



An Attempt To Examine Antibacterial Activity Of Different Organic Solvents Extract Of Marine Seaweeds

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ABSTRACT

Marine algae derivatives have showed promise as potential candidates for the development of new antibacterial drugs. There is a need for novel antibacterial medicines since drug-resistant pathogenic germs already kill countless people, necessitating the exploration of readily accessible untapped natural sources like seaweed. Seaweeds (*Gracilariaverrucosa*, *Gracilariafoliifera*, *Gracilariacorticata*, *Gracilariacrassa*, and *Gracilariaedulis*) were examined in this study for their ability to combat human bacterial infections (*K. pneumoniae*, *P. aeruginosa*, *E. aerogenes*, *S. typhi* and *S. aureus*). By using agar cut well diffusion techniques, the bactericidal activity of several extracts (Acetone, Benzene, Butanol, Chloroform, Ethanol, Ethyl Acetate, Hexane, Isoamyl Alcohol, Methanol, and Propanol) was assessed. All of the *Gracilaria* species under study exhibited antibacterial activity.

Keywords: Algae, Pathogenic, Seaweeds, Antibacterial, *Gracilaria*

I. INTRODUCTION

Bacterial plant infections have the potential to wreak havoc across the agricultural industry. Many economies place a great deal of importance on this industry. In order to fulfill the rising global food demand brought on by the expanding population, agricultural yield losses brought on by illnesses must be decreased. Intense sprays of synthetic pesticides are utilized to tackle these agricultural issues, however this present control technique is losing effectiveness as microbes acquire resistance to these poisons. More urgently, chemical pesticides have a negative impact on the ecosystem, including how they affect the fertility of the soil and non-target species. For instance, it has been noted that the application of neonicotinoid pesticide to crops has a detrimental effect on the interannual reproductive capacity of both wild and managed bees, including honeybees, in several regions. Therefore, pesticides have the potential to destroy biodiversity and have long-term negative consequences on human health. As a result, it is widely acknowledged that developing alternate strategies to lessen chemical use is necessary. Natural products are being studied as sources of possible substitutes,

especially marine algae since they may be a new supply of secondary metabolites. It is thought that seaweeds produce these compounds as a defense against microorganisms. The antibacterial activity of these compounds in seaweeds was initially discovered by Harder. Since then, a number of biological activities—including anti-inflammatory, antioxidant, antifouling, and antiviral—have been discovered in numerous seaweed species. Seaweeds have also been utilized for many generations as a source of medicine in many coastal locations and as part of the regular diet in many nations.

Therefore, these possibly new bioactive substances may help shield plants from harmful organisms. These substances also address other issues with synthetic pesticides, such as their biodegradability, potential for reduced environmental effect, and decreased risk of infections developing treatment resistance. Additionally, because they are made from natural ingredients, pesticides often only harm the target pest and closely related species.

II. INHIBITION OF FOODBORNE PATHOGENIC AND SPOILAGE BACTERIA IN FOOD PRODUCTION

Due to changes in consumer eating patterns and the increasing antibiotic resistance of spoilage and pathogenic foodborne bacteria, cases of food poisoning have substantially increased in recent years. Food producers are switching from traditionally used synthetic preservatives and excessive amounts of salt to organically derived alternatives in response to mounting public demand. Without the negative side effects of many synthetic preservatives or excessive salt consumption, these natural antimicrobials originating from terrestrial plants and marine algae offer shelf-life extension and greater safety from germs that cause food poisoning.

Himanthaliaelongata, *Saccharinalatissima*, and *Laminariadigitata* are three edible Irish brown seaweeds with antibacterial properties in both their raw and heat-processed (95°C) forms. Their effectiveness was evaluated against pathogens including *Listeria monocytogenes*, *Salmonella abony*, *Enterococcus faecalis*, and *Pseudomonas aeruginosa*, which frequently create issues in the food sector. Raw *Himanthaliaelongata* extracts in methanol at a concentration of 60 mg/mL inhibited *Listeria monocytogenes* by 98.7% in microtiter tests, compared to sodium nitrite (96.2%) and sodium benzoate (96.5%). Also more effective than the norms against *Enterococcus faecalis* and *Pseudomonas aeruginosa* was raw *Himanthaliaelongata*. *Laminariadigitata* was the next most effective against all four bacteria, followed by raw *Saccharinalatissima* extracts. All seaweeds had much less antibacterial action after being heated. These uncooked, cold-storage goods, such raw meats and fish, may benefit from the insertion of these raw seaweed extracts since they would exert their bacterial inhibition throughout this period before being inactivated by heat, when the product would be eaten.

III. MATERIALS AND METHODS

Sample collection

India was the source of the hand-picked seaweeds. The seaweeds that were gathered were identified in the Indian Centre for Marine Fisheries Research Institute.

Collection of microorganisms

According to Bergey's handbook of determinative bacteriology, the pathogenic bacteria were isolated from clinical samples obtained from diagnostic labs and identified on the basis of morphological, biochemical, and physiological characteristics. It was discovered that the isolated microbes were *K. Aeruginosa*, *E. pneumoniae*, and *P. S. aerogenes* *S. typhi*, and *Aureus*.

Extract Preparation

To get rid of unwanted material, fresh water was used to wash and distilled water was used to rinse the seaweed samples. Samples were then chopped into small pieces, ground in a mixer grinder, and shade dried. Different solvents with increasing polarity, including Hexane, Benzene, Chloroform, Ethyl acetate, Acetone, Butanol, Propanol, Ethanol, Methanol, and Isoamyl alcohol, were utilised since organic solvents are more effective. 5 g of the materials were extracted for the extraction procedure by soaking them overnight at room temperature in 50 ml of various solvents. Following incubation, the extract was filtered before being concentrated according to the solvents' boiling points. The antibacterial activity of the concentrated extracts against microorganisms was examined.

Determination of antibacterial activity

The agar well diffusion technique was used to assess the antibacterial activity. On sterile MH agar plates, the overnight-grown bacterial culture was transferred. A sterile 6 mm cork borer was used to make wells in the previously inoculated MH agar plates. Using sterile micropipette tips, 80 μ L of each *Gracilaria* extract were added to the wells and allowed to diffuse for two hours at 4°C before the plates were incubated for 24 hours at 37°C. By measuring the diameter of the inhibitory zone for each well, the findings were reported.

Determination of minimal inhibitory concentration (MIC)

For MIC determination, the extracts that had a significant amount of activity were used. Four dosage levels of the extracts, ranging from 20 to 80 L, were examined. A sterile 6 mm cork borer was used to make wells in the MH agar plate after the overnight-grown bacterial culture was put there. Different concentrations (2080 L) of the extract were added to different wells, diffused for two hours at 4 C, and then the plates were incubated at 37 C for twenty-four hours. The zone of inhibition was seen, and the MIC was calculated using the test sample's lowest dose that showed the zone of inhibition.

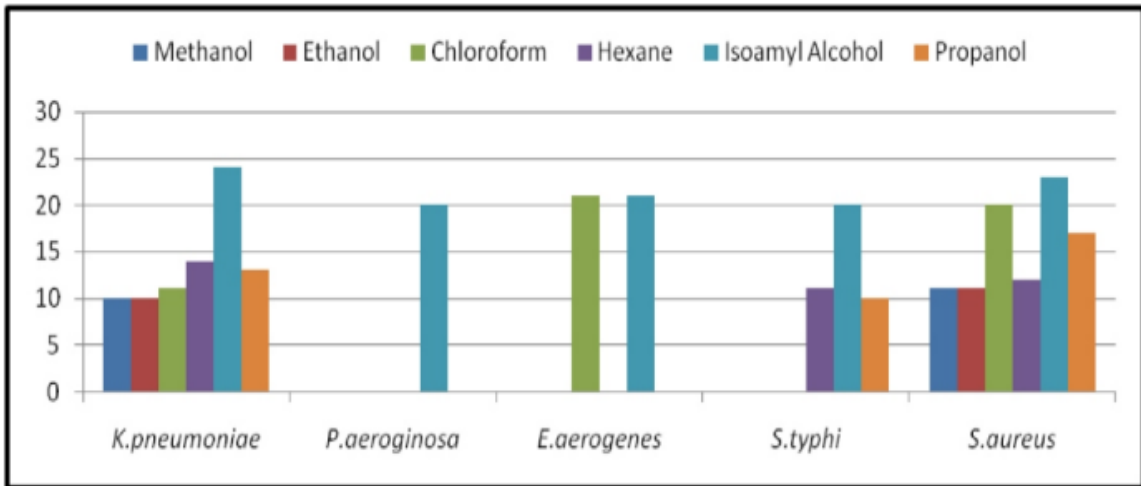


Figure 1: Antibacterial Activity of G. Verrucosa

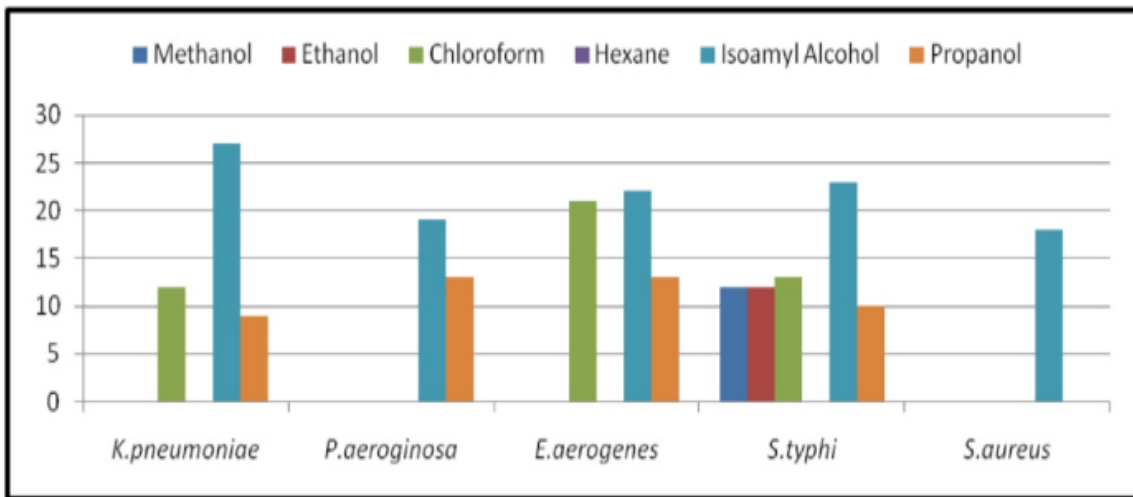


Figure 2: Antibacterial Activity of G. Foliifera

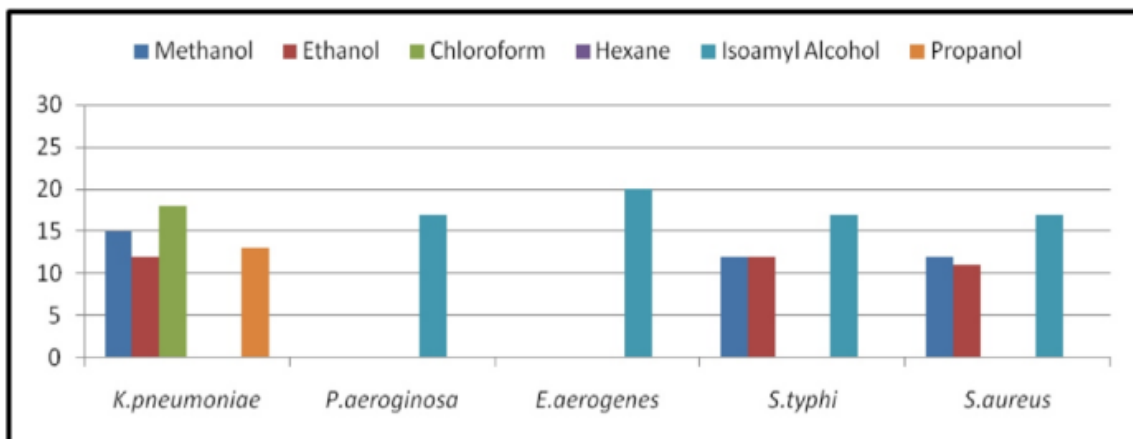


Figure 3: Antibacterial Activity of G. Corticata

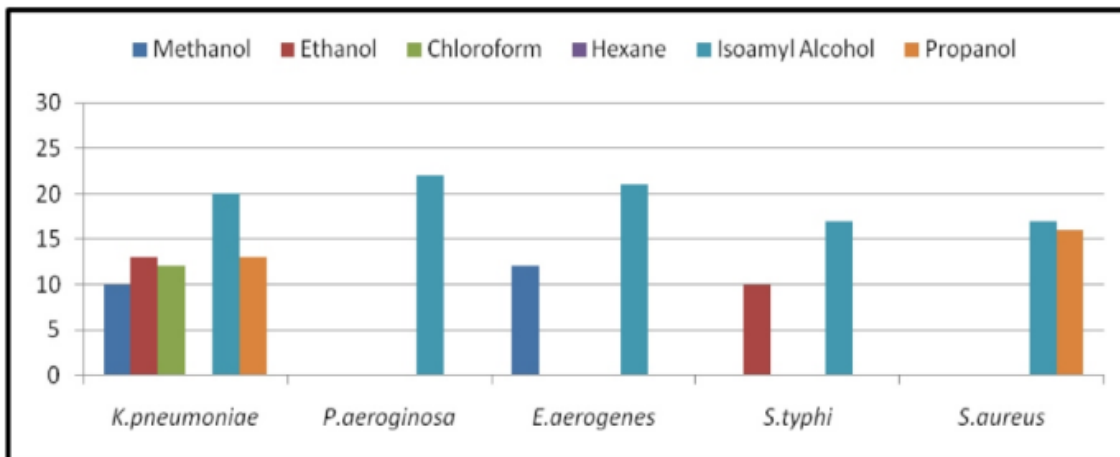


Figure 4: Antibacterial Activity of G. crassa

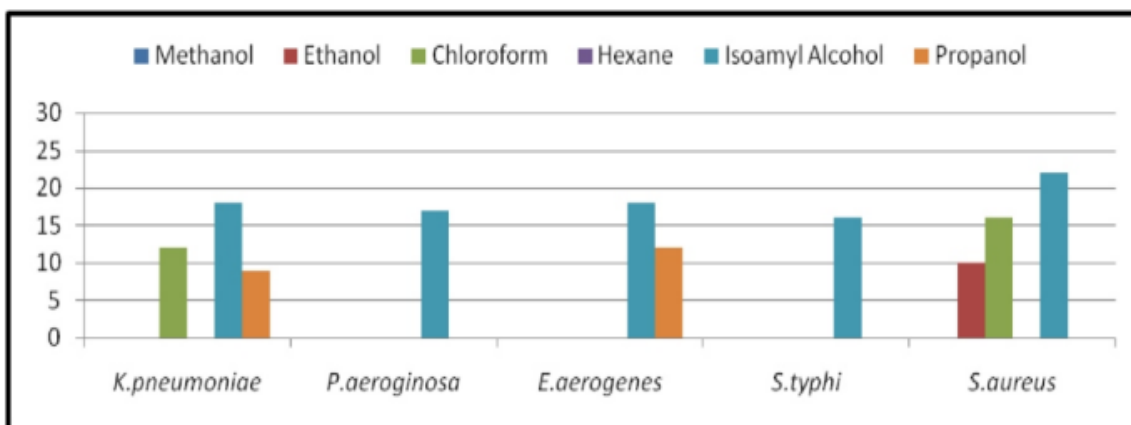


Figure 5: Antibacterial Activity of G. Edulis

IV. RESULTS AND DISCUSSION

Five different gracilaria species were tested for their antibacterial efficacy against both gram-positive and gram-negative bacteria. Ten distinct polar and non-polar organic solvents, including acetone, butanol, ethanol, ethyl acetate, isoamyl alcohol, methanol, and propanol, were tested in the preliminary testing. Only those extracts (Ethanol, Chloroform, Isoamyl alcohol, Methanol, and Propanol) displaying antibacterial activity were taken into account. Figure 1-5 summarizes the zone of inhibition induced by these extracts against pathogenic bacteria. Species of various types were sensitive to varying solvents. Among the solvents tested, the isoamyl extract showed the most activity, while the hexane extract showed the lowest. However, the isoamyl alcohol extract demonstrated the most activity and had the largest zone of inhibition against most of the pathogens tested. The G extract dissolved in isoamyl alcohol generated the largest inhibition zone (27 mm). *K. foliifera* vs. *pneumoniae*. The zone of inhibition against *E. coli* was greatest (21 mm) in chloroform, making it the second-best solvent for antibacterial activity *aerogenes*. Table 1 summarizes the findings of a MIC determination

assay performed on the extracts known to have antibacterial activity. Antibacterial activity was seen at relatively low concentrations in the isoamyl alcohol extract.

Table 1: Minimum Inhibitory Concentration of Gracilaria extracts.

Seaweed and Pathogens	Minimum Inhibitory Concentration Extract					
	Methanol	Ethanol	Chloroform	Hexane	Isoamyl Alcohol	Propanol
<i>G. verrucosa</i>						
<i>K. pneumoniae</i>	60	80	60	60	20	40
<i>P. aeruginosa</i>	-	-	-	-	20	-
<i>E. aerogenes</i>	-	-	20	-	20	-
<i>S. typhi</i>	-	-	-	60	20	60
<i>S. aureus</i>	60	60	20	60	20	40
<i>G. foliifera</i>						
<i>K. pneumoniae</i>	-	-	60	-	20	80
<i>P. aeruginosa</i>	-	-	-	-	20	60
<i>E. aerogenes</i>	-	-	40	-	20	60
<i>S. typhi</i>	40	40	40	-	20	80
<i>S. aureus</i>	-	-	-	-	20	-
<i>G. corticata</i>						
<i>K. pneumoniae</i>	40	60	40	-	-	60
<i>P. aeruginosa</i>	-	-	-	-	20	-
<i>E. aerogenes</i>	-	-	-	-	20	-
<i>S. typhi</i>	80	80	-	-	20	-
<i>S. aureus</i>	80	80	-	-	20	-
<i>G. crassa</i>						
<i>K. pneumoniae</i>	80	80	80	-	20	60
<i>P. aeruginosa</i>	-	-	-	-	20	-
<i>E. aerogenes</i>	60	-	-	-	20	-
<i>S. typhi</i>	-	80	-	-	40	-
<i>S. aureus</i>	-	-	-	-	40	60
<i>G. edulis</i>						
<i>K. pneumoniae</i>	-	-	60	-	40	80
<i>P. aeruginosa</i>	-	-	-	-	20	-
<i>E. aerogenes</i>	-	-	-	-	40	60
<i>S. typhi</i>	-	-	-	-	20	-
<i>S. aureus</i>	-	60	40	-	20	-

V. CONCLUSION

Seaweeds were tested for their antibacterial efficacy against plant diseases in quarantine. It was discovered that the raw seaweed extracts had powerful antibacterial components. Gracilaria extracts showed activity against the majority of the virulent bacteria tested. *G. verrucosa* outperformed them all in terms of inhibitory efficacy against most diseases. *K. pneumoniae* and *S. aureus* strains were more susceptible to all *G. verrucosa* solvent extracts. The potential of Gracilaria species as a source of novel bioactive chemicals with antibacterial activity against pathogenic organisms has been highlighted by these findings. However,

further phytochemical research is required to determine which chemical components in seaweed extracts have the antibacterial activity.

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