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Ethanolic extracts of shallot leaves (*Allium ascalonicum* L.) as botanical pesticide for controlling fall armyworms (*Spodoptera fugiperda* J.E. Smith)

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ABSTRACT

The application of botanical pesticide is a positive long term approach to control *S. frugiperda*. Shallot leaves contain acetogenin compound, hence it has the potential to become botanical pesticide. This study is aimed at finding out the effects of ethanolic extract of shallot leaves on *S. frugiperda* larvae in vitro. Extract is prepared using maceration method with ethanol solvent. The method of treatment is Leaf Dipping. The study applies Completely Randomized Design with concentration of shallot leaf extract as treatment at 0 ppm, 250 ppm, 500 ppm, 750 ppm dan 1000 ppm. Each treatment consists of four repetitions. Examination is conducted for 24 hours. Examination parameter is mortality of larvae and LC_{50}. Data are analyzed with One Way ANOVA (proceeded with LSD) and Probit analysis. The results suggest that shallot leaf extract at the concentration of 1000 ppm appears to be the most effective on the mortality of larvae. The obtained LC_{50} value is 263,02 ppm. The study indicates that shallot leaf extract can be applied as an alternative synthetic insecticide to control ini *S. frugiperda*.

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1. Introduction

Spodoptera frugiperda J.E. Smith is an insect originating from America and has spread to various countries. In early 2019, this pest was found to infest corn plants in Sumatera (Kementan, 2019). The pest infests plant growth area and this may result in plant failure to grow shoots and young leaves. The larvae of *S. frugiperda* have high eating ability, whereas the imagos are strong flyers and have extensive cruising range (CABI, 2019).

Spodoptera frugiperda is polyphagous, with some of its main host plants being food crops from Graminae group such as corn (*Zea mays* L.), rice (*Oryza sativa* L.), wheat (*Triticum* spp), sorghum (*Sorghum* spp), and sugarcane (*Saccharum officinarum* L). The loss caused by this pest infestation on corn plants in Africa and Europe has amounted to between 8.3 and 20.6 tons per year at economic value ranging from US$ 2.5 to 6.2 billions (FAO & CABI, 2019).

The application of botanical pesticide has been recommended as an alternative to the hazardous synthetic insecticides such as pyrethroid and organophosphate which may be harmful to the environment, cause pest resurgence and pest resistance towards insecticides (Arya & Tiwari, 2013).

Extracts from such plants as *Azadirachta indica, Milletia ferruginea, Croton macrostachyus, Phytolacea docendra, Nicotina tabacum* and *Chrysanthemum cinerariifollium* have been successfully applied to control pest insects (Jirnmci, 2014). Silva, et. al., (2015) have reported high mortality rate among *S. frugiperda* larvae treated with extract of *A. indica* seed residue. Another study suggests that ethanolic extracts of *Argemone ochroleuca* have resulted in mortality of S. frugiperda larvae due to feed deficiency and larvae growth retardation (Martínez, et. al., 2017).

Shallot leaf is known to contain high concentration of acetogenin. Acetogenin compound is stomach poisonous and may result in insect mortality (Plantus, 2008). Shallot leave as botanical pesticide for *S. frugiperda* pest is potential for development, hence the results of this study are expected to provide more information on botanical pesticides to control larvae of *S. frugiperda*.

2. Method

2.1. Extract Preparation

Preparation of botanical pesticide in this study refers to the method by Meriga, et. al., (2012). The ratio of simplicia and solvent is 1:10. An amount of 100 g simplicia powder from shallot leaves is put into maceration bottle. Then, 1000 ml of 96% ethanol is added into the bottle. Maceration is carried out for 24 hours. The maceration bottle is manually agitated every 1 hour. After 24 hours, filtering is made using filter paper. The filtering procedure results in filtrate, which is subsequently evaporated in a rotary evaporator set at 37°C for 3 hours. The filtrate is then thickened in waterbath at 60°C until it becomes paste. Coarse extracts of shallot leaves are then made in concentration levels of 250 ppm, 500 ppm, 750 ppm and 1000 ppm. Each of the concentrates is then contained in 50 ml (% w/v).

2.2. Extract Testing

Treatment of botanical pesticide on test animals refers to the method by Meriga, et. al., (2012). Testing is conducted using Leaf Dipping Methods. Test cage is prepared and provided with floor base. The fell armyworm larvae instar III are put into the cage and let hungry for 1-2 hours. Young corn leaves are washed and cut into 4 x 4 cm size. These leaf cuts are dipped into pesticide solution for 30 seconds. These dipped leaf cuts are air-dried and put into the cage already filled with *S. frugiperda* larvae. Observation on mortality is conducted for 1 x 24 hours. Dead larvae are those incapable of moving when they are touched.

2.3. Data Analysis

Variance analysis (ANOVA) is made on the mortality rate of examined fall armyworms. When difference in the average value is found, then LSD test is proceeded. Lethal concentration (LC_{50}) analysis is made using Probit analysis to decide the effective concentration capable of killing 50% of larvae in a specified time frame.

3. Findings and Discussion

3.1. Mortality of *S. frugiperda* larvae

Higher concentration of shallot leaf extract results in increased mortality of the larvae (Figure 1). Tested concentration of 250 - 1000 ppm resulting in larvae mortality of 60% - 90% in 24 hours indicates that shallot leaf extract has the capability to kill larvae in a short period. Previous reports show that insecticidal effects on Spodoptera pests are found in a number of acetogenin-containing compounds such as extracts of *Annona montana* (Blessing, et. al., 2010), *Rollinia occidentalis* (Tolosa, et. al., 2012), dan *Annona mucosa* (Massarolli, et. al., 2017).

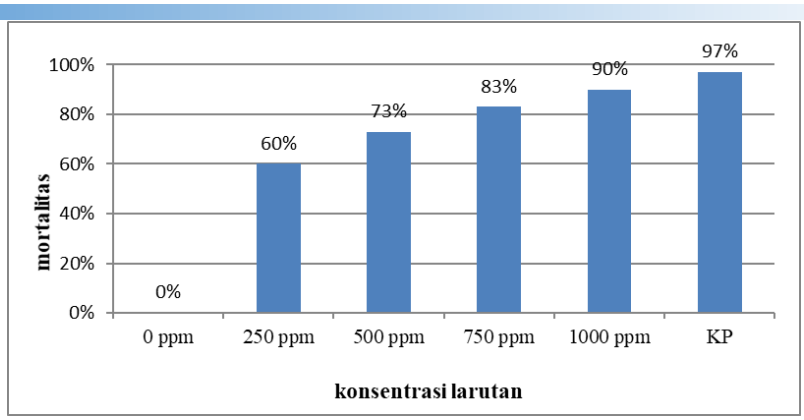

Fig. 1. Diagram of mortality rate of *S.frugiperda* larvae in 24 hours; KP is Positive Control, i.e, emamectin benzoate.

Shallot leaf extract provides significant effect on mortality of *S. frugiperda* larvae. Concentration of 1000 ppm turns out to be the most effective according on Least Significant Difference (LSD) test (Table 1). The acetogenin compound content, which is a potential stomach poison, is efficient in controlling chewing insects such as Lepidopterans (Isman, 2006). Acetogenin is a powerful inhibitor for the production ATP (adenosine triphosphate) in mitochondrial complex I (NADH ubiquinone oxidoreductase) within insects. Such decrease in the production of ATP may lead to cell apoptosis. In addition, acetogenin has the compound characteristics which disturb hormonal activities of Lepidopterans (Massarolli, et. al., 2016).

Table 1. Average mortality of S. frugiperda larvae after treatment

Treatment	Mortality
0 ppm	0.0[a]
250 ppm	3.6[b]
500 ppm	4.2[b]
750 ppm	5.0[c]
1000 ppm	5.4[d]

[a.] Note: a,b,c,d,e Different Superscript on average values indicates significant difference (P<0,05) in Least Significant Difference (LSD) Test.

3.2. Lethal Concentration (LC_{50})

Obtained LC_{50} value of 263.02 ppm is effective to kill *S. frugiperda* larvae by 50% of the population. This value is lower than the one obtained with the extract of *Annona mukosa* seed, i.e. 1479 ppm tested on *Helicoverpa armigera* insect (Souza, et. al., 2017). Both extracts of shallot leaves and *Annona mucosa* seeds contain high level of acetogenin. Blessing, et. al., (2012) reported on correlation between insecticidal characteristics of acetogenin and specific site of interaction with lipid membranes. Acetogenin interacts with phosphate from lipid membranes in different levels. Difference in antisymmetric stretching of phosphate group indicates the loss of water in hydrogen bond. Destabilization in the membranes due to dehydration around the phosphate group is caused by interaction with acetogenin. Similar study reported by Hidalgo, et. al. (2018) suggested that acetogenin compound from *Annonaceae* family gave *antifeedant* effect which resulted in decrease in growth of larvae and malformation of imagos.

4. Conclusion

Ethanolic extract of shallot leaves at the concentration of 263,02 ppm is effective in killing 50% of fall armyworms in 24 hours. This suggests that ethanolic extract of shallot leaves has the potential to become botanical pesticide to control *S. frugiperda*. This study requires further evaluation in field conditions to find out its effect on target pests, non-target pests, and host plants.

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