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INHERENT ANTIOXIDANT POTENTIAL OF AVICENNIA MARINA LEAVES

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ABSTRACT

Antioxidants inhibit the reaction caused by free radicals and prevent the cells and tissues from damage. By taking enough amounts of exogenous antioxidants, it will increase protection against free radicals. In the present investigation antioxidant properties of leaves of *Avicennia marina* was determined for their potential benefits for health. The antioxidant activity was assessed by DPPH scavenging activity. The methanolic extract of leaves of *A. marina* had shown very significant DPPH (1, 1-diphenyl-2-picrylhydrazyl) radical scavenging activity. It was observed that DPPH radical scavenging activity increased with the increasing concentration of the plant extract. The sample ML3 exhibited highest $68.12 \pm 0.03\%$ antioxidant activity at 1000 µg/mL and ML4 showed the lowest antioxidant activity 20.06 \pm 0.04 % at 125 µg/mL.Therefore, in vitro assays indicate that *A. marina* leaves extracts are a better source of natural antioxidant, which might be helpful in preventing the progress of various oxidative stresses. The results proved the potential of *A. marina* leaves for utilization as significant source of natural antioxidant.

Keywords: DPPH, Avicennia Marina, Antioxidant Activity, Oxidative Stress, Medicinal Plants.

I. INTRODUCTION

It has long been conceding that in higher plants, naturally occurring substances have antioxidant activity. Attention is being focused on the protective biochemical functions of naturally occurring antioxidants in the cells of the organisms containing them (Larson, 1988). An antioxidant is a molecule, capable of inhibiting other molecular oxidation. Antioxidants are radical scavengers which protect the human body against free radicals that may cause pathological conditions such as ischemia, anemias, asthma, arthritis, inflammation, neuro-degeneration, Parkinson's diseases, mongolism, ageing process and perhaps dementias (Oke et al, 2002). There are copious enzyme systems in the human body, for free radical scavenging, but micronutrients such as beta-carotene vitamin C and vitamin E are the major antioxidants. Human body cannot produce these nutrients and must be provided in diet. Insufficient levels of endogenous and exogenous antioxidants can cause oxidative stress and an imbalance of oxidants and antioxidants resulting in cellular damage or death (Valko et al, 2007 and Uttara et al, 2009). By taking ample amounts of exogenous antioxidants, protection can be enhanced against free radicals (Halliwell 1995).

A plentiful number of both natural and synthetic antioxidants are recommended for various human diseases (Cuzzocrea 2001). The most referenced synthetic antioxidants in the food industry are butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), propyl gallate (PG) and tetra-butyl hydroquinone (TBHQ). Nevertheless, synthetic antioxidants have shown potential toxicity and health risks, most notably carcinogenicity. There is growing interest toward natural antioxidants from herbal sources (Larson, 1988; Gazzani et al, 1988; Veliglu et al, 1988). Plant has been badly neglected for scientific studies (Islam et al, 2019). Epidemiological and in vitro studies on medicinal plants and vegetables strongly have supported the idea that plant constituents with antioxidant activity are capable of exerting protective effects against oxidative stress in biological systems (Cao et al, 1996; Block et al, 1992; Ness et al, 1997).Therefore, it is of significant important to find natural sources of safe and inexpensive antioxidants and use them in foods and pharmaceutical preparations to replace synthetic antioxidants (Lee et al. 2004, Song 2010). Therefore, in the present study the antioxidant activity of the *A. marina* was evaluated for their inherent capability for health benefits.

In our previous study we investigated that ghaf pods and leaves has a potential antioxidant activity and nutraceutical properties (Bhardwaj. V 2021b, c). Furthermore, to continue our research on plants we probed that *Avicennia marina* is a novel convivial phyto medicine for antibiotic resistant pathogenic bacteria (Bhardwaj. V 2021a). Moreover, we continue further research to detect the potency of other plants as source of antioxidative agent. With this view, the present investigation was initiated to study the antioxidant activity of methanolic extract of leaves of *Avicennia marina* which was evaluated by in vitro free radical scavenging



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activity. We need to spread awareness on the importance of these plants and the role in the functioning of a healthy ecosystem and thereby protecting the species from extinction.

II. **MATERIAL AND METHODS**

2.1 Sample collection

Samples of leaves of A. marina (samples taken from four plants) were collected from plants grown on Khuzam road, Ras Al Khaimah, UAE in the month of March 2021. The leaves were sun dried for 5-7 days or more and then oven dried for better grinding. The dried leaves and pods were then ground to a coarse powder using high capacity of grinding machine and then stored in airtight bottles.

2.2 Preparation of the extracts

About 5 g of the coarse powder was extracted with 25.0 ml of methanol followed by continuous hot extraction method (Bhardwaj V 2021c). Stirred well and kept for incubation in closed container. Then we centrifuged the tubes at 4000 rpm for 30 min. After that we transferred the supernatant extract for drying for 10 minutes until dry powder was obtained. We finally got residue of samples (leaves). All the extracts were then stored in refrigerator at -20°C till use (Al Ghais et al 2020a, b, c).

2.3 Chemicals

The chemicals used in the present investigation were of analytical grade and of high purity from Merck. Standard reagents used for analysis were purchased from Germany and USA.

2.4 DPPH photometric assay

The plant methanolic extracts were prepared in different concentrations 125, 250, 500, 1000 μg/mL to analyse the antioxidant property by using DPPH radical scavenging assay method which was described in our previous research paper (Bhardwaj, V 2021c). The measurement of the DPPH radical scavenging activity was performed according to methodology described by Brand-Williams et al. 1995. The scavenging activity percentage (AA%) was determined according to Mensor et al. 2001:

Scavenging activity (%) = ($[A_{517} \text{ of control} - A_{517} \text{ of test sample}] / A_{517} \text{ of control}) \times 100$.

Where A_{517} control is the absorbance of DPPH radical+ methanol; A_{517} test sample is the absorbance of DPPH radical+ sample extract.

2.5 Statistical analysis

Data are expressed as mean. Pair wise comparisons were performed. Standard error mean was determined for triplicate and expressed as ± SEM.

III. **RESULTS AND DISCUSSION**

3.1 Determination of DPPH scavenging activity

The DPPH Free-radical scavenging assay was significant between the concentrations of their extracts (P< 0.05) (Table 1). The decrease in absorbance of the radical was due to hydrogen donation. According to our research, we found that there was a change in colour of solution in test tubes, when samples of extracts of A. marina leaves are mixed with DPPH reagent solution the colour was turned from purple to yellow by time (Figure 1). Further, the antioxidant activity of samples was determined by measuring absorbance with a spectrophotometer at 517 nm. The percentage of DPPH scavenging activity of methanolic extract of leaves of A. marina was presented in Table 1. According to our research finding the sample ML3 exhibited highest 68.12 ± 0.03% antioxidant activity at 1000 μ g/mL as compared to other extracts and ML4 showed the lowest antioxidant activity 20.06 \pm 0.04 % at 125 μ g/mL (Table 1).



Figure 1: DPPH radical scavenging activity observed by change in the colour from purple (control) to yellow (Test).

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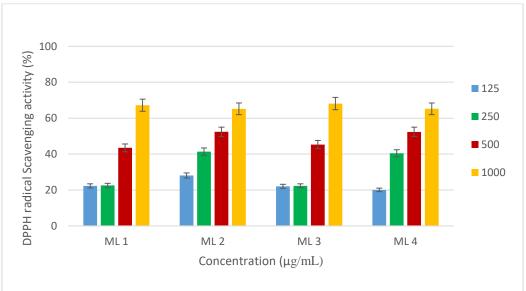
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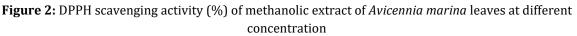
Similar results were reported by Malik et al, 2013, Banerjee et al, 2008 and Jaslin et al, 2011. According to the analysis results, the total antioxidant activity was significant between four Mangrove plant leaves sample (ML1, ML2, ML3 and ML4) and also between the concentrations of their extracts (P<0.05). Moreover, the analysis results graph (Figure 2) shows that the antioxidant activity increased with increasing concentrations of the extracts from $125(\mu g/ml)$ to $1000(\mu g/ml)$.

Table 1. DPPH Antioxidant scavenging activity of methanolic extract of Avicennia marina leaves at different concentration

SNo	Concentration (µg/ml)	*DPPH scavenging activity of different Mangrove leaves (ML) samples (%)			
		ML 1	ML 2	ML 3	ML 4
1	125	22.3 ± 3.05	28.09 ± 0.03	22.06 ± 1.05	20.06 ± 0.04
2	250	22.6 ± 0.02	41.38 ± 1.02	22.32 ± 0.042	40.43 ± 2.01
3	500	43.46 ± 2.04	52.37 ± 0.025	45.32 ± 1.04	52.34 ± 0.01
4	1000	67.199 ± 0.02	65.16 ± 1.05	68.12 ± 0.03	65.20 ± 2.02

*All values are expressed as mean ± SEM for triplicates





IV. **CONCLUSION**

In summary, this research work demonstrate that leaves of Avicennia marina has remarkable antioxidant activity. According to our research findings, we hope that the potential of Avicennia marina leaves could be best connected, towards a possible beneficial integration in food and pharmaceutical industry and also to spread awareness on the importance of medicinal plants and the role they played in the functioning of a healthy ecosystem and this will also help in protecting the species from extinction. In this regard, there would be a need of further study on purification of the crude extracts and to discover the antioxidant compounds present in the leaves of mangrove.

Ethics Approval And Consent To Participate

Not applicable.

Consent for publication

Not applicable.

Availability of data and materials

The relevant data and materials are available in the present study.



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Competing interests

The authors declare that they have no competing interests. All procedures followed were in accordance with the ethical standards (institutional and national).

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