Evaluation of Kemiri Sunan (*Reutealis trisperma* Blanco) Seed Oil Nanoemulsion as insecticide against *Planococcus minor* (Hemiptera: Pseudococcidae)

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Abstract. Estate commodities and flowering plants are mostly attacked by *Planococcus minor*. Nowadays, botanical pesticide nanoemulsion technology is developing to be used to control insect pests. To evaluate the kemiri sunan seed oil nanoemulsion as insecticide against *P. minor* was the objective of this study. Completely Randomized Design (CRD) with four replication was used on the kemiri sunan evaluation test. Each replication consist of seven concentration (0,2%; 0,39%; 0,78%; 1,56%; 3,13%; 6%; and water as control). Analysis of variance (ANOVA) at α 95% followed by Duncan multiple test at α 5% was carried out. The evaluation result showed an increase in the insecticidal effect of kemiri sunan seed oil nanoemulsion against the insect species when formulated as a nano-emulsion. The application of kemiri sunan seed oil nanoemulsion for five day after treatment was able to cause the mortality of *P. minor* as much as 90%. Within 5 days after treatment, LC<sub>50</sub> was 0.09 % and LT<sub>50</sub> was 3,7 days. It is implies that the kemiri sunan seed oil nanoemulsion when to be sprayed may kill directly to the target pest and it is needed around 3,7 days to kill 50% population of pest. The results obtained indicate that kemiri sunan seed oil based nanoemulsion formulations can be used as toxicants for the control of *P. minor*. The kemiri sunan seed oil based nanoemulsion can be developed into botanical pesticides. Low chemical residues, safe for natural enemies and can be used as component in integrated pest management are the advantages of The kemiri sunan seed oil based nanoemulsion.

Keywords: kemiri sunan seed oil nanoemulsion, *Planococcus minor*, evaluation

1. Introductions

*Planococcus minor* is a pest in various commodities and is found in various flowering plants, found in nearly 250 plant species and includes 80 families, of these 250 plants some of them are cash crops such as soybean, potato, corn, grape, coffee, and cocoa [1, 2]. The existence of this pest incidence on sesame plants is rarely reported, but due to global climate change, the presence of these pests in sesame plants began to cause problems with decreasing crop production even in severe incidence causing the death of sesame plants. To control these pests, farmers usually use chemical insecticide [3]. But then, the utilization of the insecticide has negative effect such as pest resistance, pest resurgence, mortality of natural enemies, mortality of beneficial insect, and environmental damage [4]. Recently, the most developed technology in pest control is environmentally friendly pest control. One of the environmentally friendly pest control technologies is using botanical pesticides [5]. There are several conditions to be able to develop botanical pesticides, such as the plant material is not difficult to found,
supply in large quantities is not constrained, safe for natural enemies, safe for pollinator, not toxic to humans, safe for the environment, and effective to control insect pest. Low risk of resistant and resurgent pests, safe to beneficial insect (pollinator, predator, and parasitoid), no phytotoxic effect on plant, does not cause environmental damage, and does not cause acute poisoning in humans are the advantages of utilization of botanical pesticide [6,7].

Nowadays, nanoemulsion technology is developed in some country especially in Indonesia. This technology began to develop because it uses a metastable system, and to support its use will depend heavily on synthesis or preparation methods [8]. Recently, the "top-down" process has started to develop a lot and is used in several developing and developing countries. This "top-down" process is a new low-energy emulsification method that relies more on phase behavior and properties to promote ultra-small droplet formation with constant strong stirring (Ostertag et al., 2012). In Indonesia, the used of low cost technology is easier to adapt in farmer because of the lack of funds of farmer in Indonesia.

Information about the evaluation of kemiri sunan seed oil nanoemulsion as insecticide against \textit{P. minor} has not been widely studied and has not been widely used in several countries. Consequently, it is necessary to conduct research related to evaluation of kemiri sunan seed oil nanoemulsion as an insecticide against \textit{P. minor}. From this research, it is expected to open up new research related to nanoemulsion and finally obtained a simple technology that can be applied by farmers to control \textit{P. minor} larvae which are low cost and have a low negative environmental impact. The aims of this research were to evaluate the kemiri sunan seed oil nanoemulsion as insecticide against \textit{P. minor}.

2. Materials and Methods

2.1. Experimental and test Insect

The evaluation of kemiri sunan seed oil nanoemulsion was carried out at the Laboratory of Pest and Disease of Indonesian Sweetener and Fiber Crops Research Institute from April to November 2018. For stock culture of the pest, \textit{P. minor} were obtained and collected from sesame plantations at the Karangploso Experiment Station, Malang, East Java. After collected the pest from the field, rearing pests is conducted in greenhouses and uses sesame plant as food sources for \textit{P. minor}. For bioassay in this research were used the second instar of F2 generation.

2.2. Kemiri sunan seed oil nanoemulsion extraction

The kemiri sunan (\textit{Reutealis trisperma} Blanco) seeds were collected from Purwodadi Botanical Garden, East Java, Indonesia; and the alkil gliserol ftalat was from Dow AgroSciences. Two kilograms of kemiri sunan seeds were dried to obtain a moisture content of approximately 15%. This material was put into manual oil press machine and pressed. From this machine we obtain the oils and it was used for nanoemulsion extraction. Distilled water and aqueous filtrate were part of the continuous phase of oil in water nanoemulsion. The formulation keeps the active component (90% kemiri sunan oil) constant. The clear microemulsions obtained by adding alkyl glycerol phthalate to the aqueous filtrate of kemiri sunan seed oil. 750 rpm was carried out for stirring this microemulsion. The nanoemulsion in the form of a stable and smooth dispersion of oil particles obtained by stirring for 60 minutes [9]. This kemiri sunan seed oil nanoemulsion that used in toxicity test against \textit{P. minor}.

2.3. The evaluation of kemiri sunan seed oil nanoemulsion as insecticide against \textit{P. minor}

In order to evaluation of kemiri sunan seed oil nanoemulsion as insecticide against \textit{P. minor}, a test was carried out with Completely Randomized Design (CRD) with four replication. Each of replication used seven concentration (0.2%; 0.39%; 0.78%; 1.56%; 3.13%; 6%; and water as control). On this test we used kemiri sunan seed oil nanoemulsion. Twenty five second instar larvae used for each of concentration. The sesame leaves was replaced with new one daily. The evaluation was determined by direct contact application. The evaluation of kemiri sunan seed oil nanoemulsion as insecticide against pest used parameters observed for five consecutive days the percentage of pests that died from the effects of this botanical pesticides.
2.4. Data analysis

The mortality data were analyzed by Anova and if the results were significantly different (α 5%), continued with Duncan Multiple Range Test. Estimation of lethal concentration (LC\textsubscript{25}, LC\textsubscript{50}, LC\textsubscript{75}) and lethal time (LT\textsubscript{25}, LT\textsubscript{50}, LT\textsubscript{75}) were analysed by probit analysis and the associated 95% confidence interval. This probit analysis were used Software of POLO Plus [10, 11].

3. Result and Discussions

The evaluation of kemiri sunan seed oil nanoemulsion as insecticide against \textit{P.minor} was studied by preparing nanoemulsion with different concentrations and determining the LC\textsubscript{25}, LC\textsubscript{50}, LC\textsubscript{75}, LT\textsubscript{25}, LT\textsubscript{50}, LT\textsubscript{75} of the samples. The evaluation test was conducted with concentrations of 0,2%; 0,39%; 0,78%; 1,56%; 3,13%; 6%; and water as control (Table 1). The mortality rates for \textit{P. minor} that were treated with various concentrations of kemiri sunan seed oil nanoemulsion after five days of exposure are shown in Figs. 1 and Table 1. From the average percentage mortality from definitive test obtained, it was evident that kemiri sunan seed oil nanoemulsion have insecticide effect to \textit{P. minor} larvae (Figure 1). The mortality of \textit{P. minor} was significantly (p<0.05) affected by concentration and exposure interval (Table 1). On five days after treatment, 90% of \textit{P. minor} mortality are the highest mortality after application of the kemiri sunan seed oil nanoemulsion. the mortality rate of \textit{P.minor} was increasing on the second day of treatment and after the fourth day after treatment the mortality rates reach above 80%. These data showed that kemiri sunan seed oil nanoemulsion reacted slowly in destroying pest but was able to cause the death of the pest with the percentage of above 80%. A variation in the mortality rate of \textit{P. minor} at each applied level of kemiri sunan seed oil nanoemulsion concentration was show on Table 1. At five days after the treatment showed that the increased of concentration of kemiri sunan seed oil nanoemulsion linear with increased mortality of \textit{P. minor}. This phenomenon possibly because the chances of the amount of toxins exposed to the pest would be greater so that more pest would be death.

Table 1. Effect of kemiri sunan seed oil nanoemulsion on \textit{P. minor} mortality at five days after treatment

| Concentration rate of kemiri sunan seed oil nanoemulsion (%) | Percentage of mortality \textit{P. minor} after treatment by kemiri sunan seed oil nanoemulsion (%) |
|-------------------------------------------------------------|----------------------------------------------------------------------------------------|
|                                                                 | one day | two day | three day | four day | five day |
| 0%                                                          | 0 a     | 0 a     | 0 a       | 0 a      | 0 a      |
| 0,20%                                                       | 0 a     | 11 a    | 22 b      | 57 b     | 59 b     |
| 0,39%                                                       | 0 a     | 17 a    | 23 c      | 68 c     | 68 b     |
| 0,78%                                                       | 5 a     | 19 a    | 30 d      | 72 d     | 73 c     |
| 1,56%                                                       | 6 a     | 22 a    | 43 e      | 75 d     | 76 c     |
| 3,13%                                                       | 6 a     | 34 a    | 52 f      | 80 e     | 84 d     |
| 6,00%                                                       | 8 a     | 39 a    | 55 f      | 88 e     | 90 d     |

* the same letter notation in one column shows not significantly different by DMRT (α = 5%).

LC\textsubscript{50} and LT\textsubscript{50} value is used as one of the insecticidal activity criteria to the kemiri sunan seed oil nanoemulsion. The lower the LC\textsubscript{50} and LT\textsubscript{50} value the more poisonous of an insecticide. Within 5 days after treatment, LC\textsubscript{50} was 0.09 % and LT\textsubscript{50} was 3.7 days (Table 2 and 3). It is defines that mortality of the target pest pests may occur if sprayed with the kemiri sunan seed oil nanoemulsion and it is needed around 3,7 days to kill 50% population of pest.

The insecticide activity of the kemiri sunan seed oil nanoemulsion was congruent with the results of several other researches which stated that the use of botanical pesticide oil nanoemulsion could be used as an insecticide for pest control. On one day after treatment, nanoemulsion eucalyptus oil formulation in the form of karanja and jatropha aqueous filtrate with concentrations of 300 and 1500 ppm was able to cause 88-100% mortality in \textit{T. castaneum} imago [12] In another study, it was stated that the use of
The neem oil nanoemulsion formulation with 1% azadirachtin content worked effectively causing the death of S. oryzae and T. castaneum imago with a mortality range of 85-100% and 74-100% respectively [13]. Whereas in the essential oil nanoemulsion formulation from three local plants of Ageratum conyzoides, Achillea fragrantissima and Tagetes minuta the mortality rate was 88.3-95.3% for T. castaneum imago on ten days after treatment [14]. Nanoemulsions of plant essential oils gave 82.6-99% mortality rates against Aphid species [15]. From the literature shows that nanoemulsion has advantages that can increase the toxicity and effectiveness of botanical pesticides. Nanoemulsions have the ability to optimize the delivery of compounds that are not soluble in water or active ingredients [16]. The small size of the droplets allows them to deposit or spray uniformly on target areas [17]. Few reports are available on the insecticidal activity of kemiri sunan seed oil nanoemulsion with fast and high mortality against P. minor. This study showed an increase in the insecticidal effect of kemiri sunan seed oil nanoemulsion against the insect species when formulated as a nano-emulsion. The results obtained indicate that kemiri sunan seed oil based nanoemulsion formulations can be used as an insecticide for the control of P. minor. The kemiri sunan seed oil based nanoemulsion can be recommended for simple pest control technology in integrated pest management tactics.

Table 2. Estimation of lethal concentration (LC) of kemiri sunan seed oil nanoemulsion against P. minor at five days after treatment

| Lethal concentration (LC) | LC_{25} (CI 95%) (%) | LC_{50} (CI 95%) (%) | LC_{75} (CI 95%) (%) |
|---------------------------|----------------------|----------------------|----------------------|
|                           | 0.01                 | 0.09                 | 0.97                 |
|                           | (0.008 – 0.024)      | (0.03 – 0.16)        | (0.69 – 1.39)        |

Table 3. Estimation of lethal time (LT) of kemiri sunan seed oil nanoemulsion against P. minor at five days after treatment

| Lethal time (LT) | LT_{25} (CI 95%) (%) | LT_{50} (CI 95%) (%) | LT_{75} (CI 95%) (%) |
|------------------|----------------------|----------------------|----------------------|
|                  | 2,3                  | 3,7                  | 5,9                  |
|                  | (1,6-2,8)            | (3,0-4,8)            | (4,6-10,1)           |

4. Conclusions

The nanoemulsion formulation was able to increase the effect of kemiri sunan seed oil nanoemulsion as an insecticide against Planococcus minor. The application of kemiri sunan seed oil nanoemulsion for five days after treatment was able to cause the mortality of P. minor as much as 90%. LC_{50} and LT_{50} value is used as one of the insecticidal activity criteria to the kemiri sunan seed oil nanoemulsion. The lower the LC_{50} and LT_{50} value the more poisonous of an insecticide. Within five days after treatment, LC_{50} was 0.09 % and LT_{50} was 3,7 days. It is implies that the kemiri sunan seed oil nanoemulsion when to be sprayed may kill directly to the target pest and it is needed around 3,7 days to kill 50% population of pest. From the results of this study indicate that kemiri sunan seed oil based nanoemulsion formulations can be used as an insecticide for the control of P. minor. The kemiri sunan seed oil nanoemulsion can be recommended for simple pest control technology in integrated pest management tactics.

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