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Research Article
Antifungal Activity of Crude Seaweed Extracts Collected from Latakia Coast, Syria

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Abstract

Background and Objective: Seaweed extracts displayed a broad spectrum as antimicrobial agent due to their richness in bioactive compounds. Seven seaweeds [*Codium tomentosum* (Chlorophyta), *Corallina mediterranea*, *Hypnea musciformis* and *Laurencia papillosa* (Rhodophyta) and *Padina pavonica*, *Sargassum vulgare* and *Dictyota dichotoma* (Phaeophyta)] crude extracts effectiveness had been evaluated against two fungal (*Aspergillus niger* and *Candida albicans*) strains using aqueous, methanol, ethanol, chloroform, acetone, ethyl acetate and hexane solvents. **Materials and Methods:** Seaweeds antifungal effect had been evaluated based on the zone of inhibition (ZI), Minimum Inhibitory Concentration (MIC) and Minimum Fungicidal Concentration (MFC). Amphotericin B (40 mg mL^{-1}) was used as standard for antifungal activity. **Results:** Aqueous extracts showed no activity against the two tested fungi regardless examined seaweeds species. Whereas, the other six organic extracts adversely affected fungal strains. Overall, *D. dichotoma* showed no activity against the two tested fungi regardless examined solvents. Moreover, methanolic *S. vulgare* extract was the strongest by showing the lowest MIC value of 0.11 and 0.13 mg mL^{-1} against *A. niger* and *C. albicans* fungal strains, respectively and the lowest MFC value of 1.67 mg mL^{-1} for the both fungal strains. **Conclusion:** Seaweeds seem to be promising natural resources with low cost for antifungal treatment. Further researches on methanolic *S. vulgare* extracts fractions is requested.

Key words: Antifungal activity, seaweeds, minimum inhibitory concentration, minimum fungicidal concentration

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Aspergillus and *Candida* fungal species are important medically and commercially. However, some of them cause serious disease in humans (Allergic bronchopulmonary aspergillosis, Acute and Disseminated invasive aspergillosis, Aspergilloma, Candidiasis or thrush), grain crops and animals. It worth noting that fungal resistant to commercial antibiotics is considered as an emerging public health threat. As for bacteria, few fungal pathogens became resistant to antibiotics, thereby; antifungal resistance design to treat them is requested. It has been demonstrated that some types of *Candida* fungi are increasingly resistant to first-line and second-line antifungal medications, like fluconazole and echinocandins (anidulafungin, caspofungin and micafungin). *Aspergillus* species as *Candida* fungi resistant also to antibiotic and is also an emerging issue. This problem is still unknown, in particularly for the former species. In this respect, over all *Aspergillus* resistance to azole is recorded[1] to be 3-6%. As for *Candida*, *Aspergillus* infection is related to highly mortality. Otherwise, resistant infection could be developed in people previously exposed to certain antifungal treatment[2].

Medications resistance in fungal strains like e.g. in *Aspergillus* spp. and *Candida* spp. has been infrequently reported. So, treatment failure draws the attention of scientists to look for another treatment to cure them. In this respect, plants (terrestrial and aquatic), seaweeds and lichens extracts as natural resources could be employed as an alternative or as a supplement to antibiotics treatment. Where, all the latter living organisms in particularly seaweeds have bioactive compounds namely secondary metabolites (terpenoids, tannins and phenolic compounds... etc.)[3-4] proved to be active and could be used as antibacterial , antifungal, antioxidants and anticancer. Indeed, their abundance worldwide, easily to get it with low cost make them a potent candidates to be used as a medications.

Seaweed extracts among them proved their efficacy worldwide with board spectrum as antibacterial agent e.g., in Syria[4,5], China[6], Yucatan peninsula[7], Malaysia[8], Bangladesh[9], Belgium[10] and antifungal e.g., in India[3], Syria[4], China[6], Yucatan peninsula[7], Malaysia[8], Bangladesh[9], Belgium[10], UK[11], Egypt[12], Brazil[13], Lebanon[14], Korea[15] and Libya[16].

Little is known about selected seaweeds efficacy against the two fungal strains. Thereby, the current investigation will highlight the seaweeds antifungal activity and the most potent extract will be handled in the future in performance investigation.

MATERIALS AND METHODS

Seaweeds collection: Seven [*Codium tomentosum* (Chlorophyta), *Corallina mediterranea*, *Hypnea musciformis* and *Laurencia papillosa* (Rhodophyta) and *Padina pavonica*, *Sargassum vulgare* and *Dictyota dichotoma* (Phaeophyta)] seaweeds species were collected along the Syrian Coast at 4 km North-Latakia, Syria (38°29'766"E Longitude and 34°37'734"N Latitude) and prepared for extraction as reported by Saleh *et al.*[5].

Seaweed extracts preparation: The seven seaweeds were extracted using aqueous and six solvents (methanol, ethanol, chloroform, acetone, ethyl acetate and hexane). All solvents were purchased from Sigma-Aldrich-Germany. Extraction process had been done as described by Saleh and Al-Mariri[4].

Fungal strains: *Aspergillus niger* and *Candida albicans* fungal strains were provided by the Microbiology and Immunology Division, Department of Molecular Biology and Biotechnology of Atomic Energy Commission of Syria (AECS) in Damascus, Syria. Fungal inoculations were carried out by incubation on Potato Dextrose Agar (PDA) at 28±3°C for 2 days as described by Saleh and Al-Mariri[4].

Antifungal activity test

Disc-diffusion test: Seaweeds antifungal activity has been carried out based on disc-diffusion method described by Bauer *et al.*[17] and Saleh and Al-Mariri[4]. Amphotericin B (40 mg mL^{-1}) (Sigma-Aldrich, St. Louis, USA) was used as standard for antifungal activity. For each extract, duplicate trials were conducted against each strain.

Minimum Inhibitory Concentration (MIC) and Minimum Fungicidal Concentration (MFC): The Minimum Inhibitory Concentration (MIC) and Minimum Fungicidal Concentration (MFC) were estimated using the microplate method as reported by Bauer *et al.*[17] and Saleh and Al-Mariri[4]. Amphotericin B (40 mg mL^{-1}) (Sigma-Aldrich, St. Louis, USA) was used as standard for antifungal activity.

The current study was carried out between April and August, 2016.

Statistical analysis: Statistical analysis was carried out using Statview 4.5 statistical package[18] at the 5% significance level ($p = 0.05$). Data were subjected to Analysis of Variance (Two-way ANOVA) for the determination of differences in means between different tested solvents against selected

isolates for each algae species. Differences between means were examined for significance by Fisher's Least Significant Difference (PLSD) test. Data were expressed as Mean±Standard Deviation (SD).

RESULTS

Antifungal activity of seven seaweeds had been evaluated against two fungal strains based on ZI (Table 1), MIC (Table 2) and MFC (Table 3) values.

Zone of Inhibition (ZI): This parameter ranged from 3 mm with hexane *L. papilosa* extracts against *A. niger* strain to 17 mm with acetonic *P. pavonica* extracts against *C. albicans* strain (Table 1).

Minimum Inhibitory Concentration (MIC): The MIC value varied according to the examined seaweed and solvent (Table 2). In this regards, it ranged from 0.11 mg mL^{-1} with methanol *S. vulgare* against *A. niger* and hexane against *C. albicans*, to 53.3 mg mL^{-1} with ethyl acetate *H. musciformis* against *C. albicans* (Table 2).

Minimum Fungicidal Concentration (MFC): This value ranged from 1.67 mg mL^{-1} with methanol *S. vulgare* against the both fungal strains to 20 mg mL^{-1} with *H. musciformis* (all solvents except methanol one) and also with *L. papillosa* (chloroform and acetone against *A. niger* and also with ethyl acetate and hexane against the both fungal strains) (Table 3).

Table 1: Seaweed extracts antifungal activity using disc-diffusion method expressed as Zone of Inhibition

	Micro organisms	Zone of inhibition (ZI) (mm)						
Seaweed species	Fungi	Methanol	Ethanol	Chloroform	Acetone	Ethyl acetate	Hexane	Amphotericin B
C. tomentosum	*C. albicans*	14±0.22^{Da}	12±0.19^{Ec}	12±0.3^{Cc}	13±0.25^{Db}	11±0.07^{Dd}	7±0.13^{Fe}	16.7±0.16
	A. niger	12±0.4^{Fb}	10±0.28^{Fd}	12±0.34^{Cb}	13±0.29^{Da}	11±0.29^{Dc}	7±0.11^{Fe}	14.5±0.34
C. mediterranea	*C. albicans*	15±0.45^{Ca}	13±0.35^{Dc}	12±0.27^{Cd}	14±0.39^{Cb}	14±0.37^{Bb}	11±0.4^{Ce}	16.7±0.16
	A. niger	14±0.37^{Db}	14±0.29^{Cb}	12±0.45^{Cd}	15±0.49^{Ba}	13±0.39^{Cc}	12±0.35^{Bd}	14.5±0.34
H. musciformis	*C. albicans*	15±0.55^{Ca}	13±0.22^{Db}	12±0.29^{Cc}	11±0.4^{Ed}	10±0.45^{Ee}	10±0.55^{De}	16.7±0.16
	A. niger	15±0.52^{Ca}	14±0.29^{Cb}	11±0.35^{Dd}	10±0.44^{Fe}	11±0.33^{Dd}	12±0.37^{Bc}	14.5±0.34
L. papillosa	*C. albicans*	13±0.22^{Ea}	12±0.38^{Eb}	10±0.4^{Ed}	11±0.23^{Ec}	8±0.37^{Fe}	7±0.31^{Ff}	16.7±0.16
	A. niger	12±0.18^{Fa}	12±0.44^{Ea}	10±0.38^{Eb}	9±0.53^{Gc}	4±0.2^{Hd}	3±0.15^{Ge}	14.5±0.34
P. pavonica	*C. albicans*	17±0.19^{Ab}	15±0.22^{Bd}	13±0.17^{Be}	18±0.3^{Aa}	16±0.29^{Ac}	13±0.2^{Ae}	16.7±0.16
	A. niger	16±0.22^{Bb}	17±0.15^{Aa}	15±0.24^{Ac}	13±0.09^{De}	14±0.26^{Bd}	13±0.17^{Ae}	14.5±0.34
S. vulgare	*C. albicans*	11±0.36^{Gb}	9±0.18^{Gc}	12±0.2^{Ca}	9±0.28^{Gd}	7±0.15^{Gf}	8±0.4^{Ee}	16.7±0.16
	A. niger	10±0.33^{Hc}	7±0.38^{Hf}	11±0.54^{Db}	9±0.44^{Gd}	8±0.25^{Fe}	12±0.19^{Ba}	14.5±0.34
D. dichotoma	*C. albicans*	0±0.0	0±0.0	0±0.0	0±0.0	0±0.0	0±0.0	16.7±0.16
	A. niger	0±0.0	0±0.0	0±0.0	0±0.0	0±0.0	0±0.0	14.5±0.34

Same capital letter (column) and lowercase letter (row) are not significantly different at p = 0.05 probability by Fisher's PLSD test. LSD 0.05 solvent 0.863, solvent and isolate 0.581

Table 2: Minimum inhibitory concentration (MIC) of the seven seaweed extracts against the examined microorganisms

	Microorganisms	Minimum inhibitory concentrations(mg mL^{-1})						
Seaweed species	Fungi	Methanol	Ethanol	Chloroform	Acetone	Ethyl acetate	Hexane	Amphotericin B
C. tomentosum	*C. albicans*	3.3±1.4^{Ab}	5.8±3.8^{Ad}	6.7±2.9^{Ad}	3.3±1.4^{Ad}	3.3±1.4^{Ad}	>10.0±0.0^{Ac}	2.0±0.2
	A. niger	4.2±1.4^{Ab}	5.8±3.8^{Ad}	6.7±2.9^{Ad}	4.2±1.4^{Ad}	3.8±2.2^{Ad}	>10.0±0.0^{Ac}	2.0±0.17
C. mediterranea	*C. albicans*	1.7±0.7^{Bc}	2.1±0.7^{Be}	3.3±1.4^{Ae}	1.7±0.7^{Be}	2.1±0.7^{Bd}	10.0±0.0^{Ac}	2.0±0.2
	A. niger	1.7±0.7^{Bc}	2.9±1.9^{Be}	3.3±1.4^{Ae}	2.1±0.7^{Be}	1.7±0.7^{Bd}	10.0±0.0^{Ac}	2.0±0.17
H. musciformis	*C. albicans*	16.7±5.8^{Ca}	23.3±15.3^{Ca}	26.7±11.5^{Bb}	33.3±11.5^{Ba}	53.3±23.1^{Aa}	10.0±0.0^{Dc}	2.0±0.2
	A. niger	13.3±5.8^{Ca}	26.7±11.5^{Ba}	33.3±11.5^{Ba}	33.3±11.5^{Ba}	43.3±35.1^{Ab}	43.3±35.1^{Aa}	2.0±0.17
L. papillosa	*C. albicans*	13.3±5.8^{Aa}	13.3±5.8^{Ac}	20.0±0.0^{Ac}	20.0±0.0^{Ab}	>20.0±0.0^{Ac}	10.0±0.0^{Bc}	2.0±0.2
	A. niger	13.3±5.8^{Aa}	20.0±0.0^{Ab}	20.0±0.0^{Ac}	20.0±0.0^{Ab}	>20.0±0.0^{Ac}	>20.0±0.0^{Ab}	2.0±0.17
P. pavonica	*C. albicans*	5.8±3.8^{Ab}	7.5±4.3^{Ad}	8.3±2.9^{Ad}	8.3±2.9^{Ac}	4.2±1.4^{Ad}	>10.0±0.0^{Ac}	2.0±0.2
	A. niger	2.9±1.9^{Ab}	3.8±2.2^{Ae}	3.3±1.4^{Ae}	4.2±1.4^{Ad}	4.2±1.4^{Ad}	7.5±3.5^{Ac}	2.0±0.17
S. vulgare	*C. albicans*	0.13±0.05^{Ac}	0.21±0.09^{Ae}	0.19±0.12^{Ae}	0.13±0.05^{Ae}	0.32±0.00^{Ae}	0.11±0.05^{Ad}	2.0±0.2
	A. niger	0.11±0.05^{Ac}	0.21±0.09^{Ae}	0.13±0.05^{Ae}	0.13±0.05^{Ae}	0.27±0.09^{Ae}	0.13±0.05^{Ad}	2.0±0.17
D. dichotoma	*C. albicans*	ND	ND	ND	ND	ND	ND	2.0±0.2
	A. niger	ND	ND	ND	ND	ND	ND	2.0±0.17

Same capital letter (column) and lowercase letter (row) are not significantly different at P= 0.05 probability by Fisher's PLSD test, LSD 0.05 solvent 6.742, solvent and isolate 6.742, ND: No activity

Table 3: Minimum fungicidal concentration (MFC) of the seven seaweed extracts against the examined microorganisms

	Microorganisms	Minimum fungicidal concentration (MFC) (mg mL^{-1})						
Seaweed species	Fungi	Methanol	Ethanol	Chloroform	Acetone	Ethyl acetate	Hexane	Amphotericin B
C. tomentosum	*C. albicans*	8.3±2.9^{Ac}	10.0±0.0^{Ab}	10.0±0.0^{Ab}	10.0±0.0^{Ab}	10.0±0.0^{Ab}	>10.0±0.0^{Ab}	4.0±0.32
	A. niger	8.3±2.9^{Ac}	10.0±0.0^{Ab}	10.0±0.0^{Ab}	10.0±0.0^{Ab}	10.0±0.0^{Ab}	>10.0±0.0^{Ab}	4.0±0.19
C. mediterranea	*C. albicans*	3.3±1.4^{Be}	4.2±1.4^{Be}	6.7±2.9^{Bc}	4.2±1.4^{Bc}	4.2±1.4^{Bd}	>10.0±0.0^{Ab}	4.0±0.32
	A. niger	3.3±1.4^{Be}	4.2±1.4^{Be}	5.0±0.0^{Bd}	4.2±1.4^{Bc}	4.2±1.4^{Bd}	>10.0±0.0^{Ab}	4.0±0.19
H. musciformis	*C. albicans*	20.0±0.0^{Aa}	>20.0±0.0^{Aa}	>20.0±0.0^{Aa}	>20.0±0.0^{Aa}	>20.0±0.0^{Aa}	>20.0±0.0^{Aa}	4.0±0.32
	A. niger	20.0±0.0^{Aa}	>20.0±0.0^{Aa}	>20.0±0.0^{Aa}	>20.0±0.0^{Aa}	>20.0±0.0^{Aa}	>20.0±0.0^{Aa}	4.0±0.19
L. papillosa	*C. albicans*	20.0±0.0^{Aa}	20.0±0.0^{Aa}	20.0±0.0^{Aa}	20.0±0.0^{Aa}	>20.0±0.0^{Aa}	>20.0±0.0^{Aa}	4.0±0.32
	A. niger	16.7±5.8^{Bb}	20.0±0.0^{Aa}	>20.0±0.0^{Aa}	>20.0±0.0^{Aa}	>20.0±0.0^{Aa}	>20.0±0.0^{Aa}	4.0±0.19
P. pavonica	*C. albicans*	8.3±2.9^{Ac}	7.5±3.5^{Ad}	10.0±0.0^{Ab}	10.0±0.0^{Ab}	10.0±0.0^{Ab}	>10.0±0.0^{Ab}	4.0±0.32
	A. niger	6.7±2.9^{Bd}	8.3±2.9^{Ac}	10.0±0.0^{Ab}	10.0±0.0^{Ab}	>10.0±0.0^{Ab}	>10.0±0.0^{Ab}	4.0±0.19
S. vulgare	*C. albicans*	1.67±0.6^{Cf}	2.8±0.5^{Bf}	3.33±0.6^{Be}	2.08±0.1^{Cd}	5.0±0.0^{Bc}	10.0±0.0^{Ab}	4.0±0.32
	A. niger	1.67±0.6^{Cf}	2.8±0.5^{Bf}	3.33±0.6^{Be}	2.08±0.1^{Cd}	5.0±0.0^{Bc}	10.0±0.0^{Ab}	4.0±0.19
D. dichotoma	*C. albicans*	ND	ND	ND	ND	ND	ND	4.0±0.32
	A. niger	ND	ND	ND	ND	ND	ND	4.0±0.19

Same capital letter (column) and lowercase letter (row) are not significantly different at p = 0.05 probability by Fisher's PLSD test, LSD 0.05 solvent 2.912, solvent and isolate 0.543. ND: No activity

DISCUSSION

In the current investigation, ZI, MIC and MFC values served to screen the seaweeds fungicidal effects as reported in many investigations. Data presented herein showed that aqueous extracts have not any activity against the two tested fungi regardless examined seaweeds species. Whereas, the other six organic extracts adversely affected fungal strains. It was noticed that *D. dichotoma* extracts were not active against the both fungal strains regardless tested solvent.

Susceptibility of the two tested fungal strains to the algal extracts expressed as Zone of Inhibition (ZI) was recorded as following: little susceptibility when ZI between 7-10 mm, middle susceptibility between 10.1-15 mm and high susceptibility between 15.1-31 mm. From data presented in Table 1, variance analysis showed that seaweed, solvent and interaction seaweed with solvent effect's on ZI (Table 1) values was significantly different ($p \leq 0.001$). This parameter varied according to the examined seaweed species and solvent. In this regards, little effect (ZI between 7-10 mm) has been observed with hexane *C. tomentosum*, hexane and ethyl acetate *L. papilosa*, ethyl acetate and acetone *S. vulgare* extracts against the two fungal strains (Table 1). Whereas, the remaining extracts exhibited in majority activity ranged between middle to high effectiveness.

Wefky and Ghobrial[12] reported antifungal activity of 5 seaweeds against *A. flavus* and *A. niger* fungal strains using methanol, ethanol and acetone solvents. The previous investigation showed that acetonic *L. pinnatifida* and *S. hystrix* extracts displayed the highest antifungal effect with ZI of 16 and 26 mm, respectively. Whereas, Kim *et al.*[15] reported antifungal activity of *Eisenia bicyclis* edible brown seaweed extract against 8 *Candida* strains. The previous investigation showed that ZI ranged between 20-24 mm with methanol extracts.

Khallil *et al.*[16] reported antifungal activity of 5 brown seaweeds (*S. vulgare*, *Cystoseira barbata*, *Dictyopteris membranacea*, *D. dichotoma* and *Colpomenia sinuosa*) against 8 fungal strains using acetone, ethanol, chloroform, cyclohexane and ethyl acetate solvents. The previous investigation showed that cyclohexanic *S. vulgare* extracts exhibited the highest inhibitory activity against *F. oxysporum.* Otherwise, ZI against *A. niger* was recorded to be 7 and 12 mm with *S. vulgare* ethanol and cyclohexane extracts. Whereas, no activity recorded with chloroform, acetone and ethyl acetate extracts against the same strain.

As for MIC (Table 2) and MFC (Table 3), variance analysis showed that seaweed, solvent and interaction seaweed with solvent effect's on MIC (Table 2) and MFC (Table 3) values were significantly different ($p \leq 0.001$).

Overall, methanolic *S. vulgare* extract was the strongest by showing the lowest MIC/MFC value of 0.11/1.67 and 0.13/1.67 mg mL^{-1} against *A. niger* and *C. albicans* fungal strains, respectively. Followed by *C. mediterranea*, in this respect, the lowest MIC/MFC value was recorded to be 1.7/ 3.3 mg mL^{-1} against the both tested fungal strains with methanolic extract.

Previously, Tariq[11] reported antifungal activity of 4 red seaweeds against *Aspergillus* sp. and *C. albicans*. The previous investigation showed that *A. flavus*, *A. fumigatus* or *C. albicans* were resistant to all the previous seaweeds extracts. Moreover, Ballesteros *et al.*[19] reported biological

activity of 71 Mediterranean macrophytes. The previous investigation showed that *Codium* spp. and *Corallina elongata* displayed a strong antifungal effect with a good antifungal effect recorded with *Corallina granifera*.

Whereas, Zheng *et al.*[6] reported antifungal effect of 23 species of marine seaweeds (Chlorophyta, Phaeophyta and Rhodophyta). The previous investigation showed that Rhodophyta were the most potent extracts. Morales *et al.*[7] reported antifungal effect of 6 seaweeds using ethyl acetate partition of methanol extracts. The previous investigation showed that *Trichophyton mentagrophytes* fungus growth was inhibited by *L. obtusa*, *S. filipendula* and *S. hystrix* extracts at 6.25-3.13 mg mL^{-1}. Moreover, Wefky and Ghobrial[12] reported antifungal activity of 5 seaweeds against *A. flavus* and *A. niger* fungal strains. The previous investigation showed that acetonic *S. hystrix* extract displayed the highest antifungal effect.

Nor Afifah *et al.*[8] reported antifungal activity of *Halimeda discoidea* green seaweed extract against 6 fungal strains. The previous investigation showed that all the 6 tested fungal strains were resistant to the examined extracts. Whereas, Rodloff *et al.*[20] reported that not all tested *Candida* species similarly responded to antifungal.

Kim *et al.*[15] reported antifungal activity of ethyl acetate edible brown seaweed *Eisenia bicyclis* extract against *Candida* species. The previous investigation showed that MIC/MFC value ranged between 4-32/16-64 mg mL^{-1} against *C. albicans.* Overall, ethyl acetate-soluble extract was the most potent with MIC/MFC value ranged between 4-8/16 mg mL^{-1} against the tested strain.

Khaled *et al.*[14] reported methanolic *P. pavonica* and *S. vulgare* antifungal effect against *Candida* species strains. The previous investigation showed that *S. vulgare* extract has no activity against tested fungal strains. Whereas, ethyl acetate *P. pavonica* fraction extract had antifungal effect against *C. krusei* and more noticeable against *C. glabrata* strain. Indeed, Guedes *et al.*[13] reported antifungal activity of different seaweeds species against *Candida* species. The previous investigation showed that MIC ranged between 0.008-0.016 mg mL^{-1}. Moreover, dichloromethane, methanol and ethanol extracts showed the strongest inhibition growth. Moreover, Chowdhury *et al.*[9] reported antifungal effect of 10 freshwater and marine algae against *C. albicans* strain. The previous investigation showed that *Chlorella* spout the examined algae had the highest antifungal activity. Recently, Raj *et al.*[3] reported hexane, chloroform, ethyl acetate, acetone and methanol extracts of *Ulva* sp. green algae against 10 fungal isolates. The previous investigation showed that ethyl acetate *U. lactuca* extracts were the most potent by showing the lowest MIC of 0.250 mg mL^{-1} against *C. parapsilosis*.

More recently, Saleh and Al-Mariri[4] reported the inhibitory effect of *U. lactuca* (Chlorophyta), *Dilophus spiralis* (Phaeophyta) and *Jania rubens* (Rhodophyta) marine seaweeds against 2 fungal (*C. albicans* and *A. niger*) strains using aqueous and six organic extracts. The previous investigation showed that the lowest MIC value was recorded to be 0.106 mg mL^{-1} with *U. lactuca* methanolic extract against the both fungal strains and with acetone and hexane against *C. albicans*. Moreover, the lowest MFC (0.266 mg mL^{-1}) was observed with *D. spiralis* Chloroform against both fungal strains. Moreover, Saleh *et al.*[5] reported antibacterial effect of *C. tomentosum*, *C. mediterranea*, *H. musciformis* and *S. vulgare extracts* against 10 Gram-negative bacterial isolates using aqueous and six organic solvents. The previous investigation showed that the methanolic *S. vulgare* extract had the strongest antibacterial effect by showing the lowest MIC/MBC values of 0.08/1.00 mg mL^{-1} against *E. coli* O:157 isolate. Whereas, Peres *et al.*[21] reported antifungal activity of ten seaweeds against *Aspergillus flavus* fungal strains. The previous study revealed that *A. flavus* growth did not significantly inhibited by the seaweed extracts.

As for lichens extracts, as natural resources have been proved their effectiveness as antifungal agent. In this respect, Rukayadi *et al.*[22] reported antifungal activity of methanolic *Usnea* sp. lichen extract against *Malassezia furfur* fungi. More recently, Plaza *et al.*[23] reported also antifungal effect of *Cladonia* aff. *rappii* A. Evans (Cladoniaceae) lichen against 4 genus of *Candida* and 1 genus of *Cryptococcus* using aqueous, ethanol and dichloromethane extract. The previous investigation showed that the lowest MIC value was recorded to be 2.2 and 5.9 mg mL^{-1} against *C. glabrata* with ethanol and dichloromethane extracts, respectively. Whereas, the lowest MFC value was recorded to be recorded 7.4 mg mL^{-1} against *C. neoformans* followed by 8.9 mg mL^{-1} against *C. glabrata* with dichloromethane extracts.

The strongest antifungal activity recorded in *S. vulgare* could be attributed to the abundance of phenolic components making them more active against examined strains compared to the other seaweed phyla in particularly, Chlorophyta. Indeed, this phenomenon could be attributed to the presence of other bioactive components that were not measured in the present investigation. Phenolic compounds could be used as antimicrobial agents in wild mushrooms[24] and *Ficus sycomorus*[25] against some micro-organisms resistant to antibiotics.

CONCLUSION

Antifungal effectiveness of seven seaweeds [1 (Chlorophyta), 3 (Rhodophyta) and 3 (Phaeophyta)] crude extracts has been mainly investigated against two fungal (*A. niger* and *C. albicans*) strains using aqueous, methanol, ethanol, chloroform, acetone, ethyl acetate and hexane solvents. Seaweeds antifungal activity has been screened based upon MIC/MFC value. All over, methanolic *S. vulgare* extract was the most effective by showing the lowest MIC/MFC value of 0.11/1.67 mg mL^{-1} and 0.13/1.67 mg mL^{-1} against *A. niger* and *C. albicans* fungal strains, respectively. Thereby, methanolic *S. vulgare* extracts fractions needs performance research for future.

SIGNIFICANCE STATEMENT

The current study highlighted the importance of some seaweed extracts using different solvents, as antifungal agent. Methanolic *S. vulgare* crude extracts as antifungal agent should be given more attention. Moreover, their fractions with an in-depth study regarding their fractions potency separately need more investigation. Seaweeds seem to be promising natural resources with low cost for antifungal treatment in pharmacology and medicine applications.

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