Vol 9 No. 2 (2024) 252-261 ISSN (Online) : 2502-0943

Jurnal Katalisator

ANTI-ANTIOXIDANT ACTIVITY LEAVES ANANASA COMOSUS IS MEDIUM USING THE DPPH METHOD

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Detail Artikel

Diterima : 9 September 2024
Direvisi : 17 November 2024
Diterbitkan : 17 November 2024

Kata Kunci

Pineapple leaf
Antioxidant
IC_{50}
DPPH

Penulis Korespondensi

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ABSTRACT

One of the best fruit plants in Indonesia, the pineapple (Ananas comosus), has a high antioxidant content. Ananas comosus, or pineapple leaves, are components of the pineapple plant. They have a somewhat thick leaf sheath with small brown spines pointing upwards along the edge. (Ananasa comosus). As is well known, Riau in general and Pekanbaru in particular are abundant producers of pineapples as a natural resource, with approximately 7,595 tons in 2021, resulting in a significant amount of pineapple leaves being wasted. Research on the antioxidants from pineapple leaves has not been extensively conducted. The aim of this study is to investigate the potential of pineapple leaves (Ananas comosus) as a source of natural antioxidants. The DPPH technique is being used in a descriptive quantitative research study to investigate the antioxidant capacity of pineapple leaves. Ananas comosus, or dried and crushed pineapple leaves, are macerated in a methanol solvent until the sample is fully submerged. The IC50 value determined by the linear regression equation is the parameter used in this technique. After the extract evaporated from the solvent, its absorbance was

http://doi.org/10.22216/jk.v5i2.5717
Published by LLDIKTI Wilayah X

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measured at 520 nm using a microplate reader and the DPPH technique. Pineapple leaves showed moderate antioxidant activity against DPPH with an IC50 value of 113.069 ppm, according to data from calculations and studies. According to this study, the antioxidant capacity of pineapple leaves (Ananas comosus) is moderate.

INTRODUCTION

Background Indonesia is a country rich in natural resources and has a high diversity of plants and is mostly spread in tropical forest areas known as a myriad of medicinal plants. (Jamshidi-Kia et al., 2020) . The community uses medicinal plants that can cure all diseases, which is a traditional medicine. (Juariah et al., 2024), To treat degenerative diseases such as stroke, hypertension, diabetes mellitus, and heart disease, which are caused by free radical compounds.(Jamshidi-Kia et al., 2020) A free radical is a molecule that has one or more unpaired electrons that are unstable and can cause damage to surrounding molecules to obtain electron pairs. (Jamshidi-Kia et al., 2020) . Therefore, in the human body there are compounds that can prevent free radicals, namely antioxidants. (Jayawardena et al., 2020)Antioxidants are molecules or compounds that are stable enough to donate their electrons or hydrogen to free radical molecules or compounds and neutralize them. (Ou et al., 2021). Antioxidants are divided into two parts, namely natural antioxidants and synthetic antioxidants (Ou et al., 2021) Antioxidants can be found in foods that contain vitamins such as vitamin E, vitamin C, and β-carotene, and there are several other compounds such as phenolic compounds, flavonoid compounds, or organic acids that we can obtain from plants, one of which is the pineapple plant (Phuyal et al., 2020).

Pineapple is one of the fruit-producing plants that is very popular and liked by the people of Indonesia. In addition, pineapple is also one of the natural ingredients used as traditional medicine. (Sienkiewicz et al., 2022). The pineapple plant is a short-stemmed plant and is a monocot plant and sprouts seedlings; then the leaves are long and have small brown thorns that face upwards on the edge of the pineapple leaf sheath. In addition, pineapple leaves can also be used as a medicine for urinary stones; pineapple leaf shoots are taken three times a day. (Yeow et al., 2021) Then pineapple leaf waste has not been used optimally so that many farmers only burn it, which can cause air pollution due to the smoke, and there are also those who leave it alone; therefore, the waste problem, if not overcome, will disturb and harm the environment and the local community. The solution that can be used to manage pineapple leaf waste is to process it into fiber to become a quality fiber product that has a selling value and has added value if studied from an economic perspective, so it is necessary to make efforts to deal with pineapple leaf waste. (Hamzah et al., 2021).

RESEARCH METHODS

Sample Extraction

http://doi.org/10.22216/jk.v5i2.5717
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We wash the samples, thinly chop them to separate the meat from the fruit, and then dry them at room temperature. The process involves storing 10 grams of finely peeled Dutch tuber leather in a dark bottle. The useBy using dark bottles, you can minimize exposure to light, which can destroy light-sensitive compounds like pigments or easily oxidized compound compositions, reduce extraction efficiency, and accelerate the degradation of the desired active ingredients. Next, you add methanol as a solvent to extract the active substances from the sample, soaking and silencing it for three consecutive 24 The sample results are then filtered using filter paper. Test antioxidant activity with the DPPH method. (Yunita & Sari, 2022),(Surya & Marliza, 2020).

Working Procedure

Sample Extract by Maceration The sample used in this study was pineapple leaves, and then good, good, and pest-free pineapple leaves were selected to be used as research samples. Pineapple leaf samples that have been selected are washed thoroughly using running water and cut into small pieces. Next, it is dried in the sun. The dried pineapple leaf sample was crushed and weighed as much as 10 grams. The sample was put into a dark bottle, and then methanol solvent was added until the sample was submerged and left for 72 hours. After soaking for 72 hours, see the results of soaking the sample on pineapple leaves, filtered using filter paper, and put into a vial tube, left to dry to get extract from pineapple leaves. The extracted extract was then tested for antioxidant activity.

Antioxidant Activity Test Using the DPPH Method The antioxidant activity test was carried out using a microplate reader with DPPH at a wavelength of 520 nm. A sample of 2 mg is dissolved in 2 mL of methanol so that the sample concentration becomes 1000 ppm. Row A contains a sample of 100 µL (the plate consists of rows A-H, each totaling 8 wells). A total of 50 µL of methanol was inserted into each well in the B-H row. Row A is pipetted as much as 50 µL and put into row B. Row B is pipetted. 50 µL is put into row C and done until F; row F is pipetted 50 µL and then discarded. So that concentrations of 1000 ppm, 500 ppm, 250 ppm, 125 ppm, 62.5 ppm, and 31.25 ppm were obtained. While the G-H line is filled with 50 µL of methanol, specifically in the H line, only wells 1-6 are filled. Rows A-G are added with 80 µL of DPPH at a concentration of 40 ppm, then incubated for 30 minutes. Radical capture activity was measured as a decrease in DPPH absorbance with microplate readers and data processing. As a comparison, vitamin C has a concentration of 50 ppm.

The percentage of IC_{50} inhibition is determined by using analysis by making a relationship curve between the percentage of resistance and concentration.

The value of % inhibition is calculated by the following formula:

$$\%\ \text{inhibition} = \frac{A\ control - A\ sample}{A\ control} \times 100\%$$

Remarks: A control = Absrobsi does not contain samples

A Sample = Absrobsi containing the sample

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The formula for the linear equation is as follows:

$Y=ax+b$

Description: Y = Sample absorption

x = Sample concentration

IC_{50} Formula

$y=ax-b$

Remarks: y = absorbance of the sample

x = Sample concentration

Data Analysis

Data analysis used sample absorbance measurements and the percentage formula (%) of inhibition. The data is analyzed by creating a calibration curve and then entered into the linear regression equation of % inhibition by concentration and then calculated the value.

RESULTS AND DISCUSSION

Results Analysis of antioxidant activity using the DPPH method with a microplate reader at a wavelength of 520 nm produced an IC_{50} value as seen in table 1.

Table 1 Percent inhibition of pineapple leaf sample concentration

Methanol Concentration (ppm)	Ln Concentration	% Inhibition	IC_{50}
1000	6.907	81.96	
500	6.214	74.184	
250	5.521	63.297	113.069
125	4.828	55.521	
62.5	4.135	45.412	
31.25	3.442	22.706	

Based on table .1 above, the concentration of pineapple leaf extract sample was obtained at 113.069 ppm.

http://doi.org/10.22216/jk.v5i2.5717
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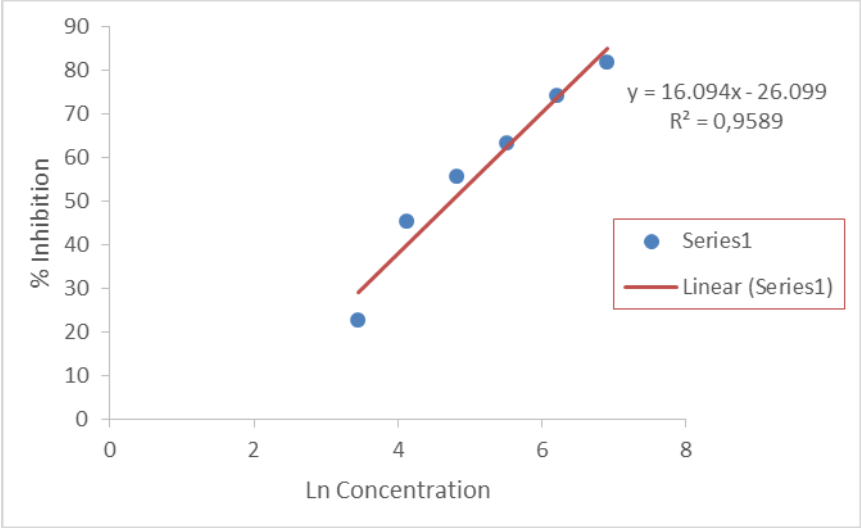

Graph 1. Relationship of % inhibition to sample concentration Calculation of IC_{50} with methanol fraction

Y = 16.094 LnX – 26.099

50 = 16.094 LnX – 26.099

50 + 26.099 = 16.094X

X = $\frac{76.099}{16.094} = 4{,}728$

IC_{50} = 113.069 ppm

Table. 2 Percent inhibition of Ascorbic Acid concentration

Ascorbic acid	Ln Concentration	% Inhibition	IC_{50}
50	3.912	96.6449	
25	3.218	87.2321	
12.5	2.525	76.0485	1.849
6.25	1.832	66.5424	
3.125	1.139	57.2227	
1.5625	0.446	48.2759	

Based on table 2 above, the concentration of ascorbic acid was obtained at 1.849 ppm.

http://doi.org/10.22216/jk.v5i2.5717
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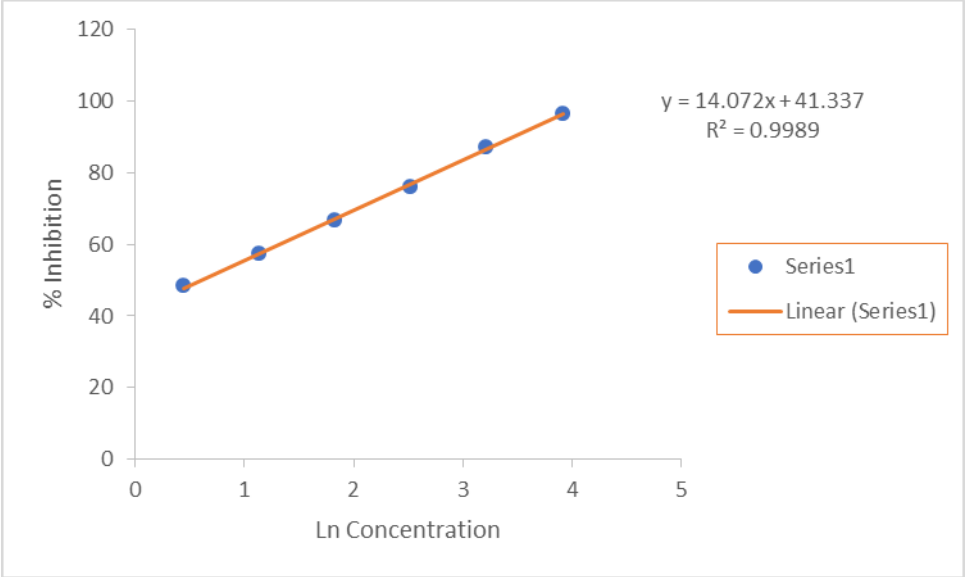
Jurnal Katalisator Vol 9 No. 1 (2024) 252-261



Y = 14.072 LnX + 41.337

50 = 14.072 LnX + 41.337

50 – 41.337 = 14.072 X

X $= \frac{8.663}{14.072} = 0{,}615$

IC_{50} = 1.849 ppm

Table 3 Phytochemical screening tests

Compound Class	Reagents	Identification Results	Conclusion
Flavonoid	Sample extract, Concentrated Magnesium + HCL.	Orange Yellow.	+
Phenolic and Tannins	Sample Extract, FeCl3 5%.	Green color.	+
Saponin	Sample extract, Aquades.	Foamed for 5 Minute.	+
Terpnoids and steroids	Sample extract, Chloroform, Acetic Acid, Sulfuric Acid	Bluish-green color steroids, terpenoid brownish ring.	–

http://doi.org/10.22216/jk.v5i2.5717
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Discussion

This study aims to determine the antioxidant activity in pineapple leaves using the DPPH method with *a microplate reader*. Pineapple leaves, first weighed as much as 10 grams and then macerated for 3 x 24 hours. The maceration method was chosen because it is simple in its work and uses only a few tools. Maceration is an aimplicia extraction process using solvents and several times of shaking or stirring at room temperature. (Zaiyar et al., 2020).

The solvent used in this study is methanol, because this solvent is a universal solvent, namely a solvent that can dissolve polar organic compounds in the sample. The sample will be macerated within 3x24 hours, after which the sample is extracted (filtered) using filter paper until methanol extract is obtained, after which an antioxidant examination is carried out using *a microplate reader* with a wavelength of 520 nm

Antioxidant activity was measured by calculating the amount of reduction in the intensity of the purple color of DPPH, which was proportional to the reduction in the concentration of DPPH solution. The immersion is produced by the reaction of the diphenyl pyrryl hydrazyl molecule with a hydrogen atom released by one molecule of the sample component so that a diphenyl pyrryhydrazyl compound is formed and causes the color change of DPPH from purple to yellow. The effect of sample concentration on the percentage of inhibition where the increase in activity is proportional to the increase in concentration. The regression equation and subsequently the 50% activity equation were determined so that the effective concentration price (IC_{50}) was obtained. IC50 is a number that indicates the concentration of the extract (ppm), which is able to inhibit the oxidation process by 50%. A compound is said to have very strong antioxidant activity if the IC_{50} value is less than 50 ppm; antioxidants are strong if the IC50 value is 50-100 ppm; antioxidants if the IC_{50} value is 101-250 ppm; antioxidants will be moderate if the IC_{50} value is 250-500 ppm; antioxidants will be weak; antioxidants if the IC50 value is 501-1000 will be very weak; and more than 1000 antioxidants are inactive. (Hidayah et al., 2019).

The IC_{50} value in ascorbic acid as a comparison is ppm, which is very strong antioxidant activity. This is because ascorbic acid is a pure solution. In the methanol extract sample of pineapple leaves, an IC_{50} value of 113.16 ppm was obtained, which is included in the category of moderate antioxidants. Based on this study, it is known that pineapple leaves can be used as antioxidants to ward off free radicals.

Based on research that has been conducted (Hamzah et al., 2021), pineapple peel contains secondary metabolites of alkaloids and steroid flavonoids. In the test of potential antioxidant activity, the ethyl acetate fraction of pineapple fruit peel has antioxidant potential with an IC_{50} value of 40.52 ppm, which is a very strong antioxidant category. The difference in results obtained by powerful antioxidants can be caused by several factors, such as solvents and those conducted in the study.

Based on Table 1 above, it is known that the antioxidant activity of pineapple leaves at the highest concentration was obtained with a percentage inhibition of 81.96%, and the lowest concentration was obtained with a percentage inhibition of 22.706%. So the concentration of the antioxidant activity test of pineapple leaf extract was obtained at 113.16 ppm. Based on table 4.2 above, it is known that the activity of ascorbic acid at the highest concentration was

http://doi.org/10.22216/jk.v5i2.5717
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obtained as a percentage inhibition of 96.6449%, and the lowest concentration was obtained as a percent of 48.2759%. So the results of the antioxidant activity test of pineapple leaf extract were obtained at 1.8517 ppm.

In Table. 3, the flavonoid test of pineapple leaf extract gave a positive result which was characterized by a yellow-orange color change after adding concentrated Mg and HCl powder to the sample solution. The positive results in flavonoid testing are due to the reduction of Mg metal flavonoids and the formation of flavilium salts that can form orange to yellow colors. In the testing of phenolic compounds and tannins, pineapple leaf extract will give positive results if there is a change in green color after adding 5% $FeCl_3$ sample extract to the sample solvent. Positive results on testing of phenolic compounds and tannins. In the Ponin test, pineapple leaf extract will give positive results if foam forms that lasts for 5 minutes. So the results were positive in the tannin senhyawa test. In the testing of Terponoids and Steroids, the pineapple leaf extract tested did not form brownish or violet rings. And in steroid testing, pineapple leaf extract also did not form a bluish-green color.

CONCLUSION AND SUGGESTIONS

Conclusion

Based on the results of the antioxidant activity test using the DPPH method on pineapple leaf methanol extract, it can be concluded that pineapple leaf methanol extract has a moderate antioxidant activity category based on the analysis results, an IC50 value of 113.16 ppm was obtained. The IC_{50} value is included in the medium level (in the range of 100-250).

Suggestion

Based on the research conducted, the author suggests that further research needs to be carried out using other parts of pineapple leaves or other plants that have the potential to be a good source of natural antioxidants, and also using other test methods such as FRAP *(Ferric Reducing Antioxidant Power)* and CUPRAC *(Cupric Ion Reducing Antioxidant Capacity)* so that a better antioxidant test method can be determined.

ACKNOWLEDGMENTS

There is no best offering that the author can give other than a sense of gratitude to those who have helped the author a lot.and thank you also to the Research Laboratory, Faculty of Pharmacy and Health Sciences, Abdurrab University for giving you the opportunity to be able to conduct research

http://doi.org/10.22216/jk.v5i2.5717
Published by LLDIKTI Wilayah X

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BIBLIOGRAPHY

Hamzah, A. F. A., Hamzah, M. H., Man, H. C., Jamali, N. S., Siajam, S. I., & Ismail, M. H. (2021). Recent updates on the conversion of pineapple waste (Ananas comosus) to value-added products, future perspectives and challenges. In *Agronomy* (Vol. 11, Issue 11). MDPI. https://doi.org/10.3390/agronomy11112221

Jamshidi-Kia, F., Wibowo, J. P., Elachouri, M., Masumi, R., Salehifard-Jouneghani, A., Abolhassanzadeh, Z., & Lorigooini, Z. (2020). Battle between plants as antioxidants with free radicals in human body. *Journal of HerbMed Pharmacology*, *9*(3), 191–199. https://doi.org/10.34172/jhp.2020.25

Jayawardena, T. U., Wang, L., Asanka Sanjeewa, K. K., In Kang, S., Lee, J. S., & Jeon, Y. J. (2020). Antioxidant potential of sulfated polysaccharides from padina boryana; protective effect against oxidative stress in in vitro and in vivo zebrafish model. *Marine Drugs*, *18*(4). https://doi.org/10.3390/md18040212

Juariah, S., Juliantari, E., Hanggara, D., Surya, A., & Endrini, S. (2024). Chemical Profiling, Antifungal and Anti-inflammatory Evaluations of Ethanol Extract of Zingiber officinale var. rubrum and Curcuma caesia Mixture from Riau, Sumatera Island, Indonesia. *Tropical Journal of Natural Product Research*, *8*(1), 5794–5800. https://doi.org/10.26538/tjnpr/v8i1.12

Ou, Q., Zhang, S., Fu, C., Yu, L., Xin, P., Gu, Z., Cao, Z., Wu, J., & Wang, Y. (2021). More natural more better: triple natural anti-oxidant puerarin/ferulic acid/polydopamine incorporated hydrogel for wound healing. *Journal of Nanobiotechnology*, *19*(1). https://doi.org/10.1186/s12951-021-00973-7

Phuyal, N., Jha, P. K., Raturi, P. P., & Rajbhandary, S. (2020). Total Phenolic, Flavonoid Contents, and Antioxidant Activities of Fruit, Seed, and Bark Extracts of Zanthoxylum armatum DC. *Scientific World Journal*, *2020*. https://doi.org/10.1155/2020/8780704

Sienkiewicz, N., Dominic, M., & Parameswaranpillai, J. (2022). Natural Fillers as Potential Modifying Agents for Epoxy Composition: A Review. In *Polymers* (Vol. 14, Issue 2). MDPI. https://doi.org/10.3390/polym14020265

Surya, A., & Marliza, H. (2020). *Comparison of Antioxican Tea Potential (Camellia Sinensis) between Green Tea and Black Tea in Datingradical 2 , 2 " Diphenyl-1-Picrylhydrazyl "(Dpph)*. *63*(6), 1179–1189.

Yeow, P. K., Wong, S. W., & Hadibarata, T. (2021). Removal of azo and anthraquinone dye by plant biomass as adsorbent – a review. In *Biointerface Research in Applied Chemistry* (Vol. 11, Issue 1, pp. 8218–8232). AMG Transcend Association. https://doi.org/10.33263/BRIAC111.82188232

Yunita, E., & Sari, D. R. A. P. (2022). Aktivitas Antioksidan dan Toksisitas Fraksi Etil Asetat dan Fraksi N-Heksan Daun Pegagan (Centella Asiatica L.). *Jurnal Mandala Pharmacon Indonesia*, *8*(1), 58–66. https://doi.org/10.35311/jmpi.v8i1.167

Jurnal Katalisator Vol 9 No. 1 (2024) 252-261

Zaiyar, Surya, A., & Marliza, H. (2020). Comparison of Antioxican Tea Potential (Camellia Sinensis) between Green Tea and Black Tea in Datingradical 2,2"Diphenyl-1-Picrylhydrazyl"(Dpph). *Solid State Technology*, *63*(6), 1179–1189. www.solidstatetechnology.us

http://doi.org/10.22216/jk.v5i2.5717
Published by LLDIKTI Wilayah X