

Preliminary Screening of Antibacterial and Phytochemical Studies of *Ocimum americanum* Linn.

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Abstract

The aim of the present research was focused on the antibacterial and preliminary phytochemical properties of *Ocimum americanum* Linn. (Lamiaceae) via *in-vitro* approach. Antibacterial activity was tested against, Gram +ve and Gram -ve organisms. Maximum activity was exhibited by alcoholic extract against *Staphylococcus aureus* followed by *Bacillus subtilis* and *Pseudomonas aeruginosa*. Chloroform and Petroleum ether leaves extract exhibited less activity. The different solvent extracts showed the presence of secondary metabolites like alkaloids, phenolic compounds, tannins, lignin, starch, saponins, flavanoids, terpenoid and anthraquinone.

1. Introduction

Ocimum americanum (Lamiaceae) is annual plant native to the African continent and grows to a height of 2 feet. It is also known as the African basil with a distinct mint flavor, with hairy leaves and scented flowers. Two types of oil are distilled from this plant one contain methyl cinnamate as principal constituent the other 2- camphor. The will yield from herb varieties' from 0.46 to 0.65 %. Oil used in perfumes and cosmetics. It its natural source of camphor. Seeds of this plant are tonic and diuretic. The will from its leaves has the property of destroying bacteria and insects. The development of drug resistance in human pathogens against commonly used antibiotics has necessitate the search for new antimicrobial substance from other sources. Screening of *O. americanum* for antimicrobial activities and phytochemical is important for finding potential new compounds for therapeutic uses. This study reports the antibacterial activity of different solvent extracts using some more strains.

2. Materials and Methods

Sample collection and authentication

The fresh, mature healthy leaves of *Ocimum americanum* (Lamiaceae) were collected from west land of Dhule away from pollution. The plant materials were identified using the Flora of Dhule and Nadurbar District [1] at Post graduate Department of Botany, S.S.V.P. Sansthas, L.K.Dr.P.R.Ghogrey Science College, Deopur, Dhule-424005 (M.S.) India.

Sample preparation

Fully grown leaves of *O. americanum* were weighed (1kg). The plant samples were shade dried

ground and sieved with 2mm iron sieve to form uniform powder and stored in airtight bottles.

Preparation of extracts and phytochemical screening

The dried plant material was pulverized into fine powder using a grinder (mixer). About 50 gm of powdered material was extracted in soxhlet extraction apparatus with 250 ml of each of the following solvents; Petroleum ether, chloroform, and Alcohol [2]. The extracts obtained with each solvent were filtered through Whatman filter paper No. 1 and the respected solvents were evaporated (at 40°C) with the help of heating mantle. The sticky greenish-brown substances were obtained and stored in refrigerator and were suspended in dimethyl sulphoxide (DMSO) for prior to use [3]. Some of the extracts were used for the qualitative phytochemical screening for the identification of the various classes of active chemical constituents, using standard prescribed methods [4-6]. The tests were noted as Present (+) and absent (-).

Tested microorganisms

Various cultures of human pathogenic, gram positive and gram negative bacteria were used. These are *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Bacillus subtilis*. The cultures were obtained from Department of Microbiology, Government Institute of Science, Aurabgabad, (M.S.) India. The microorganisms were repeatedly subcultured in order to obtain pure isolates. A loop full test organism was inoculated on nutrient broth and incubated for 24 h at 37±1°C and maintained in sterile condition.

Screening for antibacterial properties

Antibacterial activities of plant extracts were tested by Agar well diffusion method [7]. The culture plates were prepared by pouring 20 ml of sterile nutrient agar. 1 ml inoculum suspension was spread uniformly over the agar medium using sterile glass rod to get uniform distribution of bacteria. A sterile cork borer (8 mm) was used to make wells in each plate for extracts. These plates were labeled and 100 µl of each plant extracts (at concentration of 50, 100 mg/ml) was added aseptically into the well. Then the plates were incubated for 24 h at 37°C during which the activity was evidenced by the presence of zone of inhibition surrounding the well. Each test was repeated three times and the antibacterial activity was expressed as the mean of diameter of the inhibition zones (mm) produced by the plant extracts when compared to the controls.

3. Result and Discussion

The results obtained for the antibacterial tests performed on different solvent extracts of *Ocimum americanum* are presented (Table 1). Among the extracts tested, alcohol extracts showed broader

spectrum of activity, being active to both Gram-positive and Gram-negative organisms compared to chloroform and petroleum. The alcohol extract at 100mg/ml for example, 23 mm was recorded as diameter zone of inhibition highest activity against *S. aureus*. This was followed by 16 mm *B. subtilis*, 12 mm *P. aeruginosa*, and *E. coli*, 10 mm respectively. The least activity 5 mm against *B. subtilis*, *P. aeruginosa* and *E. coli* at 50mg/ml was recorded by petroleum ether extracts. Activities of the various extracts were comparable to those of standard antibacterial agent ampicillin and DMSO as control. The differences in the observed activities of the various extracts may be due to varying degree of solubility of the active constituents in the four solvents used. It has been documented that different solvents have diverse solubility capacities for different phytochemical constituents [8]. The results of preliminary phytochemical components in leaves of *Ocimum americanum* revealed the presence of alkaloids, phenolic compounds, tannins, lignin, starch, saponins, flavanoids, terpenoid and anthraquinone. (Table 2).

Table 1. Antibacterial efficacy of different solvent extracts of *Ocimum Americanum* leaves

| Sr. No. | Microorganism | Strain +/- | Concentration (mg/ml) | Zone of inhibition (mm) | | | | |
|---------|-------------------------------|------------|-----------------------|-------------------------|------------|---------|------|-----------------------|
| | | | | Petroleum ether | Chloroform | Alcohol | DMSO | Ampicillin (40 µg/ml) |
| 1. | <i>Escherichia coli</i> | -ve | 100 | 08 | 09 | 10 | 0 | 17 |
| | | | 50 | 05 | 08 | 09 | | |
| 2. | <i>Pseudomonas aeruginosa</i> | -ve | 100 | 06 | 07 | 12 | 0 | 18 |
| | | | 50 | 05 | 06 | 10 | | |
| 3. | <i>Staphylococcus aureus</i> | +ve | 100 | 08 | 11 | 23 | 0 | 29 |
| | | | 50 | 08 | 09 | 15 | | |
| 4. | <i>Bacillus subtilis</i> | +ve | 100 | 07 | 09 | 16 | 0 | 20 |
| | | | 50 | 05 | 06 | 12 | | |

Figures are diameter of zone of inhibition (in triplicates)

Table 2. Phytochemical Constituents of *O. americanum* leaves

| Phytochemical Constituents | |
|----------------------------|------|
| Parameter | Test |
| Alkaloids | + |
| Phenolic compounds | + |
| Tannins | + |
| Lignin | + |
| Starch | + |
| Saponins | + |
| flavanoids | + |
| Terpenoid | + |
| Anthraquinone | + |

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