ULVA INTESTINALIS ASSOCIATED BACTERIA: MOLECULAR IDENTIFICATION AND ANTIMICROBIAL POTENTIAL.

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Abstract

Seventeen marine bacteria associated with the green alga *Ulva intestinalis* (northern coast in Tunisia), were isolated and tested for antibacterial and antifungal activity. None of the 17 isolates displayed antifungal activity whereas two isolates displayed antibacterial activity with different antibiotic spectra. These two isolates (U1 and U5) belonged to Alpha-proteobacterial genus *Stappia*. Strain, U5, showed the strongest activity against pathogens especially *Staphylococcus aureus* and *Aeromonas salmonicida*. Algae like *U. intestinalis* may benefit from epiphytic bacteria such as *Stappia* as potential resources of natural antibiotic production and concomitant protection against undesirable prokaryotes.

Keywords: Algae, Bacteria, Biodiversity, Antibiotics

Introduction

Bacteria surviving on marine algae live in highly competitive, nutrient limited environment. This competition for both space and nutrient inflict selective pressure on the marine microorganisms that may result in the production of bioactive compounds of possible medical and biotechnological value [1]. In Tunisian coast, the green alga *U. intestinalis* is abundantly present but little is known about their epibionts and the ability of these to produce bioactive compounds. Here we investigate the culturable bacterial fraction, colonizing *U. intestinalis* and focus on their antimicrobial activity against pathogens bacteria and yeast.

Materials and methods

Marine green algae *U. intestinalis* were collected from coastal waters, north of Tunisia in Cap Zebib (37° 16.2' N, 10° 3.6' E) between January and November 2007. Seaweed samples were washed and epiphytic bacteria were isolated [2, 3]. Identification of isolates was realized by PCR amplification of 16S rRNA gene. In vitro antibacterial tests were performed against fifteen sensitive bacteria including fish and human pathogens (*Aeromonas hydrophila , Pseudomonas cepacia, Vibrio anguillarum, Vibrio tapetis* CECT 4600, *Aeromonas salmonicida, Salmonella typhymurium, Streptococcus sp., Staphylococcus aureus, Vibrio alginoliticus, E. coli* O126B16, *E. coli* ATCC 25922, *Pseudomonas fluorescens* AH2, *Pseudomonas aeruginosa* ATCC 27853, *Staphylococcus aureus* ATCC 25923, *Enterococcus feacalis* ATCC 29212) and *Candida albicans* ATCC.

For the bioassay, drops of 10µl of the algae-associated bacteria suspension were spotted on the agar plates swabbed with the test strains and allowed to dry at room temperature. After incubation for 24 h, antibacterial activity was evaluated by measuring the inhibition zone (in mm) from the edge of the colony to the edge of the clear zone.

Results and discussion

Seventeen morphological distinct isolates were selected from *U. intestinalis* surface and tested for potential antibacterial and antifungal activities. Ten isolates were identified by PCR amplification and 16sRNA gene sequencing. Among them, nine were gram-negative, assigned to *Proteobacteria* (Gamma (one strain) and Alpha subclasses (seven strains)) and *Bacteroidetes* (one strain). Only one strain was gram-positive. *Gamma-Proteobacteria* subclass was represented by the genus *Vibrio*. This isolate appears to be not closely associated to its host since it was found inhibited by *U. intestinalis* extracts (unpublished data). Only two strains U1 and U5 showed antibacterial activities. These isolates were closely related to *Stappia* sp. Strong activity was detected for U5 against gram-positive pathogens:*S. aureus and S. aureus* ATCC 25923 and gram-negative *A. salmonicida* (Table 1). Similar antimicrobial activity of epiphytic seaweed bacteria was reported previously [3]. These authors described *U. intestinalis* as the main source of antibiotic producing bacteria that were identified as *Pseudomonas-Alteromonas.* Here we isolated *antibiotic producing* strains of *Stappia* sp. from *U. Intestinalis*, a promising result in the discovery of novel antibacterial compounds. Further analyses and characterization of the antibacterial compound is in progress.

Table.1. Antibacterial spectra of the *U. intestinalis* active strains U1 and U5.

	Active strains	
Test strains	**U1**	**U5**
***S.aureus* ATCC 25923**	**-**	**13**
S.aureus	**-**	**13**
A.salmonicida	**4**	**12**
Streptococcus.sp	**3**	**-**
P.auruginosa	**4**	**-**
A. hydrophila	**4**	**-**

Zone of inhibition (mm) measured considering the spot diameter, -: No activity.

References

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