



IN VITRO ANTIBACTERIAL AND ANTIFUNGAL ACTIVITY OF AQUEOUS LEAF EXTRACT OF *TRIDAX PROCUMBENS*

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ABSTRACT

Bioavailability and the presence of secondary metabolites encourage research on medicinal plants to explore novel antimicrobial drugs. Antimicrobial activity was studied using agar disk-diffusion method (Kirby-Bauer method) against some selected microbial species *Escherichia coli*, *Klebsiella pneumoniae*, *Enterococcus species*, *Pseudomonas aeruginosa*, *Bacillus cereus*, *Salmonella typhi*, *Beta Streptococcus*, *Staphylococcus aureus*, *Candida albicans* and *Aspergillus niger* at 6.67%, 33.33%, 50%, 66.67% and 100% of its aqueous dilutions. The aqueous extract of *Tridax procumbens* leaves demonstrates significant antimicrobial activity against the selected microbial species, except for *A. niger*. Activity increases with an increase in concentration of the aqueous extract. Comparable activity has been recorded against MDR bacterial species *E. coli*, *P. aeruginosa* and *S. aureus*. The recorded activity is found to be superior to the control against

MDR *Enterococcus* (17.6mm) and *B. cereus* (19.5mm) species. The leaf extract can be a potential drug candidate for treating the microbial infections.

KEYWORDS: *Tridax procumbens*, antibacterial, antifungal, aqueous, bioactive compounds.

INTRODUCTION

Bacterial resistance against various antibacterial drugs that are being used to control infections is increasing nowadays. The presence of useful metabolites in medicinal plants

attracts researchers towards the exploration of novel chemical compounds and their pharmacological properties.

Tridax procumbens, a perennial plant belonging to the family Asteraceae, is widely distributed in Asia and Africa in almost all seasons. *Tridax procumbens* has been used traditionally to treat asthma, fever, epilepsy, cough, and diarrhea because of its pharmacological activities, which include anti-inflammatory, antihyperuricemic, wound-healing, anticancer, antidiabetic, larvicidal, cardioprotective, antiulcer, hepatoprotective, antimicrobial, antioxidant, hypolipidemic and analgesic effects.^[1-4]

Some important phytochemical compounds that have been reported include β -amyron, β -amyrin, lupeol, luteolin, stigma sterol, campasterol, fucosterol, arachidic acid, palmitic acid and lauric acid.^[4-6] Bioactive compounds contribute to its pharmacological activities, which enable its usage against various ailments in various parts of different countries.

Aqueous, ethanol, and methanol extracts of this plant have been reported to have antibacterial activity against *Escherichia coli*, while its ethyl acetate extract possesses antibacterial activity against *Escherichia coli*, *Salmonella typhi*, *Klebsiella pneumoniae*, *Staphylococcus aureus* and *Bacillus cereus*.^[6-8] The present study examines the antibacterial as well as antifungal activity of the plant species against some selected microbial species.

MATERIALS AND METHODS

COLLECTION AND PREPARATION OF PLANT EXTRACTS

The fresh young leaves of the plant samples were collected and grounded well using mortar and pestle. The fresh juicy extracts were squeezed out, filtered using Whatmann No.1 filter paper, refrigerated and used for antimicrobial activity.

TESTED MICROORGANISMS

The crude aqueous extracts of *Tridax procumbens* were tested for their antimicrobial activity against eight different bacterial strains *Escherichia coli*, *Klebsiella pneumoniae*, *Enterococcus species*, *Pseudomonas aeruginosa*, *Bacillus cereus*, *Salmonella typhi*, *Beta Streptococcus* and *Staphylococcus aureus* as well as against fungal species *Candida albicans* and *Aspergillus niger* at 6.67%, 33.33%, 50%, 66.67% and 100% of its aqueous dilutions (v/v) using agar disk-diffusion method (Kirby-Bauer method). For antibacterial studies, the control used was Amikacin and for antifungal studies, Nystatin was the control.

RESULTS AND DISCUSSION

The aqueous dilutions of the plant species exhibit significant antibacterial and antifungal activity against all the microbial strains *Escherichia coli*, *Klebsiella pneumoniae*, *Enterococcus species*, *Pseudomonas aeruginosa*, *Bacillus cereus*, *Salmonella typhi*, *Beta Streptococcus*, *Staphylococcus aureus* and *Candida albicans*, except *Aspergillus niger*.

Tridax procumbens shows an increase in activity against all the microbial species with an increase in concentration. At its lowest concentration of 6.67%, no activity was recorded against either bacterial or fungal species. Maximum activity was observed for its 100% pure extract. Compared to the control, greater activity was recorded against *Enterococcus* (17.6mm against 17mm) and *Bacillus cereus* (19.5mm against 16mm) species. Moreover, its activity was comparable with that of the control against *E. coli*, *P. aeruginosa* and *S. aureus*. Activity is minimal against beta *Streptococcus* species (Table 1).

Table 1: Antibacterial activity of aqueous leaf extract of *Tridax procumbens*.

Bacterial Species	Zone of Inhibition Diameter (mm)					
	6.67%	33.33%	50%	66.67%	100%	Amikacin
<i>E.coli</i>	-	14	15.1	16	18	19
<i>Klebsiella pneumoniae</i>	-	-	13	16	17.3	20
<i>Pseudomonas aeruginosa</i>	-	12	14	16	17.2	18
<i>Salmonella typhi</i>	-	12.3	13	14.6	16	19
<i>Enterococcus</i>	-	11.6	13	15.2	17.6	17
<i>Staphylococcus aureus</i>	-	11	13.6	15	16.4	17
<i>Bacillus cereus</i>	-	14	16	18	19.5	16
<i>Beta streptococcus</i>	-	-	-	12.5	14	20

Multidrug resistance has been increasing the focus towards plant-based antimicrobial compounds. Gram-negative bacterial species such as *A. baumannii*, *E. coli*, *K. pneumoniae*, *P. aeruginosa* and gram-positive bacterial species *S. aureus*, *E. faecium*, *E. faecalis* and *S. pneumoniae* have been documented to exhibit antibiotic resistance.^[9] Reports emphasize the importance of drugs acting against antibiotic-resistant microbial species, as patients infected by such species are more vulnerable to their consequences, which sometimes can be fatal also.

Gram-positive bacteria, which possess a thick layer of peptidoglycan over the cytoplasmic membrane, develop resistance^[10] either by their beta-lactamase secretion or by reducing the affinity and susceptibility of their target site.^[11, 12] The extract records a comparable activity against gram-positive *S. aureus* (16.4mm against 17mm) which is responsible for fetal illness

and increased mortality rates.^[13, 14] Again, the extract proves to be superior against gram-positive multidrug-resistant *Enterococcus* species and *B. cereus*, which is also considered to be an emerging antibiotic-resistant species.^[15]

Gram-negative bacteria, which produce antibiotic-inactivating enzymes, are reported to be the cause of difficult nosocomial infections. *P. aeruginosa*, a gram-negative bacterium that has been reported to be a dreadful pathogen, can also show antibiotic resistance by gene mutation apart from beta-lactamase secretion.^[9,16] Comparable activity against the species is recorded (17.2mm against 18mm) for the plant extract. Again, the extract possesses comparable activity against *E. coli*, another multidrug-resistant gram-negative species that develops resistance by altering the antibiotic's target site and reducing intracellular concentration via efflux pumps and membrane permeability changes apart from beta-lactamase secretion, to that of the control.^[17, 18]

Candida albicans and *A. niger* can cause mucosal and systemic infections in immunocompromised patients.^[19, 20] Though the extract shows no activity against *A. niger* species, the zone of inhibition diameter is significant against *C. albicans*, with a maximum activity (15.6mm) recorded for its 100% pure extract (Table 2).

Table 2: Antifungal activity of aqueous leaf extract of *Tridax procumbens*.

Fungal Species	Zone of Inhibition Diameter (mm)					
	6.67%	33.33%	50%	66.67%	100%	Nystatin
<i>Candida albicans</i>	-	11.2	13	14	15.6	19
<i>Aspergillus niger</i>	-	-	-	-	-	13

Antimicrobial activity of plant extracts can be mainly attributable to the presence of bioactive compounds. Many bioactive compounds have been identified and isolated from the plant species, which include procumbetin^[21], 8,3'-dihydroxy-3,7,4'-trimethoxy-6-O- β -d-glucopyranosyl flavone, 6,8,3'-trihydroxy-3,7,4'-trimethoxyflavone, puerarin^[22], centaureidin, centaurein^[23], methyl 14-oxononacosanoate, methyl 14-oxooctadecanoate, 30-methyl-28-oxodotriacont-29-en-1-oic acid, β -amyrone, lupeol, β -amyrin and fucosterol.^[24] Furthermore, phenolic acids, including benzoic, benzeneacetic acids, vanillic and guaiacol, have also been identified.^[25]

CONCLUSION

Tridax procumbens leaf extract possessing bioactive compounds exerts antimicrobial activity against all the selected bacterial species and against the *C. albicans* fungal species. Moreover, its activity is higher against the gram-negative *Enterococcus* and *B. cereus* species compared to that of the control. The extract can be used to develop antimicrobial compounds after investigating and isolating bioactive compounds.

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