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Antioxidant Activity Test of Ethanol Extract of Feather Plantain Fruit (*Musa paradisiaca* L) with the DPPH Method (1,1- Diphenyl-2-Picrylhydrazyl)

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ABSTRACT

Antioxidants are compounds that can counteract or reduce the negative effects of free radicals. Antioxidants work by donating one electrons to compounds that are free radicals so that the activity of these compounds can be inhibited. This study aims to determine the activity and potential of antioxidants in the extract of banana feather fruit (Musa paradisiaca) which is taken from the village of Gunungcupu, Ciamis Regency. This feather plantain extract is obtained by maceration extraction using 70% ethanol. The antioxidant potential in this study was measured using the DPPH (1,1-diphenyl-2- picrylhydrazyl) method by Uv-Vis spectrophotometry. The antioxidant activity of feather plantain extract and vitamin C were used as positive controls at concentrations of 10 ppm, 20 ppm, 30 ppm, 40 ppm, 50 ppm and 60 ppm. Absorbance measurements using Uv-Vis spectrophotometry at a maximum wavelength of 520 nm. The plantain feather extract has an IC50 value of 59.10 ppm. While vitamin C has an IC50 value of 73.20 ppm. Based on the classification of antioxidants strength, feather plantain extract and vitamin C have strong antioxidant activity.

Keywords : Antioxidant, Musa paradisiaca, DPPH.

INTRODUCTION

Plantain fruit (musa paradisiaca) is one of them of the five most bananas consumed by Indonesian people. However, the antioxidant activity of these bananas and types Other local Indonesian bananas have not been studied much. Whereas These bananas may have useful antioxidant activity to reduce the negative impact of oxidation processes on the body man. Plantain is one of them A tropical fruit that grows a lot in Asia Southeast. Plantain is a banana has a high carotene content (PKBT, 2005), which where carotene is an antioxidant compound gives yellow color to banana skin (Nuramanah, 2012).

Plantain is a type of banana commercial which has a medium size and fat with a curved fruit shape and a slight fruit base round. Beta carotene has biological activity as an antioxidant which can reduce singlet oxygen, a reactive molecule formed from exposure to ultraviolet light on the skin, so that it can prevent the development of cancer cells. Singlet oxygene like other free radicals can trigger formation of a chain of subsequent free radical reactions (Roche, 2010). Free radical compounds arise as a result of various complex chemical processes in the body, in the form of by-products of the oxidation or cell burning process that takes place during breathing, cell metabolism, excessive exercise, inflammation or when the body is exposed to environmental pollution such as vehicle exhaust motorbikes, cigarette smoke, pollutants and solar radiation or cosmic radiation. Antioxidants can also is defined as a compound that when in low concentrations are present with the substrate that can oxidized, can delay or inhibit oxidation these compounds (Sunardi, 2007). Single electron transfer is strongly influenced by stability solvent at a certain charge (Ou, Huang et al, 2002).

Studies on antioxidants are tests based on the transfer of hydrogen atoms, including the Oxygen Radical Absorption Capacity (ORAC) test, there are also mixed tests, including the transfer of hydrogen atoms and electrons, including the 2,2'-Azinobis-(3-ethylbenzothiazoline-6- sulfonic acid) test. (ABTS), and [2,2-di(4-tert)-octylphenyl)-1-picrylhydrazyl] (DPPH) assay. The test uses spectrophotometry, based on changes in the color of the solution to be analyzed (Irina et al, 2021) and (Ramdan and Wulandari 2023). Antioxidant activity using the DPPH neutralization method is notated in EC50, as the antioxidant concentration required to reduce the DPPH concentration by 50% (Foti M.C, 2015). The DPPH method is a simple and accurate

method for antioxidant testing (Rahmah, Ramdan, and Lestari 2023). In the research that has been carried out on the test antioxidant ethanol extract of winged bean leaves (PsophocarpusTetragonolubus L) using the DPPH (1,1-Diphenyl-2-Picrylhydrazyl). The results of this research concluded that The ethanol extract of winged bean leaves has strong antioxidant activity (Nurhaida, 2022). Antioxidant activity tests using the DPPH method have also been carried out on kesum leaves (Polygo-num minus Huds.) and provided strong antioxidant activity results (Indah Purwaningsih, 2018). Research on ethanol, ethyl acetate and hexane extracts of white turi stems (Sesbania grandiflora (L.) Pers.) showed strong antioxidant activity (Jamilatur Rohmah et al, 2020)

TOOLS AND MATERIALS

The materials used in this research were banana fruit simplicia, 70% ethanol, Mg Metal DPPH, HCl, Vimain C p.a, H2SO4 2 N, distilled water, 2,6-dichlorophenol indophenol. The tools used are UV-Vis Spectrophotometer, analytical scales, laboratory glassware, macerator, water bath, hot plate, oven and blender.

METHODS

a. Extract preparation

Plantain fruit extract is made by mashing the pulp of the banana fruit, then weighing 300 grams and then putting it into the The maceration vessel is then moistened with 70% ethanol solvent. for 3x24 hours. The solvent is changed every 1x24 hours and carried outstir as often as possible. Then the filtrate is filtered using filter paper and concentrating at temperature 60^{o}C until a thick extract is obtained.

b. Phytochemical Screening

Flavonoid examination with concentrated HCL and Mg metal Weigh out 0.5 grams of the extract and put it in cup, add 2 ml of 70% ethanol then stir and Add 0.5 gram magnesium powder and 3 drops of HCL concentrated. Formation of orange to red color shows the presence of flavones, red to bright redshows flavanols, bright red to red purplish indicates flavanones (Farnsworth, 1966). Flavonoid Examination with H2SO4 2N A total of 1 ml of ethanol extract of plantain fruit put into a test tube, thenAdd 2 drops of 2N H2SO4

and shake strong. Positive samples contain flavonoids when in solution experienced a very striking color change turn yellow, red or brown (Munte et al, 2015).

c. Preparation of DPPH Solution

Weighed 10 mg DPPH dissolved in ethanol and put into a 100 ml measuring flask. Volume is sufficient with ethanol up to the limit mark, so that it is obtained DPPH solution with a concentration of 100 ppm. Then DPPH solution is placed in a dark container.

d. Preparation of Vitamin C Standard Solution

The mother liquor (1000 ppm) was prepared by weighing 50 mg of vitamin C powder and dissolved in 50 ml distilled water in a 50 ml volumetric flask.

e. Preparation of 2,6-Dichlorophenol

Indophenol Solution This reagent is made by dissolving 50 mg of Na salt 2,6-Dichlorophenol Indophenol that has been stored in a desiccator in 200 ml of added water 50 mg sodium bicarbonate, then shake vigorously. Then from the mother liquor a concentration series of 10 is made, 20, 30, 40,50 and 60 ppm. To make each concentration was pipetted at 0.1 ml, 0.2 ml, 0.3 ml, 0.4 ml, 0.5 ml and 0.6 ml of mother liquor into a 10 ml volumetric flask and adjust with distilled water to 10 ml. Drizzle with solution 2,6-dichlorophenol indophenol until color changes.

f. Absorption measurement using

UV-Vis spectrophotometer Each concentration of the vitamin comparison solution 2 ml of C was put into a test tube. Add 2 ml of DPPH solution. Next, incubate indoors. Then The absorbance was measured using a blank form Aquadest.

g. Preparation of 70% ethanol extract test solution

Stock solution (1000 ppm) was prepared by weighing 50 mg banana fruit extract and dissolved in 50 ml of ethanol in a 50 ml volumetric flask. From stock solution concentration series of 10, 20, 30, 40, 50 and 60 ppm was made.

h. Absorption measurement using UV-Vis spectrophotometer.

Then transfer 2 ml of each test solution into a test tube. Add 2 ml of DPPH solution, cover with aluminum foil. Next, incubate indoors dark. Then the absorbance was measured using a blank in the form of ethanol.

i. Determination of IC50

IC50 (Inhibitory Concentration) is the concentration causing a 50% loss of DPPH activity. For Calculating the IC50 value requires percent inhibition data which can be calculated using the formula as following:

RESULTS

a. Screening Phytochemical result

Table 1. Screening flavonoid result

Analyte	Reagent	Result	Confirmation
Flavonoid	Mg powder, HCL concentrate	+	Orange
	H_2SO_4 2N	+	Yellow

b. Percent Inhibition of Vitamin C

$$\% \text{ Inhibition} = \left(\frac{Absorbance\ of\ blank - Absorbance\ of\ Sample}{Absorbance\ of\ blank}\right) x100\%$$

Table 2. Result of inhibition activity of vitamin C

Concentration (ppm)	Absorbance	Average of absorbance	% Inhibition
10	0,322	0,324	11,47
	0,327		
	0,324		
20	0,311	0,311	15,02
	0,312		
	0,311		
30	0,302	0,288	21,31
	0,281		
	0,283		
40	0,256	0,256	30,05
	0,252		
	0,261		
50	0,238	0,238	34,97
	0,237		
	0,240		
60	0,214	0,212	42,07
	0,209		
	0,215		

c. Linear standard curve of vitamin C

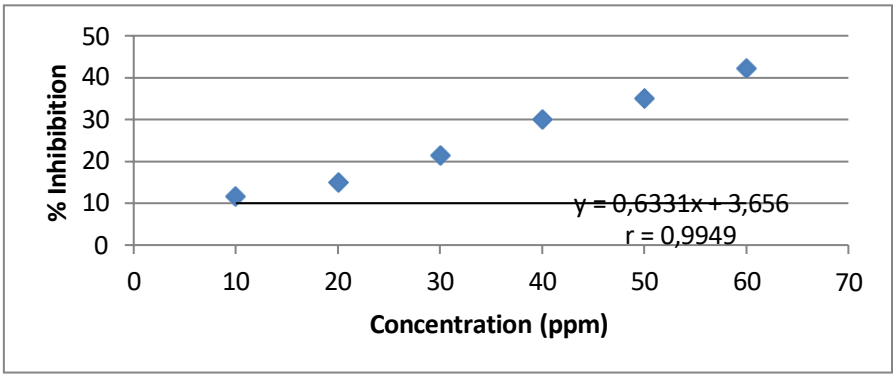

Figure 1. Linear Curve of vitamin C

$Y = bx + a$

$50 = 0{,}6331\ x + 3{,}656 \longrightarrow x = IC_{50} = 73{,}20$ ppm

d. The result antoxidant activity of banana fruite extract

Table 3. Inhibition activity of banan extract

Concentration (ppm)	Absorbance	Average of absorbance	% Inhibition
10	0,176	0,174	52,45
	0,174		
	0,174		
20	0,171	0,168	54,09
	0,167		
	0,167		
30	0,134	0,133	63,66
	0,133		
	0,133		
40	0,113	0,113	69,12
	0,114		
	0,114		
50	0,066	0,067	81,69
	0,068		
	0,068		
60	0,048	0,047	87,15
	0,048		
	0,045		

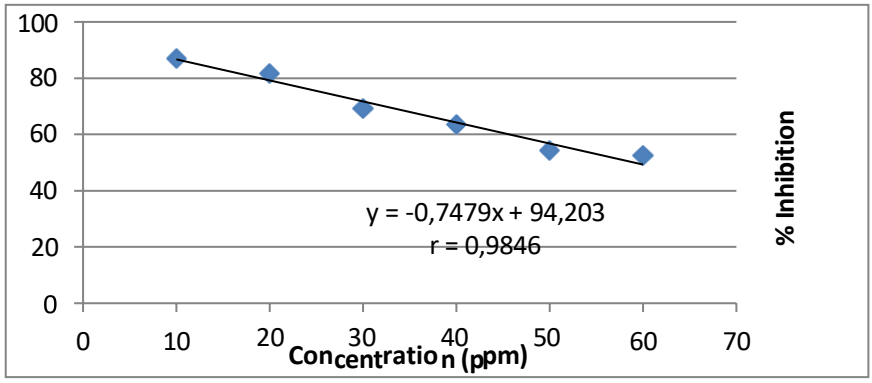

Figure 2. Linear Curve of Concentration and % Inhibition in Banana Fruit Extract

Y = bx + a

50 = 0,7479 x + 94,203 ⟶ x = IC_{50} = 59,10 ppm

DISCUSSION

The results of phytochemical screening carried out on the ethanol extract of plantain showed the presence of flavonoid compounds with the color changing from yellow to orange when added with Mg powder and concentrated HCL and changing color after being dripped with 2N H2SO4.

Based on calculations using the standard curve linear equation for pure Vitamin C, an IC50 value of 73.20 ppm was obtained. The y coefficient in the linear equation with a value of 50 is the IC50 coefficient, while the x coefficient in this linear equation is the concentration of % inhibition. The IC50 (Inhibitory Concentration) value describes the level of antioxidant power based on free radical inhibition of 50%. Based on the antioxidant strength classification, Vitamin C has strong antioxidant activity. Furthermore, the activity in the plantain fruit extract sample was obtained through a linear equation that the IC50 value was 59.10 ppm. The y coefficient in the linear equation with a value of 50 is the IC50 coefficient, while the x coefficient in this linear equation is the concentration of % inhibition. The IC50 (Inhibitory Concentration) value describes the level of antioxidant power based on free radical inhibition of 50%. Based on the classification of antioxidant power, plantain fruit extract has strong antioxidant activity.

Based on the results of determining the antioxidant activity of pure vitamin C and plantain fur extract, both have strong antioxidant activity. The presence of flavonoid compounds in the

ethanol extract of plantain bulu fruit causes the extract to have antioxidant activity. This is because flavonoid compounds are a class of polyphenolic compounds which have many hydroxyl (OH) groups. These hydrogen and hydroxy atoms can be donated to radical compounds so that the compounds can be stabilized.

CONCLUSION

Results of antioxidant activity tests using the DPPH method on fruit ethanol extract Plantain has strong antioxidant activity with an IC50 value (Inhibitory Concentration) of 59.10 ppm.

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