

ANALYSIS OF ANTIBACTERIAL AND ANTIFUNGAL ACTIVITY OF CRUDE EXTRACTS FROM SEEDS OF *CORIANDRUM SATIVUM*

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ABSTRACT

Background: Antimicrobial compounds with plant sources have numerous therapeutic potentials; not only are they effective in the treatment of infectious diseases, but also have less chances of side-effects often associated with antimicrobial compounds. This study was conducted to evaluate the antimicrobial activity of coriander seed extract.

Material & Methods: Extraction from seed of plant was carried out through maceration. Then antibacterial and antifungal activity of extract was done with Radial Diffusion Assay method. Minimum inhibitory concentration of this extract was also determined.

Results: The results demonstrated a wide antimicrobial activity of the extract against the microbes studied. Antimicrobial effects of the plant extract showed the greatest effect on *Staphylococcus aureus* resistant strain. Plant extract displayed also effective germicidal effects on *Klebsiella pneumonia*, *Pseudomonas aeruginosa* as well as *Penicillium lilacinum* and *Aspergillus niger*.

Conclusion: Coriander seed extract has effective antimicrobial activity against gram-negative and gram-positive bacteria as well as fungi.

Key Words: Antimicrobial, *Coriandrum sativum*, Crude extract.

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INTRODUCTION

Bacterial and fungal diseases have dramatically affected the health of human societies,¹ hence search on antimicrobial compounds has been an active branch in medical sciences. With increasing antibiotic-resistant microbial strains, the discovery of new antimicrobial compounds has been of double significance.² In this regard, various sources of poisonous secretion of animals such as snake, scorpion, spider, amphibians and insects, have been discovered with diverse effects.³⁻⁶ Herbal plants have a special place in traditional medicine. Antimicrobial herbal compounds are one of the valuable medical resources, and in line with the spread of infectious diseases, identification of more of these ex-

tracts and compounds will be useful in treating patients.^{12,13} Antimicrobial compounds with plant sources have numerous therapeutic potentials; not only are they effective in the treatment of infectious diseases, but also reduce a large number of side-effects that are often associated with antimicrobial compounds.¹⁴⁻¹⁶ Isolation of effective antimicrobial components has been made from various plants, showing a significant impact on a variety of pathogenic bacteria and fungi. Historically, plants have provided sources of antimicrobial compounds to fight infection.⁷

It is estimated that 250,000 to 500,000 plant species exist on Earth, some of which are widely used in medicine.¹¹ In recent years, great attention has been attracted to research on medicinal plants. Various activity-related aspects of plant compounds have been investigated against various bacteria, fungi, viruses and protozoa. Hexan extract of the stem bark of *Amona glabra*,¹⁷ alkaloid extract from dried seeds of *Semecarpus anacardium*,¹⁵ alcoholic

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extract of the stem bark of *Clausena anisata*,¹⁸ aqueous extracts of *Azadirachta indica* leaves¹⁹ and oil extract from the foliage of *Santolina chamaecyparissus* and *Aegle marmelos*²⁰ have shown antimicrobial activity against various bacteria and fungi.

Coriandrum sativum is a flavoring used in foods of China, Mexico, India and South America. The coriander seed consists of completely spherical smooth fruit with straw-yellow color including a crown-like part with five needle-shaped ridges at the top, which is composed of two parts. (Fig. 1) Coriander seed has a traditional use as flavoring, galactagogue, carminative and antiseptic.⁸ The effect of coriander seed has been approved in lipid metabolism and its subsequent reduction.^{9,10}



Fig 1: Picture of *Coriandrum sativum* and its seeds.

The present study was conducted to evaluate the antimicrobial activity of coriander seed extract.

MATERIAL AND METHODS

Methanol, formaldehyde, acetic acid, sodium chloride, sodium hydroxide, ethanol, trichloroacetic acid, acetone, glycerol and chloridric acid were obtained from Merck Company. Trypticase soy broth (TSB) was purchased from High Media Company. All the other chemicals used, including agarose, methyl green, agar, Triton X-100, Coomassie Brilliant Blue R-250 and bromophenol blue were of analytical grades.

Sample extraction was carried out through maceration. For this purpose, 15 g powder of coriander seeds was mixed with 150 ml of distilled water and set aside for 10 hours in the lab temperature. The solution was then heated for 3 hours on medium heat and was filtered afterward. The extract was concentrated to one-tenth of the original volume using the vacuum distillation and was used for evaluating antimicrobial activity.

To investigate the antimicrobial activity, two methods, including radial diffusion and macro

broth dilution, were used. The antimicrobial activity was assessed based on radial growth inhibitions and the minimum inhibitory concentration in radial diffusion and macro broth dilution approaches respectively.

The antimicrobial effects of the extract were investigated using radial diffusion assay (RDA)

One species of gram-positive bacteria (*Staphylococcus aureus* PTCC1431 (*S. aureus*)) and two species of gram-negative bacteria (*Klebsiella pneumonia* (*K. pneumonia*) and *Pseudomonas aeruginosa* (*P. aeruginosa*)) were used for primary assays. An aliquot of bacteria with a titer of 4×10^6 CFU was mixed with 10 ml of medium containing 0.03% TSB and 1% agarose and was poured into a plate. Holes were then created in the medium using a punch, the plant extract was loaded into the wells and the plates were incubated for 3 hours at 37 °C. After the three-hour incubation, the secondary medium, enriched with 6% TSB and 1% agarose, was poured into the plate, and the plates were incubated at 37 °C for 18 hours. The plate was then stained for 24 hours using a solution containing 37% formaldehyde, 15 ml; methanol, 27 ml; water, 63 ml; and Coomassie brilliant blue R-250, 2 mg. The plates were destained for approximately 10 min with an aqueous solution of 10% acetic acid and 2% dimethylsulfoxide.²¹

Antifungal activity of the plant extract was tested on *Penicillium lilacinum* (*P. lilacinum*) and *Aspergillus niger* (*A. niger*). One ml of fungal suspension was inoculated in 20 ml of Potato dextrose agar and was poured into the germ culture plates. The holes were then created by the punch in the medium and were filled by the plant extract. The plates were incubated for 7 days at 30°C to 35°C and the results were recorded during this period.²²

To determine the MIC, a particular method, used for peptides, was applied. Stock serial dilutions of 1 to 32 mg/ml of the extract were prepared and 20µl of the peptide stocks were added to a solution containing 10^6 CFU/ml of bacteria, which was then poured into a plate. The microplate was incubated at 37°C for 18 hours. After this time, the absorbance of each well was read at 630 nm using an enzyme-linked immunosorbent assay (ELISA) reader, and the results were compared to the control samples. The MIC was defined as the peptide concentration at which the absorbance of the treated bacterial sample is half of that of the untreated well of bacteria. *K. pneumonia*, *P. aeruginosa*, and *S. aureus*, were used for MIC determination. Experiments were carried out in triplicate.

To determine the fungi-associated MIC, 180 microliters of Sabouraud Dextrose Agar culture medium along with 10 microliters of fungal suspen-

sion (10^6 CFU/ml) and 10 microliters of serial concentration of the plant extract were poured in microplates which were then incubated at 37 °C for 24 hours. The MIC was similarly defined as minimum concentration at which no growth was observed. *P. lilacinum* and *A. niger* were used for MIC determination. Experiments were carried out in triplicate.²³

RESULTS

The results showed that plant extract obtained from coriander seeds has inhibitory effect on the three bacteria studied. It has also antimicrobial activity against the fungi investigated in this study.

The qualitative assessment results on antibacterial activity of the plant extract are shown in Figure 2. The results are indicative of the effective antibacterial activity of the plant extract against the bacterial strains studied. As clear from qualitative results, *S. aureus* and *P. aeruginosa* have respectively displayed the most and the least sensitivity to the extract. As shown in Figure 2, extract-related diameter of the inhibition zone is more than that of 30 µg of neomycin and kanamycin antibiotics, demonstrating more effective antibacterial activity of the extract.

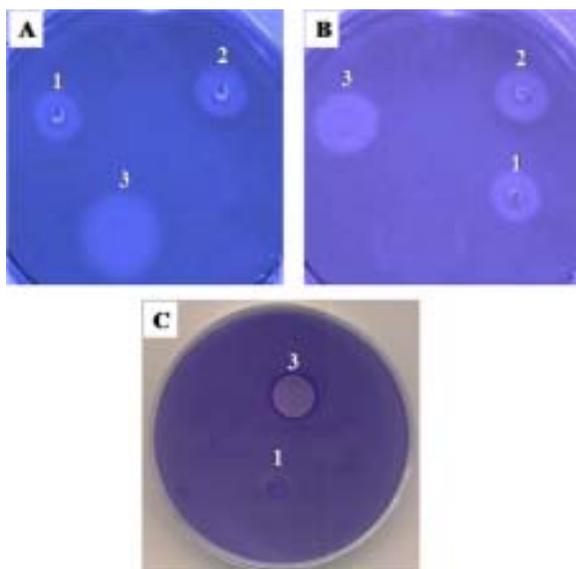


Fig 2: Antimicrobial effects of coriander seed extract against gram-positive and gram-negative bacteria.

A: *Klebsiella pneumoniae*; B: *Pseudomonas aeruginosa*; C: *Staphylococcus aureus*

1: kanamycin; 2: neomycin; 3: plant extract

The plant extract also revealed antifungal activity, as it led to significant fungal destruction in the area around the injected site at concentration of 1 mg/ml. (Fig. 3)

A: *Penicillium lilacinum*; B: *Aspergillus niger*

The MIC was determined for quantifying antimicrobial activity of coriander seed extract on the microbes investigated, and the results are presented in Figure 4 and Table 1. Figure 4 shows the growth curve of five microbes at different concentration of plant extract. Table 1 was acquired from Figure 4. As shown at Figure 4 and Table 1, the results of the quantitative test are well overlapping those of qualitative test, as *S. aureus* and *P. aeruginosa* were introduced as the most and the least sensitive strains in quantitative test.

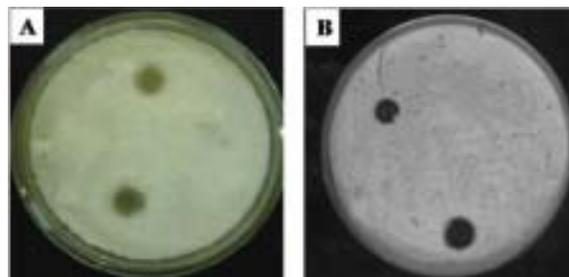


Fig 3: Antimicrobial effects of coriander seed extract against fungi.

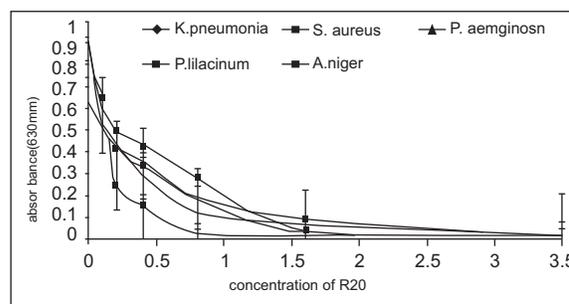


Fig 4: The growth curve of microbes studied in the presence of various concentrations of the plant extract.

Table 1: The MIC values of coriander seed extract

Microbe	MIC (mg/ml)
<i>S. aureus</i>	1.3
<i>K. pneumoniae</i>	2.65
<i>P. aeruginosa</i>	3.2
<i>P. lilacinum</i>	2.5
<i>A. niger</i>	2.3

DISCUSSION

Plant extracts are a complex combination of different chemical elements with different values. Due to significant alteration in these compounds, biological effects of the extracts are varied. Antimi-

crobial properties of some herbal extracts has been identified,²⁴ regarding these properties and other biological impacts, plant extracts have been of great attention as an appropriate substitute for antibiotics for potential therapeutic targets²⁴ and/or health-cosmetics products and food industry.²⁵ Coriander is a pharmaceutical plant with proven therapeutic potential; investigation on antimicrobial effects of the plant essential oils has shown a highly effective antibacterial activity.²⁶ The results of the present study is also demonstrating antimicrobial effects of coriander seed extract, which is in line with other researches confirming medicinal effects of this plant.^{27,28} Besides the antibacterial effects, findings also revealed antifungal activity of the extract, so as it eliminated the *P. lilacinum* and *A. niger* completely effectively at low concentrations. The antifungal activity observed is in consistence with previous studies in this field.²⁹⁻³² The results exhibited that in addition to antimicrobial activity of coriander oils as well as therapeutic potentials of other parts of the plant such as leaves, coriander seed can also be of biological effectiveness, particularly in terms of antibacterial and antifungal activity.⁸ Findings associated to antimicrobial activity of coriander seed extract indicate promising therapeutic properties of plant administration against disease pathogenesis. Antibacterial activity of coriander essential oils has been well established on the *E. coli* strain.²⁶ The present study, in which macro broth dilution and disk diffusion were applied, demonstrated more effective antibacterial property of coriander seed extract against the gram negative and gram positive bacteria strain. Antibacterial activity of the extract against resistant *S. aureus* strain represents extract effectiveness against such a resistant bacteria, for which the MIC was obtained 1.3 mg/ml.

Findings also showed effective antimicrobial activity of the extract on the three bacteria studied in comparison with neomycin and kanamycin antibiotic. In this study, it has been observed that coriander extract has less antimicrobial effect on gram negative bacteria which can be probably due to the presence of cell wall polysaccharides, preventing active compounds from reaching to cytoplasmic membrane of these bacteria.^{33,34} Fungi are more sensitive to plant essences compared to gram negative bacteria. Little information is available on essential oils and their derivatives' mode of action on fungal cells. In general, herbal products contribute to cytoplasm granulation,³⁵ cytoplasmic membranes rupture,^{36,37} and deactivation or inhibition of intracellular and intercellular enzyme activity,²⁵ cell walls disintegration³⁸ and the destruction of electron transport system,³⁹ these cellular events can independently or simultaneously reach to a maximum level while preventing the mycelial growth.^{25,37} Mechanism of antifungal activity of coriander seed extract may be owing to any of the reasons mentioned.

CONCLUSION

Coriander extract has effective antimicrobial activity against gram-negative and gram-positive bacteria as well as fungi.

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CONFLICT OF INTEREST
 Authors declare no conflict of interest.
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