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Greenhouse and field evaluation of essential oil formulations on *Nilaparvata lugens* Stal and their natural enemies

Tri Lestari Mardiningsih* and Ma'mun
Indonesian Spice and Medicinal Crops Research Institute (ISMCRI), Bogor 16111, West Java, Indonesia

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Citronella and clove (CiC) and lemongrass and clove (LeC) mixtures, each at the ratio of 1:1, citronella, lemongrass, and clove oils and solution without EOs (blank) were tested. To determine the effect of the EOs on oviposition deterrent and mortality of *N. lugens* adults, the rice plants were sprayed with each solution, allowed to dry, then the seedling was kept by a plastic cage that had the top part covered with perforated cloth. Three pairs of *N. lugens* adults were introduced into the cage. The observation was conducted on the number of eggs laid and the mortality of adults. An experiment on the mortality of nymphs was carried out by spraying the rice seedlings containing 10 nymphs and caged as above. The field experiment was conducted by spraying the rice plants with those solutions. The formulas of CiC and LeC at the concentration of 10 ml/L deterred oviposition of *N. lugens* by 64.6 and 57.4% respectively. Both EOs caused mortalities in adults at 43.3% and nymphs of *N. lugens* as much as 60-64%. Those formulas applied twice a week were effective to control *N. lugens* with the value of efficacy 70.2 and 50.4% respectively and safe to natural enemies in the field.

Keywords: Essential oil formulations, Greenhouse and field evaluation, Mortality, Natural enemies, *Nilaparvata lugens*.

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Introduction

Brown planthopper, *Nilaparvata lugens* Stal (Hemiptera: Delphacidae) is an important pest in rice plants. This pest attack caused damage by directly releasing phloem fluid, causing damage to plants by producing a phenomenon called 'hopper burn'[1]. Control of *N. lugens* by using a botanical insecticide, namely citral incorporated to guar gum-based film could be used to control *N. lugens*[2]. Essential oil and nano essential oil of star anise (*Illicium verum*) and lemongrass (*Cymbopogon citratus*) also showed insecticidal activity to *N. lugens*[3].

Controlling insect pests with synthetic insecticides gave quick results, effective, and efficient. However, the use of those insecticides unwisely caused bad side effects. Therefore, several efforts were conducted to reduce the use of synthetic insecticides. One of the efforts is the use of botanical insecticides that are considered more environmentally friendly.

Plant-based insecticides produced a lot of compounds that could act as an insecticidal activity to *N. lugens*[4]. Some aromatic plants showed good efficacy both in contact and fumigation applications[5]. Citronella, lemongrass, ageratum, java turmeric, and neem oils caused mortality and oviposition deterrent to *Crocidolomia pavonana*[6].

Lemongrass oil was toxic to *Aphis citricola* at 169,6 mg/L, 24 hours after treatment. Field trial of 20% mixture of *Ligusticum chuanxiong* (LCO) + Lemongrass oil (LO) EC against *A. citricola* caused significant control. The control effects of 500 times dilution were 90.06 and 87.24% which were equal to 1,000 times dilution of 20% imidacloprid EC. While the control at 1,000 times dilution of 20% LCO + LO EC was still more than 80% seven days after the treatment[7].

Clove oil had insecticidal activities (toxic and deterred oviposition) to *Liriomyza trifolii*, *Bemisia argentifolii*, and *Tuta absoluta* on tomato plants[8]. Clove oil influenced the gametogenesis of *S. frugiperda* ovarioles, which negatively impact its reproduction[9].

A previous study revealed that mixtures of citronella and clove oils (CiC), lemongrass and clove oils (LeC), and ageratum and clove oils in various ratios were two to three times more toxic and had oviposition deterrent effect than individual EOs on *Crocidolomia pavonana* and *Helopeltis antonii*. CiC and LeC in the ratio of 1:1 showed the lowest number of eggs laid by *C. pavonana* and

*Correspondent author
Email: tri_mardiningsih@yahoo.com

H. antonii[10]. Testing EOs formulas to another kind of insect pest should be tried, therefore a study to evaluate the mixtures of EOs formulas (citronella, clove, and lemongrass) in the ratio of 1:1 to mortality and oviposition deterrent of *N. lugens* in the greenhouse and its population and natural enemies population in the field was carried out. CiC and LeC were formulated to provide practical usage in some important pests. Evaluation of the formula is required.

Materials and Methods

Preparation of plants and test insects

Plant materials used as the food of *N. lugens* were rice plants, the variety of IR 64. Rice seeds were sown in muddy media, after around 10 days they were transplanted into buckets containing mud. Rice seedlings at the age of more than a month old were used for testing. *N. lugens* nymphs were collected from rice plantations in Bogor District, West Java, Indonesia. The nymphs were reared in the greenhouse of the Indonesian Spice and Medicinal Crops Research Institute (ISMCRI). After developed to become adults and copulation, the offsprings were used for testing.

EOs formulation

EOs were obtained by steam distillation. The EOs used in this experiment were citronella, clove, and lemongrass. Citronella leaves were distilled at the Experimental Station of Manoko, Lembang, Bandung. Clove leaves were distilled in Leuwiliang, redistilled at Test Laboratory, ISMCRI, Bogor. Lemongrass stems were distilled by a farmer in Cianjur. The chemical contents of each EOs were analyzed with GC-MS at Health Laboratory, Jakarta.

EOs tested were 2 essential oil mixtures that were effective from the result of the 2015 study[10]. Each essential oil was mixed with clove oil, the stock solution was made by following per under the ratio of EO used, i.e., 1:1, then it was mixed with the blank. Blank was in the form of solvent and surfactant/emulsifier (consisted of tween 80 (10%), turpentine (59%), and teepol (10%)). The use of each mixing was as much as 10 and 5 mL. Besides that, every single essential oil was mixed with blank so that becoming emulsifiable concentrate (EC) formula. The amount of EO in each formula was 30%. Blank was without EO.

Oviposition deterrent and mortality of *N. lugens*

The experiment was conducted in a randomized complete block design (RCBD), with 13 treatments and repeated 5 times. The treatments were formulations of i) citronella:clove oil 1:1 + blank 10 mL/L, ii) citronella:clove oil 1:1 + blank 5 mL/L, iii) lemongrass:clove oil 1:1 + blank 10 mL/L, iv) lemongrass:clove oil 5 mL/L, v) citronella oil + blank 10 mL/L, vi) citronella oil + blank 5 mL/L, vii) lemongrass oil + blank 10 mL/L, viii) lemongrass oil + blank 5 mL/L, ix) clove oil + blank 10 mL/L, x) clove oil + blank 5 mL/L, xi) blank (tween 80, turpentine, teepol) 10 mL/L, xii) blank 5 mL/L, and xiii) control.

Oviposition deterrent was tested in a greenhouse by sprayed rice plant with tested formula, let to dry up, thus the plant was caged with a plastic cage which the top was covered with perforated cloth, then 3 pairs of *N. lugens* adults were introduced into the cage. The adults were caged for one day, the next day the adults were transferred and caged to the rice plant which was treated. Transferring and caging was conducted 3 times (4 days) so that for one replication, 52 rice seedlings were needed. Mortality of adults was also observed up to four days after treatment. If an insect died, it was not replaced with a new one. The method is as per Setiawati *et al.* [11]. After that, the plant was dismantled and observed under a binocular microscope to count the number of eggs laid. The effective repellency for each EOs was calculated using the following formula[11]:

$$ER\ (\%) = \frac{NC - NT}{NC} \times 100\%$$

Where, ER is effective repellency (%), NC is the number of egg in control (+ blank), and NT is the number of egg in treatment. Mortality of adults was observed every day till four days after application.

Mortality of *N. lugens* nymphs

The experiment was arranged in a randomized complete block design (RCBD), with 13 treatments and repeated 5 times. The treatments were the same as the test of oviposition deterrent and mortality of *N. lugens*. Treatment was carried by spraying every 10 nymphs (around 2 weeks old) on rice seedling each with concentration tested. Furthermore, the nymphs and the plant were caged with a plastic cage which the top was covered with perforated cloth. Observation of the nymphs' mortality was done every day so that the mortality is constant.

Field experiment

Test of efficacy EOs in the field was carried out at farmer's rice plantation in Sindang Barang, Bogor, West Java (6°34'17, 52" S and 106°45'30, 30" E). The experiment was conducted in a randomized complete block design (RCBD), with 14 treatments and was repeated 4 times (56 plots of rice clumps). The treatments were as in the test of oviposition deterrent and mortality of *N. lugens* added with synthetic insecticide (imidacloprid). The concentration used was 10 mL/L and the frequencies for spraying were once and twice a week, except those of imidacloprid and control were once a week. The plot size was 5 m x 8 m, consisting of around 640 rice clumps.

For the two-time spraying treatment, observation of *N. lugens* population was conducted once a week. Observation of *N. lugens* population and spraying was conducted if the attack of *N. lugens* has been found. Before the first spraying was carried out, observation of *N. lugens* population was done. Observation of *N. lugens* population and spraying was done a minimum of 5 times. The observation was carried out on 20 sample clumps which were selected systematically from each plot. The parameter observed was the population of *N. lugens*. Supporting data was the population of natural enemies. Data from observation results were then used to count the value of insecticide efficacy tested. If the first observation of *N. lugens* population was not significantly different between treatment plot then the value of insecticide tested was counted with the following formula[12]:

$$EI = \left(\frac{C_a - T_a}{C_a}\right) \times 100\%$$

Where, EI is the efficacy of insecticide tested (%), C_a is *N. lugens* population at control plot after application of insecticide, and T_a = *N. lugens* population at treatment plot after application of insecticide.

If at the first observation of *N. lugens* population is significantly different between treatment plot then the value of insecticide tested was counted with the following formula[13]:

$$EI = \left(1 - \frac{C_a}{C_a} \times \frac{C_b}{T_b}\right) \times 100\%$$

Where, EI is the efficacy of insecticide tested (%), C_a is *N. lugens* population at control plot after application of insecticide, C_b = *N. lugens* population at control plot before application of insecticide, and T_b= *N. lugens* population at treatment plot before application of insecticide.

Data analysis

Data were analyzed with analysis of variance (ANOVA). Test of significance difference was done using Duncan test at the level of 5%.

Results and Discussion

Oviposition deterrent of *N. lugens*

Results on oviposition deterrent showed that treatment of a mixture of citronella and clove oils (1:1) at the concentration of 10 mL/L showed the highest suppression of the number of eggs laid up to 4 days after treatment compared with other treatments and control. Then, followed by the treatment of the mixture of lemongrass and clove oils (1:1) at the concentration of 10 ml/L, the mixture of citronella and clove oils (1:1), and the mixture of lemongrass and clove oils (1:1) at 5 ml/L (Table 1).

Table 1 showed that citronella oil deterred oviposition of *N. lugens*. This was in line with the study of Setiawati *et al.*[11], which stated that citronella oil applied on chili plant could inhibit oviposition and decreased egg hatching of *Heliothis armigera*. Citronellal showed repellent activity to nymphs and adults of *Lygus hesperus*[14]. The main active ingredient of citronella oil was observed to be citronellal (44.85%)[10]. Clove oil decreased F1 offsprings, the damage of seed, weight loss of mung bean, and raised the rate of adult emergence inhibitory[15]. Citronella and lemongrass oil deterred the oviposition in *Crocidolomia pavonana*[6]. The mixture of citronella and clove oil and the mixture of lemongrass and clove oil in alkyl glycerol phthalate caused a higher oviposition deterrent of *C. pavonana* and *Helopeltis antonii* compared to single oil application[10]. The mixture of citronella and clove oil and the mixture of clove and lemongrass oil in tween 80, turpentine, and teepol at the concentration of 10% also caused a higher oviposition deterrent in *H. antonii* on cucumber fruit (as the host)[16]. The mixture of citronella and clove oil and the mixture of clove and lemongrass oil in tween 80, turpentine, and teepol also caused a higher oviposition deterrent in *N. lugens* on rice variety of Ciherang compared to single oil application[17]. Moreover, in this study mixture of citronella and clove oil increased the activity of the oviposition deterrent in *N. lugens*. The best treatment was the mixture of citronella and clove oil which gave the highest effective repellency (64.6%). Compared with individual EO, it increased oviposition deterrent two to three times.

There were effects of blank on the formula because the main component of the solution was turpentine. The number of eggs laid on the treatment of blank (10 ml/L) was not significantly different from other treatments but significantly different from control. Turpentine also had a higher repelling influence on *Cryptorrhynchus lapathi* L. (Coleoptera: Curculionidae) than control[18]. The blank contained turpentine which contains a natural compound (β-pinene) that could be used as a larvicide in *Plutella xylostella* and *Mythimna separata*[19].

Mortality of *N. lugens* adult

Results of test on mortality of *N. lugens* adult was the highest on treatments of citronella: clove oil and lemongrass: clove oil at 10 mL/L and clove oil 10 mL/L, but not significantly different up to 4 days after treatment compared with other treatments and control (Table 2). The addition of clove oil proved to increase the mortality of *N. lugens* adults at a concentration of 10 ml/L. Citronella and lemongrass oil caused mortality of *C. pavonana* larvae[6]. The addition of clove oil proved to increase the killing activity of *C. pavonana* larvae, it could be seen that the mortalities of *N. lugens* of the two mixtures at a concentration of 10 ml/L were the highest. Citronella oil (*Cymbopogon winterianus*) showed insecticidal activity against *Acanthoscelides obtectus*[20].

Clove oil decreased respiratory rate, avoided mobility on surface treated, and reduced population growth rate of *Sitophilus zeamais*[21]. The oil caused mortality in *S. zeamais* and *A. obtectus*[22]. Clove oil also had a repellent activity to *Aedes aegypti*[23]. Eugenol was the least toxic to *Tribolium castaneum* and *Rhyzopertha dominica*[24]. Eugenol was the main active ingredient of clove oil (77.54%)[10]. Clove oil

Table 1 — The average number of eggs laid by *N. lugens*

Formulations	Ratio	The average number of eggs on the day				ER
		1	2	3	4	
Citronella:clove + blank (10 mL/L)	1:1	14.8^{a}	29.2^{d}	47.0^{d}	54.0^{e}	64.6
Citronella:clove + blank (5 mL/L)	1:1	38.6^{a}	62.6^{bcd}	80.2^{cd}	89.4^{cde}	49.0
Lemongrass:clove + blank (10 mL/L)	1:1	11.2^{a}	31.8^{cd}	53.8^{d}	65.0^{de}	57.4
Lemongrass:clove + blank (5 mL/L)	1:1	35.6^{a}	79.4^{bcd}	113.0^{bc}	117.6^{bcd}	32.9
Citronella + blank (10 mL/L)	-	48.0^{a}	68.8^{bcd}	99.4^{bcd}	121.8^{bcd}	20.2
Citronella + blank (5 mL/L)	-	68.6^{a}	93.6^{bc}	125.0^{bc}	138.8^{bc}	20.9
Lemongrass + blank (10 mL/L)	-	58.2^{a}	91.8^{bcd}	139.0^{bc}	147.8^{bc}	3.1
Lemongrass + blank (5 mL/L)		59.4^{a}	105.2^{ab}	147.4^{bc}	163.0^{b}	7.1
Clove + blank (10 mL/L)		75.2^{a}	102.4^{ab}	142.8^{bc}	165.8^{b}	-8.7
Clove + blank (5 mL/L)		73.6^{a}	111.0^{ab}	136.2^{bc}	150.4^{bc}	14.3
Blank (10 mL/L)		67.2^{a}	94.6^{bcd}	143.8^{bc}	152.6^{bc}	
Blank (5 mL/L)		84.4^{a}	119.2^{ab}	155.4^{b}	175.4^{b}	
Control		136.8^{a}	188.6^{a}	240.6^{a}	269.6^{a}	
CV (%)		1.58	3.35	3.17	2.61	

Numbers followed by the same letters are not significantly different on 5% DMRT. ER = percent effective repellency

Table 2 — The average mortality of adults and nymphs of *N. lugens*

Treatments	Ratio	Mortality of adults (%) on day				Mortality of nymphs (%) at the day of			
Formulations		1	2	3	4	1	2	3	4
Citronella:clove + blank (10 mL/L)	1:1	0	26.7^{a}	43.3^{a}	43.3^{a}	30^{a}	52^{a}	64^{a}	64^{a}
Citronella:clove + blank (5 mL/L)	1:1	0	16.7^{a}	33.3 ^{a}	33.3^{a}	30^{a}	38^{abc}	42^{abc}	42^{abc}
Lemongrass:clove + blank (10 mL/L)	1:1	0	23.3^{a}	40.0^{a}	43.3^{a}	32^{a}	52^{ab}	60^{ab}	60^{ab}
Lemongrass:clove + blank (5 mL/L)	1:1	0	16.7^{a}	20.0^{abc}	26.7^{ab}	16^{a}	28^{abc}	28^{cde}	28de
Citronella + blank (10 mL/L)	-	0	13.3^{a}	23.3^{abc}	33.3^{a}	14^{a}	36^{b}	38^{abc}	38^{abc}
Citronella + blank (5 mL/L)	-	0	3.3^{a}	6.7^{cd}	13,3^{bc}	18^{a}	32^{bc}	34^{abcd}	34^{abcd}
Lemongrass + blank (10 mL/L)	-	0	6.7^{a}	13.3^{bcd}	23.3^{abc}	12^{a}	24^{abc}	30^{bcd}	30^{bcd}
Lemongrass + blank (5 mL/L)		0	0^{a}	6.7^{cd}	13.3^{bc}	4^{a}	14^{cde}	14^{def}	14^{def}
Clove + blank (10 mL/L)		0	13.3^{a}	33.3^{ab}	40.0^{a}	22^{a}	34^{abc}	34^{abcd}	34^{abcd}
Clove + blank (5 mL/L)		0	6.7^{a}	13.3^{bcd}	23.3^{abc}	24^{a}	24^{bcd}	24^{cdef}	24^{cdef}
Blank (10 mL/L)		0	3.3^{a}	10.0^{cd}	23.3^{abc}	0^{a}	2de	2^{ef}	2^{ef}
Blank (5 mL/L)		0	0^{a}	0^{d}	10.0^{c}	0^{a}	0^{e}	0^{f}	0^{f}
Control		0	0^{a}	0^{d}	3.3^{c}	0^{a}	0^{e}	0^{f}	0^{f}
CV (%)		-	0.13	0.11	0.10	0.11	0.13	0.11	0.11

Numbers followed by the same letters are not significantly different on 5% DMRT caused mortality and repellence in adult *Sitophilus granarius*, so it has the potential to prevent insecticide resistance development[25]. It also had insecticidal activity, reduced growth rate and thwart the emergence of offspring of *Callosobruchus maculatus*[26]. Clove bud oil was toxic and had a repellent activity in *Dermanyssus gallinae*[27]. Eugenol showed acaricidal activity against *Psoroptes cuniculi*[28].

Lemongrass oil had insecticidal activities in *T. castaneum* adults[29]. The oil (*Cymbopogon citratus* (DC.) Stapf.) caused mortality (61.43%), oviposition inhibition (4.00%), and inhibition of adult emergence (10.32%) of *C. maculatus*[30]. It showed insecticidal and repellent activities in *S. granarius*[31]. The EOs of lemongrass (*C. citratus*) and citronella (*C. nardus*) were toxic and showed repellent activities in *Oryzaephilus surinamensis* and *S. zeamais*[32]. Clove and lemongrass oils had high fumigant toxicity to *Frankliniella schultzei* and *Pseudococcus jackbeardsleyi*[33].

Mortality of *N. lugens* nymphs

Results of test on mortality of *N. lugens* nymphs showed that the treatment of the mixture of citronella and clove and lemongrass and clove mixture in the ratio of 1:1 at the concentration of 10 mL/L and clove 10 mL/L showed the highest mortality of *N. lugens* adults up to 3 days after treatment compared with other treatments and control (Table 2). Treatment of blank did not affect nymph mortality, which may be because of different application methods. This case is the same as the research on Ciherang variety of rice plants on the mortality of nymphs, the two formulas were not mutually different (63.33 and 56.67 nymphs), but the two formulas were significantly different from blank (6.67 nymphs)[17]. Citronella and lemongrass oils also caused the mortality of *C. pavonana* larvae[6]. Increasing the killing activity of *C. pavonana* larvae was a result of clove oil addition, it could be seen that the mortalities of *N. lugens* of the two mixtures at the concentration of 10 mL/L were the highest. Those two formulas also caused high mortality of nymphs of *H. antonii*[16]. In a study[17] which used rice, the variety of Ciherang, the mortality of *N. lugens* at 4 days after the application was also higher (60.00 and 56.70 adults). The population of *N. lugens* at 21 days after infestation was not significantly different in IR 64 and Ciherang varieties (both are resistant varieties)[34]. Citronella oil (*C. winterianus*) changed biochemical and histological conditions of *Spodoptera frugiperda* larvae and caused reproductive failure[35]. Citronella oil resulted in changes in midgut morphology and stored resource reduction in the body fat of *S. frugiperda* larvae[36].

These studies showed that mixing the EOs could raise the mortalities of *N. lugens* nymphs and adults to nearly twice the mortality on tests with individual EO. In this mortality test, the high mortality occurred one day after application. There were some additional mortalities observed on the second and the third day after treatments. Blank did not affect the mortalities of adults and nymphs, however, it influenced to oviposition deterrent of *N. lugens*.

Field experiment

The field experiment showed that the first week after application, treatment LeC with spraying frequency of twice a week in the concentration of 10 mL/L showed the highest efficacy value (64.3%). The first week after application, for LeC with spraying frequency twice a week, the value of efficacy was the highest, the same with imidacloprid (synthetic), followed by CiC 2x. In the third week after the application, the results were the same as the previous week. In the fourth week after the application, the increase of *N. lugens* population occurred in all treatment plots, the highest efficacy value occurred for the treatment of CiC 2x (Table 3). This experiment was in line with the experiment in the greenhouse where CiC and LeC at the concentration of 10 mL/L gave the best result. The solutions of CiC and LeC at the concentration of 10 mL/L with the application interval of twice a week gave results as good as the synthetic insecticide (imidacloprid). Results of EOs test to natural enemies population of *N. lugens*, i.e., *Cyrtorhinus lividipennis* (Hemiptera:Miridae), *Paederus tamulus* (Coleoptera:Staphylinidae), *Lycosa* sp. (Lycosidae), and *Coccinella arcuata* (Coleoptera: Coccinellidae) in the field showed that the average of population addition/reduction in the 4th week after application were not significantly different in all treatments. This meant that EOs were safe for natural enemies.

Results of the study revealed that the mixture of CiC (1:1) and that of LeC (1:1) in the concentration of 10 mL/L could raise oviposition deterrent, mortalities of adults, and nymph of *N. lugens* in the greenhouse. Testing CiC (1:1) and that of LeC (1:1) with the frequency of spraying twice a week in the field also gave a high value of insecticide efficacy (more than 50%) to control the population of *N. lugens*.

Table 3 — The average population of *N. lugens* at various treatments of essential oils in the field

Treatments of formulation	Frequency of application	Population before application	The addition/ reduction of population 1 week after application	EI (%)	The addition/ reduction of population 2 weeks after application	EI (%)	The addition/ reduction of population 3 weeks after application	EI (%)	The addition/ reduction of population 4 weeks after application	EI (%)
CiC + bl 1:1	1 x	820.5^{a}	48.50^{abcd}	9.5	-606.75^{abc}	57.8	-163.00^{a}	82.5	1372.25^{a}	14.6
CiC + bl 1:1	2 x	569.0^{a}	-170.75^{ab}	58.5	-273.75^{d}	79.9	-80.50^{a}	92.2	469.75^{a}	70.2
LeC + bl 1:1	1 x	667.8^{a}	-154.25^{abcd}	45.5	-353.50^{abcd}	72.6	-118.75^{a}	90.9	1018.75^{a}	37.9
LeC + bl 1:1	2 x	717.0^{a}	-244.25^{a}	64.3	-351.25^{abcd}	80.4	-89.75^{a}	94.4	822.00^{a}	50.4
Ci + bl	1 x	756.3^{a}	296.25^{bcd}	-9.6	-819.50^{a}	62.5	-108.25^{a}	77.9	1419.00^{a}	10.4
Ci + bl	2 x	703.3^{a}	-227.75^{a}	39.0	-335.75^{bcd}	59.8	-170.00^{a}	85.9	879.00^{a}	44.3
Le + bl	1 x	573.5^{a}	150.50^{cd}	24.6	-454.00^{abcd}	56.7	-107.50^{a}	71.3	1273.75^{a}	16.6
Le + bl	2 x	723.5^{a}	-170.50^{ab}	42.4	-319.25^{cd}	62.3	-137.00^{a}	82.9	1020.25^{a}	35.2
C + bl	1 x	1426.3^{a}	-140.25^{abcd}	-33.9	-657.00^{ab}	-1.3	-421.50^{a}	63.3	1595.00^{a}	-4.6
C + bl	2 x	524.5^{a}	71.75^{abcd}	37.9	-396.00^{abcd}	67.7	-118.50^{a}	85.5	765.00^{a}	50.8
Blank	1 x	622.0 ^{a}	46.25^{abcd}	29.9	-286.25^{cd}	38,5	-163.50^{a}	61.4	996.50^{a}	29.5
Blank	2 x	449.0 ^{a}	145.00^{cd}	38.2	-352.50^{abcd}	61.1	-109.00^{a}	76.6	1196.25^{a}	22.9
Imidacloprid		626.8 ^{a}	-263.25^{a}	62.1	-264.25^{d}	84.0	-85.25^{a}	97.5	648.50^{a}	61.5
Control		678.3^{a}	282.25^{d}		-339.75^{d}		-55.25^{a}		1157.00^{a}	
CV/KK		6.39	13.86		7.67		14.12		14.72	

Numbers followed by the same letters are not significantly different at 5% DMRT. CiC = citronella : clove, LC = lemongrass : clove, Ci = citronella, Le = lemongrass, C = clove, bl = blank, - = reduction, EI = the value of insecticide efficacy tested

Besides that, those solutions were also safe against natural enemies of *N. lugens* in the field, namely *C. lividipennis* (Hemiptera: Miridae), *P. tamulus* (Coleoptera: Staphylinidae), *Lycosa* sp. (Lycosidae), and *C. arcuata* (Coleoptera: Coccinellidae). This is seen from the average population of the natural enemies' population that was significantly different in all treatments. Intercropping tea with aromatic plants also reduced the population of leafhoppers (*Empoasca onukii* Matsuda) and increased natural enemies in northern China[37]. Moreover, the use of the essential oils of *Artemisia herba-alba*, *A. campestris*, and *A. Absinthium* to control *C. maculatus* and *Bruchus rufimanus* (Coleoptera: Chrysomelidae) showed that the oils were compatible with their parasitoids wasps *Dinarmus basalis* (Hymenoptera: Pteromelidae) and *Triaspis luteipes* (Hymenoptera: Braconidae), natural enemies of those[38]. The use of essential oils of *Coriandrum sativum* and *Nerium indicum* could attract *C. lividipennis*, the main predator of *N. lugens*, which feeds on *N. lugens* eggs and nymphs[39]. Lemongrass oil was tolerant to *Podisus nigrispinus* (Heteroptera: Pentatomidae), the predator of Coleopteran and Lepidopteran species, however, it was toxic to the larvae of *P. nigrispinus*[40]. Clove oil was toxic to the aphid *Rhopalosiphum maidis* and safe to the non-target ladybeetle, *Coleomegilla maculata*[41].

Spraying the citronella and clove oils mixture (1:1) and lemongrass and clove oils (1:1) in the concentration of 10 mL/L on rice plant deterred oviposition of *N. lugens* by 64.6 and 57.4% and caused 43.3% adults mortality. Spraying those formulas on the rice plants containing nymphs caused 64 and 60% of *N. lugens* nymphs in the greenhouse. Those mixtures with the frequency of spraying twice a week were effective to control *N. lugens* with the value of efficacy 70.2 and 50.4% and were safe to natural enemies in the field. Further research is needed to increase the effectiveness in the field.

Conclusion

Spraying the formulation CiC (1:1) and CLe (1:1) in the concentration of 10 mL/L increased oviposition deterrence as well as the nymph and adult mortality of *N. lugens* in the greenhouse. These mixtures with the frequency of spraying twice a week were effective to control *N. lugens* and were safe to natural enemies in the field. The formula needs to be refined so that it can increase the effectiveness in a field test.

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Conflict of interest

The authors state that there is no conflict of interest.

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