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Antibacterial Properties of *Tchihatchewia isatidea* Boiss. from Turkey

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Abstract
Development of antimicrobial agents obtained from plant extract is among the noteworthy topics to resolve the bacterial problems associated with environment, food safety, animal and human health. In this study, antibacterial activity of methanolic extract of *Tchihatchewia isatidea* Boiss. (crude extract) was determined. Dried powder of the parts (flower, leave and stem) of *Tchihatchewia isatidea* Boiss. were kept in methanol for 3 days in dark place. The antibacterial activity of crude extract was determined by disc diffusion method. Methanol-impregnated discs were used as negative control. Also commercial antibiotic discs (Clindamycin (10 µg), Tetracycline (10 µg) and Amoxicillin-Clavulanic acid (30 µg)) were utilized as positive control. According to the experimental results of this study, the methanolic extract of *Tchihatchewia isatidea* Boiss. (At the doses of 800 µg/10µl, 400 µg/10µl, 200 µg/10µl and 100 µg/10µl) have been found to have antibacterial activity against *Staphylococcus aureus* ATCC 29213. It was determined that methanolic extract of *Tchihatchewia isatidea* Boiss. has showed no antibacterial activity against *Bacillus subtilis* ATCC 6633, *Escherichia coli* ATCC 25922, *Enterococcus faecalis* ATCC 29212, *Salmonella typhimurium* ATCC 14028, *Streptococcus mutans* CNCTC 8/77, *Streptococcus mitis* CNCTC 4/77, *Streptococcus salivarius* CNCTC 64/59 and *Staphylococcus epidermidis* ATCC 12228. The antibacterial activity of methanolic extract of *Tchihatchewia isatidea* Boiss. was investigated in this study for the first time.
Key words: Antibacterial Activity, Disc Diffusion Method, *Tchihatchewia isatidea* Boiss.

INTRODUCTION

Development of new, reliable, cost-efficient and non-toxic herbal antimicrobial agent is very important in minimizing environmental and health problems [1]. Although conventional drugs usually provide effective antibiotic therapy for bacterial infections, there is an increasing problem of antibiotic resistance and a continuing need for new solutions [2]. National center for complementary and alternative medicine (NCCAM) is the agency for scientific researches about herbal medicine [3].

The roots of *Tchihatchewia isatidea* Boiss. are used for treating wounds in folk medicine (Figure 1 and Figure 2.) [4]. The local names of this plant are allıgelin and boya çiçeği. *Tchihatchewia isatidea* Boiss.(Brassicaceae) is endemic to Turkey (Erzincan, Erzurum, Gümüşhane, Sivas, Tunceli, Malatya) and grows on eroded hills and screes [5,6].

Figure 1. *Tchihatchewia isatidea* Boiss. [4]

Figure 2. *Tchihatchewia isatidea* Boiss. [4]

The present study determined the antibacterial activity of crude extract of aerial parts of *Tchihatchewia isatidea* Boiss. against *Staphylococcus aureus* ATCC 29213, *Bacillus subtilis* ATCC 6633, *Escherichia coli* ATCC 25922, *Enterococcus faecalis* ATCC 29212, *Salmonella typhimurium* ATCC 14028, *Streptococcus mutans* CNCTC 8/77, *Streptococcus mitis* CNCTC 4/77, *Streptococcus salivarius* CNCTC 64/59 and *Staphylococcus epidermidis* ATCC 12228.

MATERIAL AND METHOD

Tchihatchewia isatidea Boiss. were obtained from Atatürk Horticultural Central Research Institute, Yalova, Turkey in May 2014. Test microorganisms used in this study were *Staphylococcus aureus* ATCC 29213, *Bacillus subtilis* ATCC 6633, *Escherichia coli* ATCC 25922, *Enterococcus faecalis* ATCC 29212, *Salmonella typhimurium* ATCC 14028, *Streptococcus mutans* CNCTC 8/77, *Streptococcus mitis* CNCTC 4/77, *Streptococcus salivarius* CNCTC 64/59 and *Staphylococcus epidermidis* ATCC 12228 which were supplied from Microorganism Culture Collections Research and Application Center of Istanbul University and Microbiology Laboratory Culture Collection of Gebze Institute of Technology.

Dried powder of flower, leave and stem parts of *Tchihatchewia isatidea* Boiss. were kept in methanol for 3 days in dark place at a rate of 1:10 (w/v). The solvent in the obtained extracts were removed by using rotary evaporator under vacuum at 50°C for 15 minutes. The extract concentrations were adjusted by adding own solvent to each extract at the determined rates (800 µg/10µl, 400 µg/10µl, 200 µg/10µl, 100 µg/10µl).

Bacterial suspension was obtained from overnight culture and was adjusted to 0.5 McFarland by using a densitometer. Disc diffusion method was used to determine the antibacterial activity of parts of *Tchihatchewia isatidea* Boiss. Sterile discs (6 mm in diameter) were impregnated with the 10 µl of prepared extracts. Solvent (methanol) impregnated discs were used as negative control and commercial antibiotic discs (Clindamycin (10 µg), Tetracycline (10 µg) and Amoxicillin-Clavulanic acid (30 µg)) were utilized as positive control. Prepared bacterial suspension was inoculated to Mueller Hinton Agar by using sterile swabs. The impregnated discs were slightly pressed onto the Agar. The incubation was performed at 37°C for 24 hours. After the incubation, the diameters of the inhibition zone (IZs) were measured.

RESULT AND DISCUSSION

Obtained experimental results showed that methanolic extract of *Tchihatchewia isatidea* Boiss. (At the doses of 800 µg/10µl, 400 µg/10µl, 200 µg/10µl and 100 µg/10µl) have been found to have antibacterial activity against *Staphylococcus aureus* ATCC 29213. Also methanolic extract of *Tchihatchewia isatidea* Boiss. has showed no antibacterial activity against *Bacillus subtilis* ATCC 6633, *Escherichia coli* ATCC 25922, *Enterococcus faecalis* ATCC 29212, *Salmonella typhimurium* ATCC 14028, *Streptococcus mutans* CNCTC 8/77, *Streptococcus mitis* CNCTC 4/77, *Streptococcus salivarius* CNCTC 64/59 and *Staphylococcus epidermidis* ATCC 12228. Experiments of this study were performed two times under aseptic conditions and the diameters of IZs were the average of two replicates. Inhibition zone diameters of methanolic extracts of *Tchihatchewia isatidea* Boiss. were presented in Table 1. The inhibition zone diameters of Clindamycin (10 µg), Tetracycline (10 µg) and Amoxicillin-Clavulanic acid (30 µg) against *S. aureus* are 31 mm, 19 mm, 30 mm respectively.

CONCLUSION

In the present study we report that the crude extracts obtained from flowers, leaves and stems of *Tchihatchewia isatidea* Boiss. have antibacterial activity against *Staphylococcus aureus* ATCC 29213. This plant extract may be used as natural antimicrobial instead of chemical preservatives used in food industry, nutraceuticals, cosmetic industry and pharmaceuticals. Further studies on this species may be performing the toxicological studies because of some potentially harmful compounds. Furthermore, this is the first study to show that *Tchihatchewia isatidea* Boiss. has antibacterial effect against *Staphylococcus aureus* ATCC 29213.

Table 1. Inhibition zone diameters (IZs) of methanolic extract of *Tchihatchewia isatidea* Boiss. against *S. aureus*

	EXTRACT (µg/disc)	IZs (mm)	INHIBITION ZONE
FLOWER	Methanolic Extract		800µg/disc
	800	8.3	
	400	8.0	
	200	6.9	
	100	6.8	
LEAVE	Methanolic Extract		800µg/disc
	800	7.9	
	400	7.7	
	200	7.3	
	100	7.1	
STEM	Methanolic Extract		800µg/disc
	800	9.1	
	400	8.9	
	200	7.5	
	100	7.1	

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