

# A Research Proposal of Antibacterial Activity of Some Volatile Oil Bearing Plants



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**Submission:** May 26, 2021; **Published:** June 28, 2021

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## Abstract

The antibacterial activity of essential oil of two Ethiopia plants will be study. The essential oil produced from different parts of plants (coriander and fennel plants) will by measuring MIC (minimum inhibitory concentration), whereas some bacterial strains are used in this study such as follows: *Staphylococcus aureus*, and *klebisela*. The two samples used in this experiment have antibacterial activity against Gram-positive and Gram-negative bacteria. Extract by different solvents and then examined by GCMS preliminary phytochemical screening. The antibacterial activities of the essential oils will determine.

**Keywords:** Extraction; Essential oil; Antibacterial

**Abbreviations:** MIC: Minimum Inhibitory Concentration; SFE: Steam Distillation and Supercritical Fluid Extraction

## Introduction

Medicinal plants contain certain active components, which have been exploited in traditional medical practice for the treatment of various illnesses over the years. Plants of both lower and higher categories are examined to produce active chemical substances with which they defend themselves against foreign agents such as invading microorganisms [1] Vegetable is an edible component of plants or its part, intended for many purposes of which includes medicinal purposes, commercial purposes [2]. Coriander (*Coriandrum sativum L.*) is a well-known aromatic/ medicinal plant, which grows in Mediterranean countries, and possesses a lot of pharmacological activities. In traditional medicine, seeds are used in the treatment of gastrointestinal problems, rheumatism and joint pains. It has been reported that they exhibit a hypoglycaemic action and influence on carbohydrate metabolism, whereas the antimicrobial effect exhibit both leaves and seeds. Besides, leaves and seeds are used to flavour various commercial foods, as liqueurs, teas, meat products and pickles [3]. Isolation of coriander essential oil using different extraction methods (steam distillation and supercritical fluid extraction – SFE) and determination of essential oil compounds (using GC-MS method, in the first place) were investigated in [3,4].

Fennel (*Foeniculum vulgare Mill*) is an annual herbaceous plant belongs to the family Apiaceae. It is widely cultivated throughout the temperate and tropical regions of the world. Fennel herb is being used as a medicinal and economic plant in Asian countries. Survey of published literature revealed that fennel herb and seeds effectively control numerous infectious disorders of bacterial, fungal, viral, mycobacterium, and protozoal origin as well as used antispasmodic, diuretic, anti-inflammatory, analgesic, secretomotor, secretolytic, galactagogue, eye lotion, and antioxidant remedy in Europe and Mediterranean areas [5,6]. The different compounds in the oil were found to act in synergistic manner (i.e. more than the mere summation of the actions of the individual compounds). This stresses the importance of using the whole oil (or the whole crude extract) of the seeds in different pharmacological and clinical studies.

## Problem Statement

The information on antimicrobial activity of given medicinal plants is very poor. Resistance to drugs by microorganisms has increased. This resistance has been attributed to overdose and under dose of drugs due to over counter prescription of drugs, ability of microorganisms to undergo genetic variability

(mutation), use of antibiotics in food preservation and general misuse of drugs [7]. Hence there is need to come up with sensitive and effective drugs specially using medicinal plants. Therefore, there is need to carry out proper identification of the medicinal plants, their antibacterial activity and know their phytochemical composition to use medicinal plants for treatment purpose.

### Aim of study

The main aim of this study is to investigate some volatile oils of plant extracts and to test on selected microorganisms.

#### Objectives

- Selection, authentication and extraction of volatile oil-bearing plants
- To identify the phytochemical constituents of medicinally active plants.
- To administer and determine the antibacterial activity of these medicinal plants.

### Justification of the study

The synergic effect of coriander and Fennel on bacteria is not well investigated before and there is need for further research to be done for all traditional medicinal plants to establish their uses and the active phytochemicals with medicinal properties. The information will help bridge the gap between modern medicine and traditional medicine making it possible to compliment the modern medicine.

### Literature Review

Medicinal and aromatic plants constitute a large part of natural flora and are considered an important resource in various fields such as the pharmaceutical, flavor and fragrance, perfumery, and cosmetic industries [8]. At present, more than 80% of the global population depends on traditional plant-based medications for treating various human health problems [9,10]. Coriander (*Coriandrum sativum L.*) is a well-known aromatic/medicinal plant, which grows in Mediterranean countries, and possesses a lot of pharmacological activities. All parts of the plant are edible but the fresh leaf and the dried seeds are the most common parts used in cooking. In the Indian traditional medicine, *C. sativum* is used in treating the disorders of digestive, respiratory and urinary systems, as it has diaphoretic, diuretic, carminative and stimulant activity. In Iranian traditional medicine, *C. sativum* has been indicated for several medical problems such as dyspeptic complaints, loss of appetite, convulsion, anxiety and insomnia [11]. The basic *C. sativum* aroma produced by a combination of the major components, 1, 8-cineole and  $\alpha$ -terpinyl acetate. *C. sativum* oil is used in food, perfumery, flavor and carminative. In medicine, it is used as a powerful aromatic, antiseptic, stimulant, carminative, stomachic, expectorant, antispasmodic and diuretic [12]. The physiochemical properties of *C. sativum* essential oil such as density (25°C), acid value, ester value, and optical rotation (25°C) were found to be 0.8310, 1.4592, 4.0, 23.7 and +11.5 g/cm<sup>3</sup> respectively [13].

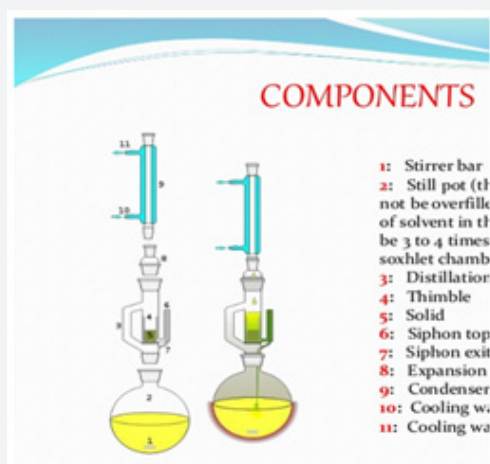


Figure 1: Component of Soxhlet extractor method.

Fennel (*Foeniculum vulgare Miller*) is a major aromatic plant belonging to the Apiaceae family, which has long been considered as a medicinal and spice herb. Fennel oil is commonly used as a favoring agent in food products and as a constituent of cosmetic and pharmaceutical products; furthermore, its antimicrobial properties against a wide range of microorganisms have been

well established [14,15]. It has long been known that some plant essential oils could affect the expression of staphylococcal exotoxins [16]. Fennel seeds essential oil is used as flavouring agents in food products such as liqueurs, bread, cheese, pickles and pastries [17] and an ingredient of cosmetics and pharmaceutical products [18]. Fennel (*Foeniculum vulgare Mill*) is an annual

herbaceous plant belongs to the family Apiaceae. It is widely cultivated throughout the temperate and tropical regions of the world. Fennel herb is being used as a medicinal and economic plant in Asian countries. Survey of published literature revealed that fennel herb and seeds effectively control numerous infectious disorders of bacterial, fungal, viral, mycobacterium, and protozoal origin as well as used antispasmodic, diuretic, anti-inflammatory, analgesic, secretomotor, secretolytic, galactagogue, eye lotion, and antioxidant remedy in Europe and Mediterranean areas [19]. Fennel seeds essential oil is used as flavouring agents in food products such as liqueurs, bread, cheese, pickles and pastries [20] and an ingredient of cosmetics and pharmaceutical products [21].

### Chemical constituent

The volatile oil of *C. sativum* contains about  $\alpha$ -pinene (1.5%),  $\beta$ -pinene (0.2%), sabinene (2.8%), myrcene (1.6%),  $\alpha$ -phellandrene (0.2%), limonene (11.6%), 1,8-cineole (36.3%),  $\gamma$ -terpinene (0.7%), terpinolene (0.5%), linalool (3%), linalyl

acetate (2.5%), terpinene (0.9%),  $\alpha$ -terpineol (2.6%),  $\alpha$ -terpinyl acetate (31.3%), citronellol (0.3%), nerol (0.5%), geraniol (0.5%), methyl eugenol (0.2%) and trans-nerolidol (2.7%) [22]. The major constituents of the essential oil from the fennel were found estragole (61.1%), fenchone (23.5%), limonene+  $\beta$ -phellandrene + 1, 8-cineole (8.7%) and  $\alpha$ pinene (1.2%) [23,24].

### Taxonomic position according to Cronquist (1981)

**Kingdom:** Plantae (Figure 2)

**Division:** Tracheophyta

**Class:** Magnoliopsida

**Order:** Apiales

**Family:** Apiaceae

**Genus:** Coriandrum

**Species:** *C. sativum*



Scientific name: *Coriandrum sativum* L.

Bangla/local name: 'Dhonia'.

**Figure 2:** Botanical description of plant (Coriander).

### Botanical description of coriander

- a) **Habit:** Coriander is an erect, glabrous, annual herb
- b) **Root:** Coriander has a well-developed fleshy taproot
- c) **Stem:** The species has solid, finely grooved stems and is richly branched
- d) **Leaf:** The leaves alternate and are compound and most of them are pinnate or bipinnate
- e) **Fruit:** The small oval and aromatic fruit is a cremocarp, yellowish brown, 3 to 5 mm in di.

### Taxonomic position according to Cronquist (1981)

**Kingdom:** Plantae (Figure 3)

**Division:** Magnoliophyta

**Class:** Magnoliopsida

**Order:** Apiales

**Family:** Apiaceae

**Genus:** *Foeniculum*

**Species:** *F. vulgare*

### Botanical description of Fennel

- a) **Habit:** Fennel is a biennial or perennial herb with erect stems, when older with hollow internodes.
- b) **Root:** The plant has an elongated and enlarged tap root. The tap root is yellowish to light brown up to 2.5 cm in diameter, which is suitable for soils that can become deeply penetrating, with few larger laterals but many side rootlets.
- c) **Stem:** There is normally one, erect, stout, main stem up to 1.5 m occasionally to 2 m, with the base 2-3 cm diameter, usually profusely branched on the higher portion

**d) Fruit:** The fruit, a schizocarp, is elliptical, yellowish brown, 4-8 mm in length and consists of 2 curved mericarps. The seeds are hairy and yellow brown in color.

#### Economic importance

Used as spices

Also used as medicinal purpose



English name: Fennel  
Scientific name: *Foeniculum vulgare* Mill.  
**Figure 3:** Fennel.

## Materials and methods

### Plant material

Coriander and fennel seeds will be obtained at local supermarkets and purchase from local herbal shop and were used without further purification.

### Method of extracts

#### Isolation of essential oil from coriander using soxhlet

30g of the cilantro seeds will extracted using 130-150mL of the solvents at 50-60°C for 3-4 cycles in a Soxhlet extractor and the crude extract will be concentrated by allowing them for evaporation. All the filtered extracts will store at -20°C in an airtight container for further analysis [25].

$$\text{Essential Oil yield (\%)} = \frac{\text{Volume of essential oil (g)}}{\text{Weight of plant sample (g)}} \times 100$$

#### Isolation of essential oil from fennel using soxhlet

100g of dried *Foeniculum vulgare* seeds will milled to form a course powder and then extracted with methanol (500ml). The aqueous extract will prepared by boiling 10g of dried ground seed in 20ml distilled water for 2h.

$$\text{Oil content (\%)} = \frac{\text{Volume of the oil}}{\text{Weight of sample}} \times 100$$

### Procedure Soxhlet Extraction

**a.** Normally a solid material containing some of the desired compound is placed inside a thimble made from thick filter paper, which is loaded into the main chamber of the Soxhlet extractor. The Soxhlet extractor is placed onto a flask containing the

extraction solvent. The Soxhlet is then equipped with a condenser.

**b.** The solvent is heated to reflux. The solvent vapour travels up a distillation arm and floods into the chamber housing the thimble of solid. The condenser ensures that any solvent vapour cools, and drips back down into the chamber housing the solid material.

**c.** The chamber containing the solid material slowly fills with warm solvent. Some of the desired compound will then dissolve in the warm solvent. When the Soxhlet chamber is almost full, the chamber

**d.** is automatically emptied by a siphon side arm, with the solvent running back down to the distillation flask. This cycle may be allowed to repeat many times, over hours or days.

**e.** 4. During each cycle, a portion of the non-volatile compound dissolves in the solvent. After many cycles the desired compound is concentrated in the distillation flask. The advantage of this system is that instead of many portions of warm solvent being passed through the sample, just one batch of solvent is recycled.

**f.** 5. After extraction the solvent is removed, typically by means of a rotary evaporator, yielding the extracted compound. The non-soluble portion of the extracted solid remains in the thimble and is usually discarded.

### Antimicrobial Activity

These techniques will used to test the anti-microbial activity: the paper disc diffusion method. The minimum inhibitory concentration (MIC) will determined by the later method.



### Papers-disc diffusion method

Paper discs (6mm in diameter) saturated with a 40mg/mL solution of plant extract were applied to the surface of agar plates that were previously seeded by spreading 0.1mL of culture overnight. The plates were incubated overnight at the appropriate temperature, and the diameter of the resulting zone of inhibition will be measured in millimeters.

### Phytochemical Screening.

#### Detection of alkaloids

**Mayer's test:** Filtrates were treated with Mayer's reagent (Potassium Mercuric Iodide). Formation of a yellow coloured precipitate indicates the presence of alkaloids. Detection of phenols.

**Ferric chloride test:** Extracts were treated with 3-4 drops of ferric chloride solution. Formation of bluish black colour indicates the presence of phenols.

#### Detection of glycoside

**Legal's test:** Extracts were treated with sodium nitropruside in pyridine and sodium hydroxide. Formation of pink to blood red colour indicates the presence of cardiac glycosides.

#### Detection of flavonoids

**Alkaline reagent test:** Extracts were treated with few drops of sodium hydroxide solution. Formation of intense yellow color, which becomes colorless on addition of dilute acid, indicates the presence of flavonoids [26-28].

#### Detection of tannins

**Gelatin test:** To the extract, 1% gelatin solution containing sodium chloride was added. Formation of white precipitate indicates the presence of tannins.

#### Detection of saponins

**Froth test:** Extracts were diluted with distilled water to 20ml, and this was shaken in a graduated cylinder for 15minutes. Formation of 1cm layer of foam indicates the presence of saponins.

### Expected Results and Outputs

The expected results will be to identify the synergic effect of coriander and fennel in selected bacterial to be used for medicinal purpose. The medicinal essential oil coriander and fennel will be identified and administered in selected bacterial to establish its chemical properties by doing lab research. The information from this study will help to understand the medicinal use of coriander and fennel will provide basic information for further study and also bridge the information gap on medicinal plant.

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DOI: [10.19080/GJPPS.2021.08.555744](https://doi.org/10.19080/GJPPS.2021.08.555744)

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