

**Toxicity of Plant-Based Insecticide Extracts of Tobacco Stem  
(*Nicotiana tabacum* L.), Babadotan Leaves (*Ageratum conyzoides* L.) and  
a Combination of These to Control *Crocidolomia pavonana* F. Larvae**

**Toksistas Insektisida Berbasis Ekstrak Batang Tanaman Tembakau  
(*Nicotiana tabacum* L.), Daun Babadotan (*Ageratum conyzoides* L.) dan Kombinasi  
dari Keduanya untuk Mengendalikan Larva *Crocidolomia pavonana* F.**

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**ABSTRACT**

*Crocidolomia pavonana* is one of the pests that attacks the Brassicaceae family. The larvae of *C. pavonana* can eat cabbage plants until they are able to reduce production by 79.81%. One of the solutions to control this pest is to use plant-based insecticides. This study aims to determine the toxicity of tobacco stem extracts, babadotan leaves and a combination of the two against *C. pavonana*. Each extract was tested to instar III *C. pavonana* larvae by leaf dip method. At 96 hours after treatment, babadotan extract (17%) had a mortality value of 93.33%, while tobacco extract (30%) had a mortality value of 93.33%. Babadotan extract had a lower LC<sub>50</sub> than tobacco extract of 6,710 and 10,640, respectively. Meanwhile, the combination of Ac:Nt (3:5) had the best mortality and LC<sub>50</sub> values, which were 96.67% and 5.245. These values indicate that the combination of extracts is more toxic than single ingredients. Based on the combination index, the combination of Ac:Nt (3:5) at the LC<sub>50</sub> and LC<sub>95</sub> levels is weak synergistic. Thus, the single-ingredient treatment of babadotan extract was more toxic than the tobacco stem extract, but the combination treatment of Ac:Nt (3:5) was more toxic than the treatment of single extract.

Key words : *Crocidolomia pavonana*, plant-based insecticide, insecticide combination, *Nicotiana tabacum*, and *Ageratum conyzoides*.

**ABSTRAK**

*Crocidolomia pavonana* merupakan salah satu hama yang menyerang tanaman dari famili Brassicaceae. Larva dari *C. pavonana* dapat memakan tanaman kubis hingga mampu menurunkan hasil produksi sampai 79,81%. Salah satu solusi pengendalian hama ini adalah menggunakan insektisida nabati. Penelitian ini bertujuan untuk menentukan toksistas dari ekstrak batang tembakau, daun babadotan dan kombinasi keduanya terhadap larva *C. pavonana*. Setiap ekstrak diuji ke larva instar III *C. pavonana* dengan metode celup daun di cawan petri dan diamati selama 96 jam. Pada 96 jam setelah perlakuan, ekstrak babadotan konsentrasi 17% memiliki nilai mortalitas sebesar 93,33%, sedangkan ekstrak tembakau konsentrasi 30% memiliki nilai mortalitas sebesar 93,33%. Ekstrak daun babadotan memiliki LC<sub>50</sub> yang lebih rendah daripada ekstrak batang tembakau masing-masing sebesar 6,710 dan 10,640. Sedangkan kombinasi ekstrak batang tembakau dan daun babadotan (3:5) memiliki mortalitas



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dan nilai  $LC_{50}$  tertinggi yaitu sebesar 96,67% dan 5,245. Nilai tersebut menunjukkan bahwa kombinasi ekstrak bersifat lebih beracun daripada bahan tunggal. Berdasarkan indeks kombinasi, kombinasi Ac:Nt (3:5) pada taraf  $LC_{50}$  dan  $LC_{95}$  bersifat sinergistik lemah. Dengan demikian, pada perlakuan bahan tunggal ekstrak babadotan lebih beracun daripada ekstrak batang tembakau, namun perlakuan kombinasi Ac:Nt (3:5) lebih beracun daripada perlakuan ekstrak tunggal.

**Kata kunci:** *Crocidolomia pavonana*, insektisida nabati, kombinasi insektisida, *Nicotiana tabacum*, dan *Ageratum conyzoides*.

## INTRODUCTION

Crop caterpillar or *Crocidolomia pavonana* F. is one of the most common pests that damage broccoli, cabbage, radish, and some *Brassicaceae* family plants. *C. pavonana* disturbs these crops in the larval phase by feeding on new leaves and can prevent cabbage from forming a crop. When the center is damaged, the larvae feed on the tip and descend to older leaves. The 1st instar larvae will group together and feed on the lower epidermis of the leaf, forming a window and producing fine white threads that cover the group. Entering instar 3, in addition to the leaves, the larvae will feed on the stalks and stems to create tunnels. The larvae of *C. pavonana* will then disperse and damage the crop until they enter the growing point of the plant (Habriantono *et al.*, 2023). The crop will be completely damaged if the caterpillar attack is not immediately controlled. The attack is detrimental to farmers because it can damage plants and reduce production yields by 79.81% (Javier *et al.*, 2018).

Pest control by farmers in the field generally uses chemical insecticides. The continuous use of chemical insecticides by farmers is aimed at suppressing pest populations quickly. Farmers do not realize that the use of insecticides in this way can have various negative impacts such as resurgence and resistance in pests, pest population explosion, killing non-target insects, and killing natural enemies (Dadang and Prijono, 2008). According to Sarbawal *et al.* (2018), chemical pesticides have been shown to be involved in the pathogenesis of Parkinson's and Alzheimer's diseases as well as various respiratory and reproductive disorders. Based on this, alternative control techniques are needed that can be used as a substitute for chemical insecticides and optimized in their use so as to reduce the level of pest attacks on plants.

One alternative to replace the use of chemical insecticides that are friendly to humans and the environment is to use botanical insecticides that do not harm the environment. Botanical insecticides are insecticides with plant-based ingredients that contain compounds derived from secondary metabolites. These compounds are used by plants for self-defense measures from pest or predator interference. These compounds have the potential to be used as botanical insecticides. Botanical have many effectivenesses to control of pest like the *Crocidolomia binotalis* on the cabbage (Habriantono *et al.*, 2023). Additionally, the botanical pesticide as characteristic the clearing and washing of residue pesticide (Hoesain *et al.*, 2023). Majority of plants that contain several secondary metabolite compounds such as saponins, flavonoids, tannins, and alkaloids have the potential to be used as larvicides (Javier *et al.*, 2018). Some types of plants that contain secondary metabolites such as tobacco at concentrations of 15% are known to have a repellent and mortality effect with values above 20% and above 70% on *Tribolium castaneum* so that they can be used as an alternative in pest control (Hanif *et al.*, 2016).

Tobacco plants (*Nicotiana tabacum* L) contain nicotine, terpenoids, flavonoids, and saponins. The nicotine content of tobacco can cause respiration disorders that cause paralysis to death in test insects (Amoabeng *et al.*, 2018). Nicotine attacks the central nervous system of insects so that the insect dies (Hanif *et al.*, 2016). Tobacco extract insecticides can be used to kill *Helicoverpa armigera* because at a concentration of 10% it provides mortality of 43.3% (Radev and Stoyanova, 2024). However, this concentration can be said to be ineffective because it provides a fairly low mortality rate, so it is necessary to combine tobacco extract with other ingredients. Combinations are made to minimize one or more ingredients that are used in excess and increase the effectiveness of a botanical insecticide. The combination can be done by combining tobacco stems with babadotan leaves (*Ageratum conyzoides* L).

Babadotan leaves have polyphenol and tannin compounds that tobacco does not have. Babadotan plants contain secondary metabolites that can be utilized as botanical insecticides such as terpenoids, alkaloids, saponins, flavonoids, polyphenols, and tannins (Pintong *et al.*, 2020). According to Pintong *et al.* (2020), the use of babadotan leaf extract at a concentration of 25% gives 100% mortality to *Helicoverpa armigera*. This is because babadotan leaves contain secondary metabolites of alkaloids, saponins, flavonoids, polyphenols, and tannins (Pintong *et al.*, 2020). Secondary metabolites produced from botanical pesticides cause changes in insect body metabolism. Symptoms can be caused to contact and oral mechanisms. Morphological signs from study Habriantono *et al.*, (2023) show changes experienced by insect pests starting from color changes, lysis and death.

The combination of tobacco stem and babadotan leaves is expected to increase the effectiveness of botanical insecticides against *C. pavonana*. Therefore, it is necessary to conduct further research related to the toxicity of botanical insecticides extracts of tobacco stems (*N. tabacum* L), babadotan leaves (*A. conyzoides* L) and the combination of both against *C. pavonana* larvae. It is intended to determine the toxicity of each ingredient and its combination against *C. pavonana*.

## MATERIALS AND METHODS

The research was conducted from April 2024 to September 2024 at the Agrotechnology Laboratory, Faculty of Agriculture, University of Jember. The research used several tools and materials, namely babadotan leaves, tobacco stems, *C. pavonana* instar 3 larvae, broccoli seeds, distilled water, 96% ethanol, tween 80, plastic boxes, digital scales, blenders, petridish, beakers, and rotary evaporators. The test used the leaf dipping method with control treatment, babadotan extract treatment of 2%; 5%; 7%; 9%; 17% concentration, tobacco stem extract treatment of 4%; 8%; 11%; 15%; 30% concentration, and Ac:Nt (3:5) extract combination treatment of 2%; 4%; 6%; 8%; 13% concentration.

The research was conducted through several procedures. The first stage was planting broccoli plants as food used during the test. Seeds were sown then transferred into polybags and treated every day. The next stage was the rearing of *C. pavonana* larvae. Larvae were propagated in plastic boxes and fed with broccoli leaves. The next stage is the preparation of botanical insecticides. Tobacco stems and babadotan leaves were washed thoroughly. Then the material was mashed and sieved, then macerated with 96% ethanol with a ratio of tobacco powder to ethanol of 1:5 for 5 days and babadotan powder to ethanol of 1:3 for 3 days. The

filtrate is then in a rotary evaporator which is then stored in the refrigerator ( $\pm 4^{\circ}\text{C}$ ). The next stage is toxicity testing. Stock solution of each extract was diluted according to the concentration of each treatment. Broccoli leaves were formed into a circle with a diameter of 5 cm and then dipped into the extract for 5 minutes. Each concentration level was tested on 30 3rd instar larvae. Larvae were put into a petridish containing broccoli leaves. The treated leaves were replaced with untreated leaves after 24 hours. Feeding changes were made every day. Every day, mortality data was recorded until the 4th day.

The variables observed were larval mortality,  $\text{LC}_{50}$  of each extract, and the activity properties of the mixture. The percentage mortality of *C. pavonana* larvae can be calculated in the following way:

$$M = \frac{A}{B} \times 100\%$$

Description:

M = Percentage mortality of *C. pavonana* larvae

A - Number of dead *C. pavonana* larvae

B = Total number of *C. pavonana* larvae in each treatment

Lethal Concentration ( $\text{LC}_{50}$ ) is the concentration that can kill 50% of the total larvae tested and is obtained from probit analysis. The nature of the activity of the mixture of tobacco and babadotan extracts was analyzed using the calculation of the combination index or IK of  $\text{LC}_{50}$  and  $\text{LC}_{95}$  levels. IK can be calculated in the following way (Dadang and Prijono, 2008):

$$IK = \frac{\text{LC}_x^{1(\text{cm})}}{\text{LC}_x^1} + \frac{\text{LC}_x^{2(\text{cm})}}{\text{LC}_x^2} + \left[ \frac{\text{LC}_x^{1(\text{cm})}}{\text{LC}_x^1} \times \frac{\text{LC}_x^{2(\text{cm})}}{\text{LC}_x^2} \right]$$

Description:

$\text{LC}_x^1$  = *N. tabacum* concentration

$\text{LC}_x^2$  = *A. conyzoides* concentration

$\text{LC}_x^{1(\text{cm})}$  = Concentration of *N. tabacum* in the mixture

$\text{LC}_x^{2(\text{cm})}$  = Concentration of *A. conyzoides* in the mixture

According to Dadang and Prijono (2008), the interaction properties of mixtures can be categorized as follows:

- 1) If the IK value is  $< 0.5$  then the mixture is strongly synergistic
- 2) If IK is  $0.5 \leq \text{IK} \leq 0.77$  then the mixture is weakly synergistic
- 3) If the IK value is  $0.77 < \text{IK} \leq 1.43$  then the mixture is additive
- 4) If the IK value  $> 1.43$  then the mixture is antagonistic

## RESULT AND DISCUSSION

### Mortality of *C. pavonana* Larvae

Mortality is the average mortality of *C. pavonana* larvae during the test. Mortality data were obtained from the 24th, 48th, 72nd, and 96th hour after application. Each treatment showed different levels of mortality on each day. Mortality data of *C. pavonana* larvae in each treatment are presented in graphical form in figures 1, 2, and 3.

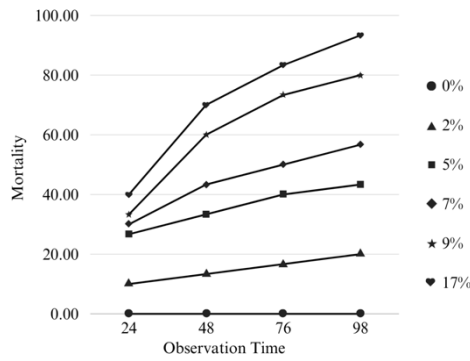


Figure 1. Percentage mortality of *C. pavonana* larvae in *A. conyzoides* leaf extract treatments

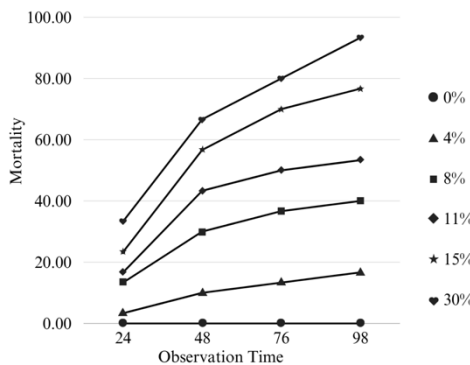


Figure 2. Percentage mortality of *C. pavonana* larvae in *N. tabacum* stem extract treatments

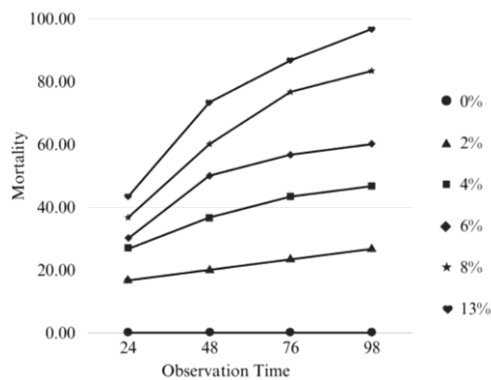


Figure 3. Percentage mortality of *C. pavonana* larvae in the combined treatment of *N. tabacum* stem and *A. conyzoides* leaf extracts

Based on figure 1. *A. conyzoides* extract gave a sharp increase in mortality at 24-48 JSP and a lower increase in mortality at 48-96 JSP. The 17% concentration at 24 JSP gave the highest mortality of 40% followed by 2-9% concentrations with mortality values of 10-33.33%. The increase continued at 48 JSP with the highest mortality of 70% at 17% concentration and mortality of 13.33-60% at 2-9% concentration. The 17% concentration resulted in 83.33% mortality of test larvae and 2-9% concentration with 17-73.33% mortality. The highest mortality of *A. conyzoides* extract treatment occurred at 17% concentration at 96 JSP with 93.33%, while 2-9% concentration gave a mortality rate of 20-80%. Based on Figure 2, a sharp increase in mortality also occurred at 24 JSP to 48 JSP. At 24 and 48 JSP, the 30% concentration of *N.*

*tabacum* extract gave the highest mortality rate of 33.33% and 66.67%, respectively. Whereas at 24 and 48 JSP, mortality due to the 4-15% concentration treatment was 3.33-23.33% and 10-56.67%, respectively. The 30% concentration resulted in the highest mortality of test larvae at 72 JSP by 80% followed by the 4-15% concentration by 13.33-70%. Observations at 96 JSP obtained the highest mortality value of 93.33% at 30% concentration and 16.67-76.67% mortality at 4-15% concentration. The mortality rate of the test larvae increased sharply at 24 and 48 JSP with the highest mortality in the combination treatment of *A. conyzoides* and *N. tabacum* extracts at the highest concentration (13%) of 43.33% and 73.33%, respectively. Meanwhile, concentrations of 2-8% gave mortality of 16.67-36.67% and 20-60%, respectively. The mortality rate at 72 JSP with 13% concentration and 2-8% concentration resulted in 86.67% and 23.33-76.67% mortality of the test larvae, respectively. The highest mortality rate of *C. pavonana* larvae occurred at 96 JSP with 96.67% at 13% concentration followed by 2-8% concentration treatment with 26.67-83.33% mortality.

Treatment of single extracts of babadotan leaves and tobacco stems as well as their combinations had a mortality effect on *C. pavonana* larvae. The results showed that the treatment of extracts singly or in combination of tobacco stems and babadotan leaves resulted in the death of *C. pavonana* larvae. Figures 1, 2, and 3 show the increase in mortality that occurs due to the accumulation of toxicity in the test larvae. The combination treatment had a higher mortality rate than the single extracts of babadotan leaves and tobacco stems at 96 JSP, namely a concentration of 13% with 96.67% mortality. In addition, the treatment with a higher concentration gave a higher mortality rate than the low concentration treatment. This is in accordance with the statement of Rioba and Stevenson (2017) that the mortality rate of the test larvae will increase in line with the increase in the concentration of the extract used in the treatment because the active compounds that enter will be more and more effective in killing the test larvae. The death of *C. pavonana* larvae is thought to be caused by the active ingredients contained in each material.

The content of active ingredients in babadotan leaves is more than tobacco stems, so the mortality of the combination treatment and the single ingredient babadotan is higher than tobacco stems. According to Pintong *et al.* (2020) babadotan leaves have secondary metabolites in the form of alkaloids, saponins, flavonoids, polyphenols, and tannins with high enough amounts, while according to Amoabeng *et al.* (2018) tobacco in the stem part also contains nicotine, saponins, and flavonoids although not as much as in the tobacco leaf part. The amount of nicotine contained in the stem ranges from 0.08-0.16% (Peševski *et al.* 2010). According to Astuti *et al.* (2018) after these secondary metabolite compounds enter the body, the compounds will diffuse with the blood and flow so that the larval body's defenses are disrupted and the recreation of secondary metabolites that are disturbed will stop. Larval death due to botanical insecticides combined with babadotan leaf extract and tobacco stems occurs when leaves containing botanical insecticides are eaten by larvae until the larvae become less mobile or limp and eating activity decreases, then the larvae will die. The content of secondary metabolites contained in both extracts causes disruption of the digestive process, damages the digestive system, disrupts the absorption of nutrients, and causes death.

Based on observations in the study, the death of *C. pavonana* larvae caused by extract treatment has the characteristics of a softened body, drying and wrinkling of the larval body so

that the body wall appears to shrink, and changes in body color to brown to blackish (Figure 4.). Larvae that died due to the combination of extracts were characterized by blackened larval bodies, dried and wrinkled bodies (Figure 4). Larvae mortality due to babadotan leaf extract is characterized by a brown-black body and a slightly shrunken and softened body, while larval mortality due to babadotan stems is characterized by a brown body, drying, and slightly wrinkled (Figure 4.). According to Astuti *et al.* (2018), the body of larvae that die from botanical pesticides will experience changes in color and shape, become soft and crumble when touched, blackish brown, and the size of the larvae shrinks and shrinks.



Figure 4. Differences in the mortality of *C. pavonana* larvae in the control and larvae killed by the extracts. (a) Ac:Nt extract combination (3:5), (b) single *A. conyzoides* extract, (c) single *N. tabacum* extract, (d) live larvae in control treatment

The metabolic damage to the test larvae was caused by the secondary metabolites contained in the plant-based insecticide. Alkaloids, flavonoids, and saponins are compounds that act as stomach poisons. Alkaloids act as stomach poisons by entering the larval body through the treated feed and then will interfere with the performance of the digestive apparatus and can reduce insect feeding activity with its bitter taste and inhibit the work of taste receptors so that the larvae die of hunger due to not being able to recognize their food (Astuti *et al.*, 2018). Saponins contained in both ingredients will cause the walls of the digestive tract to become corrosive, destroy the digestive tract and reduce the performance of enzyme activity so that the digestive system is disrupted and skin turnover to inhibit insect growth (Javier *et al.*, 2018). The performance of digestive enzymes will decrease due to saponins so that food absorption is disrupted and the insect stomach is irritated, this is indicated by a change in the color of the insect to darken and shrink the body of the insect (Kanmani *et al.*, 2021). Flavonoids contained in the treatment food will enter and cause disruption of the digestive system so that the test larvae can experience death (Javier *et al.*, 2018). Flavonoids that have entered will be absorbed by the mesenteron which functions in the absorption of nutrients and causes poisoning in the larvae, besides that flavonoids cause inhibition of taste receptors in the body of the test larvae so that the larvae lose their ability to recognize their food and the larvae will slowly die (Khan *et al.*, 2015).

Tannin and polyphenol compounds act as stomach poisons and disrupt the digestive process and inhibit the absorption of nutrients. Digestive activity in test larvae can be disrupted by the presence of tannin compounds that can inhibit the process of protein absorption by binding to proteins needed by larvae, so that larval nutritional needs are not met and cause death, besides that tannin acts as a food inhibitor because it has a bitter taste (Pintong *et al.*, 2020). Proteins, carbohydrates, vitamins, and minerals present in insects will be bound to polyphenols so that these nutrients cannot be digested by the stomach, this causes weakening of larval movement until the larvae die (Sakadzo *et al.*, 2020). According to Ragesh *et al.* (2016)

polyphenolic compounds in botanical insecticides can cause changes in the color of the larvae to brown or black when they die. Meanwhile, nicotine can act as a stomach poison that disrupts the digestive process and neurotoxins by attacking the muscle nerves in the nervous system so that the nerves become inactive and cause symptoms of poisoning such as not feeling stimulation, disability, and death (Radev and Stoyanova, 2024).

The mortality rate of *C. pavonana* larvae treated with babadotan leaf extract with a concentration of 17% was 93.33%. This result is lower than the results of research by Ragesh *et al.* (2015). The study resulted in 50% mortality of *H. armigera* larvae at a concentration of 0.17%. While the difference with previous research also occurred in the treatment of tobacco stem extract with a concentration of 11% and 30% which caused mortality of 53.33% and 93.33%. This mortality is higher than the research on botanical pesticides of tobacco stem extract against *H. armigera* by Radev and Stoyanova (2024) which reached 43,3%% with an extract concentration of 10%.

Differences in mortality rates can be caused by differences in the species used in the test, the location where the extract was taken, the feed used during the test, the length of maceration of the material, and the storage of the extract. The location of plant growth affects the content of active compounds because it is influenced by genes as internal factors and external factors which include temperature, humidity, light, and altitude where it grows (Khurseed *et al.*, 2022). According to Nailufar and Prijono (2017), *C. pavonana* and *P. xylostella* have different levels of immunity and immunity that can affect the number of larval mortality rates. Radev and Stoyanova's tobacco extract research (2024) used mustard leaf feed, while this study used broccoli leaves which according to Nailufar and Prijono (2017) have a waxy coating so that active compounds in botanical insecticide extracts are difficult to adhere to the leaves. The maceration of tobacco stems carried out in this study is with the ratio of simplisia and 96% ethanol solvent of 1: 5 for 5 days based on the recommendations of Radev and Stoyanova (2024), while in the research of Ramasamy *et al.* (2021) used the maceration method for 60 hours with the ratio of simplisia: 96% ethanol solvent, namely 1: 4. The difference in the length of maceration time also occurred in babadotan extract, in the research of Ragesh *et al.* (2015) maceration was carried out for 4 days with 96% ethanol solvent, while this study macerated babadotan extract for 3 days with 96% ethanol solvent. The longer maceration will increase the touch process between the simplisia and the solvent so that the extracted compounds will increase (Yooboon *et al.*, 2019). In addition, according to Yooboon *et al.* (2019), the greater the amount of solvent in the maceration process will produce a greater pressure so that the more extracts are produced. According to Nenotek *et al.* (2022), the results of extracts that have been rotated should be immediately stored in the refrigerator with a temperature of  $\pm 4^{\circ}\text{C}$  and immediately used for research. However, in this study, the extract was stored outside the room for several hours before being put into the refrigerator. In addition, the extract was stored too long in the refrigerator for more than one month before finally being used in testing. Storage of extracts is influenced by the length of storage time and temperature because it can affect the toxicity performance of the extract during testing to test larvae (Syahputra and Minarti, 2022).

**LC<sub>50</sub> of Tobacco Stem and Babadotan Leaf Extracts and their Combination against *C. pavonana* Larvae**

The toxicity of the extracts of *A. conyzoides*, *N. tabacum* and their mixture can be known through the LC<sub>50</sub> value that has been analyzed through probit analysis. Lethal concentration 50 or LC<sub>50</sub> is the concentration of an ingredient needed to kill the test larvae by 50%. LC<sub>50</sub> values are presented in table 1. Based on table 1. the highest LC<sub>50</sub> value is in *N. tabacum* extract which is worth 10.640%, then *A. conyzoides* extract has an LC<sub>50</sub> value of 6.710%. The lowest LC<sub>50</sub> value was found in the combination of *A. conyzoides* and *N. tabacum* extracts at 5.245%.

Table 1. LC<sub>50</sub> value of *N. tabacum* stem extract and *A. conyzoides* leaf extract and its combination against *C. pavonana* larvae

Extract	LC <sub>50</sub> (%)	Confidence Interval 95%		Slope	SE Slope
		Lower Bound	Upper Bound		
<i>A. conyzoides</i>	6,71	3,962	8,531	1,071	0,566
<i>N. tabacum</i>	10,64	4,573	14,309	1,031	0,783
Ac:Nt (3:5)	5,245	2,895	6,747	1,455	0,637

The results of probit analysis obtained during the study showed that the LC<sub>50</sub> of the mixture of *A. conyzoides* and *N. tabacum* extracts (3:5) had a lower value than the LC<sub>50</sub> value of each extract (Table 1). This indicates an increase in toxicity in the mixture of *A. conyzoides* and *N. tabacum* extracts so that it is more efficient than the use of single extracts because in the mixture of the two extracts a lower concentration level is used than the concentration in the single extract. The lower LC<sub>50</sub> value on a botanical insecticide material indicates that the more toxic the extract is (Priyono *et al.*, 2021).

**Nature of Activity of Tobacco Stem and Babadotan Leaf Extracts against *C. pavonana* Larvae**

The mortality data of *C. pavonana* larvae were analyzed by probit analysis to determine the LC<sub>50</sub> and LC<sub>95</sub> values. The LC<sub>50</sub> and LC<sub>95</sub> values were used in the calculation of the combination index to determine the activity properties of the mixture of *N. tabacum* and *A. conyzoides* extracts. The IK value and the nature of the mixture interaction are presented in table 2. Based on table 2, the IK value of the mixture of *A. conyzoides* and *N. tabacum* extracts at the LC<sub>50</sub> and LC<sub>95</sub> levels is 0.691 and 0.563, respectively. Both levels are weakly synergistic. This is because the IK value is between 0.5 and 0.77 so it is classified as a mixture that is weakly synergistic.

Table 2. Toxicity of Tobacco Stem and Babadotan Leaf Extracts against *C. pavonana* Larvae

Extract	Combination Index		Nature of Activity	
	LC <sub>50</sub>	LC <sub>95</sub>	LC <sub>50</sub>	LC <sub>95</sub>
Ac+Nt (3:5)	0,691	0,563	Weak Synergistic	Weak Synergistic

The weak synergistic properties of the mixture of *A. conyzoides* and *N. tabacum* extracts showed that the mixture treatment gave a slightly higher mortality rate of *C. pavonana* larvae than the treatment of each extract (Priyono *et al.*, 2021). According to Priyono *et al.* (2021), pest control using a botanical insecticide mixture of several extracts that have synergistic properties

can reduce the application dose of the insecticide so as to minimize the negative impact received by organisms other than the target. This can be shown through the research of Astuti *et al.* (2018) the combination treatment of *A. indica* and *C. inophyllum* (3:1) at 0.1% concentration gave a higher mortality rate of 69% compared to the single extract treatment of *A. indica* at 0.1% concentration which only gave a mortality rate of 62%. It can be seen that the concentration of the combination is smaller and gives the highest mortality compared to the concentration of a single ingredient so that the combination of extracts can be said to reduce the dose of single ingredient extracts so that the raw materials used are also reduced. Mixing several extracts in insecticides also helps farmers in overcoming the limited amount of extract raw materials whose availability is not always available (Dadang and Prijono, 2008).

Synergistic properties are thought to arise due to the addition of secondary metabolites in the combination of extracts. Tobacco extract has nicotine that babadotan does not have, while babadotan extract has tannins and polyphenols that tobacco does not have. Nicotine is a neurotoxin that can affect the central nervous system in insects, causing interference with the transmission of impulses and muscle nerves until muscle performance decreases, muscle nerves are inactive, and death (Amoabeng *et al.*, 2023). Tannin compounds that enter insects will bind to proteins in digestion and reduce the activity of digestive enzymes so that the performance of insects in digesting food will be disrupted (Pintong *et al.*, 2020). Polyphenolic compounds will also mengikan nutrients needed by insects such as proteins, carbohydrates, vitamins, and minerals so that these nutrients cannot be digested and the movement of insects becomes weak and eventually dies (Sakadzo *et al.*, 2020).

### CONCLUSIONS AND SUGGESTION

Based on research, babadotan extract is more toxic than tobacco extract because it has a mortality of 93.33% at a concentration of 17% and an  $LC_{50}$  value of 6.710. The combination treatment of Ac:Nt (3:5) was more toxic than the single extract treatment because it had a mortality of 96.67% at a concentration of 13% and an  $LC_{50}$  value of 5.245. The combination of Ac:Nt (3:5) is weak synergistic at the  $LC_{50}$  and  $LC_{95}$  levels with IK values of 0.691 and 0.563.

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