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POTENTIAL ANTICOAGULATION ACTIVITY OF RAMBOT BUE (*Passiflora foetida*) PLANT LEAF JUICE

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ABSTRACT

Substances that can be used to prevent blood clots are called anticoagulants. The anticoagulant drug used to treat blood clotting disorders that is commonly used today is heparin. Heparin is reported to have side effects of bleeding, osteoporosis, and thrombocytopenia. Based on these conditions, research is needed to look for anticoagulants from natural ingredients such as plants. Several plants in Indonesia are reported to have anticoagulant activity. The Rambot Bue plant (*Passiflora foetida*), commonly known as rambosa, whose leaves are often used by the people of Aceh as a mixture in vegetable stew. This plant is efficacious as a traditional medicine such as an analgesic and is reported to have many secondary metabolic activities. Therefore, it is necessary to carry out research to find out whether the Rambot Bue plant has the potential to have anticoagulant activity. In this research, the anticoagulant activity of Rambot Bue plant leaf juice will be tested in vitro in the laboratory using the modified Lee and White method. The anticoagulant activity test to determine the potential anticoagulant activity of Rambot Bue leaf juice gave results that no coagulation occurred in five blood specimens to which the juice of Rambot Bue plant leaves was added with juice in volumes of 450 µL, 400 µL, 350 µL and 300 µL. This condition shows that Rambot Bue leaf juice has the potential to have anticoagulant activity up to a volume of 300 µL.

Keywords: *anticoagulant, Passiflora foetida, Lee and White.*

INTRODUCTION

Substances that can be used to prevent blood clots are called anticoagulants (Gunawan et al., 2022). Anticoagulant drugs are used to treat coagulopathy, namely disorders of the coagulation or blood clotting system

which can manifest as blood clots (thrombus) in veins, arteries or overall (systemic) (Susi Endrawati dan Anom Parmadi, 2022). Some of the causes of coagulopathy are endothelial damage due to inflammation in viral infections, atherothrombosis such as myocardial infarction and ischemic stroke (Pratiwi & Adhityasmara, 2021), (Mende et al., 2022). The anticoagulant drug currently frequently used is heparin (Mende et al., 2022), (Pratiwi & Adhityasmara, 2021), (Rahmawati Rahmawati et al., 2018). However, the anticoagulant drug heparin is stated to have side effects including causing bleeding, osteoporosis and thrombocytopenia (Rohmah & Fickri, 2020). Based on these conditions, research is needed to look for anticoagulants from natural ingredients such as plants. Based on research that has been conducted, there are 58 plants in Iraq that contain secondary metabolic compounds with anticoagulant activity (Pour et al., 2017).

Indonesia is a country that is rich in various kinds of plants that have the potential to be used as raw materials for medicine (Gunawan et al., 2022). Several plants in Indonesia have also been studied to have anticoagulant activity, including garlic (*Allium sativum*) (Rahmawati Rahmawati et al., 2018), kayu ular (*Strychnos lucida*), (Gunawan et al., 2022), papaya (*Carica papaya L*) (Rohmah & Fickri, 2020) and many more.

Currently, many studies study the relationship between plants and humans, which is known as Ethnobotany. Ethnobotany studies how to use plants in everyday life, namely a descriptive form of botanical knowledge possessed by people in certain areas (Dewantari et al., 2018). One of the plants that is often used by the people of Aceh is the Rambot Bue Plant (*Passiflora foetida*). The Rambot Bue plant is generally known as rambosa. Acehnese people often use Rambot Bue leaves as a mixture in vegetable stew.

Rambot Bue is a plant that grows in plantation areas, grasslands, edges and roads. The leaves and fruit of this plant contain various nutrients including potassium, sodium, calcium, flavonoids and high iron content, while the fruit is rich in vitamin C and fiber (Rahmandani et al., 2021). The Rambot Bue plant contains bioactive components which have been used as traditional medicinal plants in several places in Indonesia and are reported as an analgesic drug. The phytochemical components of the Rambot Bue plant include alkaloids, phenols, glycosides, flavonoids, cyanogenic compounds, passiflorisin, polypeptides, alpha-pyrone, tetrafilin A, tetrafilin B, tetrafilin B sulfate, deidacin, and volkenin (Rohmania et al., 2024), (Saputri et al., 2021). Several studies also state that the Rambot Bue plant has several secondary metabolic activities including antioxidant, antidiabetic, anticholesterol and antibacterial activities (Sari & Puspitasari, 2021).

Based on the extensive secondary metabolic activity of the Rambot Bue plant, it is necessary to carry out research to find out whether the Rambot Bue plant has the potential to have anticoagulant activity. In this research, the anticoagulant activity of Rambot Bue plant leaf juice will be tested in vitro in the laboratory using the modified Lee and White method.

METHODS

This research is a type of laboratory experimental research to test the activity of Rambot Bue leaf juice as an anticoagulant in vitro. The anticoagulant activity test method is the modified Lee and White method (Gandasoebrata, 2011). The test was carried out with 4 variations in volume of Rambot Bue leaf juice (450 µL, 400 µL, 350 µL and 300 µL)

Material

A 500 gram sample of Rambot Bue leaves was collected from the Lambaro Skep Village area, Kuta Alam District, Banda Aceh City, Aceh Province. Whole blood specimens of 5 cc were obtained from 5 volunteers with the assumption that the volunteers were healthy and not undergoing treatment. The equipment used is a erlenmeyer flask 250 cc, a glass funnel, 5 cc syringe, a 3 cc test tube with a diameter of 7 mm and a tourniquet. The materials needed are alcohol cotton, dry cotton, plaster and tissue.

Preparation of Rambot Bue (*Passiflora foetida*) Leaf Juice

A 500 grams of Rambot Bue leaves are washed thoroughly and drained at room temperature until there are no water droplets. Next, it is crushed using mortal until it becomes a slurry and squeezed using gauze, the juice is collected in a erlenmeyer flask.

Whole Blood Specimen Preparation

Palpate the veins in the elbow crease, clean with 70% alcohol and leave to dry. Place the tourniquet 3-4 inches above the elbow crease and the volunteer is asked to clench his fist so that the vein is clearly visible. The skin is punctured with a syringe needle until the tip of the needle enters the lumen of the vein. Remove the tourniquet and slowly withdraw the syringe pump until the blood volume is exactly 5 cc. Place dry cotton wool on the skin where the needle is pierced and remove the needle and syringe. Place a band aid on the puncture mark. Remove the needle from the syringe and pass it through the wall of the test tube (Nugraha et al., 2022).

In Vitro Anticoagulant Activity Test using The Lee And White Method

Prepare 5 test tubes, place them on the test tube rack and give each an identity : Control, 450 µL, 400 µL, 350 µL and 300 µL. The tube with the control identity is left empty. Tubes with volume identities, each filled with Rambot Bue leaf juice with a volume according to identity. Perform a venous phlebotomy and add 1 ml of whole blood specimen to each tube. Each test tube was homogenized slowly and left for 3 minutes on the test tube rack. Use a stop watch to measure time. After 3 minutes, lift each tube and tilt it 45° to observe blood clotting, with the stopwatch still running. If the blood in the test tube has not yet clotted, then place the test tube back on the rack with the stop watch still running. Every 30 seconds lift each tube and tilt it 45° to observe blood clotting, with the stopwatch running. If blood clots occur in one of the test tubes, immediately record the time. The test is stopped when the blood in the tube coagulates or after a test time of 120 minutes (Gunawan et al., 2022).

RESULTS AND DISCUSSION

Research on the potential anticoagulant activity of Rambot Bue (Passiflora foetida) leaf juice was carried out on 5 blood specimens from 5 volunteers using the modified Lee and White method. This research used 4 variations in the volume of Rambot Bue leaf juice (450 µL, 400 µL, 350 µL and 300 µL). The results want to see whether clots occur in the blood given Rambot Bue leaf juice within a certain period of time. The results of each blood specimen can be seen in Table. 1.

Table 1. Observation Of The Clotting Time Of Blood Specimens With The Addition Of Rambot Bue Plant Juice

No	*Blood Specimen Code*	*Blood Specimen Coagulation Time Period (Minutes)*				
		Tube Test Identity				
		Controle	*450 µL*	*400 µL*	*350 µL*	*300 µL*
1.	I	6,5	-	-	-	-
2.	II	7,5	-	-	-	-
3.	III	6.0	-	-	-	-
4.	IV	7,5	-	-	-	-
5.	V	6,5	-	-	-	-

Based on Table. 1. It can be seen that there are differences in the coagulation times of blood specimens from 5 volunteers. Based on references, coagulation time should not exceed 15 minutes and is normally 3–9.5 minutes for arm vein blood specimens. Thus, the volunteers' blood coagulation time was still within normal limits.

The coagulation process did not occur in five blood specimens that were added to the juice of the leaves of the Rambot Bue plant (Passiflora foetida) in volumes of 450 µL, 400 µL, 350 µL and 300 µL. This condition shows that Rambot Bue leaf juice has the potential to have anticoagulant activity up to a volume of 300 µL.

The anticoagulant activity of the juice from the leaves of the Rambot Bue plant is probably due to several active ingredients contained in the leaves of the Rambot Bue plant. The leaves of the Rambot Bue plant contain alkaloids, steroids, saponins, flavonoids, tannins and steroids, which are secondary metabolite compounds that can be found in plants and have biological activity (Mulyani, 2019).

In several studies, the alkaloid compound pelitorin is an active amide which has the role of inhibiting or prolonging the activity of thrombin and Factor Xa as well as inhibiting fibrin polymerization. Research related to the alkaloid compounds piperine and pelitorin has been proven to have activity inhibiting thrombin in the formation of platelet fibrin polymerization and Factor Xa. Meanwhile, flavonoids in the form of phytoestrogens are compounds from plants which are known to contain non-steroids with a structure and function similar to estrogen. Phytoestrogens have the potential to act as anticoagulants by preventing the formation of atherosclerosis, preventing vasoconstriction, and inhibiting fibrinogen. (Rohmah & Fickri, 2020). Tannin is a secondary metabolite compound that has an effect on pro-coagulation of blood in an extract. When tannin is used orally it is vasoprotective. Tannins also have an adjuvant effect, namely vasoconstriction in small blood vessels, which is an important parameter in hemostasis, so tannins can be useful as a hemostatic agent in the blood. Meanwhile, Saponin is a secondary metabolite compound which has biological properties such as hemolytic abilities. The red onion plant contains the active compound saponin which is effective as an anticoagulant (Rama, 2020).

CONCLUSION

The anticoagulant activity test to determine the potential anticoagulant activity of Rambot Bue leaf juice gave results that no coagulation occurred in five blood specimens to which the juice of Rambot Bue plant leaves was added with juice in volumes of 450 µL, 400 µL, 350 µL and 300 µL. This condition shows that Rambot Bue leaf juice has the potential to have anticoagulant activity up to a volume of 300 µL.

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