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Antimicrobial and antioxidant properties of *Coriandrum sativum* L. seed essential oil

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Abstract

In the present study the essential oil of *Coriandrum sativum* L. seed was extracted by hydrodistillation and investigated for the antimicrobial activity against clinical pathogens *E. coli*, *Enterobacter aerogenes*, *Klebsiella*, *P. aeruginosa* and *C. albicans* by microdilution method, Minimum bactericidal concentration and antioxidant activity by DPPH assay. Results showed that MIC was minimum 0.02mg/ml and 0.04mg/ml for *C. albicans* and *Klebsiella pneumoniae* and maximum 1.28 mg/ml for *P. aeruginosa*, followed by *E. coli* 0.64 mg/ml. The minimum bactericidal concentration results showed that the MBC value ranges from 0.02mg/ml to 2.56mg/ml. The MBC and or MFC value is low for *C. albicans* 0.04mg/ml and high for *P. aeruginosa* 2.56mg/ml. The essential oil of *Coriandrum sativum* L. seed showed significant antioxidant activity and the percentage of inhibition were 66.2% and 87.8% for standard ascorbic acid. The IC₅₀ value were 0.147 mg/ml and 0.108 mg/ml.

Keywords: *Coriandrum sativum*, essential oil, MIC, MBC, DPPH assay

1. Introduction

The use of traditional medicine is widespread in India [1]. A number of medicinal plants, traditionally used for over 1000 years named rasayana are present in herbal preparations of Indian traditional health care systems [2, 3]. India has been identified as a major resourceful area in the traditional and alternative medicines globally [4]. Beyond this pharmaceutical approach to plants, there is a wide tendency to utilize herbal product to supplement the diet, mainly with the intention of improving the quality of life and preventing the diseases of elderly people [5]. Aromatic plants and spices's essential oils are used in natural therapies, alternative medicine, pharmaceutical drugs and preservation of food [6]. To improve the human health it is inevitable to investigate the physical and chemical properties of essential oil (EO) and its biological properties.

C. sativum L (Coriander) belongs to the family Umbelliferae/ Apiaceae is a annual culinary herb contain rich source of aroma compounds and essential oils with biologically active components unveiling antibacterial, antifungal and antioxidant properties [7-9]. They are valuable in the preparation of food products, perfumes, cosmetics and to prevent food borne diseases and food spoilage as food preservatives too. Coriander has been used for the treatment of cough, bronchitis, dysentery, diarrhea, gout, rheumatism, intermittent fevers and as antiseptic [10-12]. Coriander seed essential oil have been reported to have antibacterial activity against gram positive and gram negative bacteria as well as candida species [13-15]. Thus in the present study essential oil was extracted from coriander seed by hydrodistillation and investigated for the antimicrobial activity by micro broth dilution method and antioxidant activity by DPPH scavenging assay.

2. Materials and methods

2.1 Extraction of Coriander seed Essential oil

Coriandrum sativum dried seeds were procured from local Uzhavvar sandhai market, Perambalur, Tamil Nadu. The plant material was identified with the help of different floras [16-18] and documented properly. The dried seeds were blended in a mixer grinder and powdered. Then about 75gm of the powder material was taken in a one litre flask and 750ml water was added. Hydrodistillation was performed to extract essential oils by cleverger apparatus for about four hours. The essential oil collected were dried with anhydrous sodium sulphate and stored at 4 °C until further use. The percentage of essential oil yield was calculated [19].

2.2 Physicochemical properties

The physicochemical properties of Coriander seed essential oil such as colour, density, refractive index, acid value and saponification value per AOAC standard methods [20].

2.3 Antimicrobial Activity of Essential oil

2.3.1 Minimum Inhibitory concentration

2.3.1.1 Test organisms

Clinical isolates of bacteria were used for the bioassay studies. The isolates included *Escherichia coli*, *Enterobacter aerogenes*, *Pseudomonas aeruginosa* and *Klebsiella pneumoniae*. They were identified and confirmed on the basis of morphological, biochemical and physiological characters according to Bergeys Manual of Systematic Bacteriology [21]. *Candida albicans* was identified by colony and growth characteristics in culture medium SDA, CHROM agar, germ tube test and carbohydrate fermentation test [22-26].

2.3.1.2 Preparation of Inoculum suspension

A single well isolated colony of each bacterial isolates were transferred to 4 ml of trypticase soy broth and incubated at 35 °C for 4 hrs to attain a turbidity of a 0.5McFarland standard. The inoculum suspension was diluted (1:20 dilution) by transferring 2ml of inoculum suspension to 38ml of sterile saline to get a concentration of 5×10^5 CFU/ml.

2.3.1.3 Preparation of essential oil

Antimicrobial activity of essential oil was determined by broth microdilution method in a 96 well microplate. Essential oils were dissolved in 5% DMSO to a stock concentration of 10.24 mg/ml and filter sterilized. The essential oil was diluted by serial two fold dilution in sterile Muller Hinton broth medium 5.12, 2.56, 1.28, 0.64, 0.32, 0.16, 0.08, 0.04, 0.02, 0.01mg/ml.

2.3.1.4 MIC Assay

Different concentration of *Coriandrum sativum* seed essential oil ranges from 5.12mg/ml to 0.01mg/ml in Muller Hinton broth was added to ten microwell. A positive control containing Muller Hinton broth without essential oil and a negative control containing uninoculated Muller Hinton broth devoid of essential oil were used in this assay. To each microwell except negative control 0.01ml of inoculum was added and incubated 18- 24 hrs. MIC of *C. sativum* seed essential oil is the lowest concentration at which no visible growth of bacteria by naked eye in the microwell.

2.3.2 Minimum Bactericidal and fungicidal Concentration

Inoculum preparation, concentration of essential oil and assay were performed as described for MIC. MBC was determined by subculture of 20 µl, from each microwell which didn't show visible growth to a sterile plate count agar plate and incubated at 18- 24 hrs. The plates were observed for absence of visible growth. MBC is defined as the minimum concentration of the essential oil where 99.9% or more of the initial inoculum was killed by showing no growth after inoculation on the agar medium after incubation and the MIC, MFC and MBC values were examined.

2.4 In vitro antioxidant DPPH scavenging assay

The antioxidant activity of *C. sativum* L. seed essential oil was determined by the DPPH assay. The DPPH radical scavenging activity was estimated for five different concentrations (0.025, 0.05, 0.100, 0.150 and 0.200 mg/ml) of essential oil and standard ascorbic acid by veerapagu *et al.* [27]

(2018). The antioxidant activity was expressed as inhibition percentage (I%) of DPPH radical by following.

$$\% \text{ scavenged [DPPH]} = \frac{[(Ac-As)/Ac]}{1} \times 100$$

where Ac was the absorbance of the control, and As was the absorbance of sample or standards.

3. Results and discussions

In the present study *Coriandrum sativum* seed essential oil was extracted by hydrodistillation method and the yield of *C. sativum* seed essential oil was 1.2% (Table 1). Essential oil from coriander seed was $1.1 \pm 0.1\%$ [28]. Essential oil yield percentage may vary from 0.03 -2.6%. [29-30].

3.1 Physicochemical properties of *C. sativum* seed oil

The results of the physicochemical properties of CSEO were represented in table 1. The colour, density, refractive index, acid value and saponification values were found to be pale yellow, 0.84, 1.427, 10.56 and 184.3. The density and refractive index were important in the evaluation of purity of oils. Considerable variations are reported in the physicochemical characteristics of coriander seed essential oils since factors such as type and origin of cultivar, seed maturity at harvest, storage conditions and method of extraction can influence such properties [31, 32].

3.2 Antimicrobial activity

3.2.1 Minimum Inhibitory concentration

Antimicrobial activity of *Coriandrum sativum* seed essential oil was evaluated against clinical pathogens. The bacteria and yeast were identified based on morphological, cultural and biochemical characteristics. The pathogens were identified as confirmed to be *E. coli*, *Enterobacter aerogenes*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa* and *Candida albicans* and the results are presented in the table 2. The MIC values of CSEO are represented in table 3. The results showed that MIC was maximum 1.28 mg/ml for *P. aeruginosa*, followed by *E. coli* 0.64 mg/ml and it was minimum 0.02mg/ml and 0.04mg/ml for *C. albicans* and *Klebsiella pneumoniae* (Table 3). Similar results was reported that *P. aeruginosa* was the most resistant to growth inhibition showing highest MIC [33]. MIC values of *C. sativum* essential oil ranges from <0.195 to 1.562 µg/mL and showed strong antimicrobial activity against *B. subtilis*, *C. albicans*, *E. faecalis*, *E. faecium*, *K. pneumonia*, *L. innocua*, *P. aeruginosa*, *S. enteritidis*, *S. infantis*, *S. kentucky*, and *S. typhimurium* [34]. Presence of antibacterial substances in the higher plant is well established [35]. Plants have provided a source of inspiration for novel drug compounds as plants derived medicines have made significant contribution towards human health. Phytochemistry can be used for the treatment of diseases as is done in case of Unani and Ayurvedic system of medicines or it can be the base for the development of a medicine, a natural blueprint for the development of a drug [36].

3.2.2 Minimum Bactericidal and fungicidal concentration

The results of the *C. sativum* essential oil MBC values are as in table 3. The MBC value ranges between 0.02mg/ml to 2.56mg/ml. The MBC value is low for *C. albicans* 0.02mg/ml and high for *P. aeruginosa* 2.56mg/ml. The MBC/MIC and MFC/MIC ratio were 2,1,4,2 and 2 for *E. coli*, *E. aerogenes*, *Klebsiella pneumoniae*, *P. aeruginosa* and *C. albicans*. The bactericidal activity of coriander essential oil was higher in gram negative bacteria *E.coli*, *Pseudomonas* and *Salmonella*

typhimurium than gram positive [33]. Similarly reported that MBC value of coriander was 62.5mg/ml for MRSA [37]. Essential oils are highly rich in lipophilic compounds that could dissolve in the biomembrane of microorganisms and interact with membrane lipids and proteins; this can result in cell disruption, leakage of cell content and finally cell death [38, 39]. This may explain the antimicrobial activity of coriander.

3.3 antioxidant activity DPPH assay

Antioxidants inhibit oxidation of food also quench dreaded free radicals produced due to environmental and physiological stress which leads to aging, atherosclerosis and cancer [40]. Antioxidation and oxidation processes in plants are complex and therefore it is difficult to measure each antioxidant component separately. DPPH free radical has been used to determine the radical scavenging activity of antioxidants [41, 42]. In the present study the essential oil of *Coriandrum sativum* L. seed showed significant antioxidant activity. The percentage of seed essential oil was 66.2% and 87.8% for standard ascorbic acid (Fig1). The IC₅₀ value were 0.147 mg/ml and 0.108 mg/ml. Our results agreed with the results reported by shawar *et al.* [28] that the essential oil from coriander seeds showed significant radical scavenging activity (66.48 ± 0.80%) at a concentration of 500 µg/ml and might be due to the presence of high concentrations of linalool in seed

essential oil. Hassanen *et al.* [43] found that DPPH scavenging activity (%) of coriander seed essential oil was 87.5%. Similar result of DPPH scavenging activity percentage of inhibition of 30.1% for coriander seed essential at a concentration of 0.125mg/ml was reported by Ahmed *et al.* [32].

Table 1: Physicochemical properties of *C. sativum* essential oil

| Properties | Result |
|------------------------------------|-------------|
| Yield% (g/100g) | 1.2 |
| Colour | Pale yellow |
| Density (g/cm ³ , 25°C) | 0.84 |
| Refractive index | 1.427 |
| Acid value | 10.56 |
| Saponification value | 184.3 |

4. Conclusion

In the present study essential oil of *Coriandrum sativum* seed extracted by hydrodistillation was evaluated for antimicrobial activity and antioxidant activity. The essential oil exhibited high activity against *Candida albicans* and *Klebsiella pneumoniae* and significant antioxidant activity. It may concluded that the *Coriandrum sativum* seed essential oil could be a potential source of natural antimicrobial agents to combat clinical human pathogens and as antioxidants it may be a suitable food additives to prevent oxidative stress related degenerative diseases.

Table 2: Identification of bacteria and yeast

| S. No | Identification of bacteria | | | | | Identification of yeast | |
|-------|----------------------------|--------|--------|------------|--------|---------------------------|------------------------------|
| | Test | Result | | | | Test | Result |
| | | A | B | C | D | | |
| 1 | Gram stain | -ve | -ve | -ve | -ve | Gram stain | Positive |
| 2 | Shape | Rods | Rods | Rods | Rods | Shape | Budding yeast |
| 3 | Motility | Motile | Motile | Non-motile | Motile | Sabouraud's dextrose agar | smooth creamy white colonies |
| 4 | Indole | + | - | - | - | Corn meal agar | terminal chlamydo spores |
| 5 | MR | + | - | - | - | Hichrome candida agar | glistening green colony |
| 6 | VP | - | + | + | - | Germ tube test | Presence of germ tube |
| 7 | Citrate | - | + | + | + | Dextrose | + ve |
| 8 | Oxidase | - | - | - | + | Maltose | + ve |
| 9 | Catalase | + | + | + | + | Sucrose | - ve |
| 10 | Lactose fermentation | + | + | + | - | Lactose | - ve |

A. *Escherichia coli*, B. *Enteobacter aerogenes* C. *Klebsiella pneumoniae*, D. *Pseudomonas aeruginosa*, E. *Candida albicans*

Table 3: Minimum inhibitory concentration and minimum bactericidal and fungicidal activity of CSEO

| | A | B | C pneumoniae | D | E |
|-----------------------------|------|------|--------------|------|------|
| MIC (mg/ml) | 0.64 | 0.16 | 0.04 | 1.28 | 0.02 |
| MBC (mg/ml), MFC (mg/ml), (| 1.28 | 0.16 | 0.16 | 2.56 | 0.04 |
| MBC/MIC and MFC/MIC | 2 | 1 | 4 | 2 | 2 |

A. *Escherichia coli*, B. *Enteobacter aerogenes* C. *Klebsiella pneumoniae*, D. *Pseudomonas aeruginosa*, E. *Candida albicans*

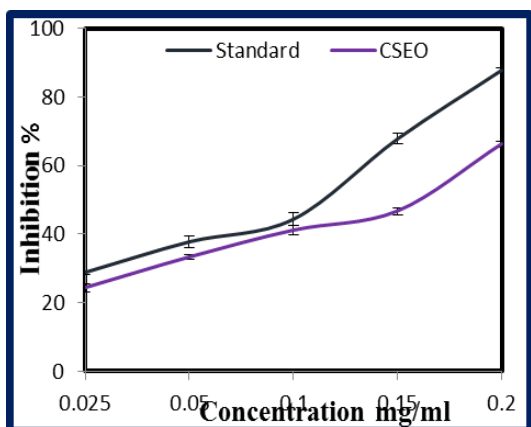


Fig 1: Antioxidant activity of CSEO

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