Development of herbal bag for herbal bath during postnatal care from Temuan traditional knowledge

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Abstract. This study is about the development of the herbal bath bag during postpartum care. Herbal bathing important for mother because it can fresh mother’s body and gave a therapy during that period. Lack of knowledge about the herbal bathing is the main factor on why this study was conducted. The objectives for this study are to formulate the plant remedies to be used in the 2 in 1 mini pocket herbal bath bag production, to investigate the phytochemical that presence in the formulation and determine the presence of anti-bacterial properties in the plant extract based on the formulation and to develop 2 in 1 mini pocket herbal bath bag for traditional herbal bath based on traditional knowledge from the Temuan community in Taman Negara Gunung Ledang. This study focused on two different plant which are Annona muricata (durian belanda) and Psidium guajava (jambu batu). Both plants were used during postpartum care for herbal bathing by indigenous people. Moreover, the plant part used was the leaves. There are five formulation of the plant that a based on the Design Expert software. Basically, this study consists of two tests which are phytochemical screening and antibacterial testing. Phytochemical screening included terpenoids, steroid, flavonoid, quinone and alkaloids test. The result showed that formulation 3 has higher numbers of phytochemical presence which are flavonoid, terpenoid and quinone. In addition, formulation 3 also have higher number of means ± std for result anti-bacterial activity. As a conclusion, the three objectives were successful achieved and formulation 3 was chosen to develop the herbal bath bag.

1. Introduction
The postnatal period is period where the phase for the life of mother and new-born babies are in critical condition. The hardest time that mother and babies should face is started from the first day after the delivery of the babies. Based on the previous research, there is an issue where the mortality rate of the mother and new-born is increasing during this period around the worldwide. Lack of care during this time can cause the death to happen or disability to promote healthy and behavior of the mothers and babies [2]. Therefore, the postnatal care is importance for both mother and babies to ensure that the mother and baby are in good health. Mostly, mothers and babies spend most of the postnatal period at home. Therefore, the father should alert and concern about the health of the mother and babies. The postnatal care at home needed the nurse to come and visit mother’s house according to the standard schedule of postnatal care visit. The mother and babies should go to the health clinic for postnatal check-up and get family counseling planning when they reached at day 30.

However, However, postpartum care more focused on the mother only. It started after the mother
had been delivers their baby and mostly the period took about 6 to 8 weeks. The reason on why mothers should be taking care is because mothers faced changes such as emotional and physical. Beside mother, fathers also needed together learn how to care the baby. This may reduce mother’s stress and burden. During this period, mother need to rebuild their strength by having a plenty of rest and getting nutrition food. As we know, baby wakes up about every three hours because they need to be fed and changed for their comfort. Therefore, mother and father should always concern and change the routine to care the baby. Don’t let the mother to do all the things without any helping. Moreover, mother’s body has changed due to the pregnancy and delivers birth. The mother should get a healthy diet to recover from the changing. By this healthy and balanced diet, mothers can be able to be active and able to care for their baby. Food that mother should consumed are grain (whole wheat, brown rice, oatmeal), vegetables (peas, beans, dark green vegetables), fruits, dairy (fat free, low-fat product) and protein (fish, nuts). By doing exercises, mother also can burn calories.

Herbal bathing is one of the traditional practices during the postpartum period. It concerned with cleanliness and hygiene related to rituals of purification and separation. Herbal baths promote the people’s physical health and their psychological well-being [2] and according to references [3], herbal bath can improve people’s health in many ways such as relax tense muscles, open pores, encourage digestion, improve circulation, soften the skin, assist in natural detoxification, boost the immune system and promote restful sleep.

2. Materials and methods

2.1. Plant selection
For this study, it will focus on two different type of plant sample which are Annona muricata (durian belanda) and Psidium guajava (jambu batu) (Othman et al., 2017). The plant part that used for each sample was leaves. This selection of plants was due to the traditional knowledge that was practiced by Temuan Community in Taman Negara Gunung Ledang during their postnatal care. This 2 plant samples also have same function which are to treat flatulence and freshen body. Also, Temuan Community used this plant for bathing

2.2. Preparation of plant extracts
First of all, the Design Expert software was used to formulate the plant extraction. The software displayed that there were 5 formulations that include this 2-plant sample. Each formulation has different ratio of the plant sample. After formulations were determined, the leaves then were dried and grinded to make a powder form. Next, this study only consists of aqueous extraction where the solvent used was water. The method used for extraction was infusion method where 5g of sample infused in 100ml distilled water. The time required to infuse this extract was 40 minutes. Then the extract was filtered using Whatmann filter paper.

2.3. Phytochemical analysis
Phytochemical screening was performed to identify phytochemicals present in the aqueous extracts of plant leaves for each formulation. The phytochemicals were detected by color tests [10].

2.3.1. Test of alkaloids. 1 mL of the extract was prepared. Then 2 mL of Mayer’s reagent was added. Appearance of dull white precipitate indicated the presence of alkaloids [5]

2.3.2. Test of flavonoids. 1 mL of extract was prepared. Then 1 mL of Ferric Chloride was added. The formation of brown colour indicated the presence of flavonoids [5]

2.3.3. Test for steroids. The extracts were dissolved in 2 mL of chloroform to which 10 drops of acetic acid and five drops of concentrated sulphuric acid was added and mixed. The change of red colour through blue to green indicated the presence of steroids [5]
2.3.4. **Test for terpenoids.** 5 mL of extract was mixed in 2 mL of chloroform and 3 mL of concentrated sulphuric acid was added to form a layer. A reddish-brown precipitate of the interface indicated the presence of terpenoids [5]

2.3.5. **Test for quinone.** 1 mL of extract was prepared. A few drops of concentrated hydrochloric acid were added. A yellowish-brown colour indicated the presence of quinone [5]

2.4. **Antibacterial activity**
Antibacterial activity is the most common type of antimicrobial activity. In this study, the disk diffusion assay method was used to do the anti-bacterial activity and there were 2 different types of bacteria which are *Staphylococcus aureus* and *Pseudomonas aurigenosa*. The disk diffusion susceptibility method is simple and practical and has been well-standardized. The agar used for this method is Mueller-Hinton agar and it was poured in the petri dishes. The preparation of agar was done by mixing 19 g of Mueller- Hinton agar powder with 500 mL distilled water and the preparation for nutrient broth was done by mixing 0.39g of nutrient broth powder with 30 mL distilled water. Both solutions were autoclaved at 121 °C. The function of nutrient broth was to put the bacteria into the nutrient broth solution. The bacteria cultured by getting the first colony and then the bacteria were putting into the nutrient broth and incubated at 40 °C for 24 hours. For the paper disc, the extract for all formulation was dropped to the filter paper that acts as paper disc. Then, the paper disc was dried by waiting about a few hours before the paper disks placed on the inoculated agar surface. The agar plates that consist of bacteria and plant extract were incubated for 16–24 h at 35°C prior to determination of results. The zones of growth inhibition around each of the disks were measured to the nearest millimeter.

3. Result and discussion

3.1. **Formulation using design expert software**
Based on the Design Expert software, there were 5 formulations for this study which the formulation consists of *Annona muricata* and *Psidium guajava*. Table 1 showed the 5 formulation and its ratio that used in this study. The total weigh was 5g for each formulation.

| Formulation | Annona muricata | Psidium guajava | Ratio |
|-------------|----------------|----------------|-------|
| Formulation 1 | 0.00 g | 5.00 g | 0:1 |
| Formulation 2 | 5.00 g | 0.00 g | 1:0 |
| Formulation 3 | 1.25 g | 3.75 g | 1:3 |
| Formulation 4 | 3.75 g | 1.25 g | 3:1 |
| Formulation 5 | 2.50 g | 2.50 g | 1:1 |

The ratio for the formulation were 0 *Annona muricata*: 1 *Psidium guajava*, 1 *Annona muricata*: 0 *Psidium guajava*, 1 *Annona muricata*: 3 *Psidium guajava*, 3 *Annona muricata*: 1 *Psidium guajava* and 1 *Annona muricata*: 1 *Psidium guajava*.

3.2. **Phytochemical screening**
The phytochemical screening was done tested for all the formulation with same solvent which is water (aqueous extract). The tests were carried out to detect the presence of secondary metabolite such as alkaloid, flavonoid, steroid, terpenoid and quinone. The results for this testing were presented in the Table 2.
Based on the results, flavonoid, terpenoid and quinone presence in the formulation 1 and formulation 3, meanwhile formulation 5 only presence flavonoid and quinone where the terpenoid was absence. However, alkaloid and steroid were found to be absent in all the formulation. Also, formulation 2 and formulation 4 does not absent any phytochemical. The presence of the phytochemical compounds in these formulations enhances their pharmaceutical and therapeutic potentials [8].

The main role for phytochemical is to prevent diseases caused and the result of oxidative stress and release reactive oxygen species has single oxygen of various radicals as a damaging side effect of aerobic metabolism [10]. First of all, flavonoids are the important phytochemicals which are responsible for the free radical scavenging activity [9]. Besides, terpenoid act as antimicrobial, antifungal, antiviral, antihyperglycemic, anti-inflammatory, antioxidants, antiparasitic, immunomodulatory, and as skin permeation enhancer [7]. Lastly, Hydroquinone is a skin-lightening agent that can bleaches the skin, which can be helpful when treating different forms of hyperpigmentation such as acne scars, age spots, freckles, melasma and post-inflammatory marks from psoriasis and eczema.

3.3. Antibacterial activity

The bacterial strains used for the study were Staphylococcus aureus (Gram-positive bacteria) and Pseudomonas aeruginosa (Gram-negative bacteria). Bacterial cultures were obtained from Food Microbiology Laboratory and stored at 37°C. Both of the cultures were revived on nutrient broth and were given the required incubation conditions [9]. Petri dishes were prepared with 20 mL of sterile Muller- Hinton Agar and culture suspension containing bacteria was swabbed on solidified MHA medium and allowed to dry for 10 minutes [10]. Each bacterium was test against two different concentration of the extract (1g sample: 20ml of distilled water and 1g sample: 10ml of distilled water). There, there were 20 plate of petri dishes used in this study. Unfortunately, the bacteria S. aureus was contaminated because the incubator used also contaminated. The stock culture of S. aureus unsuccessfully saved and there was no testing for anti-bacterial activity against S. aureus. Meanwhile, bacteria P. aeruginosa that provided in the lab was from the cotton swab and fortunately, it can be cultures for the anti-bacterial activity. The results for antimicrobial activity of different concentration for plant extracts against bacteria are shown in Table 3.

Table 3 showed the result for both concentration of plant extract against bacteria Pseudomonas aeruginosa. The control used which is distilled water was maintained in each petri dish plates. The antimicrobial activity was evaluated by measuring the inhibition zone diameter in millimeters (mm) around the wells [10]. Based on the result obtained, the zones of inhibition range were from 10mm to 14mm. The results were presented in tables as mean ± standard deviation [4]. The data obtained were analyzed using Microsoft Excel 2010 where the mean and standard deviation were determined. The reason on why using this Microsoft Excel because the data obtained were so little data and it is not suitable for using analysis of variance (ANOVA). The result showed that formulation 3 with concentration 1g sample: 10ml distilled water has the highest number of means ± standard deviation which is 13.500 ± 0.5774 followed by formulation 5 with same concentration (11.500 ± 0.5774).
However, there were negative results in formulation 2 where the zone of inhibition is zero. Both concentration of the extract does not have anti-bacterial activity.

Table 3. The comparison of diameter zone inhibition between different plant extraction concentrations for *Pseudomonas aeruginosa* with distilled water as control

| Concentration of plant extract | Formulation 1 Zone of inhibition | Formulation 2 Zone of inhibition | Formulation 3 Zone of inhibition | Formulation 4 Zone of inhibition | Formulation 5 Zone of inhibition |
|--------------------------------|----------------------------------|----------------------------------|----------------------------------|----------------------------------|----------------------------------|
| 1g sample: 20ml distilled water| Replicate 1 14.00                | -                                | 12.00                            | 10.00                            | 10.00                            |
|                                 | Replicate 2 14.00                | -                                | 12.00                            | 10.00                            | 11.00                            |
|                                 | Replicate 3 -                    | -                                | 13.00                            | -                                | 11.00                            |
|                                 | Replicate 4 -                    | -                                | -                                | -                                | 12.00                            |
| Control                         | -                                | -                                | -                                | -                                | -                                |
| Mean ± std                      | 7.00 ± 8.083                     | 0.00 ± 0.00                      | 9.25 ± 6.185                     | 5.00 ± 5.774                     | 11.00 ± 0.817                    |

| 1g sample: 10ml distilled water | Replicate 1 12.00                | -                                | 14.00                            | -                                | 11.00                            |
|                                 | Replicate 2 12.00                | -                                | 13.00                            | -                                | 11.00                            |
|                                 | Replicate 3 12.00                | -                                | 14.00                            | -                                | 12.00                            |
|                                 | Replicate 4 -                    | -                                | 13.00                            | -                                | 12.00                            |
| Control                         | -                                | -                                | -                                | -                                | -                                |
| Mean ± std                      | 9.00 ± 6.00                      | 0.00 ± 0.00                      | 13.50 ± 0.577                    | 0.00 ± 0.00                      | 11.50 ± 0.577                    |

*Pseudomonas aeruginosa* are the bacteria that have a wide physiological potential that allow them to populate diverse environments and lead to severe infections of humans such as septicemia, leg ulcers, and burn wounds [1]. *Pseudomonas aeruginosa* can alter repair processes, leading to chronic wounds and infections [7]. Also, it infects organs (skin, ear, eye, heart, blood, soft tissue, and bone), joints and respiratory, urinary, gastrointestinal and central nervous systems. However, epithelial barriers direct contact with the external environment, have increased probability of infection [7]. Moreover, there is a clinical *P. aeruginosa* strains resistant to all available antibiotics in clinical use [7].

### 4. Conclusion

As a conclusion, there were five formulations that used for this study. Each of the formulation consists of different ratio. These five formulations were used in further analysis and testing which are phytochemical screenings and anti-bacterial testing. The result showed that formulation 3 was the best formulation compared to other formulation. This is because, formulation 3 consist of phytochemical flavonoids, quinone and terpenoid. Also, formulation 3 with the extract 1g sample: 10ml of distilled water has the higher result for antibacterial activity where the mean ± std was 13.500 ± 0.5774. Therefore, the formulation 3 was chosen to develop the herbal bath bag for postpartum care. Overall, the three objectives for this study were successfully achieved.

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### References

[1] A Buivydas, T Pasanen, A Senčilo, R Daugelavičius, M Vaara, and D. H. Bamford 2013 “Clinical isolates of *Pseudomonas aeruginosa* from superficial skin infections have
different physiological patterns,” FEMS Microbiology Letters 343 2 183–189
[2] Charlotte I E A Van, T Klooster, V Haabo, S Ruyschaert, T Vossen and T R V Andel 2018 “Herbal bathing: an analysis of variation in plant use among Saramaccan and Aucan Maroons in Suriname,” Journal of Ethnobiology and Ethnomedicine 14 1
[3] E McIntosh and E M I E McIntosh 2019 “Self-Care Secrets: How to Create an Herbal Bath,” LearningHerbs, 12-Aug-2019. [Online]. Available: https://learningherbs.com/remedies-recipes/herbal-bath/
[4] E Obianuju, O Pamela, I Eghosa, U Princewill and A. Bond 2017 “Effects of Aqueous Leaf Extract of Annona muricata on Pregnancy and Pregnancy Outcome,” Journal of Advances in Medical and Pharmaceutical Sciences 12 1 1–6
[5] G V and K D 2014 “Preliminary Phytochemical Analysis of Leaf Powder Extracts of Psidium guajava L.” International Journal of Pharmacognosy and Phytochemical Research
[6] M Ruffin and E Brochiero 2019 “Repair Process Impairment by Pseudomonas aeruginosa in Epithelial Tissues: Major Features and Potential Therapeutic Avenues,” Frontiers in Cellular and Infection Microbiology 9
[7] O Rashidi, H S F A Abdul, H F A Mohd and J M Aizat 2017 “Carotenoid Content and Composition in 20 Medicinal Plant Species of Traditional Malay Midwifery Postnatal Bath,” Journal of Pharmacy and Nutrition Sciences 7 4 193-196
[8] P P Brahmkshatriya and P S Brahmkshatriya 2013 “Terpenes: Chemistry, Biological Role, and Therapeutic Applications,” Natural Products 2665–2691
[9] S A, K P and R G “Phytochemical Screening and Antimicrobial Assay of Various Seeds Extract of Cucurbitaceae Family,” International Journal of Applied Biology and Pharmaceutical Technology 3 3 ISSN: 0976-4550
[10] S E Priya and R Ravindhran 2015 “Phytochemical Analysis and Antimicrobial Properties of Extracts from Aerial Parts of Phyla nodiflora (L) Greene,” International Journal of Current Microbiology and Applied Science, ISSN: 2319-7706 4 2 347- 358
[11] T T, A R, V D and D R 2015 “Preliminary Phytochemical Screening Of Different Solvent Mediated Medicinal Plant Extracts Evaluated,” International Research Journal of Pharmacy, 4 6 246–248