

Antipathetic Effect of *Euphorbia Hirta* on *Staphylococcus Aureus*

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ABSTRACT

The use of plant compounds to treat infection is an age old practice in a large part of the world, especially in developing countries, where there is dependence on traditional medicine for a variety of diseases. *Euphorbia sp.* are used in traditional medicines. Hypotensive and tonic properties are also reported in *E. hirta*. The aqueous extract exhibits anxiolytic, analgesic, antipyretic, and anti-inflammatory activities. Methanolic extract of leaves have antifungal and antibacterial activities. In this work different concentration shows as 100, 75, 50, 25% and under controlled gave inhibition zone as 9.33, 6.02, 4.48, 3.66 but controlled patriplaite full of microorganism.

Keyword: *Euphorbia Hirta*, Antimicrobial, *Staphylococcus Aureus*

I. INTRODUCTION

Medicinal plants have played an essential role in the development of human culture. This is the reasons for the usage of specific medicinal plants for treatment of certain diseases were being discovered; thus, the medicinal plants' usage gradually abandoned the empiric framework and became founded on explicatory facts. Plants had been the source of treatment and prophylaxis (Huang, *et.al.*2012).). Since time immemorial people have tried to find medications to alleviate pain and cure different illnesses. In every period, every successive century from the development of humankind and with the advancement of civilizations, the healing properties of certain medicinal plants were identified, noted, and conveyed to the successive generations Tarek A *et. al.*(2017).

The use of plant compounds to treat infection is an age old practice in a large part of the world, especially in developing countries, where there is dependence on traditional medicine for a variety of diseases (Martins Ekor, 2013). According to World Health Organization, medicinal plants would be the

best source of Ayurveda In recent years, antimicrobial properties of medicinal plants are being increasingly reported from different parts of the world (Haidan,2016). Antimicrobial plant extracts have been recognized as a future source of new antimicrobials in the event of the current downturn in the pace at which these are being derived from micro-organisms.(Desalegn Amenu 2014).

Euphorbia hirta is a small annual herb common to tropical countries Mohammad ,(2010). It can grow to a height of 40 cm. The stem is slender and often reddish in colour, covered with yellowish bristly hairs especially in the younger parts. The leaves are oppositely arranged, lanceolate and are usually greenish or reddish underneath measuring about 5 cm long. In the axils appear very small dense round clusters of flowers. The small green flowers constitute the inflorescence characteristic of the *euphorbia's*. The stem and leaves produce white or milky juice when cut Anononymous.(2005). *Euphorbia hirta* is annual weed mostly found on road side. The plant widely used in traditional medicinal to cure various disease especially for gastrointestinal disorder, skin disease etc.(Huang *et al.*, 2012). *E. hirta* is mostly

contain gallic acid, quercetin and a phenolic substance C H O . Several studies conclude that *E. hirta* possess 28 18 15 anti-anaphylactic, antioxidant, anticancer, antifeedant, antiplatelet aggregation and anti-inflammatory, aatoxin inhibition, antifertility, antiplasmodial, antiameobic, larvicidal, and insect repellent activities (Sandeep and Chandrakant, 2011).

Plant material : Morphology of plant material:

E. hirta belongs to the plant family *Euphorbiaceae* and genus *Euphorbia*. It is a slender-stemmed, annual hairy plant with many branches from the base to top, spreading upto 40 cm in height, reddish or purplish in color. Leaves are opposite, elliptic - oblong to oblong-lanceolate, acute or subacute, dark green above, pale beneath, 1- 2.5 cm long, blotched with purple in the middle, and toothed at the edge. The fruits are yellow, three-celled, hairy, keeled capsules, 1-2 mm in diameter, containing three brown, four-sided, angular, wrinkled seeds Rahman *et. al.*2013), Hussain *et. al.* 2014.) The collected plants were dried under shade, crushed and subject to soxhlet extraction with methanol.

Morphology of bacteria

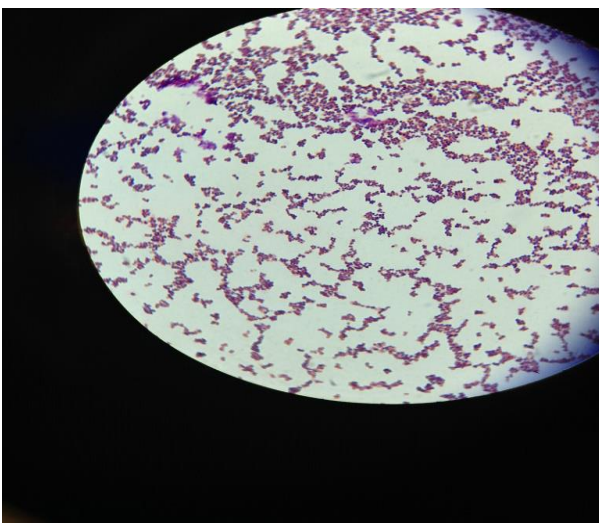


Figure 1. Strain of *S.aureus*.

Staphylococcus was first identified in 1880 in Aberdeen, Scotland, by surgeon Sir Alexander Ogston in pus from a surgical abscess in a knee joint. This name was later amended to *Staphylococcus aureus* by Friedrich Julius Rosenbach, who was credited by the official system of nomenclature at the time (Ogston ,1984). *S. aureus* can cause a range of illnesses, from minor skin infections, such as pimples, impetigo, boils, cellulitis, folliculitis, carbuncles, scalded skin syndrome, and abscesses, to life-threatening diseases such as pneumonia, meningitis, osteomyelitis, endocarditis, toxic shock syndrome, bacteremia, and sepsis. It is still one of the five most common causes of hospital-acquired infections and is often the cause of wound infections following surgery (Singh *et.al.*2018). *S.aureus* is not always pathogenic, it is a common cause of skin infections including abscesses, respiratory infections such as sinusitis, and food poisoning. Pathogenic strains often promote infections by producing virulence factors such as were collected potent protein toxins, and the expression of a cell-surface protein that binds and inactivates antibodies (Abeer2016). The emergence of antibiotic-resistant strains of *S. aureus* such as methicillin-resistant *S. aureus* is a worldwide problem in clinical medicine. Despite much research and development there is no approved vaccine for *S. aureus* (Tracey *et.al.*2018)

II. MICRODILUTION METHOD

PRINCIPLE

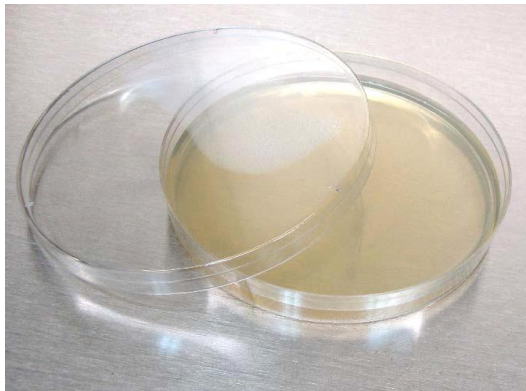
Dilution susceptibility testing methods are used to determine the minimal concentration of antimicrobial needed to inhibit or kill the microorganism. This can be achieved by dilution of antimicrobial in either agar or broth media Vineetha *et. al.*(2015).

PROCEDURE

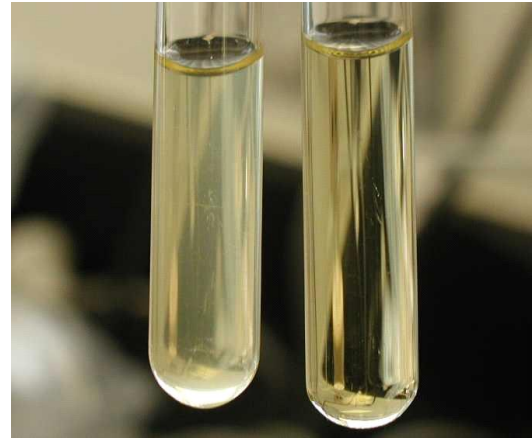
The minimum inhibitory concentration (MIC) was determined by micro dilution method using serially diluted plant extracts. The methanol extract were diluted to get series of concentrations from 25%,50%,75% and 100% respectively in distilled water. The microorganism suspension of 50 μ l was added to the broth dilutions. These were incubated for 18 hours at 37°C. MIC of each extract was taken as the lowest concentration that did not give any visible bacterial growth. E. L. Chuah(2014).



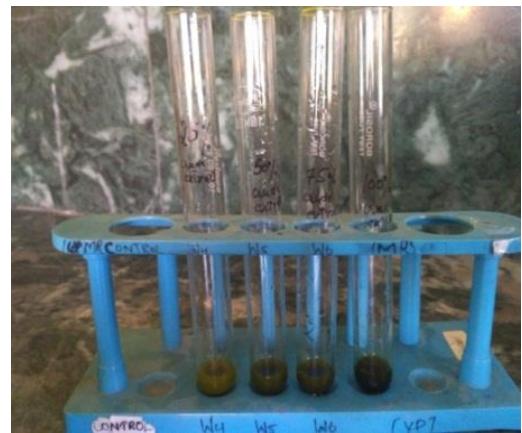
(A)



(B)



(C)



(D)

Figure 2 (a) MHA (250ml), (b) MHA Plate, (c) Nutrient broth & (d) Test tubes containing crude methanolic extract of *E. hirta* with different concentrations.

Evaluation of antibacterial activity

The effect of various plant extracts on the several bacterial strains were assayed by Agar well diffusion method and further confirmed by disc diffusion method. The minimum concentrations of the plant extracts to inhibit the microorganisms were also determined by microdilution method using plant fractions serially diluted in sterile nutrient broth.

Bacterial cultures of that were used for antimicrobial assay of test organisms were obtained from the culture collection, UCBMSH, Dehradun. The bacteria

were maintained on nutrient broth (NB) at 37°C till required for analysis.

AGAR WELL DIFFUSION METHOD PRINCIPLE

The antimicrobials present in the plant extract are allowed to diffuse out into the medium and interact in a plate freshly seeded with the test organisms. The resulting zones of inhibition will be uniformly circular as there will be a confluent lawn of growth. The diameter of zone of inhibition can be measured in millimeters.

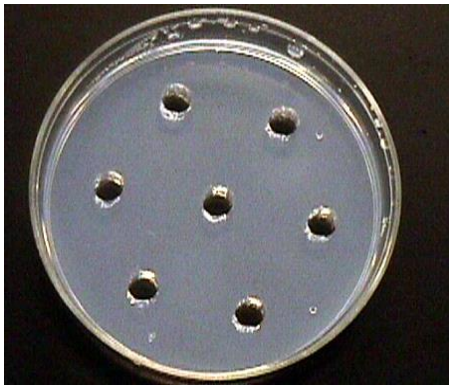


Figure 3. Plate showing wells made with borer

PROCEDURE

Petriplates containing 20ml Muller Hinton medium were seeded with 24hr culture of bacterial strains. A sterile swab stick was used to spread about 0.2 ml of the standardized test inoculum evenly on the surface of the solidified media. Five equidistant wells of 5 mm in diameter were then made on the seeded agar plate using a sterile cork borer and the plant extracts with concentrations ranging from 25%, 50%, 70% and 100% respectively, were introduced into the bored holes. A 5 ml of tween 20 was used in reconstituting the extracts. The plates were then incubated at 37°C for 24 hours. The antibacterial activity was assayed by measuring the diameter of the inhibition zone formed around the well (NCCLS, 1993).



(a)



(b)

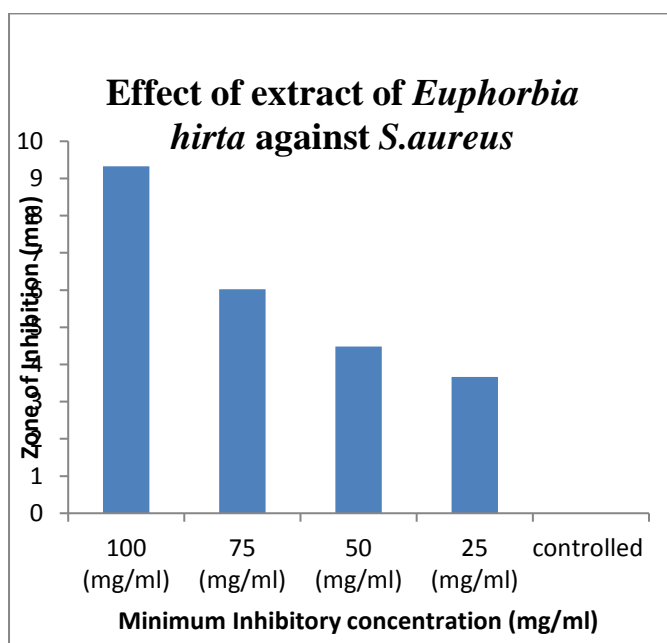
Figure 4. (a) culture plate (b) inhibition zone

III. RESULTS AND DISCUSSION

Results of the antibacterial screening of different concentrations of the extracts on the test isolates are shown in Table 1 and Graph 1. The results show that the increase in concentration of the extract increased the zones of growth inhibition of the bacteria (Fig. 5 b). The assessment of the antibacterial activity was based on the measurement of diameter zone of inhibition (mm) that formed around the hole made by the borer filled with the extract. Maximum inhibition zone was recorded at 100 mg/ml and the minimum inhibition zone at 25 mg/ml in both the bacteria for all the extracts and controlled petriplates showed no zone of Inhibition (Table 1 and 2; Fig 5a and b). This proved that the extract is effective against microbes.

| Concentrations of <i>Euphorbia hirta</i> extract (mg/ml) | Zone of Inhibition (mm) in <i>Staphylococcus aureus</i> |
|--|---|
| 100 | 9.33 |
| 75 | 6.02 |
| 50 | 4.48 |
| 25 | 3.66 |

Table 1. Zone of Inhibition in *S.aureus*



Graph 1. Effect of extract of *Euphorbia hirta* against *S.aureus*

IV. CONCLUSION

In this study, the results indicated that the methanolic extract of the plants inhibited the growth of the test bacteria. This therefore, showed that the extract contained substances that can inhibit the growth of the selected bacteria. Other workers have also shown that extracts of plants inhibit the growth of various microorganisms at different concentrations (Faraja *et al.* 2018).

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