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GC-MS ANALYSIS OF ACANTHOPHORA SPICIFERA

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ABSTRACT

Acanthophora spicifera is one of the bioactive compound rich seaweed, which exhibit potent antitumor and antibacterial activity against human cancer cell lines and bacteria. In the present study, the methanolic extract of *A. spicifera* has been subjected to GC-MS analysis. Twenty four chemical constituents have been identified. The major constituents are octanol, piperazine, benzoic acid and octadecenoic acid. These compounds are invariably having pesticidal, antimicrobial and anti-inflammatory properties.

KEYWORDS: **Cancer, antimicrobial, anti-inflammatory, *Acanthophora spicifera***

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INTRODUCTION

Marine environment occupies about two third of the earth's surface and the future of the world population depends mainly on this environment for its food, industrial raw materials and other live saving drugs. Seaweeds are marine macro algae and primitive type of plants, growing abundantly in the shallow waters of sea estuaries and back water. They flourish wherever rocky foral or suitable substrate are available for their attachment. The seaweed flora of India is highly diversified and comprises mostly of tropical species. The rock beaches, mud flats, estuaries, coral reefs and lagoons along the Indian coast provide an ideal habitat for growth of seaweeds. A total of 25 species of green seaweeds 90 species of brown and 350 species of red seaweeds are found in the world sea area that are commercially important because of their protein, amino acids and mineral contents [1]. These marine algae have evolved unique and highly specialized biochemical pathways to adapt to their sea water medium and survival pressures which gave rise to unparallel variety of biochemical composition in marine algae. For human these nutritional biochemical constituents have been used for centuries. Many species of algae have been used in the industry for extractions of phycocolloids (algin, carrageenan and agar). Certain edible seaweeds contain significant quantities of protein, lipids, minerals, vitamins [2] and 20-50% minerals in their dry weight [3]. Marine algae serve as important resources of bioactive natural products [4]. The production of antimicrobial activities was considered to be an indicator of the capacity of the seaweeds to synthesize bioactive secondary metabolites [5]. There are numerous reports of compounds derived from macro algae with a broad range of biological activities, such as antibacterial, antifungal [6], antiviral [7], antitumoral [8] anticoagulant [9]. The vast varieties of seaweeds were found to possess useful untapped biochemical compounds, which might be a potential source of drug leads in the future. *Acanthophora spicifera* (Vahl) Borgessen is more known as drift red algae that locally so called "bulung tumbung bideng" in Malaysian tropical seawater. It is an erect edible plant possess numerous spines along the branches except for main branches. It occurs in wide range of habitats, as an epiphyte on other algae, on hard bottom or normally as drift algae due to its tolerance to high motions of wave [10]. The colour can be shades of red, orange, dark brown depending on the water level or wave motion. *A. spicifera* is utilized by human as raw foods for diets, raw salads and as flavouring and thickening ingredients in cooking [11]. Several previous studies have revealed the bioactivity of active compounds isolated from the *Acanthophora sp.* such as antibacterial [12], antioxidant, anti-viral, anti-implantation [13] and anti-fouling activity. Steroids and fatty acids ester of *A. spicifera* were reported to exhibit potent antitumor and antibacterial activity against human cancer cell lines and bacteria [14]. Hence it is intended to identify the phytochemical constituents with the aid of GC-MS technique.

GC–MS ANALYSIS

Preparation of extract [15]

Acanthophora spicifera was shade dried and powdered. 20 g of the powdered seaweed was soaked in 95% ethanol for 12 h. The extracts were then filtered through Whatman filter paper No.41 along with 2 gm sodium sulfate to remove the sediments and traces of water in the filtrate. Before filtering, the filter paper along with sodium sulphate was wetted with 95% ethanol. The filtrate was then concentrated by bubbling nitrogen gas into the solution. The extract contained both polar and non-polar phytocomponents. 2 µl of the solution was employed for GC-MS analysis.

Analysis

GC-MS analysis was performed using GC clarus 500 Perkin Elmer system comprising a

AOC-20i autosampler and gas chromatograph interfaced to a mass spectrometer instrument employing the following conditions: column elite-1 fused silica capillary column (30 × 0.25 mm ID ×1EM df, composed of 100% dimethyl polysiloxane), operating in electron impact mode at 70 eV;helium (99.999%) was used as carrier gas at a constant flow of 1ml/min and an injection volume of 0.5 EI was employed (split ratio of10:1) injector temperature 250°C; ion-source temperature 280°C. The oven temperature was programmed from 110°C (isothermal for 2 min), with an increase of 10°C/min, to 200°C, then 5°C/min to280°C, ending with a 9 min isothermal at 280°C. Mass spectra were taken at 70 eV; a scan interval of 0.5s and fragments from 40 to 550 Da.

IDENTIFICATION OF COMPONENTS

From the spectrum obtained by GC-MS analysis the components were identified to possible extent by using the database of National Institute Standard and Technology (NIST) having more than 62,000 patterns. The spectrum of the unknown component was compared with the spectrum of the known components stored in the NIST library. The name, molecular weight and the structure of the components of the test materials were ascertained.

RESULTS AND DISCUSSION

A high resolution mass spectrum equipped with a data system in combination with Gas chromatography was used for the analysis of bioactive components present in them ethanolic extract of *A. spicifera.* Based on spectral data it was found that the extract contained a mixture of volatile compounds. A total of 24 peaks were observed with retention times as presented in Fig.1. and Table-1. The active constituents were 1-hexanol 2- ethyl, 1 – octanol 3,7 – diemethyl, piperazine, benzoic acid, hexadecane, hexadecanoic acid and octadecenoic acid (Table-2). These compounds are invariably having pesticidal, anthelmentic, antifungal, antimicrobial, antioxidant, insect repellent, nematicidal and cancer preventive properties,which could be further isolated, purified and confirmed to be utilized in medical and agricultural industries.

Table4

Biocomponents identified in the methanolic extract of Acanthophora spicifera by GC-MS

S.No.	RT	Name of the compound	Molecular Formula	MW	Peak Area %
1	2.24	Propane ,1-(1,1 – dimethylethoxy)-2, 2-dimethyl-	C9H20O	144	1.64
2	2.32	D-Fructose, diethyl mercaptal, pentaacetate	C20H32O10S2	496	3.02
3	2.49	Octane, 3,5- dimethyl-	C10H22	142	13.35
4	264	1-Hexanol, 2- ethyl-	C8H18O	130	4.79
5	3.05	1 – Octanol, 3,7 – diemethyl-	C10H22O	158	5.29
6	3.12	Piperazine, 1 – nitroso	C4H9N3O	115	3.40
7	3.21	Octadecane, 3 – ethyl – 5 – (2 – ethylbutyl)-	C26H54	366	1.51
8	3.98	Benzoic Acid	C7H6O2	122	10.71
9	4.18	9,12,15 – Octadecatrienoic acid, 2-phenyl-1, 3-dioxan-5-yl ester	C28H40O4	440	2.64
10	4.5	1-Propanol, 3-(octadecyloxy)-	C21H44O2	328	2.14
11	4.68	Benzaldehyde, 3,4 – dimethyl-	C9H10O	134	2.64
12	4.85	Ethanol, 2- (octadecyloxy)-	C20H42O2	314	1.01
13	5.53	1-Dodecanol, 3,7,11 – trimethyl	C15H32O	228	2.52
14	5.64	17 – Pentatriacontene	C35H70	490	3.40
15	5.75	Hexadecane, 1,1-bis (dodecyloxy)-	C40H82O2	594	3.15

16	8.98	Triazene, 1,3-bis(4-methoxyfurazan-3-yl)-3-propyl-	C9H13N7O4	283	1.26
17	10.36	Nonadecane	C19H40	268	1.89
18	11.08	Eicosanoic acid	C20H40O2	312	3.15
19	11.35	Ethyl iso – allocholate	C26H44O5	436	1.01
20	13.42	n-Hexadecanoic acid	C16H32O2	256	8.44
21	21.42	Di-n-octyl phthalate	C24H38O4	390	6.68
22	25.07	9-Octadecenoic acid, (2-phenyl-1, 3-dioxolan – 4-yl) methyl ester, cis-	C28H44O4	444	4.66
23	25.48	4,8,12- Tetradecatrien – 1-ol, 5,9,13-trimethyl-	C17H30O	250	7.81
24	29.82	Cholesta – 8, 24 – dien- 3 – ol, 4-methyl-, (3a,4a)-	C28H46O	398	3.80

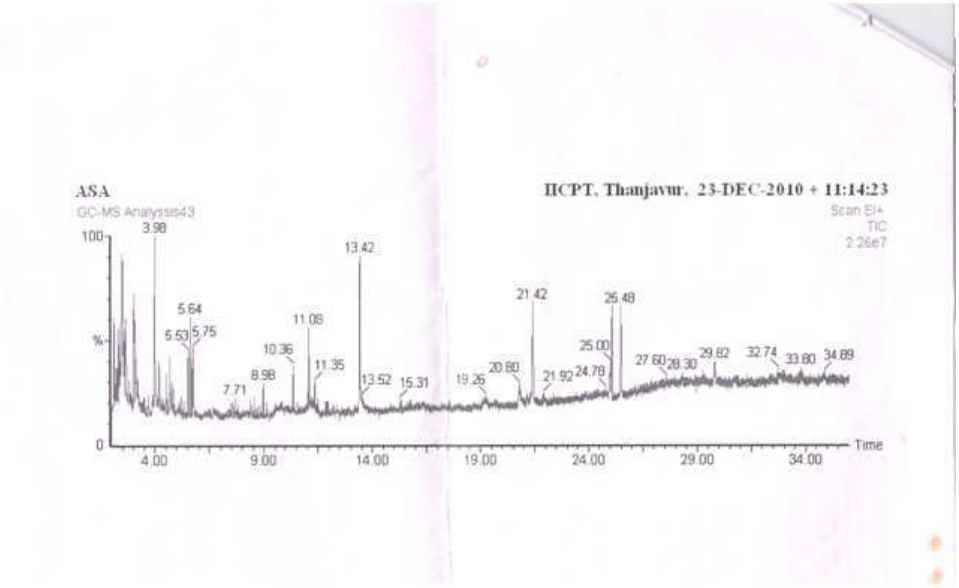
Table5

Bioactivecomponents identified in A. spicifera extract by GC-MS.

RT	Name of the compound	Compound nature	Activity
2.64	1-Hexanol, 2-ethyl	Fatty alcohol	Flavor and fragrance agent
3.05	1-Octanol, 3,7 dimethyl	Esters in essential oil	neurological tremors
3.12	Piperazine, 1-nitroso		anthelmintic, antifungal pesticide
3.98	Benzoic acid	Benzoic acid	Antimicrobial, insect repellent
5.75	Hexadecane, 1,1 bis (dodecyloxy)	Palmitic acid ester	Antioxidant hyocholesterolemic pesticide
11.35	Ethyl iso – allocholate	Steroid	
13.42	n-Hexadecanoic acid	Palmitic acid	Antioxidant, hypocholesterolemi, nematicides, pesticide
25.07	9 – Octadecenoic acid (2 – phenyl -1, 3-dixitan – 4 – ye)methyl ester	Linolenic acid	Anti inflammotry hypocholesterolmic, cancer preventine, nematicide, insectifuge, antizemic

Figure 1

.***Chromatogram of A.spicifera by GC-MS.***



REFERENCE

1. Santhanam, R.N., Remananthan and G. Jagathusan. 1990. *Coastal Aquaculture in India*. C.B.S. Publishers and Distributors. pp. 159-162.
2. Norziah M.H. and Y. Ching 2002. Nutritional composition of edible seaweeds *Gracilaria changgi*. *Food Chemistry*. 68: 69-76
3. Kazutosi N 2002. Seaweeds kaiso: bountiful harvest from the seas: sustenance for health & well being by preventing common life-style related diseases .
4. Metzger P, Roger M. N. and Largean C 2002. Botryolins A ans B, two tetramethyl sequalene triethers from the green microalga *Botryoccus braunic*. *Phytochemistry*. 59: 839-843.
5. González del Val A, G. Platas, A. Basilio, A. Cabello, J. Gorrochategui, I. Suay, F. Vicente E. Portillo, Jiménez del Rio M, Garcia RG, Peláez F 2001. Screening of antimicrobial activities in red, green and brown macroalgae from Gran Canaria (Canary Islands, Spain). *Int. Microbial*. 4: 35-40.
6. Lima-Filho J.V.M, Carvalho A.F.F.U, Freitas S.M, Melo V.M.M (2002). Antibacterial activity of extracts of six macroalgae from the northeastern Brasilian coast. Braz. *J. Microbiol.* 33: 311-314.
7. Richards J.T, Kern E.R, Glasgow L.A, Overall J.C, Deign E.F, Hatch M.T 1978. Antiviral activity of extracts from Marine algae. *Antimicrob. Agents Chemother*.14(1): 24
8. Espeche M. E, E. R.Fraile, A. M. S. Mayer 1984. Screening of Argentine marine algae from antimicrobial activity. *Hydrobiologia* 116/117: 525- 528.
9. Athukorala Y, Ki-Wan Lee, K. Se-Kwon, J. You-Jin 2006. Anticoagulant activity of marine green and brown algae collected from Jeju Island in Korea. *Bioresour. Technol.* 98(9): 1711-1716.
10. Kilar J.A. and J. McLachlan 1986. Ecological Studies of the Alga, *Acanthophora spicifera* (Vahl) Borg. (Ceramiales: Rhodophyta): Vegetative Fragmentation. *J. Experimental Marine Biology and Ecol*.104: 1-21.
11. Phang S.M., 2006. Seaweed resources in Malaysia: (Current status and future prospects Aquatic Ecosystem Health and Manage. 9: 185-202.
12. Ganesan P, C.S. Kumar and N. Bhaskar 2008. Antioxidant properties of methanol extracts and its solvent fractions obtained from India red seaweeds. *Bioresource Technol.* 99: 2717- 2723.
13. Duarte M.E.R, J.P. Cauduro, G.D. Noseda, M.D.Noseda, A.G. Goncalves, C.A. Pujol, E.B. Damonte and A.S. Cerezo 2004. The agaran sulfate from *Acanthophora spicifera* (Rhodomelaceae, Ceramiales) and its antiviral activity. Relation between structure and antiviral activity in agarans. *Carbohydrate Res*. 339: 335-347.
14. Han L, D. Shi, F. Xu, Z. Yuan, J. Sun and J. Shi 2009. Bioactive sterols from red alga *Acanthophora spicifera* boergesen. Zhongguo Zhongyaozazhi Zhongguozhongyaozazhi. *China J. Chinese MateriaMedica*.34: 60-63.
15. Merlin, N. J., V. Parthasarathy, R. Manavalan and S. Kumaravel 2009. Chemical investigation of aerial parts of *Gmelina asiatica* Linn.by GC-MS. *Pharmacog. res.*1(3): 152-156.