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INTRODUCTION

The Jordan Journal of Pharmaceutical Sciences (JJPS) is a peer-reviewed Journal, which publishes original research work that contributes significantly to further the scientific knowledge in pharmaceutical sciences' fields including pharmaceutical/medicinal chemistry, drug design and microbiology, biotechnology and industrial pharmacy, instrumental analysis, phytochemistry, biopharmaceutics and Pharmacokinetics, clinical pharmacy and pharmaceutical care, pharmacogenomics, bioinformatics, and also JJPS is welcoming submissions in pharmaceutical business domain such as PharmacoEconomics, Pharmaceutical Marketing, and Management. Intellectual property rights for pharmaceuticals, regulations and legislations are also interesting topics welcomed from our colleagues in Schools of Law.

On a current topic in Pharmaceutical Sciences are also considered for publication by the Journal. JJPS is indexed in SCOPUS (Q3). The Editorial Team wishes to thank all colleagues who have submitted their work to JJP). If you have any comments or constructive criticism, please do not hesitate to contact us at jjps@ju.edu.jo. We hope that your comments will help us to constantly develop JJPS as it would be appealing to all our readers.

Prof Ibrahim Alabbadi
Editor-in-Chief
School of Pharmacy- The University of Jordan
Amman 11942- Jordan

Letter from the Editor-in-Chief

Another year went by. It was an extraordinary year that none of us will soon forget, not only because of hard health times, but also because of the bad economic crisis. However, after every dusk comes the light, hoping that 2021 would be the start of the dawn. Jordan Journal of pharmaceutical Sciences (JJPS) completed 2020 publishing 4 issues on regular times; one issue per quarter (achieving an extra issue than the years before), besides having 10 articles per issue (instead of 5) in order to decrease the waiting time for the accepted articles to be published; trying to serve as much researchers as we can.



One of the achievements is the diversity areas of submissions to JJPS, the latter makes JJPS distinguished with an added value of a different taste that hopefully matches the journal readers' desires in Jordan as well as in the region. Nowadays we have submissions not only in the pharmaceutical chemistry, pharmacognosy and pharmacology, but also in pharmacy practice, clinical pharmacy, pharmaceutical care and behavioral areas related to humans and patients such as psychological considerations during the COVID-19 pandemic. Furthermore, JJPS received submissions from all around the world; giving readers the opportunity to be exposed more to different scientific research patterns worldwide with an increase in number of submissions by 62% in 2020 compared to 2019. Moreover, citations increased in 2020.

The new members in the editorial board are distinguished professors representing almost all fields of pharmaceutical sciences from different backgrounds coming from diversified research schools from USA, Canada, Europe, Australia, and Jordan. Also, they came from different work environments: governmental and private higher education institutions. The latter started smart and hard work toward becoming one of your choices to submit your article in any of the pharmaceutical fields.

In the new issues of JJPS in 2021, we will see an editorial commentary written by one of our colleagues in JJPS expressing one of the interests and thoughts related to the status que in general from their point of view.

Finally, it is really a great honor to have a new advisory board consisting of well-known scientists from different regional and international countries representing almost all pharmaceutical fields; the JJPS family is sure that the respected scientists will have a positive impact and will add value particularly in the quality of manuscripts accepted for publication. Looking forward to more achievements in 2021.

Prof Ibrahim Alabbadi
Editor-in-Chief

Editorial Commentary

Dear Colleagues and Researchers,

As one of the Editorial Board members of the Jordan Journal of Pharmaceutical Sciences (JJPS), it is my pleasure to write for all my colleagues in the field of pharmaceutical analysis. Although JJPS is a peer-reviewed journal for original research articles, review articles, and short communications related to all aspects of pharmaceutical sciences, I am encouraging you to submit your most recent advances and research work in pharmaceutical and biomedical analysis fields, including drug analysis, analytical methodology, quality control, and instrumentation.

JJPS is indexed in Scopus (Elsevier), Ulrich's Periodicals Directory, Google Scholar, and EBSCO. Furthermore, JJPS has an open access policy, which enables your research to become available to all scientists in the field and thus, increases exposure and citation.

It has to be mentioned that the journal is overseen by an Editorial Advisory board, which consists of eminent and competent researchers in all fields of pharmaceutical sciences. In addition, the editorial board members have rich academic qualifications. Therefore, with the network of both boards, the widespread of the journal is distinguished.

At the end, I encourage all scientists involved in the pharmaceutical analysis field to submit their work, or their postgraduate students work and sense the professionalism that they will encounter from JJPS editorial staff and board. At JJPS, we believe in a quick scientific reviewing process since responding to authors promptly is ideal for pursuing widespread scientific knowledge.

Hoping for more collaborations between us in the future.

Professor Wael Abu Dayyih

Department of Pharmaceutical Medicinal Chemistry and Pharmacognosy
Faculty of Pharmacy and Medical Sciences
University of Petra
wabudayyih@uop.edu.jo

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The bleeding and clotting time analysis of the stem extract of *Musa paradisiaca* var. *sapientum* (L.) Kunze on hemostatic response

Hendrik Setia Budi^{1*}, Wisnu Setyari Juliastuti¹, Yuniar Putri Sulistyowati¹

¹Department of Oral Biology, Faculty of Dental Medicine, Universitas Airlangga

*Correspondence : Hendrik Setia Budi, Department of Oral Biology, Faculty of Dental Medicine, Universitas Airlangga. Jl. Prof. DR. Moestopo No 47 Surabaya, Indonesia, 60132.

ABSTRACT

A herbal medicine that can be used to accelerate bleeding is the Ambonese banana stem sap (*Musa paradisiaca* var. *Sapientum*) through secretion of Adenosin Diphosphate (ADP) and von Willebrand Factor (vWF) in the process of platelet adhesion on the injury vessels. The aim of this study to test the influence of Ambonese banana stem extract in a concentration of 25%, 50% and 100% against hemostatic response on mice. 24 animal study were randomly assigned into four groups. Group 1 was the control, group of 2 was given 25% extract concentration, group of 3 was given 50% extract concentration and group of 4 was given 100% extract concentration of Ambonese banana stem per-oral. Then all the groups were examined their bleeding and clotting time. The administration of Ambonese banana stem extract at the concentrations of 25%, 50 %, and 100% significantly shortened both bleeding time and clotting time compared to control group ($p = 0.00$). The group with 100% extract of the Ambonese banana stem having the shortest bleeding time and clotting time. The conclusions of this study showed Ambonese banana stem extract has potential as the hemostatic agent by shortening bleeding and clotting time on mice.

Keywords: Ambonese banana stem extract, bleeding time, clotting time, hemostatic.

1. INTRODUCTION

The surgical procedures come with a risk of complications which may include pain, nerve injury, swelling, excessive bleeding (hemorrhage), and infections. Severe intraoperative or postoperative hemorrhage is one of the few life-threatening complications for which a dentist may have to initiate management.^{1,2} The bleeding disorder, it doesn't clot fast enough. This results in too much bleeding or long-lasting bleeding, and become port de entry of infection.^{3,4}

Hemostasis is a complex process that leads to the formation of a blood clot at the site of vessel injury and three phases can be distinguished by vascular spasm,

formation of a platelet plug and coagulation. Primary hemostasis starts immediately after damage of the vessel wall with vasoconstriction as a result of local contraction of vascular smooth muscle cells.⁵ The von Willebrand factor (VWF) binds to the exposed subendothelial collagens. Platelets are tethered to the site of endothelial cell injury through the binding of VWF to the glycoprotein Ib receptor of the platelet.⁶

In major oral and maxillofacial surgical procedures, electrocautery and suture ligatures are most commonly used to control bleeding from small and major vessels. However, when the use of pressure is not effective, the use of electrosurgical instruments could endanger teeth or nerves, systemic and, or topical hemostatic agents may be needed.^{7,8} The study shown the effectiveness of Plasma Rich in Growth Factors (PRGF) in controlling bleeding after dental extractions in patients with haemophilia. In this fascinating study there were no risks related to the use

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of blood derivatives, yet good haemostasis and healing of soft tissues were achieved. In other words, the post-extraction socket healed better and faster which can prevent further bleeding.⁹

Indonesia is a rich-tropical country in natural resources with the second highest biodiversity in the world after Brazil. Natural resources, such as plants and animals in Indonesia, are very varied, which can be used for improving natural products. The importance of biodiversity, e.g., in traditional medicine and agriculture, is deep-rooted in Indonesian society.¹⁰ A plant that can be efficacious as a drug and has long been used to accelerate wound healing and tooth pain relief by the people of the village of Trunyan, Bali is Ambonese banana stem extract.¹¹ In banana plants, there are significant compounds, such as saponins, flavonoids, anthraquinones, and tannins contained in both its fruit and sap, as well as lectin compounds found in banana stems. Lectin plays a role in stimulating mitotic cells which has an effect on accelerating wound healing and bleeding cessation.^{12,13} Meanwhile, tannins are astringent, stopping bleeding in the wound.¹⁴ This study aimed to prove Ambonese banana stem extract (*Musa paradisiaca* var. *Sapientum*) administration can accelerate wound healing through hemostatic response.

Materials and Methods

Plant material and methods

Collected from the Plant Conservation Center Botanical Gardens Purwodadi, Pasuruan, Indonesia, Ambonese banana tree will be able to be harvested at age 12-13 months, with the height is about 2.5-3 m, and the stem diameter is about 17.3-18.9 cm wide. Determination was carried out by the Indonesian Institute of Sciences (LIPI) Purwodadi, Pasuruan, Indonesia number 1036/IPH.UPT.03.4/HM/IX to prove that the type of banana tree used is a type of Ambonese banana.

Preparation of the sample

To get Ambonese banana stem's sap in large quantities, the stem need to be cut at the lower end part, then clean it to remove the impurities. After that, made it into small

pieces weighing 200 grams, add 200 ml of sterile distilled water and blend until smooth. The sap and water that have been mixed together are then filtered using a Buchner funnel connected with a vacuum pump (Gast, USA brand) with whatman filter paper number 1. The filtration results are dried using a freeze dryer so that it can be stored for a long time. The making of Ambonese banana stem extracts with concentration of 25%, 50% and 100% was carried out by dissolving dried preparations in sterile aquadest.

Experimental design

The ethical approval to conduct the study was taken from the Research Ethics Committee, Faculty of Dentistry, Universitas Airlangga. This research used a randomized post-test only control group design. We used 24 male mice divided into 4 groups. The first group was the control group with the administration of vehicle. The second group was given Ambonese banana stem extract at a concentration of 25%. The third group was given Ambonese banana stem extract at a concentration of 50%. The fourth group was given Ambonese banana stem extract at a concentration of 100%. Those extracts were given for 3 consecutive days. Bleeding and clotting time examinations then were conducted in each group.

Bleeding time experimental

The mice (*Mus musculus*), male, 20-30 gram of body weight were anesthetized with diethyl ether using inhalation method. Mice are put in a transparent box for easy viewing, which contains cotton with ether, and within a few seconds the activity of the mice begins to decline and then falls asleep. The mice's tail was cut 3 cm long from the tip of the tail, blood that came out after cutting was dropped on absorbed paper. The stopwatch was started to run along with the visible blood coming out of the mice that had been cut. Blood dripped on absorbent papers that should not touch the wound. The stopwatch was then stopped when blood did not drip anymore on the absorbent papers.¹⁵ Mice are generally feed containing low fiber (5%), protein (20%) and fat (5-10%) in pellet.

Clotting time experiment

Blood was taken from the heart of anesthetized mice by surgery using a scalpel, and then 1cc of blood was taken using a tuberculin syringe directly from the heart. The stopwatch was started to run along with the entry of blood on the syringe. The blood that had been taken was then inserted into capillary tube to measure the clotting time. Afterwards, the tubes were tilted slowly and re-stood up at 10-second intervals until the blood in the tubes did not flow again when they were tilted to a tilt angle of 90°. The stopwatch was stopped when the blood had not flowed even though the tubes were tilted at an angle of 90°. Surgical wounds then were stitched and oral antibiotics and analgesics were given.¹⁶

Statistical analysis

All the data were tabulated and assessed using the

Statistical Package for Social Sciences (SPSS) version 20.0. Comparison of various time among four study groups were done using ANOVA test and post hoc Tukey test. A p-value ≤ 0.05 was considered to be statistically significant.

Results

Phytochemical components

The Saponin, flavonoid, tannin, anthraquinone and lectin levels were analyzed using UV-Visible Spectrophotometer by comparing the absorbance values of standard compounds with the extract of Ambon banana stem sap. Detection of saponin compounds with a wavelength of 215 nm, flavonoids with wavelengths of 226 nm, tannins with wavelengths of 275 nm, anthraquinones with wavelengths of 285 nm and lectins with wavelengths of 228 nm (see table 1).

Table 1. The concentration of each component in Ambonese banana stem extract

<i>Components</i>	<i>Number of replicant</i>	<i>Concentration (mg/100mL)</i>
<i>Saponin</i>	3	1.30
<i>Flavonoid</i>	3	0.28
<i>Tannin</i>	3	1.50
<i>Antraquinone</i>	3	0.30
<i>Lectin</i>	3	0.11

Hemostatic properties of plant extract

Bleeding time is a clinical laboratory test performed to evaluate platelet function. It involves the creation of a standardized incision and timing the cessation of bleeding. At sites of vascular injury, platelets are recruited to exposed subendothelial extracellular matrix components via specific platelet receptors involved in adhesion and aggregation. The herbal medicines contain a lot of different compounds which some of them have great complexities. Plants substances such as polysaccharides, mucilages and tannins may modulate and modify the effects of "active components".

Results of the One-way ANOVA test showed that the administration of Ambonese banana stem extract at the concentrations of 25%, 50 %, and 100% significantly shortened both bleeding time and clotting time compared to control group ($p = 0.00$). Ambonese banana stem extract at the concentrations of 100% even had the shortest bleeding time compared to the other groups. In other words, the higher the concentration is, the shorter the bleeding time is. Similarly, in the clotting time examination, the group given Ambonese banana stem extract at the concentration of 100% had the shortest clotting time compared to the other groups (see table 2).

Table 2. Bleeding and clotting time test

Groups	$\bar{X} \pm SD$	
	Bleeding Time (second)	Clotting Time (second)
1	149.5 ^a ± 33.2	183.3 ^a ± 19.5
2	75.2 ^b ± 23.2	128 ^b ± 25.8
3	53.2 ^c ± 7.9	94.7 ^c ± 17.1
4	35 ^d ± 8.53	86.33 ^c ± 13.2

The values with different superscript letters in a column are significantly different ($p < 0.05$).

Discussion

The results of this research showed that bleeding time and clotting time in Group 1 (control) had a significant difference when compared with Group 2 (given Ambonese banana stem extract at the concentration of 25%), Group 3 (given Ambonese banana stem extract at the concentration of 50%) and Group 4 (given Ambonese banana stem extract at the concentration of 100%). In other words, the

administration of Ambonese banana stem extract at the concentrations of 25%, 50%, and 100% can accelerate both bleeding time and clotting time in mice. Moreover, it is also known that Group 1 needed the longest bleeding time compared to the other groups (see figure 1). Ambonese banana stem extract actually contain various active compounds, namely lectins and tannins.^{17,18}

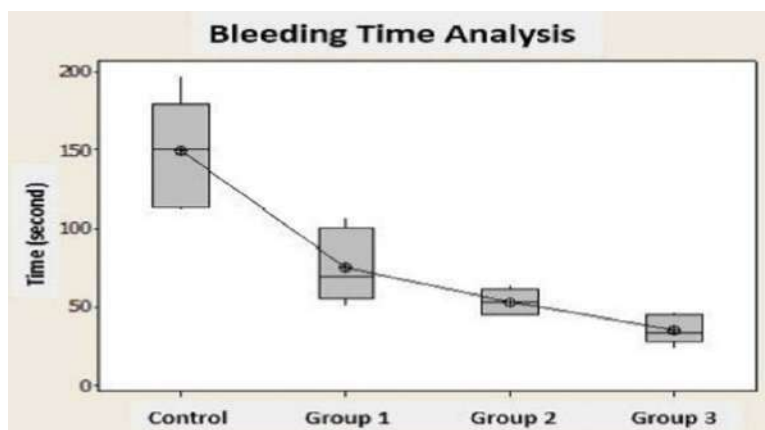


Figure 1. Examination of bleeding time by cutting the mice's tail.

C-type lectins are a subgroup of lectins, carbohydrate-recognizing proteins, which recognize and bind carbohydrates in a Ca^{2+} ion-dependent manner. They play a crucial role in hemostasis, a complex system to stop bleeding, which encompasses coagulation, platelet activation, and thrombus formation.¹⁹ The mechanism of lectin in accelerating the process of hemostasis is through some stages.²⁰ After an injury to a blood vessel, platelets

normally in a state of silence will turn into active and move towards an injured blood vessel to form a platelet plug at an initial phase. This phase is called platelet adhesion, which is the attachment of platelets to injured tissue.²¹⁻²³

Tannic acid is a commercial compound which is similar to the plant polyphenol tannin, that stops bleeding from mucous membrane via vasoconstriction.² Tanin is another active compound contained in Ambonese banana

stems playing a role in increasing ADP secretion in blood. ADP compound is a vasoactive chemical substance released by the body during vascular injury. ADPs and other chemicals, including serotonin, then trigger platelets to deform irregularly and become sticky with each other. Thus, one platelet and another will be easier to bind. An increase ADP levels in the blood then will result in high platelet aggregation so that platelet plugs as a mechanism for body hemostasis will soon be formed and will accelerate the bleeding time indicated in this research by the rapid cessation of blood coming out of the mice that were cut off.²⁴

Ambonese banana stem extract at the concentration of 100% is thought to have higher active compounds of tannin and lectin than concentrations of 25% and 50%. This high level of the active compounds, tannins and lectins, can increase platelet activity to adhesion and aggregate platelets when a vascular injury occurs. This is in accordance with the results of this research that showed

that the group of mice with Ambonese banana stem extract at the concentration of 100% had the shorten bleeding compared to the other groups.

In addition to accelerating the bleeding time, the administration of Ambonese banana stem extract at the concentrations of 25%, 50% and 100% can also accelerate the process of clotting time in mice (see figure 2). According to Ogle (2008), clotting time is an ability of blood to change from liquid to semi-solid. After the platelet plug is formed, the blood clotting cascade begins.⁷ This process runs through extrinsic and intrinsic blood clotting pathways involving 13 blood clotting factors. After the process, a prothrombin activator is formed which has a task of converting prothrombin to thrombin. Next, thrombin converts soluble fibrinogen into solid fibrin polymer threads. These fibrin threads will later become nets that will capture plasma, blood cells, and platelets to become a clot.^{9,25}

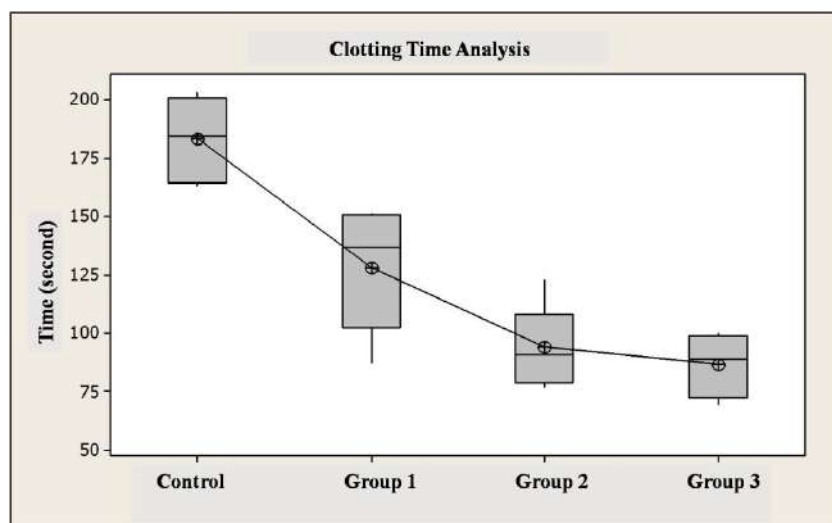


Figure 2. Examination of clotting time with modified Lee and White tube methods.

High platelet aggregation activities, furthermore, were found in the group with the administration of Ambonese banana stem extract since the effects of the active compounds, tannins and lectins, will be directly

proportional to the increased clotting time in mice. The solid fibrin threads formed then will attract more platelets carrying out more aggregation. As a result, the blood clotting process will occur faster. The results of this

research are the same as those reported by Bamidele, et al. (2010), examining the effects of methanol *ageratum conyzoides* leaf extract on hemostatic responses in mice. The research conducted by Bamidele, et al. (2010), finds that a decrease in bleeding time will compensate for a decrease in clotting time in mice.²⁶

Based on the previous hypothesis, the higher the concentration of Ambonese banana stem extract is, the higher the platelet aggregation activity occurs. If more platelets are attached to one another, then the fibrin threads that have formed in the initial phase of blood clotting will attract more thrombocytes, and blood clots will also occur faster. But, the decrease in clotting time in the group with Ambonese banana extract at the concentration of 100% was not significantly different from the group with Ambonese banana extract at the concentration of 50%. Even though at the concentration of 100% had the fastest clotting time compared to the other groups. However, based on the results of Duncan's distance test, Ambonese banana extract at the concentration of 50% did not differ significantly compared to that at the concentration of 100%.

This may occur due to the influence of blood clotting factors working on blood clotting cascade. Before the formation of fibrin threads which will later capture blood cells and plasma, blood clotting cascade occurs first, affected by various factors including 13 intrinsic and extrinsic blood clotting factors, such as fibrinogen, prothrombin, thromboplastin tissue, calcium, and vitamins K. In this study, unfortunately, there was no observation on the blood clotting factors. Hence, it is assumed that there was less significant difference between the group with the administration of Ambonese banana stem extract at the 100% and the group with that at the concentration of 50%. This finding may be influenced by the blood clotting factors.

Finally, based on the results, it can be said that the study hypothesis has been proven. The administration of Ambonese banana stem extract has an effect on accelerating the hemostasis response in mice characterized by an increase in both bleeding time and clotting time. The active compounds of tannins and lectins working synergistically in mice can lead to an effect on accelerating both bleeding time and clotting time.

Indonesia is a rich-tropical country which plants are used as an alternative medicines. However, deeper research needs to be done to prove the efficacy of drugs from plants in order to be used by the community. Although this research has proven the benefits of Ambonese banana stem sap as hemostatic, the platelet activity on the wound needs to be known to support this study.

The use of plant-derived medicines continues to be developed through biomolecular research so that they can be used as a standardized medicine. Ambonese banana stem has the potential to be developed into hemostatics through several studies that have been done before, and even has been patented for cancer therapy. Topical and hemostatic drugs are also capable for tooth extraction or scaling and root planning in gum disease that are needed for patients with blood disorders, such as hemophilia, which are still common in Indonesia, even in the world.

Conclusions

Ambonese banana stem extract (*Musa paradisiaca* var. *Sapientum*) have a potential as hemostatic agent by shortening bleeding and clotting time on mice.

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REFERENCES

1. Akolkar AR, Kulkarni DG, Gangwani KD, Shetty L, Channe SP, Sarve PH. Bleeding control measures during oral and maxillofacial surgical procedures: a systematic review. *J Dent Res Rev*. 2017;4(4):79-89.
2. Mani A, Anarthe R, Kale P, Maniyar S, Anuraga S. Hemostatic Agents in Dentistry. *Galore Int JHealth SciRes*. 2018;3(4):40-6.
3. Minno GD, Canaro M, Ironside JW, Navarro D, Perno CF, Tiede A, Gürtle L. Pathogen safety of long-term treatments for bleeding disorders: still relevant to current practice. *Haematol*. 2013;98(10):1495-8.
4. Verma DK and Rajan R. A case of thrombocytopenia due to odontogenic infection. *J Dent Res Dent Clin Dent Prospect*. 2011;5(4):144-7.
5. Van Ommen CH and Peters M. The bleeding child. Part I: primary hemostatic disorders. *Eur J Pediatr*. 2011;171(1):1-10.
6. Bryckaert M, Rosa JP, Denis CV, Lenting PJ. Of von willebrand factor and platelets. *Cell Mol Life Sci*. 2014;72(2):307-326.
7. Ogle OE, Swantek J, Kamoh A. Hemostatic agents. *Dent Clin North Am*. 2011;55:433-9.
8. Santhosh K. Local hemostatic agents in the management of bleeding in oral surgery. *Asian J Pharm Clin Res*. 2016;9(3):35-41.
9. Christian B. Risk of bleeding in oral surgery in patients with disorders of haemostasis. *Blood Transfus*. 2015;13:176-7.
10. Von Rintelen K, Arida E, Häuser C. A review of biodiversity-related issues and challenges in megadiverse Indonesia and other Southeast Asian countries. *Res Ide Outc*. 2017;3:e20860.
11. Sudirga, SK. 2000. Utilization of plants as traditional medicines in Trunyan village, Kintamani district, Bangli regency. *J Bumi Lestari Udayana*. 2000;12(5):7-18.
12. Coelho LC, Silva PM, Lima VL, Pontual EV, Paiva PM, Napoleao TH, et al. Lectins, interconnecting proteins with biotechnological/pharmacological and therapeutic applications. *Evid Based Complement Alternat Med*. 2017;1-22.
13. Budi HS, Soesilowati P, Imanina Z. Histopathological of macrophages and neovascular of tooth extraction wound healing on ambonese banana stem sap administration. *Maj Kedokt Gigi Ind*. 2017;3(3):121-27.
14. Prasetyo BF, I Wientarsih, Priosoeryanto BP. Activity and stability test of ambon banana stem extract gel (*Musa paradisiaca* var *sapientum*) in wound healing process in mice (*Mus musculus albinus*). *J Vet*. 2010;11(2):70-3
15. Herbert JM and Bernat A. Aprotinin reduces clopidogrel-induced prolongation of the bleeding time in the rat. *Thromb Res*. 1993;71:433-41.
16. Fareed J, Thethi I and Hoppensteadt D. Old versus new oral anticoagulants: focus on pharmacology. *Annu Rev Pharmacol Toxicol*. 2012;52:79-99.
17. Kattuo M, Issa R, Beitawi S. Commonly used herbal remedies for the treatment of primary dysmenorrhea and heavy menstrual bleeding by herbalists in Amman, Jordan: A cross-sectional survey. *Jordan J Pharm Sci*. 2020;13(4):467-83.
18. Priosoeryanto BP, Putriyanda N, Listyanti AR, Juniantito V, Wientarsih I, Prasetyo BF et al. The Effect of Ambonese banana stem sap (*Musa paradisiaca* forma *typica*) on the acceleration of wound healing process in mice (*Mus musculus albinus*). *J Agri Rural Dev Trop Subtrop*. 2007;4(2):35-49.
19. Swanson MD, Winter HC, Goldstein IJ, Markovitz DM. A lectin isolated from bananas is a potent inhibitor of hiv replication. *J Biol Chem* 2010;285(12):8646-55.
20. Johannes A. Eble. Structurally robust and functionally highly versatile C-type lectin (-related) proteins in snake venoms. *Toxins*. 2019;136:1-25
21. Inoue KS, Osada M, Ozaki Y. Physiologic and pathophysiologic roles of interaction between C-type lectin-like receptor 2 and podoplanin: partners fromin uteroto adulthood. *J Thromb Haemost*. 2017;15:219-29.
22. Stokes KY, Granger DN. Platelets: a critical link between

- inflammation and microvascular dysfunction. J Physiol 2012;590(5):1023-34.
23. Ghoshal K and Bhattacharyya M. Overview of platelet physiology: its hemostatic and nonhemostatic role in disease pathogenesis. Sci WorldJ. 2014;781857.
24. Periyah MH, Halim AS, Mat Saad AZ. Mechanism action of platelets and crucial blood coagulation pathways in hemostasis. Int J Hematol Oncol Stem Cell Res. 2017;11(4):319-27.
25. Gupta S. Selective activation of platelets by surfaces and soluble agonists. In: Ph.D. Thesis. The University of the Basque Country, Bilbao, Spain, 2014.
26. Kulkarni R. Alternative and topical approaches to treating the massively bleeding patient. Clinic Adv Hematol Oncol 2004;2(7):428-31.
27. Bamidele O, Akinnuga AM, Anyakudo MMC, Ojo OA, Ojo GB, Olorunfemi JO et al. Haemostatic effect of methanolic leaf extract of *Ageratum conyzoides* in albino rat. J of Med Plants Res 2010;4(20):2075-9.
28. Elayah E, Abu-rish E, Yousef A. Effect of dividing low dose aspirin on platelet reactivity and the correlation with patient-related factors. Jordan J Pharm Sci. 2020;13(2):185-95.

تحليل زمن النزف والتخثر لمستخلص ساق *Musa paradisiaca var. sapientum* (L.) Kunze على الاستجابة المرفقة

هندريك بودي، ويسنو جولياستوتي، يونيار سوليستيواتي*

*كلية طب الأسنان، جامعة إيرلانجا

ملخص

أحد الأدوية العشبية التي يمكن استخدامها لتسريع النزيف هو عصارة جذع الموز الأمبونية (*Musa paradisiaca var. Sapientum*) من خلال إفراز Adenosin Diphosphate (ADP) وعامل von Willebrand (vWF) في عملية التصاق الصفائح الدموية على الأوعية المصابة. تهدف هذه الدراسة إلى اختبار تأثير مستخلص جذع الموز الأمبوني بتركيز 25% و 50% و 100% استجابة مرقى على الفئران. تم تقسيم 24 دراسة حيوانية بشكل عشوائي إلى أربع مجموعات. المجموعة الأولى كانت المجموعة الضابطة، المجموعة الثانية أعطيت 25% تركيز المستخلص، المجموعة الثالثة أعطيت تركيز المستخلص 50% والمجموعة الرابعة أعطيت تركيز 100% من مستخلص جذع الموز الأمبوني عن طريق الفم. تم فحص جميع المجموعات وقت النزيف والتجلط. أدى إعطاء مستخلص جذع الموز الأمبوني بتركيزات 25% و 50% و 100% إلى تقصير وقت النزف ووقت التخثر بشكل كبير مقارنة بمجموعة التحكم ($p = 0.00$). أظهرت استنتاجات هذه الدراسة أن مستخلص جذع الموز الأمبوني لديه القدرة على أن يكون عامل مرقى عن طريق تقصير وقت النزف والتخثر على الفئران.

الكلمات الدالة: مستخلص جذع الموز الأمبوني، زمن النزف، زمن التخثر، مرقى.

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