Effectiveness Test of *Piper methysticum* Extract Against *Crocidolomia pavonana* larvae

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Abstract. Application synthetic insecticide with high frequency and continuously can give to negative impact. Alternative control which secures is botanical insecticide. This research aimed to effectiveness tests of *P. methysticum* plant against *C. pavonana* larvae using root and leaves this plant. The tested insecticide activity including mortality and feeding inhibition tests. The extract was tested at five concentration levels and repeated five times. The results showed that *P. methysticum* root extract was able to cause *C. pavonana* larvae mortality of 94% with a concentration of 419.7 gram/100 ml in water meanwhile, the testing using leaves extract causes mortality 98% from concentration 342 gr/100 ml in water. The feeding inhibition test showed the *P. methysticum* roots extract give effect with very weak criteria at some concentrations used a no choice method while Leaves extract *P. methysticum* does not show feeding inhibition.

Keywords : Piper methysticum, Mortality, Feeding inhibition, *C. pavonana*

1 Introduction

Crocidolomia pavonana (Lepidoptera: Crambidae) is one of the pest in the cabbage plants. *C. pavonana* is able to reduced cabbage production due to the destructive activity of larvae at the growing point of the plant. Therefore, farmers use synthetic insecticides to control these pests. However, when applying synthetic insecticides intensively and excessively it will cause very dangerous negative impacts such as insect pest resistance, pollution to the surrounding environment which is indicated by the death of organisms non-target such as natural enemies, livestock, and other organisms.

Piper methysticum Forst belongs to the Piperaceae family. The *P. methysticum* plant is known as *WATI* by the indigenous people of Merauke, the Marind tribe. This plant is used by the community in several important events such as funerals, pig feasts, weddings, religious events, peace events and is used as a dowry for women who are getting. The part of the plant that is used for important occasions is the root part. In addition, the local community also uses the plant as traditional medicine, such as the part of the stem which is used as cough medicine and wound medicine [1]. The leaves are used as wound medicine for insect bites or prickling of the spines of several types of fish. Treatment is done by chewing the part and extracting the juice or attaching it to the injured part [2]. Kava has considerable potential as a source of pharmaceutical compounds. The kavalactones act as sedatives, soporifics, analgesics, anticonvulsives, local

anaesthetics, muscle relaxants, and diuretics. They also show strong antimicrobial activity, particularly against fungi, including yeasts [3].

The technology currently being developed is to utilize plant extracts as a botanical insecticide. botanical insecticides are insecticides with active ingredients derived from plants, extracts that are expected to hurt the life of insect pests in terms of the behavior and physiology of insect pests. The compound components contained in the roots and leaf of *P. methysticum* are thought to act as insecticides. Therefore, the researchers are interested in testing the effectiveness of the root and leaf extracts of *P. methysticum* as a botanical insecticide against *C. pavonana* larvae.

2 Methodology

2.1 Time and Place

The research was conducted from July to December 2018 at the Laboratory of the Department of Agrotechnology, Faculty of Agriculture, Musamus Merauke University.

2.2 Tools and Materials

The tools used were brushes, plastic boxes, measuring cups, tweezers, cotton wool, digital scales, filters, scissors, mortal and mortal children. The materials used

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were the roots and leaves of the *P. methysticum* plant, the larvae of *C. pavonana* instar 2nd, 10% honey, clean water.

2.3 Research Procedure

2.3.1 Preparation of extract

- 1) The roots and leaves of *P. methysticum* were cut into small pieces.
- 2) Weighed using digital scales.
- 3) The wet material is then crushed using a separate mortal
- 4) Each ingredient is then immersed in 100 ml of clean water and 0.1-gram detergent for 24 hours.
- 5) The extract preparation is filtered using a fine sieve and the extract is ready for use.

2.3.2 Maintenance and multiplication of *C. pavonana*

The test insects included that the larvae would be kept in a plastic box with screen windows (10 cm x 7cm x 5 cm) and given mustard feed. Towards the pupae, sterile sawdust will be prepared as a medium for pupae. The pupae formed will be transferred into a plastic tube and left until the adult insect appears. The adult insect will be given 10% honey liquid feed applied to cotton, meanwhile, the female insect will be given mustard leaves as a medium for laying eggs ; the stalk is inserted into the tube and wrapped in wet cotton. The group of eggs obtained are then taken and kept in a plastic box until the eggs hatch.

2.4 Methods

2.4.1 Mortality test.

The test was carried out using the leaf dipping method. The extract was tested at five concentration levels plus control. The concentration levels of the root extract were 26.24 g/100 ml water, 52.47 g/100 ml water, 104.93 g/100 ml water, 209.85 g /100 ml water and 419.7 g/100 ml water. Meanwhile, the concentration of leaf extract was 11.38 g/100 ml water, 42.75 g/100 ml water, 85.50 g/100 ml water, 171 g/100 ml water, 342 g/100 ml water. The test larvae were ten of *C. pavonana* instar 2nd.

2.4.2 Feeding inhibition test.

Two methods were used, namely choice and non-choice which were tested against the same concentration as the mortality test. Ten 2nd instar *C. pavonana* larvae and each treatment were repeated five times. The formula used to calculate the percentage feeding inhibition with the choice method is:

$$PM = \frac{BKK - BKP}{BKK + BKP} \times 100\% \quad (1)$$

The percentage of feeding inhibition with a method non-choice is calculated using the formula :

$$PM = \frac{BKK - BKP}{BKK} \times 100\% \quad (2)$$

Information :

PM : feeding Inhibition (%)

BKK : weight leaves dry control (gr)

BKP : weight leaves dry treatment (gr)

The calculated percentage of will be followed by determining the criteria for feeding inhibition, namely ≥ 80 (strong); $61 \leq x < 80$ (moderate); $40 \leq x < 60$ (weak); < 40 (very weak).

3 Result and Discussion

3.1 Mortality test

The mortality percentage of *C. pavonana* larvae treated with *P. methysticum* roots extract on 24 HAT observations showed that the mortality of test larvae was 6-20% at low concentrations to the highest concentrations. Observations of 48 HAT showed an increase in the percentage of larval mortality at each test concentration by 12 - 72%. Meanwhile, the 72 HAT observations showed an increase in the number of dead test larvae by 38 - 94%, while the control treatment did not show larval mortality at each observation time (Fig. 1). The mortality value of *C. pavonana* due to *P. methysticum* leaves extract treatment showed increased mortality at each observation time and showed a percentage of 98 % at a concentration of 342 g/100 ml water (Fig. 2).

Fig. 1. Percentage of mortality *C. pavonana* larvae due to roots extract treatment *P. methysticum*

Fig. 2. Percentage of mortality *C. pavonana* larvae due to leaves extract *P. methysticum*

3.2 Feeding inhibition

3.2.1 No choice method

The value of feeding inhibition *P. methysticum* roots extract showed very weak criteria at a concentration of 52.37 g/100 ml water, 104.93 g/100 ml water, 209.85 g/100 ml water, and 419.7 g/100 ml water but a concentration of 26.24 g/100 ml water showed no effect of feeding inhibition (Table 1). Meanwhile, Table 2 shows that there is no feeding inhibition effect due to *P. methysticum* leaves extract.

Table 1. Criteria feeding inhibition *P. methysticum* root extract against *C. pavonana* larvae.

Extract	Concentration (gr/100 ml water)	Feeding Inhibition (%)	Criteria
Root <i>P. methysticum</i>	26.24	-13.17	no reaction
	52.47	4.73	very weak
	104.93	11.32	very weak
	209.85	16.74	very weak
	419.7	22.48	very weak

Note: number followed by a sign (-) indicates that there are not feeding inhibition

Table 2. Criteria feeding inhibition *P. methysticum* leaves extract against *C. pavonana* larvae

Extract	Concentration (gr/100 ml water)	Feeding Inhibition (%)	criteria
Leaf <i>P. methysticum</i>	21.38	-59	no reaction
	42.75	-98	no reaction
	85.5	-164	no reaction
	171	-287	no reaction
	342	-220	no reaction

3.2.2 Choice method

The results of the test with the no-choice method are shown in Tables 3 and 4, that do not show an inhibitory effect on eating.

Table 3. Criteria feeding inhibition *P. methysticum* root extract against *C. pavonana* larvae

Extract	Concentration (gr/100 ml water)	Feeding Inhibition (%)	criteria
Root <i>P. methysticum</i>	26.24	-14,33	no reaction
	52.47	-22,40	no reaction
	104.93	-20,90	no reaction
	209.85	-23,45	no reaction
	419.7	-14,63	no reaction

Note: number followed by a sign (-) indicates that there are not feeding inhibition

Table 4. Criteria feeding inhibition *P. methysticum* leaf extract against *C. pavonana* larvae

Extract	Concentration (gr/100 ml water)	Feeding Inhibition (%)	Criteria
Leaves <i>P. methysticum</i>	21.38	-60	no reaction
	42.75	-24	no reaction
	85.5	-25	no reaction
	171	-8	no reaction
	342	-6	no reaction

Note: number followed by a sign (-) indicates that there are not feeding inhibition

The results of the mortality test for *C. pavonana* larvae in Fig. 1 and Fig. 2 show that the mortality value is not much different. The difference in mortality is caused by differences in concentrations and active compounds in the root and leaf extracts of *P. methysticum*. According to [4], *P. methysticum* leaves extract was able to mortality *C. pavonana* larvae by 22% using a water concentration of 35 g/100 ml. the research results of [5] the n-hexane extract of *P. methysticum* leaves has insecticidal properties, at a mortality rate of 63.33% and can inhibit eating of *Plutella xylostella* larvae at a concentration of 0.015-0.38% of 11.69-85.54%. So it is suspected that the mortality that occurs after the treatment of *P. methysticum* roots and leaves extracts is due to the action of the compounds contained.

The active compounds of *P. methysticum* that have been identified are kavalactones, Pyron, flavonoids, and alkaloids. Besides, [2] added that there are seven main compounds of kava, namely lactone, and glutathione in kava root which were extracted using several types of organic solvents and water with a total of lactone compounds with acetone organic solvent 286.2 mg/g extract) while the water solvent showed total lactone and glutathione 108.6 mg/g, and 26.3 mg/g. In other island, extract *P. methysticum* greatly used for the production of medicines. The active ingredients responsible for these properties are mainly six kavalactones that have been extracted from Kava and reportedly include demethoxy-yangonin, dihydrokavain, yangonin, kavain, dihydromethysticin, and methysticin [6]. Meanwhile, the relationship between mortality and concentration can be drawn from the regression parameters on tables 5 and 6 shows the values of LC₅₀ and LC₉₅ as predictive numbers of concentrations at each observation time. The toxicity of the extract can be seen from the predictive value of the LC₅₀ and LC₉₀ on the 72 HAT. When result analysis showed the concentration value that lower so exhibiting high toxicity.

The value of feeding inhibition which is classified as a very weak criterion with a choice method is thought to be due to the activity of the active compounds contained in the extract which can inhibit the work of the test insect's sensory organs such as tasting. so that the leaves of the control treatment were mostly eaten by the larvae.

Therefore, it can be stated that *P. methysticum* has potential as a botanical insecticide because it can kill and feeding inhibition of the test insects. However, more testing is needed to determine the right concentration, the best solvent for extraction, and extract formulation so that the effectiveness of the extract can be increased.

4 Conclusion

The root extract of *P. methysticum* was able to mortality of *C. pavonana* larvae to 94% with a concentration of 419.7 gr/100 ml water and mortality due leaves extract to 98% concentration of 342 gr/100 ml water on the 72 HAT observation. The feeding inhibition test, roots extract showed weak criteria at several concentrations using the no-choice method, and the choice method did not show any feeding inhibition. While leaves extract show no feeding inhibition reaction.

Table 5. The estimator of toxicity parameters of *P. methysticum* root extract against *C. Pavonana* larvae were tested using the leaf dip method

Extract	Observation Time (HAT)	$b \pm$ GB	LC ₅₀ (SK 95%) (%)	LC ₉₀ (SK 95%) (%)
Root <i>P. methysticum</i>	24	0.61 ± 0.25	9662.7 (-)	1144934. 110 (-)
	48	1.60 ± 0.22	219.42 (136.91- 513.73)	1383.8 (556.59 - 18352.00)
	72	1.29 ± 0.21	49.70 (17.71- 083.27)	482.81 (229.90 – 4690.21)

Note: b : Slope;
GB : Default Error;
LC : Lethal Concentrate;
SK : The interval of trust
HAT : Hour After Treatment

Table 6. The estimator of toxicity parameters of *P. methysticum* leaves extract against *C. pavonana* larvae were tested using the leaf dip method

Extract	Observation Time (HAT)	$b \pm$ GB	LC ₅₀ (SK95%) (%)	LC ₉₀ (SK95) (%)
Leaves <i>P. methysticum</i>	24	0.21 ± 0.26	-	-
	48	0.17 ± 0.19	-	-
	72	1.17 ± 0.28	6.73 (0.90- 14.46)	168.67 (100.56- 555.18)

Note: b : Slope;
GB : Default Error;
LC : Lethal Concentrate;
SK : The interval of trust
HAT : Hour After Treatment

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