Eco-friendly application of vermiwash obtained from different types of waste with neem seed kernel extract against papaya mealybug, *Paracoccus marginatus* Williams and Granara de Willink

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Abstract— *Excessive agro-chemical usage has created a serious threat to the environment and human health. On the other hand, inappropriate bio solid waste management has given rise to social and environmental issues. Vermicomposting can be one of the solutions for both problems. Vermiwash is a product from vermicomposting which is generally used as a foliar fertiliser. Besides, it can also be used as a mild biopesticide in organic farming. In this study, three different substrates (cow dung, goat manure and vegetable waste) were used to produce vermiwash. Neem seed kernel (NSK) extract was combined with the vermiwash obtained from the different substrates to treat papaya mealybug (Paracoccus marginatus) and its predators on Carica papaya. 40% vermiwash and 10% NSK extract were used for the field study. The foliar application of vermiwash (cow dung) + NSK extract (1:1), vermiwash (goat manure) + NSK extract (1:1), vermiwash (vegetable waste) + NSK extract (1:1), vermiwash (cow dung), vermiwash (goat manure), vermiwash (vegetable waste) and control treatment resulted 75.72%, 67.19%, 57.62%, 45.00 %, 40.90%, 33.78% and 00.00% of papaya mealybug population reduction respectively. The combination of vermiwash obtained from cow dung and neem seed kernel extract (1:1) showed a higher performance in suppressing papaya mealybug population in field condition. There was no remarkable population reduction of the mealy bug predators after the application.*

Keywords— Vermiwash, Paracoccus marginatus, Natural predators, Neem seed kernel extract, Organic farming.

I. INTRODUCTION

The concern of organic farming and sustainable agriculture has increased in recent years. Unrestrained applications of fertilisers and pesticides in agriculture have caused adverse effect on the environment [1]. Meanwhile the rate of solid waste generation is escalating with population increase and industrialisation [2]. Improper management of bio solid waste cause serious problems in the society [3]. Vermicomposting is one of the environmental friendly methods in converting various bio wastes through earthworms into vermicompost and vermiwash [4]. These eco-friendly products can be an alternative to agro-chemicals. Vermiwash is a brownish-red liquid extract collected after the passage of water through a column of vermiculture. It is a collection of coelomic fluid and excretory products of the earthworms. The liquid fertiliser vermiwash helps to enhance crop productivity. Besides, it is effective against pests and diseases [5]. Vermiwash is rich in several nutrients, vitamins and plant growth regulators [6]. It also comprises insecticidal and antifungal bioactive compounds which suppress pest and diseases [7]. Earthworms express coelomic fluid when under severe stress condition. The coelomic fluid of the earthworm is secreted through dorsal pores in the form of mucus as a response of defense mechanism. The biological activity of the earthworm depends on the type of waste material provided as the feed for vermicomposting. The performance of *Eisenia foetida* varies in different types of waste used for vermicuture [8]. A number of studies have been undertaken on vermiwash as a liquid fertiliser in many crops. However, the study on application of vermiwash obtained from different types of waste in pest management is scarce.

There are various number of plant based pesticides used to manage the pests in organic farming. Neem (*Azadirachta indica A. Juss*) has remarkable insecticidal properties [9]. Azadirachtin is a component of neem which is an effective plant based pesticide and also has antifeedant and toxic effects on insects [10, 11]. In this study neem seed kernel extract was selected as a plant based pesticide to combine with vermiwash to manage soft body insects in papaya cultivation.

Papaya mealybug (*Paracoccus marginatus* Williams and Granara de Willink) of order Hemiptera, family Pseudococcidae is a severe pest of *Carica papaya* [12]. It is an invasive pest to Sri Lanka; it was first reported in Colombo and Gampaha districts in July 2008 [13]. It is a pest in fruit and vegetable crops and ornamental plants. It has a very wide host range extending up to 22 families of plants in Asia [14]. The insect feeds on leaves, stems and fruits and sucks the sap causing stunting of the plants, chlorosis, early fruit drop and death. Sooty mould is formed by the honeydew secreted by the insects while feeding [15]. There were many natural enemies of papaya mealybug recorded in the field. Insect like green lacewing (*Chrysoperla carnea* Stephens - Neuroptera: Chrysopidae) and coccinellid beetle (*Anegleis cardoni* Weise) and a spider species (*Phintella vittata* - Arachnida: Araneae: Salticidae) were recorded to predate on papaya mealybugs [16, 17]. Frequent and excess use of chemical insecticides results in depletion of natural enemies [18]. Thus, there is a need to use alternative environmental friendly insecticides to sustain the management. The objective of the study is to test the effect of different types of vermiwash and neem seed kernel extract on papaya mealybugs and its predators on *Carica papaya* as an environmental friendly solution.

II. METHODOLOGY

Vermiwash and neem seed kernel extract were prepared at the Department of Agricultural Biology, Faculty of Agriculture, University of Jaffna, Ariviyal nagar, Kilinochchi, Sri Lanka. Different types of vermiwash were obtained by using *Eisienia foetida* in the vermicompost unit.

2.1 Collection of vermiwash

Three identical 250L one side opened empty barrels were taken for the preparation of vermicompost. A hole was made at the lower end of each barrel to accommodate a vertical limb of 'T' jointed tube in a way that one inch of the tube was inside the barrel. A tap was attached to the outer end of the tube in order to collect the vermiwash. 25cm of broken bricks were placed at the bottom of the barrel as a filter unit. Above the brick layer, a 30cm of loamy soil was placed. On top of this, a 30cm layer of cow dung in the first barrel, goat manure in the second and vegetable wastes in the third were added separately. The substrates were moistened with water. *Eisienia foetida* earthworms of 0.5kg were released in the barrels. The vermicompost barrels were covered with muslin cloth for protection. Freshwater was sprinkled daily to maintain the moisture (80%). The mixture was manually turned up once a week for 3 weeks. After 28 days, the three different types of vermiwash from each barrel were collected and labelled as vermiwash (cow dung), vermiwash (goat manure) and vermiwash (vegetable waste).

2.2 Preparation of neem seed kernel extract

200g of cleaned, shade dried kernels of neem were ground properly, soaked for 24 hours in 1L of distilled water and squeezed through muslin cloth to obtain the extract. The volume of the extract was then made up to 4L by adding 3L of distilled water. A concentration of 10% of this extract was used for the treatments. A detergent powder (0.1%) was added as a spreading agent just prior to the treatment [19].

2.3 Field studies

A field study was conducted in a farmer field in Urumpirai, in the Jaffna district in June 2017 to evaluate the efficacy of selected treatments in completely randomized block design. Red lady variety *Carica papaya* trees of the age of 15 months were selected for the study. The treatments were applied when the crop reached 25-50% of leaves/fruits damage (Table 1). The total number of papaya mealybugs in $16cm^2$ per fruit was counted using hand lens. The initial count of the papaya mealybugs was recorded a day before the first treatment. The respective treatments were applied using hand sprayers in a 14 day interval. Each treatment was replicated five times with each replicate being an individual plant. Treatments were evaluated 7, 14, 21 and 28 days after the initial spray. Additionally, the number of predators was recorded for each treatment.

2.4 Statistical analysis

Pest and predator population reduction levels at all treatments were subjected to analysis of variance (ANOVA) and the means were separated by Duncan's Multiple Range Test (DMRT) using SAS. Percentage of reduction was calculated according to Henderson-Tilton formula.

TABLE 1
DETAILS OF TREATMENTS USED AGAINST PAPAYA MEALYBUG

Treatment Number	Treatment
T1	40% Vermiwash (goat manure)
T2	40% Vermiwash (cow dung)
T3	40% Vermiwash (vegetable waste)
T4	40% Vermiwash (goat manure) + 10% NSK extract (1:1)
T5	40% Vermiwash (cow dung) + 10% NSK extract (1:1)
T6	40% Vermiwash (vegetable waste) + 10% NSK extract (1:1)
T7	Control

NSK – Neem seed kernel

III. RESULTS AND DISCUSSION

In the field experiment all treatments showed a gradual reduction in papaya mealybug population compared to the control (Table 2).

TABLE 2
THE MEAN NUMBER OF PAPAYA MEALYBUGS AND PERCENTAGE OF REDUCTION

Treatment Number	Treatments	Mean ± SE of papaya mealybug					Population reduction % in week 4
			1st application		2nd application		
		PTC	7 DAT	14 DAT	7 DAT	14 DAT	
T1	40% Vermiwash (goat manure)	32.00	31.2 ± 1.3^{b}	30.40 ± 0.8^{b}	28.20 ± 0.9^{c}	27.00 ± 0.8^{c}	40.90
T2	40% Vermiwash (cow dung)	32.60	31.00 ± 1.5^{b}	29.80 ± 1.5^{b}	27.20 ± 1.1^{c}	25.60 ± 0.8^{d}	45.00
T3	40% Vermiwash (vegetable waste)	33.00	33.00 ± 0.8^{b}	32.60 ± 1.7^{b}	31.00 ± 0.8^{b}	31.20 ± 0.8^{b}	33.78
T4	40% Vermiwash (goat manure) + 10% NSK extract (1:1)	31.60	24.00 ± 1.3^{cd}	22.00 ± 1.7^{c}	18.80 ± 1.5^{e}	14.80 ± 1.2^{g}	67.19
T5	40% Vermiwash (cow dung) + 10% NSK extract (1:1)	30.00	21.60 ± 0.8^{de}	19.00 ± 1.5^{c}	14.60 ± 1.2^{e}	10.40 ± 0.8^{f}	75.72
T6	40% Vermiwash (vegetable waste) + 10% NSK extract (1:1)	31.40	25.60 ± 1.6^{bc}	24.80 ± 2.4^{c}	22.00 ± 2.4^{d}	19.00 ± 2.2^{e}	57.62
T7	Control	31.80	38.40 ± 1.8^{a}	39.40 ± 1.6^{a}	40.20 ± 0.9^{a}	45.40 ± 1.0^{a}	-

Means having same letters in a column indicate that the values are not significantly different at 0.05 α by DMRT, SE – standard error, NSK- neem seed kernel, PTC- pre-treatment count, DAT- day after treatment

Applications combined with neem seed kernel extract showed a higher pest population reduction level than the sole applications of vermiwash. Neem seed kernel extract contains insecticidal compound called Azadirachtin which has repellent and antifeedant effects against insect pest [10, 11]. In the 4th week, the mixed application of 40% vermiwash (cow dung) + 10% NSK extract (1:1) showed a higher pest reduction of 75.72% followed by the combined applications of 40% vermiwash (goat manure) + 10% NSK extract (1:1) with 67.19% and 40% vermiwash (vegetable waste) + 10% NSK extract (1:1) with 57.62%. A similar study on insect pest management reported that, combined application of vermiwash with plant extracts suppressed the infestation of thrips and mites and improved productivity of the crop [20]. Vermiwash contains numerous enzymes, vitamins, macro and micro nutrients, hormones, insecticidal and antimicrobial properties which support plant health and protection [6, 7]. Tracy in 1951 reported the activity of chitinase in the posterior part of the intestine of earthworm's mid gut [21]. Chitinase in earthworms not only supports digestive function but also engages in defense mechanism against chitin containing organisms [22]. Chitinase present in the vermicast repels insects; the enzyme degrades the chitin in the arthropods exoskeleton [23, 24, 25]. However, the mechanism of vermiwash in reduction of population

density of mealybugs was less clear. In other related studies on soft body arthropods, applications of vermiwash have reduced the pest population of sucking insects [26, 27, 28].

Sole applications of 40% vermiwash obtained from cow dung, goat manure and vegetable waste resulted 45.00%, 40.90% and 33.78% of pest suppression respectively. There was a significant difference in mealybug population reduction between vermiwash prepared by using cow dung and goat manure. This may be due to the performance of the earthworms in different animal wastes. *Eisienia foetida* biomass, reproduction performance, cocoon production per worm and growth rate were reported high in cattle manure than goat manure [8, 29]. Vermiwash obtained from the vegetable substrate showed the least efficacy in insect suppression level. It may be due to the poor growth of the earthworms in the vegetable waste. Growth and reproduction of the earthworms depend on the biochemical quality of the substrates provided as feed [30]. The biochemical quality of the vermiwash varies based on the types of waste used in vermicomposting [31].

TABLE 3
POPULATION REDUCTION OF PREDATORS OF MEALYBUG BY TREATMENTS

Treatment Number	Treatments	Reduction of population %					
		Coccinellid		Green lacewing		Spider	
		14 DAT	28 DAT	14 DAT	28 DAT	14 DAT	28 DAT
T1	40% Vermiwash (goat manure)	-40.00*	-20.00*	-68.00*	-40.00*	-33.33*	-25.00*
T2	40% Vermiwash (cow dung)	-16.67*	00.00	-40.00*	00.00	-77.78*	-33.33*
T3	40% Vermiwash (vegetable waste)	-28.33*	-10.00*	-54.00*	-28.33*	-33.33*	-20.00*
T4	40% Vermiwash (goat manure) + 10% NSK extract (1:1)	-40.00*	-20.00*	-68.00*	-40.00*	-33.33*	-25.00*
T5	40% Vermiwash (cow dung) + 10% NSK extract (1:1)	-16.67*	00.00	-16.67*	00.78	-66.67*	-25.00*
T6	40% Vermiwash (vegetable waste) + 10% NSK extract (1:1)	-40.00*	-20.00*	-68.00*	-40.00*	-55.56*	-33.33*
T7	Control	-	-	-	-	-	-

NSK- neem seed kernel, DAT- day after treatment,* negative sign (-) means no reduction in population

The data collected on predators revealed that there were no reduction in coccinellid beetle (*Anegleis cardoni*) and spider (*Phintella vittata)* population in the field after the application (Table 3). However, 40% Vermiwash (cow dung) + 10% NSK extract showed a slight population reduction of 0.78% on soft body insect green lacewing (*Chrysoperla carnea* Stephens). Overall, the sprays showed less harm to the mealybug predators in the habitat.

IV. CONCLUSION

The efficacy of vermiwash mixed with neem seed kernel extract against papaya mealybugs was higher than the treatments solely with vermiwash. Since vermiwash is a mild bio pesticide it can be combined with neem seed kernel extract in pest management. Mixed foliar application of 40% vermiwash (cow dung) and 10% neem seed kernel extract (1:1) in an interval of 2 weeks curtailed the population build up of papaya mealybugs. This cost effective combination of application was identified as a remedy for pest problem on papaya crops in organic farming and home gardening. Vermiwash obtained from cow dung showed a higher pest population reduction level than the vermiwash obtained from goat manure and vegetable waste. The type of waste used for vermiwash production influenced the efficacy level of pest suppression. The eco-friendly treatments showed negligible effect on predators of papaya mealybugs.

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