

Antimicrobial Effect of Seed Extract of Cardamom (*Elettaria cardamomum* Maton)Sema AĞAOĞLU¹✉ Nursel DOSTBİL² Süleyman ALEMDAR¹¹Yüzüncü Yıl University, Veterinary Faculty, Department of Food Hygiene and Technology, 65080 Van, TURKEY.²Yüzüncü Yıl University, Science Faculty, Department of Biology, 65080 Van, TURKEY.

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SUMMARY

In this study, antimicrobial effects of seed extract of cardamom (*Elettaria cardamomum* Maton) on some microorganisms including pathogens were investigated. For this purpose extract of cardamom seed which is prepared in diethyl ether were tested on bacterial and fungal cultures such as *Pseudomonas aeruginosa*, *Mycobacterium smegmatis*, *Klebsiella pneumoniae*, *Staphylococcus aureus*, *Escherichia coli*, *Salmonella typhimurium*, *Enterococcus faecalis*, *Micrococcus luteus* and *Candida albicans* by the paper disc agar diffusion method. According to findings, it is determined that inhibitory activity was detected on *M. smegmatis*, *K. pneumoniae*, *S. aureus*, *E. coli*, *E. faecalis*, *M. luteus* and *C. albicans* where no such activity was detected on *P. aeruginosa*. While *S. aureus* was detected to be the most sensitive strain, the least inhibitory effect was found on *E. coli*.

Keywords: Cardamom, Antimicrobial activity, Pathogen.

Kakule Tohumu (*Elettaria cardamomum* Maton) Ekstraktının Antimikrobiyal Etkisi**ÖZET**

Bu çalışmada, kakule tohumunun (*Elettaria cardamomum* Maton) bazı mikroorganizmalar üzerindeki inhibitör etkinliği araştırıldı. Bu amaçla kakule tohumunun diethyl eterde hazırlanmış ekstraktı *P. aeruginosa*, *M. smegmatis*, *K. pneumoniae*, *S. aureus*, *E. coli*, *E. faecalis*, *S. typhimurium*, *M. luteus* ve *C. albicans* üzerinde disk difüzyon metodu kullanılarak in vitro olarak denendi. Sonuç olarak; kakule tohumundan elde edilen ekstraktın test suşlarından *M. smegmatis*, *K. pneumoniae*, *S. aureus*, *E. coli*, *E. faecalis*, *M. luteus* ve *C. albicans* üzerinde farklı düzeylerde inhibitör aktivite gösterdiği, ancak *P. aeruginosa* üzerinde benzer etkinin oluşmadığı tespit edildi. Kakuleye karşı en hassas suşun *S. aureus* olduğu belirlenirken, en zayıf inhibitör etki *E. coli*'de gözlemlendi.

Anahtar kelimeler: Kakule, Antimikrobiyal etki, Patojen.

INTRODUCTION

Cardamom is the dried fruit of the tall perennial herbaceous plant, *Elettaria cardamomum* Maton, and belonging to the family *Zingiberaceae* (3). This herb is cultivated commercially in India, Sri Lanka, Guatemala and Tanzania. The leaves are lanceolate, green or dark green, glabrous on both surfaces with acuminate apex. The fruit are tri-ocular, ovoid, oblong or greenish-brown capsules containing about 15-20 reddish brown seeds. The cardamom seeds have a warm, slightly pungent and highly aromatic flavour. Therefore, it is used as a spice in meat products such as Bologna and Frankfurter (5).

The chemical composition of cardamom varies considerably with variety, region and age of the product. The content of volatile oil in the seeds is strongly dependant on storage conditions, but may be as high as 8%. The volatile oil contains about 1.5% α -pinene, 0.2% β -pinene, 2.8% sabinene, 1.6% myrcene, 0.2% α -phellandrene, 11.6% limonene, 36.3% 1,8-cineole, 0.7% γ -terpinene, 0.5% terpinolene, 3% linalool, 2.5% linalyl acetate, 0.9% terpinen 4-01, 2.6% α -terpineol, 31.3% α -terpinyl acetate, 0.3% citronellol, 0.5% nerol, 0.5% geraniol, 0.2% methyl eugenol and 2.7% trans-nerolidol (11). The basic cardamom aroma produced by a combination of the major components, 1,8-cineole and α -terpinyl acetate (12).

Cardamom oil is used in food, perfumery, and

liquor a pharmaceutical industries as a flavour and a carminative. In medicine, it is used as a powerful aromatic, antiseptic, stimulant, carminative, stomachic, expectorant, anti-spasmodic and diuretic (5,11). In some parts of the world, especially the Near East and Saudi Arabia, cardamom is used mostly in the preparation of "Gahwa" a strong cardamom coffee concoction (5).

In Turkey, consumption of coffee containing cardamom seed is common in southeast regions. In studies carried out in Turkey, the antimicrobial activities of different plants and their extracts used as spices or aromatic herbs including *Nigella sativa*, nettle, onion, garlic, peppermint, cumin, cinnamon and thyme have been investigated (1,2,7,10,13).

This study was carried out to determine whether seed extract of cardamom (*Elettaria cardamomum* Maton) has inhibitory activity on some pathogens and saprophytic microorganisms.

MATERIALS and METHODS

The cardamom (*Elettaria cardamomum* Maton) seed, material of this study were obtained from whole sales and retail organic food stores. Samples were ground in a breaker until they would pass a 1 mm sifter and they were preserved in cloth bags in the laboratory until extraction procedure (9).

Microorganisms

Standard strains of microorganisms used in the present study (*Pseudomonas aeruginosa* ATCC 27853, *Mycobacterium smegmatis* CCM 2067, *Klebsiella pneumoniae* FML 5, *Staphylococcus aureus* ATCC 25923, *Escherichia coli* ATCC 25922, *Salmonella typhimurium* KUEN 1357, *Enterococcus faecalis* ATCC 15753, *Micrococcus luteus* A 2971 and *Candida albicans* ATCC 60192) were obtained from culture collection of Yüzüncü Yıl University, Faculty of Medicine, Department of Microbiology Laboratory.

Trypticase Soy Broth (TSB, DIFCO 0369-01-4) was used for activation of cultures of microorganisms. Antibacterial activity assays were carried out using Mueller-Hinton Agar (MHA, OXOID CM337).

Preparation of model extracts

The method suggested by Hanafy and Hatem (9) was used to prepare model extracts. For this purpose, 200 g of ground samples of cardamom seeds were soaked in 500 ml of diethyl ether for 6 h. During this period the mixture was agitated every 15 min intervals and following filtration, diethyl ether was removed using an evaporator (60 °C). In analysis, dark brown and green colored, oily extracts were used without any dilution. Sample extracts were kept in freezer (+4 °C) until analysis were concluded.

Antimicrobial activity test

In the present study, disc diffusion technique as described by Hanafy and Hatem (9) was applied. The diethyl ether extracts of cardamom seed, were transferred into sterile bottles containing filter paper (Whatman No:1; 6 mm diameter). Bottles were then placed into a water bath (50 °C) for complete removal of diethyl ether with periodical shakings to allow an even distribution of the extract between discs.

All strains used in the study were inoculated to TSB agar and incubated at 35±0.1 °C for 24 h and were allowed to grow until they reach 10⁸-10⁹ cfu/ml. The 0.1 ml of inoculum from the prepared culture was transferred to MHA medium. The inoculum was spread to surface of plates with a sterile swab and the inoculated plates were dried at room temperature. Paper discs embedded within a plant extract were placed on previously inoculated plates and were incubated at 35±0.1 °C for 48 h. After incubation the zones of growth inhibition around disks were measured in mm (4). Antibacterial activity studies were carried out for each test strains in duplicate and average measurement were calculated.

RESULTS

The results of the antimicrobial activity assays indicated that cardamom seed had inhibitory activity on *M. smegmatis*, *K. pneumoniae*, *S. aureus*, *E. coli*, *E. faecalis*, *M. luteus*, and *C. albicans*; however no inhibitory activity was observed against *P. aeruginosa*. Examining Table 1; *S. aureus* which is an important pathogen in food-poisoning has been identified as the most sensitive strain against cardamom. Results of antimicrobial activity assays are represented in Table 1 and Figure 1.

Table 1. Antimicrobial activity of seed extract of cardamom

Test strains	Inhibition zone (mm)
<i>P. aeruginosa</i> (ATCC 27853)	-
<i>M. smegmatis</i> (CCM 2067)	21
<i>K. pneumoniae</i> (FML5)	14
<i>S. aureus</i> (ATCC 25923)	30
<i>E. coli</i> (ATCC 25922)	13
<i>S. typhimurium</i> (KUEN 1357)	17
<i>E. faecalis</i> (ATCC 15753)	15
<i>M. luteus</i> (A 2971)	18
<i>C. albicans</i> (ATCC 60192)	20



Figure 1. The inhibition zones of seed extract of cardamom, which were determined with standard test strains.

A) *M. smegmatis* B) *K. pneumoniae* C) *S. aureus* D) *E. coli*
E) *S. typhimurium* F) *E. faecalis* G) *M. luteus* H) *C. albicans*

DISCUSSION

The beneficial health effects of extracts from many types of plants that are used as seasoning agents in foods and beverages have been claimed for centuries (5). In this study, the purpose was to examine the inhibitory effects of cardamom (*Elettaria cardamomum* Maton) seed extract, some pathogens causing food poisoning and different illnesses in humans, and some microorganisms causing spoilage in foods were used as test strains. For this purpose, the diethyl ether extracts of cardamom seeds were tested on *P. aeruginosa*, *M. smegmatis*, *K. pneumoniae*, *S. aureus*, *E. coli*, *S. typhimurium*, *E. faecalis*, *M. luteus* and *C. albicans* with disc diffusion method as in vitro.

In this study, extract of cardamom seed displayed a variable degree of antimicrobial activity on different microorganisms. *S. aureus* was found to be more sensitive strain than the others. On the other hand *P. aeruginosa* was found to be most resistant bacteria against the cardamom seed. Examining findings, the widest inhibition zone was formed around *S. aureus* followed by *M. smegmatis*, *C. albicans*, *M. luteus* and *S. typhimurium*. The least inhibitory effects were observed for *E. coli*, *K. pneumoniae* and *E. faecalis* (Table 1).

Some investigators noted that sensitivity of microorganisms to chemotherapeutics differs according to type of strain (6,8). Similiar results have been observed in our study.

Antimicrobial characteristics of the herbs are due to various chemical compounds including volatile oils, alkaloids, tannins and lipids that are presented in their tissue (5,7). The inhibitory effect of cardamom seeds detected in the present study may be due to the presence of volatile oils.

In conclusion, our results indicated that extract of the cardamom seed which was prepared using diethyl ether, has a strong inhibitory activity on some pathogens. According to us, using cardamom as antimicrobial additives in food may be useful.

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