

ANTIOXIDANT AND ANTIBACTERIAL PROPERTIES ON DIFERENT TISSUE OF *Syzygium aromaticum*

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ABSTRACT

This paper focuses on the effect of three drying treatments (microwave-, oven- and air-drying) on the antioxidant and antibacterial properties of different tissue of *Syzygium aromaticum*. The microwave-oven was found to be effective drying method to maintain the antioxidant and antibacterial properties of unripe fruits, leaves and stems of *S. aromaticum*. The unripe fruits exhibit higher TPC, CQAC, Vitamin C and antibacterial activity, followed by leaves and stems. The increasing order for the TFC is stems, unripe fruits and leaves. In this study, all the antioxidant compounds correlate well with antioxidant activities that were determined using DPPH and FRAP assay.

KEYWORDS: *Syzygium Aromaticum*, Antioxidant, Antibacterial, Microwave-Oven.

INTRODUCTION

The search of healing powers in plants is an idea of ancient (Craig, 1999). Since ancient time, plant extracts are used by man to safeguard himself against certain diseases as well as to promote his health. Recently, the demand for herbs and medicinal plants are becoming high as the number of people that using herbal remedies in their daily life inclined. It was reported that approximately 80% of the world's population especially those in third world countries largely rely on herbs for maintaining their health (Rahmat et al., 2006). Many aromatic, medicinal and spice plants contain confirmed strong antioxidant possessive components (Mishra et al., 2007). Researches and development nowadays in the field of scientific technique explore several non-nutritive chemicals of plants such as terpenoids and flavanoids that were initially thought to be not significant to human diet carries antioxidant properties (Mishra et al., 2007). Antioxidant as matter of fact are complex compounds that provides barrier for the body against several devastating diseases such as cardiac or arterial diseases, arthritis, cataracts, premature ageing and other chronic diseases. Seeing that plants are susceptible to damage caused by active oxygen, it develops many antioxidant defense systems which eventually results in the formation of potential antioxidants. Besides, plants have served well humans as a precious component of seasoning, beverages, cosmetics, dyes, and medicines. *Syzygium aromaticum* have been accepted as one of the most ancient and valuable spices of the Orient (Leena and Sapna, 2008) and looked upon as a source of natural antioxidants.

Recently, Deepa and Milliard (2011) stated that *Syzygium aromaticum* (tropical cloves) considered as a champion of all the antioxidants known till date. Moreover, Dorman et al., (2000) reported that *S. aromaticum* exhibits powerful antioxidant activity that can be compared to the activities of BHA and pyrogallol which are the synthetic antioxidants. Leena and Sapna (2008) also stated that the essential oil extracted from *S. aromaticum* leaf exhibits scavenging activity over 2, 2-diphenyl-1-picryl hydrazyl (DPPH) radical at lower concentration compared to eugenol, butylated hydroxytoluene (BHT), butylated hydroxyanisole (BHA) as well as a considerable inhibition towards hydroxyl radicals (Gulcin et al., 2004 and Jirovetz et al., 2006). The antioxidant activity of *S. aromaticum* bud extract and its aromatic compounds are equivalent to the natural antioxidant, α -tocopherol (Lee and Shibamoto, 2001). In addition, *S. aromaticum* also plays an important role in medicine for its antibacterial, antiseptic and antibiotic properties. According to a study conducted by Shafi et al., (2002), it is found that *S. aromaticum* possess the antibacterial activity. On the other hand, anti-inflammatory activity of *S. aromaticum* was reported by Muruganadan et al., (2001). The studies confirmed that spices including *S. aromaticum* able to inhibit the growth and activities of both Gram-negative and Gram-positive bacteria (Hoque et al., 2008).

This study which focused on antioxidant and antibacterial properties on different tissue of *S. aromaticum* namely unripe fruits, leaves and stems was conducted as there was not much research have been done on this Malaysian grown species with medicinal value especially by combining effect of different drying treatments on the total phenolic content (TPC), total flavanoid content (TFC), caffeoylquinic acid content (CQAC), total vitamin C content, antioxidant activities (AOA) and antibacterial activities. Besides, the present study introduced the effective drying method that can sustain most partial loss of antioxidant activity during the

drying treatment. Thus, the present study represents the first systematic analysis that combines the effect of drying treatments, total phenolics content (TPC), total flavanoid content (TFC), caffeoylquinic content (CQAC), total vitamin C content, antioxidant activities (AOA) and antibacterial activities on different tissue of *S. aromaticum* which fulfilled the objectives of the study to determine the antioxidant content from tissues of *S. aromaticum* and its antioxidant activity, as well as determine the the antioxidant content from tissues of *S. aromaticum* and its antioxidant activity, as well as determine the correlation between both of the antioxidant compounds and its activity. other than that, antibacterial activity of those tissues using Gram-negative and Gram-positive bacteria also being investigated in this study.

MATERIALS AND METHODS

Sample Collection

Fresh leaves, stems and unripe fruits samples of *S. aromaticum* were collected from the trees at Universiti Putra Malaysia (UPM), Bangi, Malaysia. The handpicked samples of the leaves, stems and unripe fruits were subjected to various drying treatments within the same working day.

Drying Treatments

Three different drying methods namely, microwave-, oven- and air-drying methods was carried out. The different tissue of *S. aromaticum* was thoroughly washed 2-3 times with running tap water before subjected to drying treatments in order to clean debris and contaminant. In microwave-drying, the plant materials were subjected to the 800W power input for 2 min in a microwave oven (Triple distribution system, Samsung). In oven-drying, the plant materials were subjected to 40°C for 24 hour in oven (Memmert). In air-drying, the plant materials were air-dried for three days in the laboratory at ambient temperature of 25–30° C. The fresh samples were subjected to drying treatment until a constant weight is obtained. Once, the drying process over, the plant materials were blendered separately using cutting machine (Fritch) and were stored at room temperature for further analysis.

Sample Extraction

The three replicates for each sample of grinded leaves, stems and unripe fruits of 0.1g were weighed out using an electronic balance (Mettler Toledo). Then, 25mL of distilled water was used to extract the samples and boiled at 50°C-60°C for 30 minutes separately. The extracted samples were filtered through Whatman No.1 filter paper. Then, filtrates obtained were stored at 4°C to be used for further analysis of antioxidative activity. The soxhlet extraction method was also used to extract the different tissue of *S. aromaticum* for the determination of antibacterial activity. The, 30g grinded leaves, stems and unripe fruits samples of *S. aromaticum* were extracted with 175mL of methanol in a soxhlet extractor. After four hours of extraction, the honey form extract was stored at 4°C for further use.

Determination of Antioxidant Compounds

Total phenolic content (TPC) of different tissue of *S. aromaticum* extracts were conducted using Folin-Ciocalteu assay (Chan et al., 2007). The calibration equation obtained for gallic acid was $y = 0.007x + 0.018$ ($R^2 = 0.9817$), where y-axis represents the absorbance readings and x-axis represents the concentration of gallic acid used in µg/mL. The total phenolics content was determined in triplicate.

The total flavonoid content of different tissue of *S. aromaticum* extracts was determined using a method reported by Chang et al. (2002) on each drying method. The calibration equation used for quercetin was $y = 0.018x + 0.039$ ($R^2 = 0.9751$) where y-axis represents absorbance readings and x-axis represents the concentration of quercetin used in µg/mL. The total flavonoids content was determined in triplicate.

Caffeoylquinic acid content (CQAC) of different tissue of *S. aromaticum* extracts was conducted using the molybdate assay (Clifford and Wright, 1976). The calibration equation used for chlorogenic acid was $y = 0.004x + 0.007$ ($R^2 = 0.9724$) where y-axis represents absorbance readings and x-axis represents the concentration of chlorogenic acid used in µg/mL. The caffeoylquinic acid content was determined in triplicate.

The content of total vitamin C in different tissue of *S. aromaticum* extracts was determined using a modified method reported by Davis and Masten (1991) where 1g of different tissues of *S. aromaticum* was extracted with 5mL of phosphate citrate buffer (1% at pH 3.5) in a mortar and pestle. The calibration equation used for ascorbic acid was $y = -0.012x + 1.790$ ($R^2 = 0.9725$) where y-axis represents absorbance readings and x-axis

represents the concentration of ascorbic acid used in $\mu\text{g/mL}$. The total vitamin C content was determined in triplicate.

Determination of Antioxidant Activity

The DPPH free radical-scavenging (FRS) assay was determined method used by Miliauskas et al. (2004) with some modifications. The 1g of different tissue of *S. aromaticum* was extracted in 25mL of distilled water. FRS ability was calculated as IC_{50} (half maximal inhibitory concentration) and determined as ascorbic acid equivalent antioxidant capacity (AEAC) in g AA/100 g (Leong & Shui, 2002) using the formula given below:

$$\text{AEAC (mg AA/100g)} = \text{IC}_{50(\text{ascorbate})} / \text{IC}_{50(\text{extract})} \times 100,000$$

The IC_{50} of ascorbic acid that adapted for the calculation of AEAC was 0.002688 mg/mL. The DPPH assay was conducted in triplicate and was repeated thrice. The experiment also was conducted in dark condition.

The FRAP assay was determined by following method reported by Wong et al. (2005). The 1g of different tissue of *S. aromaticum* was extracted in 25mL of distilled water. Antioxidant capacity that depends on ferric ions reducing ability of the plant extracts was expressed as percent of antioxidant. The calculation for percent of antioxidant is shown below,

$$\text{Percent of antioxidant (\%)} = [(\text{A593 of sample} - \text{A593 of control}) / \text{A593 of sample}] \times 100.$$

The FRAP assay was conducted in triplicate and was repeated thrice.

Determination of Antibacterial Activity

The Kirby-Bauer disc-diffusion method was used to inoculate the Mueller-Hinton agar plate. The organism was suspended in 2 ml of sterile saline. The turbidity of the bacterial suspension was standardized using a spectrophotometer. The absorbance reading at 625 nm should be 0.1 for the 0.5 McFarland standard. A sterile swab was dipped into the inoculum tube and the surface of a Mueller-Hinton agar plate was inoculated with streaking the swab over the entire agar surface three times and each time the plate was rotated nearly 60 degrees. Filter paper disc (6 mm in diameter, Whatman No. 1) was picked up with lightly flamed forcep and was dipped with soxhlet extracted leaves, stems and unripe fruits of *S. aromaticum* separately (0.5g of extract in 0.1mL of methanol). The filter paper disc was then transferred onto the inoculated agar. Streptomycin immersed filter paper disc and methanol immersed filter paper disc was used as positive and negative controls. The working solution of control antibiotics was prepared with the concentration of 1mg/mL. Then, the plates was inverted and incubated for 24 hours at 37°C. After incubation overnight at 37°C, inhibition zones was measured on bottom of plate and recorded to nearest millimeters. The test was carried out in triplicate and mean value was calculated. The antibacterial activity of the extracts was expressed based on inhibition percentage of streptomycin and was classified as strong (+++), moderate (++) and weak (+) for the inhibition percentage of $\geq 70\%$, $50 < 70\%$ and $< 50\%$ respectively.

Statistical Analysis

All the experimental results of the parameters was determined in triplicate and repeated three times. Moreover, all the results were expressed in means \pm standard deviation (SD). The data were analyzed using one way ANOVA. The mean values were compared using Duncan's multiple range test at 5% ($p = 0.05$) significance level with the aid of Statistical Package for the Social Sciences (SPSS) software version 16.0 for windows.

RESULT AND DISCUSSION

The study of antioxidant and antibacterial properties on different tissue of *Syzygium aromaticum* by combining the effect of different drying treatments has not been reported previously. Extraction efficiency of water was determined from different tissue of *S. aromaticum* on their total phenolic content (TPC) accordingly. The first extractions of microwave oven dried tissues yield 87% to 89% range, followed by the second extractions that yielded 8-12%. For oven dried tissues first extraction, the outcome showed a range from 87% to 89% that homologous to the first extraction of microwave oven extract. Second extraction on the other hand yielded higher percentage than the microwave oven extract which is 9% to 10%. Third extractions remained the same that is nearly 3% for both oven dried and microwave oven. Together with air dried tissues, triple extractions of different tissues yielded are 87-90%, 7-9% and 2-3% respectively. Above all, the first extraction gave superior yield over second and third extractions. Preliminary study show that using the water solvent

system is efficiently to extract the antioxidant properties in all the three tissue as reported by Kim *et al.*, (2011) that hot water extracted *S. aromaticum* exhibits phenolic content (108.28 µg CE/g) among all other 13 types of spices that they analyzed. Apart from that, Abdou *et al.*, (2011) concluded in a research on *S. aromaticum* that it gave the highest percentage of extracting phenolic compound over 10 other edible plants tested with range 57% to 86% by water-based extraction. It is comparable to Li *et al.*, (2006) research on Citrus peel that concludes polar solvent extracts polar compound which definitely proved the extraction theory. In the present study, it was found that microwave-oven drying method is superior to other drying methods in retaining the antioxidant and antibacterial properties of different tissue of *S. aromaticum*. The advantage of microwave system which applies heat at all directions has obviously shown their effectiveness in drying in this study. The effectiveness of microwave-oven drying also observed for leaves of *Vitex* species (Chong and Lim, 2011). Oven-drying is found to be the next effective drying method and followed by air-drying method. The influences of drying method on antioxidant activities were tested using two assays; 2, 2-diphenyl-1-picrylhydrazyl (DPPH) and Ferric Reducing Antioxidant Power (FRAP) whereas the antioxidant content was tested using caffeoylquinic acid content (CQAC), total phenolic content (TPC), total flavanoid content (TFC) and total vitamin C content. The TPC was first tested to determine the presence of phenolic compound in *S. aromaticum*.

From this study, it was found that unripe fruits of *S. aromaticum* exhibits highest phenolic compound regardless on the drying treatments, followed by leaves and stems in TPC test (Table 3.1). This result obviously shows the positive correlation of phenolic compound with the antioxidant activities in *S. aromaticum* (Table 3.3). With the increasing amount of phenol compound found in a plant, it clearly indicates the ability of the plant as an antioxidant source. Previous research done by Ronald *et al.*, (1998) on their *Vaccinium* berries plant also shows a remarkable correlation between phenolic compounds with the antioxidant behaviour. Additionally, Kim *et al.*, (2011) in their study on extraction using hot water extract, found a large quantity of phenolic compound in the *S. aromaticum*. TPC as matter of fact was carried out in the first place to provide an estimation of phenolic content in the sample. The usage of Follin ciocalteu reagent was able to characterize the compound in fast and simple way. On the other hand, TFC shows a contradict outcome with TPC which having leaves as the greatest contributor of phenol compounds which the same pattern of result was obtained by Hsu *et al.*, (2006) that reported Doum Palm leaves exhibits higher amount in TFC as compared to *Strobilanthes crispus* plant. This result is in agreement with our present study (Table 3.1) where flavonoid content were 100 fold lower than phenolic content. Thus, it is proven that *S. aromaticum* species exhibits lower amount of flavanoid in comparison to its phenolic content. A research reported by Ismail *et al.*, (2004) show the relationship between phenolic compound and flavanoid, where the higher the amount of phenolic, the higher the amount of flavanoid compound. CQAC presents similar result with the TPC that put unripe fruits as the champion among other two, leaves and stem. As shown in Table 3.1, microwave oven dried samples retains higher amount of caffeoylquinic acid contents compared to oven and air-dried samples. In addition, caffeoylquinic acid can be categorized under phenolic acids because the formation of hydroxycinnamic acid which is one of phenolic category, are made up of caffeic and quinic acid (Manach *et al.*, 2004). A research conducted by Chan *et al.*, (2009) has shown proportionality between TPC and CQAC in five leaves extracts of *Etligeria* species where extracts with higher TPC contains higher CQAC.

From total vitamin C assay, unripe fruits of *S. aromaticum* also found to have the highest antioxidant content apart from leaves and stems. Referring to the result, it is found that microwave oven dried fruits and stems capable to preserve higher amount of vitamin C followed by oven and air dried samples. In contradict, the oven dried leaves were competent to uphold high quantity of vitamin C than microwave oven and air dried leaves accordingly. In this study, water extracted *S. aromaticum* possess significant amount of vitamin C by virtue of its nature of water solubility. Previous study by Bhowmik *et al.*, (2012) has shown that in 100 g of *S. aromaticum* consisting 11.7 mg of vitamin C. Moreover, Leena and Sapna (2008) also found 80.81 mg/100 g of vitamin C in *S. aromaticum*. These reports were unambiguously proved *S. aromaticum* as plant having antioxidant properties. To recapitulate, among the different tissue of *S. aromaticum*, the unripe fruits contain higher level TPC, CQAC and Vitamin C, followed by the leaves and stems. Whereas, the leaves of *S. aromaticum* contains higher level of TFC, followed by unripe fruits and stems. From this study, it is clearly shown that phenolic, flavonoid, caffeoylquinic and Vitamin C content are well correlated with antioxidant activities.

The antioxidant activities were then continued by DPPH and FRAP assays. In obtaining the amount of scavenging ability in vitro, DPPH assay was used to determine the potential of the substance to donate hydrogen atom or electrons because with the presence of antioxidants mean there are the presences of hydrogen donating groups (Rohman *et al.*, 2010). The process continues as the hydrogen donating groups hence will break the chain of the free radicals (Endo *et al.*, 1985). This hydrogen groups eventually will remove the odd electron feature that responsible for radical formation activity; and also hydrogen was donated to the free radical in its reduction to unreactive species (Hsu *et al.*, 2006). Loganayaki *et al.*, (2011) and Brand-William *et al.*, (1995), prove the same concept either that hydrogen donating ability contributed by the nature of phenolics related to

the scavenging activity. Others authors such as Satisha *et al.*, (2011) concluded the same result stated the scavenging ability is proportional to the amount of hydrogen donating present. From the present study, it was found that the unripe fruits of the *S. aromaticum* exhibits highest ascorbic acid equivalent antioxidant capacity (AEAC) in DPPH assay followed by leaves and stems (Table 3.2). DPPH was also observed from the IC₅₀ of the extract that shows inversely proportional to the AEAC where the lower the IC₅₀, the higher the AEAC. Thus, it gives the same resemblance to the ginger species that shows a directly proportional between TPC and AEAC; the higher the TPC, the higher AEAC (Chan *et al.*, 2009). The results show microwave oven dried unripe fruits, leaves and stems of *S. aromaticum* exhibits lower IC₅₀ than ascorbic acid (0.002688 mg/mL) which are 0.002456 mg/mL, 0.002557 mg/mL, and 0.002626 mg/mL respectively. IC₅₀ was defined by Rohman *et al.*, (2010) as the antioxidant concentration required scavenging 50 % of DPPH radical in the sample at specific time. These are comparable with a research conducted by Kim *et al.*, (2011) that significantly obtained 84.22% of inhibition in DPPH assay from *S. aromaticum*, by hot water extract. Loganayaki *et al.*, (2011) proved that antioxidant activity in DPPH increases with the increasing amount of phenolic compound due to the double bond existing in C-ring. Torane *et al.*, (2011) also suggested a theory that the ability of antioxidant as a radical scavenger is due to the amount of hydroxyl group and chemical structure in its phenolic compound. Together with DPPH, FRAP assay also demonstrates unripe fruits having highest reducing ability of ferric ions. To wrap up, the increasing order of antioxidant activity with DPPH and FRAP assay are stems, leaves and unripe fruits.

Table 3.1: Antioxidant compounds of unripe fruits, leaves and stems of *S. aromaticum*.

Drying Treatment	Tissue	TPC (mg GAE/100g)	TFC (mg QE/100g)	CQAC (mg CGAE/100g)	Total Vitamin C Content (mg AA /100g)
Microwave oven	Leaves	264.93 ± 15.46 ^b	2.35 ± 0.08 ^a	74.79 ± 3.45 ^{bc}	8.58 ± 0.06 ^e
	Stems	252.19 ± 2.17 ^c	1.84 ± 0.03 ^d	60.13 ± 2.05 ^d	6.98 ± 0.05 ^e
	Unripe fruits	325.19 ± 1.54 ^a	2.23 ± 0.05 ^{ab}	86.71 ± 3.09 ^a	9.47 ± 0.02 ^a
Air	Leaves	228.00 ± 4.12 ^d	1.89 ± 0.11 ^{cd}	56.71 ± 1.73 ^d	8.14 ± 0.05 ^f
	Stems	213.05 ± 2.07 ^e	1.05 ± 0.27 ^f	55.46 ± 1.71 ^d	6.59 ± 0.04 ⁱ
	Unripe fruits	319.57 ± 0.87 ^a	1.50 ± 0.17 ^e	72.63 ± 1.44 ^c	8.88 ± 0.04 ^c
Oven	Leaves	252.52 ± 2.21 ^c	2.09 ± 0.05 ^{bc}	73.04 ± 2.02 ^c	8.72 ± 0.05 ^d
	Stems	216.24 ± 6.38 ^e	1.70 ± 0.06 ^{de}	59.25 ± 2.88 ^d	6.78 ± 0.04 ^b
	Unripe fruits	323.76 ± 3.75 ^a	1.74 ± 0.10 ^d	80.00 ± 7.73 ^b	9.05 ± 0.04 ^b

Note: Values of TPC, TFC, CQAC and Total Vitamin C are means ± SD (n = 3). The different letters within the column indicate that the values are significantly different (p≤0.05)

Table 3.2: Antioxidant activities of unripe fruits, leaves and stems of *S. aromaticum*.

Drying Treatment	Tissue	FRAP (% of antioxidant)	DPPH	
			IC ₅₀ (mg/mL)	AEAC (g AA/100g)
Microwave oven	Leaves	81.89 ± 0.07 ^b	0.002557	105.24 ± 4.16 ^{ab}
	Stems	80.19 ± 0.06 ^a	0.002626	102.38 ± 1.03 ^{abc}
	Unripe fruits	82.19 ± 0.07 ^a	0.002456	109.58 ± 4.67 ^a
Air	Leaves	79.56 ± 0.18 ^f	0.003293	82.30 ± 9.19 ^d
	Stems	78.46 ± 0.03 ^e	0.004455	63.91 ± 19.96 ^c
	Unripe fruits	80.37 ± 0.09 ^d	0.003151	85.91 ± 9.00 ^{cd}
Oven	Leaves	81.38 ± 0.09 ^c	0.002968	90.91 ± 6.99 ^{bcd}
	Stems	80.08 ± 0.16 ^c	0.003027	89.20 ± 7.50 ^{bcd}
	Unripe fruits	81.91 ± 0.11 ^b	0.002759	97.56 ± 4.30 ^{bcd}

Note: Values of FRAP and AEAC are means ± SD (n = 3). The different letters within the column indicate that the values are significantly different (p≤0.05).

Table 3.3: Correlation of antioxidant compounds and antioxidant activities on unripe fruits, leaves and stems of *S. aromaticum*

Correlation	TPC (mg GAE/100g)	TFC (mg QE/100g)	CQAC (mg CGAE/100g)	Total Vitamin C Content (mg AA /100g)
FRAP (% of antioxidant)	0.721	0.768	0.888	0.796
AEAC (mg AA/100g)	0.506	0.716	0.513	0.475

Table 3.4: Antibacterial activity of soxhlet extracted unripe fruits, leaves and stems of *S.aromaticum* using Gram-positive and Gram-negative bacteria.

Drying Treatment	Tissue	Zone of inhibition (mm)			
		Gram-positive bacteria		Gram-negative bacteria	
		<i>B. subtilis</i>	<i>S. aureus</i>	<i>E. coli</i>	<i>P. aeruginosa</i>
Microwave oven	Leaves	4.63 (66) **	10.47 (118)***	5.50 (67)**	9.1 (98)***
	Stems	3.50 (50)**	9.67 (109)***	5.37 (66)**	8.4 (87)***
	Unripe fruits	5.30 (74)***	12.33 (137)***	6.43 (77)**	10.0 (107)***
Air	Leaves	3.30 (47),	7.50 (94)***	5.10 (64)**	8.2 (85)***
	Stems	2.30 (33),	7.43 (89)***	4.37 (53)**	8.0 (80)***
	Unripe fruits	3.90 (55)**	10.00 (109)***	5.50 (67)**	9.0 (93)***
Oven	Leaves	3.80 (53)**	9.33 (104)***	5.17 (63)**	8.7 (90)***
	Stems	3.43 (48),	10.00 (111)***	5.07 (62)**	8.0 (87)***
	Unripe fruits	4.63 (66)**	11.63 (129)***	5.77 (72)***	10.0 (100)***
Streptomycin		7.06	8.80	8.15	9.62
Methanol		-	-	-	-

Apart from that, the different tissue of *S.aromaticum* also shows a significant inhibition against both the gram-positive and gram-negative bacteria (Table 3.4). The inhibition was found to be effective against *P. aeruginosa*, *E.coli* and *S. aureus* compare to *B.subtilis*. The unripe fruit of *S.aromaticum* was found exhibit higher antibacterial activities compare to leaves and stems. According to the antibacterial activity test result, it was also found the effect of drying treatment which shows microwave oven and oven dried different tissue of *S. aromaticum* exhibits higher percentage of inhibition over air dried extracts. Khalid and Kiong (2010) reported on the inhibition of *Eschericia coli*, *Salmonella spp*, *Klebsiella pneumonia*, *Staphylococcus aureus*, *Streptococcus* and *Bacillus subtilis* by *S. aromaticum* where undeniably presented its high antibacterial properties. Likewise, Aneja and Joshi (2010) supported the same pattern of result having *S. aromaticum* which was extracted using methanol extract, competent in inhibiting *Staphylococcus aureus*. In addition, Pandey and Singh (2011), reported the ability of *S. aromaticum* to inhibit *S. aureus* (24mm), *P. aeroginosa* (19mm) and *E. Coli* (20 mm) in their study. These previous research are assuredly confirmed the antibacterial properties in *S. aromaticum*. Moreover, eugenol is the main constituent of *S. aromaticum* that responsible for its antioxidant and antibacterial properties (Kamataoet al., 2012). The amount of eugenol was able to be retained in the samples by cause of microwave oven and oven drying ability in conserving high amount of antioxidant compounds. The results for the antioxidant content and activities comparison are as follows.

CONCLUSIONS

The effects of three different drying treatments on the antioxidant and antibacterial properties of *S.aromaticum* were studied. The most effective drying treatment was microwave-drying as it able to retain higher amount of antioxidant compounds than oven-drying and air-drying. Additionally, the antioxidant compounds were largely found in unripe fruits of *S.aromaticum*, followed by the leaves and stems. The strong correlations were observed for FRAP assay and moderate correlations were observed for DPPH assay. *Syzygium aromaticum* also showed effective inhibition against both gram-positive and gram-negative bacteria. In addition, the antioxidant compound correlate well with antioxidant activities. Sample with higher antioxidant compounds exhibit higher antibacterial activity that proves *S. aromaticum* is rich with antioxidant and antibacterial properties.

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