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Evaluation of Antifungi activity and Bioactive Compounds of *Sargassum sp.* extract

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Abstract:
The aim of this study was to evaluate the efficacy of compounds and the antifungal vitality of *Sargassumsp*. against pathological fungi which are *Candidaalbicans* , *Candida krusei* , *Aspergillusniger* . as the effective compounds were extracted from this algae solvents were used (hot water and ethanol alcohol) with the qualitative detection of secondary metabolites from the aqueous extract, which indicated the presence of chemical compounds such as flavonoids, phenols, glycosides, terpenes, carotenoids and saponins. The chemical compounds were determined using gas chromatography - mass spectrometry (Gc-mass) of the ethanolic extract of seaweed. These compounds are represented , n-Hexadecanoic acid , Oleic acid , Fucostero , Cholesta-5,20,24-trien-3-ol, (3.beta.)- , Tetradecanoic acid , Palmitoleic acid and Pentane,2-(1-methylethyl)thio] . The aqueous extract of the seaweed did not show any biological activity against all the fungal species under study. While it was noted that the ethanolic extract was more effective in its effect on pathogenic fungal species. As the rates of inhibition zones of the extract (22 , 21.2 , 20.76) against *C.albicans*, *A.niger*, and *C.krusei*, respectively.
Keywords: Bioactive compounds , Gc-mass , Antifungal activity , Sargassum sp..

Introduction:

Fungi are eukaryotic organisms, as there are approximately (50,000) types of fungi in nature, as about (80) types of molds and yeasts have the ability to cause various diseases for humans and animals alike, and diseases caused by fungi are known as (Mycosis) The infection is usually chronic because the fungus grows slowly and includes superficial, systemic and opportunistic infections that usually affect people who are immunocompromised (1) . The genus Candida is one of the natural flora of the body, as it is found naturally in the oral cavity, in addition to its presence in the vagina and respiratory tract (2). However, when an imbalance occurs in the microbial balance due to a change in conditions, the candida grows in excess of its normal limit, which causes human diseases known as Candiaiasis, and this term is applied to a fungal infection of any kind of the genus *Candida* (3)

Aspergillus fungus is one of the pathogens of opportunistic fungal diseases as a result of its spread in the air, soil and various surfaces, as humans inhale the spores of this genus continuously, and the pathogenicity of the fungus depends on the amount of spores entering the body, as the infections range from mild to causing acute and chronic pulmonary diseases, in addition to The immune status of a person,

and the chances of infection increase in people who suffer from weak immunity and chronic diseases, and who take immunosuppressive drugs, and *A. flavus* , and *A. nigar* are among the most pathogenic types of this genus (4) . One of the main reasons for using algae extracts as antimicrobials is their natural origin, as they have minimal harmful side effects on humans and animals and have less environmental risks compared to synthetic alternatives (5) . The increase in microbial resistance to common antibiotics that occurred as a result of excessive and indiscriminate use of them forced researchers to study new resources for antibiotics from different sources, as marine organisms, including algae, are a rich source of bioactive compounds, as studies showed the isolation of more than (15000) natural products Navy in the time period (1965-2005) (6) .

Materials and Methods:

-Samples Collection

The algae sample was obtained in the form of a powder from the American Amazon company. The solvents (ethanol at a concentration of 80% and hot water 100%) were used ,Diagnosed pathogenic fungal isolates were obtained from Al-Ameen Center for Research and Advanced Biotechnology

-prepevation of brown alga extracts

The hot extraction was carried out using a Soxhlet extractor at a temperature of (70) °C for the ethanolic extract. And a temperature of (100) C ° for the aqueous extract for a period of (24) hours.

-Gas chromatograph mass spectrometry

Identification of some chemical compounds in ethanolic extracts of *Sargassum sp*. By (Gc-mass) of the Basra Oil Company / Department of Laboratory and Quality Control / Nahran Omar site.

-Antifungi activity assay

A. niger, C. albicans, and *C. krusei* were used in the experiment. Culture media (potatoes and saproids) were used. As the concentrations of extracts were prepared (100, 200, 300) mg / ml. The antifungal test was carried out using the diffusion method by digging according to the method (7). Inhibition zones in all directions of the pits were calculated by ruler in millimeters. Experiments were repeated three times and the mean zones of inhibition were taken.

-Statistical analysis

The data were analyzed statistically according to the randomized complete block design (CRBD) with three replications using (Tow-Way-ANOVA) and using the statistical program (Spssver 23) and the Least significant differences (L.S.D) test was used to compare the averages under the probability level $P \leq 0.05$

Results and discussion

The qualitative detection results recorded in Table [1] showed that the aqueous extract of *Sargassumsp*. On secondary metabolites, flavonoids, phenols, saponins, terpenoids, carotenoids, and glycosides .

Table [1]: Phytochemicals analysis of *Sargassumsp*. extract

Phytochemicals	Result
alkaloids	-
Flavonoids	+
Glycosides	+
Phenols	+
Terpenoids	+
Carotenoids	+
Saponins	+
Tannins	-

+ = Present ; - = Obsent

This result agreed with(8). and conducted on the brown moss *Sargassumcrassiflium* . The results presented in Table [2] revealed the identified compounds through gas chromatography - mass spectrometry of the ethanolic extract of *Sargassum sp*. The presence of a number of active chemical compounds .. The compounds occupied the largest area among the diagnosed compounds are, n-Hexadecanoic acid that took up space (125061416) It is a carboxylic acid and has antifungal activities, as indicated by a study (9). It is possible to attribute the counter activity to it as it occupies the largest part of the total area .secondly the compound was Oleic acid, which took up an area (44086904), which is an unsaturated fatty acid and has antioxidant and antifungal activity, according to the study of (10). As for the compound Fucosterol , which occupied an area of (31488409) , it is one of the steroid compounds and has antifungal activities, as indicated by (11).

Table [2]: Phytochemicals identified by the Gc-mass of *Sargasssum sp*. algae extract Biological activity

Compound name	Molecular formula	M W g/mol	R T	Area
Tetradecanoic acid	$C_{14}H_{28}O_2$	228.37	17.996	22833127
Palmitoleic acid	$C_{16}H_{30}O_2$	254.41	19.765	10055719
n-Hexadecanoic acid	$C_{16}H_{32}0_2$	256.42	20.106	125061416

Pentane,2-(1-methylethyl)thio]	$C_8H_{18}S$	146.30	21.322	11893569
Oleic acid	$C_{18}H_{34}O_2$	282.468	21.619	44086904
Fucosterol	$C_{29}H_{48}O$	412.7	30.072	31488409
Cholesta-5,20,24-trien-3-ol, (3.beta.)-	$C_{27}H_{42}O$	382.6	31.424	25187468

The results showed the hot aqueous extract of this alga did not show any activity against the studied fungi in the collection of the used concentrations. This result is consistent with the findings of (12) in his study on the brown alga *Sargassumdenticulatum*. It is possible that the ineffectiveness of the aqueous extract against fungi is due to the fact that all the identified substances are insoluble in water (13). The results recorded in Table [3] showed the difference in the effectiveness of the ethanolic extract of *Sargassum sp*. against the types of fungi used in the study. As the concentrations were used (100, 200, 300) mg / ml . There is a correlation between the used concentrations algae and the average inhibition diameters of the studied fungi. The higher the concentration, increased the inhibition zone.

Table [3]: Antifungal activity of *Sargassumsp*. against the types of fungi used in the study

Fungal species	**concentration mg/mL**			**Mean**
	100	**200**	**300**	
C.albicans	19.0±1.0	22.0 ± 1.0	25.0 ± 1.0	22
C.krusei	19.0±1.0	21.0 ± 1.0	22.3 ± 1.0	20.76
A.niger	17.3 ± 0.57	22.0 ± 1.0	24.3 ± 0.57	21.2
Mean	18.4	21.66	23.86	

L.S.D = 2.3 ($P \leq 0.05$)

These results are consistent with the findings of (14) in his study on the brown alga *Sargassumpolycystum* against pathogenic species of the genus *Candida* spp.. It also agreed with the study of (15) which was conducted on the brown alga *Sargassum sp*. against *A. .niger* And with the study of (12) . There are many factors that affect the results reached by the researcher in testing the activity of algae extracts against fungal growth. As there is a difference in the results reached by researchers in studying the same type of algae. This is due to the difference in regions, harvest time and preservation of the samples used in the test. In addition to the stage of algae growth, the method of extraction and the type of solvent used in the extraction.

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