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Full Length Article

Antioxidant and Antibacterial Activities of Different Extracts and Fractions of a Mangrove Plant *Sonneratia alba*

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Abstract

A study was carried out to investigate the antioxidant potential and antibacterial activities of ethanol, methanol and chloroform extracts from soil grown and tissue culture explants of the mangrove plant *Sonneratia alba,* as well as its ethyl acetate and water extracted fractions. Antioxidant effects were determined by DPPH and SOD assays while antibacterial activities were determined using the agar disk diffusion method. LC-MS/MS analysis was also conducted in an attempt to identify active compounds in the extracts and fractions. The results showed that both the methanol and ethanol extracts of leaf and bark exhibited potentially good antioxidant and antibacterial activities compared to the chloroform extracts. There was a positive correlation between the total phenolic content and the antioxidant effects of the ethanol and methanol extracts of leaf and bark ($R^2 = 0.72$). Among the crude extracts, the ethanol extract of bark exhibited the best antioxidant and antibacterial activities. Chloroform extracts of both leaf and bark samples exhibited lower antioxidant and antibacterial activities. On the other hand, water extracted fraction showed better antioxidant activity compared to ethyl acetate extracted fraction. All the extracts showed good antioxidant activity with IC_{50} values ranging between 0.019-0.37 mg mL^{-1}. LC-MS/MS analysis revealed 8 peaks at m/z values of 331, 345, 452, 463, 473, 480 and 494 with fragmentation patterns corresponding to the high antioxidant activity of the water extracted fraction. This study has shown that there are metabolites in *S. alba* that have the potential to serve as antioxidant and antibacterial compounds that may serve as a lead to the development of new pharmaceuticals.

Keywords: *Sonneratia alba*; Crude extract; Fractionated constituent; Antioxidant effect/potential; Antibacterial activity

Introduction

Mangrove is generally used to describe the assemblage of woody trees and shrubs which grow in saline coastal habitats in the tropics and subtropics. Extracts and chemicals from mangroves are used mainly in folkloric medicine (e.g., bush medicine) for treatment of several diseases like contagious diseases, cardiovascular disorders and cancer, as well as in agriculture as pesticides (insecticides, herbicides and fungicides) where these practices continue until today (Bandaranayake, 2002). Promising antibacterial activity of ethyl acetate extract of *Avicennia marina* mature leaves (Abeysinghe *et al.,* 2006), methanol extract of *Excoecaria agallocha* leaves and shoots (Chandrasekaran *et al.,* 2009) and antifungal activity of methanol extract of *Exoecaria agallocha* and *Bruguiera gymnorrhiza* trunks (Kazuhiko, 2002) are some examples of pharmaceutical potential of mangrove plants. It has also been reported that plant crude extracts and natural compounds has antioxidant and antimicrobial activity (Barla *et al.,* 2007; Chua *et al.,* 2008; Keskin *et al.,* 2010; Jahan *et al.,* 2011). It is well documented that many types of plants contain compounds like phenols, alkaloids, peptides, fatty acid, essential oils, aldehydes and ethanol or water soluble compounds which have significant therapeutic application against viral, fungal and bacterial pathogens (Khan *et al.,* 2003; Pavrez *et al.,* 2005).

The antioxidants acting in the body defense system are categorized into three defense line systems according to their functions: preventive antioxidants, radical-scavenging antioxidants and repair and *de novo* antioxidants (Noguchi and Niki, 1999). In recent years, scientists tried to replace synthetic antioxidants with natural antioxidants from plant materials due to the carcinogenic properties of the synthetic antioxidants (Sasaki *et al.,* 2002).

Sonneratia (Sonneratiaceae) is a genus of mangrove plants belonging to the Lythraceae family. The genus *Sonneratia* consists of nine species in the tropical and subtropical regions worldwide (Wang and Chen, 2002). Different parts of *Sonneratia* are used as traditional

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medicine. In Malaysia, ripen fruits are used to remove intestinal parasites while half ripen fruits are use for the treatment of coughs. *Sonneratia alba* (also known as the mangrove apple) is the most widespread of the mangrove trees. They are found from East Africa through the Indian subcontinent, Southeast Asia, northern Australia, Borneo and Pacific Islands. The sepal of *S. alba* was reported as having antioxidant and anti-lipid peroxidation properties (Nuntavan *et al.,* 2003). The leaves, trunks and barks have also been reported for its antioxidant properties (Wada *et al.,* 2002). The ripen fruits of *S. alba* are said to taste like cheese.

To the best of our knowledge, no research on the antioxidant properties of *in vitro* grown explants and active fractions and antibacterial activities of *S. alba* has been carried out. In this study, we investigated the antioxidant effects/properties and antibacterial activity of methanol, ethanol and chloroform extracts and fractions of wild type and tissue culture explants of *S. alba* and carried out liquid chromatography tandem mass spectrometry (LC-MS/MS) analysis to identify active compounds.

Materials and Methods

Chemicals and Reagents

All the chemicals and reagents used in this study were purchased from Sigma Chemical Co. Ltd (USA). All other chemicals and solvents used in this study were of analytical grades obtained from BDH (Poole, UK).

Plant Material Collection and Extraction

Samples of leaves, barks and fruits of *S. alba* were carefully collected from a tree in Carey Island, Selangor, Malaysia. Seeds were collected after blooming of the fruits and were immediately sterilized. Seeds were cultured under *in vitro* and *in vivo* conditions. Under *in vivo* condition (uncontrolled condition), seeds were grown on sand and soil mixture. Under *in vitro* conditions, seeds were cultured on Murashige and Skoog (MS) and Woody Plant Medium (WPM) media. Three hormones were used, namely; 6-Benzylaminopurine (BAP), Gibberellic acid (GA_3) and Kinetin, with three concentrations and combinations of 100, 500 and 1000 mg L^{-1} for each. Seeds were soaked for 24 h in water. Hormone solutions were made using sterilized water (120°C for 20 min) before the seeds were soaked. Control cultures were also prepared for both wild type and tissue culture explants without any hormone. Seeds were cultured aseptically on 5 mL 2.8% phytagel medium with MS and WPM in 16 mL culture tubes. They were maintained under light conditions (photosynthetic photon flux density 80 µE m^{-2} s^{-1}) at 23–24°C. One seed was cultured in one culture tube covered with autoclaved translucent film. Both MS and WPM medium contained 3% (w/v) sucrose. We found that WPM media with 100 ppm BAP was the best condition for seed germination. Hundreds of seeds were then germinated with the same conditions and medium (5 mL of 2.8% phytagel medium with MS and WPM) to obtain enough plant materials to carry out all the experiments.

Explants Extracts

All the explants extracts were prepared according to the procedure of Haq *et al*. (2011). Roots of the seedlings were removed and the aerial parts were used as plant materials. Aerial parts were dewatered and dried in incubator at 40°C. Dried seedlings were ground using an electric blender and made into powder form which was then soaked in three selected solvents (95% ethanol, methanol and chloroform) and preserved in a dark place at room temperature for three days. Two L of solvents were used for extraction of 500g powder of each sample. Samples were filtered through Whatman No. 1 filter paper (Whatman International, England). Following filtration, the solutions were evaporated using evaporator at 40°C and the resulting plant extracts were dissolved in dimethylsulfoxide (DMSO). The procedures described herein were also used for leaves and barks collected from wild type *S. alba*.

Fractionation of Active Crude Ethanol Extracts

Solvent-solvent partitioning was done using the protocol designed by Kupchan (1973) and modified version of Van Wagenen (1993). Given that the sample containing ethanol extract of barks was the best extract amongst all the crude extracts, it was chosen for fractionation into polar, semi polar and non polar fractions. Firstly, ethanol extract was soaked in hexane solvent and was separated into hexane fraction and hexane insoluble fraction. Secondly, hexane insoluble fraction was partitioned with water and ethyl acetate in 1:1 ratio. To carry out the bioactivity tests, the resulting fractions were filtered through a fresh cotton plug followed by a Whatman No.1 filter paper and evaporated using a rotary evaporator at low temperature (40-45°C) and pressure.

Determination of Total Phenolic Content

Total phenolic content of various extracts of *S. alba* was determined through a protocol based on Folin-Coicalteau method described by Slinkard and Singleton (1977).

Determination of Superoxide Dismutase (SOD) Antioxidant Activity

Superoxide dismutase (SOD) assay was carried out using SOD determination kit according to the protocol provided by Sigma-Aldrich, at 37ºC for 20 min with double distilled water as blank. Absorbance was read at 450 nm using a

micro plate reader. The SOD activity was calculated as percentage inhibition.

Determination of DPPH antioxidant activity

Reducing power assay of all extracts was determined based on the method described by Sorenson *et al.* (1986). According to Haq *et al.* (2011) radical scavenging capacity assay was conducted, where DPPH* (1, 1-diphenyl-2-picrylhydrazil) was used as the free radical. Radical Scavenging Capacity (%) = [(Blank – Sample A)/Blank] x 100. The IC_{50} value for each extract was extrapolated from graphs plotted using the optical density (OD) value obtained.

Antibacterial Activities of Crude Extracts and Extracted Fractions of *S. alba*

The antibacterial activities of crude extracts and extracted fractions (5, 25, 50, 100, 250 µg/mL) of *S. alba* were tested against two Gram-positive (*Staphylococcus aureus*, *Bacillus cereus*) and two Gram-negative (*Escherichia coli*, *Pseudomonas aeruginosa*) laboratory bacteria. The bacterial strains were obtained from the Microbiology Division, Institute of Biological Sciences, University of Malaya. The agar disk diffusion method as described by Kil *et al*. (2009) was used. The sets of five dilutions (5, 25, 50, 100, and 250 µg/mL) of *S. alba* extracts were prepared in double-distilled water. Mueller-Hinton sterile agar plates were seeded with bacterial strains (108 cfu) and allowed to stay at 37°C for 3 h. Control experiment was carried out using tetracycline as the standard drug. The zones of growth inhibition around the disks were measured after 18 to 24 h of incubation at 37°C. The sensitivities of the microorganism species to the plant extracts were determined by measuring the sizes of inhibitory zones (including the diameter of disk) on the agar surface around the disks, and values <8 mm were considered as not active against microorganisms.

Liquid Chromatography-tandem Mass Spectrometry (LC-MS/MS) Analysis

LC-MS/MS analysis of water extracted fraction of *S. alba* was performed using an Applied Biosystems 3200Q Trap LC-MS/MS instrument. A Phenomenex Aqua C18 (50 mm × 2.0 mm × 5 µm) column was used for separation. The column temperature was set at 30°C. The mobile phase consisted of water (A) and acetonitrile (B) with 0.2% formic acid and 2mM ammonium formate. The initial condition was set at 10% A to 90% B from 0.01 min to 5.0 min, remained at this condition for 2 min, then back to 10% A in 0.1 min and re-equilibrated for 3 min. The total run time was 10 min at a flow rate of 0.25mL/min.

Statistical analysis

All experiments were carried out in triplicate. One-way analysis of variance (ANOVA) was used to analyze the data using MSAT-C statistical software. The means were compared by Duncan's Multiple Range Test (DMRT) and $p < 0.05$ was considered to indicate statistical significance.

Results

Germination of *Sonneratia alba* Seeds

Fig. 1 shows germination of *S. alba* seeds in different medium and hormone concentrations. Amongst all the medium and concentration used, WPM medium with BAP at a concentration of 100 ppm showed the highest germination rate. GA_3 with MS medium and WPM at the concentration of 500 ppm showed the lowest germination rate, which was also lower than the control.

Total Phenolic Contents and Antioxidant Activities of Crude Extracts of *S. alba*

Antioxidant activities of methanol, ethanol and chloroform extracts of wild type and tissue culture explants of *S. alba* were examined using SOD and DPPH reducing power

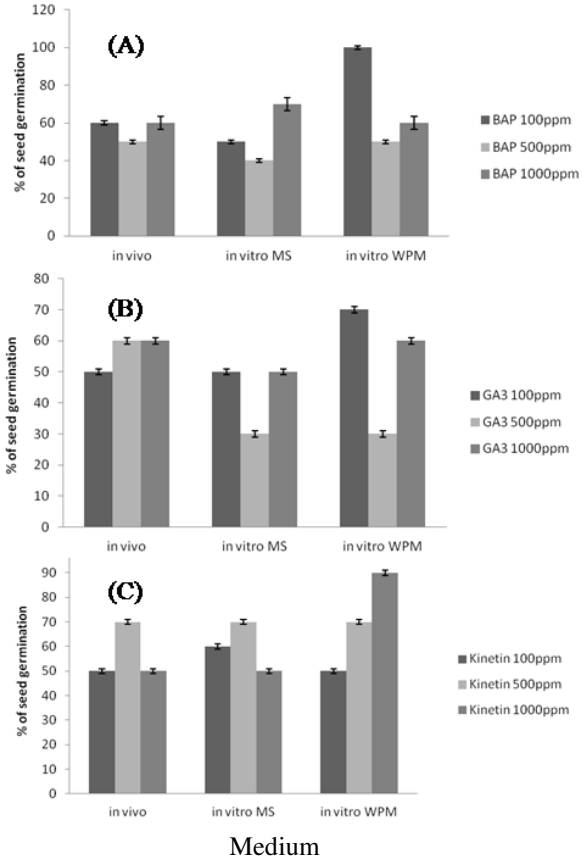

Fig. 1: Seed germination of *S. alba* in different tissue culture medium with different hormone concentrations. *In vivo* refers to soil grown seeds whilst *in vitro* refers to seeds gown in MS medium or WPM containing culture tubes

Table 1: Total phenolic contents and DPPH assay IC_{50} values of ethanol, methanol and chloroform extracts of wild type and tissue culture explants of *S. alba.*

Crude extracts of *S. alba*	Total phenolic contents (mg of GAE/g)	IC_{50} (mg/mL)
ML	216.53 ± 3.09 c	0.038 ± 0.003e
EL	205.93 ± 4.278cd	0.029 ± 0.004abcd
CL	23.60 ± 4.02 h	0.27 ± 0.017hi
MB	299.53 ± 4.91 ab	0.025 ± 0.003abc
EB	313.81 ± 5.54 a	0.0197 ± 0.001ab
CB	38.83 ± 2.95 g	0.193 ± 0.015h
MV	123.87 ± 3.24 e	0.073 ± 0.002f
EV	101.66 ± 2.58 f	0.092 ± 0.003fg
CV	07.82 ± 1.49 I	0.37 ± 0.004j
		0.018 ± 0.003a (Vit-C)

Samples presented with different alphabetic letters are significantly different ($p<0.05$).

Table 2: Antibacterial activity of methanol, ethanol and chloroform extracts of wild type and tissue culture explants of *S. alba*

	Zone of inhibition (cm)									
Bacteria	ML	MB	MV	EL	EB	EV	CL	CB	CV	Positive control
B. cereus	12.78	13.56	9.43	11.98	16.98	8.76	6.34	7.54	-	17.93
S. aureus	14.87	16.89	11.57	13.78	19.56	9.08	7.45	8.78	-	21.10
E. coli	10.65	12.34	8.43	9.65	13.78	9.45	5.78	6.24	-	14.30
P.aeruginosa	9.87	11.23	6.45	8.25	12.02	5.76	-	5.45	-	12.60

All the data are means of three replicates, Positive control: Tetracycline
MV= Methanol extract of explants, EV= Ethanol extracts of explants and CV= Chloroform extract of explants, ML= Methanol extract of leaves, EL= Ethanol extract of leaves, CL= Chloroform extract of leaves, MB= Methanol extract of barks, EB= Ethanol extract of barks and CB= Chloroform extract of barks.

Table 3: IC_{50} values obtained from DPPH assay of water and ethyl acetate extracted fractions of *S. alba.*

Sample	IC_{50} (mg/mL)
Water extracted fraction	0.024 ± 0.001 ab
Ethyl acetate extracted fraction	0.124 ± 0.001 c
Ascorbic acid	0.018 ± 0.004 a

Samples presented with different alphabetic letters are significantly different ($p<0.05$).

Table 4: Antibacterial activity of water and ethyl acetate extracted fractions of *S. alba*

	Zone of inhibition (cm)		
Bacteria	Water extracted fraction	Ethyl acetate extracted fraction	Positive control
B. cereus	13.55	9.65	17.93
S. aureus	16.75	13.35	21.10
E. coli	10.45	7.97	14.30
P. aeruginosa	9.85	7.85	12.60

All the data are means of three replicates, Positive control: Tetracycline

assays. Total phenolic content was also measured of all the extracts by using the Folin-Coicalteau method described by Slinkard and Singleton (1977). As shown in Table 1; all the methanol and ethanol extracts exhibited high phenolic content, whereas chloroform extracts exhibited low phenolic content. Ethanol extract of bark showed the highest

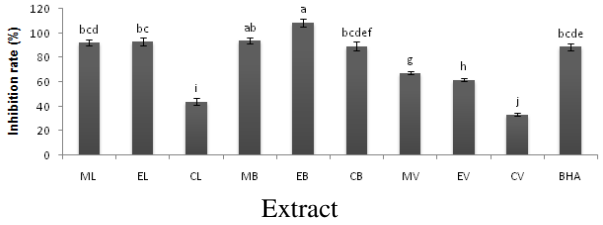

Fig. 2: Antioxidant activity of ethanol, methanol and chloroform extracts of wild type and tissue culture explants of *S. alba* using SOD assay. BHA was used as the positive control. Samples presented with different alphabetic letters are significantly different ($p<0.05$).

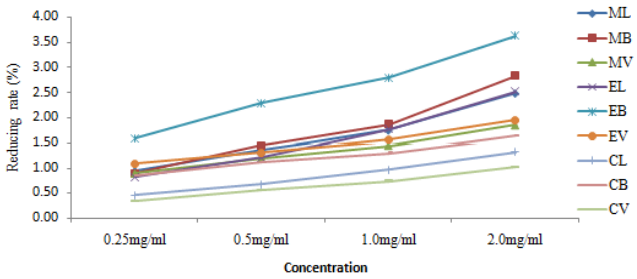

Fig. 3: Antioxidant activity of ethanol, methanol and chloroform extracts of wild type and tissue culture explants of *S. alba* using reducing power assay.
MV= Methanol extract of explants, EV= Ethanol extracts of explants and CV= Chloroform extract of explants, ML= Methanol extract of leaves, EL= Ethanol extract of leaves, CL= Chloroform extract of leaves, MB= Methanol extract of barks, EB= Ethanol extract of barks and CB= Chloroform extract of barks.

phenolic content, which was not significantly higher than methanol extract of bark but significantly higher than all other extracts. *In vitro* explants extracts showed comparatively lower phenolic contents than *in vivo* explants. Ethanol extract of bark and chloroform extract of *in vitro* explants exhibited significantly highest and lowest antioxidant activity in SOD assay (Fig. 2). The ethanol extract of bark also showed higher antioxidant activity than the positive control BHA.

Four concentrations have been used to evaluate the reducing power assay. The result of reducing power assay (Fig. 3) showed that when the concentrations increased the reducing rates also increased. Among all the extracts, ethanol extract of barks showed the highest reducing rate and chloroform extract showed the lowest reducing rate. Low concentration of extracts showed low reducing rate and high concentration of extracts showed high reducing rate in this assay. The IC_{50} values of methanol, ethanol and chloroform extracts of wild type and tissue culture explants is shown in Table 1. Ethanol extract of bark exhibited low IC_{50} value among all the crude extracts. However, ascorbic acid showed the lowest IC_{50} value but there was no

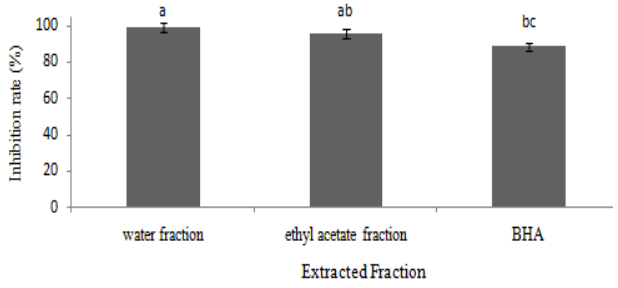

Fig. 4: Antioxidant activity of water and ethyl acetate extracted fractions of *S. alba* using SOD assay. BHA was used as the positive control. Samples presented with different alphabetic letters are significantly different ($p<0.05$)

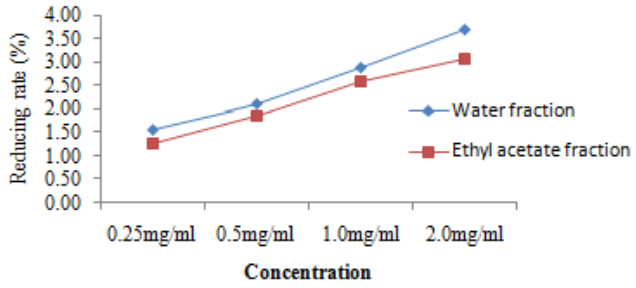

Fig. 5: Antioxidant activity of water and ethyl acetate extracted fractions of *S. alba* using DPPH reducing power assay

significant difference between ascorbic acid and ethanol extract of bark. Chloroform extracts of bark showed significantly high IC_{50} value compared with methanol and ethanol extracts of leaf and bark.

Antibacterial Activities of Crude Extracts of *Sonneratia alba*

The antibacterial activity of crude extracts of *S. alba* was evaluated against four laboratory strains of bacteria, including two Gram positive (*Staphylococcus aureus*, *Bacillus cereus*) and two Gram negative bacteria (*Escherichia coli*, *Pseudomonas aeruginosa*) (Table 2). All the extracts except chloroform extracts of tissue culture explants inhibited the growth of the tested bacteria. Ethanol extracts exhibited higher antimicrobial activity among all the crude extracts. The ethanol extract of bark showed the highest percentage reduction (92.7%) against *S. aureus*. Chloroform extract of leaves do not show any antimicrobial activity against *P. aeruginosa*. All the crude extracts and positive control showed high activity against *S. aureus* but low activity against *P. aureginosa*. Positive control (30 µg tetracycline, total volume of 50 µL) inhibited the growth of all tested bacteria significantly compared to the examined extracts. The presence of antibacterial activity in different extracts indicated that the extracts possess active compounds, which have different activities.

Antioxidant Activities of Ethyl–Acetate and Water Extracted Fractions

As shown earlier, the ethanol extract of bark was the best extract among all the extracts. The ethanol extract was then fractionated into polar, semi polar and non-polar fractions. The water extract showed better antioxidant and antimicrobial activities in all the tests compare with ethyl acetate extract. In superoxide dismutase (SOD) antioxidant assay (Fig. 4), water extracted fraction showed higher antioxidant activity than ethyl acetate and BHA. However, there was no significant difference between all the examined samples.

The IC_{50} value was calculated following DPPH assay experiments. Ascorbic acid was used as positive control. No significant difference between the IC_{50} values of water extracted fraction and ascorbic acid was observed (Table 3). The DPPH reducing power assay also showed that water extracted fraction of *S. alba* was a better fraction in antioxidant activity compared with ethyl acetate extracted fraction (Fig. 5). The reducing rate increased with increasing concentration, with 2 mg/mL showed the highest reducing rate both for water and ethyl acetate fraction.

Antibacterial Activity of Ethyl Acetate and Water Extracted Fractions

Table 4 shows the results obtained for the antibacterial activity analysis of the water and ethyl acetate extracted fractions. Though the activity was not as significant as the positive control, both fractions exhibited activity against all the four gram positive and negative bacteria. The percentage inhibition was not as high as the positive control (tetracycline). The highest inhibition zone was found for the water fraction against *S. aureus* (16.75 cm) while the lowest inhibition zone was exhibited by the ethyl acetate fraction (7.85 cm) against *P. aeruginosa*. The ethyl acetate fraction showed less antibacterial activity than the water fraction.

LC-MS/MS Analysis of Water Extracted Fraction

Liquid chromatography tandem mass spectrometries (LC-MS/MS) was used for detection of nominal mass of the active compound of water fraction of *S. alba* (Fig. 6). We studied the nominal masses of the major active components in the water fraction, and we observed 8 such peaks at m/z values of 331, 345, 452, 456, 463, 473, 480 and 494. Each of the 8 major peaks observed were found to have rich fragmentation patterns from the MS/MS spectrum and this unique patterns (also known as fingerprint) corresponds to the high antioxidant activity of the extracted water fraction of the plant sample.

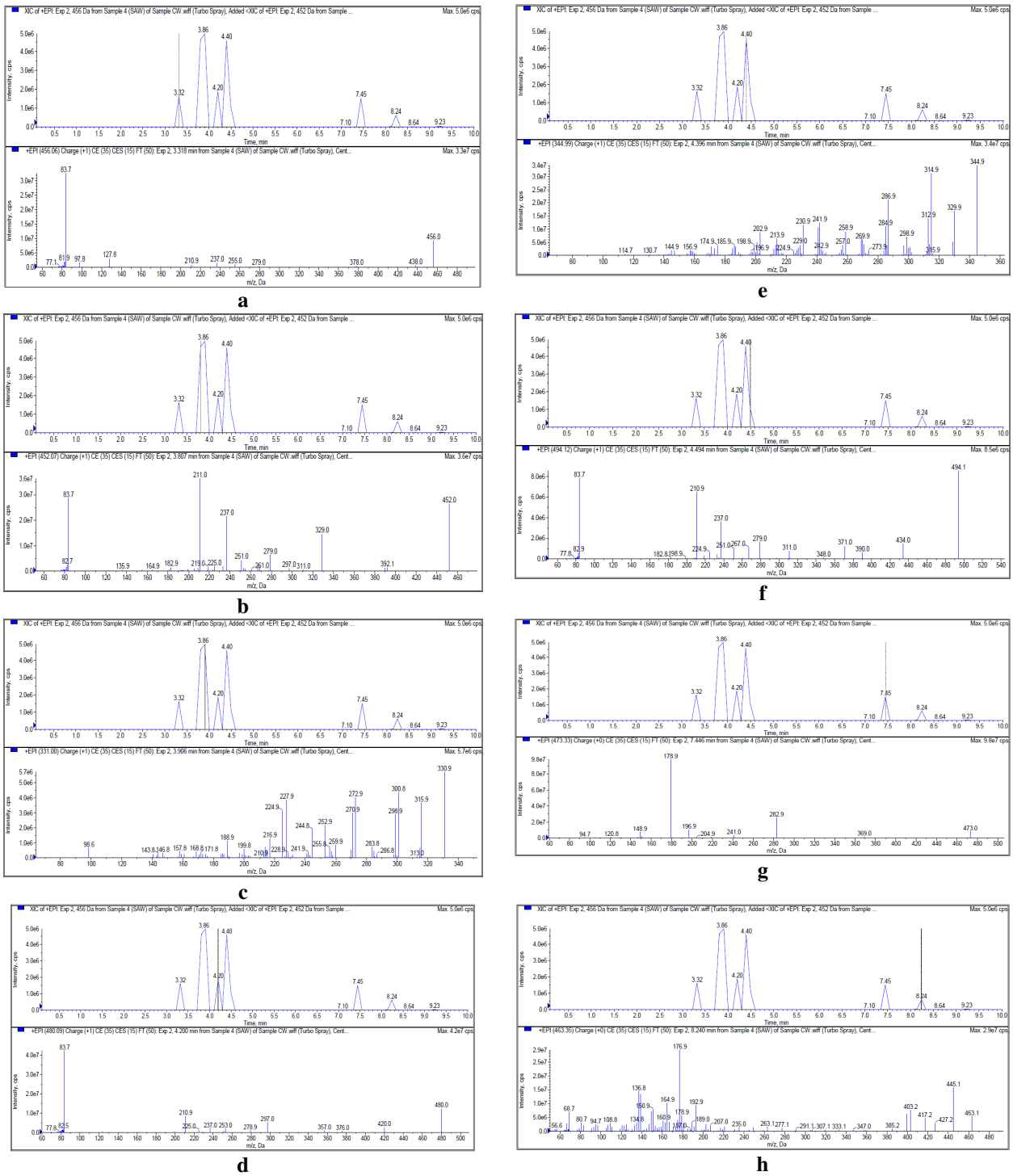

Fig. 6 (a-h): Fragmentation pattern of the major active components in the water fraction from MS/MS spectrum of the major peaks with m/z values of (a) 456; (b) 452; (c) 331, (d) 480; (e) 345; (f) 494; (g) 473; (h) 464.

Discussion

In our study, we have found that cytokinins such as BAP and kinetin are required for seed germination of *Sonneratia alba. M*edium supplemented with BAP can play important role in dormancy breaking and enhance the germination rate in seeds (Otroshy *et al.,* 2009).

Our results also showed that the extraction solvent significantly affected the total phenolic content of the resulting extracts. For example, methanol and ethanol

extracts exhibited better activity compared with chloroform extracts. These results were agreement with previous studies, which showed that methanol can extract the highest amount of phenolic compounds compare to petroleum ether, ethyl acetate and water from both their examined samples, *Lentinus edodes* and *Volvariella volvacea* (Cheung *et al.,* 2003). *S. alba* can be introduced as a high value source of phenolic compound due to the high amount of this group of compounds. Comparison of amounts of phenolic compounds in different parts of Indian *Sonneratia* spp. was carried out previously by Banerjee *et al*. (2008) and it was shown that leaf explants of *S. apetala* contain the highest amounts of phenolic compounds compared to the stem barks and roots. However, in our study we found that the phenolic compound is higher in barks than leaves. Banerjee *et al.* (2008) also reported that *Bruguiera gymnorrhiza* and some other mangrove plants contain the higher amount of phenols in barks than the leaves.

Earlier research on *Sonneratia* species mainly focused on morphological and anatomical features (Baker and Van Steenis, 1951; Duke and Jackes, 1987; Ko, 1993; Chen, 1996; Wang and Chen, 2002; Qin *et al.,* 2007). More recently, studies on the phytochemistry and pharmacology of *Sonneratia* species have also been reported, for example triterpenoids and sterols from the aerial parts of *S. apetala* (Ji *et al*., 2005); two flavonoids and their antioxidant activity from the leaves of *S. caseolaris* (Sadhu *et al*, 2006); five triterpenoids together with two sterols from the stems of *S. ovata* and unknown chemical constituents from the fruits of *S. caseolaris* and *S. ovata* (Zheng and Pei, 2008); and nine compounds from the fruits of *S. caseolaris* and seven from the fruits of *S. ovata* (Wu *et al*., 2009).

The study showed a positive correlation between the total phenolic contents and the antioxidant activity of wild type and tissue explants of *S. alba*, as was reported recently in *Teucrium stocksianum* flower (Rahim *et al.,* 2013). The study also confirmed that the extraction solvent in preparation of sample has an important effect on the antioxidant activity.

Zhou and Yu (2004) obtained four pure and solvent mixture extracts and they found that pure ethanol had the lowest polyphenols content in wheat bran grain sample. However, we report here that ethanol extract exhibited the highest antioxidant activities in both leaves and barks samples. Ethanol extract of barks of *S. alba* has been fractionated into three fractions which were water, ethyl acetate and hexane extracted fractions. The amount of hexane extracted fraction was too little (0.3 g from 50 g of crude extract) and thus omitted for further bioactivity tests. Both the water and ethyl acetate extracted fractions were used to evaluate the antioxidant effects and antibacterial activities. Water extracted fraction showed the highest bioactivities compared with ethyl acetate extracted fraction. Khokhar and Magnusdottir (2002) reported that water was the best solvent for extraction of tea catechins, followed by 80% methanol or 70% ethanol. We found that the water extract fraction of ethanol extracts of leave of *S. alba* exhibited better antioxidant activity based on both the enzymatic (SOD activity assay) and non-enzymatic (reducing power) assays. In order to understand more about the nature of the active compounds in the water extracted fractions, we studied the nominal masses of the major active components in the water fraction, and we observed 8 such peaks at m/z values of 331, 345, 452, 463, 473, 480 and 494. Each of the 8 major peaks observed were found to have rich fragmentation patterns from the MS/MS spectrum and this unique patterns (also known as fingerprint) corresponds to the high antioxidant activity of the water extracted fraction of the plant sample.

All the crude extracts and fractions of *S. alba* exhibited antioxidant properties and antibacterial activities. High phenolic content could be responsible for the high activity of all assays. Extracts from *S. alba* has the potential to as antioxidant and antibacterial natural sources for the biomedical, healthcare, food and agriculture industries. The ethanol extract of the bark and water extracted fractions exhibited promising antioxidant properties and antibacterial activities. LC-MS/MS analysis revealed the nominal mass of the yet unidentified compounds of the extracted water fraction that exhibited high antioxidant activity. The identification of these active compounds from *S. alba* merit further study.

Acknowledgements

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