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ANTIMICROBIAL ACTIVITIES OF SOME MEDICINAL PLANTS FROM TURKEY

ABSTRACT

In this study, the antimicrobial activities of *Achillea teretifolia* Wild., *Origanum acutidens* (Hand.-Mazz.) Letswaart, *Nepata italica* L., *Stachys lavandifolia* Vahl, var. *lavandifolia*, *Mentha spicata* L. subsp. *spicata* were determined which grown as naturally in Turkey. These plants were prepared with methanol and antimicrobial activities of these extracts have been examined on test microorganisms as follows: *Pseudomonas aeruginosa* DMS 50071 SCOTTA, *Klebsiella pneumoniae* FMC 5, *Staphylococcus aureus* COWAN 1, *Bacillus megaterium* DSM 32, *Candida albicans* FMC 17, *Candida globrata* ATCC 66032, *Epidermophyton* sp. and *Trichophyton* sp. by disk diffusion methods. Result of present study showed that the extracts of plant inhibited the growth of microorganisms (9-27 mm) in the different ratio. However, They had no effect against some bacteria and yeasts used in study.

Keywords: Antimicrobial Activity, Agar Disc Diffusion Method, Medicinal Plants, Plant Materials, Test Microorganisms

TÜRKİYE'DE BAZI TIBBİ BİTKİLERİN ANTİMİKROBİYAL AKTİVİTELERİ

ÖZET

Bu çalışmada, Türkiye de doğal olarak yetişen *Achillea teretifolia* Wild., *Origanum acutidens* (Hand.-Mazz.) Letswaart, *Nepata italica* L., *Stachys lavandifolia* Vahl, var. *lavandifolia*, *Mentha spicata* L. subsp. *spicata*'nın antimikrobiyal aktiviteleri belirlendi. Metanol ile hazırlanan bu bitki ekstraktlarının antimikrobiyal aktiviteleri, *Pseudomonas aeruginosa* DMS 50071 SCOTTA, *Klebsiella pneumoniae* FMC 5, *Staphylococcus aureus* COWAN 1, *Bacillus megaterium* DSM 32, *Candida albicans* FMC 17, *Candida globrata* ATCC 66032, *Epidermophyton* sp. and *Trichophyton* sp. 'ye karşı disk diffüzyon metodu ile incelendi. Bu çalışmanın sonucu bitki ekstraktlarının mikroorganizmaların gelişmelerini farklı oranlarda engellediğini göstermiştir (9-27 mm arasında inhibisyon zonları). Bununla beraber, bitki ekstraktları çalışmada kullanılan bazı mikorganizmalar üzerinde etki etmemiştir.

Anahtar Kelimeler: Antimikrobiyal Aktivite, Agar Disk Diffüzyon Metodu, Tıbbi Bitkiler, Bitki Materyalleri, Test Mikroorganizmaları

1. INTRODUCTION (GİRİŞ)

Various medical plants have been used for years in daily life to treat disease all over the world. According to a study performed by the WHO based on publications on pharmacopoeias and medical plants in 91 countries, the number of medicinal plants is nearly 20.000 [1 ve 2]. Traditional medical treatments in daily life are now being used with empiric methods [1].

In particular, the antimicrobial activity of these plants has formed the basis of many applications, including raw and processed food preservation, pharmaceuticals, alternative medicine, and natural therapies [3].

The effects of different plants extracts on pathogens microorganism studied by a very large number of researchers in different parts of the world [4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16 ve 17]. Plants used in the present study; herbal parts of some Achillea species are used as a folk remedy to treat abdominal pain, wounds and stomachache also these species are used in cosmetics, fragrances and agriculture, e.g., plant protection [18]. Origanum species are traditionally used as sedative, diuretic, degasifier, sweater and antiseptic, and also in the treatment of gastrointestinal diseases and constipation [19]. Several species of Nepeta genus are utilized in folk medicine for treatment of contusions, rheumatic pains, fever, cutaneous eruptions [20]. The genus Stachys, which belongs to the Lamiaceae family, consists of about 300 species widespread throughout the world are used for diuretic, gastric effects [19 ve 21]. The boiled leaves of Mentha spicata is being used to relieve hiccup, flatulence, giddiness and as remedy for inflammation, bronchitis, and to control vomiting during pregnancy [19 ve 22].

2. RESEARCH SIGNIFICANCE (ÇALIŞMANIN ÖNEMİ)

The aim of this work is to evaluate the potential antimicrobial activities of *A. teretifolia*, *O. acutidens*, *N. italica*, *S. avandifolia* var. *lavandifolia*, *M. Spicata* subsp. *spicata* which used in folklore medicine of Turkey.

3. MATERIAL- METHOD (MATERYAL-METOT)

3.1. Obtained of Plant Materials and Preparation (Bitki Materyalinin Elde Edilmesi ve Hazırlanması)

A. teretifolia, *O. acutidens*, *N. italica*, *S. lavandifolia* var. *lavandifolia*, *Mentha spicata* subsp. *spicata* were collected from east Anatolia region (Elazığ, Tunceli, Malatya) in the Eastern Anatolia of Turkey. The taxonomic identification of plant materials was determined using by flora of Turkey [23]. The dried and powdered plant materials (20g) were extracted in 400 ml methanol solvent by keeping on a rotary shaker for 24 h. The aqueous extracts were filtered using Whatman filter paper (No.1) and then consantrated *in vacuo* at 37°C using a Rotary evaporator. They were stored at 4°C for further studied. Then, All the extracts obtained were injected into blank antibiotic disks of 6 mm diameter (Oxoid) in amounts of 80 µl.

3.2. Test Microorganisms (Test Mikroorganizmaları)

A total 4 bacteria (*Pseudomonas aeruginosa* DMS 50071, *Klebsiella pneumoniae* FMC 5, *Staphylococcus aureus* COWAN 1, *Bacillus megaterium* DSM 32), 4 yeasts (*Candida albicans* FMC 17, *Candida globrata* ATCC 66032, *Epidermophyton* sp. and *Trichophyton* sp.) species were used in the present investigetion. The list of microorganisms used is given in table 1. Microorganisms were provided by the Department of Biology,

Faculty of Science and Arts, Firat University, Microbiology Laboratory, Elazig-Turkey.

3.3. Antimicrobial Activity (Antimikrobiyal Aktivite)

Antimicrobial tests were carried out by disc diffusion method using 100 µl of suspension containing 10^6 per/ml of bacteria, 10^4 per/ml yeast and 10^4 per/ml dermatofit fungi inoculated into mueller hinton agar (Difco), malt extract agar (Difco) and Glukoz Sabouroud agar (Difco), respectively. The discs (6 mm diameter) were impregnated with 80 µl placed on the inoculated agar. Petri dishes were placed at 4°C for 2 h. Then, the inoculated plates were incubated at 37±0.1°C at 24 h for bacterial strains and also at 25±0.1°C at 72 h for yeast and dermatofit fungi. Antimicrobial activity was determined by measuring the zone of inhibition against the test organisms [24].

4. RESULTS AND DISCUSSIONS (BULGULAR VE TARTIŞMA)

The in vitro antimicrobial activities of plant extracts, control groups and standart antibiotic are showed on Table 1.

The methanolic extracts of *A. teretifolia*, *O. acutidens*, *N. italica*, *S. lavandifolia* var. *lavandifolia*, *Mentha spicata* subsp. *spicata* exhibited activity against bacteria, fungi (Table 1). it seems that the antimicrobial activity of those plant extracts are variable as seen in Table 1 and plant extracts of generated inhibition zones smaller than those generated by have a higher activity usually as to comparasion antibiotic.

The extracts of *A. teretifolia* showed all of the tested microorganisms from high to low respectivelly; *B. megaterium* (18 mm), *S. aureus* (15 mm), *P. aeruginosa* (15 mm), *K. pneumoniae* (13 mm), *C. albicans* (13 mm), *Epidermophyton sp.* (11 mm), *Trichophyton sp.* (10 mm), *C. globrata* (9 mm) (table 1). These findings are congrued with several literatures. For example, Unlu et al. [25] found that air-dried aerial parts of *Achillea setacea* and *Achillea teretifolia*, exhibited inhibitory effects on *Clostridium perfringens*, *Acinetobacter lwoffii* and *Candida albicans*. In another study done by Barış et al. [18] indicated that the essential oil from *Achillea biebersteinii* Afan. (Asteraceae)exhibited antimicrobial activity against 8 bacteria, 14 fungi and the yeast, whereas methanolic extract of these species remained inactive.

The extracts of *O. acutidens* was more effective against *P. aeruginosa* (27 mm), *K. pneumoniae* (25 mm), *Epidermophyton sp.*(27 mm) and *Trichophyton* sp.(24 mm) (Table 1). But it was observed less activity against *S. aureus* and *B. megaterium* (13 mm, 17 mm inhibition zone respectivelly). But this plant extract did not show any activity to the other tested microorganisms; *C. albicans* and *C. globrata* as seen in table 1. In a previous study was demonstrated that 10 µl of plant extracts generally have antimicrobial activity against gram positive bacteria but had no antimicrobial activity against gram negative bacteria and yeast [5]. Antifungal assays showed that *O. acutidens* oil, carvacrol and thymol completely inhibited mycelial growth of 17 phytopathogenic fungi of which antifungal effects were higher than commercial fungicide, benomyl as reported by Kordali et al [16].

The extracts of *N. italica* showed inhibition zone above all of the tested microorganisms except for *C. globrata* and *Epidermophyton sp.* which from high to low respectivelly; *P. aeruginosa* (17 mm), *S. aureus* (16 mm), *K. pneumoniae* (15 mm), *B. megaterium* (13 mm), *Trichophyton sp.* (12 mm), *C. albicans* (11 mm) (Table 1).

The extract of *S. lavandifolia* var. *lavandifolia* did not show any antimicrobial activity against to *B. megaterium*, *Trichophyton sp.* (Table 1). However, it showed activity against to other tested microorganisms; *P. aeruginosa*, *K. pneumoniae*, *S. aureus*, *C. albicans*, *C. globrata* and *Epidermophyton sp.* (11 mm, 13 mm, 11 mm, 15 mm, 10 mm, 13 mm inhibition zone respectivelly), Several studies determined similar effects [12 ve 17].The methanolic extracts of *Phlomis bruguieri*, *P. herba-venti*, *P. olivieri*, *Stachys byzantina*, *S. inflata*, *S. lavandulifolia* and *S. laxa* exhibited concentration-dependent (10, 50,100, 250, 500, 750, and 1000 µg/disc) activity against *S. aureus*, *Streptococcus sanguis*, *Escherichia coli*, *P. aeroginosa*, *K. pneumoniae*, while the extracts did not show any activity to *Aspergilus niger* and *C. albicans* [12]. The extracts of *Stachys byzantina*, *S. inflata*, *S. lavandulifolia* and *S. laxa* exhibited concentration-dependent activity against against Gram (+) microorganisms. The extracts, however, did not show any antifungal activity [17].

The extract of *M. spicata* subsp. *spicata* showed each other not far values inhibition zone to *P. aeruginosa* (11 mm), *K. pneumoniae* (9 mm), *S. aureus* (10 mm), *B. megaterium* (9 mm), *C. albicans* (11 mm), while were not able to inhibit any of the *C. globrata*, *Epidermophyton sp.* and *Trichophyton sp.* [Table 1]. In a previous study was demonstrated that the methanol extract from *Mentha longifolia* subsp. *longifolia* which one of *Mentha* species almost remained inactive against all 30 microorganisms tested whereas the essential oil from *M. longifolia* subsp. longifolia showed strong antimicrobial activity [14].

Present results revealed that the extract of *A. teretifolia*, *O. acutidens*, *N. italica*, *S. lavandifolia* var. *lavandifolia*, *M. spicata* subsp. *spicata* have antimicrobial activitiy against some microrganisms. However; it was no observed any antagonistic effect against to other some bacteria and yeasts.

According to our results, *A. teretifolia*, *O. acutidens*, *N. italica*, *S. lavandifolia* var. *lavandifolia*, *Mentha spicata* subsp. *spicata* could be used as raw materials for phytotherapy because of their antimicrobial activities. Moreover, phytochemical studies are required to identify the types of natural compounds and active agents responsible for the antimicrobial effects of these medicine plants. In addition, the results from present study confirmed the use of these plants in traditional medicine for the treatment of infectious disease.

Table 1. Antimicrobial activity of some medicinal plants taxa
(Tablo 1. Bazı tibbi bitki taksonlarının antimikrobial aktivitesi)

Materials	A. t	O. a	N. i	S. l	M. s	Control	Standart
Microorganisms	İnhibition zone (mm)						
P. aeruginosa	15	27	17	11	11	na	11**
K. pneumoniae	13	25	15	13	9	na	9**
S. aureus	15	13	16	11	10	na	13**
B. megaterium	18	17	13	na	9	na	9**
C. albicans	13	na	11	15	11	na	18*
C. globrata	9	na	na	10	na	na	12*
Epidermophyton sp.	11	27	na	13	na	na	NT
Trichophyton sp.	10	24	12	na	na	na	NT

A. t: *A. teretifolia*,
O. a: O. *acutidens* Letswaart,
N. i: *N. italica*,
S. l: *S. lavandifolia* var. *lavandifolia*,
M. s: *M. spicata* subsp. *spicata*,
na: not active,
*: Nystatin, (30 µg/disk)
**: Streptomysin sülfat(10 µg/disk),
Control: methanol,
NT: not tested.

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