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RESEARCH ARTICLE

Phytochemical screening and antioxidant activities of Methanol extracts from eight *Syzygium* species

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Abstract

This study was carried out to investigate the phytochemical screening of the methanol crude extracts of the leaves, twigs, and heartwoods of eight *Syzygium* species (*S. filiforme* var. *filiforme*, *S. papillosum*, *S. griffithii*, *S. pseudoformosum*, *S. claviflorum* var. *claviflorum*, *S. glaucum*, *S. syzygioides,* and *Eugenia* Sp. 57). The analysis revealed the presence of alkaloids, flavonoids, phenols, steroids, terpenes, and tannins except saponins in all plant extracts. Evaluation of the antioxidant activities were tested using DPPH, FRAP, and ABTS assays. All plant extracts showed good DPPH scavenging activity with the percentage inhibition more than 70 % at 100 ppm. The leaves and twigs of the methanol extracts of *S. griffithii* exhibited the most potent ferric ion reducer ranging from 1.03 ± 0.03 mM to 7.52 ± 0.49 mM and 1.43 ± 0.12 mM to 9.68 ± 0.38 mM FRAP equivalent, respectively. The heartwoods crude extract of *S. syzygioides* showed the most potent ferric ion reducer ranging from 1.20 ± 0.01 mM to 10.47 ± 0.12 mM FRAP equivalent. All of the leaves crude extracts showed potential antioxidant activities towards ABTS assay. The methanol extracts of the leaves of *S. glaucum* showed good SC_{50} value (116.90 mM) followed by *S. papillosum* (S2LM, SC_{50} value = 132.70 mM) *and Eugenia* Sp. 57 (S4LM, SC_{50} value = 143.10 mM). The twigs crude extracts of *S. syzygioides* (S8TM) and *S. griffithii* (S3TM) showed excellent antioxidant activity towards ABTS assay with the SC_{50} values of 130.60 mM and 145.40 mM, respectively. The crude extracts of *S. syzygioides* exhibited good antioxidant activity among the heartwoods extracts with the SC_{50} value of 96.20 mM.

Keywords: *Syzygium*, phytochemical screening, antioxidant, FRAP, ABTS, DPPH

INTRODUCTION

The Myrtaceae is in the major group of flowering plants which comprised about 100 genera and 3000 species throughout the tropics and subtropics especially in Australia. In Malaysia, Myrtaceae consists of about 9 genera and 210 species which are distributed from the lowland to mountain forests [1]. The leaves are simple, estipulate, opposite or alternate, and finely dotted with oil glands. The flowers are small to medium sized, regular, and bisexual [1]. All the Malaysia's species are evergreen but they flush new leaves and flowers seasonally. There are eight other genera in Myrtaceae family of Malaysia such as *Syzygium (Eugenia), Baeckea* (Chuchor atap), *Rhodomyrtus* (Kemunting), *Leptospermum* (China maki), *Melaleuca* (Gelam), *Tristania* (Pahlawan/Pelawan), *Rhodamnia* (Mempoyan), *Pseudoeugenia*, and *Decaspermum*. This family has commercial importance for its flower, edible fruits, oil-yielding glands, and timbers. *Syzygium* (kelat) is the largest genus in Malaysia and it occurs in all parts of the country from high tide level to the summit of Gunung Tahan. Kelat or Kayu kelat is the local name for most wild species which have small or inedible fruits and it refers to its peculiarly rigid bark [1]. Malaysia's *Syzygium* is an evergreen tree with pale green or pinkish leaves while their flowers are pollinated by flies, beetles, and butterflies [1]. The *Syzygium* plants have been reported to have many phenolic compounds such as alkaloids, flavonoids, anthocyanins, glucosides, ellagic acid, isoquercetin, kaempferol, and myrecetin which have been proven to have significant antioxidant activities [2]. Previous work on this genus reported significant bioactivities especially antimicrobial and antioxidant [3,4]. Traditionally, this genus is used for the treatment of numerous ailments such as fever, diarrhea, stomachache, dysentery, catarrh, diabetes, asthma, and as a stimulant for the nerves [4,5]. To the best of our knowledge, there is no report on phytochemicals and bioactivity studies towards these eight *Syzygium* species (*S. filiforme* var. *filiforme*, *S. papillosum*, *S. griffithii*, *S. pseudoformosum*, *S. claviflorum* var. *claviflorum*, *S. glaucum*, *S. syzygioides*, and *Eugenia* Sp. 57). Hence, the study aimed to analyze the phytochemical of methanol extracts from the aforementioned *Syzygium* species and their antioxidant activities against DPPH, FRAP, and ABTS assays.

EXPERIMENTAL

Plant materials

The leaves, twigs, and heartwoods of eight *Syzygium* species *S. filiforme* var. *filiforme* (SK2946/16), *S. papillosum* (SK2947/16), *S. griffithii* (SK2948/16), *S. pseudoformosum* (SK2951/16), *S. claviflorum* var. *claviflorum* (SK2950/16), *S. glaucum* (SK2952/16), *S. syzygioides* (SK2953/16), and *Eugenia* Sp. 57 (SK2949/16) were collected from Gunung Cakah, Taman Botani Negara Shah Alam, Selangor in August 2015. The plant samples were authenticated by Dr. Shamsul bin Khamis, a botanist from Forestry Department, Universiti Putra Malaysia. The specimens were deposited in the Biodiversity Unit, Herbarium, Institute of Bioscience, Universiti Putra Malaysia. All parts of the plants were dried for a few weeks.

Extraction method

The dried leaves, twigs, and heartwoods of *Syzygium* species were ground into powdered form and extracted using cold extraction method with MeOH solvent. The extracts were filtered and the solvent was removed under vacuum using a rotary evaporator to obtain the MeOH extracts.

Phytochemical screening

The MeOH extracts of the samples were analysed to identify the presence of flavonoids, alkaloids, glycosides, steroids, phenols, saponins, terpenoid, and tannins according to standard method [6,7].

Alkaloid test

0.3 g of MeOH crude extracts of each parts of *Syzygium* species were dissolved in 15 mL of ammoniacal chloroform and the obtained solutions were filtered. About five to ten drops of sulphuric acid (2M) were added to the filtrate and shake well, then the entire solution was allowed to stay for a while. Two layers were formed. An acidic upper layer and an organic lower layer were formed. The acidic layer was pipetted out into a test tube (5×10 mm) and two drops of Mayer reagent were added. Any changes in the turbidity of the solution were observed.

Flavonoid test

0.3 g of each crude extract was dissolved in MeOH and the solution was filtered into two test tubes (5×10 mm) labelled as A and B. Test tube A was referred as blank. A magnesium plate was added into test tube B followed by 0.5 mL of concentrated hydrochloric acid. Colour changes of the solution were observed. Appearance of reddish pink or dirty brown colour indicated the presence of flavonoid.

Steroid test

About 100 mg of dried MeOH crude extract was dissolved in 2 mL of chloroform. About two to three drops of sulphuric acid were carefully added to form a lower layer. A reddish-brown colour at the interface was an indicative of the presence of steroidal ring.

Saponin test

A drop of sodium bicarbonate was added in a test tube containing about 50 mL of the MeOH extract. The mixture was shaken vigorously and kept for 3 mins. A honey comb-like froth was formed, indicating the presence of saponins.

Phenol test

In a test tube containing 2 mL of MeOH crude extract, 2 mL of distilled water followed by a few drops of 10 % aqueous ferric chloride solution were added. Formation of blue or green colour indicated the presence of phenols.

Tannin test

A few drops of 1 % solution of ferric chloride was added to the test tube containing 5 mL of MeOH extract. The mixture was observed to note the formation of blue or green colour which indicated the presence of tannins.

Terpenoid test

2 mL of chloroform and 1 mL of concentrated sulphuric acid was added to the test tube containing 1 mg of MeOH extract. The solution was observed for reddish brown colour formation which indicated the presence of terpenoids.

Antioxidant activity

The antioxidant activity was determined using three assays, 1,1-Diphenyl-2-picrylhydrazyl (DPPH) free radical scavenging, ferric reducing antioxidant potential (FRAP), and 2,2'-azino-bis (3-ethylbenzthiazoline-6-sulphonic acid) (ABTS).

Solvents and chemicals

DPPH, ABTS, 6-hydroxy-2,5,7,8-tetramethylchroman-2-carboxylic acid (Trolox), butylated hydroxytoluene (BHT), and butylated hydroxyanisole (BHA) were obtained from Sigma-Aldrich. Analytical grade methanol, glacial acetic acid, dilute hydrochloric acid (HCl), and dimethyl sulfoxide (DMSO) were purchased from Merck. Sodium acetate trihydrate, ferric sulphate solution, 2,4,6-tripyridyl-s-triazine (TPTZ), and potassium persulfate were purchased from Fluka.

DPPH free radical scavenging assay

The free radical scavenging assay was conducted based on method described by Najihah *et al.* [8] with minor modification. Briefly, 100 µM DPPH (1 mL) dissolved in MeOH was added to the MeOH solution (3 mL) of the tested samples at different concentrations. An equal volume of MeOH was added in the control test. The mixture was shaken vigorously and allowed to stand at room temperature for 30 min. BHA and ascorbic acid (AA) were used as standard references. Then, the absorbance at 517 nm was measured with Epoch microplate reader. The percentage of scavenging of DPPH was calculated using the following equation:

$$S\% = [A_{\text{blank}} - A_{\text{sample}} / A_{\text{blank}}] \times 100$$

where A_{blank} is the absorbance value of the control reaction (containing all reagents except the test compound) and A_{sample} is the absorbance value of the test compound. The sample concentration providing 50 % scavenging (SC_{50}) was calculated by plotting scavenging percentages against concentrations of the sample. All tests were carried out in triplicate and scavenging values were reported as means (scavenging percentage).

FRAP assay

The FRAP assay was carried out according to Channarong *et al.* and Shahwar *et al.* with minor modification [9,10]. FRAP reagent was freshly prepared, consisted of stock solution with ratio 10:1:1 of 300 mM acetate buffer, 10 mM TPTZ in 40 mM HCl, and 20 mM $FeCl_3.6H_2O$ solutions. 5 µL of sample, 15 µL of MeOH, and 150 µL of FRAP reagent were added to the 96-well plates. The absorbance at 573 nm was measured after 10 min of incubation at 37 °C. $FeSO_4.7H_2O$ solution (0.1 mM–1.0 mM) was used to build up calibration curves of standard antioxidant.

ABTS assay

The ABTS assay was performed as described by Zou *et al.* [11] with minor modification. ABTS and potassium persulfate were dissolved with distilled water to obtain concentrations of 7 mM and 4.9 mM, respectively. The two solutions were mixed and incubate in the dark for 12–16 hours at room temperature. After incubation time, the ABTS radical was added with distilled water until the absorbance was 0.7 at 734 nm. 10 µL of sample and 190 µL of ABTS solutions were added to the 96-well plates. The absorbance was recorded after 30 minutes incubation in the dark at room temperature. The percentage of antioxidant activity was calculated using the following formula:

$$\text{Scavenging Percentage} = \frac{\text{Abs (ABTS)} - \text{Abs (ABTS+Sample)}}{\text{Abs (ABTS)}} \times 100$$

RESULTS AND DISCUSSION

Phytochemical screenings

The preliminary phytochemical screening of the MeOH crude extracts were conducted in order to determine the presence of secondary metabolites in all parts of eight *Syzygium* plants (*S. filiforme* var. *filiforme, S. papillosum, S. griffithii, S. pseudoformosum, S. claviflorum* var. *claviflorum, S. glaucum, S. syzygioides, and Eugenia* Sp. 57). Analysis of different parts of the plant extracts (leaves, twigs, and heartwoods) revealed the presence of alkaloids, flavonoids, phenols, steroids, terpenes, and tannins except saponins. The result of phytochemical analysis of eight Syzygium species is shown in Table 1 while Table 2 shows the observation scale for the phytochemicals analysis. The various phytochemical compounds detected are known to have beneficial importance in medicinal sciences such as flavonoids and alkaloids. Flavonoids and phenols are known to exhibit good bioactivities such as antioxidant, antifungal, cytotoxicity, and antibacterial in *Syzygium* species [12-32]. Alkaloids have been associated with medicinal uses for centuries owing to their bioactivities such as cytotoxicity, antibacterial, and antioxidant [33]. Plant steroids and terpenes are known for their importance as antimicrobial and insecticidal activities [19, 34]. Tannins are reported to exhibit antiviral, antibacterial, and anti tumor activities [34].

Table 1 Result of phytochemical analysis of eight *Syzygium* species.

TEST	OBSERVATION SCALES					
	Alkaloid	Flavonoid	Steroid	Phenol	Tannin	Terpenoid
Leaves						
S1LM	+1	+2	+1	+2	+2	+1
S2LM	+3	+2	0	+2	+2	0
S3LM	+1	+1	0	+2	+2	0
S4LM	+2	+1	0	+2	+2	0
S5LM	+2	+2	+1	+2	+2	+1
S6LM	+2	+2	0	+1	+1	0
S7LM	+1	+1	+1	+2	+2	+1
S8LM	+1	+1	0	+1	+1	0
Twigs						
S2TM	+3	+2	0	+2	+2	0
S3TM	+1	+2	0	+2	+2	0
S4TM	+2	+2	+1	+2	+2	+1
S5TM	+2	+2	+1	+2	+2	+1
S6TM	+3	+2	+2	+2	+2	+2
S7TM	+2	+2	+2	+2	+2	+2
S8TM	+3	+2	+1	+2	+2	+1
Heartwoods						
S1HM	+3	+1	0	+2	+2	0
S2HM	+2	+2	0	+2	+2	0
S3HM	+2	+2	+1	+2	+2	+1
S4HM	+2	+2	+1	+2	+2	+1
S5HM	+2	+2	0	+2	+2	0
S6HM	+3	+2	+1	+2	+2	+1
S7HM	+3	+2	0	+2	+2	0
S8HM	+3	+2	+1	+2	+2	+1

S1=*S. filiforme var. filiforme*; S2=*S. papillosum*; S3=*S. griffithii*; S4=*Eugenia* Sp. 57; S5=*S. claviflorum var. claviflorum*; S6=*S. pseudoformosum*; S7=*S. glaucum*; S8=*S. syzygioides*.

Table 2 Observation scale for phytochemical test [6-7].

Observations	Scale	Results
Alkaloid Test		
No changes	0	Negative
Transparent solution with faint trace of precipitate	+1	Positive
Mostly transparent solution with some precipitate	+2	Positive
Translucent solution with definite precipitate visible	+3	Positive
Immediate precipitate formation	+4	Positive
Large amount of precipitate formed immediately	+5	Positive
Flavonoid Test		
No changes	0	Negative
Formation of slightly brown or reddish pink colour	+1	Positive
Formation of dirty brown or reddish pink colour	+2	Positive
Steroid and Terpenoid Tests		
No changes	0	Negative
Thin layer of reddish brown colour at the interface	+1	Positive
Thick layer of reddish brown colour at the interface	+2	Positive
Saponin Test		
No changes	0	Negative
Formation of honey comb like froth	+1	Positive
Phenol and Tannin Tests		
No changes	0	Negative
Formation of slightly blue or green colour	+1	Positive
Formation of dark blue or green colour	+2	Positive

Antioxidant activities

Antioxidant assay was done to screen the antioxidant property of the MeOH crude extracts from the leaves, twigs and heartwoods of eight *Syzygium* species (*S. filiforme* var. *filiforme, S. papillosum, S. griffithii, S. pseudoformosum, S. claviflorum* var. *claviflorum, S. glaucum, S. syzygioides and Eugenia* Sp. 57). It can be measured using three different methods; DPPH, FRAP, and ABTS assays. The antioxidant activity cannot be evaluated by only a single method due to the complex nature of phytochemicals and the antioxidant activity determination is reaction-mechanism dependent [11]. Therefore, it is important to employ multiple assays to evaluate the antioxidant activity of plant extracts and pure compounds.

DPPH *assay*

In this study, ELISA microplate reader was used to determine the radical scavenging activity of the crude extracts of *Syzygium* species against free radical of DPPH. ELISA microplate reader measured the reduction in optical absorbance of each pure compound due to the scavenging of stable free radical DPPH at 517 nm [8]. BHA and AA were used as standard controls. Table 3 shows the percentage of scavenging activity at 100 ppm of the leaves, twigs, and heartwoods of eight *Syzygium* species. From the results, all of the plant extracts show good DPPH scavenging activity with the percentage inhibition more than 70 % at 100 ppm. Previous study on plant extracts of *S. cumini*, *S. paniculatum*, and *S. aromaticum* also exhibited strong DPPH scavenging activity [22,24,30].

Table 3 Scavenging percentage of eight *Syzygium* species.

Heartwoods	S (%)	Leaves	S (%)	Twigs	S (%)
S1HM	76.15	S1LM	95.55	-	-
S2HM	94.34	S2LM	76.55	S2TM	95.92
S3HM	96.22	S3LM	94.87	S3TM	89.06
S4HM	95.29	S4LM	87.71	S4TM	93.13
S5HM	94.85	S5LM	95.99	S5TM	95.10
S6HM	95.12	S6LM	95.88	S6TM	95.39
S7HM	90.73	S7LM	94.13	S7TM	87.39
S8HM	93.92	S8LM	95.44	S8TM	91.66
Standards					
BHA	84.25	-	-	-	-
AA	83.95	-	-	-	-

S1=*S. filiforme var. filiforme*; S2=*S. papillosum*; S3=*S. griffithii*; S4=*Eugenia* Sp. 57; S5=*S. claviflorum var. claviflorum*; S6=*S. pseudoformosum*; S7=*S. glaucum*; S8=*S. syzygioides;* BHA=Butylated hydroxyanisole; AA=Ascorbic acid;(*p < 0.05); Dash (-) means no activity.

FRAP assay

Determination of the ferric reducing antioxidant power towards the crude extracts of eight *Syzygium* species were tested in order to measure the reductive ability of antioxidant by evaluating the transformation of Fe (III) to Fe (II) in the presence of reductants in the samples [9]. The reducing power of the samples was monitored at the absorbance of 573 nm using ELISA microplate reader. BHT, Trolox, and AA were used as standard controls. Table 4 shows the FRAP results of the leaves crude extracts of eight *Syzygium* plants. *S. griffithii* (S3LM) exhibits the most potent ferric ion reducer ranging from 1.03 ± 0.03 to 7.52 ± 0.49 mM FRAP equivalent among all the tested leaves extracts followed by *S. pseudoformosum* (1.17 ± 0.07 to 7.31 ± 1.48 mM) and *S. glaucum* (1.09 ± 0.18 to 6.54 ± 0.24 mM).

The crude extracts of the twigs of *S. griffithii* (S3TM) also exhibit good FRAP activity with the value range from 1.43 ± 0.12 to 9.68 ± 0.38 mM FRAP equivalent. Crude extracts of the twigs of *S. syzygioides* (S8TM) also show the potential as antioxidant activity with the FRAP equivalent value ranging from 0.93 ± 0.05 to 8.48 ± 0.25 mM as shown in the Table 5.

Table 4 FRAP assay of the leaves extracts of eight *Syzygium* species.

Samples	mM FRAP EQUIVALENT to $FeSO_4.7H_2O$		
	0.1 mM	**0.6 mM**	**1.0 mM**
Leaves			
S1LM	0.99 ± 0.04	2.37 ± 0.09	3.04 ± 0.21
S2LM	1.29 ± 0.06	4.16 ± 0.03	5.58 ± 0.25
S3LM	1.03 ± 0.03	3.23 ± 0.48	7.52 ± 0.49
S4LM	1.34 ± 0.05	4.09 ± 0.15	5.02 ± 0.26
S5LM	1.16 ± 0.01	4.00 ± 0.64	5.57 ± 0.31
S6LM	1.17 ± 0.07	3.63 ± 0.10	7.31 ± 1.48
S7LM	1.09 ± 0.18	4.38 ± 0.20	6.54 ± 0.24
S8LM	0.73 ± 0.06	2.70 ± 0.31	4.46 ± 1.04
Standards			
Trolox	0.43 ± 0.08	1.23 ± 0.20	2.43 ± 0.40
BHT	0.33 ± 0.09	1.24 ± 0.07	1.89 ± 0.02
AA	0.07 ± 0.08	0.33 ± 0.08	0.60 ± 0.06

S1=*S. filiforme var. filiforme*; S2=*S. papillosum*; S3=*S. griffithii*; S4=*Eugenia* Sp. 57; S5=*S. claviflorum var. claviflorum*; S6=*S. pseudoformosum*; S7=*S. glaucum*; S8=*S. syzygioides;* BHT=Butylated hydroxytoluene; AA=Ascorbic acid;(*p < 0.05).

Table 5 FRAP assay of the twigs extracts of eight *Syzygium* species.

Samples	mM FRAP EQUIVALENT to $FeSO_4.7H_2O$		
	0.1 mM	**0.6 mM**	**1.0 mM**
Twigs			
S2TM	1.07 ± 0.01	2.80 ± 0.40	4.74 ± 0.45
S3TM	1.43 ± 0.12	4.61 ± 0.28	9.68 ± 0.38
S4TM	1.46 ± 0.08	4.44 ± 0.32	6.04 ± 0.82
S5TM	1.28 ± 0.01	4.05 ± 0.24	6.86 ± 0.55
S6TM	0.84 ± 0.02	3.34 ± 0.16	6.02 ± 1.09
S7TM	0.96 ± 0.15	2.68 ± 0.14	4.48 ± 1.33
S8TM	0.93 ± 0.05	4.03 ± 0.44	8.48 ± 0.25
Standards			
Trolox	0.43 ± 0.08	1.23 ± 0.20	2.43 ± 0.40
BHT	0.33 ± 0.09	1.24 ± 0.07	1.89 ± 0.02
AA	0.07 ± 0.08	0.33 ± 0.08	0.60 ± 0.06

S1=*S. filiforme var. filiforme*; S2=*S. papillosum*; S3=*S. griffithii*; S4=*Eugenia* Sp. 57; S5=*S. claviflorum var. claviflorum*; S6=*S. pseudoformosum*; S7=*S. glaucum*; S8=*S. syzygioides;* BHT= Butylated hydroxytoluene; AA=Ascorbic acid;(*p < 0.05).

For the heartwoods crude extracts, *S. syzygioides* (S8HM) showed the most potent ion reducer ranging from 1.20 ± 0.01 to 10.47 ± 0.12 mM FRAP equivalent as shown in Table 6. The crude extracts of the *Eugenia* sp. 57 also have a potential as antioxidant activity with the FRAP equivalent value range from 1.56 ± 0.19 to 8.81 ± 0.46 mM.

Table 6. FRAP assay of the heartwoods extracts of Eight *Syzygium* species.

Samples	mM FRAP EQUIVALENT to $FeSO_4.7H_2O$		
	0.1 mM	**0.6 mM**	**1.0 mM**
Heartwoods			
S1HM	1.12 ± 0.04	3.94 ± 0.54	4.70 ± 0.13
S2HM	1.02 ± 0.08	3.03 ± 0.43	5.96 ± 0.58
S3HM	1.40 ± 0.06	4.84 ± 0.14	6.62 ± 0.27
S4HM	1.56 ± 0.19	5.94 ± 0.16	8.81 ± 0.46
S5HM	1.46 ± 0.09	3.03 ± 1.38	6.17 ± 0.50
S6HM	1.56 ± 0.44	5.00 ± 0.34	6.78 ± 0.74
S7HM	0.95 ± 0.01	2.80 ± 0.04	4.83 ± 0.35
S8HM	1.20 ± 0.01	5.39 ± 0.81	10.47 ± 0.12
Standards			
Trolox	0.43 ± 0.08	1.23 ± 0.20	2.43 ± 0.40
BHT	0.33 ± 0.09	1.24 ± 0.07	1.89 ± 0.02
AA	0.07 ± 0.08	0.33 ± 0.08	0.60 ± 0.06

S1=*S. filiforme var. filiforme*; S2=*S. papillosum*; S3=*S. griffithii*; S4=*Eugenia* Sp. 57; S5=*S. claviflorum var. claviflorum*; S6=*S. pseudoformosum*; S7=*S. glaucum*; S8=*S. syzygioides;* BHT=Butylated hydroxytoluene; AA=Ascorbic acid;(*p < 0.05).

ABTS assay

The determination of the ABTS radical was tested towards all the crude extracts of *Syzygium* species in order to evaluate the antioxidant activity by delocalization the $ABTS^{\bullet+}$ through measuring the reduction of the radical cation as the percentage inhibition of absorbance at 734 nm [11]. BHA, BHT, and Trolox were used as standard controls. Table 7 shows the ABTS radical scavenging activity from the leaves, twigs, and heartwoods of eight *Syzygium* species which is expressed as Scavenging Concentration at 50 % (SC_{50}). From the results, the crude extracts of *S. syzygioides* (S8HM) exhibit good antioxidant activity among the heartwoods extracts with the SC_{50} value of 96.20 mM. All leaves crude extracts show the potential as antioxidant activity towards ABTS assay. *S. glaucum* (S7LM) has obtained good SC_{50} value (116.90 mM) followed by *S. papillosum* (S2LM, SC_{50} value = 132.70 mM) and *Eugenia* Sp. 57 (S4LM, SC_{50} value = 143.10 mM). The twigs crude extracts of *S. syzygioides* (S8TM) and *S. griffithii* (S3TM) show excellent antioxidant activity towards ABTS assay with the SC_{50} values of 130.60 mM and 145.40 mM, respectively.

Table 7 Scavenging Concentration (SC_{50}) of ABTS assay.

Sample	SC_{50} values (mM)
Heartwoods	
S1HM	120.00
S2HM	145.70
S3HM	152.50
S4HM	130.70
S5HM	115.40
S6HM	161.00
S7HM	218.30
S8HM	96.20
Leaves	
S1LM	221.20
S2LM	132.70
S3LM	176.30
S4LM	143.10
S5LM	144.40
S6LM	186.70
S7LM	116.90
S8LM	226.90
Twigs	
S2TM	266.40
S3TM	145.40
S4TM	193.70
S5TM	135.50
S6TM	195.00
S7TM	244.10
S8TM	130.60
Standards	
Trolox	385.00
BHT	175.00
BHA	100.00

STATISTICAL ANALYSIS

Data obtained from biological activities were expressed as mean values. The statistical analyses were carried out by employing one-way ANOVA ($p < 0.05$). A statistical package (SPSS version 16.0) was used for the data analysis.

CONCLUSION

In summary, the present paper reports for the first time on the phytochemical screening of eight Malaysian *Syzygium* species (*S. filiforme var. filiforme, S. papillosum, S. griffithii, S. pseudoformosum, S. claviflorum var. claviflorum, S. glaucum, S. syzygioides*, and *Eugenia* Sp. 57) on their antioxidant activities. All plant extracts showed good DPPH scavenging activity with the percentage inhibition of more than 70 % at 100 ppm. The crude extracts of *S. syzygioides* showed the most potent ion reducing activity ranging from 1.20 ± 0.01 to 10.47 ± 0.12 mM FRAP equivalent. The plant also showed the most potential antioxidant activity towards ABTS assay with the SC_{50} value of 96.20 mM. These findings are important to search for novel phytochemicals from Malaysian *Syzygium* species with bioactivities and to provide scientific evidence that can be used in future for the development of new active agents.

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REFERENCES

[1] Ng, F. S. P., Phil, D. (1978). *Tree flora of Malaysia*, third ed., Forest Department, Ministry of Primary Industries Malaysia, Malaysia: pp. 119.

[2] Ravi, K., Ramachandran, B., Subramanian, S. (2004). Protective effect of *Eugenia jambolana* seed kernel on tissue antioxidants in streptozotocin-induced diabetic rat. *Biological and Pharmaceutical Bulletin*, 27(8): 1212 - 1217.

[3] Bajpai, M., Pande, A., Tewar, S. K., Prakash, D. (2005). Phenolic contents and antioxidant activity of some food and medicinal plants. *International Journal of Food Sciences and Nutrition*, 56(4): 287–291.

[4] Mohanty, S., Cock, I. E. (2010). Bioactivity of *Syzygium jambos* methanolic extracts: Antibacterial activity and toxicity. *Pharmacognosy Research*, 2(1): 4-9.

[5] Tiwari, P., Verma, R., Ahirwar, D., Chandy, A., Dwivedi, S. (2014). Evaluation of anxiolytic effect of *Syzygium aromaticum*: A traditional herb of India. *Asian Pacific Journal of Tropical Disease*, 4(1): 77-80.

[6] Gowri, S. S., Vasantha, K. (2010). Phytochemical screening and antibacterial activity of *Syzygium cumini* (L.) (Myrtaceae) leaves extracts. *International Journal of Pharmatech Research*, 2(2): 1569-1673.

[7] Kamal, A. (2014). Phytochemical screening of *Syzygium cumini* seeds. *Indian Journal of Plant Sciences*, 3(4):1-4.

[8] Hashim, N. M., Rahmani, M., Ee, G. C., Sukari, M. A., Yahayu, M., Amin, M. A., Ali, A. M., Go, R. (2012). Antioxidant, antimicrobial and tyrosinase inhibitory activities of xanthones isolated from *Artocarpus obtusus* F. M. Jarret, *Molecules*. 17(5): 6071-6082.

[9] Shahwar, D., Raza, M. A., Bukhari, S., Bukhari, G. (2012). Ferric reducing antioxidant power of essential oils extracted from *Eucalyptus* and *Curcuma* Species. *Asian Pacific Journal of Tropical Biomedicine*, 2(3): S1633-S1636.

[10] Channarong, S., Jutiviboonsuk, A., Korsanan, S. (2012). Total reducing antioxidant capacity of Thai herbal aromatic powder (Ya Hom) measured by FRAP assay. *Thai Pharmaceutical and Health Science Journal*, 7(3): 111-114.

[11] Zou, Y., Chang, S. K. C., Gu, Y., Qian, S. Y. (2011). Antioxidant activity and phenolic composition of Lentil *(Lens culinaris var. Morton)* extracts and its fractions. *Journal of Agricultural and Food Chemistry*, 59(6): 2268-2276.

[12] Kumar, A., Ilavarasan, R., Jayachandran, T., Decaraman, M., Aravindhan, P., Padmanabhan, N., Krishnan, M. R. V. (2009). Phytochemicals investigation on a tropical plant, *Syzygium cumini* from Kattuppalayam, Erode district, Tamil Nadu, South India. *Pakistan Journal of Nutrition*, 8(1): 83-85.

[13] Balyan, U. and Sarkar, B. (2016). Integrated membrane process for purification and concentration of aqueous *Syzygium cumini* (L.) seed. *Food Bioproducts Processing*. 9(8): 29–43.

[14] Muruganandan, S., Srinivasan. K. S., Chandra, S. K., Tandan, J. L., Paviprakash, V. (2001). Anti-inflammatory activity of *Syzygium cumini* bark. *Fitoterapia*, 72(4): 369-375.

[15] Saroj, A., Pragadheesh, V. S., Palanivelu, Yadav, A., Singh, S. C., Samad, A., Negi, A. S., Chanotiya, C. S. (2015). Anti phytopathogenic activity of *Syzygium cumini* essential oil, hydrocarbon fractions and its novel constituents. *Industrial Crops and Products*, 74: 327–335.

[16] Samy, M. N., Sugimoto, S., Matsunami, K., Otsuka, H., Kamel, M. S. (2014). One new flavonoid xyloside and one new natural triterpene rhamnoside from the leaves of *Syzygium grande*. *Phytochemistry Letters*, 10: 86–90.

[17] Osman, H., Rahim, A. A., Isa, N. M., Bakhir, N. M. (2009). Antioxidant activity and phenolic content of *Paederia foetida* and *Syzygium aqueum*. *Molecules*, 14(3): 970–978.

[18] Manaharan, T., Appleton, D., Cheng, H. M., Palanisamy, U. D. (2012). Flavonoids isolated from *Syzygium aqueum* leaf extract as potential anti hyperglycaemic agents. *Food Chemistry*, 132(4): 1802-1807.

[19] Djoukeng, J. D., Abou-Mansour, E., Tabacchi, R., Tapondjou, A. L., Bouda, H., Lontsi, D. (2005). Antibacterial triterpenes from *Syzygium guineense* (Myrtaceae). *Journal of Ethnopharmacology*, 101(1-3): 283–286.

[20] Simirgiotis, M. J., Adachi, S., To, S., Yang, H., Reynertson, K. A., Basile, M. J., Gil, R. R., Weinstein, I. B., Kennelly, E. J. (2008). Cytotoxic chalcones and antioxidants from the fruits of *Syzygium samarangense* (wax jambu). *Food Chemistry*, 107(2): 813–819.

[21] Tavares, I. M. C., Vanzela, E. S. L., Rebello, L. P. G., Ramos, A. M., Alonso, S. G., Romer, E. G., Silva, R. D., Gutierrez, I. H. (2016). Comprehensive study of the phenolic composition of the edible parts of jambolan fruit (*Syzygium cumini* (L.) Skeels). *Food Research International*, 82: 1–13.

[22] Bitencourt, P. E. R., Ferreira, L. M., Cargnelutti, L. O., Denardi, L., Boligon, A., Fleck, M., Brandao, R., Athayde, M. L., Cruz, L., Zanette, R. A., Alves, S. H., Moretto, M. B. (2016). A new biodegradable polymeric nanoparticle formulation containing *Syzygium cumini*: phytochemical profile, antioxidant and antifungal activity and in vivo toxicity. *Industrial Crops and Products*, 83: 400–407.

[23] Ayyanar, M., Babu, P. S. (2012). *Syzygium cumini* (L.) skeels: A review of its phytochemical constituents and traditional uses. *Asian Pacific Journal of Tropical Biomedicine*, 2(3): 240–246.

[24] El-Maati, M. F. A., Mahgoub, S. A., Labib, S. M., Al-Gaby, A. M. A., Ramadan, M. F. (2015). Phenolic extracts of clove (*Syzygium aromaticum*) with novel antioxidant and antibacterial activities. *European Journal of Integrative Medicine*, 8(4): 1-11.

[25] Nishikava, H., Hata, T., Funakami, Y. (2004). A role for corticotropin-releasing factor in repeated cold stress-induced anxiety-like behavior during forced swimming and elevated plus-maze test in mice. *Biological and Pharmaceutical Bulletin*, 27(3): 352-356.

[26] Millan, M. J., Hjorth, S., Samanin, R., Schreiber, R., Jaffard, R., Ladonchamps, B. (1997). S 15535. A novel benzodioxopiperazine ligand of serotonin (5-HT) 1A receptors: II. Modulation of hippocampal serotonin release in relation to potential anxiolytic properties. *Journal of Pharmacology and Experimental Therapeutics*, 282(1): 148-161.

[27] Mansourian, A., Boojarpour, N., Ashnagar, S., Beitollahi, J. M., Shamshiri, A. R. (2014). The comparative study of antifungal activity of *Syzygium aromaticum*, *Punica granatum* and nystatin on *Candida albicans;* An in vitro study. *Journal de Mycologie Medicale*, 24(4): 163-168.

[28] Krishnasamy, G., Muthusamy, K. (2015). In vitro evaluation of antioxidant and antidiabetic activities of *Syzygium densiflorum* fruits. *Asian Pacific Journal of Tropical Disease*, 5(11): 912-917.

[29] Tupe, R. S., Kulkarni, A., Adeshara, K., Shaikh, S., Shah, N., Jadhav, A. (2015). *Syzygium jambolanum* and *Cephalandra indica* Homeopathic preparations inhibit albumin glycation and protect erythrocytes: An in vitro study. *Homeopathy*, 104(3): 197-204.

[30] Vuong, Q. V., Hirun, S., Chuen, T. L. K., Goldsmith, C. D., Bowyer, M. C., Chalmers, A. C., Phillips, P. A., Scarlett, C. J. (2014). Physicochemical composition, antioxidant and anti-proliferative capacity of a lilly pilly (*Syzygium paniculatum*) extract. *Journal of Herbal Medicine*, 4(3): 134–140.

[31] Arumugam, B., Manaharan, T., Heng, C. K., Kuppusamy, U. R., Palanisamy, U. D. (2014). Antioxidant and antiglycemic potentials of a standardized extract of *Syzygium malaccense*. *Food Science and Technology*, 59(2): 707-712.

[32] Arumugam, B., Palanisamy, U. D., Heng, C. K. and Kuppusamy, U. R. (2016). Potential antihyperglycaemic effect of myricetin derivatives from *Syzygium malaccense*. *Journal of Functional Foods*, 22: 325-336.

[33] Nobori, E., Miurak, K., Wu, D. J., Takabayashik, L. A. and Carson, D. A. (1994). Deletions of the cyclin-dependent kinase-4 inhibitor gene in multiple human cancers. *Nature*, 368(6473): 753-756.

[34] Aiyelaagbe, O. O. and Osamudiamen, P. M. (2009). Phytochemical screening for active compounds in *Mangifera indica* leaves from Ibadan, Oyo State. *Plant Sciences Research*, 2(1): 11-13.