

Evaluation of banana (*Musa* sp.) flowers of selected varieties for their antioxidative and anti-hyperglycemic potentials

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Article history

Received: 10 August 2015
Received in revised form:
12 January 2016
Accepted: 28 January 2016

Keywords

Antioxidant activity
Phenolic compounds
Antihyperglycemic activity
Banana flower

Abstract

Consumption of banana flower as a vegetable is popular among many countries in Southeast Asia. In this study, banana flowers of six different Malaysian cultivars namely, pisang Abu (*Musa balbisiana* cv P. Abu), pisang Berangan (*Musa acuminata* cv P. Berangan), pisang Nipah (*Musa balbisiana* cv P. Nipah), pisang Susu (*Musa acuminata* cv P. Susu), pisang Mas (*Musa acuminata* cv P. Mas) and pisang Rastali (*Musa paradisiaca* cv P. Rastali) were investigated for their antioxidant and anti-hyperglycemic properties. The total poly phenolic content and antioxidant activities, the α -amylase and α -glucosidase inhibitory potentials of the banana flower extracts were studied in vitro using relevant assays. Among the six cultivars, cultivar Susu was found to have the highest phenolic content (80.13 ± 4.64 mg of GAE/g of extract) and displayed the highest ABTS+ and DPPH radical scavenging activities (24.73 ± 0.04 and 25.10 ± 0.15 μ mol of Trolox equivalent/g of extract). The anti-amylase and anti-glucosidase activity of the banana flowers extracts were in the range of 47.31-62.58% and 74.98-91.62%, respectively. All banana flower extracts inhibited the activity of α -glucosidase better than α -amylase at the concentration of 200 μ g/ml. This study concluded that the extracts of Malaysian banana flowers were potent sources of natural antioxidants, which can be used as postprandial hyperglycemia regulators.

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Introduction

Agricultural sector plays an important role in boosting the economy and GDP growth of Malaysia. Since Malaysia is located in the peninsula of the Southeast Asia, it is blessed with a conducive climate to nourish various types of tropical fruit crops such as papayas, bananas, guavas, pineapples and so on. Banana (*Musa* sp.) plant, belonging to the genus of *Musa* from the family of Musaceae, is a large, perennial monocotyledonous herb. It can grow from 2 to 4 meter in height at maturity. A host of different cultivars of banana have been cultivated in several parts of Malaysia. Based on the consumption pattern of people, banana has been ranked as the fourth most important fruit crop in Malaysia (Darvari *et al.*, 2010). According to Sulaiman and co-workers (2011), there are more than 20 edible banana cultivar types and most of them are derived from two wild species, known as *Musa acuminata* and *Musa balbisiana*.

Banana plant is not only a food crop but it is also known for medicinal uses. Apart from the banana fruit, other parts of the banana tree are known to have

medicinal properties beneficial to mankind. Because of the protective action against food spoilage, green banana leaves are commonly used in Malaysia and Indonesia for wrapping up of a popular breakfast food known as nasi lemak. The fiber extracted from the banana pseudostem has been used in different applications. The plant sap can be applied externally to sooth the pain of stings and bites. According to Kumar *et al.* (2012), roots are also known for medicinal applications. The flower of banana tree is also a popular vegetable among the people living in countries such as Malaysia, Philippines, Indonesia, and Si Lanka. It is usually red or purple red in color and attached to the end of the banana fruit bunch. In the red or purple red bracts, lots of small whitish flowers, which would turn into the mature edible banana fruit could be found. In Malaysia, banana flower is cooked to serve in preparing different types of cuisines. Apart from its food uses, banana flowers are also believed to possess some medicinal properties. In China, for instance, banana flower is traditionally used for certain illness such as heart pain, diarrhea, asthma and stomach cramps

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(Sumathy *et al.*, 2011). According to Kumar *et al.* (2012), it can be used for the treatment of bronchitis, dysentery, ulcers etc. A number of studies conducted in vivo and in vitro showed the health benefits of the consumption of banana flowers (Bhaskar *et al.*, 2011; China *et al.*, 2011). For instance, the consumption of cooked banana flower is believed to be beneficial to diabetic patients.

Over the years, several studies have been conducted on different aspects of Malaysian banana plant. Sulaiman *et al.* (2011) studied the antioxidant property and mineral contents of banana fruits of several cultivars. Shian *et al.* (2012) investigated the relationship between extracting solvents and antioxidant properties of three Malaysian banana cultivars namely, pisang Berangan, pisang Mas and pisang Raja. According to Tee and Hassan (2011), oral administration of both green and yellow banana peel extracts at different dose levels (200 and 400 mg/kg) can be used to treat depression. Fadhilah *et al.* (2014) reported that the cultivars of native banana namely pisang Berangan, pisang Mas and pisang Nipah had potent antibacterial activity against gram negative bacteria. In a subsequent study, Sumathy *et al.* (2011) also confirmed the antimicrobial properties of a Malaysian banana cultivar. However, there is hardly any study on the pharmacological properties of Malaysian banana flowers, particularly in relation to anti-hyperglycemic activity. Hence, the objective of this study was to compare banana flowers of locally available cultivars namely, pisang Abu (*Musa balbisiana* cv P. Abu), pisang Berangan (*Musa acuminata* cv P. Berangan), pisang Nipah (*Musa balbisiana* cv P. Nipah), pisang Susu (*Musa acuminata* cv P. Susu), pisang Mas (*Musa acuminata* cv P. Mas) and pisang Rastali (*Musa paradisiaca* cv P. Rastali) for their anti-oxidative capacities and carbohydrate hydrolyzing enzyme inhibitory activities.

Materials and Methods

Plant materials

Samples of banana flowers of six different cultivars namely, *Musa balbisiana* cv P. Abu (Abu), *Musa balbisiana* cv P. Nipah (Nipah), *Musa acuminata* cv P. Berangan (Berangan), *Musa acuminata* cv P. Mas (Mas), *Musa paradisiaca* cv P. Rastali (Rastali) and *Musa acuminata* cv P. Susu (Susu) were collected in triplicate from the banana plantation areas located at Johor. The collected samples were rinsed under running water to remove solid particles from the surface. The samples were then sliced and dehydrated at temperature between 40 - 45°C for one week using

an oven (Memmert 854, Schwabach, Germany). The dried samples were ground into fine powder using a grinder and then kept at 4°C for further analysis.

Chemicals and reagents

α -amylase and α -glucosidase were purchased from Sigma-Aldrich company. All chemicals used in the experiments were of analytical grade.

Banana flower extract preparation

A 100 g portion of dried banana flower was placed in a 1 L conical flask and a 500 ml portion of 80% ethanol was used as the solvent system for extraction. The conical flask was then placed in a orbital shaker to enhance the extraction efficiency through shaking. After two consecutive days of extraction, the mixture was filtered through Whatman No. 1 filter paper. The residue was re-extracted with same volume of 80% ethanol for at least two more times until the filtrate became colorless. The extract was concentrated under reduced pressure using a rotary evaporator (Büchi Model R-205 rotary evaporator, Switzerland) kept at 50°C water bath. The final weight of the extract was determined and the recovery percentage was calculated based on the formula below (Fatiha *et al.*, 2012).

$$\text{Yield of extract (\%)} = \frac{\text{Weight of extracts (g)}}{\text{Weight of dried material used (g)}} \times 100$$

Assay of α -glucosidase inhibition

α -glucosidase inhibitory activity was assayed according to Matsui *et al.* (2001) with little modification in a 96-well microplate. A 40 μ L portion of BFEs in 1 ml of 50 mM sodium acetate buffer of pH 6.0 was added to a reaction mixture of 30 μ L of 4 mM p-nitrophenyl- α -D-glucopyranoside and 30 μ L of 50 mU/ml α -glucosidase. The reaction was terminated by the addition of 50 μ L of 0.1 M Na₂CO₃ after 30 minutes of incubation at 37°C and the absorbance was measured at 405 nm. The inhibition of α -glucosidase in percentage was calculated using the relation;

$$\text{Inhibition (\%)} = [\text{Ac} - (\text{As} - \text{Ab})/\text{Ac}] \times 100$$

Where Ac = absorbance of control, Ab = absorbance of blank and As = absorbance of sample.

Assay of α -amylase inhibition

The α -amylase inhibition was performed according to the method of Bernfeld with little modification in a 96-well microplate (Premakumara *et al.*, 2013). The reaction mixture containing 50 μ L

portion of BFEs, 40 μL of starch (1% w/v) and 50 μL α -amylase (5 mg/ml) in a total reaction volume of 1 ml of 100 mM sodium acetate buffer (pH 6.0) was incubated at 40°C for 15 min. A 0.5 mL portion of DNS was then added and the reaction was subjected to further incubation for 5 min in a boiling water bath. It was then cooled in a cold water bath and the absorbance was recorded at 540 nm.

The percentage inhibition of α -amylase was calculated using the following formula.

$$\text{Inhibition (\%)} = [\text{Ac} - (\text{As} - \text{Ab}) / \text{Ac}] \times 100$$

Where Ac = absorbance of control, Ab = absorbance of blank and As = absorbance of sample.

Determination of total polyphenolic content (TPC)

The TPC was assayed by Folin-Ciocalteu reagent in a 96-well microplate (Singleton *et al.*, 1999). A 20 μL portion of the BFEs were mixed with 110 μL of freshly prepared Folin-Ciocalteu reagent. A 70 μL portion of 20% Na_2CO_3 was added and the mixture was incubated for 30 min at room temperature. Absorbance was measured at 765 nm. The results are expressed as mg of Gallic acid equivalent (GAE) per 1g of the dry material.

Determination of flavonoid content

Determination of flavonoid contents of samples was carried out according to the method described by Ebrahinzadeh *et al.* (2008). Generally, 1 ml portion of different BFE or standard solution was added to 4 ml of distilled water. A 300 μL portion of NaNO_2 (5% w/v) was added and incubated at room temperature for 5 min. Next, a 300 μL portion of AlCl_3 (10% w/v) was added. At 6th minute, 2 ml of 1.0 M of NaOH was added and the total reaction volume was made up to 10 ml with distilled water. The absorbance was read at 510 nm. The absorbance value of each sample was recorded in four replicates (n=4). Quercetin was used as the standard. A 100 mg portion of quercetin was measured and dissolved in 20 ml of methanol to make up of 5 mg/ml of stock solution. A series of dilution from stock solution was performed to make up 0.1 to 0.5 mg/ml of quercetin standard solution to generate the calibration curve. The flavonoids content of each sample was expressed as mg of Quercetin equivalent (QE) / g of extract.

ABTS assay

The radical ABTS was used to measure the radical scavenging activity of the BFEs (Re *et al.*, 1999). The stock of the ABTS radical cation was produced by reacting equal proportions of the 7.8 mM ABTS and

2.45 mM potassium persulfate at 37°C for 16 h in the dark to activate the ABTS. A working solution of the ABTS was prepared by diluting the stock to produce an absorbance of 0.7 ± 0.02 at 734 nm (Re *et al.*, 1999). A 40 μL portion of the ABTS working solution was reacted with 160 μL of the BFE and incubated for 10 min at 25°C after which the absorbance was recorded at 734 nm. The results were expressed as mmol trolox equivalent antioxidant power (TEAC).

DPPH assay

The DPPH radical scavenging assay was also performed in order to further ascertain the radical scavenging activities of the BFEs. A 160 μL portion of 125 μM DPPH radical dissolved in ethanol was mixed with 40 μL of the BFEs. The absorbance was measured at 517 nm after 15 minutes of incubation at room temperature. The results are expressed as mmol of trolox per 1 g crude bran (Blois, 1958).

FRAP assay

The FRAP assay was performed according to the method described by Benzie and Szeto (1999) with some modification. A 20 μL portion of BFEs was added to the mixture of 150 μL of the working FRAP reagent and 30 μL of acetate buffer. The whole reaction mixture allowed standing for 8 min and the absorbance was recorded at 600 nm. The results are expressed in mM FeSO_4 per gram of the dry bran. The FRAP reagent was prepared by mixing 300 mM acetate buffer (pH 3.6), 10 mM TPTZ solution and 20 mM $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$ in the ratio of 10:1:1 and subjected to heating up to 37°C just prior to use. The TPTZ solution was prepared by dissolving 10 mM TPTZ in 40 mM HCl (Premakumara *et al.*, 2013).

Statistical analysis

All measurements were carried out at least in triplicate data (n=3). All results were presented in the form of mean \pm standard deviation (SD). Data were analyzed statistically using one-way analysis of variance (ANOVA) with IBM SPSS software package (version 21.0). When F values were significant, mean differences were compared using Duncan's multiple range test at the 5% level of probability.

Results and Discussions

Total phenolic contents (TPC)

The TPC of the different BFEs determined by the Folin-Ciocalteu assay are presented in Table 1. The TPC values were found to be between 45.06 to 80.13 mg of GAE / g of extract and in the decreasing order of Susu > Mas > Rastali > Berangan > Abu

Table 1. The antioxidant properties of banana flowers of six different Malaysian cultivars

Banana cultivar	Phenolic content (PC)	Flavonoid content (FC)	ABTS radicals scavenging activity (ABTS)	DPPH radical scavenging activity (DPPH)	Ferric reducing antioxidant power (FRAP)
<i>Abu</i>	60.17±0.50 ^b	29.33±0.26 ^b	9.18±0.13 ^c	2.62±0.20 ^a	70.70±3.27 ^a
<i>Berangan</i>	61.77±0.77 ^b	34.65±3.23 ^c	15.55±0.09 ^e	25.10±0.14 ^c	336.00±22.30 ^d
<i>Nipah</i>	45.06±2.40 ^a	27.49±0.78 ^b	9.68±0.02 ^d	16.80±2.82 ^b	159.71±5.44 ^c
<i>Susu</i>	80.13±4.64 ^d	38.15±1.59 ^d	24.73±0.04 ^f	25.10±0.15 ^c	98.58±1.03 ^b
<i>Mas</i>	78.31±0.65 ^d	23.73±0.66 ^a	5.23±0.34 ^b	22.10±0.42 ^c	158.39±1.97 ^c
<i>Rastali</i>	68.65±1.03 ^c	51.12±1.60 ^e	3.72±0.11 ^a	25.10±0.14 ^c	404.59±6.05 ^e

Each value in the table represents mean±SD. Means within each column bearing different superscripts are significantly ($p<0.05$) different. The phenolic content was expressed as mg of GAE/g of extract (n=3); the flavonoid content was expressed as mg of QE/g of extract (n=4); the antioxidant capacity (ABTS & DPPH radical scavenging activity) was expressed as μ mole of Trolox/g of extract (n=3); the reducing power was expressed as μ mole of FeSO₄/g of extract (n=3).

> Nipah. When compared to the cultivars grown in India, banana flowers of cultivars used in this study showed relatively higher phenolic content. The observed difference in phenolic contents might be due to either varietal difference or use of different extracting solvents. According to China *et al.* (2011), the variation in TPC was quite general among different cultivars of banana grown in India. The highest phenolic content of Indian banana flower was noted for cultivar Kacha (11.94 ± 0.03 mg of GAE / g of extract) (China *et al.*, 2011). In a different study, Sulaiman *et al.* (2011) also observed the variation of the TPC of banana fruits of different Malaysian cultivars. According to Table 1, the highest total phenolic content was recorded for cultivar Susu (80.13 ± 4.65 mg of GAE / g of extract) while the lowest phenolic content was observed for cultivar Nipah (45.06 ± 2.40 mg of GAE / g of extract). Although the phenolic content of Mas (78.31 ± 0.65 mg of GAE / g of extract) was little lower than that of Susu, their differences were not significant ($p>0.05$). However, the phenolic contents of both Mas and Susu cultivars were significantly ($p<0.05$) higher than those of the other cultivars. Banana flowers of different cultivars might possess different levels of phenolic compounds due to a number of reasons. The high yield of crude extract might neither indicate the highest phenolic content nor antioxidant activity since several constituents with little or no antioxidant activity could be solubilized by aqueous based

solvents (Anwar *et al.*, 2013). For instance, cultivar Susu showed the lowest yield of crude extract, but it contained the highest amount of phenolics. However, Sulaiman *et al.* (2011) previously pointed out that the extracting solvents had significant influence on the phenolic concentration of crude plant extracts.

Flavonoids content

The total flavonoid contents of the different BFEs are presented in Table 1. Flavonoids are the most commonly distributed phenolic constituents of the plant kingdom. They are a group of low molecular weight phenolic compounds that are responsible for the aroma and antioxidant properties of food. Assessing the flavonoid contents of various food varieties is a common practice because of their radical scavenging abilities (Anwar *et al.*, 2013). In this study, the total flavonoid contents of different banana flowers were found to vary from 23.74 to 51.12 mg of quercetin equivalent [QE] / g of the extract. According to Table 1, the flavonoid content of cultivar Rastali was significantly higher ($p<0.05$) than those of any other cultivar with the mean value of 51.12 ± 1.60 mg of QE / g of extract. In contrast, the lowest flavonoid content was observed for cultivar Mas (23.73 ± 0.66 mg of QE / g of extract). The flavonoid contents of the rest of the banana cultivars were found to be within this range. Haytowitz *et al.* (2013) pointed out that the cultivar and growing conditions accounted for about 25 to 33% of the

Table 2. The antihyperglycemic properties of banana flowers of six different Malaysian cultivars

Banana cultivar	α -amylase activity inhibition (%)	α -glucosidase activity inhibition (%)
<i>Abu</i>	56.55±3.18 ^b	82.07±0.37 ^c
<i>Berangan</i>	62.58±2.32 ^c	76.58±0.25 ^b
<i>Nipah</i>	58.50±2.27 ^{b,c}	83.33±0.05 ^c
<i>Susu</i>	55.05±1.97 ^b	91.62±0.05 ^d
<i>Mas</i>	47.31±3.05 ^a	77.30±1.78 ^b
<i>Rastali</i>	55.91±1.49 ^b	74.98±0.15 ^a

Each value in the table represents the mean±SD, n=3. Means within each column bearing different superscripts are significantly (p<0.05) different. The inhibitory effect of the banana flower extracts was measured at the concentration of 200µg/mL

variability in the flavonoid content of foods. Other factors such as climatic stress, insect infestation, and agricultural practices may be accounted for the remaining variability. According to previous studies, flavonoids were well distributed in various parts of the banana tree. For instance, a monomeric flavonoid known as leucocyanidin was detected in banana fruit (Darsini *et al.*, 2012). Someya *et al.* (2002) found that the peel of the banana fruit was rich in gallic acid. According to some other phytochemical analyses, rutin existed as major bioactive component of banana leaves extract (Someya *et al.*, 2002) while rutin and quercetin were present in significant amounts in various other vegetables (China *et al.*, 2011).

ABTS⁺ radical scavenging activity

The results of the ABTS assay are presented in the Table 1. ABTS⁺ radical scavenging activity measurement is commonly used in the determination of the antioxidant capacity of a wide variety of food samples. As ABTS⁺ radical is soluble in both aqueous and organic solvents, it is a popular chromophore used in evaluating antioxidant activities of both hydrophilic and lipophilic antioxidants. The values of the ABTS radical scavenging activities of different BFEs were found to vary from 3.72 to 24.73 TEAC (mmol/g dry bran). The values were significantly (p<0.05) different and tended to follow the order of Susu > Berangan > Nipah > Abu > Mas > Rastali. The highest ABTS⁺ radical scavenging activity was observed for cultivar Susu with the mean value of 24.73 ± 0.03 µmole

of Trolox equivalent / g of extract while the lowest value was observed for cultivar Rastali (3.72 ± 0.09 µmole of Trolox equivalent / g of extract). According to China *et al.* (2011), flower of the Indian banana variety called Kacha was found to have the highest ABTS⁺ radical scavenging activity. The antioxidant capacity of plant extracts would also vary based on the type and amounts phytochemicals. The results of the present study showed that the antioxidant capacity was directly related to the TPC values of the extracts. Apart from cultivar differences, other factors such as maturity, environmental conditions, and extracting solvent might affect the antioxidant capacity (Hue *et al.*, 2012). For instance, Sulaiman *et al.* (2011) pointed out that the antioxidant activity of plant extracts would significantly be affected by the type of extracting solvent. Apart from these, climatic stress and agricultural practices may also influence the variability of antioxidant activities in plant based food products.

DPPH radical scavenging activity

The DPPH radical scavenging activities of the BFEs are shown in Table 1. DPPH radical scavenging assay is the most frequently used methods in *in vitro* antioxidant capacity studies (Alam *et al.*, 2013). These radicals are stable owing to their spare electron delocalization over the whole molecule (Irshad *et al.*, 1981). According to Table 1, the lowest value was observed for cultivar Abu with the mean values of 2.62 ± 0.18 µmole of Trolox equivalent / g of extract.

The observed values for cultivars namely, Nipah and Mas were 16.8 ± 2.83 and 22.1 ± 0.14 μmole of Trolox equivalent / g of extract, respectively. Cultivars Berangan, Susu and Rastali were found to display roughly similar values. Being recorded with the highest phenolic content and ABTS⁺ radical scavenging activity, cultivar Susu was found to show the highest DPPH radical scavenging activity as well. According to a previous report, an Indian cultivar known as Kacha was found to display the highest DPPH radical scavenging activities (China *et al.*, 2011).

FRAP reducing power

The results of the reducing power of the different BFEs are given in Table 1. The FRAP assay estimates the antioxidant power, which is the reducing ability of the substance involved in the transfer of electron in the reaction (Ediriweera and Ratnasooriya, 2009). Although FRAP assay was initially developed to measure the plasma antioxidant capacity, it is now widely used for evaluating the antioxidant capacity of food samples due to its simplicity, convenience and reproducibility (Pulido *et al.*, 2000). According to Table 1, BFEs of four cultivars, namely Abu, Berangan, Susu, and Rastali exhibited significant ($p < 0.05$) differences in their reducing powers except the cultivar Mas and Nipah, whose reducing power values were 158.39 ± 1.97 and 159.71 ± 5.44 μmole of FeSO_4 / g of extract, respectively. Cultivar Abu exhibited the lowest reducing power among all varieties (70.70 ± 2.67 equivalent to μmole of FeSO_4 / g of extract). On the other hand, cultivar Rastali showed the highest value (404.59 ± 4.94 equivalents to μmole of FeSO_4 / g of extract) that was about six times higher than that of cultivar Abu. According to some reports, high FRAP values observed in grains such as red bean were due to the high content of condensed tannins (Zou and Chang, 2014).

α -amylase inhibitory activity

The α -amylase inhibitory potentials of the different BFEs are compared as shown in Table 2. Inhibition of carbohydrate degrading enzymes is one of the approaches adapted to decrease the rate of glucose release into bloodstream. Previously, the inhibitory effect of traditional medicinal plant extracts on the α -amylase and α -glucosidase activities has been documented in a number of reports (Ashok Kumar *et al.*, 2011; Nair *et al.*, 2013). Among the medicinal plants of Malaysia, *Gynura procumbens* (Sambung Nyawa), *Ficus deltoidea* (Mas Cotek), *Cosmos caudatus* (Ulam Raja), *Phyllanthus niruri* (Dukung Anak), *Orthosiphon stamineus* (Misai

Kucing) and *Hibiscus rosa-sinensis* (Pokok Bunga Raya) were found to be useful for treatment of diabetes (Sekar *et al.*, 2014). According to Table 2, the highest α -amylase inhibition was observed for cultivar Berangan with the mean value of $62.58 \pm 2.32\%$, while the lowest α -amylase inhibition was observed for cultivar Mas with the mean value of $47.31 \pm 3.05\%$. The rest of the cultivars were in the order of Nipah > Abu > Rastali > Susu. However, there was no significant difference ($p > 0.05$) among the cultivars namely Abu, Rastali, Susu and Nipah on the α -amylase inhibition.

α -glucosidase inhibitory activity

The results of the α -glucosidase inhibitory potentials of the BFEs are shown in Table 2. Generally, inhibition of α -glucosidase activity might help to decrease the rate of absorption of glucose during digestion. According to Table 2, the percentage inhibitions of α -glucosidase by different varieties of banana flowers were tended to follow the order of Susu > Nipah > Abu > Mas > Berangan > Rastali. The variation of α -glucosidase inhibition was not exactly similar to the pattern of inhibition of α -amylase as noted before. The highest α -glucosidase inhibition was observed for cultivar Susu with the mean value of $91.62 \pm 0.05\%$, while the lowest α -glucosidase inhibition was observed for cultivar Rastali with the mean value of $74.98 \pm 0.15\%$. According to statistical analysis, the α -glucosidase inhibition exerted by cultivar Susu was significantly higher ($p < 0.05$) than those of any other banana cultivar in this study. However, there was no significant difference ($p > 0.05$) between cultivars namely, Berangan and Mas. As pointed out earlier, the TPC and free radical scavenging activities (ABTS⁺ and DPPH) of the cultivar Susu was the highest among the cultivars selected in this study (Table 1). These might contribute to the outstanding inhibitory effect on enzyme α -glucosidase. Previously, the strong correlation between antioxidant capacity and antihyperglycemic activity was reported in the ethanolic extract of *Peltophorum pterocarpum* (Manaharan *et al.*, 2012). Although cultivar Susu exhibited a lower inhibition of α -amylase activity, it exerted the highest inhibition for α -glucosidase activity. This could be due to differences in the types of bioactive compounds, modes of inhibition and affinity of inhibitor for the enzyme binding site.

By overall, the anti-amylase and anti-glucosidase activities of the Malaysian banana flowers were found in the range of 47.31 - 62.58% and 74.98 - 91.62%, respectively. Obviously, the inhibitory effect of BFEs on α -glucosidase was stronger than the α -amylase at

the concentration of 200 µg/ml. This finding was in accordance with the previous findings obtained in several types of teas and wines, which were found to exert a stronger inhibition effect on α -glucosidase (Kwon *et al.*, 2008). The anti-amylase and anti-glucosidase activities of the banana flowers could be attributed partly to their high flavonoid contents (23.73 - 51.12 mg of QE / g of extract). According to previous reports, the anti-diabetic potential of flavonoid such as quercetin, myricetin, and rutin have been confirmed through animal model studies (Hussain *et al.*, 2012; Kandasamy *et al.*, 2012).

Conclusion

The antihyperglycemic activities and antioxidant capacities of the banana flowers of six Malaysian cultivars were assessed in this study. Among the six cultivars, Susu was found to possess the highest phenolic content (80.13 ± 4.64 mg of GAE / g of extract) and the highest ABTS⁺ and DPPH free radical scavenging activities. This showed that a positive correlation existed between the phenolic contents and the free radical scavenging activity of the banana flowers. All six banana cultivars inhibited the α -glucosidase activity better than α -amylase activity at concentration of 200 µg/ml. Among them, the highest α -amylase inhibitory activity was shown by cultivar Berangan while the highest α -glucosidase inhibitory activity was displayed by cultivar Susu. By overall, Susu was the best candidate for antioxidant capacity and antihyperglycemic activity among the six Malaysian banana flowers and a potent source of food supplement for health benefits. Further studies on the isolation and characterization of individual antioxidant compounds of banana flowers will be carried out using different chromatographic techniques.

Acknowledgement

The authors acknowledge the financial support from a fundamental research grant (FRGS/2/2013/SG01/UPM/02/5) awarded by Ministry of Higher Education Malaysia.

References

- Anwar, F., Kalsoom, U., Sultana, B., Mushtaq, M., Mehmood, T. and Arshad, H. A. 2013. Effect of drying method and extraction solvent on the total phenolics and antioxidant activity of cauliflower (*Brassica oleracea* L.) extracts. *International Food Research Journal* 20: 653–659.
- Ashok Kumar, B. S., Lakshman, K., Nandeesh, R., Arun Kumar, P. A., Manoj, B., Kumar, V. and Sheshadri Shekar, D. 2011. *In vitro* alpha-amylase inhibition and *in vivo* antioxidant potential of *Amaranthus spinosus* in alloxan-induced oxidative stress in diabetic rats. *Saudi Journal of Biological Sciences* 18: 1–5.
- Benzie, I. F. F. and Szeto, Y. T. 1999. Total antioxidant capacity of tea by the ferric reducing/antioxidant power assay. *Journal of Agricultural and Food Chemistry* 47: 633–636.
- Bhaskar, J. J., Shobha, M. S., Sambaiah, K. and Salimath, P.V. 2011. Beneficial effects of banana (*Musa* sp. var. elakki bale) flower and pseudostem on hyperglycemia and advanced glycation end-products (AGEs) in streptozotocin-induced diabetic rats. *Journal of Physiology and Biochemistry* 67: 415–425.
- Blois, M. 1958. Antioxidant determinations by the use of a stable free radical. *Nature* 181: 1199–1200.
- China, R., Dutta, S., Sen, S., Chakrabarti, R., Bhowmik, D., Ghosh, S. and Dhar, P. 2011. *In vitro* antioxidant activity of different cultivars of banana flower (*Musa paradisiacus* L.) extracts available in India. *Journal of Food Science* 76: C1292–C1299.
- Darsini, D. T. P., Maheshu, V., Vishnupriya, M. and Sasikumar, J. M. 2012. *In vitro* antioxidant activity of banana (*Musa* spp. ABB cv. Pisang Awak). *Indian Journal of Biochemistry and Biophysics* 49: 124–129.
- Darvari, F. M., Sariah, M., Puad, M. P. and Maziah, M. 2010. Micropropagation of some Malaysian banana and plantain (*Musa* sp.) cultivars using male flowers. *African Journal of Biotechnology* 9: 2360–2366.
- Ebrahimzadeh, M. A., Pourmorad, F. and Bekhradnia, A. R. 2008. Iron chelating activity, phenol and flavonoid contents of some medicinal plants from Iran. *African Journal of Biotechnology* 7: 3188–3192.
- Ediriweera, E. and Ratnasooriya, W. 2009. A review on herbs used in treatment of diabetes mellitus by Sri Lankan ayurvedic and traditional physicians. *Ayurveda* 19: 593–601.
- Fadhilah, F., Jalani, M., Mohamad, S., Nazatul, W. and Shahidan, S. 2014. Antibacterial effects of banana pulp extracts based on different extraction methods against selected microorganisms. *Asian Journal of Biomedical and Pharmaceutical Sciences* 4: 14–19.
- Fatiha, B., Khodir, M., Farid, D., Tiziri, R., Karima, B., Sonia, O. and Mohamed, C. 2012. Optimisation of solvent extraction of antioxidants (phenolic compounds) from Algerian mint (*Mentha spicata* L.). *Pharmacognosy Communications* 2: 72–86.
- Haytowitz, D. B., Bhagwat, S. and Holden, J. M. 2013. Sources of variability in the flavonoid content of foods. *Procedia Food Science* 2: 46–51.
- Hue, S., Boyce, A. N. and Somasundram, C. 2012. Antioxidant activity, phenolic and flavonoid contents in the leaves of different varieties of sweet potato (*Ipomoea batatas*). *Australian Journal of Crop Science* 6: 375–380.
- Hussain, S. A., Ahmed, Z. A., Mahwi, T. O. and Aziz, T. A. 2012. Effect of quercetin on postprandial glucose excursion after mono- and disaccharides challenge in normal and diabetic rats. *Journal of Diabetes Mellitus*

- 2: 82–87.
- Irshad, M. and Sharma, C. B. 1981. Purification and properties of an S-amylase protein-inhibitor from *Arachis hypogaea* seeds. *Biochimica Biophysica Acta* 659: 326–333.
- Kandasamy, N. and Ashokkumar, N. 2012. Myricetin, a natural flavonoid, normalizes hyperglycemia in streptozotocin-cadmium-induced experimental diabetic nephrotoxic rats. *Biomedicine & Preventive Nutrition* 2: 246–251.
- Kumar, S., Kumar, V., Rana, M., and Kumar D. 2012. Enzyme inhibitors from plants: an alternate approach to treat diabetes. *Pharmacognacy Communications* 2: 18–33.
- Kwon, Y. I., Apostolidi, E. and Shetty, K. 2008. *In vitro* studies of eggplant (*Solanum melongena*) phenolics as inhibitors of key enzymes relevant for type 2 diabetes and hypertension. *Bioresource Technology* 99: 2981–2988.
- Manaharan, T., Palanisamy, U. D. and Ming, C. H. 2012. Tropical plant extracts as potential antihyperglycemic agents. *Molecules* 17: 5915–5923.
- Matsui, T., Ueda, T., Oki, T., Sugita, K., Terahara, N. and Matsumoto, K. 2001. α -glucosidase inhibitory action of natural acylated anthocyanins. 1. Survey of natural pigments with potent inhibitory activity. *Journal of Agricultural and Food Chemistry* 49: 1948–1951.
- Nair, S. S., Kavrekar, V. and Mishra, A. 2013. *In vitro* studies on alpha amylase and alpha glucosidase inhibitory activities of selected plant extracts. *European Journal of Experimental Biology* 3: 128–132.
- Premakumara G. A. S., Abeysekera W. K. S. M., Ratnasooriya, W. D., Chandrasekharan, N. V. and Bentota, A. P. 2013. Antioxidant, anti-amylase and anti-glycation potential of brans of some Sri Lankan traditional and improved rice (*Oryza sativa* L.) varieties. *Journal of Cereal Science* 58: 451–456.
- Pulido, R., Bravo, L. and Saura-Calixto F. 2000. Antioxidant activity of dietary polyphenols as determined by a modified ferric reducing/antioxidant power assay. *Journal of Agricultural and Food Chemistry* 48: 3396–3402.
- Re, R., Pellegrini, N., Proteggente, A., Pannala, A., Yang, M. and Rice-Evans C. 1999. Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radical Biology and Medicine* 26: 1231–1237.
- Sekar, M., Zulhilmi, M., Abdullah, B. I. N., Yasser, A., Bin, H., Azlan, N. O. R. and Syafiq, M. 2014. Ten commonly available medicinal plants in Malaysia used for the treatment of diabetes - A review. *Asian Journal of Pharmaceutical and Clinical Research* 7: 1–5.
- Shian, T. E., Abdullah, A., Musa, K. H., Maskat, M. Y. and Ghani, M. A. 2012. Antioxidant properties of three banana cultivars extracts. *Sains Malaysiana* 41: 319–324.
- Singleton, R. M., Orthofer, V. L. and Lamuela-Raventos, R. 1999. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. *Methods in Enzymology* 299: 152–178.
- Someya, S., Yoshiki, Y. and Okubo, K. 2002. Antioxidant compounds from bananas (*Musa cavendish*). *Food Chemistry* 79: 351–354.
- Sulaiman, S. F., Yusoff, N. A. M., Eldeen, I. M., Seow, E. M., Sajak, A. A. B. and Ooi, K. L. 2011. Correlation between total phenolic and mineral contents with antioxidant activity of eight Malaysian bananas (*Musa* sp.). *Journal of Food Composition and Analysis* 24: 1–10.
- Sumathy, V., Lachumy, S. J., Zakaria, Z. and Sasidharan, S. 2011. *In vitro* bioactivity and phytochemical screening of *Musa acuminata* flower. *Pharmacology online* 2, 127: 118–127.
- Tee, T. P. and Hassan, H. 2011. Anti-depressant-like activity of banana peel extract in mice. *American Medical Journal* 2: 59–64.
- Zou, Y. P. and Chang S. K. C. 2014. Antioxidant and anti-proliferative properties of extract and fractions from small red bean (*Phaseolus vulgaris* L.). *Journal of Food Nutrition* 1: 1-11.