



Polyphenol content and antioxidant activity of nipa palm (*Nypa fruticans* Wurmb.) fruit husk at different extraction temperatures

Sabri Sudirman^{1,*} , Bernica Toyba¹, Herpandi Herpandi¹, Indah Widiastuti¹, Puspa Ayu Pitayati¹, Miftahul Janna¹ and Rahmadiana Utami¹

1, Fisheries Product Technology, Faculty of Agriculture, Universitas Sriwijaya, Indralaya - 30662, South Sumatra, Indonesia

ARTICLE INFO

Original Article

Article history:

Received 9 February 2025

Revised 2 July 2025

Accepted 7 July 2025

DOI: 10.22077/jhpr.2025.8912.1474

P-ISSN: 2588-4883

E-ISSN: 2588-6169

*Corresponding author:

Fisheries Product Technology, Faculty of Agriculture, Universitas Sriwijaya, Indralaya - 30662, South Sumatra, Indonesia

Email: sabrisudirman@unsri.ac.id

© This article is open access and licensed under the terms of the Creative Commons Attribution License <http://creativecommons.org/licenses/by/4.0/> which permits unrestricted, use, distribution and reproduction in any medium, or format for any purpose, even commercially provided the work is properly cited.

A B S T R A C T

Purpose: Oxidative stress is a condition caused by an imbalance between free radicals and the body's antioxidant defense system. Antioxidants are substances that reduce or inhibit the formation of free radicals. This study aimed to evaluate the effects of different extraction temperatures on the polyphenol content of nipa palm (*Nypa fruticans*) fruit husk and to determine its antioxidant activity. **Research method:** The nipa palm fruit husk was extracted using different temperatures (30°C, 45°C, and 60°C) and the antioxidant activity was determined using the 2,2-diphenyl-1-picrylhydrazyl method. The data were presented as mean \pm standard deviation and analyzed by one-way analysis of variance (one-way ANOVA) followed by Duncan multiple comparison tests. **Findings:** The results showed that an extraction temperature of 60°C produced the highest total phenolic (44.50 mg Gallic acid equivalent per g of sample), flavonoid (8.75 mg Quercetin equivalent per g of sample), and tannin (50.30 mg Tannic acid equivalent per g of sample). Fourier transform infrared analysis detected hydroxyl groups, indicating the presence of polyphenol compounds in the nipa palm fruit husk extract. Furthermore, the extraction temperature of 60°C exhibited the highest radical scavenging activity (52.79%) compared to other temperatures (30°C and 45°C). These findings suggest that nipa palm fruit husk extract can serve as a natural antioxidant and has potential applications as a pharmaceutical agent. **Research limitations:** There were no limitations identified. **Originality/Value:** This study is the first to report the effect of temperature on the extraction of polyphenol content from nipa fruit husks. It also highlights the potential of nipa fruit husk extract as a natural source of antioxidant agents.

Keywords:Antioxidant, Fruit husk, *Nypa fruticans*, Tannin

INTRODUCTION

A free radical is a molecule that is highly reactive with other molecules due to an unpaired electron in the outer orbital. These molecules can be produced through various endogenous processes, such as mitochondrial respiration and immune cell activation, as well as from exogenous sources like radiation, pollution, and smoking (Chandimal et al., 2025). When the free radicals are higher than potential antioxidants in the body, this condition is known as oxidative stress (Rad et al., 2020). In this condition, the body needs to take antioxidants from outside the body, also known as exogenous antioxidants. An antioxidant is a substance that can reduce or inhibit the free radical formation. It can be obtained from natural sources, such as dietary supplements or functional foods (Mukherjee et al., 2024). Polyphenol compounds, vitamins, and polysaccharides are widely recognized as sources of antioxidants that can be extracted from natural resources, particularly plants, where they predominantly accumulate in the leaves, flowers, fruits, and seeds (Lang et al., 2024; Wilczyńska & Žak, 2024).

Polyphenol compounds are a type of secondary metabolite that is generally found in plants. Polyphenol substances include phenolic acids, flavonoids, tannins, stilbenes, and lignans (Lang et al., 2024). Polyphenol groups are known as antioxidant agents because they inhibit free radical formation. It can act as a radical scavenger by donating hydrogen atoms or electron transfer mechanisms (Andrés et al., 2023). Polyphenols can be extracted from terrestrial and aquatic plants, such as water lettuce (*Pistia stratiotes*) (Herpandi et al., 2021; Sudirman et al., 2022) and *Limnocharis flava* (Sudirman et al., 2024a). The nipa palm (*Nypa fruticans*) is an aquatic plant found in mangrove areas and can serve as a source of bioactive components. Previous research has reported that palm leaf and fruit extracts contain bioactive compounds, including polyphenols, which have antioxidant activity (Astuti et al., 2020; Hermanto et al., 2020). More recent research by Sudirman et al. also reported that nipa fruit husk extract contains polyphenolic compounds (Sudirman et al., 2024b).

Polyphenols are thermolabile compounds (Antony & Farid, 2022). Thermal degradation is the primary mechanism cited to explain the reduction in polyphenol yield during high-temperature extractions (Duarah et al., 2024). A previous study reported that different extraction temperatures affect the total phenol and flavonoid contents of *L. flava* and *P. stratiotes* leaf extracts (Sudirman et al., 2024a). Additionally, high temperatures were found to reduce the polyphenol content in *Hibiscus sabdariffa* extract (Singh et al., 2021). Different extraction temperatures also have varying effects on the polyphenol content of *Utricularia aurea* and its antioxidant activity (Sudirman et al., 2023). Based on these references, temperature is a crucial factor in the polyphenol extraction process for determining the optimal conditions. However, the effects of temperature on the extraction of polyphenol compounds from nipa fruit husk have not been reported. Therefore, this study aimed to investigate the optimal extraction temperature for polyphenol compounds from nipa (*N. fruticans*) fruit husks.

MATERIALS AND METHODS

Preparation and extraction

The small fruit husk pieces were dried in a dehydrator at 45°C for 24 hours and then ground into dried fruit husk powder using a Microphyte disintegrator B-One DM-120M grinding machine. The extraction was performed using the maceration method at three different temperatures: 30°C, 45°C, and 60°C. A 50% ethanol solution (ethanol in water) was used as the solvent, following the procedure described by a previous study (Sudirman et al., 2024b). In brief, 20 g of the dried sample was placed in an Erlenmeyer flask containing 200 mL of

50% ethanol. After 2 hours of maceration, the filtrate and residue were separated using Whatman No. 42 filter paper. The filtrate was transferred to a new collection tube, and the residue was re-extracted with fresh solvent under the same conditions, continuing for a total of five extractions. After the extraction process, the filtrates were combined in a new collection tube, and the solvent was removed using a rotary vacuum evaporator at 40°C to form a paste, which was then freeze-dried.

Total phenolic, flavonoid, and tannin contents analysis

The total phenolic content was determined using Folin-Ciocalteu's method, following the procedure outlined by the previous study (Molole et al., 2022). Briefly, 0.2 mL of each extract (10 mg/mL) was mixed with Folin-Ciocalteu's phenol reagent in a reaction tube and allowed to react for 5 minutes. After the reaction, 8% sodium carbonate was added, and the volume was adjusted to 3 mL with distilled water (dH₂O). The mixture was then incubated in the dark for 30 minutes. The supernatant was separated by centrifugation at 3,000 rpm for 30 minutes. The absorbance of the supernatant was measured using a spectrophotometer (Genesys 150 ThermoScientific, Massachusetts, USA) at 750 nm. Gallic acid was used as a standard to quantify the total phenolic content, which was expressed as mg of gallic acid equivalent (GAE) per gram of dried sample (mg GAE/g dry weight).

The total flavonoid content was determined using the aluminium chloride colorimetric method, as described in a previous study (Shraim et al., 2021). Briefly, 0.5 mL of the extract solution was mixed with a 2% aluminium chloride solution (1:1) and incubated at room temperature for 60 minutes. After the reaction, the absorbance was measured immediately using a spectrophotometer (Genesys 150 ThermoScientific, Massachusetts, USA) at 420 nm. Quercetin was used as a standard to quantify the total flavonoid content, which was expressed as mg quercetin equivalent (QE) per gram of dried sample (mg QE/g dry weight).

Tannin content was measured using the Folin-Ciocalteu's method, as described by a previous study (Balaky et al., 2021), with tannic acid used as the standard. A stock solution of tannic acid was prepared at a concentration of 100 µg/mL and diluted to various concentrations to create a standard curve. For the analysis, 0.1 mL of the sample solution (1 mg/mL) was pipetted into a 10 mL volumetric flask containing 7.5 mL of distilled water, 0.5 mL of Folin-Ciocalteu's reagent, and 1 mL of 35% sodium carbonate. Distilled water was added to bring the total volume to 10 mL. The mixture was then stirred and incubated at room temperature for 30 minutes. Absorbance was measured at 700 nm using a spectrophotometer. The tannin concentration was calculated and expressed as mg tannic acid equivalent (TAE) per gram of dried sample (mg TAE/g dry weight).

Functional group analysis

The Fourier transform infrared spectroscopy (FT-IR) analysis was used to detect the functional group of polyphenols from nipa palm fruit husk. FT-IR spectra of polyphenols were obtained by mixing polyphenol extract with potassium bromide, further it was pressed to form pellets. The analysis used a Fourier transform infrared (InfraRed Bruker Tensor 37) and was performed according to a previous method (Bhateja et al., 2020).

Antioxidant activity assay

The antioxidant activity of nipa palm extract was analyzed by the 2,2-diphenyl-1-picrylhydrazyl (DPPH) method (Sudirman et al., 2022). Briefly, the extract was dissolved in ethanol to make a concentration of 0.25 mg/mL. Then, 1 mL of each sample (0 mg/mL and 0.25 mg/mL) was mixed with 0.2 mM DPPH solution (1:1, v/v) and incubated at 37°C for 30 minutes. The absorbance was immediately measured using a UV-Vis spectrophotometer

(Genesys 150 ThermoScientific) at 517 nm. The antioxidant activity was calculated as the inhibition of the extract on the DPPH radical according to this formula (1):

$$\text{Percentage (\% inhibition)} = \frac{(\text{Abs}_{\text{blank}} - \text{Abs}_{\text{sample}})}{\text{Abs}_{\text{blank}}} \times 100\% \quad (1)$$

Whereas: $\text{Abs}_{\text{blank}}$, the absorbance at 517 nm without sample (0 mg/mL); $\text{Abs}_{\text{sample}}$, the absorbance at 517 nm with sample (0.25 mg/mL).

Statistical analysis

The data were presented as mean \pm standard deviation (SD) and analyzed by one-way analysis of variance (one-way ANOVA) followed by Duncan multiple comparison tests. Whereas, $p < 0.05$ was considered significantly different. Statistical analysis was performed with Statistical Product and Service Solution (SPSS) software ver22.0 (IBM Corporation, Armonk, NY, USA).

RESULTS AND DISCUSSION

Total phenolic, flavonoid, and tannin contents

In this study, polyphenol compounds were successfully extracted from the *N. fruticans* (nipa palm) fruit husk using different temperature extraction methods. The nipa fruit husk contains polyphenols, such as phenolic acids, flavonoids, and tannins. The total phenol content of nipa fruit husk extracts is shown in [Figure 1](#). The total phenolic content at an extraction temperature of 60°C (44.50 mg GAE/g) was significantly higher ($p < 0.05$) compared to 30°C (35.03 mg GAE/g), but there was no significant difference ($p > 0.05$) when compared to 45°C (40.26 mg GAE/g). The highest phenolic content was observed when the extraction was conducted at 60°C. A previous study reported the highest total phenolic content in *U. aurea* leaf when extracted at 60°C (50.80 mg GAE/g), compared to other extraction temperatures (30°C and 45°C) ([Sudirman et al., 2023](#)). Additionally, the total phenolic content in *Funtumia elastica* stem bark increases with rising extraction temperatures (25-65°C) ([Frempong et al., 2021](#)). The total phenolic content of grape marc also increases with extraction temperatures ranging from 45°C to 60°C ([Spigno et al., 2007](#)). The total phenol of mate tea leaves increases from 8.4 mg GAE/g to 11.8 mg GAE/g when extracted at 40°C and 60°C, respectively ([Rebocho et al., 2022](#)).

The total flavonoid content of nipa fruit husk extracts is shown in [Figure 2](#). The flavonoid content of nipa palm fruit husk at an extraction temperature of 60°C (8.75 mg QE/g) was significantly higher ($p < 0.05$) compared to an extraction temperature of 30°C (2.71 mg QE/g), with no significant difference ($p > 0.05$) when compared to 45°C (4.28 mg QE/g). The highest flavonoid content was observed when the extraction was conducted at 60°C. This study reports a similar finding to a previous study, where the extraction temperature of 60°C yielded the highest flavonoid content in *U. aurea* leaf, compared to other temperatures (30°C and 45°C) ([Sudirman et al., 2023](#)). Previous studies reported that the flavonoid content of *Cannabis sativa* increases from 14 mg LUE/100 g to 25 mg LUE/100 g when extracted at temperatures ranging from 40 to 60°C ([Teh & Birch, 2014](#)). Additionally, the flavonoid content of *Tabernaemontana heyneana* also increases with rising extraction temperatures (55 to 65°C) ([Sathishkumar et al., 2008](#)). A previous study also reported that the flavonoid content of *Daphne genkwa* increases as the extraction temperature rises from 40 to 60°C ([Xiang et al., 2024](#)).

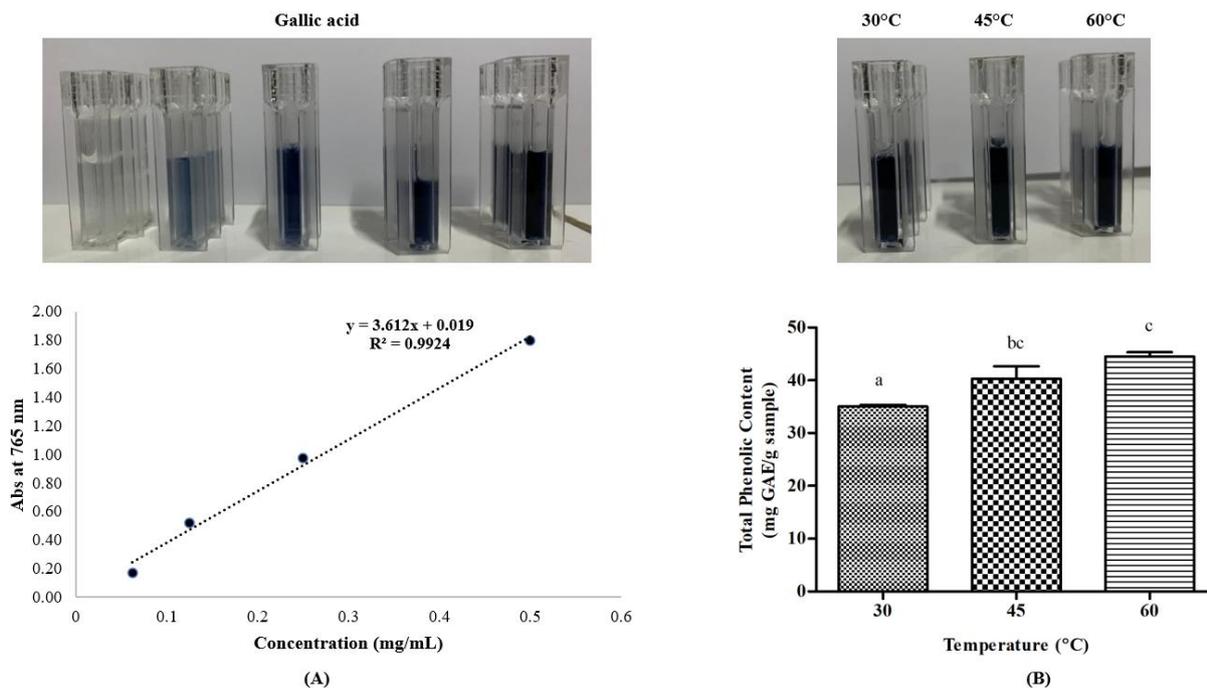


Fig. 1. (A) Gallic acid standard curve and (B) total phenolic content of nipa palm (*N. fruticans*) fruit husk extract at different extraction temperatures. Data are shown as mean ± standard deviation ($n=3$). The values with different letters represent significantly different ($p<0.05$) as analyzed by Duncan’s multiple range test.

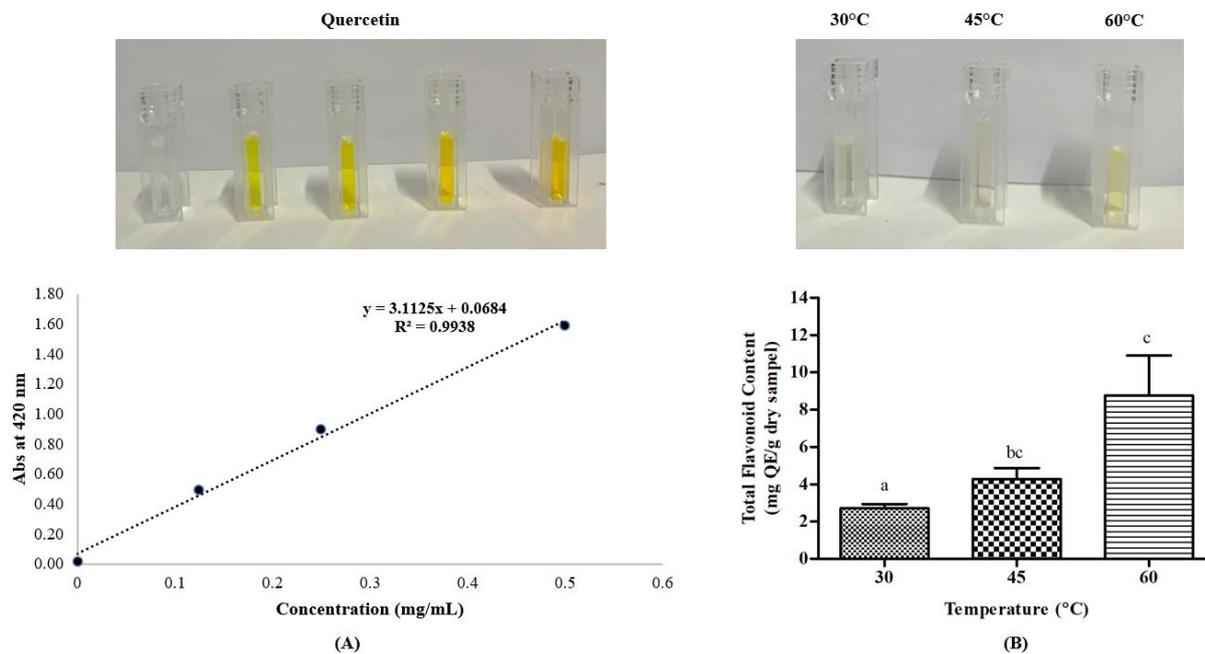


Fig. 2. (A) Quercetin standard curve and (B) total flavonoid content of nipa (*N. fruticans*) fruit husk extract at different extraction temperatures. Data are shown as mean ± standard deviation ($n=3$). The values with different letters represent significantly different ($p<0.05$) as analyzed by Duncan’s multiple range test.

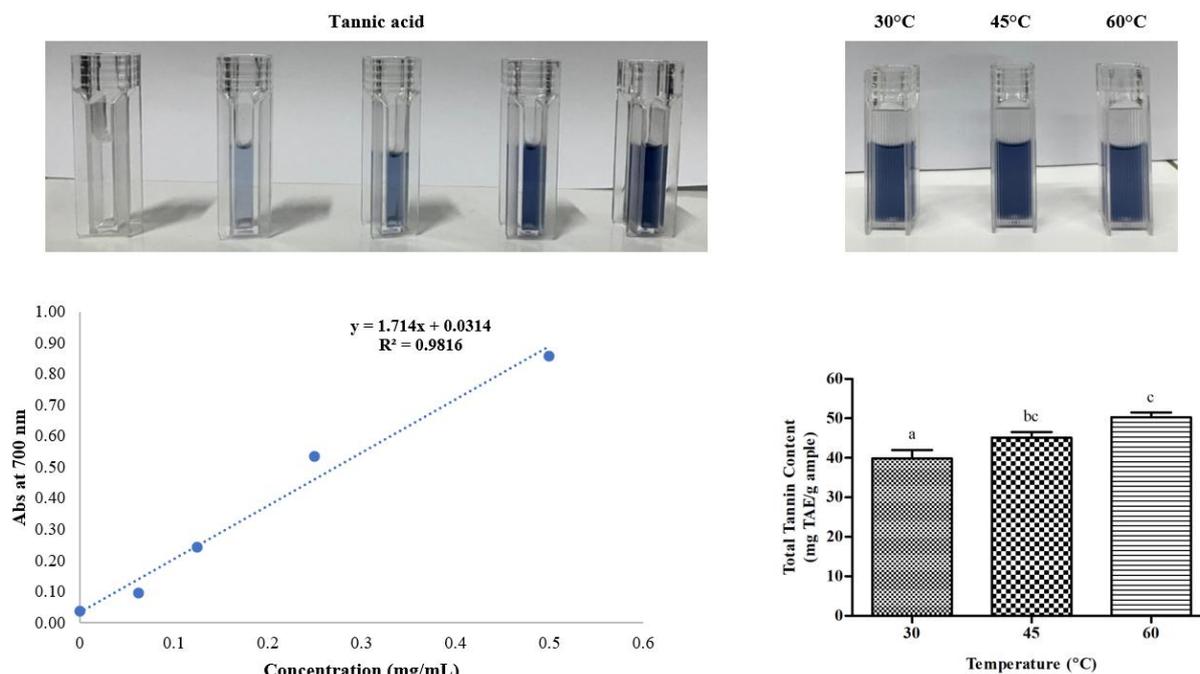


Fig. 3. (A) Tannic acid standard curve and (B) total tannin content of nipa (*N. fruticans*) fruit husk extract at different extraction temperatures. Data are presented as mean \pm standard deviation ($n=3$). The values with different letters represent significantly different ($p<0.05$) as analyzed by Duncan's multiple range test.

The total tannin content of nipa fruit husk extracts is shown in Figure 3. The tannin content of nipa fruit husk at an extraction temperature of 60°C (50.30 mg TAE/g) was significantly higher ($p<0.05$) compared to 30°C (39.85 mg TAE/g), with no significant difference ($p>0.05$) when compared to 45°C (45.11 mg TAE/g). The highest tannin content was observed when the extraction was conducted at 60°C. A previous study reported that the total tannin content of coffee husk increased from 13.03 TAE/g to 22.83 TAE/g with an increase in extraction temperature (40°C – 60°C) (Maimulyanti et al., 2023). Additionally, the total tannin content of *Quercus infectoria* galls also increased with rising extraction temperatures (50-75°C) (Arina & Harisun, 2019).

Overall, the total phenolic, flavonoid, and tannin contents increase with rising extraction temperatures. An increase in temperature promotes the release of secondary metabolites, such as polyphenolic compounds, from the plant matrix into the solvent. This is due to the reduction in solvent viscosity and the increase in diffusivity at higher temperatures (Liu et al., 2019). A previous study also reported that increasing the temperature during the extraction process softened plant tissues and weakened the interactions between proteins and phenols, as well as between phenols and polysaccharides, leading to greater migration of polyphenols into the solvent (Shi et al., 2003; Ma et al., 2022). However, polyphenols are thermolabile compounds and higher temperatures may cause the degradation of polyphenolic compounds (Antony & Farid, 2022). A previous study reported that the phenolic compounds in *Clinacanthus nutans* leaves decreased when extracted at 80°C (Sulaiman et al., 2017). Similarly, the total flavonoid content of seed cake extracts decreased when the extraction temperature exceeded 60°C (Teh & Birch, 2014). Additionally, the total tannin content in *Q. infectoria* galls decreased when the extraction temperature was increased from 75 to 100°C (Arina & Harisun, 2019). Additionally, the total phenolic content of *Cymbopogon citratus* decreased when the extraction temperature exceeded 90°C (Muala et al., 2021). These changes are attributed to high temperatures causing the oxidation and degradation of the

desired compounds. Thus, temperature plays a crucial role in the polyphenol extraction process. In this present study, the optimum extraction temperature was found to be 60°C.

Functional group of polyphenol compound

The polyphenol functional groups of nipa fruit husk extract are shown in Figure 4. The FT-IR spectra revealed peaks at 3268.62 cm^{-1} , 2201.17 cm^{-1} , 2097.65 cm^{-1} , 1990.11 cm^{-1} , 1608.68 cm^{-1} , 1520.54 cm^{-1} , 1439.45 cm^{-1} , 1365.86 cm^{-1} , 1256.04 cm^{-1} , and 1090.98 cm^{-1} . These peaks indicate the stretching of the polymeric hydroxyl group (O-H) and hydrogen-bonded stretching at 3268.62 cm^{-1} , and O-H bending at 1365.86 cm^{-1} (Nandiyanto et al., 2019). Figure 4 also shows C-O stretching vibration absorption bands at 1608.68 cm^{-1} and aromatic ethers at 1256.04 cm^{-1} . Additionally, the alkyne group ($\text{C}\equiv\text{C}$) was detected at 2201.17 cm^{-1} . The region between 3400 and 3200 cm^{-1} is associated with the asymmetric and symmetric stretching of the polymeric hydroxyl (O-H) group and hydrogen bonding, which is a characteristic feature of polyphenolic compounds (Wongsa et al., 2022).

Antioxidant activity

The antioxidant activity of nipa palm (*N. fruticans*) is shown in Figure 5. As indicated in Figure 5, the extract at 0.25 mg/mL exhibits antioxidant activity ranging from 43.55% to 52.79%. The extract at an extraction temperature of 60°C (52.79%) shows significantly ($p < 0.05$) higher inhibition compared to the other temperatures (30 and 45°C). Whereas, no significant difference ($p > 0.05$) between extraction temperature of 30°C (43.55%) and 45°C (45.41%). A previous study reported that the percentage inhibition of *Aloiampelos striatula* leaves-methanolic extract at 0.20 mg/mL was approximately 34.83% (Mokoroane et al., 2020). Additionally, the 0.2 mg/mL extract of *Grewia carpinifolia* exhibited an inhibition percentage of about 11.85%, with a total phenol content of 19.08 mg GAE/g (Adebiyi et al., 2017). The present study indicates that nipa palm fruit husk extract is more effective in antioxidant activity compared to these previous studies. This may be due to its higher polyphenol content compared to the extracts in the studies cited.

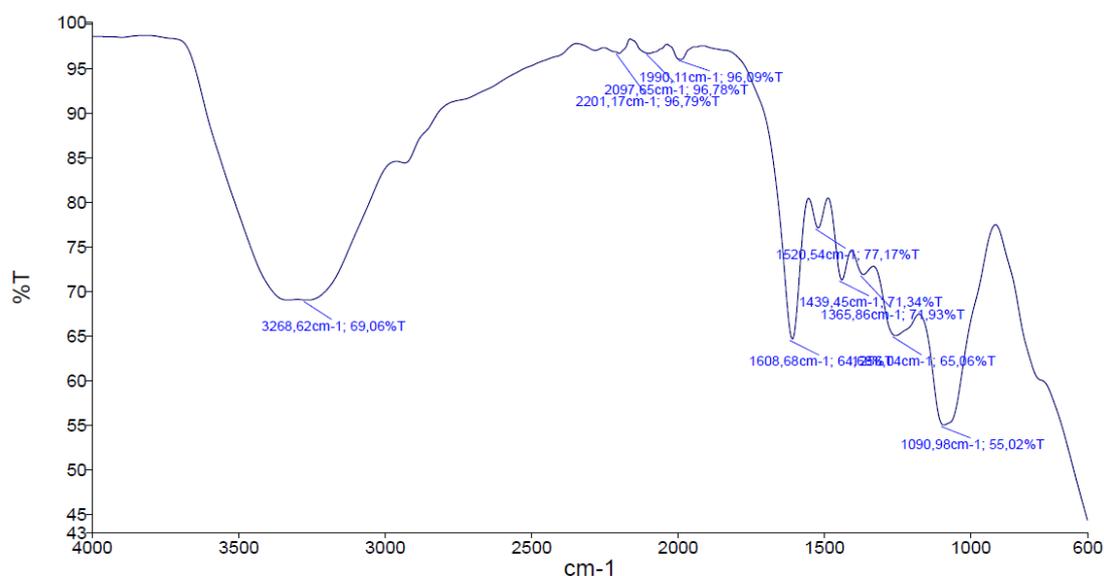


Fig. 4. The representative Fourier transform infrared spectroscopy (FT-IR) spectra peaks of nipa palm (*N. fruticans*) fruit husk extract at an extraction temperature of 60°C.

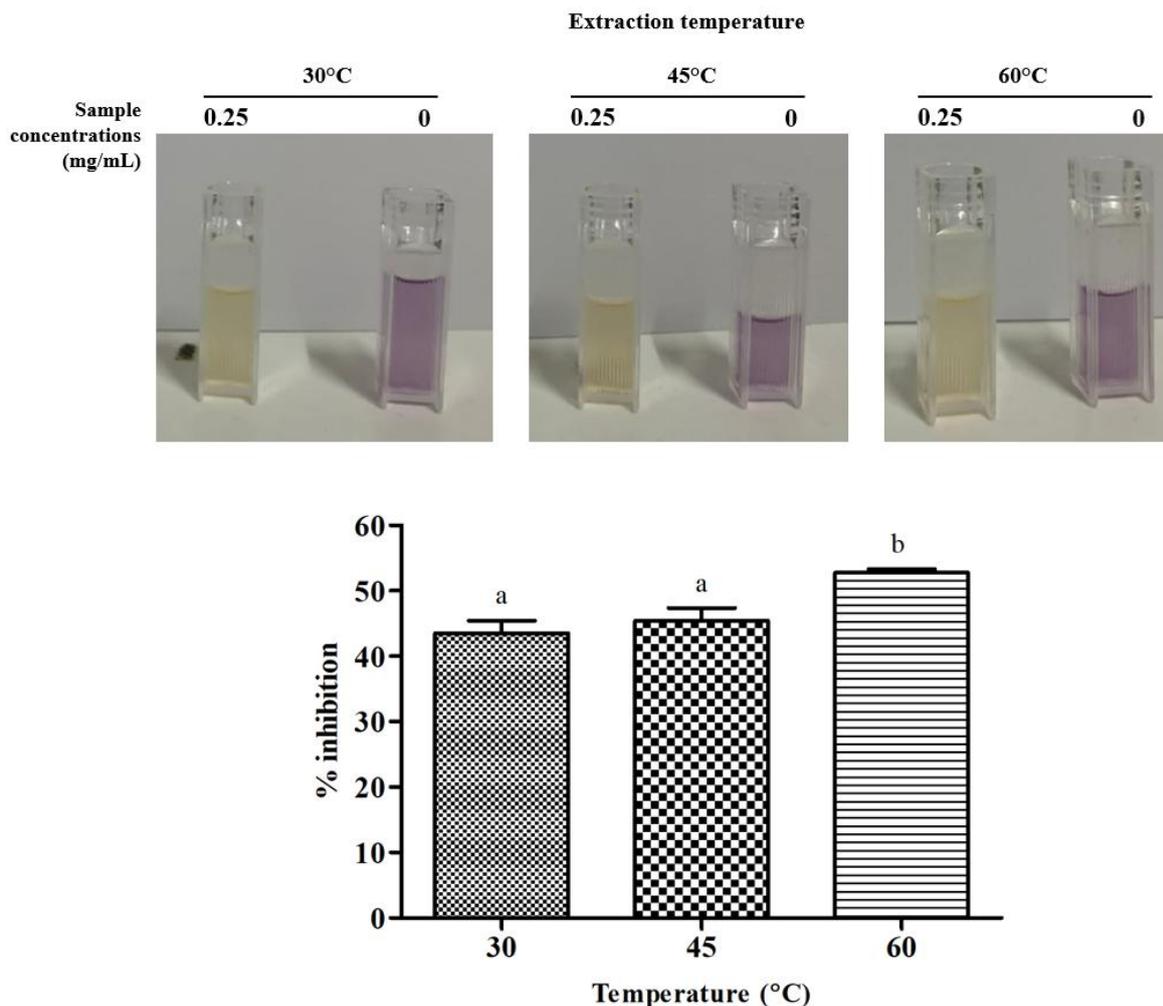


Fig. 5. Antioxidant activity of nipa palm (*N. fruticans*) fruit husk extract at different extraction temperatures. Data are presented as mean \pm standard deviation ($n=3$). The values with different letters represent significantly different ($p<0.05$) as analyzed by Duncan's multiple range test.

When the free-radical DPPH interacts with an unpaired electron, it shows the highest absorption at 517 nm, producing a purple color. A free-radical scavenging antioxidant reacts with DPPH to form DPPHH, which absorbs less light due to its reduced hydrogen content. As more electrons are captured, the DPPHH radical causes decolorization, then shifting the color from purple to yellow (Baliyan et al., 2022). The antioxidant activity of nipa palm husk extracts is due to the presence of polyphenol compounds, such as phenolic acid, flavonoid, and tannin. The extraction temperature of 60°C shows the highest antioxidant activity due to the presence of the highest polyphenol compounds especially phenolic acid and tannins. A previous study reported that polyphenol groups are known as antioxidant agents because they inhibit free radical formation. It can act as a radical scavenger by donating hydrogen atoms or electron transfer mechanisms (Andrés et al., 2023; Lang et al., 2024).

CONCLUSION

The polyphenol compounds were successfully extracted from the nipa palm (*Nypa fruticans*) fruit husk using 50% ethanol at various extraction temperatures. The extraction at 60°C yielded the highest total phenolic (44.50 mg GAE/g), flavonoid (8.75 mg QE/g), and tannin

(50.30 mg TAE/g) contents compared to the 30°C and 45°C extractions. Consequently, it also exhibited the highest antioxidant activity, with a percentage inhibition of approximately 52.79% at 0.25 mg/mL. Fourier transform infrared analysis identified hydroxyl groups, confirming the presence of polyphenol compounds in the nipa palm fruit husk extract. These results suggest that nipa palm fruit husk extract could potentially be used as a source of nutraceutical food, particularly as a natural antioxidant agent.

Conflict of interest

The authors declare that there is no conflict of interest.

Acknowledgments

The research or publication of this article was funded by DIPA of Public Service Agency of Universitas Sriwijaya 2024. SP DIPA Number 023.17.2.677515/2024 on November 24, 2023. In accordance with the Rector's Decree Number: 0013/UN9/LP2M.PT/2024, on May 20, 2024.

REFERENCES

- Adebiyi, O. E., Olayemi, F. O., Ning-Hua, T., & Guang-Zhi, Z. (2017). *In vitro* antioxidant activity, total phenolic and flavonoid contents of ethanol extract of stem and leaf of *Grewia carpinifolia*. *Beni-Suef University Journal of Basic and Applied Sciences*, 6(1), 10-14. <http://dx.doi.org/10.1016/j.bjbas.2016.12.003>.
- Andrés, C. M. C., Lastra J. M. P. D. L., Juan, C. A., Plou, F. J., & Pérez-Lebeña, E. (2023). Polyphenols as antioxidant/pro-oxidant compounds and donors of reducing species: Relationship with human antioxidant metabolism. *Process*, 11(9), 2771. <http://dx.doi.org/10.3390/pr11092771>.
- Antony, A. & Farid, M. (2022). Effect of temperatures on polyphenols during extraction. *Applied Sciences*, 12(4), 2107. <http://dx.doi.org/10.3390/app12042107>.
- Arina, M. Z. I., & Harisun, Y. (2019). Effect of extraction temperatures on tannin content and antioxidant activity of *Quercus infectoria* (Manjakani). *Biocatalysis and Agricultural Biotechnology*, 19, 101104. <http://dx.doi.org/10.1016/j.bcab.2019.101104>.
- Astuti, M. D., Nisa, K., & Mustikasari, K. (2020). Identification of chemical compounds from nipah (*Nypa fruticans* Wurmb.) endosperm. *BIO Web of Conferences*, 20, 03002. <https://doi.org/10.1051/bioconf/20202003002>.
- Balaky, H.H., Khalid, K., Hasan, A., Tahir, S., Ubur, S., & Khedir, A (2021). Estimation of total tannin and total phenolic content in plant (*Crataegus azarolus* L.) by orbital shaker technique. *International Journal of Agriculture, Environment and Food Sciences*, 5(1), 1-6 (2021). <https://doi.org/10.31015/jaefs.2021.1.1>.
- Baliyan, S., Mukherjee, R., Priyadarshini, A., Vibhuti, A., Gupta, A., Pandey, R. P., & Chang, C, M. (2022). Determination of antioxidants by dpph radical scavenging activity and quantitative phytochemical analysis of *Ficus religiosa*. *Molecules*, 27(4), 1326. <https://doi.org/10.3390/molecules27041326>.
- Bhateja, P. K., Kajal, A., & Singh, R. (2020). Amelioration of diabetes mellitus by modulation of GLP-1 via targeting alpha-glucosidase using *Acacia tortilis* polysaccharide in streptozotocin-nicotinamide induced diabetes in rats. *Journal of Ayurveda and Integrative Medicine*, 11(4), 405-413. <https://doi.org/10.1016/j.jaim.2019.06.003>.
- Chandimali, N., Bak, S. G., Park, E. H., Lim, H.-J., Won, Y.-S., Kim, E.-K., Park, S.-I., & Lee, S. J. (2025). Free radicals and their impact on health and antioxidant defenses: A review. *Cell Death Discovery*, 11, 19. <https://doi.org/10.1038/s41420-024-02278-8>.
- Duarah, P., Joardar, S., Debnath, B., & Purkait, M. K. (2024). Optimized extraction of polyphenols from tea factory waste and cost-effective drying methods for sustainable utilization. *Bioresource Technology Reports*, 26, 101833. <https://doi.org/10.1016/j.biteb.2024.101833>.

- Frempong, F. T., Boadi, N. O., & Badu, M. (2021). Optimization of extraction conditions for polyphenols from the stem bark of *Funtumia elastica* (Funtum) utilizing response surface methodology. *AAS Open Research*, 4, 46. <http://dx.doi.org/10.12688/aasopenres.13284.2>.
- Hermanto, H., Mukti, R. C., & Pangawikan, A. D. (2020). Nipah (*Nypa fruticans* Wurmmb.) fruit as a potential natural antioxidant source. *IOP Conference Series: Earth and Environmental Science*, 443(1), 012096. <http://dx.doi.org/10.1088/1755-1315/443/1/012096>.
- Herpandi, Lestari, S. D., Bastian, & Sudirman, S. (2021). Antioxidant activity of the fractions from water lettuce (*Pistia stratiotes*) extract. *Food Research*, 5(2), 451-455. [http://dx.doi.org/10.26656/fr.2017.5\(2\).578](http://dx.doi.org/10.26656/fr.2017.5(2).578).
- Lang, Y., Gao, N., Zang, Z., Meng, X., Lin, Y., Yang, S., Yang, Y., Jin, Z., & Li, B. (2024). Classification and antioxidant assays of polyphenols: A review. *Journal of Future Foods*, 4(3), 193-204. <http://dx.doi.org/10.1016/j.jfutfo.2023.07.002>.
- Liu, Y., Li, J., Fu, R., Zhang, L., Wang, D., & Wang, S. (2019). Enhanced extraction of natural pigments from *Curcuma longa* L. using natural deep eutectic solvents. *Industrial Crops and Products*, 140, 111620. <http://dx.doi.org/10.1016/j.indcrop.2019.111620>.
- Ma, Y., Meng, A., Liu, P., Chen, Y., Yuan, A., Dai, Y., Ye, K., Yang, Y., Wang, Y., & Li, Z. (2022). Reflux extraction optimization and antioxidant activity of phenolic compounds from *Pleioblastus amarus* (Keng) shell. *Molecules*, 27, 362. <https://doi.org/10.3390/molecules27020362>.
- Maimulyanti, A., Prihadi, A. R., Mellisani, B., Nurhidayati, I., Putri, F. A. R., Puspita, F., & Widarsih, R. W. (2023). Green extraction technique to separate tannin from coffee husk waste using natural deep eutectic solvent (Nades). *RASAYAN Journal of Chemistry*, 16(3), 2002-2008. <http://dx.doi.org/10.31788/rjc.2023.1638334>.
- Mokoroane, K. T., Pillai, M. K., & Magama, S. (2020). 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity of extracts from *Aloiampelos striatula*. *Food Research*, 4(6), 2062-2066. [http://dx.doi.org/10.26656/fr.2017.4\(6\).241](http://dx.doi.org/10.26656/fr.2017.4(6).241).
- Molole, G. J., Gure, A., & Abdissa, N. (2022). Determination of total phenolic content and antioxidant activity of *Commiphora mollis* (Oliv.) Engl. resin. *BMC Chemistry*, 16, 48. <https://doi.org/10.1186/s13065-022-00841-x>.
- Muala, W. C. B., Desobgo, Z. S. C. D., & Jong, N. E. (2021). Optimization of extraction conditions of phenolic compounds from *Cymbopogon citratus* and evaluation of phenolics and aroma profiles of extract. *Heliyon*, 7, e06744. <https://doi.org/10.1016/j.heliyon.2021.e06744>.
- Mukherjee, S., Chopra, H., Goyal, R., Jin, S., Dong, Z., Das, T., & Bhattacharya, T. (2024). Therapeutic effect of targeted antioxidant natural products. *Discover Nano*, 19, 144. <https://doi.org/10.1186/s11671-024-04100-x>.
- Nandiyanto, A. B. D., Oktiani, R., & Ragadhita, R. (2019) How to read and interpret FT-IR spectroscopy of organic material. *Indonesian Journal of Science and Technology*, 4(1), 97-118. <http://dx.doi.org/10.17509/ijost.v4i1.15806>.
- Rad, M. S., Kumar, N. V. A., Zucca, P., Varoni, E. M., Dini, L., Panzarini, E., Rajkovic, J., Fokou, P. V. T., Azzini, E., Peluso, I., Mishra, A. P., Nigam, M., El-Rayess, Y., Beyrouthy, M. E., Polito, L., Iriti, M., Martins, N., Martorell, M., Docea, A. O., Setzer, W. N., Calina, D., Cho, W. C., & Sharifi-Rad, J. (2020). Lifestyle, oxidative stress, and antioxidants: Back and forth in the pathophysiology of chronic diseases. *Frontiers in Physiology*, 11, 694. <http://dx.doi.org/10.3389/fphys.2020.00694>.
- Rebocho, S., Mano, F., Cassel, E., Anacleto, B., Bronze, M. D. R., Paiva, A., & Duarte, A. R. C. (2022). Fractionated extraction of polyphenols from mate tea leaves using a combination of hydrophobic/hydrophilic nades. *Current Research in Food Science*, 5, 571-580. <http://dx.doi.org/10.1016/j.crfs.2022.03.004>.
- Sathishkumar, T., Baskar, R., Shanmugam, S., Rajasekaran, P., Sadasivam, S., & Manikandan, V. (2008). Optimization of flavonoids extraction from the leaves of *Tabernaemontana heyneana* Wall. using L₁₆ orthogonal design. *Nature and Science*, 6(3), 10-21.
- Shraim, A. M., Ahmed, T. A., Rahman, M. M., & Hijji, Y. M. (2021). Determination of total flavonoid content by aluminum chloride assay: A critical evaluation. *LWT*, 150, 111932. <https://doi.org/10.1016/j.lwt.2021.111932>.

- Shi, J., Yu, J., Pohorly, J., Young, J. C., Bryan, M., & Wu, Y. (2003). Optimization of the extraction of polyphenols from grape seed meal by aqueous ethanol solution. *Food, Agriculture & Environment*, 1(2), 42-47.
- Singh, M., Thrimawithana, T., Shuklaa, R., & Adhikari, B. (2021) Extraction and characterization of polyphenolic compounds and potassium hydroxycitrate from *Hibiscus sabdariffa*. *Future Foods*, 4, 100087. <https://doi.org/10.1016/j.fufo.2021.100087>.
- Spigno, G., Tramelli, L., & Faveri, D. M. D. (2007). Effects of extraction time, temperature and solvent on concentration and antioxidant activity of grape marc phenolics. *Journal of Food Engineering*, 81(1), 200-208. <http://dx.doi.org/10.1016/j.jfoodeng.2006.10.021>.
- Sudirman, S., Baehaki, A., Fathullah, F., & Janna, M. (2023). Effects of extraction temperature on polyphenol compounds and antioxidant activity of golden bladderwort (*Utricularia aurea*). *Agritech*, 43(4), 308-313. <http://dx.doi.org/10.22146/agritech.75223>.
- Sudirman, S., Herpandi, Rinto, Lestari, S., Harma, M., & Aprilia, C. (2024a). Effects of extraction temperature on bioactive compounds and antioxidant activity of yellow velvetleaf (*Limnocharis flava*) and water lettuce (*Pistia stratiotes*) leaf extract. *Food Research*, 8(1), 136-139. [http://dx.doi.org/10.26656/fr.2017.8\(1\).113](http://dx.doi.org/10.26656/fr.2017.8(1).113).
- Sudirman, S., Herpandi, Safitri, E., Apriani, E. F., & Taqwa, F. H. (2022). Total polyphenol and flavonoid contents and antioxidant activities of water lettuce (*Pistia stratiotes*) leave extracts. *Food Research*, 6(4), 205-210. [http://dx.doi.org/10.26656/fr.2017.6\(4\).484](http://dx.doi.org/10.26656/fr.2017.6(4).484).
- Sudirman, S., Wardana, A. K., Herpandi, Widiastuti, I., Sari, D. I., & Janna, M. (2024b). Antioxidant activity of polyphenol compounds extracted from nipa palm (*Nypa fruticans*) fruit husk with different ethanol concentrations. *International Journal of Secondary Metabolite*, 11(2), 355-363. <https://doi.org/10.21448/ijsm.1360736>.
- Sulaiman, I. S. C., Basri, M., Masoumi, H. R. F., Chee, W. J., Ashari, S. E., & Ismail, M. (2017). Effects of temperature, time, and solvent ratio on the extraction of phenolic compounds and the anti-radical activity of *Clinacanthus nutans* Lindau leaves by response surface methodology. *Chemistry Central Journal*, 11(1), 54. <https://doi.org/10.1186/s13065-017-0285-1>.
- Teh, S. S., & Birch, E. J. (2014). Effect of ultrasonic treatment on the polyphenol content and antioxidant capacity of extract from defatted hemp, flax and canola seed cakes. *Ultrasonics Sonochemistry*, 21(1), 346-353. <https://doi.org/10.1016/j.ultsonch.2013.08.002>.
- Wilczyńska, A. & Żak, N. (2024). Polyphenols as the main compounds influencing the antioxidant effect of honey—A Review. *International Journal of Molecular Sciences*, 15, 10606. <https://doi.org/10.3390/ijms251910606>.
- Wongsa, P., Phatikulrungsun, P., & Prathumthong, S. (2022). FT-IR characteristics, phenolic profiles and inhibitory potential against digestive enzymes of 25 herbal infusions. *Scientific Reports*, 12, 663. <https://doi.org/10.1038/s41598-022-10669-z>.
- Xiang, Y., Liu, Z., Liu, Y., Dong, B., Yang, C., & Li, H. (2024). Ultrasound-assisted extraction, optimization, and purification of total flavonoids from *Daphne genkwa* and analysis of their antioxidant, anti-inflammatory, and analgesic activities. *Ultrasonics Sonochemistry*, 111, 107079. <https://doi.org/10.1016/j.ultsonch.2024.107079>.