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ORIGINAL ARTICLE



Phyto-nutritive component identification of selected mangroves (Avicennia officinalis and Rhizophora mucronata) through HPLC and GC

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ABSTRACT

Mangroves are characteristic elements of the coastal areas in the tropical and subtropical regions forming essential components of the intertidal ecosystems and are self-maintaining estuarine components of the biosphere. In this study, Avicennia officinalis and Rhizophora mucronata were investigated and found the presence of nutritive compounds such as carbohydrates, proteins, amino acids, lipids, fatty acids and minerals. The carbohydrate content of R. mucronata stem, lipid content of A. officinalis root, amino acid content of R. mucronata leaf and protein content of R. mucronata leaf were showed high values respectively. This study is also revealed the presence of various kinds of minerals from A. officinalis and R. mucronata.

Keywords: Avicennia officinalis, Rhizophora mucronata, GC, HPLC, Punnakayal.

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INTRODUCTION

Mangroves are a taxonomically varied genus of halophytic plants found in tropical and subtropical intertidal zones between land and sea [1]. The mangrove forest is a unique, rare, and threatened habitat with a diverse range of species [2]. Mangrove wetlands are one of the world's most prolific biomes [3]. Fevers, bodily ache, boils, and tumours are among the illnesses for which Indian mangroves are employed as a traditional treatment [4]. Mangroves are biochemically unique, producing a diverse spectrum of natural compounds that are frequently used for a variety of human ailments [5]. Natural chemicals derived from mangrove plants are less expensive and may have a more holistic effect; therefore, evaluating traditional knowledge for medicine development is critical [6]. Many bioactive chemicals with toxicological, pharmacological, and ecological significance can be found in them. These substances are made by a living organism's primary or secondary metabolism. Secondary metabolites are a group of chemically and taxonomically varied molecules with unknown functions [7]. Mangroves are a source of various bioactive substances that have been utilised in folkloric remedies, and extracts from them have been shown to be effective against human, animal, and plant infections [8] [9]. Phytochemicals, anti-diabetic, and free radical scavenging activities are found in the mangrove, A. officinalis [10]. The leaf extracts of R. mucronata, a mangrove plant, show antibacterial and antioxidant properties [5]. Hence the current study was conducted to measure the nutritional content of a variety of mangrove plants in Punnakayal estuary.

MATERIAL AND METHODS

Collection of mangroves

A. officinalis and *R. mucronata* were collected from Punnakayal estuary and washed with water, air-dried and powdered in an electrical miller, sieved and stored.

Estimation of biomolecules and nutrients

Total carbohydrate [11], total soluble protein [12], free amino acids [13] were studied using respective methods. Free protein amino acids were estimated by O-pthaldialdehyde method described by [14] using high performance liquid chromatography (HPLC) (LACHROM L-700 and D-70000 HPLC). Lipid content of selected plants was determined using [15]. Fatty acids were estimated [16] and fatty acid methyl esters (FAMEs) evaluated using gas chromatography (ASHMACO, Japan; Model No: ABD20A). Estimation of K, Ca,

Mg and Na were done using flame photometer Model 125 [17]; iron, zinc, copper, mercury and cadmium were analyzed by atomic absorption spectrophotometer (AAS 6300) [18].

RESULT AND DISCUSSION

All human being requires a number of complex organic compounds as added caloric requirements to meet the need for their muscular activities. In the present study, reveals the presence of nutritive compounds in the selected mangroves (*A. officinalis* and *R. mucronata*).

Carbohydrate

Carbohydrates are a common source of energy in living organisms [19]. In this study high amount of carbohydrate were observed in *R. mucronata* stem (90.92 mg/g DW) and low amount of carbohydrates was noted in *A. officinalis* root (14.02 mg/g DW). Moreover, the other studied parts also had a considerable amount of carbohydrates. The results of the present investigation comply with that of Suganthy and Devi [20]. Plant sugars can be used as artificial sweetener and they can even help in diabetes by supporting the body in its rebuilding [21].

Protein

Proteins are complex organic compounds of high molecular weight which are involved to make up different tissues and organs in the body. In this present study, high protein content was seen in *R. mucronata* leaf (541.99 mg/g DW) and very low content was present in *A. officinalis* root (221.43 mg/g DW). The presence of higher protein level in the plant parts towards their possible increase food value or that a protein base bioactive compound could also be isolated in future [20].

Amino acid

Amino acid content indicated that more amount of free amino acid present in R. mucronata leaf (43.1 mg/100g DW) and R. mucronata root (33.76 mg/100g DW). Less amount of amino acid was noted in A. officinalis root (10.3 mg/100g DW) and R. mucronata stem (17.7 mg/100g DW). The nutritive evaluation of mangroves on the basis of amino acid composition was carried out by using high performance liquid chromatography. Data of essential amino acids and non-essential amino acids of investigated mangroves were presented in the Table 2. The quantity of essential and non-essential amino acids was estimated. The significant amount essential amino acids like isoleucine (0.97 mg/100g DW), leucine (4.25 mg/100g DW), lysine (23.66 mg/100g DW), cystine (0.2 mg/100g DW), methionine (23.29 mg/100g DW), tyrosine (5.46 mg/100g DW), phenylalanine (4.39 mg/100g DW), threonine (1.71 mg/100g DW), valine (3.44 mg/100g DW), histidine (2.56 mg/100g DW), and arginine (2.12 mg/100g DW) was identified in A. officinalis. Nonessential amino acids like glutamic acid (11.66 mg/100g DW), serine (3.74 mg/100g DW), proline (9.45 mg/100g DW), glycine (0.68 mg/100g DW) and alanine (0.47 mg/100g DW) were also estimated. R. mucronata showed slight variation in amino acid composition. For example, in R. mucronata, the isoleucine (0.87 mg/100g DW), leucine (3.81 mg/100g DW), lysine (22.5 mg/100g DW), cystine (0.32 mg/100g DW), methionine (21.88 mg/100g DW), tyrosine (4.69 mg/100g DW), phenylalanine (3.27 mg/100g DW), threonine (0.78 mg/100g DW), valine (2.67 mg/100g DW), histidine (2.81 mg/100g DW), and arginine (1.6 mg/100g DW) were identified. Non-essential amino acids like glutamic acid (11.35 mg/100g DW), serine (7.12 mg/100g DW), proline (9.65 mg/100g DW), glysine (0.95 mg/100g DW) and alanine (0.56 mg/100g DW) were also estimated. Comparatively R. mucronata showed good composition of amino acids than A. officinalis. Similar results have been obtained in previous studies [20]. Amino acids are essential in the synthesis of proteins and precursors in the formation of secondary metabolites that participate in cell signaling, gene expression and homeostasis regulation, protein phosphorylation, synthesis of hormones and antioxidant capacity [22].

Lipid

Lipids are providing energy in oxidation processes than any other biological compounds. It constitutes a convenient storage material for living organisms. Lipid content was more in *A. officinalis* root (0.237 mg/g DW) followed by *A. officinalis* leaf (0.0305 mg/g DW) whereas least amount of lipid presents in *R. mucronata* leaf (0.0043 mg/g DW). The results of the present investigation comply with that of Suganthy and Devi [20].

Fatty acid composition of selected mangroves

Fatty acids are widely occurring in natural fats and dietary oils and they play an important role as nutrition and metabolism of living organism [23]. In the present study, the fatty acids profile of two mangroves plants such as *A. officinalis* and *R. mucronata* were identified. The chromatograms of fatty acids profile mangroves (*A. officinalis* and *R. mucronata*) were shown in the figure 1 and 2 and detailed data were reported in table (3). The fatty acids detected in selected mangroves were palmitic (C 16:0), stearic acid (C18:0), oleic acid (C18:1 n-9), linoleic acid (C18:2n-6) and alpha linolenic acid (C18:3, n-3) which totally ranged between (2.84 mg/100g DW) to (9.58 mg/100g DW). Highest amount of total fatty acids was present in *R. mucronata* (28.3 mg/100g DW) and low amount was present in *A. officinalis* (26.28 mg/100g DW). The fatty acids

distribution of mangrove tissues proclaims the terrestrial character of these plants in that carbon number ranges from 12:0 to at least 30:0; 16:0, 18:0, 18:1, 18:2 and 18:3 are the major fatty acids present in selected mangroves. The present data supported by earlier results. Palmitic acid is high amount in *R. mucronata* (9.58 mg/100g DW) and *A. officinalis* (8.61 mg/100 g DW). Stearic acid was found to be more in *A. officinalis* (4.37 mg/100g DW) and the fewer amounts in *R. mucronata* (3.21 mg/100g DW). Oleic acid is one of the mono unsaturated fatty acids. Monounsaturated fatty acid (oleic acid) was predominantly present in *R. mucronata* (7.04 mg/100g DW). Linoleic acid (C18:2) and alpha linolenic acids (C18:3) are two poly unsaturated fatty acids which cannot be synthesized by humans and other vertebrates. Highest linoleic acid was found in *R. mucronata* (5.63 mg/100g DW). Alpha linolenic acid was present in both mangroves. In food and nutrition evaluation, fatty acids have immense biological importance. In pharmacology and disease diagnosis, fatty acids also have key significance. The unsaturated (monounsaturated and poly unsaturated) fatty acids are frequently used for detecting heart disease risks, inflammation and increasing the immunity [24] [25] [26].



Fig. 2 Chromatogram showing the fatty acid profile of *R. mucronata*

Minerals

Minerals play several important roles in human physiology and biochemistry as cofactors for enzymes, and are related to energetic efficiency, fertility, mental stability, and immunity. The present study investigated many minerals which were reported in Table. 4. This study revealed that A. officinalis (0.611 ppm) and R. *mucronata* contains (380.21 ppm) showed significant amount of iron. In humans, iron is essential for hundreds of enzymes and proteins. Iron deficiency causes anemia, weakness, depression, poor resistance to infections [27] [28]. Chromium is vital element as it works with insulin to stabilize blood sugar level, help to absorb energy from blood and increase muscle mass by reducing fat mass in human body [29]. More amount of chromium was noted in A. officinalis (0.044 ppm) while R. mucronata contains fewer amounts (0.025 ppm). Zinc maintains various reaction of body, which helps to construct and maintain DNA, required for growth of body tissues, important element of ligaments and tendons. The high amount of Zinc recorded in A. officinalis (0.259 ppm) and low in R. mucronata (0.130 ppm). Copper is an important component of many enzyme systems such as cytochrome oxidase, lysyl oxidase and ceruloplasmin, an iron oxidizing enzyme in blood. The amount of Copper (Cu) is present in A. officinalis (0.030 ppm) and R. mucronata (0.022 ppm). Cu deficiency has been associated with cardiac abnormalities in human and animal; causes anaemia and neutropenia [30]. Manganese is an essential element in the bone development and in the metabolism of amino acids, carbohydrates, and cholesterol. Manganese was found in A. officinalis (0.127 ppm) and R. mucronata (562.21 ppm). Deficiency of manganese in human causes myocardial infection and other cardiovascular diseases, also disorder of bony cartilaginous growth in infants and children and may lead to immunodeficiency disorder and rheumatic arthritis in adults [31]. Both mangrove species showed same

amount of Nickel (i.e) 0.022 ppm. Its deficiency results in the disorder of liver in higher animals [31]. Arsenic is an essential element for animals and human body metabolism. Arsenic level reported in A. officinalis (0.020 ppm) and R. mucronata (0.024 ppm). Arsenic recently has been considered essential in human diets [32]. Level of Molybdenum reported in A. officinalis (0.016 ppm) and R. mucronata (0.003 ppm). Molybdenum assists the body by fighting nitrosamines, which are associated with cancer, may prevent cavities and may help to prevent aneamia [33]. R. mucronata (5.98 ppm) alone have cadmium of the selected mangrove species whereas cadmium is absent in A. officinalis. Cadmium causes acute and chronic poisoning, adverse effect on kidney, liver, vascular and immune system [34] [35]. The level of lead (Pb) is observed in *A. officinalis* (0.017 ppm) and *R. mucronata* (0.012 ppm). However, for medicinal herbs limit was 10 ppm set by China, Malaysia, Thailand and WHO. Lead causes both acute and chronic poisoning and also poses adverse effects on kidney, liver, vascular and immune system [34]. Lead is non-essential trace elements having functions neither in human's body nor in plants. They induce various toxic effects in humans at low doses [31]. Selenium is observed in A. officinalis (0.410 ppm) and it was absent in R. *mucronata*. Potassium is an essential plant nutrient and is required in large amounts for proper growth and reproduction of plants. A. officinalis contained high amount of potassium (1.765 ppm) and R. mucronata (0.917 ppm) contained low amount. Magnesium is a macronutrient that is necessary to both plant growth and health [36]. The result of the present study clearly indicated that A. officinalis (0.359 ppm) contained comparatively higher amount of magnesium content than R. mucronata (0.196 ppm). Sodium concentrations of selected mangrove plants are 0.285 ppm and 0.55 ppm in *R. mucronala* and *A. officinalis* respectively. Sodium and potassium involved in acid - base balance and transfer of nutrients in and out of individual cells [37]. Beryllium can reduce the magnesium requirement of plants by some 60% within a certain range of magnesium deficiency. The maximum level of Beryllium was observed in A. officinalis (0.69 ppm) and minimum level in *R. mucronata* (0.10 ppm). The residual obligatory magnesium requirement is probably accounted for by chlorophyll since beryllium appears to have no primary effect on chlorophyll or chlorophyll production [20] [38]. The good mineral composition of selected mangroves was suggested that the selected mangrove plants are best source for pharmaceutical and nutraceutical supplement in both lower and higher-level living organisms.

Tuble 1. Nutrients of Scietted mangiove species					
S. No.	Samples	Carbohydrates mg/g DW	Protein mg/g DW	Amino acids mg/g DW	Lipids mg/g DW
1.	A. officinalis leaf	39.02 ± 0.13	436.11± 0.176	20.3 ± 0.0765	0.0305 ± 0.0232
2.	A. officinalis stem	37.02 ± 0.17	512.3 ± 0.0129	23.3 ± 0.0578	0.0165 ± 0.0711
3.	A. officinalis root	14.02 ± 0.09	221.43 ±0.076	10.3 ± 0.0913	0.237 ± 0.0654
4.	R. mucronata leaf	41.54 ± 0.016	541.66 ± 0.0531	43.1 ± 0.0013	0.0043 ± 0.167
5.	R. mucronata stem	90.92 ± 0.0059	326.59 ± 0.0194	17.7 ± 0.0043	0.015 ± 0.00123
6.	<i>R. mucronata</i> root	14.33 ± 0.0078	244.44 ± 0.0620	33.76 ± 0.0127	0.029 ± 0.0923

Table 1: Nutrients of selected mangrove species

Values are the mean of 3 replicates ±SD and are expressed in mg/g DW.

Table 2: Amino acids	composition in A.	officinalis	and R. mucronata

Amino acids mg/100g DW		A. officinalis	R. mucronata
	Isoleucine	0.97 ± 0.01	0.87 ± 0.06
	Leucine	4.25 ± 0.22	3.81 ± 0.04
	Lysine	23.66 ± 0.03	22.5 ± 3.96
	Cystine	0.2 ± 0.06	0.32 ± 0.03
Econtial	Methionine	23.29 ± 0.22	21.88 ± 3.06
Essential	Tyrosine	5.46 ± 0.13	4.69 ± 0.11
amino acius	Phenylalanine	4.39 ± 0.32	3.27 ± 0.10
	Threonine	1.71 ± 0.11	0.78 ± 0.31
	Valine	3.44 ± 0.20	2.67 ± 0.19
	Histidine	2.56 ± 0.39	2.81 ± 0.13
	Arginine	2.12 ± 0.07	1.6 ± 0.49
	Glutamic acid	11.66 ± 0.03	11.35 ± 0.50
Non	Serine	3.74 ± 0.02	7.12 ± 0.11
essential	Proline	9.45 ± 0.04	9.65 ± 0.16
amino acid	Glycine	0.68 ± 0.46	0.95 ± 0.02
	Alanine	0.47 ± 0.04	0.56 ± 0.02
Total AA		97.8	94.93
EAA		71.85	64.88
Non-EAA		2.62	29.95
EAA/Non-EAA		2.7423	2.166

Carbon no.	Components	A. officinalis	R. mucronata	
Saturated fatty acid				
C16:0	Palmitic	8.61	9.58	
C18:0	Stearic	4.37	3.21	
Mono unsaturated fatty acid				
C18:1	Oleic	5.23	7.04	
Poly unsaturated fatty acid				
C18:2	Linolenic	4.89	5.63	
C18:3	Alpha Linolenic	3.18	2.84	
Total saturated fatty acid		12.98	12.79	
Total unsaturated fatty acid		13.3	15.51	
Total fatty acid		26.28	28.3	

Table 3: Fatty acids composition of A. officinalis and R. mucronata

Table 4: Mineral	l element in selected	mangroves of Punn	akaval. Gulf of Manna	ar

Mineral elements mg/g DW	A .officinalis (ppm)	<i>R. mucronata</i> (ppm)
Beryllium	0.69	0.10
Sodium	0.55	0.285
Magnesium	0.359	0.196
Aluminum	1.468	0.601
Potassium	1.765	0.917
Chromium	0.044	0.025
Manganese	0.127	562.21
Iron	0.611	380.21
Nickel	0.022	0.022
Copper	0.030	0.022
Zinc	0.259	0.130
Arsenic	0.020	0.024
Selenium	0.410	Nil
Molybdenum	0.016	0.003
Cadmium	Nil	5.98
Lead	0.017	0.012

Dried sample powder was used for analysis

CONCLUSION

The present investigation found that the selected mangroves are rich in bioactive phytoconstituents and nutrients, hence both have good antioxidant and antibacterial activity. The identified bioactive compounds may cure many disorders and it also will be a boon of pharma industry for development of lead drugs to treat microbes.

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