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Alkaloid compound from Tombili (*Caesalpinia bonduc*) as biopesticide agent on rice plants

Weny J.A Musa[1], Suleman Duengo[1], Ahmad Kadir Kilo[1] and Boima Situmeang[2,*]

[1]Department of Chemistry, Faculty of Mathematic and natural Sciences, Gorontalo State University, Gorontalo, Indonesia
[2]Department of Chemistry, Sekolah Tinggi Analis Kimia Cilegon, Banten, Indonesia
*Corresponding author: BS, boimatumeang@gmail.com

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Abstract: The purpose of these study is to determine the effect of active compound from tombili seed extract as a vegetable pesticide in the prevention of soil pest in rice plants. The method of these researches is isolation to obtain pure isolates, characterization by using infrared spectrophotometer (IR) and UV-VIS spectrophotometer then following biological test. The isolates obtained were tested positive for phytochemical containing alkaloid compounds and characterized as amorphous crystals. Identification of IR spectrophotometer generate a functioning -NH wave group of the wavelengths region of 3423.4 cm^{-1}, bend C-N at wave number 1242.1 cm^{-1}, C-H stretch at wave number 2925.8 and 2854.5 cm^{-1}, bend C-H at 1475 wave numbers 1540 cm^{-1}, carbonyl group (C = O) at the 1720.4 cm^{-1} wave number which is probably an alkaloid compound. The UV-VIS spectrophotometer results in a maximum absorption at 209.9 nm wavelengths indicated the presence of double bonds between the C = O functional group suspected as a result of the successive electronic transitions to n-π * and n-σ *. The result of biological test showed that methanol extracts of tombili seed can be used as a vegetable pesticide in pest prevention on rice plants, the most effective concentration used in this biological test is at 0.1% concentration, with very high effectiveness in eradicating pest on rice plants.

Keywords: Tombili, Alkaloid, Biopesticide

1. Introduction

Farmers have been dependent on chemical pesticides to control plant pests and diseases. Gapoktan (2009) reported that the application of insecticides of synthetic chemicals are less prudent and incompatible with integrated pest control (IPC) can provide various negative impacts such as pest resites, secondary pest, killing of non-target organisms, insecticide residues in foodstuffs, environment, and danger to the

user (Gapoktan, 2009). As an alternative, now began to be developed the use of plant materials to be used as a vegetable pesticide (Kardinan, 2011).

Vegetable pesticides are pesticides used for pest and disease control for plants made from natural materials, such as plant organs, or oils produced by plants. Vegetable pesticide has several advantages such as easily decomposing by sunlight and does not cause environmental disturbance, while for the loss for the use of this vegetable pesticide, the way its application must be repeatedly because it is easily decomposed by sunlight, price is not affordable by farmers because the manufacturer of these pesticides using materials from nature that has insufficient stock for mass-producing vegetable pesticides (Kardinan, 2011).

Vegetable pesticides have several functions, among others: repellent; Rejects the presence of insects, for example with a strong odor, antifidant, prevents insects from eating sprayed plants, destroys the development of eggs, larvae, inhibits the reproduction of female insects, nerve toxins, disrupts the nervous system in insects, insects, which can be used as an insect trap, control of fungi or bacteria (Syakir, 2011; Pandya, 2018; Simorangkir et al. 2020).

The use of vegetable pesticides are expected to suppress the population of pests that attack the plant without deadly pests. However, the use of pesticides are sometimes (very rare) deadly pests but only cause toxins in these pests such as stomach poison, reducing appetite pests and others (Moon et al. 2010). Tombili, is one type of plant that has the opportunity to be used as a vegetable pesticide because it contains many secondary metabolites. This plant is a type of thorn bush plant that is widely spread mainly in India, Sri Lanka, and Andaman and Nicobar Islands (Pandonou et al. 2015; Amudha et al. 2016). Based on the above, the researcher is interested to apply to rice plants by using isolated from methanol fraction of tombili seeds. As a vegetable pesticide in pest controls in rice crops.

2. Methods

2.1 Materials

The materials used in this research are tombili seed, collected from Bobohu vilages, Gorontalo province, Indonesia, rice pest, rice plant, methanol container, aquatic, n-hexane, ethyl acetate, silica gel, dragendrof reagent, wagner reagent, mayon reagent, concentrated hydrochloric acid, magnesium powder, sodium hydroxide, concentrated sulfuric acid, methylene chloride, acetone, and chloroform.

2.2 General Method

Extraction: The smooth of 1.5 kg tombili beans are macerated using methanol as solvent, to obtained methanol extract. The extracts were then evaporated at a temperature of 30-40°C to obtain a thickened extract of methanol. The extract obtained was phytochemical test.

Phytochemical Screening: Phytochemical tests was done to know the class of secondary metabolite compounds contained in tombili seed extract. Phytochemical tests to include flavonoid test, alkaloid test, steroid test, terpenoids, tannin test, and saponin (Situmeang et al. 2019).

The Process of Separation and Purification: The separation and purification processed is carried out using Thin Layer Chromatography (TLC) and column chromatography.

Characterization of Compounds: The characterization of the active compound class was performed by UV-Vis spectrophotometer and infrared spectrophotometer.

2.3 Biological Testing

In biological test about conducted application of rice plants aimed to determine the effectiveness of isolation from fraction of methanol tombili seeds as a vegetable pesticide. Tested using a jar-shaped container in which the filter papers was inserted and three sheets having been cut in the same form were weighed and immersed for ± 5-10 seconds in isolate of methanol fraction of tombili seeds with different concentrations of 0.1%, 0.05% and 0.01% coupled with variation of control (control without leaf, control + leaf, and leaf + methanol) then inserted pest on prepared container. The pests used are pests which obtained from farmers in the area around Gorontalo, with the type of pest is earthen wall. This pest is the biggest enemy of the farmers because when the pest wall pest attacks the rice plants, the plants become brown or yellow. As a result farmers have decreased production. Applications and observations were performed for 1 x 24 hours, with the first pest being used for ± 8 hours. Isolate methanol fraction of each tomato seed concentration.

3. Results and Discussion

3.1 Extraction

Tombili seeds sample, which is finely extracted by maceration. The purpose of maceration is to extract the chemical compound components contained in the sample. The maceration process is done for 3 x 24 hours by using methanol as solvent. Methanol used as a solvent because it can bind all chemical components contained in plants, both non polar and polar. The obtained macerate was evaporated using a rotary evaporator at a temperature of 45°C. Extract of methanol from tombili seeds obtained as much as 68 g.

3.2 Phytochemical Test

Phytochemical tests were performed on methanol viscous extract from tombili seeds including flavonoid test, alkaloid test, steroid test, terpenoids, saponin, and tannins. The results of phytochemical test of viscous extract showed positive flavonoids, alkaloids, terpenoids, saponins, tannins and showed negative results on the steroid test.

3.3 Separation and Purification

Separation and purification is done by adsorption gravity column chromatography. The column used is a column that has a length of 50 cm. The column is filled with n-hexane solvent and then inserted a silica gel of 30 g with a height of 20 cm in a dry stand slowly as its stationary phase. The column is eluted for approximately 4 hours and left for 1 night. A total of 10 g of viscous methanol extracted was added with gel silica, open faucet state. The extract was eluted with combination n-hexane: methylencloride (MTC): Acetone gradually increased with solvent polarity by 10% and obtained by 34 vials. The vials were analyzed by thin layer chromatography (TLC) using eluen n-hexane: methylencloride (MTC): acetone (6: 2: 2). Obtained 3 fraction, fraction of fraction is given symbol of MT which mean methanol tombili. There are three fractions of the methanol fraction of tombili 1 (MT1) (1-8) 0.9 g, methanol tombili 2 (MT2) (9-17) 0.15 g, and methanol tombili 3 (MT3) (18-34) 3 g. At the fraction of MT3 further separation was performed using a smaller column chromatography, using combination of ethyl acetate: methanol gradually with a 10% polarity increase. The separation results obtained 20 vials, then analyzed by TLC to see the stain pattern of each vial obtained 2 fractions ie the MT3.1 (1-10) 0.68 g and MT3.2 (11-20) 0.98 g. The result of this separation yields 40 vials. Each vial is done by TLC analysis, to see the stain pattern. The same stain patterns were combined and obtained 3 fractions of the MT3.1.1 (1-11) 0.18 g fractions and the MT3.1.2 (12-20) 1.78 g and MT3.1.3 (21-40) 1.86 g. Analyze of thin layer chromatography (TLC) show a single stain indicating the presence of pure isolating.

3.4 Purity Test

In the fraction of MT3.1.1 indicates the possibility of pure isolating, so do the purity test using thin layer chromatography (TLC) method using various eluent comparisons of n-hexane: MTC: acetone (9: 1: 0,5), n-hexane : Ethyl acetate (8: 2), n-hexane: acetone (8: 2), ethyl acetate: methanol (9: 1). Based on the analysis of thin layer chromatography (TLC), the MT3.1.1 fraction indicates the presence of pure isolating, this is also seen in the existing stain pattern giving only a single stain pattern on various eluent variations. The MT3.1.1 fraction showing pure isolates was tested for its purity with a two dimensional thin layer chromatographies analysis. Two dimensional thin layer chromatography was performed using different eluent (mobile phase), where the sample (MT3.1.1 fraction) was bottled on KLT plate and eluted using eluen n-hexane: acetone (8: 2) as E1, then The TLC plate was removed, dried and rotated 90°, and eluted using a different eluent chloroforom: methanol (8: 2) as E2.

3.5 Pure Isolate Phytochemical Test

The fraction of the resultant column is a pure isolate phytochemical tested, to see the compounds contained therein. Phytochemical results of pure isolate showed that

pure isolates to contain alkaloid compounds. This is evidenced by the formation of precipitate on the Dragendrof reagent. While the flavonoid test, steroid, and terpenoid showed negative results with no absence of discoloration of each reagent.

3.6 Characterization of Compounds

The characterization of the active compound class was performed by infrared spectrophotometer and UV-Vis spectrophotometer.

3.7 Infrared Spectrophotometer

Infrared spectrophotometer is used to identify functional groups contained in isolating. The result of infrared spectrophotometer is shown in Fig 1.

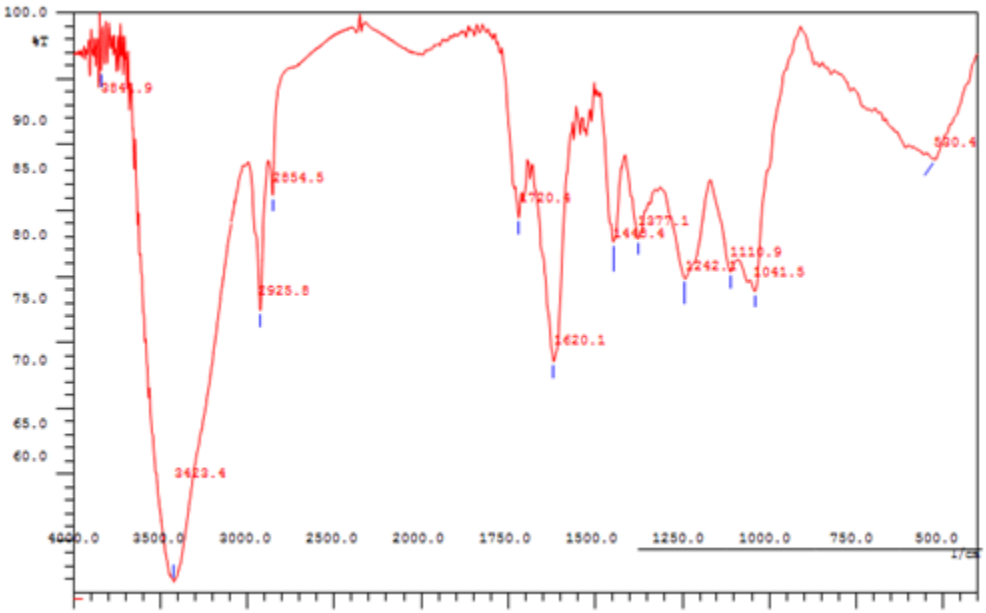

Fig. 1 infrared spectrophotometer of sample

According to Fig. 1. infrared spectrophotometer results show that isolates from the MT3.1.1 fraction indicate the presence of uptake -N-H group at wavelength λ 3423.4 cm-1 wave numbers seen in 3300-3500 cm-1 regions (Prachayasittikul et al. 2010; Susilawati at al. 2015). This absorption is supported by the presence of absorption at 1242.1 cm-1 wave numbers indicating a C-N bending uptake seen in the 1020-1250 cm-1 region (De Leon et al. 2010). The presence of sharp bonds with weak intensity at the wavelengths of 2925.8 cm-1 and 2,2854.5cm-1 represents CH stretching in the region of 2850-2950 cm^{-1} and supported by the presence of sharp absorption and weak intensity at 1475 wave numbers -1540 cm-1 which is the vibration of CH (Tijjani et al. 2012).

The carbonyl group (C = O) is present in the 1720.4 wave number seen in the 1870-1540 cm-1 region (Silverstein. 1984), which is supported by the presence of a sharp and weak absorption at the wave number 1620.1 cm-1 in the region of 1500 -1675cm-1 .

It can be concluded that isolate has characteristic of functional group -N-H, C-H, C-N, and C = O (Harbone, 1987).

3.8 Biological Testing

This biological test was conducted to determine the effectiveness of isolation from methanol fraction of tombili seeds as a vegetable pesticide in pest prevention on rice plants. The isolates used as biological samples was the result of the separation and purification of MT3.1 showing the presence of a single stain indicating the presence of pure isolating and obtaining three fractions of the fraction of MT3.1.1 (1-11) 0.18 g and the fraction of MT3.1.2 (12-20) 1.78 g and MT3.1.3 (21-40) 1.86 g. Each fraction was diluted with concentrations of 0.1%, 0.05%, and 0.01%, supplemented by control variation (leafless control, leaf + control, and leaf control + methanol) after each concentration was prepared and prepared Container and inserted filter paper that has been adjusted to the size of the container. Then include 3 pieces of fresh rice leave that have been applied with concentration preparations. After all the containers are filled with rice leaves with each concentration are then inserted pests. Pests used are pests obtained from farmers in the area around Gorontalo, with the type of pest is the earthenwall, and observed 1 × 24 hours.

From the results of the study showed that the concentration of isolating that has excellent effectiveness in the biological test is the concentration of 0.1%. This can be seen from the death of pests (wall bones).

Ground wall is one of the pest of rice plants, which can cause loss for farmers. The characteristics of this bamboo are foul, attack the plant by sucking liquid in plants, body size with a length of 1 cm and 1 cm wide. How to eat is done by mouth suckers, suck stem liquid, plant leaves. The walls suck the juices of the food in the stems, the leaves of plants, so as to reduce the energy and nutrients needed by plants (Paendong et al. 2011). Aspiration by the soil wall in the tillers phase causes the number of tillers to decrease and the growth is stunted (stunted), and the grain of the hampah, the sucking pitch turns brown or yellow. The leaves become dry and roll longitudinally. If the attack occurs after the pregnancy phase, the plants show symptoms of stunting in formation of panicles to the absence of panicles, or forming a dwarf panicle, and empty grain (Paendong et al. 2011). The death of the pest (earthen wall) on the biological test is caused by the presence of toxic compounds in tombile seeds that work as toxins, the alkaloid compounds. These alkaloids are the most common organic compounds found in nature. Alkaloids can generally be defined as a basic substance having one or more alkaline nitrogen atoms and incorporated into a cyclical system, ie, a heterocyclic ring. Alkaloids will taste bitter like the tongue, Alkaloid categorized as toxic, the compound exhibits a wide physiological activity, almost without exception, alkaline-containing nitrogen in heterocyclic rings (Harbone, 1987). Alkaloids can inhibit the feeding power of insects, alkaloid compounds can act as stomach poisoning or stomach poison. Therefore,

when the alkaloid compounds into the insect body, the digestive tool will be disrupted. In addition, the compound inhibits the taste receptor in the insect's mouth area. Stomach toxins will also affect larval metabolism after eating toxins. The poison will enter into the body and circulated with the virgin. Blood-borne toxins will affect the nervous system and then will cause death (Muaddibah, 2016).

4. Conclusion

The optimum concentration of methanol extracted of the most effective tombili seed is used as a vegetable pesticide that is at a concentration of 0.1%. The results of the phytochemical isolate test of methanol extract of tombili seed are alkaloids.

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References

Amudha, P., Bharathi, N. P., & Vanitha, V. (2016). Caesalpiniabonducella – A REVIEW ON pharmacological and phytochemical activity of seeds. *International Journal of Pharma and Bio Sciences*, 7(4), 674-680. **DOI:**10.22376/ijpbs.2016.7.4.b674-680.

Gapoktan, 2009. *Kebijakan pertanian dalam pengembangan gabungan kelompok tani gapoktan sebagai keembagaan ekonomi diperdesaan*. (http://pse.litbang.pertanian.go.id) acces 1 august 2016.

Harborne, J. (1987). *Metode fitokimia: penuntun cara modern menganalisis tumbuhan*. Cetakan kedua. Padmawinata K, Soediro I, Penerjemah. Bandung: Penerbit ITB.

Syakir, M. (2011). Status penelitian pestisida nabati pusat penelitian dan pengembangan tanaman perkebunan. *Seminar Nasional Pesnab IV, Jakarta*.

Kardinan, A. (2011). Penggunaan pestisida nabati sebagai kearifan lokal dalam pengendalian hama tanaman menuju sistem pertanian organik. *Pengembangan Inovasi Pertanian*, 4(4), 262-278.

Muaddibah K. 2016. Pengaruh ekstrak daun legetan (*Synedrella nodiflora*) terhadap perkembangan ulat daun kubis (*Plutella xylostella*). [Skripsi]. Malang (ID): UIN Maulana Malik Ibrahim.

De León, L., López, M. R., & Moujir, L. (2010). Antibacterial properties of zeylasterone, a triterpenoid isolated from Maytenus blepharodes, against Staphylococcus aureus. *Microbiological Research*, 165(8), 617–626. **DOI:**10.1016/j.micres.2009.12.004

Moon, K., Khadabadi., Deokate, U., and Deore. (2010). Caesalpina bonducella. Report and opinion: 2(3).

Paendong, E., Pelealu, J., & Rimbing, J. (2011). Penyebaran hama kepinding tanah dan musuh alaminya pada pertanaman padi sawah di Sulawesi Utara. *EUGENIA*, 17(3), 178-186. **DOI:**10.35791/eug.17.3.2011.3541

Padonou, E. A., Ahossou, O. D., Okou, F. O., Assogbadjo, A. E., Kakaï, R. G., Lykke, A. M., & Sinsin, B. (2015). Impact of climate on seed morphology and plant growth of Caesalpinia bonduc L. in West Africa. *International Journal of Agronomy and Agricultural Research (ijaar)*, 6(3), 86-96.

Pandya, I. Y. (2018). Pesticides and their applications in agriculture. *Asian J Appl Sci Technol*, 2(2), 894-900.

Prachayasittikul, S., Saraban, P., Cherdtrakulkiat, R., Ruchirawat, S., & Prachayasittikul, V. (2010). New bioactive triterpenoids and antimalarial activity of Diospyros rubra Lec. *Excli Journal*, 9, 1-10.

Silverstein, Bassler, dan Morill 1984. Penyidikan spektrometrik senyawa organik. Edisi ke-4, Jakarta : Erlangga

Situmeang, B., Ibrahim, A. M., Bialangi, N., Musa, W. J., & Silaban, S. (2019). Antibacterial activity and phytochemical screening of Kesambi (Sapindaceae) against Eschericia coli and Staphylococcus aureus. *Jurnal Pendidikan Kimia*, 11(1), 14–17. **DOI:**10.24114/jpkim.v11i1.13078

Simorangkir, M., Nainggolan, B., Doloksaribu, J. F., & Silaban, S. (2020). Effect of sarang banua (*Clerodendrum fragrans* Vent Willd) leaves extract on serum globulin levels of rabbit (*Oryctolagus cuniculus*). Journal of Physics: Conference Series, **1485**, 012016. **DOI:**10.1088/1742-6596/1485/1/012016

Susilawati, Y., Nugraha, R., Muhtadi, A., Soetardjo, S., & Supratman, U. (2015). (S)-2-Methyl-2-(4-methylpent-3-enyl)-6-(propan-2-ylidene)-3,4,6,7-tetrahydropyrano[4,3-g]chromen 9(2H)-one. *Molbank*, 2015(2), M855. **DOI:**10.3390/m855

Tijjani, A., Ndukwe, I., & Ayo, R. (2012). Isolation and characterization of lup-20(29)-ene-3, 28-diol (Betulin) from the stem-bark of adenium obesum (Apocynaceae). *Tropical Journal of Pharmaceutical Research*, 11(2). **DOI:**10.4314/tjpr.v11i2.12