Antimicrofouling activity of some common plants found in Chennai (India)

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Abstract

Four different panels such as copper coated steel, aluminium coated steel, mild steel and wood which are widely used to construct ship hulls, boats and other structures were submerged in the sea water and the biofilm formed on these panels were collected after 24 hrs. The organisms were isolated and cultured in the nutrient agar medium. The organisms were identified as Klebsiella sp., Aeromonas sp., Pseudomonas sp. and Bacillus sp. and were confirmed by various biochemical tests. Four different plants viz., Senna auriculata L, Prosopis juliflora (Sw.) DC, Morinda pubescens J.E. Smith and Adhatoda Vasica NEES were selected as they are commonly found in and around Chennai. Their leaf extracts were prepared using ethanol. Antimicrobial activity test was done by well diffusion method. It was found that Senna auriculata L and Prosopis juliflora (Sw.) DC showed maximum inhibitory effect on these organisms. Prosopis juliflora (Sw.) DC showed maximum inhibition zone on the Bacillus sp. and Aeromonas sp. than the other organisms while Senna auriculata L showed maximum inhibition zone on Pseudomonas sp. and Klebsiella sp. However, the extract of Morinda pubescens J.E. Smith and Adhatoda vasica NEES did not show any inhibition on these organisms. Further work is required to examine the precise role of such antifouling activity in nature, and to determine the specific antifouling activity against marine bacteria implicated in biofilms.

Keywords: biofilm, anti-fouling, Senna auriculata L, Prosopis juliflora (Sw.) DC, Morinda pubescens

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Introduction

A biofilm is an aggregate of microorganisms in which cells adhere to each other and/or to a surface. Biofilms may form on living or non-living surfaces, and represent a microbial life in natural, industrial and hospital settings. The microbial cells growing in a biofilm are physiologically distinct from planktonic cells of the same organism. Biofouling is especially economically significant on ships’ hulls where high levels of fouling can reduce the performance of the vessel and increase its fuel requirements. Biofouling is also found in almost all circumstances where water based liquids are in contact with other materials. Industrially important examples include membrane systems, such as membrane bioreactors and reverse osmosis spiral wound membranes cooling water cycles of large industrial equipments and power stations. Biofouling can also occur in oil pipelines carrying oils with entrained water especially those carrying used oils, cutting oils, soluble oil or hydraulic oils. Individually small, accumulated biofoulers can form enormous masses that severely diminish ships’ maneuverability and carrying capacity. Fouling causes huge material and economic costs in maintenance of mariculture, shipping industries, naval vessels, and seawater pipelines. Anti-fouling is the process of removing the accumulation, or preventing its accumulation. In industrial processes bio-dispersants can be used to control biofouling.

Anti-fouling is the process of removing the accumulation, or preventing its accumulation. In order to minimize the impacts of foulers, many underwater structures are protected by antifouling coatings. Many types of coatings, however, have been found to be toxic to marine organisms. Lack of knowledge of taxonomy and ecology of tropical fouling organisms is a major stumbling block to development of effective antifouling management. This has led to the unnecessarily heavy handed use of broad spectrum toxins which damage the environment and kill native wildlife. Hence, using eco-friendly antifouling agents are necessary to protect the ecosystem and yet commercially viable as well. The potential of higher plants as source of antifouling agents is still largely unexplored. Among the estimated 250,000-500,000
plant species, only a small percentage has been investigated phytochemically and the fraction submitted to biological or pharmacological screening is even smaller. Thus, any phytochemical investigation of a given plant will reveal only a very narrow spectrum of its constituents. Plants represent a rich source of antimicrobial agents. Although hundreds of plant species have been tested for antimicrobial properties, the vast majority of have not been adequately evaluated. Considering the vast potentiality of plants as sources for antimicrobial drugs a systematic investigation was undertaken to screen the local flora of Chennai where the selected plants (Senna auriculata L, Prosopis juliflora (Sw.) DC, Morinda pubescens J.E. Smith and Adhatoda Vasica NEES) were found in abundance.

**Materials and methods**

**Collection of Plant material**

Fresh leaves of four different plants viz. Senna auriculata L Prosopis juliflora (Sw.) DC, Morinda pubescens J.E. Smith and Adhatoda Vasica NEES) free from suburban parts of Chennai, Tamilnadu. The leaves were washed thoroughly 2-3 times with running water and once with sterile distilled water and was then air-dried on sterile blotter under shade.

**Solvent extraction**

Thoroughly washed dried leaves of four different plants viz. Senna auriculata L Prosopis juliflora (Sw.) DC, Morinda pubescens J.E. Smith and Adhatoda Vasica NEES) were dried in shade for five days and then powdered with the help of blender. 50 g of dried powder was soaked in 300 ml ethanol for 48 hours with intermittent shaking. The plant extracts were filtered through Whatman No. 1 filter paper into pill vials. The filtrates were dried until a constant dry weight of each extract was obtained. The residues were stored at 4°C for further use. Isolation of microfoulers: Four different panels such as copper coated steel, aluminium coated steel, mild steel and wood which are widely used to construct ship hulls, boats and other structures were used as fouling patterns. Each panel was placed in a tray containing 1litres of seawater sample collected from Marina beach, Chennai. The panels were arrested for 48 hrs to 72 hrs and the biofilm was scrapped and washed by using sterile brushes and distilled water. The biofilm collected was then inoculated on a nutrient agar medium and incubated at 37°C for 24 hrs. The number of colonies was counted for each panel and was found to be different for each panel (Fig. 1).

**Results and discussion**

The antibacterial activity of Senna auriculata, Prosopis juliflora, Morinda pubescens and Adhatoda vasica is shown in table 3. Maximum inhibitory effect was shown by leaf extracts of Senna auriculata and Prosopis juliflora on these organisms. Prosopis juliflora showed maximum inhibition zone on Bacillus sp. and Aeromonas sp. than the other organisms while Senna auriculata showed maximum inhibition zone on Pseudomonas sp. and Klebsiella sp. However, the extract of Morinda pubescens and Adhatoda vasica did not show any inhibition on these organisms. Further phytochemical studies are required to determine the types of compounds responsible for the antifouling effects of these species. Although Adhatoda vasica and Morinda pubescens are well known for their medicinal properties they did not have any effect on these microorganisms by forming biofilm.
From the above results it can be concluded that plant extracts have great potential as antifouling compounds against microfoulers that cause enormous economic loss in shipping and other industries. *Prosopis juliflora*, which is a weed and *Senna auriculata* have shown maximum antifouling activity and further research may help to discover bioactive natural products that may serve as leads for the development of new antifouling agents that are eco-friendly. Such screening of various natural organic compounds and identifying active agents is the need of the hour as our country is blessed with enormous bioresources which are still to be tapped and utilised.

**Table 3.** Inhibitory effect of ethanol extract on biofilm organisms (in mm) (K- *Klebsiella* sp.A- *Aeromonas* sp. P- *Pseudomonas* sp.B- *Bacillus* sp.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Motility</th>
<th>Gram stain</th>
<th>Oxidase</th>
<th>Catalase</th>
<th>Citrate</th>
<th>Indole</th>
<th>MR</th>
<th>VP</th>
<th>Urease</th>
<th>Nirate</th>
<th>TSI</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Klebsiella</em> sp.</td>
<td>Motile</td>
<td>-Rods</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>Acid butt Alkaline slant</td>
</tr>
<tr>
<td><em>Aeromonas</em> sp.</td>
<td>Non Motile</td>
<td>-Rods</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>Acid butt Alkaline slant</td>
</tr>
<tr>
<td><em>Pseudomonas</em> sp.</td>
<td>Non Motile</td>
<td>-Rods</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>Acid butt Acid slant</td>
</tr>
<tr>
<td><em>Bacillus</em> sp.</td>
<td>Non Motile</td>
<td>+Rods</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>Acid butt Acid slant</td>
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**References**


