PHYTOCHEMICAL SCREENING AND ANTIOXIDANT ACTIVITY OF UNRIPE CANVEDISH AND DREAM BANANA (*Musa* sp.) FRUITS PEELS

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Abstract

Banana fruit which belongs to the family *Musaceae* is one of the most widely distributed and consumed fruit in the world especially in tropical and subtropical countries. People only consume the banana's pulp and dump its peel as solid waste. This gives a very serious agricultural waste disposal and eventually cause environmental problems that keep worsen each day. Thus, this study concern on the phytochemical content and the antioxidant in peel extracts of unripe Cavendish and Dream banana. The dried peels of banana fruit were grinded and extracted by using ethanol as the extraction solvent. The percentage yield of the ethanol extract of unripe Cavendish and Dream banana fruit peels were 17.765 % and 17.081 % respectively. The banana peel extracts were screened for the presence of bioactive compounds which showed the presence of phenols, flavonoids and tannins. Antioxidant activity of the banana peel extracts was evaluated using 1,1-diphenyl-2-picrylhydrazyl (DPPH) free radical scavenging assay. The concentration of the peel extracts required for 50 %

inhibition of DPPH radical scavenging activity (IC_{50}) were recorded at 90.28 µg/ml for Cavendish and 113.09 µg/ml for Dream banana. The extracts of both banana peels definitely showed potential as a sources of natural antioxidant.

Keywords: Cavendish banana fruit peel, Dream banana fruit peel, antioxidant, (DPPH) free radical scavenging assay, phytochemical screenings.

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Introduction

The most widely distributed and consumed fruit in the world especially in tropical and subtropical countries which is Banana fruit belongs to the family of *Musaceae*. Nowadays, banana is among the world's leading fruit crops because it in terms of economic value, it is the fifth agricultural crop in the world trade (Baskar *et al.*, 2011). Apart from that, banana is originated from the tropical region of Southern Asia including countries like Malaysia, Philippines and Indonesia (Anhwange *et al.*, 2009).

Banana is used fresh or processed to manufacture many commercial products including banana chips, jams, juice, biscuits and more. In order to make those kinds of products, people usually used banana pulp instead of its peel. Thus, most of the peels are mostly dump as solid waste at large expense because they are not being used for any other commercial application. In fact, there are about 40% of the total weight of fresh banana is generated as waste product in industries producing banana based products (Nagarajaiah and Prakash, 2011). It leads to the cause of serious environmental problems. The banana peel must be utilized and exploit as a natural, eco-friendly and economic source of valuable components which can benefits to human being in order to solve the environmental problems.

Banana peel was found to be a potential source of antioxidant and antimicrobial activities (Mokbel *et al.*, 2005). The potential applications of banana peels depend on its chemical composition as different fruit parts contain different antioxidant and antimicrobial components. Unripe banana peels contain copper, zinc, sodium, potassium, calcium, phosphorus, and iron. (Selema and Farago 1996). Apart from that, banana peel is rich in nutrients such as dietary fibre (50 % on a dry matter basis), essential amino acids, proteins (7 % dry matter basis), polyunsaturated fatty acids, and potassium (Gonzalez-Monteleongo *et al.*, 2010) The finding from Sundaram *et al.*, 2011 suggest that the unripe banana peel sample had higher antioxidant potency than ripe and leaky ripe. Therefore, in this research phytochemical analysis tests on ethanol extract of unripe Cavendish (*Musa acuminata L, cvcavendshii*) and Dream banana (*Musa acuminatacolla. AAA cv 'Berangan'*) fruit peels had been carried out and the antioxidant properties of two different cultivars of unripe banana fruit peels investigated.

Material and methods

Collection of plant materials

The unripe banana peels of Cavendish and Dream variety were purchased fresh market of Pasar Orang Kaya, Kepala Batas, Pulau Pinang, Malaysia

Sample Preparation

The unripe Dream and Cavendish banana were peeled off and sliced into smaller pieces then dried in an oven at 60°C until all the water molecules evaporate. The dried banana peels were then ground into powder by using Waring electronic blender at 18, 000 rpm for 10 minute. Next, 40.0 g of each powdered banana peels were mixed with 400 ml of ethanol solvent. Then, the mixtures were inserted into an orbital shaker and allowed to stand for 48 hours. After that, the mixtures were filtered using Whatman Filter paper no. 1. The filtrates were evaporated under reduced pressure by using rotatory evaporator at the temperature ranging between 35 °C to 50 °C in order to obtain the crude extract of the plant materials. Lastly, the crude extract of both Cavendish and Dream banana fruit peel were kept in the refrigerator until required for use.

Phytochemical screenings

The ethanol extract of unripe Cavendish (*Musa acuminata L, cv cavendshii*) and Dream (*Musa acuminata colla. AAA cv* 'Berangan') banana peels were tested by using specific reagents in order to detect the presence of bioactive compounds such as flavonoids, phenols, tannins and saponins.

Flavonoid test

Sample crude extract was mixed separately with 10 ml of ethyl acetate and warmed for 3 minutes. Then, the mixtures were filtered and 4 ml of the filtrate were mixed with 1 ml of dilute ammonia. The mixtures were shaken vigorously. The presence of flavonoids was indicated by the formation of yellow coloration of the entire mixture (Kham *et al.*, 2011).

Phenol test

For phytochemical screening test of phenols, 0.5 ml of Folin-cicoalteau reagent and 2.0 ml of 20 % Na₂CO₃ were added to 2.0 ml of ethanol extract of banana peels. The formation of blue coloration of the mixture was used as the indicator for the presence of phenols (Sumathy *et al.*, 2011).

Tannin test

Approximately 0.5 g of crude extract was dissolved in 20 ml of distilled water and filtered. Then, 2.0 ml of 2 % ferric chloride (FeCl₃) solution were added to the filtrate. Dark blue or dirty green coloration was used as the indicator for the presence of tannins (Ehiowenwenguan *et al.*, 2014).

Saponin test

About 0.5 g of powdered banana peels were mixed with 5 ml of distilled water. The mixture was then shaking vigorously. The formation of stable persistent froth indicated the presence of saponins (Sumathy *et al.*, 2011).

Antioxidant activity

Preparation of stock solution

100 mg of crude extract of Cavendish and Dream banana fruit peels were dissolved in 100 ml of ethanol in order to prepare 1.0 mg/ml of stock solution. Five different concentration of stock solutions were then prepared via dilution technique consisting of 0.04 mg/ml, 0.08 mg/ml, 0.12 mg/ml, 0.16 mg/ml and 0.20 mg/ml respectively.

Preparation of standard solution

100 mg of ascorbic acid were added to 100 ml of ethanol in order to prepare a 1.0 mg/ml standard solution. The standard solution was then diluted into five concentrations which were 0.04 mg/ml, 0.08 mg/ml, 0.12 mg/ml, 0.16 mg/ml and 0.2 mg/ml respectively.

DPPH free radical scavenging activity assay

Starting with the first concentration of 0.04 mg/ml, 2 ml of the 0.04 mg/ml stock solution were mixed with 1 ml of 0.1 mg/ml DPPH solution in a cuvette. The test solution and the standard solution were then incubated in dark place for 30 minutes at 24 °c. The absorbance was measured after 30 minutes incubation at 517 nm by using UV-6100 Split Beam UV-Vis spectrophotometer. The steps above were repeated thrice for each concentration of stock and standard solution. The percentage of inhibition of the sample was determined by comparing it with the standard antioxidant which was the ascorbic acid. The concentration of the banana peels extract (sample) required to give 50 % of optical density were represented as IC_{50} value. In addition, ethanol was used as blank to calibrate the UV-Vis spectrophotometer. The DPPH radical scavenging activity was calculated by using the formula as follows:-

Radical scavenging activity =
$$\begin{bmatrix} A_{control} - A_{sample} \\ A_{control} \end{bmatrix} \times 100 \%$$

A = Absorbance
 $A_{control}A_{control}$ = DPPH solution + methanol
 $A_{sample 1}A_{sample 1}$ = DPPH solution + unripe banana peels extract
 $A_{sample 2}A_{sample 2}$ = DPPH solution + standard solution (ascorbic acid)

Result and Discussion

The powdered banana fruit peel of Dream and Cavendish was extracted by using ethanol solvent and the percentage yields were measured.

Table 1.	Mass	of crude	extract and	Percentage	Yield

Banana peel cultivars	Crude mass (g)	Percentage Yield (%)
Dream	7.1061	17.765
Cavendish	6.8324	17.081

Number of replicate = 3

According to Table 1, the percentage yield of Dream cv. banana fruit peel crude extract was slightly higher (17.765 %) as compared to Cavendish cv. In previous study by Nagarajaiah *et. al.*, 2001, it stated that the usage of ethanol as extraction solvent will give higher percentage yield of ethanol extract of banana fruit peel which in ranged from 11 to 18% for dry powder compared to methanol and aqueous solution extract. It can be said that the percentage yield for both Dream and Cavendish

banana fruit peel obtained from the experiment were relevant as the percentage yield were located within the range given in the previous study.

Phytochemical analysis of ethanol extract of unripe Cavendish and Dream banana fruit peel was done in order to identify the presence of bioactive compounds such as flavonoids, phenols, tannins and saponins. Table 2 showed both banana fruit peels contained with flavonoids, phenols and tannins.

Bioactive	Crude extract of unripe banana fruit peels		
Compounds	Cavendish	Dream	
Flavonoids	+	+	
Phenols	+	+	
Tannins	+	+	
Saponins	-	-	

Table 2.Bioactive compounds present in both ethanol extract

The antioxidant activity of unripe Dream and Cavendish banana fruit peels were investigated by using DPPH radical scavenging assay. The activities were measured based on the reduction at 517 nm absorbance (A) to scavenge the stable DPPH free radical. DPPH radical scavenging assay was used in this research as it helps to determine the scavenging potential of antioxidant extract based on its capability as hydrogen donator and electron transfer. The reaction that occur between antioxidant compounds and the stable DPPH free radical will result in the reduction of absorbance and the discoloration of DPPH from deep purple (1,1-diphenyl-2-picrylhydrazyl) to light yellow (1,1-diphenyl-picrylhydrazine) (Abdullah *et. al.*, 2012).



Figure 1. Percentage of inhibition of ascorbic acid, Cavendish and Dream banana peel

By using the linear equation of y = 0.4683x - 2.959 for Dream banana peel, and y = 0.4558x + 8.852 for Cavendish banana peel, the value of IC_{50} can be calculated. Table 3 showed the percentage inhibition of DPPH radical scavenging and IC_{50} value of the ethanol extract of sample.

Samples	% Inhibition	(µg/ml)	
Ascorbic acid	98.17±1.65	2.04	
Cavendish banana fruit peel	95.17±2.39	90.28	
Dream banana fruit peel	84.51±1.47	113.09	

Table 3. The percentage inhibition and ^{IC}₅₀ values of the ethanol extract of samples

The lower the IC_{50} value, the greater the antioxidant activity of the sample. According to Fidrianny *et al.*, (2014), sample that had IC_{50} less than 50 µg/ml is a very strong antioxidant, sample that had IC_{50} in the range of 50 to 100 µg/ml is a strong antioxidant, whereas sample that had IC_{50} in the range of 101 to 150 µg/ml is a medium antioxidant and lastly sample that had IC_{50} more than 150 µg/ml is a weak antioxidant. The value obtained for Dream banana fruit peel is 113.09 µg/ml which indicate moderate antioxidant activity, whereas for Cavendish banana fruit peel the value is 90.28 µg/ml which indicate strong antioxidant activity.

Conclusion

In conclusion, the percentage yield of ethanol extract of unripe Dream banana fruit is slightly higher than percentage yield of Cavendish and the phytochemical screening showed the presence of bioactive componds in both the ethanol extract of unripe Dream and Cavendish banana fruit peel. Based on IC_{50} value, Cavendish banana fruit peel had a stronger antioxidant activity than Dream banana fruit

peel. Thus it can be concluded that this plant peels possess valuable antioxidant properties.

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