculture, COA, CSK Himachal Pradesh Krishi Vishvavidyalaya, Palampur, Himachal Pradesh.

Sayyad NR. 2017. Utilization of vermiwash potential against insect pests of tomato. International Research Journal of Biological Sciences **6** (1): 44-46.

Sharma S, Sood AK and Ghongade DS. 2021. Assessment of losses inflicted by the aphid, *Myzus persicae* (Sulzer) to sweet pepper under protected environment in north western Indian Himalayan region. Phytoparasitica (Published online https:://doi.org/10.1007/s12600-021-00951-7).

Sood AK and David BV. 2012. The greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood). In: *The Whitefly or Mealywing Bugs* (BV David, ed.). LAP Lambert Academic Publishing Gmbh and Co. KG, Germany, pp 411.

Sood AK, Singh V and Mehta PK. 2018. Current status and management strategies of insect-pests of vegetable crops under protected cultivation in Himachal Pradesh. In: *Technologies and Sustainability of Protected Cultivation for Hi-Valued Vegetable Crops* (Sanjeev Kumar, NB Patel, SN Saravaiya and BN Patel, eds). Navsari Agricultural University, Navsari, Gujarat, India. pp 339-354.

Sood AK, Sood S and Anjana Devi. 2014. Morphometrics and annual life cycle of greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood) in Himachal Pradesh. Himachal Journal of Agricultural Research **40** (1): 50-57.

Sood S, Sood AK and Verma KS. 2006. Determination of baseline toxicity of some insecticides to greenhouse whitefly, *Trialeurodes vaporariorum* (Westwood) population from North-Western Indian Himalayas. Pest Management in Horticultural Ecosystems **12** (1): 67-70.

Sudeshna. 2017. Management of red spider mite using natural products in cucumber under protected environment. MSc thesis, CSK Himachal Pradesh Krishi Vishvavidyalaya, Palampur, India.

Thakur S and Sood AK. 2019. Lethal and inhibitory activities of natural products and biopesticide formulations against *Tetranychus urticae* Koch (Acarina: Tetranychidae). International Journal of Acarology **45** (6–7): 381–390.

Thakur S and Sood AK. 2021. Deterrent activity of natural products to red spider mite, *Tetranychus urticae* Koch. Indian Journal of Entomology **84** (3): 551-555.

Thakur S and Sood AK. 2022. Foliar application of natural products reduces population of two-spotted spider mite, *Tetranychus urticae* Koch on parthenocarpic cucumber *(Cucumis sativus* L.) under protected environment. Crop Protection **160**: 106306.

Van Lenteren JC. 2000. A greenhouse without pesticides: fact or fantasy. Crop Protection **19** (6): 375-384.

Venkateswarlu V, Sharma RK and Sharma K. 2011. Evaluation of eco-friendly insecticides against major insect pests of cabbage. Pesticide Research Journal **23** (2): 172-180.