



Evaluation of Some Botanical Extracts Against Major Insect Pests (Leafminer, Armored scale and Woolly Whitefly) of Citrus Plants in Central Zone of Tigray, North Ethiopia

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ABSTRACT

Citrus fruit production is suffering from various yield-limiting factors particularly the sucking pests viz., citrus leafminer *Phyllocnistis citrella*, woolly whitefly, *Aleurothrixus floccosus* and scale insects especially armored scales *Aonidiella aurantii*. The present paper tries to identify the effective botanical insecticides against these insect pests on citrus orange. An experiment was conducted in the established citrus orange farm in Kolla Temben at two farmer's fields, Adiha and Agibe during the off-season of 2018 under irrigation growing condition in a randomized complete block design with six treatments and replicated thrice. The findings in both experimental sites showed that the neem seed extract had a significantly ($P=0.001$) lower leafminer infestation levels compared to the untreated control which is on par to the insecticide treatment of dimethoate. On the other hand, in both areas, whitefly mortality of more than 81% on average was recorded from neem seed extract followed by Tree tobacco (70%) ($P=0.001$). For scale insects however, in both experimental sites, the highest mean percent mortality was recorded from the insecticide dimethoate 40% EC (86.9%, 87.2 & 86.0% on average) followed by neem seed and tree tobacco extracts, 70.1 and 65.4% respectively. The botanicals particularly the neem seed extracts followed by tree tobacco are as effective as the chemical insecticide, dimethoate 40% EC, even superior in some cases, in controlling the target pests. Therefore, these botanicals could be used as an IPM component for against the target pests.

Keywords: Citrus, Sucking insect pests, Damage, Botanical insecticides, Control.

1. INTRODUCTION

Ethiopia is one of the countries in Africa which have huge potential for the development of different varieties of horticultural crops. The country is endowed with natural resources in different agro-ecological zones which are suitable for the cultivation of horticultural products. Accordingly, large varieties of flowers, vegetables, fruit and herbs are being grown currently in various areas of the country (Kassa, 2015). In addition, the country has an ample resource of irrigation water from different rivers or streams, small dams and ground water which could be used to grow various horticultural crops.

Horticultural crop production in the country in general and in Tigray in particular is increasing both in area coverage as well as production both in small scale farmers and

commercial farms. This significant positive change in productivity is mainly due to its high profitability per unit area and its role for improving income and nutrition status of the people. In general, horticultural crop production is widely recognized for its importance in food security, human nutrition and health (source of vitamins, minerals, antioxidants, dietary fiber and for having anti-carcinogenic properties).

The orange (specifically, the sweet orange) is the fruit of the citrus species *Citrus sinensis* in the family Rutaceae. The fruit of the *Citrus sinensis* is considered a sweet orange, whereas the fruit of the *Citrus aurantium* is considered a bitter orange. The orange is a hybrid, possibly between pomelo (*Citrus maxima*) and mandarin (*Citrus reticulata*), which has been cultivated since ancient times (Lacey et al., 2009). Probably originating in Southeast Asia, oranges were already cultivated in China as far back as 2500 BC. Arabo-Phone peoples popularized sour citrus and oranges in Europe; Spaniards introduced the sweet orange to the American continent in the mid-1500s (Lazaneo, 2008).

Orange trees have been the most cultivated tree fruit in the world since 1987. Orange trees are widely grown in tropical and subtropical climates for their sweet fruit. The fruit of the orange tree can be eaten fresh, or processed for its juice or fragrant peel. Sweet oranges currently account for approximately 70% of citrus production. In 2010, 68.3 million metric tons of oranges were grown worldwide, production being particularly prevalent in Brazil and the US states of California and Florida (CABI Crop Protection Compendium, 2010). Still the information regarding the status of citrus in Ethiopia is missed.

The central zone of Tigray at large and the Kola Tembien (Adiha and Agibe areas) “woreda” (district) in particular is one of the potential areas for cultivation of different fruits (mango, orange and papaya). However, citrus fruit production in the areas is suffering from various yield-limiting factors among others, limited availability of improved varieties and agronomic practices, outbreak of insect pests and diseases, limited knowledge and skill on horticultural crops production techniques and utilization and weak research and extension support.

Insect pests particularly the sucking pests of citrus Leafminer, *Phyllocnistis citrella* (Gracillariidae: Lepidoptera), woolly whitefly, *Aleurothrixus floccosus* (Aleurodidae: Hemiptera) and scale insects especially armored scales, *Aonidiella aurantii*, reduce both the quantity and quality of produces. The former causes damages both in nursery and in grown up stages of the

citrus plant. The larvae attack tender leaves and feed in the epidermal layers of the leaf by making serpentine mines in which air gets trapped and gives them silvery appearance. The affected leaves turn pale yellow, get distorted and crumpled. Such leaves gradually dry and die away. The attack of this pest also encourages the development of citrus canker disease. On the other hand, whiteflies have naturally a piercing-sucking mouthpart that they insert into the plant tissue to feed on leaves and during this process, secrete honeydew due to which sooty mould develops on the leaves. Sometimes, the infestation is so severe that the whole orchard looks black. Fruits turn black in color and have insipid taste. The attack is more intense on the shady side of the tree. Leaves are wilted and dropped as a result of feeding damage as the populations were high. The armored scales also damage the fruits and form as blemishes at low levels of infestation and in severe cases, they damage the tree badly. Armored scales secrete armor wax in an oyster shell or circular pattern that covers their body. They remain attached to the host plant when shells are lifted up and they are difficult enough to control them because of these too hard body coverings provide protection against insecticides. Insect pest problems in general, are aggravated by different prevailing environmental stresses. For example, susceptibility of citrus crops to these pests may increase due to moisture stresses, which in turn increases yield loss in a given cropping system. The emergence of new and outbreak of existing insect pests also increase with climatic change. Moreover, there has been uncontrolled introduction of planting materials (seeds, vegetative materials) into the experimental area, without effective quarantine checking. Such introduction of plant materials may harbor disease causing agents and insect pests such as the woolly whitefly (*Aleurothrixus floccosus*), which previously have not been known in the area. All these problems do have a negative influence on the livelihood of the farmers.

Several synthetic insecticides have been widely used to control citrus insect pests in the field (Mayer et al., 1987) and though effective in bringing down pest populations, these chemical insecticides are costly and unsafe to non-target organisms such as natural enemies of the target pests, humans and the environment (Jensen and Simko, 2001; Nault et al., 2013). Particularly small holder farmers who fail to follow the practices of safe handling and application of pesticide may suffer more from the negative effects of synthetic pesticides. Moreover, most small holder famers may not afford the ever increasing costs of pesticides, therefore, alternative non – chemical insecticides, such as botanicals (Ayalew, 2005; Shiberu et al., 2013) could be the best option for small holder fruit and vegetable producers. The applications of synthetic insecticides

may have residual effect, accumulating in different components of the environment, and have adverse effects on non-target organisms, ecosystems and human health. Therefore, bio-insecticides often recognized as “Green pesticides” are an alternative to synthetic insecticides in agriculture and public health sectors (Mossa, 2016). The present study focused on the effectiveness of plant extracts as bio-insecticides for the management of the three sucking insect pests mentioned above. The botanicals are a complex of chemical compounds with multiple modes of action that enhances their activity due to the synergistic action between constituents. They can offer a safe and effective alternative to conventional insecticides for controlling insect pests within an integrated pest management program. The use of botanical insecticides is ideal for organic farming and is safe for both the environment and human health. Hence, due to the accessibility of botanical plants in the study area and sufficient earlier evidences (Birhane, personal communication, February 2018), the present study was carried out with the objective of identifying effective botanical insecticides against the sucking insect pests vis., citrus leafminer, armored scale and citrus whitefly on citrus orange.

2. MATERIALS AND METHODS

2.1. Description of the Study Area

The study was conducted in Kolla Temben at two farmer’s fields in Adiha and Agibe areas during the off-season of 2018 under irrigation growing condition. The areas are found in central zone of Tigray regional State. Agibe is found about 65 kilometres (km) north-west of Mekelle, while Adiha is 120 km away from Mekelle in the same direction. The agro-ecological zone of the study areas is characterized by warm sub-moist mid highlands, with short growing season, erratic and low amount of rain fall. The average annual rainfall of the area is 500 - 700 mm, which is concentrated in one season that is from June to beginning of September. The mean annual maximum and minimum temperature is 28°C and 18°C respectively. The soils of the study area are broadly categorized as vertisols (black clay soils) in Agibe and sandy loam in Adiha (Kolla Temben WoANRD, 2015, unpublished data).

The experiment was conducted in the established citrus orange farm in randomized complete block design (RCBD) with six treatments and replicated thrice. The treatments were four botanical extracts; neem (*Azadiracthaindica*) leaf and seed, tree tobacco (*Nicotiniaglaucea*) leaf, Mexican marigold (*Tagetesminuta*) leaf, synthetic insecticide (dimethoate 40% E.C) and

untreated control. Three years old citrus orange trees grown at four meter spacing were used for the experiment. Individual trees were considered as experimental plots.

2.2. Method of Botanicals Extraction

2.2.1. Preparation of Extracts from Neem Leaf

Sufficient matured neem leaves were collected from Tanqua Abergele, washed, chopped and dried in the laboratory under shade for five days. The dried leaves were ground using electrical blender to make fine powder. The powder was sieved using 200 µm mesh and then mixed with water at a rate of 50g/l of water and left overnight. The next day the mixture was filtered using cheese cloth and made ready for spraying.

2.2.2. Preparation of Neem Extracts from Dried Neem Kernel

Dried fruits of neem collected from the same location as that of the leaf were cracked to remove kernels inside. The kernels were dried under shade and ground with an electrical grinder. The resulting powder was sieved using 200 µm mesh, mixed with water at a rate of 50gm per 1 liter of water and soaked overnight. The next morning the mixture was filtered through cheesecloth and made ready for application.

2.2.3. Preparation of Leaf Extracts from Mexican Marigold and Tree Tobacco

Leaves of these two plant species were collected from the farm field around Mekelle town and were dried in the laboratory for seven days. Dried leaves of the species were blended using electrical blender. Leaf powder of these species was mixed water at a rate of 50gm per 1 liter of water and left overnight for soaking. The leaf powder –water mixture was filtered the next morning using cheesecloth to remove plant debris after which liquid soap was added as an emulsifying agent.

5gm soap was added before application in all the bio-pesticides for the purpose of serving as later sticky agent. The synthetic systemic insecticide, dimethoate 40% EC was sprayed at the manufacturers recommended rate of 1L/ha after being mixed with water in 15 L knapsack sprayer.

2.2.4. Spraying Frequency of Botanicals in the Field

Each of the botanical preparations and the chemical insecticide (dimethoate 40% EC at the rate of 1L/ha) were applied three times during the orange plants vegetative growth period at about two week's interval in the field.

2.3. Data Collected

Three orange branches which contain 20 leaves on average were randomly selected and examined for leafminer damage one day before spraying as well as on the 7th and 14th days after each spray treatment application. The percentages (0 – 100%) of mined leaves per plant were determined for the different treatments computing the proportions of the minded leaves against the unmined leaves in percent. Similarly, the number of insects (adults and nymphs of whitefly, and scale insects) were counted on each plant from three randomly selected branches, containing 20 leaves on average, at one day before spraying as well as on the 7th and 14th days after spraying (DAS). Whether the adults and/or the nymphs are dead is checked by closely using hand lenses and touching slightly on the pest and if dead are remain stick to the leaves being shrinked. Also the leaves beneath the infested leaves are checked in the same manner. Percent mortality (reduction in the number of insect population after treatment) was recorded by pest scouting before and after treatment by using the following equation:

$$M = (\sum IB - \sum IA) * 100 / \sum IB$$

Where M = mortality percentage; IB = Insect population before treatment; IA = Insect population after treatment (Iqbal, 2018).

2.4. Data Analysis

Data on insect count was square root transformed before the analysis of variance, while data on percentage of leafminer damage were transformed using $\text{Log}_{10}(x + 1)$ transformation in order to normalize the data. Data analysis (ANOVA) was performed using Minitab software version 18. Mean separation was carried out according to Tukey's Honestly Significant Difference (HSD) test at 0.05 probability level.

3. RESULTS AND DISCUSSION

3.1. Effect of Botanicals against Citrus Leafminer (*Phyllocnistis citrella*)

The results in Adiha showed that the lowest leafminer infestation was recorded from plants treated with neem seed extract and the insecticide dimethoate 40% EC. The average (mean of 7 days after spray - DAS and 14 DAS) leafminer infestation levels on neem seed treated plants were 19%, 23.7% and 29.5 %, in the first, second and third sprays compared to the untreated control that had 20%, 54.7% and 65.7% infestation during the same assessment periods, respectively. Particularly, referring the record at 14 days after spray of the last spraying time,

there was 43.3 % (74-30.7%) reduction of leafminer infestation in plants treated by neem seed extract compared to the untreated control (Table 1). The infestation trends was observed to have increased more in the untreated control through the initial to the end of sprays, while the infestation on botanical seed extract treated plants although shown to increase the infestation of the leaves, it was at a very low levels. Such increment in pest infestation of the treated plot might have resulted from cross-infestation by adult moths from unsprayed fields laying eggs, as the pest has short developmental time (2-3 weeks), and as many as 7-8 generations per year (Elizabeth, 2010).

Table 1. Effect of different botanicals against citrus leafminer (*Phyllocnistis citrella*) infestation of orange plants in Adiha during 2018.

Treatments	Mean percentage of infested orange tree leaves								
	First spray			Second spray			Third spray		
	1DBS *	7DAS **	14DAS	1DBS	7DAS	14DAS	1DBS	7DAS	14DAS
Neem seed	17.7 ^a	18.3 ^d	19.7 ^d	21.0 ^d	22.7 ^d	24.7 ^d	25.7 ^c	28.3 ^{cd}	30.7 ^{cd}
Neem leaf	20.0 ^a	32.0 ^b	32.3 ^b	34.0 ^b	35.7 ^b	36.8 ^{bc}	38.7 ^b	40.0 ^b	42.3 ^b
Tree tobacco	19.0 ^a	24.0 ^c	26.0 ^c	28.3 ^c	29.3 ^c	31.7 ^c	34.6 ^b	36.7 ^{bc}	39.0 ^{bc}
Mexican marigold leaf	20.7 ^a	30.0 ^b	32.0 ^b	34.7 ^b	36.7 ^b	39.0 ^b	40.3 ^b	42.3 ^b	43.8 ^b
Dimethoate 40% EC	18.3 ^a	18.3 ^d	20.0 ^d	20.3 ^d	21.5 ^d	22.2 ^d	22.7 ^c	23.3 ^d	23.6 ^d
Untreated control	20.0 ^a	45.0 ^a	52.0 ^a	54.7 ^a	57.0 ^a	63.0 ^a	65.7 ^a	68.3 ^a	74.0 ^a
St. Dev	1.2	2.0	1.5	2.1	2.2	2.4	3.0	3.4	3.5
Mean	19.6	27.9	30.3	32.2	33.8	36.2	37.9	39.8	42.2
p-value	0.007	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
CV (%)	6.2	7.1	5.0	6.4	6.4	6.7	7.8	8.6	8.3

Note: *DBS: mean percent leafminer infestation 1 day before insecticide spraying; **DAS: days after spraying; St. Dev: standard deviation; CV: coefficient of variation. [Within each column, means followed by the same letter do not differ significantly according to Tukey's Honestly Significant Difference (HSD) test at 0.05 probability level].

As the larvae of leaf miner attacks the newly growing tender leaves and feed in the epidermal layers of the leaf by making serpentine mines, it is often difficult to reach it hence, unable to control fully with such neem – botanical pesticides that contribute to the reason that in the treated leaves increment of infestation observed. However, still the neem seed treatment is effective and was found to be on par to the insecticide treatment of dimethoate in controlling

leafminer damage. The leaf extracts of tree tobacco was the second best performing among the botanical treatments, next to neem seed extract, in controlling leafminer infestation, with an average infestation level of 25%, 30.5% and 37.8% in the 1st, 2nd and 3rd sprays, respectively and all are statistically significant at P=0.001 (Table 1). Tree tobacco, *N. glauca*, contains Anabasine, the predominant alkaloid compound isomeric with nicotine (that is, containing the same proportions of carbon, hydrogen, and nitrogen), with an insecticidal properties that does have a toxic effect on the larva and thus minimize the leaves infestation (Louis 2010).The application of botanical insecticides integrated with timely inspection of trees to see if there are any pests on them and early action can be much more effective and achieve great success than trying to get rid of a pest once it has multiplied and caused damage on the trees.

Table 2. Effect of different botanicals against citrus leafminer (*Phyllocnistis citrella*) infestation of orange plants in Agibe, during 2018.

Treatments	Mean percentage of infested orange tree leaves								
	First spray			Second spray			Third spray		
	1DBS *	7DAS **	14DAS	1DBS	7DAS	14DAS	1DBS	7DAS	14DAS
Neem seed	14.0 ^a	15.3 ^d	15.7 ^d	16.0 ^d	20.7 ^d	21.7 ^d	18.6 ^d	24.3 ^d	27.7 ^d
Neem leaf	17.6 ^a	29.0 ^b	28.3 ^b	29.0 ^b	33.7 ^b	33.8 ^{bc}	35.7 ^b	39.3 ^b	43.3 ^b
Tree tobacco	16.3 ^a	21.0 ^c	22.0 ^c	23.3 ^c	27.3 ^c	28.7 ^c	29.6 ^c	31.3 ^c	36.9 ^c
Mexican marigold leaf	16.7a	27.0 b	28.0 b	29.7 b	34.7 b	36.0 b	37.2 b	38.8 b	40.8 bc
Dimethoate 40% EC	14.3 ^a	15.3 ^d	16.0 ^d	15.3 ^d	19.5 ^d	19.2 ^d	19.5 ^d	20.2 ^d	21.1 ^d
Untreated control	14.0 ^a	42.0 ^a	48.0 ^a	49.6 ^a	55.0 ^a	60.0 ^a	60.7 ^a	64.3 ^a	71.0 ^a
St. Dev	2.0	2.0	1.5	2.1	2.2	2.4	2.0	2.5	2.7
Mean	15.5	24.9	26.3	27.2	31.8	33.2	33.6	36.4	40.0
p-value	0.160	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
CV (%)	12.7	8.0	5.7	7.6	6.8	7.3	5.9	6.9	6.7

Note: * DBS: mean percent leafminer infestation 1 day before insecticide spraying; **DAS: days after spraying; St. Dev: standard deviation; CV: coefficient of variation. [Within each column, means followed by the same letter do not differ significantly according to Tukey's Honestly Significant Difference (HSD) test at 0.05 probability level].

On the other hand, the selected systemic insecticide, dimethoate 40% EC, constantly have had a record of the highest and significant reduction of the insect mean percentage infestation in

all the spray intervals to 19.2%, 21.9% and 23.5 % compared to the leafminer infestation levels recorded before spraying in the same plants (Table 1).

In Agibe, in a similar fashion to Adiha, the neem seed extract and the systemic insecticide – dimethoate 40% EC had a significant ($P=0.001$) reduction of leafminer infestation levels compared to the other treatments. Considering the mean of leafminer infestations after 14 DAS, the levels in the neem seed extract treated plants were 15.7%, 21.7% and 27.7% after the 1st, 2nd and 3rd sprays, respectively. The highest leafminer infestations were recorded from the untreated check, which were on average 45%, 57.5 and 67.7 % (average of 7 DAS and 14 DAS) after the three spraying periods, respectively (Table 2). The leafminer infestation level record in the neem seed extract and the insecticide dimethoate were on par to each other. The chemical insecticide also had an outstanding performance in Agibe, in controlling leafminer, reducing infestations by 26.7%, 35.5% and 44.1% (untreated-sprayed) at 7 DAS of the while it was 32.0%, 40.8% and 49.9.0% at 14 DAS of the 1st, 2nd and 3rd sprays respectively, compared to the untreated check. Tree tobacco, leaf extract treated orange leaves had also less infestation, which was significantly lower compared to the untreated ones as well as lower than the other botanical treatments except neem seed extract in all assessment dates after the spray applications. In both locations the infestation trends of the leafminer insect pest and the performance ranking of the treatments were in a very similar pattern (Table 1 and 2). In this study, it is clearly observed that leafminer infestation was greatly associated with emergence of new leaf flush. Therefore, spraying activities should target this crop stage.

The insect is a tiny moth that lays an egg under the leaf from which a larvae hatches that burrows into the leaf creating tunnels around in a serpentine pattern between the leaf surfaces leaving a whitish, silvery winding pathway through the leaf (Elizabeth 2010). Damage from this pest causes citrus foliage to cup and twist, creating a malformed leaf thus makes the insect difficult to control unless you apply a systemic insecticide like the one included as a standard check in this study—dimethoate 40% EC.

Rao et al. (2014) in their evaluation of plant products against citrus leafminer, in a field experiment, reported that, neem seed kernel extract (NSKE) at 5 percent, tobacco leaf extract at 5 percent and azadirachtin 10,000 ppm at 1 ml and 2 ml/L provided high levels of larval mortality ranging from 82.68% to 86.25%. In other similar experiment, Rao et al. (2015) indicated that as prophylactic measure against leafminer, the neem seed kernel, azadirachtin, and tree tobacco

extracts tested protected the sweet orange plants completely from leafminer attack up to three days, while neem seed kernel extract, azadirachtin and *Bacillus thuringiensis* (Bt)(0.005%) offered protection up to five days. In their curative experiment using natural products, *Bacillus thuringiensis* (Bt)(0.005 and 0.0025%), they observed 100 percent mortality of leafminer larvae upto 10 days. Among all botanicals they tested, the best treatments in terms of efficacy were azadirachtin and NSKE, which offered 79.97% and 78.39% larval mortality at five days after spraying.

Muhammad et al. (2016) observed that foliar application of 30% neem leaf extracts gave up to 12% reduction in the population infestation of citrus leafminer at 72 hrs post application. And therefore, they recommended neem extract as dependable to manage and control citrus leafminer infestation on citrus seedlings in nursery. The azadirachtin technical material isolated from neem seed or generally neem seed extract is most potent tetranotriterpenoid effective against several insect pests. It is most potent insect repellent, anti-feedant and an insect growth regulator and thus it is effective to combat insect pests including leafminer. Valand et al. (1992) also indicated that effective citrus leafminer control can be achieved with the systemic insecticide - dimethoate 40% EC.

3.2. Effect of Botanicals on woolly whitefly, *Aleurothrixus floccosus*

The results of the effectiveness of different botanicals on the whiteflies in Adiha are indicated in table 3. The woolly whitefly count (adult and nymph) in the untreated check one day before 1st spray was 367, and the population were increased in the second and third spray schedule assessments from the initial count to 490 (33.5%) and 606.7 (65.2%) respectively. The neem seed sprays performs with the highest mortality percent in whitefly population that is more than 80% in the three sprays followed by tree tobacco greater than 75% (Table 3) (P=0.001). This is a promising result for growers as these plants are easily accessible in their locality and can be used widely with no negative side effects to human health and the environment. The chemical insecticide, dimethoate 40% EC, had the highest mean mortality of whitefly. This chemical insecticide is registered in Ethiopia to be used for the control of sucking insect pests and our results confirms that it is best performing insecticide shortly after spraying. The other botanical pesticides leaf extracts of neem and Mexican marigold also displayed a substantial effect in controlling whiteflies, but less effective than the neem seed and Tree tobacco extracts. The woolly whitefly has several generations a year, the greatest populations usually occurs in hottest

seasons, coinciding with growth of flushes. Tender newly sprouted young leaves are particularly attractive and nutritious for citrus whiteflies. Therefore, spray schedules should be considered with the crop bud breaking cycle and new leaf flush growth. Besides, great number of woolly whiteflies was observed to occur in the underside of leaves and spraying activities should cover the undersides of all infested leaves.

Table 3. Effect of various botanicals on citrus whitefly (*Dialeurodes citri*) in orange trees at Adiha, during 2018.

Treatments	Percent mortality of whitefly insects (reduction in whitefly population compared to the count made before sprays)								
	First spray			Second spray			Third spray		
	CBS*	7DAS**	14DAS	CBS	7DAS	14DAS	CBS	7DAS	14DAS
Neem seed	391 ^a	80.6 ^{ab}	80.2 ^{ab}	83 ^{bc}	80.7 ^{ab}	83.2 ^{ab}	36.0 ^b	80.2 ^b	83.1 ^b
Neem leaf	390 ^a	69.8 ^d	71.9 ^d	107 ^b	67.8 ^c	69.6 ^d	41.3 ^b	66.9 ^d	71.8 ^c
Tree tobacco	379 ^a	75.9 ^{bc}	78.2 ^{bc}	88 ^{bc}	75.5 ^{bc}	79.0 ^{bc}	44.8 ^b	78.3 ^{bc}	81.3 ^b
Mexican marigold leaf	379 ^a	73.9 ^{cd}	74.4 ^{cd}	95 ^{bc}	69.8 ^c	73.3 ^{cd}	45.3 ^b	72.7 ^{cd}	74.1 ^c
Dimethoate 40% EC	378 ^a	85.5 ^a	85.9 ^a	71 ^c	86.2 ^a	86.7 ^a	47.6 ^b	88.4 ^a	89.9 ^a
Untreated control	367 ^a	0.0 ^e	0.0 ^e	490 ^a	0.0 ^d	0.0 ^e	606.7 ^a	0.0 ^e	0.0 ^d
St. Dev	15.5	2.2	2.1	10.8	3.9	2.6	9.2	2.7	2.5
Mean	380.5	64.1	65.1	155.7	63.3	65.3	120.6	64.4	66.7
p-value	0.452	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
CV (%)	4.1	3.4	3.2	7.8	6.1	3.9	8.9	4.2	3.7

Note: CBS: whitefly insects count 1 day before spraying;

**DAS: days after spraying; St. Dev: standard deviation; CV: coefficient of variation.

[Within each column, means followed by the same letter do not differ significantly according to Tukey's Honestly Significant Difference (*HSD*) test at 0.05 probability level].

Results for the effectiveness of the various botanicals tested on the population of woolly whitefly (*Aleurothrixus floccosus*) on orange trees in Agibe was as indicated in table 4. There was highly significant variation among treatments in controlling the citrus whitefly population, but all treatments showed high level reduction of insect count (causing mortality) compared to the untreated control. The whitefly insect count in the untreated check before application of the 1st spray was 118 per branch containing 20 leaves on average, and the pest population increased by 28.8% and 65% in counts made one day before the second and third spray, respectively (Table 6). Whereas, the neem seed extract and tree tobacco extract treatments had significant effect in killing the target pest with a mean mortality of 78 and 75.6% on average considering all

first to third sprays, respectively; while, the pest population in the untreated check keeps on increasing in the same period. The effectiveness of the tested bio-pesticides was lower in comparison to Adiha experimental site. This could be due to variation in environmental conditions, as temperature in Adiha is higher than that of Agibe and it may contribute in affecting the performance of the botanicals as well as the pest population dynamics. However, still they are effective, promising and potential in controlling the pest particularly if integrated with other control techniques through the integrated pest management perspectives. The same way with the results observed in Adiha, the chemical insecticide, dimethoate 40% EC showed the highest effectiveness in reducing the whiteflies population, with an average 87.15% mortality recorded on the first to third spray (Table 4).

Table 4. Effect of various botanicals on the population of woolly whitefly, (*Aleurothrixus floccosus*) on orange trees in Agibe, Tigray, Northern Ethiopia, during the 2018 dry season.

Treatments	Percent mortality of woolly whitefly insects (reduction in whitefly population compared to the count made before sprays)								
	First spray			Second spray			Third spray		
	CBS [*]	7DAS ^{**}	14DAS	CBS	7DAS	14DAS	CBS	7DAS	14DAS
Neem seed	128 ^a	83.6 ^b	82.0 ^b	28 ^{bc}	76.1 ^b	77.1 ^b	9.3 ^b	74.7 ^b	74.7 ^b
Neem leaf	111 ^a	70.9 ^c	71.2 ^c	32 ^{bc}	67.8 ^d	69.8 ^c	9.3 ^b	64.7 ^b	64.3
Tree tobacco	117 ^a	81.6 ^b	81.7 ^b	21 ^c	73.1 ^{bc}	75.8 ^{bc}	8.0 ^b	70.6 ^b	70.6 ^b
Mexican marigold leaf	122 ^a	72.3 ^c	72.0 ^c	37 ^b	70.9 ^{cd}	70.9 ^c	10.7 ^b	64.6 ^b	64.3 ^b
Dimethoate 40% EC	124 ^a	89 ^a	88.7 ^a	22 ^c	81.0 ^a	83.9 ^a	6.7 ^b	89.8 ^a	90.5
Untreated control	118 ^a	0.0 ^d	0.0 ^d	152 ^a	0.0 ^e	0.0 ^d	195.3 ^a	0.0 ^c	0.0 ^c
St. Dev	8.3	1.9	1.9	5.0	1.6	2.2	2.2	4.4	4.2
Mean	119.9	66.2	65.9	48.5	61.5	62.9	39.9	60.7	60.7
p-value	0.257	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
CV (%)	6.9	2.9	2.8	10.3	2.7	3.6	5.6	7.2	6.9

Note: ^{*}CBS: whitefly insects count 1 day before spraying;

^{**}DAS: days after spraying;

[Within each column, means followed by the same letter do not differ significantly according to Tukey's Honestly Significant Difference (HSD) test at 0.05 probability level].

The present study is in-line with the findings of Iqbal et al. (2015) that reported indigenous plant extracts viz., tumha (*Citrullus colosynthis*), datura (*Daturainnoxia*), neem (*Azadirachta indica*), castor (*Ricinus communis*), hing (*Ferulaasafetida*), eucalyptus (*Eucalyptus*

spp.) bitter melon (*Momordica charantia*) and garlic (*Allium sativum*) are effective in reducing the population of sucking insect pests, among which neem leaf extract was the most effective followed by garlic. McKenna et al. (2013) also reported that *Melia* fruit extracts, neem oil and abamectin, decreased the larvae population significantly to lower numbers than that of the control at 10 days after each spray application. However, they observed that the decrease in larval population caused by neem oil and abamectin was significantly higher than that of *Melia* extracts. Another experiment conducted in Ghana, indicated that the number of whiteflies were significantly lower in the neem seed extract- treated plants compared to the control plants but significantly higher than the karate - sprayed plants (Francis, 2010). Similarly, in line to findings of the present study, in Florida it was indicated that a sucrose ester extract surfactant derived from tobacco provided the best control, followed by sunspray (Ultra-Fine Spray Oil) in killing and repelling whiteflies (*Bemisia argentifolii*) on tomatoes (William and Pat, 1995). Rashid et al. (2003) indicated that dimethoate 40% EC has effectively controlled spiraling whitefly of guava.

Table 5. Effect of different botanicals on the population of armored scale insects (*Aonidiella aurantii*) on orange plants in Adiha, during 2018.

Treatments	Percent mortality of scale insects (reduction in the number of scale insects compared to the count recorded before spraying)								
	First spray			Second spray			Third spray		
	CBS *	7DAS **	14DAS	CBS	7DAS	14DAS	CBS	7DAS	14DAS
Neem seed	395 ^a	79.3 ^b	78.1 ^b	101 ^{bcd}	64.6 ^b	66.7 ^b	34 ^{bc}	64.4 ^b	67.5 ^b
Neem leaf	406 ^a	71.0 ^c	72.3 ^c	119 ^{bc}	60.5 ^b	63.1 ^b	45 ^b	61.6 ^b	64.0 ^b
Tree tobacco	392 ^a	78.1 ^b	77.9 ^b	84 ^{cd}	62.9 ^b	66.6 ^b	28 ^{bc}	64.7 ^b	67.9 ^b
Mexican marigold leaf	394 ^a	70.5 ^c	71.0 ^c	128 ^b	60.3 ^b	62.2 ^b	49 ^b	57.3 ^b	60.8 ^b
Dimethoate 40% EC	418 ^a	86.6 ^a	86.3 ^a	68 ^d	84.9 ^a	87.4 ^a	9 ^c	83.8 ^a	85.1 ^a
Untreated control	408 ^a	0.0 ^d	0.0 ^d	453 ^a	0.0 ^c	0.0 ^c	497 ^a	0 ^c	0.0 ^c
StDev	13.9	1.7	2.0	15.5	2.3	2.7	9.9	2.8	3.7
Mean	402.1	64.2	64.3	158.8	55.5	57.7	109	89.0	55.3
p-value	0.23	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
CV (%)	3.5	2.6	3.1	9.8	4.1	4.7	9.2	3.2	6.6

Note: * CBS: scale insects count 1 day before spraying;

** DAS: days after spraying; St. Dev: standard deviation; CV: coefficient of variation.

[Within each column, means followed by the same letter do not differ significantly according to Tukey's Honestly Significant Difference (HSD) test at 0.05 probability level].

3.3. Effectiveness of different Botanicals on Armored Scale Insects *Aonidiella aurantii*

As shown in table 5, there was statistically significant ($P=0.001$) difference among treatments with regard to the records at seven and fourteen days after treatment in Adiha of all the spray applications. There was high reduction in the number of insect pest compared to the count before spray treatment applications. The overall mean number of armored scales before spray was 402.1, was reduced by 60.5% and 73% after the consecutive sprays at seven days intervals. Taking averages of the records on the three sprayings, the highest mean percent mortality (reduction in the number of scale insects compared to the count recorded before spraying) was obtained from the insecticide dimethoate 40% EC (85.7%) followed by neem seed and tree tobacco extracts that resulted in 70.1 and 65.4% mortality, respectively. Interesting enough, these botanicals performed well in both locations displaying moderate to high mortality records of the armored scale insects, which were comparable to the effectiveness of the synthetic insecticide.

In a similar study, Yingfang et al. (2016) conducted over two years, revealed that products based on a mineral oil (SuffOil-X[®]), azadirachtin (Molt-X[®]), significantly reduce the number of armored scales.

Table 6. Effect of different botanicals on armored scale insects (*Aonidiellaaurantii*) on orange trees in Agibe, during 2018.

Treatments	Percent mortality of scale insects								
	First spray			Second spray			Third spray		
	CBS *	7DAS **	14DAS	CBS	7DAS	14DAS	CBS	7DAS	14DAS
Neem seed	713 ^a	73.3 ^b	71.5 ^b	185 ^{bc}	73.0 ^b	76.0 ^b	42 ^{bc}	69.0 ^b	73.7 ^b
Neem leaf	694 ^a	62.4 ^c	63.9 ^c	250 ^{bc}	62.6 ^c	68.2 ^c	79 ^{bc}	61.4 ^c	64.6 ^c
Tree tobacco	746 ^a	64.9 ^c	67.4 ^{bc}	170 ^{bc}	71.2 ^b	75.3 ^b	50 ^{bc}	66.7 ^{bc}	72.3 ^b
Mexican marigold leaf	749 ^a	62.4 ^c	64.1 ^c	269 ^b	64.1 ^c	66.5 ^c	86 ^b	62.3 ^c	62.0 ^c
Dimethoate 40% EC	672 ^a	86.6 ^a	88.0 ^a	109 ^c	87.8 ^a	88.7 ^a	13 ^c	86.6 ^a	88.5 ^a
Untreated control	680 ^a	0.0 ^d	0.0 ^d	847 ^a	0.9 ^d	0.0 ^d	898 ^a	0.0 ^d	0.0 ^d
StDev	31.6	2.1	2.5	56.9	1.3	2.3	24.6	2.3	2.8
Mean	709.1	58.2	59.12	304.9	59.8	62.5	194.4	57.7	60.2
p-value	0.043	0.001	0.001	0.001	0.001	0.001	0.001	0.001	0.001
CV (%)	4.5	3.5	4.2	18.7	2.2	3.7	13.2	4.0	4.6

Note: * CBS: scale insects count 1 day before spraying;

** DAS: days after spraying. St. Dev: standard deviation; CV: coefficient of variation.

[Within each column, means followed by the same letter do not differ significantly according to Tukey's Honestly Significant Difference (HSD) test at 0.05 probability level].

An assessment was made to observe the effect of different botanicals and a standard chemical insecticide on the armored scale insect pest. As shown in table 6, there was statistically significant ($P = 0.001$) difference among treatments at 7 DAS and 14 DAS during all the spray applications in Agbe. All treatments display high level of reduction of the scale insect population compared to the count made before treatment applications. The mean count of the armored scale before spray begins was 709.1 which latter reduced by 57% and 72.6 % after the two consecutive sprays at the seven days intervals. Considering all the first, second and third sprays, the highest mean percent mortality of scale insects were observed from spray of the insecticide dimethoate 40% EC (88.4%) followed by neem seed extract (73.7%) and tree tobacco extracts (71.7 %)(Table 6). However, synthetic insecticides possess inherent toxicities in general and dimethoate in particular that endanger the health of the farmers, consumers and the environment. Thus, hazardous effects on human health led to resurgence in interest in botanical insecticides due to their minimal costs and lesser environmental side effects (Prasannath, 2016). Botanicals used in this experiment that showed a good performance therefore are advantageous over the broad-spectrum conventional insecticide. They affect only target pests and closely related organisms, decomposed quickly and leave the food residue free and provide a safe environment to live. As a component in the integrated pest management system, these botanical insecticides can greatly reduce the quantity of synthetic insecticides applied and contribute to sustainable crop cultivation.

4. CONCLUSIONS

The experiment has had a focus to investigate the effectiveness of plant origin bio-pesticides on insect pests of orange trees. The present study showed that all the tested plant extracts were effective in controlling the target pests. It can be concluded that the botanicals particularly the neem seed extracts followed by tree tobacco are as effective as the chemical insecticide, dimethoate 40 % EC, even superior in some cases, in reducing the target pests vis., citrus leafminer, armored scale and citrus whitefly. They can hence reduce the damage to the citrus orange if used properly, as they perform superior compared to the untreated control as well as than the other treatments in both experimental sites. The fact that plants treated with neem leaf and tree tobacco had the lowest insect pest counts comparable to the synthetic insecticide (dimethoate), it indicates that these botanicals may have some anti-feeding (neem seed extract)

and active ingredients with toxic properties (Tree tobacco) that has helped to effectively act against the target pests and minimize damage to the plants (Bushey, 1951). Therefore, these botanicals can be used as part of the integrated management strategy for controlling the target pests, namely citrus leafminer, whiteflies and scales.

As infestations were associated with bud breaking and new leaf flush growth, spray applications should coincide with this stage of the crop to minimize damage. Particularly, spraying activities for controlling whitefly should cover the undersides of leaves, as great numbers of nymphs were found there.

These botanicals could be better options for the management of scale insects by farmers, as they are locally available, less costly and environmentally safe. However, further work is required to determine the appropriate frequency of application and concentration (rate/dosage of application) of these botanicals in order to come out with the best method of application that will provide the farmer with the best benefit other than the use of chemical pesticides.

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6. CONFLICT OF INTERESTS

There are no conflicts of interest.

7. REFERENCE

- Ayalew, G., 2005. Comparison among some botanicals and synthetic insecticides for the control of onion thrips (*Thrips tabaci*, Lind.) (Thysanoptera: Thripidae). Proceedings of the 13th Annual Conference of the Crop Protection Society of Ethiopia (CPSE), Addis Ababa, Ethiopia.
- Bushey, R.L. 1951. Plants That Help Kill Insects. Year book of Agriculture CABI Crop Protection Compendium. 2010. Navel orange datasheet.

- Elizabeth Grafton-Cardwell. 2010. Citrus leafminer research. Agriculture and Natural Resources, University of California.
- Francis, A. 2010. Effects of two neem kernel extracts in the control of whitefly (*Bemisia tabaci*) on tomato. M.Sc thesis, Department of Theoretical and Applied Biology, College of Science, Kwame Nkrumah University of Science and Technology, PG1814107.
- Iqbal, J., Ali, H., Hassan, W & Jamil, M.2015. Evaluation of indigenous plant extracts against sucking insect pests of okra crop. *Pakistan Entomologist*, **37**: 39-44.
- Jensen, L & Simko, B. 2001. Alternative Methods for Controlling Onion Thrips (*Thrips tabaci*)in Spanish Onions. Malheur County Extension Service, Clint Shock and Lamont,Saunders, Malheur Experiment Station, Oregon State University, Ontario.
- Kassa Melese A. 2015. Opportunities and Potential in Ethiopia for Production of Fruits and Vegetable. *International Journal of African and Asian Studies*, ISSN: 2409-6938, **15**: 41-48
- Lacey, K., Ramsey, H & Hoffman, H. 2009. Growing healthy citrus. Government of Western Australia Department of Agriculture and Food. <http://archive.agric.wa.gov.au/objtwr> [Accessed 02 Feb. 19].
- Lazaneo, V. 2008. Citrus for the home garden. University of California Cooperative Extension. Available at: <http://www.mastergardenerssandiego.org> [Accessed 02 March 2018].
- Louis Feinstein. 2010. Insecticides from plants. Process for Extracting Alkaloidals from Plants with Aqueous Ammonia-Ethylene Bichloride Mixture. P.1-6
- Mayer, D. F. Lunden, J. D & Rathbone, L. 1987.Evaluation of insecticides for *Thrips tabaci* (Thysanoptera:Thripidae) and effects of thrips on bulb onions. *Journal of economic entomology*, **80**: 930-932.
- Mc Kenna, M. M. Hammad, E. M & Farran, M. T. 2013. Effect of *Melia azedarach* (Sapindales: Meliaceae) fruit extracts on citrus leafminer *Phyllocnistis citrella* (Lepidoptera: Gracillariidae). *SpringerPlus*, **2(1)**: 144. doi:10.1186/2193-1801-2-144
- Mossa, A.T.H. 2016. Green pesticides: Essential oils as bio pesticides in insect-pest management. *Journal of Environmental Science Technology*, **9**: 354-378.
- Muhammad, F. S., Abubakar, R., Muhammad, M., Kanwer, S. A., Waqas, R & Hafiz, F.H. 2016. Effect of botanicals on the infestation of citrus leaf miner. *Phyllocnistis citrella* stainton. *Journal of Entomology and Zoology Studies*, **4(4)**: 1335-1340

- Nault, B. A., Hsu, C. L & Hoepting, C. A. 2013. Consequences of co-applying insecticides and fungicides for managing *Thrips tabaci* (Thysanoptera: Thripidae) on onion. *Pest management sciences*, **69(7)**: 841-849, doi: 10.1002/ps.3444 (Epub 2012 Nov 29).
- Prasannath, K. 2016. Botanical insecticides - special reference to horticultural insect pest management: a review. *International Journal of Agricultural Research Review*, **1(5)**: 14-18.
- Rao, R. T., Rajanikumari, V.S., Devakil, K & Sarada, G. 2014. Evaluation of plant products against Citrus leaf miner, *Phyllocnistis citrella* Stainton. *Pest management in horticultural ecosystems*, **20(2)**: 234-235
- Rao, R. P., Rao, K & Pavana, J. K. 2015. Efficacy of certain natural insecticides against citrus leaf miner, *Phyllocnistiscitrella* Stainton as prophylactic and curative measures on *Sathgudi* sweet orange. *Pest management in horticultural ecosystems*, **21(1)**: 11-15
- Rashid, M. M., Hossain, M. M., Alam, M. Z., Ibrahim, M & Bhuiyan, M. K. A. 2003. Seasonal abundance and control of spiraling whitefly, *Aleurodicus disperses* russel on guava. *Pakistan Journal of Biological Science*, **6(24)**: 2050-2053.
- Shiberu, T., Negeri, M & Selvaraj, T., 2013. Evaluation of Some Botanicals and Entomopathogenic Fungi for the Control of Onion Thrips (*Thrips tabaci* L.) in West Showa, Ethiopia. *Journal of Plant Pathology Microbiology*, **4**: 161, doi:10.4172/2157-7471.1000161.
- Valand, V.M., Patel, J.C & Patel, M.C. 1992. Bio-efficacy of insecticides against citrus leafminer on Kagzi lime. *Indian Journal of Plant Protection*, **20(2)**: 212-214.
- Williams, G & Pat. 1995. Oil, soap, surfactant, and garlic vs. whiteflies on tomatoes. *Horticulture ideas*. May, p. 55-56.
- Yingfang, Xiao., Runqian, Mao., Singleton, L & Arthurs, A. 2016. Evaluation of Reduced-Risk Insecticides for armored scales (Hemiptera: Diaspididae), infesting Ornamental Plants. *Journal of agricultural and urban entomology*, **32(1)**: 71-90.