Antibacterial effect of *Salvia officinalis* Lam extract

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**ABSTRACT**

Introduction: bacterial diseases are of common diseases in human life and treatment of the diseases by means of antibiotics creates some other problems like drug resistance and side effects. So, using new herbal drugs with fewer side effects can help worthy in treatment of this kind of infections. The effect of *Salvia officinalis* Lam extract on four species of gram positive and gram negative i.e. *Bacillus cereus*, *Bacillus anthracis*, *E.coli*, *Shigella sonnei*. Extracting from herbs by distillation of pool water and by sink method was studied and chemical compound of the extract was analyzed by Gas Chromatography (GC) and gas chromatography connected to mass spectrometer. By calculating suppressive indices it was clear that there are 10 various compounds in the extract and the most important of them are: globulin (9.3%), α-humulene (8.4%), borneol (9.4%), α-pinen1, 8 (5.5%) and cineol (18.9%). Efficient volatile oil against the mentioned bacterial suspensions was identified with 10⁸ microorganisms per ml in order to study the rate of its antibacterial property. The antibacterial activity of *Salvia officinalis* Lam was observed as non-growth haloes after 24-28 hours. The haloes diameters were obtained as 40, 25, 24, and 20 mm for *Bacillus cereus*, *Bacillus anthracis*, *Shigella sonnei*, *E.coli*.

**Key words:** Salvia, *Bacillus cereus*, *Bacillus anthracis*, *E.coli*, *Shigella sonnei*.

**INTRODUCTION**

The main part of infectious diseases in medicine area is attributed to their prevalence, and the kind of their contagious in relation with public hygiene. Although there are exceptions, infectious diseases can be prevented and cured easier than other disorders (1, 2). *Salvia* is one of the herbs that belong to mint family and 700 to 800 species of the herb grow across the world. The herb has 58 grassy species in Iran which spread across the country and 17 species of them are confined to Iran (3, 6, 8). Ancient physicians like Casin and contemporary physicians like Dr.H.Leclere expressed it as an efficient herb in removing excessive weakness with neural reason, removing neural weakness and general fatigue, neural vertigos and body frisson. The herb has anti-tumor and anti-inflammation property and is used vastly in cosmetic and perfumery industries. So, the herb is used for cold, bronchitis, digestive disorders and tuberculosis (2,3). *Salvia* extract has antibacterial property due to 1,8-cineol (4).
MATERIALS AND METHODS

Gathering, drying, and extraction
Salvia officinalis Lam were gathered from woody region of Arasbaran of Ahar town and dried in a shady place in the laboratory. The dried herbs were powdered by an electrical mill and extracted by distillation by means of klonjer machine. The prepared powders were transferred to microbiological laboratory for microbial examinations and analyzed with GC and GC/MS systems.

Analyzing with gas chromatography system
Gas chromatography system, model 9A equipped with a bar of DB-5 with the height of 30m, internal diameter of 0.25 mm and layer thickness of 0.25 mm, made by Shimodzo Co.was used in the present study. The oven temperature was kept at 40°C for 5 min. then the temperature was increased to 250°C with the rate of 4°C/m. the temperature of injection part was 260°C and gas containing helium was used at the rate of 32 cm/s.

Analyzing with gas chromatography system connected to mass spectrometry
The used system was Varian 3400 equipped with a bar of DB-5, with the length of 30 m, internal diameter of 0.25 mm and thin layer thickness of 0.25 µ. Its temperature was increased from 60 to 250°C with the rate of 4°C/min. gas containing helium with the rate of 1.1 mm/min was used as well as 70 electron volt ionization energy. Spectra identification were done by means of suppression indices by injection of normal hydrocarbons under identical conditions by extracts injection and computer program (Basic) and their comparison was conducted by obtained rates (5). The comparisons and calculations were confirmed by means of mass spectra of standard compounds as well as using termonilogies available in trepnoids laboratory of system's computer. Quantitative calculation was conducted by means of Euro chrom 2000 data processor and Area normalization and Response factors related to spectra.

Understudied bacteria
Microbial strains are Bacillus cereus (PTCC 1015), Bacillus anthracis (PTCC 1010), E.coli (PTCC 1037), Shigella sonnei (PTCC 1035). Microbial cultural media are: molohinton, notermit agar (merck of Germany). Sink method was used for studying antimicrobial effects. The concentration of microbial suspension was identified as $1.5 \times 10^8$ cfu/ml using tube No. 0.5 of Mc farland. After culturing, considered microbe was slide cultured in plates with the diameter of 8 cm diameter contained noler heniton agar culturing media. Some holes were created by sterile Pastor Pipette. The holes have suitable distance from each other and the edge of plate. Then 50 ml of extract was purred in to holes by sterile sampler. Then the plates were placed in 37°C incubators; after 24-28 hours the non-growth halos were measured with scaled ruler (the experiment was reviewed five times).

RESULTS

In order to identify chemical compound and evaluating antibacterial activity, the volatile oil of Salvia officinalis Lam essence was extracted by means of water distillation and its antimicrobial activity on Bacillus cereus, Bacillus anthracis, E.coli, Shigella sonnei was studied. Antimicrobial effect of the herb's essence was demonstrated by hole method against four microorganisms. In the present study Salvia officinalis Lam essence had the most influence on Bacillus cereus in fact positive gram bacteria, Bacillus cereus, has greater susceptibility compared with three other bacteria. Bacillus anthracis, E.coli, Shigella sonnei also demonstrate susceptibility to Salvia officinalis Lam essence. This kind of susceptibility which is observed as non-growth halos has
been measured as 40, 25, 20, and 20 mm in Bacillus cereus, Bacillus anthracis, E.coli, Shigella sonnei, respectively. Based on obtained results the susceptibility of positive gram bacteria is more than negative gram bacteria (Figure 1). Analyzing and identifying the essence compounds of Salvia officinalis Lam show that the most important of them are: globulin (9.3%), α-humulene (5%), borneol (9.4%), α-pinen1, 8 (5.5%) and cineol (18.9%), β-pienene (16%), Camphene (5%), 1,8 cineal (18.9%), α-thujone (16.4%).

![Figure 1: Antibacterial effect of salvia officinalis extract](image)

**DISCUSSION**

There are many reports about the extract of various species of salvia most of which are presented by Iranian and foreign researchers. Some of them will be mentioned for comparing. In a study by Habibi et al. monool (37.3%) was identified in S.persepolitoma extract and Teripinolene (27%) and limonene (14.9%) in S.rhytiden extract. The study conducted by Balchin et al. demonstrated that 1,8-cineale has not antimicrobial effect. They confirmed that herbs containing α-pienene have antimicrobial property (7). Adams et al. evaluated the antibacterial activity of salvia cryptantha and salvia muticoulis essence and methanol extract by disk plate and hole spread on Bacillus cereus and E.coli. they observed that the essence and methanol extract of this species demonstrated positive antibacterial effect. Availability of some compounds like α-piene, camphor, 1,8-cineole, borneol in these two species as well as salvia officinalis shows that antibacterial effect can be attributed to these compounds. 1,8 cineole and camphor are well known compounds which have antimicrobial effect (10). Based on Pattnaik et al. α-piene and borneole have an slow activity against microorganisms (9). Antibacterial effects of borneol was reported in another study; so, based on these findings the antimicrobial activity of salvia cryptantha and salvia muticoulis can be attributed to camphor and 1,8 cineole and derivatives of borneale. In the present study it was identified that the essence of salvia contains 1,8 cineole (18.9%), α-pinen (5.5%), β-pinene (96%), camphor (2.9%), borneol (9.4%) and camphen (5%)

**REFERENCES**