

Comparison of Saponin Levels of Lerak Extract (*Sapindus rarak*) Maceration and Soxhletation Results Based on UV-Vis Spectrophotometry Analysis

Umi Fitria^{1*}, Sulisetijono Sulisetijono¹, Mardiana Lelitawati¹, Muh. Wahyudi Jasman¹, Zahra Firdaus¹, and Achmad Muktafi¹

¹Department of Biology, Faculty of Mathematics and Science, Universitas Negeri Malang, Semarang street 5, Malang City, 65145, Indonesia.

Abstract. The potential of Lerak fruit (*Sapindus rarak*) as a basic ingredient for medicine has been recognized because of its content, which is rich in saponin compounds. Saponin compounds are known to have antimicrobial effects, the ability to inhibit fungal growth, antidiabetic properties, and anti-inflammatory properties. To maximize the content of active compounds, optimization is required, including variations in extraction methods. This study aims to compare the levels of saponins produced by the maceration and Soxhletation extraction methods. Saponin levels were tested using the UV-Vis spectrophotometry method with standard Diosgenin. Data were analyzed using independent t-test statistics. The yield of the maceration and Soxhletation extraction methods was $70.59 \pm 0.505\%$ and $84.71 \pm 1.956\%$ respectively. The results of saponin levels obtained from the maceration and Soxhletation extracts were $1.395 \pm 0.005 \mu\text{gde/ml}$ and $1.904 \pm 0.015 \mu\text{gde/ml}$ respectively. The statistical test results showed a significant value of 0.000, which is less than 0.05, with a 95% confidence level. The conclusion of this study was that there were significant differences in saponin levels in the macerated and soxhleted lerak extract, with the highest levels found in the Soxhletation method with levels of $1.904 \pm 0.015 \mu\text{gde/ml}$.

1 Introduction

The Sapindaceae family is one of the main sources of saponins that are widespread in all tropical and subtropical regions of Asia [1]. The most common Sapindaceae plant found in Indonesia is *Sapindus rarak*. Lerak fruit is a family of Sapindaceae which has the Latin name *Sapindus rarak* [2]. *Sapindus rarak* fruit contains allelochemical compounds in the form of saponins and other active substances such as alkaloids, flavonoids, polyphenols, and tannins [3]. Saponins are hydrophobic and hydrophilic so they can interact with water and fat at once [4]. Lerak fruit has a high percentage of saponin content which is 12% [5].

* Corresponding author: umi.fitriyati.fmipa@um.ac.id

The high content of saponins makes lerak fruit widely used in various sectors. In the medical and pharmaceutical fields, saponin compounds are used in the treatment of certain diseases, such as diabetes, high blood pressure, cardiovascular problems, and high cholesterol [6]. Saponins function as antibacterial, antifungal, antioxidant, and anti-inflammatory substances [7]. In the environmental field, saponins are known as secondary metabolite compounds included in triterpenoid glycosides or steroidal glycosides that form foam or foam so that they can be used as natural detergents, insecticides and pesticides [8][9]. Previous research has shown that saponins have the potential to replace synthetic cleaning chemicals [10], able to dissolve synthetic dyes [11] and polycyclic aromatic hydrocarbons [12] in aqueous solution. Saponin compounds contained in lerak fruit can be obtained through extraction.

The maceration method is an extraction technique that involves immersing the material in a solvent corresponding to the active compound to be extracted. This process is carried out with low heating or even no heating at all. Some of the factors that affect the extraction process include time, temperature, solvent type, comparison between material and solvent, as well as material particle size [13]. The advantage of the maceration method is that it can prevent degradation of compounds because it does not apply extraction conditions that are at high temperatures and pressures [14] so that the active substance to be taken remains intact [15]. During the soaking process, there is a breakdown of the cell wall and cell membrane due to the pressure difference between outside and inside the cell, this makes secondary metabolites in the cytoplasm be released and dissolved in the organic solvent used [16]. The socletation extraction method is a method of separating substances from their mixtures through a heating process, the socletation method provides more extract results than the maceration method [17]. The principle of socletation is repeated filtering so that the results obtained are perfect and the solvent used is relatively small [18].

The choice of maceration and socletation extraction methods because it has many advantages compared to other extraction methods. The main advantage of maceration extraction method is that the procedure and equipment used are simple and not heated so that natural materials do not become decomposed [19]. Cold extraction allows many compounds to be extracted, although some compounds have limited solubility in solvents at room temperature [20]. While the socletation method is a hot method that can produce more extracts, less solvent is used (material efficiency), the time used is faster, and the sample is extracted perfectly because it is done repeatedly [21]. In addition, biological activity is not lost when heated so this technique can be used in the search for drug brood.

The principle of socletation is repeated filtering so that the results obtained are perfect and the solvent used is relatively small [22]. Methanol p.a is the best solvent that produces the highest total extraction of saponins from lerak fruit. Saponin extract will be produced more if extracted using methanol because saponins are polar so they will dissolve more easily than other solvents [9]. Determination of saponin levels is carried out by the UV-Vis spectrophotometry method. UV-Vis spectrophotometry analysis has been recognized as the main method for identification, characterization, purity checking and determination. The advantage of the UV-Vis spectrophotometry method as a method of determining levels is that it can be used for the analysis of substances in small, fast, simple, specific and sensitive amounts or levels.

2 Experimental details

Comparative study of saponin levels of lerak extract (*Sapindus rarak*) maceration and socletation results based on UV-Vis spectrophotometry analysis and is an experimental-based study.

2.1 Materials

Tools and materials that used to facilitate this research are analytical balances, vessels for maceration, parchment, soxhlet, round table bath, rotary evaporator, waterbath, steam cup, test tube, test tube rack, oven, stirring rod, beaker glass, 4 spectrophotometer UV-Vis. The ingredients used are lerak fruit, methanol p.a, diosgenin, vanillin, H₂SO₄ 72%, HCl 2N, aquadest, chloroform, ethanol.

2.2 Research Phase

The research stage is as follows.

1. The determination of *Sapindus rarak* plants was carried out at the Microbiology Laboratory of State University of Malang, East Java.
2. Making simplisia powder, then extracted using maceration and socleation extraction methods, then concentrated using a rotary evaporator.
3. Preliminary test for the presence of saponin compounds, using foam test methods and color tests.
4. Determination of saponin levels using UVVis spectrophotometry.

3 Results and discussion

The results of the determination showed that the samples used in this study accurately showed (*Sapindus rarak*) namely with the genus *Sapindus* and the species *Sapindus rarak*. The results of viscous extracts obtained by maceration and socleation methods were 17.6523 grams and 21.1848 grams, respectively. The organoleptic results of lerak extract can be seen in Table 1.

Table 1. Organoleptic Lerak Fruit Extract

No	Organoleptik	Information	
		Maceration Results	Socleation Results
1.	Texture	Viscous liquid	Viscous liquid
2.	Color	Light brown	Dark brown
3.	Construction	Special fruit <i>Sapindus rarak</i>	Special fruit <i>Sapindus rarak</i>

The yield value of thick extract of lerak fruit (*Sapindus rarak*) obtained from maceration and socleation extraction results can be seen in Table 2, Table 3 and Table 4.

Table 2. Maceration Extract Yield

No	Weight of extracted Simplisia Powder (grams)	Maceration Extract Weight (grams)	Yield Value (%)
1.	25.0061	17.7345	70.92
2.	25.0043	17.5702	70.26
Average		17.6523	70.59±0.505

Table 3. Socletation Extract Yield

No	Extracted Simplisia Powder Weights (grams)	Socletation Extract Weight (grams)	Yield Value (%)
1.	25.0072	21.7561	86.99
2.	25.0055	20.6134	82.43
Average		21.1848	84.71±1.956



Fig. 1. Viscous Extract Foam Test Results Maceration and Socleation Results (a) Maceration (b) Socleation



Fig. 2. Color Test Results of Lerak Fruit Extract Maceration and Socleation Results (a) Maceration (b) Socleation

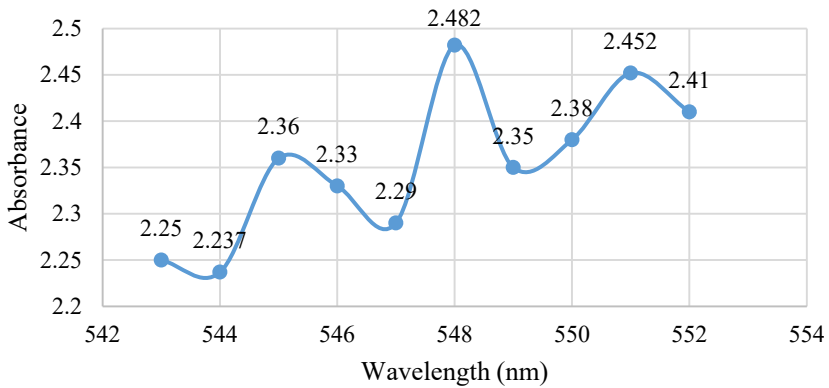


Fig. 3. Maximum Wavelength Curve Result

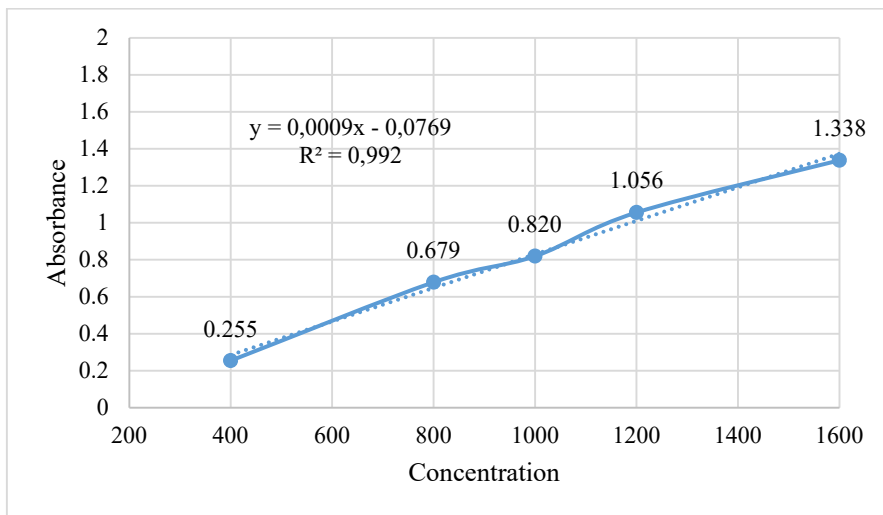


Fig. 4. Diosgenin Standard Curve Results

4 Discussion

The results of organoleptic tests on lerak fruit extract (*Sapindus rarak*) are listed in Fig. 1. The organoleptic test is seen from the color and foam produced from both extraction methods, namely maceration and socleation. Both extraction methods produce a brown color, which is the original color of lerak fruit (*Sapindus rarak*) [23]. In addition to testing the color and foam, the texture of lerak fruit extract (*Sapindus rarak*) obtained is a slightly viscous liquid. The resulting smell is the characteristic smell of lerak fruit. Both methods have similarities from the extraction results.

Based on the extraction results on lerak fruit (*Sapindus rarak*), the maceration extraction method acquires a light brown color. While the socleation method gets a dark brown color. The yield yield of maceration and socleation extraction methods was $70.59 \pm 0.505\%$ and $84.71 \pm 1.956\%$ respectively. It can be concluded that the use of the socleation extraction method produces a greater yield than using the maceration method [24]. Temperature treatment can increase the solvent's capacity to extract insoluble compounds at room temperature [25]. High temperatures affect the continuous circulation of solvents when interacting with simplisia, thereby maximizing compound extraction and increasing yield yield. Heat treatment has an important role in the extraction process [26]. When the temperature is increased, the solvent has a better ability to extract compounds that at room temperature may be difficult or insoluble [27]. This relates to changing the physicochemical properties of compounds and solvents at higher temperatures, allowing for more efficient interactions between them [28]. The size of the yield indicates the effectiveness of the extraction process [29]. The effectiveness of extraction is influenced by solvent type, temperature, extraction time, particle size simplisia, temperature, and extraction method [30].

The results of organoleptic tests on foams, known in samples with maceration and socleation extraction respectively in Fig. 2 and Fig. 3, obtained heights of 6.3 cm and 6.8 cm. The foam is allowed to stand for 30 seconds, so that it appears that the maceration extraction sample drops to 6.2 cm while the socleation extraction sample remains 6.8 cm. As shown in Fig. 4, the results of this foam can be concluded the content of saponins in both samples.

Table 4. Results of Saponin Levels in Extract

Lerak Fruit Thick Extract (<i>Sapindus rarak</i>)	Saponin Content (µg/mL)	Average of Saponin Content (µgDE/mL)
Maceration Extract	1.389	1.395±0.005
	1.401	
	1.389	
	1.401	
	1.389	
	1.401	
Socletation Extract	1.921	1.904±0.015
	1.887	
	1.921	
	1.887	
	1.921	
	1.887	

The results of saponin levels obtained from the extract using the uv-vis specophotometry test, maceration and socletation results were 1.395±0.005 µgde/ml and 1.904±0.015 µgde/ml, respectively. The results of statistical tests show a sig value of 0.000 smaller than 0.05 with a confidence level of 95%. Socletation is more efficient in extracting certain compounds because heating the solvent can increase the solubility of the target compound [31]. The temperature regulation at socletation allows the solvent to reflux through the extracted material [32]. Socletation requires a shorter time compared to maceration due to its heating properties. While the maceration process is carried out at room temperature or relatively low temperature [33].

5 Conclusion

The results of saponin levels obtained from extracts by maceration method and socletation method were 1.395±0.005 µgde/ml and 1.904±0.015 µgde/ml, respectively. The results of the statistical test showed a significant value of 0.000, lower than 0.05, with a confidence level of 95%. The conclusion of this study is that there is a significant difference in the level of saponins in macerated and extracted lerak extract by the Socletation method, with the highest level found in the Socletation method of 1.904±0.015 µgde / ml.

The author would like to thank all related parties, supervisors, all of the assistant and especially to State University of Malang. This research is an Internal PNBPN project of State University of Malang based on the rector's decree No. 5.4.1169/UN32.20.1/PM/2023.

References

1. R. Li, Z. L. Wu, Y. J. Wang, and L. L. Li, "Separation of total saponins from the pericarp of *Sapindus mukorossi* Gaerten. by foam fractionation," *Ind Crops Prod*, vol. 51, pp. 163–170, Nov. 2013, doi: 10.1016/j.indcrop.2013.08.079.
2. G. 'Tjitrosoepomo, *Morfologi Tumbuhan*. Yogyakarta: Gajah Mada University Press, 1994.
3. Y. Syahroni and D. Prijono, "Aktivitas insektisida ekstrak buah *Piper aduncum* L. (*Piperaceae*) dan *Sapindus rarak* DC. (*Sapindaceae*) serta campurannya terhadap larva *Crocidolomia pavonana* (F.) (*Lepidoptera*: *Crambidae*)," *J Entomol Indones*, vol. 10, no. 1, pp. 39–50, Apr. 2013, doi: 10.5994/jei.10.1.39.

4. I. Arslan and A. Çelik, "Saponin Rich Fractions (SRPs) from Soapwort Show Antioxidant and Hemolytic Activity," *APCBEE Procedia*, vol. 7, pp. 103–108, 2013, doi: 10.1016/j.apcbee.2013.08.019.
5. V. E. Tyler, L. R. Brady, and J. E. Robbers, *Pharmacognosy*. USA: Lea & Febiger, 1989.
6. C. Y. Cheok, H. A. K. Salman, and R. Sulaiman, "Extraction and quantification of saponins: A review," *Food Research International*, vol. 59, pp. 16–40, May 2014, doi: 10.1016/j.foodres.2014.01.057.
7. A. C. Keller, J. Ma, A. Kavalier, K. He, A.-M. B. Brillantes, and E. J. Kennelly, "Saponins from the traditional medicinal plant *Momordica charantia* stimulate insulin secretion in vitro," *Phytomedicine*, vol. 19, no. 1, pp. 32–37, Dec. 2011, doi: 10.1016/j.phymed.2011.06.019.
8. S. Arum and T. Wikaningrum, "Study of Lerak (*Sapindus Rarak*) Biochar Application for Andosol Agricultural Soil Remediation," *IOP Conf Ser Earth Environ Sci*, vol. 1065, no. 1, p. 012020, Jul. 2022, doi: 10.1088/1755-1315/1065/1/012020.
9. J. B. Harborne, *Phytochemical Methods*. London: Academic Press, 1987.
10. A. Nafiunisa, N. Aryanti, and D. H. Wardhani, "Kinetic study of saponin extraction from *sapindus rarak* DC by ultrasound-assisted extraction methods," *Bulletin of Chemical Reaction Engineering & Catalysis*, vol. 14, no. 2, pp. 468–477, 2019.
11. K. Samal, C. Das, and K. Mohanty, "Eco-friendly biosurfactant saponin for the solubilization of cationic and anionic dyes in aqueous system," *Dyes and Pigments*, vol. 140, pp. 100–108, May 2017, doi: 10.1016/j.dyepig.2017.01.031.
12. T. Kariyawasam, P. D. Prenzler, J. A. Howitt, and G. S. Doran, "Eucalyptus saponin- and sophorolipid-mediated desorption of polycyclic aromatic hydrocarbons from contaminated soil and sediment," *Environmental Science and Pollution Research*, vol. 30, no. 8, pp. 21638–21653, Oct. 2022, doi: 10.1007/s11356-022-23562-z.
13. S. Khalid *et al.*, "Extraction and Quantification of *Moringa oleifera* Leaf Powder Extracts by HPLC and FTIR," *Food Anal Methods*, vol. 16, no. 4, pp. 787–797, 2023, doi: 10.1007/s12161-023-02470-z.
14. G. Nunes Mattos, M. C. Pessanha de Araújo Santiago, A. C. Sampaio Doria Chaves, A. Rosenthal, R. Valeriano Tonon, and L. M. Correa Cabral, "Anthocyanin Extraction from Jaboticaba Skin (*Myrciaria cauliflora* Berg.) Using Conventional and Non-Conventional Methods," *Foods*, vol. 11, no. 6, p. 885, Mar. 2022, doi: 10.3390/foods11060885.
15. E. Pratiwi, "Perbandingan Metode Maserasi, Remaserasi, Perkolasi Dan Reperkolasi Dalam Ekstraksi Senyawa Aktif Andrographolide Dari Tanaman Sambiloto (*Andrographis paniculata* Nee)," Institut Pertanian Bogor, Bogor, 2010.
16. K. Ngibad, "Phytochemical Screening of Sunflower Leaf (*Helianthus annuus*) and Anting-Anting (*Acalypha indica* Linn) Plant Ethanol Extract," *Borneo Journal of Pharmacy*, vol. 2, no. 1, pp. 24–30, May 2019, doi: 10.33084/bjop.v2i1.689.
17. R. S. Irianty and S. R. Yenti, "Pengaruh perbandingan pelarut etanol-air terhadap kadar tanin pada sokletasi daun gambir (*Uncaria gambir* Roxb)," *Sagu*, vol. 13, no. 1, pp. 1–7, 2014.
18. R. Riniati, A. Sularasa, and A. D. Febrianto, "Ekstraksi Kembang sepatu (*Hibiscus Rosa Sinensis* L) Menggunakan Pelarut Metanol dengan Metode Sokletasi untuk Indikator Titrasi Asam Basa," *IJCA (Indonesian Journal of Chemical Analysis)*, vol. 2, no. 01, Mar. 2019, doi: 10.20885/ijca.vol2.iss1.art5.

19. H. Nurhasnawati, S. Sukarmi, and F. Handayani, "Perbandingan Metode Ekstraksi Maserasi dan Sokletasi Terhadap Aktivitas Antioksidan Ekstrak Etanol Daun Jambu Bol (*Syzygium malaccense* L.)," *Jurnal Ilmiah Manuntung*, vol. 3, no. 1, pp. 91–95, Jun. 2017, doi: 10.51352/jim.v3i1.96.
20. A. Teresa, "Effectiveness of Garlic Bulb (*Allium Sativum* L.) Ethanol Extract Cream on the Number of Fibroblast Cells in the Healing Process of Male White Rat (*Rattus norvegicus* L.) Wistar Strain Incision Wound," *NeuroQuantology*, vol. 18, no. 8, pp. 10–18, Aug. 2020, doi: 10.14704/nq.2020.18.8.NQ20199.
21. E. V. Arsita, D. E. Saragih, and K. Aldrin, "Anticancer potential from ethanol extract of *Zanthoxylum acanthopodium* DC. seed to against MCF-7 cell line," *IOP Conf Ser Earth Environ Sci*, vol. 293, no. 1, p. 012016, Jun. 2019, doi: 10.1088/1755-1315/293/1/012016.
22. P. Yuliana, "Ekstraksi Senyawa Tanin dan Saponin Dari Tanaman Serta Efeknya Terhadap Fermentasi Rumen dan Metanogenesis in Vitro," Institut Pertanian Bogor, Bogor, 2014.
23. R. Singh and B. Sharma, *Biotechnological Advances, Phytochemical Analysis and Ethnomedical Implications of Sapindus species*. Singapore: Springer Singapore, 2019. doi: 10.1007/978-981-32-9189-8.
24. H. Riasari, S. N. Fitriansyah, R. Hartati, K. Anggadiredja, and S. Sukrasno S, "Comparison of Extraction Methods, Antioxidant Activities, Total Phenol in Seeds and Seed Shells of Kabau (*Archidendron bubalinum* (Jack) I.C. Nielsen) From Lampung and South Sumatra," *Pharmacognosy Journal*, vol. 11, no. 6, pp. 1278–1284, Oct. 2019, doi: 10.5530/pj.2019.11.198.
25. A. Mokrani and K. Madani, "Effect of solvent, time and temperature on the extraction of phenolic compounds and antioxidant capacity of peach (*Prunus persica* L.) fruit," *Sep Purif Technol*, vol. 162, pp. 68–76, Apr. 2016, doi: 10.1016/j.seppur.2016.01.043.
26. F. Chemat *et al.*, "A review of sustainable and intensified techniques for extraction of food and natural products," *Green Chemistry*, vol. 22, no. 8, pp. 2325–2353, 2020, doi: 10.1039/C9GC03878G.
27. J. Shi, J. Yu, J. Pohorly, J. C. Young, M. Bryan, and Y. Wu, "Optimization of the extraction of polyphenols from grape seed meal by aqueous ethanol solution," *J. Food Agric. Environ*, vol. 1, no. 2, pp. 42–47, 2003.
28. B. E. Richter, B. A. Jones, J. L. Ezzell, N. L. Porter, N. Avdalovic, and C. Pohl, "Accelerated Solvent Extraction: A Technique for Sample Preparation," *Anal Chem*, vol. 68, no. 6, pp. 1033–1039, Jan. 1996, doi: 10.1021/ac9508199.
29. D. Y. Ali, Harijono, V. Fathuroya, S. D. Wijayanti, and H. F. Razi, "Effect of extraction method and solvent ratio on antioxidant activity of Dayak onion extract," *IOP Conf Ser Earth Environ Sci*, vol. 475, no. 1, p. 012024, Apr. 2020, doi: 10.1088/1755-1315/475/1/012024.
30. E. Brglez Mojzer, M. Knez Hrnčič, M. Škerget, Ž. Knez, and U. Bren, "Polyphenols: Extraction Methods, Antioxidative Action, Bioavailability and Anticarcinogenic Effects," *Molecules*, vol. 21, no. 7, p. 901, Jul. 2016, doi: 10.3390/molecules21070901.
31. O. R. Alara, N. H. Abdurahman, and C. I. Ukaegbu, "Extraction of phenolic compounds: A review," *Curr Res Food Sci*, vol. 4, pp. 200–214, 2021, doi: 10.1016/j.crfs.2021.03.011.
32. L. Shi, W. Zhao, Z. Yang, V. Subbiah, and H. A. R. Suleria, "Extraction and characterization of phenolic compounds and their potential antioxidant activities,"

- Environmental Science and Pollution Research*, vol. 29, no. 54, pp. 81112–81129, Nov. 2022, doi: 10.1007/s11356-022-23337-6.
33. I. Fajriati, A. Y. Ikhsani, A. Monitasari, M. Zamhari, B. Kartika, and J. R. Subba, “The Effect of Extraction Method on the Extract Yield in the Carotenoid Pigment Encapsulation for Halal Natural Pigment,” *Indonesian Journal of Halal Research*, vol. 4, no. 2, pp. 97–106, Aug. 2022, doi: 10.15575/ijhar.v4i2.17188.