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Antioxidant Activity Evaluation of Polar, Semi-Polar, and Non-Polar Fractions of Custard Apple (*Annona Squamosa* L.) Leaf Extract Using the DPPH Method

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Abstract: Antioxidants are compounds that can donate hydrogen atoms to free radicals so that they can stop the chain reaction and convert free radicals into a stable form. custard apple leaves are known to have various benefits, one of which is as an antioxidant. This study aims to determine the potential antioxidant activity contained in custard apple leaves (*Annona Squamosa* L) with polar, semi-polar and non-polar fractions through the DPPH method. The sample used in this study was custard apple leaf extract (*Annona Squamosa* L) extracted with 96% ethanol solvent, and phytochemical screening tests, fractionation, antioxidant activity tests using UV-Vis spectrophotometer and IC50 value calculation using Microsoft exel were carried out. The results of the sample phytochemical screening showed that there were four compounds, namely phenolics, flavonoids, tannins and alkaloids while the results of the antioxidant activity test had a very strong category with the IC50 value of ethyl acetate, n-hexane and aquadest fractions of (3.950, 4.651, and 11.009 ppm) and the activity results of the IC50 value of custard apple leaf ethanol extract with a strong category of 22.280 ppm.

Keywords: Custard apple leaves; DPPH; Vitamin C; Free radicals; Antioxidants.

Introduction

Free radicals are unstable and highly reactive molecules because they contain one or more unpaired electrons in their outer orbits, the number of free radicals if not stopped will cause various diseases such as premature aging, cancer, liver and other degenerative diseases (Li et al., 2025; H. Zhang et al., 2025).

Data from GLOBOCAN (Global Burden of Cancer) released by the WHO (World Health Organization) (2018) stated that the number of cases and deaths due to degenerative diseases (cancer) until 2018 was 18.1 million cases and 9.6 million deaths (Bahadori et al., 2025; Gibson & McAuley, 2025). Degenerative diseases are increasing as they are characterized by decreased physical activity, exercise, unhealthy lifestyle, diet, work environment factors, and stress levels (Li et al., 2025; Rubio-Senent et al., 2025). Antioxidants are compounds capable of donating hydrogen atoms to free radicals. Through this mechanism, antioxidants can terminate chain reactions and convert free radicals into more stable forms (Galano, 2025; Gulcin, 2025).

Free radicals are highly reactive molecules that can cause oxidative damage to cells and tissues. Excessive oxidative stress has been associated with the development of various chronic diseases. Therefore, antioxidants play an essential role in protecting the body from cellular damage. Synthetic antioxidants have been widely used due to their strong effectiveness. However, the long-term use of synthetic antioxidants may cause harmful side effects. Some synthetic antioxidants have been linked to toxic and carcinogenic effects. These concerns highlight the need for safer alternatives (Jain et al., 2025).

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Natural antioxidants derived from plants are considered a promising option. Plant-based antioxidants are generally associated with fewer adverse effects. They are also more acceptable for long-term consumption. The use of natural ingredients aligns with the growing demand for safer and sustainable health products. Developing natural antioxidants can reduce dependence on synthetic compounds (Elhag et al., 2025). Several medicinal plants have been identified as potential sources of natural antioxidants. Phytochemicals such as flavonoids and phenolic compounds contribute to antioxidant activity. Research on plant-based antioxidants continues to increase worldwide. Such studies aim to identify effective and safe antioxidant sources. According to (Suhag et al., 2025), natural antioxidants may serve as alternatives to synthetic antioxidants. Therefore, the development of natural antioxidant treatments is essential to minimize side effects and improve public health outcomes.

Custard apple (*Annona Squamosa* L.) is a plant that is widely used by local communities. Empirically, custard apple leaves have long been used to eradicate head lice. However, the broader medicinal potential of custard apple leaves is not widely recognized by the public (Ashraf et al., 2024; Sanshita et al., 2025). Limited awareness has restricted their utilization beyond traditional practices. Therefore, scientific research is needed to explore the potential health benefits of custard apple leaves. One promising application is their use as a natural antioxidant. Previous studies have reported that various parts of the custard apple plant contain diverse bioactive compounds. These compounds include alkaloids, flavonoids, glycosides, tannins, and phenolic substances (Fathallah et al., 2024; Mubeen et al., 2025). Such phytochemicals are known to exhibit antioxidant properties. Antioxidants play an important role in preventing oxidative stress-related diseases. Custard apple leaves have been reported to contain high levels of flavonoids. Flavonoids are recognized for their strong free radical scavenging activity (Gawade, 2025).

The high total flavonoid content indicates that custard apple leaves possess significant antioxidant potential. Flavonoids are known for their strong free radical scavenging activity. According to (Lele et al., 2025; S. Priyanka et al., 2024), custard apple leaves exhibit promising antioxidant properties. These findings suggest that custard apple leaves can be developed as a natural source of antioxidants. However, antioxidant activity may vary depending on the polarity of extracted compounds. Fractionation using solvents of different polarities allows the separation of diverse bioactive constituents. Therefore, evaluating antioxidant activity across polar, semi-polar, and nonpolar fractions is necessary (Kumar Saroj et al., 2025). The DPPH method is widely used to assess free radical scavenging activity. This method provides reliable and reproducible measurements of antioxidant capacity. Accordingly, this study was conducted to determine the antioxidant potential of custard apple leaf fractions using the DPPH assay (Alkhalidy et al., 2023; Shehata et al., 2021).

Method

The method in this study is a laboratory experimental method. The sample of this study used custard apple leaves (*Annona Squamosa* L) obtained from the Lombok area, West Nusa Tenggara, precisely in Sesaot Village, Narmada District (Aly et al., 2025). The leaves of custard apple are selected from the young leaves. The materials used in this study are DPPH powder, aquadest, n-hexane, ethyl acetate, 96% ethanol, Mg powder, concentrated HCl, dragendroff, FeCl3 1%, FeCl3 5%. The tools used in this study are blenders, 40 mesh sieves, analytical scales, maceration vessels, stirring rods, tissues, 1000 ml glass beaker, 500 ml Erlenmeyer, wathman filter paper, 1000 ml measuring cups, measuring gourds, aluminum foil, glass funnels, porcelain cups, rotary evaporators, split funnels, micro pipettes, wooden tongs, test tubes, uv-vis, cuvettes (Qi et al., 2025).

Plant Determination

The leaves of custard apple are determined to know the identity of the plant (Bapat et al., 2020). The determination was carried out at the Mataram State Islamic University Laboratory.

Custard Apple Leaf Extraction (Annona Squamosa L.)

The samples of custard apple leaves that have been picked are then washed and sorted wet, then sliced and then dried by drying in the sun and covered with a black cloth for 5 days. Dried custard apple leaves are sorted dry and mashed until a fine powder (simplicia) is obtained, after which simplicia are sifted with a mesh of size 40, then as many as 400 grams of custard apple powder are extracted macerated with 96% ethanol solvent as much as 4000 mL and stored in a maceration container with room temperature for 24 hours. Maceration was carried out for 3 days with 2 remacerations.

The extracts obtained are filtered with wathman No. 1 paper until phytrates are obtained. Furthermore, the extracts were separated by solvents with a rotary evaporator at a temperature of 50OC and concentrated using a water bath with a temperature of 50OC (Ahmad Shiekh et al., 2021; Kumar et al., 2021)

Phytochemical Screening

The phytochemical screening carried out in this study included the identification test of phenolic

compounds, flavonoids, tannins and alkaloid tests using color reagents (Ahmad Shiekh et al., 2021).

Liquid-Liquid Fractionation

A total of 10 grams of thick extract was dissolved with 60 ml of aquadest and 60 ml of n-hexane solvent was added to the separate funnel, then cornered until 2 layers were formed, the two layers were separated into different glass beaker. Furthermore, the results of the aquadest fractionation were put back into the separate funnel and 60 ml of ethyl acetate was added and cornered until 2 layers were formed, the two layers were separated into different glass beaker, and the results of 3 fractions have been obtained, namely the aquadest fraction, the n-hexane fraction and the ethyl acetate fraction. Then each fraction was concentrated with a waterbath at a temperature of 50OC (Ferreira et al., 2025).

Manufacture of DPPH 100 Ppm Master Solution

DPPH powder is weighed as much as 10 mg, then put it in a measuring gourd and dissolve it with 96% ethanol until the limit mark then shake the measuring gourd until homogeneous, after which a 100 ppm DPPH solution is obtained (Azizah et al., 2025)

Activity Testing
Antioxidant
Creation and measurement of blank curves

A pipette of 2 mL of 100 ppm DPPH solution is then put into the test tube and added 2 mL of 96% ethanol and then beaten until homogeneous, then measured its absorbance value at a wavelength of 517 nm. The results obtained will be obtained with the maximum wavelength and absorbance value of the DPPH standard solution of 100 ppm (Azizah et al., 2025)

Manufacture and measurement of vitamin C comparator solutions

A total of 2.5 mg of vitamin C is weighed and then dissolved with 96% ethanol, sufficient volume up to the mark line (concentration 100 ppm). Vitamin C raw parent solution is piped as much as 0.5ml, 1 ml, 1.5 ml, 2 ml, 2.5 ml, (to obtain a concentration of 2 ppm, 4 ppm, 6 ppm, 8 ppm and 10 ppm) add with solvent. Each solution was filled with 2 mL of water in a test tube covered with aluminum foil and 2 mL of 100 ppm DPPH solution was added. The solution is incubated for Operating Time, its absorbance was further measured at a maximum wavelength of 517 nm using Uv-Vis spectrophotometry .(Azizah et al., 2025)

Determining the operating time

The sample used is colored so that it can be known at what minute stability occurs. Operating time is indicated by the constant absorbance value obtained at the measurement of a given time range of 0-30 minutes (dos Santos et al., 2025)

Measurement of DPPH absorption after addition of fractions
Measurement of ethyl acetate fraction

The ethyl acetate fraction is made into a parent raw solution of 2.5 mg dissolved with 96% ethanol so that a 100 ppm raw solution is obtained. Extracted 500 µl, 1000 µl, 1500 µl, 2000 µl and 2500 µl so that concentrations of 2 ppm, 4 ppm, 6 ppm, 8 ppm, 10 ppm were obtained. Each concentration in the 2 ml pipette is then put into the test tube, then a 100 ppm 2 ml DPPH solution is added.

The solution is incubated during Operating Time and absorption measurements were carried out at a wavelength of 517 nm (Aryal et al., 2021).

Measurement of n-hexane fraction

The n-hexane fraction is made into a parent raw solution of 2.5 mg dissolved with 96% ethanol, so that a raw solution of 100 ppm is obtained. Extracted 500 µl, 1000 µl, 1500 µl, 2000 µl and 2500 µl so that concentrations of 2 ppm, 4 ppm, 6 ppm, 8 ppm, 10 ppm were obtained. Each concentration in the 2 ml pipette is then put into the test tube, then a 100 ppm 2 ml DPPH solution is added.

The solution is incubated during Operating Time and absorption measurements were carried out at a wavelength of 517 nm (Aryal et al., 2021).

Measurement of aquadest fraction

The aquadest fraction is made of a parent raw solution of 50.0 mg dissolved with 96% 50 ml ethanol in a measuring flask, so that a raw solution of 1000 ppm, 250 µl, 500 µl, 1000 µl, 1250 µl, 1500 µl is obtained so that a concentration of 5 ppm, 10 ppm, 20 ppm, 25 ppm, 30 ppm is obtained.

Each concentration is placed in a 2 ml pipette into the test tube and then added to a 100 ppm 2 ml DPPH solution. The solution is incubated during the operating time and absorption measurements are carried out at a wavelength of 517 nm (Aryal et al., 2021).

Manufacture of extract stock solutions and measurement of antioxidant activity

The thick extract is made into a parent raw solution of 50.0 mg dissolved with 96% ethanol as much as 50.0 ml in a measuring flask until the solution concentration of 1000 ppm is obtained. A solution of 250 µl, 500 µl, 1000 µl, 1250 µl and 1500 µl is obtained so that the concentration of 5 ppm, 10 ppm, 20 ppm, 25 ppm, 30 ppm is obtained.

Each concentration was placed in a pipette of 0.1 ml into the test tube and then added a 100 ppm dpph solution of 2.0 ml. The solution is incubated during.

Operating Time and measured absorption at a maximum wavelength of 517 nm (Aryal et al., 2021).

Data Analysis

Determination of antioxidant activity

The results of the vitamin C, extract and fraction test of custard apple leaves were calculated with the following formula:

$$\%\ Inhibisi = \{Abs\ kontrol - Abs\ sampel\}/\{Abs\ kontrol\} \times 100\%$$

Calculation of IC50 values

The IC50 value is an illustration of the concentration of the test solution that can ward off free radicals by 50%. The calculation of the IC50 value uses the formula:

$$Y = ax + b$$
$$Y = 50$$

Result and Discussion

Yield results of maceration and fractionation

Extraction is carried out with 96% ethanol solvent using the maceration method. The selection of 96% ethanol because the 96% ethanol solution is universal and capable of attracting polar and non-polar compounds (Prayitno et al., 2020). The reason for using the maceration method is because the method is simple, cheap and does not use heating so that it can minimize damage to compounds that are not heat-resistant. The use of a temperature of 50OC to avoid damage to compounds at the highest temperatures, especially in flavonoid compounds, there is a condensation process to remove solvents so that they do not affect the active substances (Prayitno et al., 2020).

Table 1. Yield % of Custard apple Extract and Leaf Fraction

Sample	Yield value %
extract	14%
ıyl acetate fraction	3,8%
N-hexane fraction	13,05%
Aquadest faction	5,5%

The fractionation method used is liquid-liquid extraction, Fractionation is a method of separating mixed components derived from macerated extracts. Fractionation is done to separate the main group of contents from one from the the other main groups based on polarity differences (Ahmad Shiekh et al., 2021; Maheshwaran et al., 2024). Fractionation was carried out successively with the solvents Aquadest (polar), n-hexane (non-polar) and ethyl acetate (semi-polar).

The polar compounds contained in the ethanol extract of custard apple leaves will be distributed into the aquadest solvent, while the semi-polar compounds will be distributed into the ethyl acetate solvent and the non-polar compounds will be distributed into the n-hexane solvent (dos Santos et al., 2025; Prayitno et al., 2020).

The randemen results in table 1 show the difference in randemen results due to polarity. The solvents to attract the compounds in custard apple leaves are different. The highest randemen for the fraction is found in the n-hexane fraction of 13.05% which is non-polar can attract compounds such as steroids, fats, phenyl propanoids. The aquadest fraction is 5.5% which attracts compounds such as flavanoid glycosides, polysaccharides that are polar in nature. The 3.8% ethyl acetate fraction is semipolar and can attract glycon, alkaloid and polyphenol compounds (Prayitno et al., 2020).

Phytochemical screening

Phytochemical screening is carried out with the aim of identifying the content of secondary metabolite compounds of a natural material. Phytochemical screening is a preliminary stage that can provide an idea of the content of certain compounds to be studied. The phytochemical screening method is carried out by color testing using a color reagent (Kumar et al., 2021; Maheshwaran et al., 2024).

Table 2. Results of Phytochemical Screening of Custard apple Leaf Ethanol Extract

Compound	Reagents	Result	Ket
Phenolic	FeCl3 5%	Color green	+
Flavonoids	Concentrated	Color orange + yellow	+
Tannins	Mg+HCl powder	Blackish green color	+
Alkaloids	FeCl3 1%	Orange Color	+

Based on table 2, the results of phytochemical screening of custard apple leaf ethanol extract show the presence of secondary metabolite compounds in the form of phenolics, flavonoids, tannins and alkaloids. In the phenolic compound test, the positive result of FeCl 3 reagent 5% was marked by green deposits, in the flavonoid test positive results were marked by orange and yellow deposits in the sample. Positive results in the tannin test were characterized by the formation of a blackish-green color after the addition of 1% FeCl3%, in the alkaloid test, orange deposits were formed in the sample after the addition of the dragendroff reagent.

Results of Antioxidant Activity Analysis
Measurement of vitamin C antioxidant activity

The positive control used in this study was vitamin C, the use of positive controls was shown to compare how strong the potential of synthetic antioxidants was with natural antioxidants in this study. The measurement of DPPH absorbance after the addition of vitamin C standard was carried out at a wavelength of 517 nm, with concentrations of 2 ppm, 4 ppm, 6 ppm, 8 ppm, and 10 ppm, and three repetitions were admitted in the absorption measurement (Azizah et al., 2025).

Table 3. Table of Percent DPPH Inhibition in Various Types Vitamin C concentration

Concentration (ppm)	Abs 1	Abs 2	Abs 3	Means	(100%) Inhibition	IC 50
0	0,00	0,00	0,00	0,00	0,00	
2	0,119	0,515	0,355	0,329	46,677	
6	0,118	0,359	0,353	0,276	55,267	6,152
4	0,116	0,355	0,352	0,274	55,591	
8	0,114	0,355	0,352	0,272	55,915	
10	0,112	0,355	0,351	0,271	56,077	
Bank DPPH			**0,617**	**0,617**	**0,617**	**0,00**

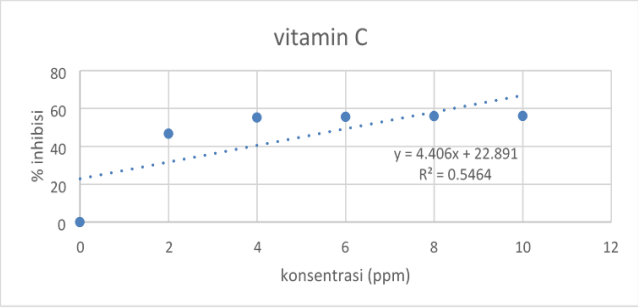

Figure 1. Vitamin C antioxidant activity curve

A compound is said to be a very strong antioxidant when the IC50 value is <50 ppm, strong when the IC50 value is between 50-100 ppm, it is said to be moderate when the IC50 value ranges from 100-150 ppm, and weak when the IC50 value ranges from 150-200 ppm. If the substance has an IC50 of more than 200 ppm, the substance is declared less active or very weak but still has the potential to be an antioxidant substance (Ashraf et al., 2024). The results of the vitamin C antioxidant activity test based on table 3 and figure 1 show that vitamin C has a very strong antioxidant activity with an IC50 value of 6.152 ppm.

Measurement of antioxidant activity Semi-polar fraction

The measurement of antioxidant activity in the three fractions was set at a wavelength of 517 nm which refers to a study conducted by (Ferreira et al., 2025). The measurement of antioxidant activity in fractions aims to determine the ability of the fractions of custard apple plant extract in warding off free radicals.

Table 4. Table of percent inhibition of DPPH in various Ethyl Acetate Faction Concentration

Concentration (ppm)	Abs 1	Abs 2	Abs 3	Average Absorbance	(100%) Inhibition	IC 50
0	0,00	0,00	0,00	0,00	0,00	
2	0,385	0,319	0,328	0, 344	44,246	
6	0,335	0,284	0,293	0,304	50,729	3,590
4	0,345	0,231	0,222	0,266	56,888	
8	0,331	0,224	0,221	0,258	58,184	
10	0,261	0,216	0,206	0,227	63,209	
Bank DPPH			**0,617**	**0,617**	**0,617**	**0,00**

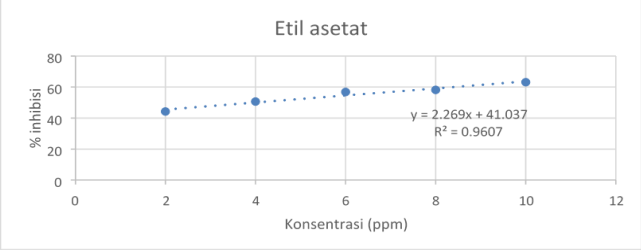

Figure 2. Antioxidant activity curve of ethyl acetate fraction

The graph shows a very strong IC50 value of 3.950 which means that the concentration of ethyl acetate solution is qualified to inhibit 50% of free radicals. compared to the results of the research conducted by (Kumar Saroj et al., 2025; Sanshita et al., 2025)that the results of IC50 of the ethyl acetate fraction were relatively strong with a value of 82.09 ppm. This shows that the comparison of the IC50 value of the ethyl acetate fraction in this study is different from the previous study. This is because it is influenced by the results of the total phenolic content in the sample. (total phenolics have an influence on antioxidant activity) (Ibrahim et al., 2025; M. Zhang et al., 2025).

Measurement of antioxidant activity of non-polar fractions

Antioxidant activity test in n-hexane fraction at concentration variations of 2 ppm, 4 ppm, 6 ppm, 8 ppm and 10 ppm (Huynh et al., 2026; Ibrahim et al., 2025). Based on the data from the measurement of the absorption of the n-hexane fraction with UV-Vis spectrophotometry and the absorption value of each of the test solutions has been obtained (Mustanir et al., 2021; Suhag et al., 2025).

Table 5. Table of percent inhibition of DPPH in various n-hexane fraction concentration

Concentration (ppm)	Abs 1	Abs 2	Abs 3	Average Absorbance	(100%) Inhibition	IC 50
0	0,00	0,00	0,00	0,00	0,00	
2	0,487	0,481	0,48	0,482	21,880	
6	0,323	0,302	0,28	0,301	51,215	4,651
4	0,311	0,17	0,166	0,215	65,153	
8	0,126	0,145	0,102	0,124	65,153	
10	0,130	0,102	0,100	0,110	82,171	
Bank DPPH			**0,617**	**0,617**	**0,617**	**0,00**

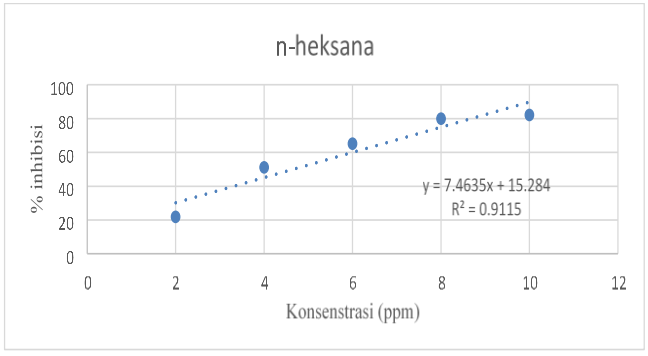

Figure 3. Antioxidant Activity Curve N-hexane Fraction

The graph shows that the IC50 value is very strong at 4.651 which means that the concentration of n-hexane is qualified to inhibit 50% of free radicals, compared to the results of the study conducted by (Elhag et al., 2025)that the results of IC50 of the n-hexane fraction were relatively strong with a value of 95.60 ppm. This shows that the comparison of the IC50 value of the n-hexane fraction in this study is different from the previous study, because it is influenced by the results of the total phenolic content in the sample. (total phenolics have an influence on antioxidant activity).

Measurement of the antioxidant activity of the polar fraction

Aquadest fraction antioxidant activity was tested at concentration variations of 5 ppm, 10 ppm, 15 ppm, 20 ppm and 25 ppm (Liza, 2021). Based on the data from the measurement of the absorbent of the aquadest fraction with UV-Vis spectrophotometry and the absorbance value of each of the test solution has been obtained, then the absorbance that has been known to be valued, a calculation is carried out to find the percent of inhibition or free radical capture activity in the aquadest fraction, which is calculated as the reduction in DPPH color (Galano, 2025; Gulcin, 2025). The results of the calculation and the percent of inhibition can be seen in the table below:

Table 6. Table of percent inhibition of DPPH in various Aquadest concentration

Concentration (ppm)	Abs 1	Abs 2	Abs 3	Average Absorbance	(100%) Inhibition	IC 50
0	0,00	0,00	0,00	0,00	0,00	
5	0,021	0,252	0,337	0,236	61,750	
10	0,108	0,189	0,332	0,209	66,126	11,009
20	0,108	0,151	0,322	0,197	68,071	
25	0,106	0,116	0,33	0,184	70,178	
30	0,105	0,104	0,232	0,147	76,175	
Bank DPPH			**0,617**	**0,617**	**0,617**	**0,00**

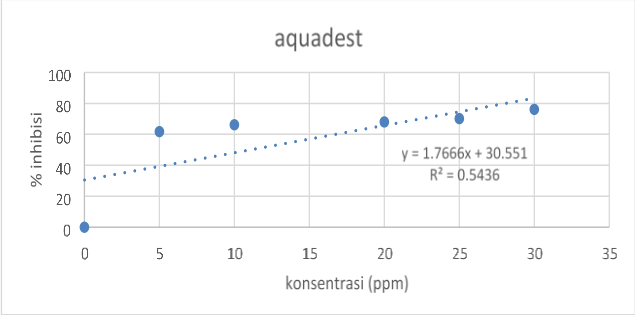

Figure 4. Antioxidant activity curve of Aquadest fraction solution

Measurement of the antioxidan t activity of the polar fraction

The graph shows a very strong IC50 value of 11,009 which is the concentration of the aquadest fraction needed to inhibit 50% of free radicals, compared to the results of the study conducted by (Kumar Saroj et al., 2025)that the results of IC50 of the aquadest fraction are relatively strong with a value of 60.77 ppm. This shows that the comparison of the IC50 value of the aquadest fraction in this study is different from the previous study, this is influenced by the results of the total phenolic content in the sample. (total phenolics have an influence on antioxidant activity) (M. Zhang et al., 2025).

According to the overall data, the results of the graph reinforce that the aquadest fraction has effective antioxidant potential at certain concentrations, where according to the resulting data, higher free radical inhibition effects are found at lower concentrations (Ibrahim et al., 2025)

Measurement of antioxidant activity of custard apple leaf ethanol extract

Measurement of antioxidant activity in custard apple leaf samples was carried out on ethanol extract with five concentration variations, namely 5 ppm, 10 ppm, 15 ppm, 20 ppm, and 25 ppm, at a wavespan of 517 nm in accordance with the research reference of (Lele et al., 2025; Qi et al., 2025)and three repeats of the measurement of antioxidant activity. The graph of the absorption measurement results on the custard apple leaf ethanol extract test solution is shown in Table 7 below:

Table 7. Table of percent inhibition of DPPH in various Ethanol Extract Concentration of Custard apple Leaves

Concentration (ppm)	Abs 1	Abs 2	Abs 3	Average Absorbance	(100%) Inhibition	IC 50
0	0,00	0,00	0,00	0,00	0,00	
5	0,530	0,530	0,551	0,357	12,965	
10	0,420	0,515	0,432	0,455	26,256	
20	0,333	0,412	0,332	0,359	41,815	22,280
25	0,225	0,313	0,281	0,273	55,753	
30	0,214	0,220	0,197	0,198	67,909	
Bank DPPH			**0,617**	**0,617**	**0,617**	**0,00**

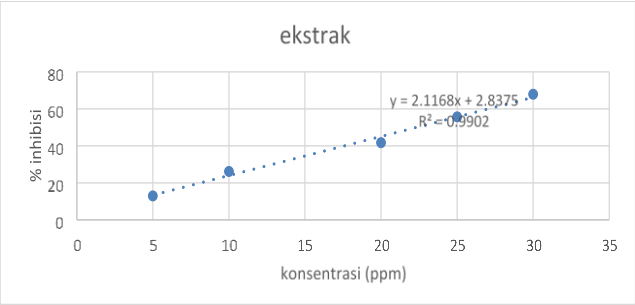

Figure 5. Antioxidant Activity Curve of Custard apple Leaf Extract

The graph shows that the IC50 value is very strong, which is 22,280 which is the concentration of the extract needed to inhibit 50% of free radicals. compared to the results of the research conducted by (Huynh et al., 2026; Tamane et al., 2025)that the results of IC50 of Chinese betel leaf ethanol extract are relatively strong with a value of 23.08514 ppm. This shows that the comparison of the IC50 extract values in this study is different from the previous study due to the difference in concentrations used in testing antioxidant activity (Khan et al., 2026; Leong et al., 2025). According to the overall data and the results of the graph reinforce that custard apple leaf extract with polar, semi-polar and non-polar fractions has effective antioxidant potential at certain concentrations, where according to the resulting data that higher free radical inhibition effects are found at lower concentrations (Prayitno et al., 2020).

Conclusion

Based on the results of the study, it was concluded that custard apple leaf ethanol extract has a strong antioxidant activity potential with an IC50 value of 22,280 ppm, while the IC50 value of ethyl acetate fraction is 3,950 ppm, n-hexane fraction has an IC50 of 4,651 ppm and the IC50 yield of aquadest fraction is 11,009 ppm. The IC50 value is not much different from the IC50 value of pure Vitamin C used as a comparison, which is 6.152 ppm.

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Conflicts of Interest
The authors declare no conflict of interest.

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