



Research Article

ANTIMICROBIAL ACTIVITY OF AQUEOUS EXTRACT OF *Terminalia chebula* RETZ. ON GRAM-POSITIVE AND GRAM-NEGATIVE MICRO-ORGANISMS

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ABSTRACT

The present study describes the anti-microbial activity of *Terminalia chebula* Retz fruit extract against microorganism. *Bacillus subtilis*, *Staphylococcus aureus*, *Staphylococcus epidermis*, *Escherichia coli*, *Staphylococcus flexineria* and *Pseudomonas aeruginosa*. For this purpose aqueous extract of fruit were prepared and tested by "Disc Diffusion Method". As a result of this study it was found that the extract of fruit generally revealed anti-microbial activity against both gram-positive bacteria (*B. subtilis*, *S. aureus* and *S. epidermis*) and gram-negative bacteria (*E. coli*, *S. flexineria* and *P.aeruginosa*).

Keywords: Anti-microbial activity, *Terminila chebula*, Disc diffusion method.

INTRODUCTION

Herbal medicines are in great demand in the developed as well as in developing countries for primary health care because of their wide biological and medicinal activities, higher safety margin and lower costs. *Terminila chebula* Retz (*combretaceae*) is medicinal plant widely distributed through out India, Burma and Srilanka. The dried ripe fruit *T. chebula* also known as black myrobalan has widely been used in the treatment of asthma, sore throat, vomiting, hiccup, bleeding, piles, diarrhoea, gout, heart and bladder diseases.¹ Black myrobalan has reported to have antioxidant and free radical scavenging activities.² It is active against cancer cells³ and *Helicobacter pylori*.⁴ It is also useful as an anticarcinogenic agent.⁵ It is used in dermal wound healing⁶ and improving gastrointestinal motility.⁷ It is reported to use in anaphylactic shock⁸ and in diabetes mellitus.⁹ One group of researchers found that both aqueous and ethanolic extract of *T. chebula* have strong antimicrobial activity against the uropathogen *Escherichia coli*.¹⁰ Gallic acid and its ethyl ester isolated from ethanolic extract of

T.chebula showed antimicrobial activity against methicillin resistance *Staphylococcus aureus*. It has also growth inhibitory action against *Salmonella typhi* and intestinal bacteria. To the best of our knowledge, there have been no published reports concerning the antimicrobial activity of aqueous extract of *T.chebula* fruits against *B. subtilis*, *S. aureus*, *S.epidermis*, *E.coli*, *S. flexineria* and *P.aeruginosa*. We have, therefore focused on its anti-microbial activity in our study. This was achieved with the disc diffusion method.

MATERIAL AND METHODS

Plant materials

T.chebula fruits were collected locally and were identified by the department of Botany, Bharathidasan University, Thiruchirappalli, India.

Preparation of extract

The collected *T.chebula* fruits were dried to get completely dried product. The dried fruits were then milled to fine powder and extracted with water and ethanol mixture (1:1) for 24 Hrs. in a separating funnel. The aqueous layer of the extract was collected in a beaker. The solvents were removed in

rotary evaporator and the crude extracts were dried at room temperature in a steady air current. The dried extracts were then stored in an air tight jars at 4°C for anti-microbial analysis.

Formulation of extract

For anti-microbial activity study on the day of experimentation, the different amount of powder was suspended in distilled water to get different concentration of suspension.

Microorganism

The microorganisms used in this study were *B.subtilus* (NCIM2493), *S.aureus* (NCIM2079), *S.epidermis* (NCIM2493), *E.coli* (NCIM2065), *S.flexinaria* (NCIM2418), *P.auriginosa* (NCIM2200).

Anti-microbial activity

The anti microbial activity of the aqueous extract was carried by disc diffusion method.¹¹ A suspension of tested microorganisms was spread on Muller-Histon Agar (MHA) (Oxoid) medium. The filter paper discs (5mm in diameter) were individually impregnated with different concentration of extract and then placed into the agar plates which had previously been inoculated with the tested microorganisms. The plates were subsequently incubated at 37° C for 24 Hrs. After incubation the growth inhibition rings were quantified by measuring the diameter of the zone of inhibition in mm. All the tests were performed in triplicate. Tobramycin, cephalixin, erythromycin and ampicillin served as positive control.

RESULT AND DISCUSSION

In-vitro preliminary screening of the antimicrobial activity of the plant extracts from *T.chebula* was studied against some micro-organisms using the filter paper disc diffusion method. The antimicrobial affect of plant extract against the different strains are illustrated in Table 1 and Figures 1.

The extract of *T.chebula* at the concentration of 100% has antimicrobial activity on the tested microorganism form high to low respectively. *P.auriginosa* (19mm), *E.coli* (16mm), *S. epidermis* (15mm), *S. aureus*(15mm), *S. flexinaria* (14mm), and *B.subtilus* (14mm) showed in Table -1.

Table1 shows that the extract of *T. chebula* have activity on *B.subtilus*, *S.aureaus*, *S. epidermis*, *E. coli*, *S. flexinaria* , *P. auriginosa* (14mm, 15mm, 15mm, 16mm, 14mm, and 19mm inhibition zone respectively).

The data indicated that gram negative *P. auriginosa* was the most sensitive strain of those tested with the extract of *T. chebula*, with strongest inhibition zone of 19mm. The extract concentration of 100% also exhibit high antimicrobial activity against *E.coli.*, with modest activity against *S.epidermis*, *S. aureaus*, *B.sustilus*, *S. flexinaria*. The 75% concentration of the extract of *T.chebula* also show strongest inhibition zone against different strains of microorganisms. The data indicates that anti-microbial activity of extract (at 75% concentration) with strongest inhibition zone of 14mm for the strain of *P.auroginosa*, *E.coli*, and *S.epidermis*. However, the inhibition zone for the species of *B.subtilus*, *S. aureaus* and *S.flexinaria* was found 13mm, 12mm and 11mm respectively.

The Table-1 shows that different concentration (50%, 75% and 100%) of plant extract were having good anti-microbial activity against *B.subtileaus*, *S.aureaus*, *S.epedermis*, *E.coli*, *S.flexinaria* and *P.auriginosa*.

The minimum inhibitory concentration (MIC) of the *T. chebula* fruit extract was measured which is depicted in the Table-1. It was observed that *S. aureaus* and *P. auriginosa* have shown MIC value at 1% concentration of plant extract.

Other bacteria have shown very small zone at 1% concentration of the extract.

On comparing the inhibition zone of the extract to that of standard antibiotics (tobramycin, cephalexin, erythromycin and amoxicillin) extract showed better activity than tobramycin and cephalexin. However,

extract is not potent than erythromycin and amoxicillin in these condition.

In the end of study we have found the extract *T.chebula* revealed antimicrobial activities against microorganisms.

Table 1: Anti-microbial activity of *T. chebula* aqueous extract of different microorganisms

Sample concentration in %	microorganisuns (inhibition zone in mm)					
	Gram-positive			Gram-negative		
	<i>B.subtilus</i>	<i>S. aureus</i>	<i>S. epiderms</i>	<i>E.coli</i>	<i>S.flexineria</i>	<i>P.auriginosa</i>
100	14	15	15	16	14	19
75	13	12	14	14	11	14
50	12	11	12	13	10	13
25	11	9	11	12	10	12
20	10	9	10	11	8	11
15	10	5	10	9	5	10
10	6	4	10	7	4	6
5	5	3	5	6	3	5
1	-	1	-	-	-	-

Table 2 : Anti-microbial activity of antibiotics on different micro-organisms

Antibiotics	Micro-organisms (inhibition zone in mm)					
	Gram-positive			Gram-negative		
	<i>B.subtilus</i>	<i>S. aureus</i>	<i>S. epiderms</i>	<i>E.coli</i>	<i>S.flexineria</i>	<i>P.auriginosa</i>
Cephalexin	15	20	16	-	-	-
Tobramycin	-	-	-	-	-	-
Ampicillin	7	10	7	-	-	-
Erythromycin	7	8	6	-	-	-
Absence of inhibition						

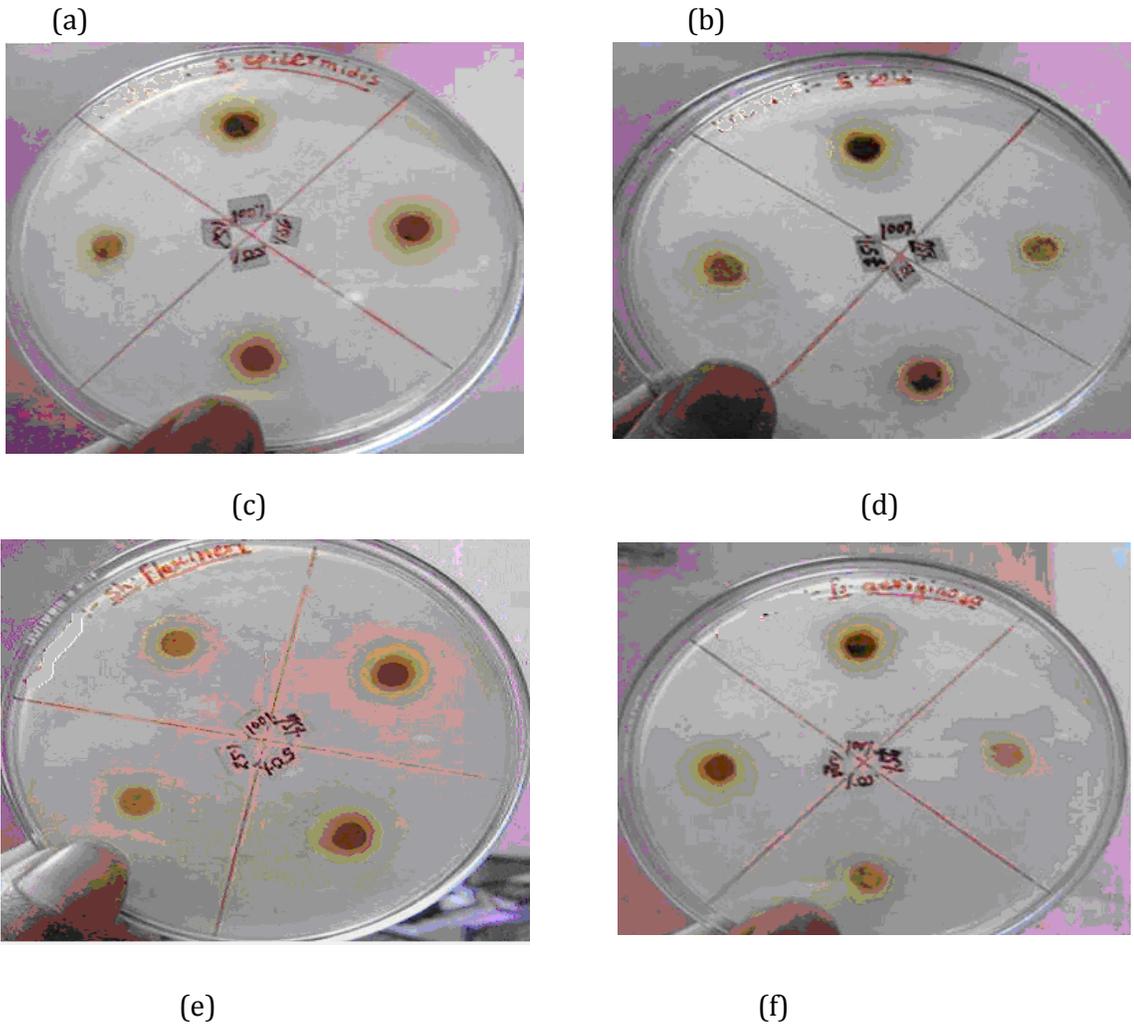
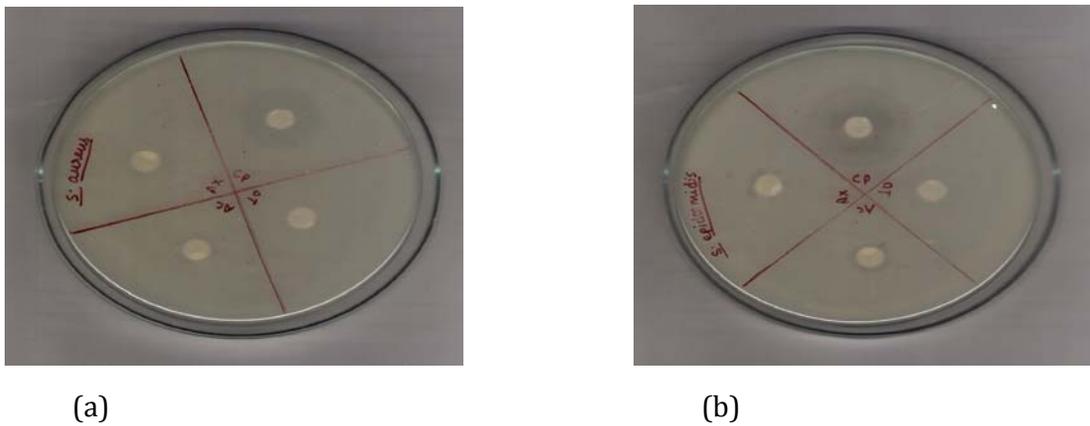
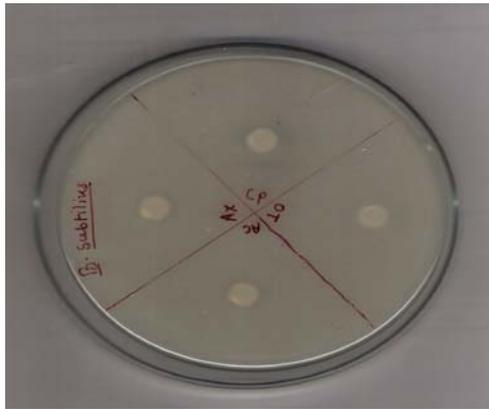


Fig. 1: Photographs showing the zone of inhibition of *T. chebula* extract against various micro-organisms *B.subtilis* (a), *S.aureus* (b), *S.epidermis* (c), *E. coli* (d), *S. flexinaria* (e), *P. auriginosa* (f).





(c)

Fig. 2: Antimicrobial activity of standard antibiotic against different micro organism: *S.aureus* (a), *S. epidermis* (b), *B.subtilis* (c).

CONCLUSION

In conclusion it may be said that whole extract can be used for protection against gram positive and gram negative bacteria. However, further studies on the extract are needed to pinpoint the finding. This report may serve as a footstep on this aspect.

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