Antimicrobial Potential of Maldivian Marine Macroalgae Against Clinically Relevant Bacterial Pathogens

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Abstract
The problem of antimicrobial resistance (AMR) is one of the major health problems globally predicted to claim lives of millions each year by 2050 unless new agents will be discovered. The marine environment such as Maldives archipelago with nearly 99% water coverage contains a variety of benthic macroalgae which still have not been explored to their full potential as a source of antimicrobials. This research deals with the assessment of antibacterial properties of two most common in Maldives benthic algae – Padina pavonica (brown algae) and Caulerpa racemosa (green algae). Fresh algae samples obtained from the intertidal areas of Hulhumale were shade-dried and extracted with 95% ethanol and distilled water under cold maceration technique to obtain crude extracts. After the experiment was performed and results were analyzed, it became clear that the ethanol extracts have shown greater extraction efficiencies and better antibacterial effects than aqueous extracts, which were not able to show any zones of inhibition. The strongest zone of inhibition was achieved with the ethanolic extract of P. pavonica on E. coli bacteria, while C. racemosa showed good suppression effect. While there was no visible inhibition effect against Klebsiella sp. and Enterococcus sp., the results have proved that the bioactivity of Maldivian algae depends on the solvent and strain and are very important to perform MIC/MBC assays.

Keywords: *Antimicrobial Resistance; Padina pavonica; Caulerpa racemosa; Ethanol Extracts; Maldivian Algae.*

Introduction

Antimicrobial resistance (AMR) constitutes a serious worldwide health issue that is expected to lead to at least 1.91 million AMR-attributable fatalities per year by 2050 if no measures are undertaken [1, 2]. In view of the fact that classical antibiotic pipelines have become unproductive, marine bioprospection is becoming increasingly popular [3]. It is worth noting that the marine environment constitutes a source of promising bioactive natural products, namely phlorotannins, terpenoids, alkaloids, and sulfated polysaccharides [4, 1]. Almost 99% covered by the ocean, Maldives constitutes an archipelago rich in benthic algae which have the potential to yield novel antimicrobials [3, 10]. Benthic macroalgae thrive in stressful intertidal conditions and produce bioactive secondary metabolites to resist herbivores and microbes [8]. In this paper, Padina pavonica (brown alga) and Caulerpa racemosa (green alga, sea grapes) were screened for antibacterial effects [5, 7]. Brown and green algae differ in their chemical composition, which enables them to damage bacterial membranes, produce reactive oxygen species and prevent biofilm formation [6, 4]. To enhance the efficiency of extraction of bioactive compounds, a double solvent system, comprising polar (aqueous) and hydroalcoholic/lipophilic (ethanol) solvent was used [8, 10].

Related work

Research carried out worldwide on the species of *Padina* and *Caulerpa* reveals that they have antimicrobial potential against Gram-positive and Gram-negative pathogens [6, 8]. Phlorotannins and polyphenolics from P. pavonica inhibit the growth of pathogens by disrupting their cell walls and biofilm formation in *S. aureus, P. aeruginosa, and E. coli* [7, 5]. In a similar manner, bis-indole alkaloids (e.g. caulerpin) and sulfated polysaccharides produced by Caulerpa species interfere with the colonization process and exert a dose-dependent bactericidal effect [9, 6]. Nonetheless, the previous literature on these species is limited to non-Maldivian chemotypes [10].

Table 1 compares this study with existing literature.

Reference	Geographic Origin	Solvents Tested	Bioactivity Against E. coli	Evaluation of Maldivian Flora
Nour et al. (2025)	Mediterranean / Global	Water / Eco-solvents	Yes	No
Al-Katib et al. (2025)	Red Sea	Methanol / Ethanol	Yes	No
Thacharoen et al. (2025)	Pacific Ocean	Organic Solvents	Moderate	No
This Work	Hulhumale, Maldives	Ethanol & Water	Yes (Ethanol extract)	Yes

Key Contribution

The present study offers the first empirical study on the antibacterial activity of the benthic marine algae Padina pavonica and Caulerpa racemosa, which were collected directly from the intertidal areas of the Maldives (Hulhumale reef zone) against the bacterial pathogens Escherichia coli, Klebsiella sp., and Enterococcus sp. [3, 5, 10].

Methodology, Experiments, and Results

Padina pavonica and Caulerpa racemosa were collected from Hulhumale intertidal reef flat, thoroughly washed with distilled water to remove epiphytes, sands, and salts, dried to constant weight under shade-drying, and powdered [5]. Cold maceration was performed within 7 days using 95% ethanol and distilled water at room temperature due to heat-sensitive nature of active compounds [8, 10]. The solvent was allowed to evaporate naturally to obtain crude extract [6]. Extraction yield was calculated and presented in Table 2.

Table 2. Extraction yields of algal samples

Species	Starting Powder (g)	Solvent	Dried Extract (g)	Percentage Yield (%)
Padina pavonica	1.91 g	Ethanol	0.05 g	2.62%
Padina pavonica	1.91 g	Water	0.02 g	1.05%
Caulerpa racemosa	0.81 g	Ethanol	0.07 g	8.64%
Caulerpa racemosa	0.81 g	Water	0.04 g	4.94%

The antimicrobial susceptibility test was carried out through the agar well diffusion test on Nutrient Agar, Blood Agar, and CLED Agar using Escherichia coli, Klebsiella sp., and Enterococcus sp. [10, 9]. Ciprofloxacin and Azithromycin acted as the positive control, and ethanol and distilled water were used as the negative control [8].

It was observed that ethanol extracts were more concentrated and had stronger antibacterial activity compared to aqueous extracts [6]. The ethanol extracts of both P. pavonica and C. racemosa inhibited the growth of E. coli while aqueous extracts did not show any inhibition [7, 9]. P. pavonica ethanol extracts had the highest level of inhibition against E. coli while C. racemosa ethanol extracts showed moderate inhibition [5, 8]. No inhibition was observed against Klebsiella sp. and Enterococcus sp. at all levels of extract concentration [10].

Discussions

The greater yields and antimicrobial activity of the ethanol extracts against E. coli agree with previous researches showing that non-polar and moderately polar bioactives, including phlorotannins, terpenoids, sterols, and lipophilic phenolics are well extracted using organic/hydroalcoholic solvents [4, 6, 8]. Alcohol

is able to break down algae cell membranes better during maceration, thus releasing the membrane-bound bioactive substances [8, 2].

The absence of any inhibition against Klebsiella sp. and Enterococcus sp. under current conditions does not prove the complete inactivity, but reflects strain-specific sensitivities, concentrations requirements or even diffusion restrictions of crude lipophilic substances through the agar gel matrix [10, 21]. Besides, proteins present in Blood Agar can bind to polyphenolic substances during the tests on Enterococcus [7, 2].

Conclusions Motivation/ProblemStatement:

Explored the poorly understood Maldivian benthic macroalgal flora as new possible therapeutic leads for AMR [1, 3].

Approach Employed:

Ethanol/water cold maceration, then agar well diffusion assay on clinical bacterial strains [5, 10].

Major Results:

Antibacterial activity of Maldivian algal species depends on the solvent used and is species-specific. The extractives in P. pavonica and C. racemosa from ethanol inhibited E. coli growth (P. pavonica had the most potent activity), while aqueous extracts had no antibacterial effects [6, 7, 8]. None of them inhibited Klebsiella sp. or Enterococcus sp. at present [10].

Limitations/Future Directions:

Current results require follow-up with MIC/MBC quantitative microdilution, seasonal sampling and phytochemical fractionation/LC-MS [4, 9, 10].

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