

ANTIBACTERIAL ACTIVITY OF AQUEOUS ALCOHOLIC EXTRACT OF *ABUTILON INDICUM* AERIAL PARTS AGAINST *ENTEROCOCCUS FAECALIS* – AN *IN VITRO* STUDYJANNATHUL FERDIOZ¹, ANITHA ROY^{2*}¹Department of Dental Surgery, Saveetha Dental College and Hospitals, Chennai, Tamil Nadu, India. ²Department of General Pharmacology, Faculty of Pharmacology, Saveetha Dental College and Hospitals, Chennai, Tamil Nadu, India. Email: anitharoy2015@gmail.com

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ABSTRACT**Objective:** To evaluate the antibacterial activity of the aqueous alcoholic extracts of *Abutilon indicum* aerial parts against *Enterococcus faecalis*.**Methods:** Agar well diffusion assay, as well as microbroth dilution assays, were used for determination of antibacterial activity. The assay was done in triplicate, and chlorhexidine 0.2% was used as the control.**Results:** Different concentrations (200, 400, and 800 mg/mL) of the ethyl acetate extract of *A. indicum* aerial parts showed dose-dependant antibacterial activity. The maximum zone of inhibition was 30 mm at the maximum concentration used (800 mg/mL), and the minimum inhibitory concentration/minimum bactericidal concentration was found to be 200 mg/mL.**Conclusion:** This study suggests that the aqueous alcoholic extract of *A. indicum* aerial parts contains promising antibacterial substances which are having activity against *E. faecalis*. *E. faecalis* being one of the major threats for root canal failure during endodontic treatment; plants with antibacterial activity against *E. faecalis* will be promising.**Keywords:** *Abutilon indicum*, *Enterococcus faecalis*, Antibacterial activity, Root canal failure.© 2017 The Authors. Published by Innovare Academic Sciences Pvt Ltd. This is an open access article under the CC BY license (<http://creativecommons.org/licenses/by/4.0/>) DOI: <http://dx.doi.org/10.22159/ajpcr.2017.v10i5.15636>**INTRODUCTION**

Endodontic infections are one of the common problems associated with oral cavity, and bacteria are the most common microorganisms occurring in endodontic infections. More than 400 different microbial species have been identified in different types of endodontic infections. *Enterococcus faecalis* is significantly more associated with asymptomatic cases of primary endodontic infections and is commonly found in cases of failed endodontic therapy [1]. *E. faecalis* is a normal inhabitant of the oral cavity. It is a facultative organism, that is, non-fastidious and easy to grow [2,3]. *E. faecalis* is the most resistant microorganism in endodontic infections and is a possible cause of root canal treatment failure and persistent infections [1,4-6]. Regular procedures use root canal irrigation. The irrigants that are currently used during cleaning and shaping include sodium hypochlorite, chlorhexidine, ethylenediaminetetraacetic acid, and mixture of a tetracycline isomer, acid and detergent. All the ideal properties required for an ideal irrigant is not with any of these irrigants. Hence, there is a continuous search for an ideal irrigant to completely clean infected root canals [7]. In such a scenario, plant products with activity against *E. faecalis* will be promising.

Abutilon indicum L. (*Malvaceae*), commonly known as Indian Abutilon or Indian mallow, is a small shrub, native to tropical and subtropical regions [8]. The plant is used in inflammation, piles, gonorrhoea treatment, and as an immune stimulant. It also has diuretic, antihelminth, sedative, analgesic, antioxidant, antidiabetic, and antibacterial properties. The plant contains many flavonoids, β -sitosterol, gallic acid, geraniol, caryophyllene, etc. [9,10]. As the plant is known for its antimicrobial property, in this study, we have explored its activity against *E. faecalis*.

METHODSAqueous alcoholic extract of *A. indicum*

- Microbial strain used: *E. faecalis* ATCC
- Culture media: Mueller-Hinton agar.

Standardization of bacterial suspension

The bacterial suspension was standardized following the CLSI guidelines and was grown in Mueller-Hinton broth (HiMedia) for 18-24 hrs, followed by the matching of bacterial suspension to the turbidity equivalent to 0.5 McFarland solution ($1-2 \times 10^8$ CFU/mL) with the addition of sterile saline.

Agar well diffusion method

Evaluation of the antimicrobial activity of the extracts was conducted according to the agar well diffusion method [11,12]. The different concentrations (200, 400, and 800 mg/mL) of the plant extract was prepared and from this 100 μ L was used for this study. 0.2% chlorhexidine was used as the control. The study was carried out in triplicate.

Minimum inhibitory concentration and minimum bactericidal concentration [13,14]

The MIC of the aqueous alcoholic extract of *A. indicum* was determined by microbroth dilution method using 96-well plates. The MIC value of the extract was determined as the lowest concentration of the extract that completely inhibited bacterial growth after 48 hrs of incubation at 37°C. For the determination of MBC, a portion of liquid (5 μ L) from each well that exhibited no growth were taken and then subcultured and incubated 37°C for 24 hrs. The lowest concentration that revealed no visible bacterial growth after sub-culturing was taken as MBC.

RESULTS AND DISCUSSION

Medicinal plants represent rich sources of antimicrobial agents used medicinally in different countries and are a source of many potent drugs used for traditional medicine [15]. Medicinal plants show antimicrobial activity by different mechanisms. They may inhibit cell wall synthesis, cause energy depletion by getting accumulated in the cell membrane, interfere with the permeability of cell membrane, cause

Table 1: Zone of inhibition produced at different concentration of aqueous alcoholic extract of *A. indicum* against *E. faecalis*

Extract	Concentration (mg)	Zone of inhibition (in mm diameter)
<i>A. indicum</i>	200	21
	400	26
	800	30
Chlorhexidine 2%		35

E. faecalis: *Enterococcus faecalis*, *A. indicum*: *Abutilon indicum*

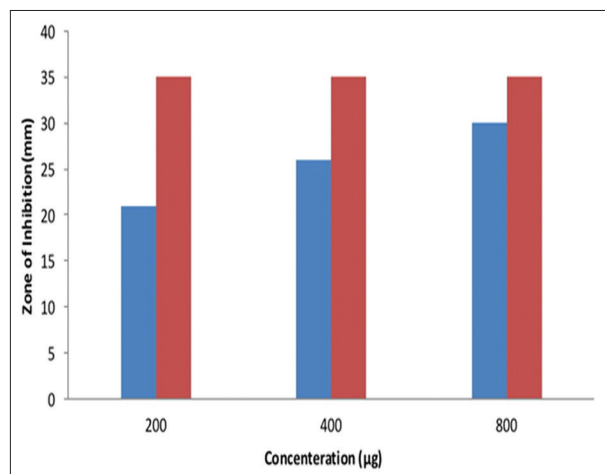


Fig. 1: Antibacterial activity of aqueous alcoholic extract of *Abutilon indicum* against *Enterococcus faecalis*

membrane disruption, modifying cellular constituents, cell damage or cell mutation [15].

In this study, different concentrations of the aqueous alcoholic extract of *A. indicum* showed maximum zone inhibition in a dose-dependent manner (Table 1). Maximum zone of inhibition was found to be 30 mm at 800 mg/mL (Fig. 1). The MIC/MBC was found to be 200 mg/mL. *A. indicum* is rich in β -sitosterol, fumaric, p-coumaric, vanillic, caffeic, and p-hydroxybenzoic, p- β -D-glucosyloxybenzoic acids, and glucovanilloyl glucose, fructose, aspartic acid, histidine, threonine, serine, leucine, galactose, and galacturonic acids, and this may be the reason for its antibacterial activity.

CONCLUSION

This study suggests that the aqueous alcoholic extract of *A. indicum* aerial parts contains promising antibacterial substances which are

having activity against *E. faecalis* and may be considered for the clinical purpose for management of *E. faecalis* infections.

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REFERENCES

- Rôças IN, Siqueira JF Jr, Santos KR. Association of *Enterococcus faecalis* with different forms of periradicular diseases. *J Endod* 2004;30(5):315-20.
- Orstavik D, Haapasalo M. Disinfection by endodontic irrigants and dressings of experimentally infected dentinal tubules. *Endod Dent Traumatol* 1990;6(4):142-9.
- Haapasalo M, Orstavik D. *In vitro* infection and disinfection of dentinal tubules. *J Dent Res* 1987;66(8):1375-9.
- Molander A, Reit C, Dahlén G, Kvist T. Microbiological status of root-filled teeth with apical periodontitis. *Int Endod J* 1998;31(1):1-7.
- Sundqvist G, Figdor D, Persson S, Sjögren U. Microbiologic analysis of teeth with failed endodontic treatment and the outcome of conservative re-treatment. *Oral Surg Oral Med Oral Pathol Oral Radiol Endod* 1998;85(1):86-93.
- Distel JW, Hatton JF, Gillespie MJ. Biofilm formation in medicated root canals. *J Endod* 2002;28(10):689-93.
- Available from: https://www.aae.org/uploadedfiles/publications_and_research/endodontics_colleagues_for_exc_ellence_newsletter/rootcanal_irrigantsdisinfectants.pdf.
- Kirtikar KR, Basu BD. *Indian Medicinal Plants*. 2nd ed., Vol. 1. New Delhi, India: Bishen Singh, Mahendra Pal Singh; 1980. p. 314-5.
- Kashmiri MA, Yasmin S, Ahmad M, Mohy-ud-Din A. Characterization, compositional studies, antioxidant and antibacterial activities of seeds of *Abutilon indicum* and *Abutilon muticum* grown wild in Pakistan. *Acta Chim Slov* 2009;56:345-52.
- Lakshmayya R, Nelluri NR, Kumar P, Agarwal NK, Gouda TS, Setty SR. Phytochemical and pharmacological evaluation of leaves of *Abutilon indicum*. *Indian J Tradit Knowledge* 2003;2:79-83.
- Pottumarthy S, Fritsche TR, Jone RN. Activity of gemifloxacin tested against *Neisseria gonorrhoeae* isolates including antimicrobial-resistant phenotypes. *Digno Microbiol Infect Dis* 2006;54:127-34.
- Bishnu PM, Pankaj B, Pratibha A, Kashi RG, Sanjiv N, Nabara D *et al*. Evaluation of antibacterial activity of some traditionally used medicinal plants against human pathogenic bacteria. *BioMed Res Int* 2015;2015:1-6.
- Anitha R, Geetha RV, Lakshmi T. Evaluation of antimycotic activity of aqueous and ethanolic extracts of *Aesculus hippocastanum* – An *in vitro* study. *Int J Drug Dev Res* 2011;3:335-8.
- Kang CG, Hah DS, Kim CH, Kim YH, Kim E, Kim JS. Evaluation of activity of the methanol extracts from 8 traditional medicinal plants. *Toxicol Res* 2011;27(1):31-6.
- Achika JI, Ndukwe GI, Ayo RG. Phytochemical screening and antimicrobial studies of aerial part of *Aeschynomene uniflora Mey*. *Ind Chem* 2016;2:113.