J. Plant Develop.
22(2015): 59 – 76

COMPOSITION OF HERB AND SEED OIL AND ANTIMICROBIAL ACTIVITY OF THE ESSENTIAL OIL OF TWO VARIETIES OF *OCIMUM BASILICUM* HARVESTED AT SHORT TIME INTERVALS

Pandu Sastry KAKARAPARTHI[1*], K. V. N. SATYA SRINIVAS[1], J. KOTESH KUMAR[1], A. NIRANJANA KUMAR[1], Ashish KUMAR[1]

Abstract: An experiment was conducted to study the changes in the chemical composition of the essential oil of two varieties of *Ocimum basilicum* over a period of six months at short harvest intervals for two crop seasons.

In variety Vikarsudha, GC/MS analysis revealed the presence of eighteen essential oil constituents. Linalool (23.5-40.1% and 22.8-33.7%) and methyl chavicol (25.4-51.9% and 40.0-52.7%) were the major constituents in main and ratoon crops.

Similarly, in variety Kuhmohak GC/MS analysis revealed the presence of linalool (19.2-25.4 % and 16.1-31.3%) and methyl chavicol (34.7-53.4% and 39.4-59.2%) in large quantities in main and ratoon crops, respectively. β myrcene, limonene, 1,8 cineole, ocimene, camphor, terpinen-4-ol, bornyl acetate, eugenol, methyl eugenol, β elemene, β caryophyllene, α humulene, γ Cadinene and cadinol were present in small quantities.

Results pertaining to the zone of inhibition in the antimicrobial activity of essential oil indicated that *Chromobacterium violaceum* is more sensitive compared to *Staphylococcus aureus.* Among the fungal strains *Aspergillus niger* was found to be more sensitive.

GC-MS analysis of the fixed oils obtained from the seeds in the ratoon crop revealed the presence of unsaturated and saturated fatty acids. The unsaturated fatty acids averaged 89% consisting of α-linolenic (49.3%-52.4%), linoleic (23.4%-26.0%), and oleic (10.3%-12.3%) acids. The most abundant saturated fatty acids were palmitic and stearic acids.

Key words: *Ocimum*, Lamiaceae, Eugenol, β caryophyllene, methyl eugenol, relative humidity

Introduction

The genus *Ocimum* belongs to the family *Lamiaceae* consists of many species of herbs and shrubs and these are collectively called basils [SIMON & al. 1992]. The number of species reported in the genus varies from 50-60 [HEGNAUER, 1966; SUCHORSKA & OSINSKA, 1992] to 150.

The most important species of *Ocimum* genus is *O. basilicum* L., this species, usually named common basil or sweet basil, is considered economically useful because of their basic natural characteristics as essential oil producers [LAWRENCE, 1993]. Sweet

[1] CSIR-Central Institute of Medicinal and Aromatic Plants, Research Centre, Boduppal, Hyderabad-500092, Andhra Pradesh – India.
[*] Corresponding author. E-mail: sastry.kakaraparthi@gmail.com, crchyd@cimap.res.in

basil is a tender herbaceous annual plant, which originates from tropical and warm areas, such as India, Africa and southern Asia. It is naturalized almost all over the world [HALVA, 1987; SORENSEN & HENRIKSEN, 1992]. The herb on steam distillation yields a bright yellow, volatile oil possessing a pleasant odour. The essential oils are composed of aroma compounds such as eugenol, methyl eugenol, citral, linalool, geraniol and thymol which are required as raw materials for the pharmaceutical, comestics and food industries [GUPTA, 1994; BIZZO & al. 2009]. Basil essential oils are known to possess antimicrobial [BERRINGTON, 2012], and insecticidal activities. Due to its pleasant aroma and antimicrobial activity, basil essential oil is a major aroma chemical with applications in various industries such as the food, pharmaceutical, cosmetic, and aromatherapy.

The variations in the essential oil compositions in *O. basilicum* cultivated in different geographical localities led to the classification of basil into chemotypes on the basis of the prevalent chemical components [LAWRENCE, 1992] or components having composition greater than 20 percent [GRAYER & al. 1996]. When grown for essential oil production, basil is harvested in full bloom, because the content and the composition of the oil are optimal at that stage [BERRINGTON & LALL, 2012; NYKÄNEN, 1989]. Depending on the climate, basil can be harvested one to three times during the cropping season. The essential oils with the finest aroma are obtained from European basil that contains linalool and methylchavicol as the main components. Reunion basil is characterized by high levels of methylchavicol, whereas the tropical chemotype of basil is known to have methyl cinnamate as the main component. Another basil chemotype that is high in eugenol is grown in North Africa, Russia, Eastern Europe, and India.

There is a great variation of essential oil composition (and aroma) among basil cultivars currently on the international market. There is a significant interest in basil as a new high-value essential oil crop in many countries. The objective of this study is to study essential oil content, composition, and bioactivity of two varieties of sweet basil (*O. basilicum*) harvested at short intervals of time in two crop seasons (main crop and ratoon crop). Stage of harvesting plays a dominant role in obtaining good quality oil. Chemical profiles of the essential oils harvested at short intervals of time in *Ocimum* sp are limited. Furthermore, this study also aims at analyzing the fatty acid composition of seed oils and antimicrobial activity of the essential extracted at different harvests.

The information provides scientific background for successful cultivation and production of quality essential oil. Considering these aspects the content and quality of essential oil obtained from two varieties of *Ocimum basilicum* harvested at short intervals of time were studied for two crop seasons.

Materials and methods

Experimental site and design of the experiment

The present study was undertaken to study the seasonal variation in the essential oil composition of two varieties of *Ocimum basilicum* (Vikarsudha and Kushmohak) during a six month period from October, 2012 to March, 2013 at the research farm of Central Institute of Medicinal and Aromatic Plants (CIMAP), Research Centre, Boduppal, Hyderabad, Telangana, India. The experimental site is located at an altitude of 542 m

above sea level with a geographical bearing of 78°8′ E longitude and 17°32′ N latitude. The mean annual rainfall of this region is generally 750 mm.

The soil is a red sandy loam (alficusto chrept) with pH 8.27 (1.25 soils to solution ratio), EC – 1.21 ds/m, organic C – 0.58%, available N (215.40 kg/ha), available P (10.30 kg/ha), and exchangeable K (103.08 kg/ha).

The experimental field was ploughed, harrowed and levelled with tractor drawn implements before starting the nursery. Seeds of CSIR-CIMAP varieties Vikarsudha and Kushmohak were sown in nursery and healthy well grown seedlings (25 days old) were transplanted in rows following a row spacing of 60 cm between rows and 45 cm between plants in 4.8 m x 6.0 m plots. The crop was planted during first week of October, 2012. A fertilizer dose of 60:40:40 Kg/ha of N:P:K was applied to the crop. Uniform doses of P and K were applied during ploughing. Nitrogen was applied in four splits. The crop was managed as per standard practices under irrigated conditions in 40 plots cultivated uniformly under similar conditions. The plots were kept weed free.

Between first week of October and middle of December 2012, the crop was harvested five times in different plots. The days of harvesting constituted the treatments and the treatments were replicated five times in a randomized complete block design. This is designated as the main crop. The field plots were again harvested commonly up to a height of 20 cm above ground level in the third week of December 2012. During the seventy days period from 15th December 2012 to the end of February 2013 the crop was harvested seven times at 10 days interval up to the end of February 2013. The dates of harvesting constituted the treatments and replicated five times. This was designated as ratoon crop.

Basilicum varieties (Kushmohak and Vikarsudha) were harvested five times at 15 days interval at 15,30,45,60 and 75 days in the main crop and seven times at ten days interval (10, 20, 30, 40, 50, 60 and 70 days) during the ratoon crop. Observations were recorded at each harvest on the morphological characters, essential oil content, and composition.

Observation on morphometric traits

The herb was harvested from ten randomly selected plants in each treatment plot replication wise and data were recorded for morphometric traits viz., number of leaves/plant, leaf area, and leaf weight/plant. The fresh weight of plant was also taken for calculating the essential oil yield/plant.

Essential oil extraction/distillation

The aerial parts of ocimum were collected from ten random plants in each plot. For the extraction of essential oils, freshly collected herbage was subjected to hydro-distillation using a Clevenger-type apparatus for 4.0 h. The essential oils obtained were dried over anhydrous sodium sulphate and stored at 4 °C until the GC analysis was carried out. The oil content and quality were observed five times in the first phase and seven times in the second phase replication wise.

Gas Chromatography analysis of essential oil

The essential oils were analyzed on a Varian CP-3800 model gas chromatograph with Galaxy software system equipped with flame ionization detector (FID) and an electronic integrator. Separation of the compounds was achieved employing a Varian CP-Sil 5CB capillary column (ID: 50 m X 0.25 mm; film thickness 0.25 μm). Nitrogen was

used as the carrier gas at a constant flow rate of 0.4 ml/min. The column temperature was programmed from 120 °C (held for 2 min.) to 240 °C (held for 5 min.) at a rate of 8 °C/min. The injector and detector temperature were set at 250 °C and 300 °C respectively. Samples of 0.2 µL were injected with a 20:100:20 split ratio. Retention indices were generated with a standard solution of n-alkanes (C_6-C_{19}). The composition was reported as a relative percentage of the total peak area without FID response factor correction.

Gas Chromatography/Mass Spectrometry (GC/MS) analysis of essential oil

GC-MS analysis was carried out on a SHIMADZU GCMS-QP2010 PLUS using a Zebron ZB5MS capillary column (ID: 30 m X 0.32 mm; film thickness 0.25 µm). The column initially held at 90 °C for 4.5 min, then heated to 150 °C at a rate of 7 °C /min and to 170 °C at a rate of 10 °C /min, held for 8 min. Injector and detector temperatures were kept at 250 °C. Helium was used as carrier gas at 86.1 KPa (12.48 Psi). Mass detection was performed by an electron ionization mode with ionization energy of 70 eV and ion source temperature of 250 °C.

Chemical compounds identification

The identification of the essential oil constituents was based on comparison of their retention indices relative to homologous series of n-alkanes (C_6-C_{19}; Poly Science; Niles, USA) and published data. Chemical constituents were further confirmed by correlating the GC data with GC-MS data and compared to the NIST mass spectral library.

Evaluation of antibacterial activity

Staphylococcus aureus and *Chromobacterium violaceum were* used in the present study were collected from Microbial Type Culture Collection and Gene Bank (M.T.C.C) Institute of Microbial Technology Sector, Chandigarh, India. The antibacterial activity of these ocimum oils against *Staphylococcus aureus* (MTCC No: 9542) and *Chromobacterium violaceum* (MTCC No: 8071) was determined using the disk diffusion method.

The cultures were cultivated on Nutrient Agar (HiMedia) at 25 °C for 48 hours. The medium and petridishes were autoclaved at a pressure of 15 lb/inc^2 for 20 min. The medium was poured into sterile petridishes under aseptic conditions in laminar flow chamber. When the medium in the plate solidify, 0.5 ml of 24 h cultured of test organism was inoculated and uniformly spread over the agar surface with a sterile L-shaped rod. Controls were maintained with DMSO and pencillin G (for gram-positive) and streptomycin (for gram-negative). The treated materials and the controls were kept in an incubator at 37 °C for 24 h. Ocimum essential oils were applied on filter paper (2.0 and 4.0 µL/disk) disks of 6 mm in diameter separately. 10 mg/mL of pencillin G (for gram-positive) and streptomycin (for gram-negative) were used. These disks were placed on the surface of seeded agar plates at equal distance. Inhibition zones were measured and diameter was calculated in mm. Two replicates were maintained for each treatment.

Evaluation of antifungal activity

Aspergillus niger (MTCC No: 9687) and *Candida albicans* (MTCC No: 7253) were used for Antifungal assay and collected form Institute of Microbial Technology Sector, Chandigarh, India. The method followed for antifungal bioassay is similar to that followed for antibacterial assay where in the medium is Potato Dextrose Agar (HiMedia) and the control is Fluconazole. Control was maintained with DMSO with Fluconazole. The treated and the control were kept in an incubator at room temperature for 48 h. Incubation

zones were measured and diameter was calculated in mm. Two replicates were maintained for each treatment.

Seed oils extraction

Seeds of CIMAP developed two *Ocimum* varieties were cleaned and powdered. Each powdered material was exhaustively extracted for fixed oil by solvent hexane in a soxhlet apparatus. All the extracts were concentrated at reduced pressures to obtain crude residues.

Preparation of fatty acid methyl esters

All the samples were treated with 2% H_2SO_4 in methanol (10 ml) at reflux temperature (65 °C) for 4 hr for the conversion of lipid component to its fatty acid methyl esters. The reaction mixture was extracted with ethyl acetate (3 x 15 ml) and the washed with water. The organic layer was dried over anhydrous sodium sulphate and concentrated to get the fatty acid methyl ester (FAME) mixture.

GC/GC-MS Analysis for the Determination of Fatty acid composition: FAME mixture was dissolved in minimum amount of chloroform and analyzed by GC and G-MS for fatty acid composition.

GC analysis fatty acids

Agilant 6890 N Series gas chromatograph equipped with a FID detector. The GC was performed using DB-225 capillary column (30 m x 0.25 mm x 0.25 µm) and the oven temperature: 160 °C (2min)-5 °C.min-230 °C (20 min); N2 flows: 1.0 mL/min and the split ration – 50:1.

GC-MS analysis fatty acids

Agilant 6890 N series gas chromatograph connected to Agilant 5973 mass spectrometer (Palo Alto, USA). The GC-MS detection was performed at 70 eV (m/z 50-550; source at 230 °C and quadruple at 150 °C) in the EI mode attached with DB-225 ms capillary column (30 m x 0.25 mm x 0.25 µm). The oven temperature: 2 min at 160 °C (2min)-5 °C/min °C (20 min): He flow: 1.0 mL/min and the split ration 50:1.

Statistical analysis

Analysis of variance was performed to determine the effect of different times of harvest on morphological traits, essential yield and quality parameters using statistical software IRRISTAT [IRRI, Manila, Philippines]. Means were compared using least significant differences [LSDs] at 5% probability levels.

Results

Leaf number, leaf weight /plant and leaf area

In Vikarsudha leaf weight (58.4-368.4 g/plant) and leaf number (163-553/ plant) increased up to 60 days harvest in the main crop (Tab. 1). In the ratoon crop leaf weight and leaf number /plant increased up to 60 days harvest and later decreased, whereas leaf area increased up to 75 days harvest.

In the variety Kushmohak, leaf weight (55-350 g/plant) and leaf number (32-314/plant) increased up to 60 DH where as leaf area showed significant improvement up to 70 DH (14-32 cm^2) in the main crop (Tab. 1). Similar results were noticed in the ratoon crop also. Results of the present investigations confirm morphological and

developmental variability of basil with respect to leaf characteristics which are inconformity with those reported earlier [SIMON & al. 1992; GRAYER & al. 1996].

Oil content and oil yield/plant

In Vikarsudha, oil content decreased from 0.6 to 0.3 with advancement in the harvest date (DH) from 15 to 75 days in the main crop (Tab. 1) where as oil yield/plant increased from 1.1 to 4.4 g/plant up to 60 DH and then decreased to 2.8 g/plant at 75 DH during the same period in the main crop.

In the ratoon crop oil content increased up to 40 DH and later showed a study decline where as oil yield showed a study increase from 0.3 to 4.17 g/plant at 60 DH and later decreased.

In the variety Kushmohak, oil content exhibited an increase decrease pattern in both the seasons and varied around 0.45 on an average (Tab. 1). Oil yield/plant showed a significant improvement up to 60 DH and decreased later in both the seasons.

Differences in the essential oil content in different varieties of the same sps. and variable composition of the essential oil with time and changes in weather parameters in the same variety were reported by many workers. The oil content of sweet basil varieties reported in this study was similar to several literature reports [SUCHORSKA & OSIŃSKA, 2001; MAROTTI & al. 1996; WETZEIL & al. 2002; SUCHORSKA & OSINSKA, 1992].

Essential oil composition at different days of harvest

Vikarsudha: In *Ocimum basilicum* variety Vikarsudha, the essential oil distilled at different harvest dates was subjected to GC/GCMs analysis and nineteen chemical constituents in the oil (> 94% of the total) were identified. The constituents identified are (Tab. 2) camphene, β myrcene, limonene, 1-8-cineole (2.7 to 5.2), ocimene, linalool (23.5 to 40.1 %), camphor, terpinen-4-ol, methyl chavicol (25.4 to 51.9 %), neral, geranial, bornyl acetate, eugenol, methyl cinnamate, methyl eugenol, β caryophyllene, α humulene, δ cadinene, and cadinol in the main crop. Similar constituents were observed in the ratoon crop also (Tab. 2). In the ratoon crop the major constituents in the essential oil were linalool (22.8-33.7%) and methyl chavicol (39.0-52.7%).

Linalool exhibited an increase-decrease pattern (28.0-40.1-23.5%) in the main crop and showed a decrease-increase pattern in the ratoon crop (33.7-22.8-30.6%). Methyl chavicol exhibited an erratic pattern in the main crop and an increase-decrease pattern in the ratoon crop (40.0-52.7-38.9%).

Kushmohak: In *Ocimum basilicum* variety Kushmohak, the essential oil distilled at different harvest dates was subjected to GC/GCMs analysis and 92% of the chemical constituents in the oil were identified. The constituents identified were (Tab. 3) camphene, β myrcene, limonene, 1,8 cineole (3.2-4.5%), ocimene, linalool (19.2 to 25.5 %), camphor, terpinen-4-ol, methyl chavicol (34.7-53.4%), bornyl acetate, eugenol (2.1-5.4%), methyl eugenol (1.9-10.1 %), β elemene, β caryophyllene, α humulene, γ Cadinene and cadinol (1.4 to 2.1%).

During the main crop season, linalool remained constant around 25% up to 45 DH and later it decreased. Other major constituent methyl chavicol showed a definite increase from 47.1 to 53.4 at 45 DH and later it decreased to 34.7% at 75 DH. Ocimene, β

caryophyllene, γ cadinene and cadinol showed decreasing pattern with time, whereas limonene, terpinen-4-ol, bornyl acetate (0.16-1.2%), eugenol (2.1-5.4%) and methyl eugenol (1.9-10.1%) showed increasing pattern with time (Tab. 3).

In the ratoon crop, though the content of linalool showed a variable pattern with time of harvest the overall trend was a decrease in linalool content of the oil with late harvesting up to 70 DH. Methyl chavicol increased from 52.7 to 59.2 at 30 DH and it decreased in the later harvests. β pinene (0.4-1.4%), limonene (0.1- to 0.3%) and 1,8 Cineole (2.2-5.1%) showed increasing pattern (Tab. 3).

The chemical composition of basil oil has been the subject of considerable studies. In basil cultivars from Australia, methyl chavicol, linalool, methyl cinnamate, a mixture of linalool/methyl cinnamate, and linalool/ methyl chacivol were reported as the main components [LACHOWICZ & al. 1997]. In the oils obtained from aerial parts of basil grown in Colombia and Bulgaria, linalool and methyl cinnamate were reported as major components of oils, respectively [VIŇA & MURILLO, 2003].

Antimicrobial activity

The essential oil of *O. basilicum* extracted from both the varieties exhibited strong antimicrobial activity against both bacterial strains whereas antifungal activity (Tab. 4) was noticed at higher dose of application only. Results pertaining to the zone of inhibition indicated (Tab. 4) that among bacterial strains tested, *Chromobacterium violaceum* is more sensitive organism compared to *Staphylococcus aureus.* Similarly among the fungal strains *Aspergillus niger* was found to be more sensitive than *Candida albicans*.

Among the pure compounds eugenol exhibited strong antibacterial activity compared to methyl chavicol, β caryophyllene and linalool (Tab. 5). Eugenol also exhibited strong antifungal activity compared to methyl chavicol.

Essential oil obtained from plants harvested at 10 and 20 days exhibited higher antibacterial activity compared to the oil extracted from old plants. The activity was more at higher dose (4 µl) compared to lower dose in case of all organisms tested in both the varieties.

The activity of the oil decreased with advancement in age of harvest in case of bacterial strains *Staphylococcus aureus* and *Chromobacterium violaceum.* Both the varieties of basil exhibited significant antibacterial activity compared to control pencilin G and streptomycin at the early stage harvested oils. Only in case of fungal strain *Aspergillus niger* higher activity was noticed with the essential oil obtained from late harvests.

Seed fixed oil

Composition of basil seed oils evaluated in this study are shown in Tab. 6. Oil content in the seeds varied from 21.6 % in Vikarsudha to 12.4 % in Kushmohak. The seed yield observed was 230 kg/ha in Vikarsudha and 250 kg/ha in Kushmohak.

Unsaturated fatty acids averaged 85.6-88.1%, including α-linolenic (49.3-52.4%), linoleic (23.6-26%), and oleic acids (10.3-12.3%). The most abundant saturated fatty acids were palmitic (8.0 % -9.2%) and stearic (3.6-3.8%).

Discussion

Essential oil content and oil yield/plant

Oil content: In Kushmohak, the oil content ranged from 0.4 to 0.5% during the five harvest dates in the main crop and, the oil content increased from 0.2 to 0.5% at 30 DH and remained the same up to 60 DH and later decreased to 0.30 at 70 DH in the ratoon crop.

In Viukarsudha the oil content decreased with age whereas in the ratoon crop oil content exhibited a variable pattern and ranged from 0.3 to 0.7% and cantered around 0.45 % in the ratoon crop.

Differences in the essential oil content in different varieties of the same sps. and variable composition of the essential oil with time and changes in weather parameters in the same variety were reported by many workers. The oil content of sweet basil varieties reported in this study was similar to several literature reports. The oil content in sweet basils from different sources (Germany, Romania, Hungary and Egypt) was reported to be varied from 0.1 to 0.55% [SUCHORSKA & OSIŃSKA, 2001]. The content of essential oil in herb of ten Italian basil cultivars ranged from 0.3 to 0.8% [MAROTTI & al. 1996]. In another study essential oil content varied from traces to 2.65% in 270 sweet basil accessions studied in Germany [WETZEIL & al. 2002]. Such variations in the essential oil content of basil across countries might be attributed to the varied agroclimatic conditions of the regions besides their genetic makeup [SUCHORSKA & OSINSKA, 1992; GALAMBOSI & SZEBENI, 2002; SEIDLER-ŁOŻYKOWSKA & KRÓL, 2008].

Oil yield/plant: In both the varieties, oil yield per plant showed an increasing tendency with age due to the increase in leaf weight and area and when there was a drop in leaf weight after 60 days there was a decrease in oil yield per plant which was a product of oil content and herb weight. Though the oil content was very high at early stages the oil yield was less due to low herb weight. The results obtained in this study are in agreement with most of previous works which reported that the full-flowering stage is characterized by the highest essential yield. Oliveria and his coworkers (2005) reported that the essential oil yield increased during plant development to reach a maximum during the flowering stage. In earlier studies highest essential content and oil yield at full flowering stage was reported in *Thymus vulgaris* [OZGUVEN & TANSI, 1998], *Artemisia pallens* Wall [MALLAVARAPU & al. 1999] peppermint [ROHLOFF & al. 2005], *Satureja rechingeri* [SEFIDKON & al. 2007], and in oregano [KIZIL & al. 2008]. The essential oils yields in another crop *Melissa officinalis* were reported to vary considerably from month-to-month and vary significantly with harvesting stages [KEIVAN SAEB & al. 2012].

Essential oil composition

Essential oil distilled at different harvest dates was subjected to GC/GCMs analysis and 92% of the chemical constituents in the oil were identified. In Vikarsudha variety, a total seventeen constituents were identified and linalool and methyl chavicol were found to be the major constituents in the essential oil.

Linalool exhibited an increase – decrease pattern (28.0-40.1-23.5%) in the main crop and showed a decrease – increase pattern in the ratoon crop (33.7-22.8-30.6%). Methyl chavicol exhibited an erratic pattern in the main crop and an increase – decrease pattern in the ratoon crop (40.0-52.7-38.9%).

In *Ocimum basilicum* variety Kuhmohak, linalool (19.2 to 25.5%) and, methyl chavicol (34.7-53.4%) were found to be the major constituents. During the main crop season, linalool remained constant around 25% up to 45 DH and later it decreased. Other major constituent methyl chavicol showed a definite increase from 47.1 to 53.4 at 45 DH and later it decreased to 34.7 % at 75 DH.

In the ratoon crop, though the content of linalool showed a variable pattern with time of harvest the overall trend was a decrease in linalool content of the oil with late harvesting up to 70 DH. Methyl chavicol increased up to 30 DH and it decreased in the later harvests.

In both the varieties methyl chavicol is present in large quantities compared to linalool and both exhibited variable patterns with date of harvest. Linalool exhibited a decreasing tendency with age whereas methyl chavicol exhibited an increase in the vegetative phase and with the onset of flowering it started declining.

The chemical composition of basil oil has been the subject of considerable studies. Methyl chavicol, linalool, methyl cinnamate, a mixture of linalool/methyl cinnamate, and linalool/methyl chacivol were reported as the main components in basil cultivars from Australia [LACHOWICZ & al. 1997], linalool and methyl cinnamate were reported as major components of oils from Colombia and Bulgaria respectively [VIŇA & MURILLO, 2003].

Due to the high content of linalool, and methyl chavicol the studied cultivars may become applied in food and perfume industries, food seasoning and flavouring, aromatheraphy, and medicinal application.

In the present study higer seasonal variation was not noticed in the essential oil composition, whereas high percentage of linalool (60.6%) [HUSSAIN & al. 2008] and estragole (52.6 and 58.26%) were reported in winter grown basil compared to summer [CHALCHAT & OZCAN, 2008]. Higher solar irradiance level increased the contents of linalool and eugenol in *Ocimum basilicum* [CHANGA & al. 2008]. The hydro-distilled essential oils content ranged from 0.5% to 0.8%, the maximum amounts were observed in winter while minimum in summer [HUSSAIN & al. 2008].

Anti microbial activity of essential oil

The major constituents in the essential oil linalool and methyl chavicol showed increasing g pattern up to 30-40 DH and later showed a decreasing pattern. The decrease in antimicrobial activity against the bacterial strains with a later harvest dates may be due to the decrease in the concentration of the major constituents. In the varity Kushmohak exhibited a increased activity agains fungal strains with essential oil obtained from later harvests, which might be due to increase in eugenol and 1-8 cineole which showed increasing pattern with late harvest dates.

Some earlier reports showed that the changes in chemical composition of an essential oil directly affected their biological activities [CELIKTAS & al. 2007; VAN VUUREN & al. 2007; SUPPAKUL & al. 2003]. Literature indicates that basil essential oils exhibited good to moderate antimicrobial activity against a wide range of microorganisms [SUPPAKUL & al. 2003; WANNISSORN & al. 2005]. Gram-positive strains of bacteria showed higher sensitivity to *O. basilicum* essential oils than those of their counterpart [BOZIN & al. 2006; LOPEZ & al. 2005].

Linalool in the essential oil was reported to be responsible for the antifungal [SOKOVIC & VAN GRIENSVEN, 2006] and antimicrobial activities of essential oils from *O. basilicum* [KOUTSOUDAKI & al. 2005; SARTORATOTTO & al. 2004]. Evaluation of antimicrobial activity of the essential oils and linalool, the most abundant component, against bacterial strains: *Staphylococcus aureus*, *Escherichia coli*, *Bacillus subtilis*, *Pasteurella multocida* and pathogenic fungi *Aspergillus niger*, *Mucor mucedo*, *Fusarium solani*, *Botryodiplodia theobromae*, *Rhizopus solani* indicated that all the tested microorganisms were affected [HUSSAIN & al. 2008].

The essential oils from *O. basilicum* varieties showed broad activity against bacteria and pathogenic fungi. The production of essential oils and their utilization as potential natural food preservants could be of economical value. However, further investigation to establish how components interact to provide the antioxidant activity is needed.

Seed oils

Oil content in the seeds varied from 21.6% in Vikarsudha to 12.4% in Kushmoh and seed yield from 230 kg/ ha in Vikarsudha and 250 kg/ha in Kushmohak.

Unsaturated fatty acids averaged 85.6-88.1%, including α-linolenic (49.3-52.4%), linoleic (23.6-26%), and oleic acids (10.3-12.3%). The most abundant saturated fatty acids were palmitic (8.0-9.2%) and stearic (3.6-3.8%). Values from the literature range from 5.3% to 15.4% for oleic acid, 14.0% to 66.1% for linoleic acid, and 15.7% to 65.0% for linolenic acid [PATWARDHAN, 1940; HENRY & GRINDLEY, 1944].

Basils are multi harvest multi utility crops. The main crop can be used for extraction of essential oil and the second crop can be used for production of dry leaf and seed oil. The seed oil content is high and the seed yield /ha is in the range 230-250 kg/ha a good quality seed oil to the extent 34.5 to 37.5 kg/ha can be obtained in the ratoon crop. This provides more income to the cultivator as this system facilitates production of essential oil for aroma chemicals, seed oil for food industry and also dry herb for traditional medicine industry and herbal tea manufacturers.

Basil cultivation technology is a very well known process through modern plant breeding techniques seed and oil content yields could be increased. Seeds of basil do not readily dehisce and can be harvested using a combine. A high linolenic acid oil, such as that found in *O. basilicum* and *O. canum*, could be used in the paint, varnish and ink industries, and as a source of linolenic acid, while oils with lower linolenic acid contentcan be used by the food industry.

Acknowledgements

The authors are thankful to the Director, CIMAP, and Lucknow for the encouragement and facilities.

References

BERRINGTON D. & LALL N. 2012. *Anticancer activity of certain herbs and spices on thecervical epithelial carcinoma (HeLa) cell line. eCAM*. 1-11.

BIZZO H. R., HOVELL A. M. C. & REZEND E. C. M. 2009. Óleos essenciais no Brasil: Aspectos gerais, desenvolvimento e perspectivas. *Quim Nova*. **32**: 588-594.

BOZIN B., MIMICA-DUKIC N., SIMIN N. & ANACKOV G. 2006. Characterization of the volatile composition of essential oil of some lamiaceae species and the antimicrobial and antioxidant activities of the entire oils. *Journal of Agriculture and Food Chemistry*. **54**: 1822-1828.

CELIKTAS O. Y., KOCABAS E. E. H., BEDIR E., SUKAN F. V., OZEK T. & BASER K. H. C. 2007. Antimicrobial activities of methanol extracts and essential oils of Rosmarinus oficinalis, depending on location and seasonal variations. *Food Chemistry*. **100**(2): 553-559.

CHALCHAT J. C. & O ZCAN M. M. 2008. Comparative essential oil composition of flowers, leaves and stems of basil (*Ocimum basilicum* L.) used as herb. *Food Chem*. **110**: 501-503.doi:10.1016/j.foodchem.2008.02.018.

CHANGA X., ALDERSON P. G. & WRIGHT C. J. 2008. Solar irradiance level alters the growth of basil (*Ocimum basilicum* L.) and its content of volatile oils. *Environ. Exp. Bot*. **63**: 216-223. doi:10.1016/j.envexpbot.2007.10.017.

GALAMBOSI B. & SZEBENI Z. S. 2002. Chemical analyses of Hungarian basil varieties in Finland. *Kertgazdasag*. **XII**(5): 44-53.

GRAYER R. J., KITE G. C., GOLDSTONE F. J., BRYAN S., PATON A. & PUTIEVSKY E. 1996. Infraspecific taxonomy and essential oil chemotypes in sweet basil, *Ocimum basilicum. Photochemistry*, **43**: 1033-1039.

GUPTA R. 1994. Basil (*Ocimum* spp.). *G-15 Gene Banks for Medicinal and Aromatic Plants Newsletter*. **1**: 1-3.

HÄLVÄ S. 1987. Studies on fertilization of dill (*Anethum graveolens* L.) and basil (*Ocimum bastticum* L.) III. Oil yield of basil affected by fertilization. *J. of Agric. Sci. in Finland.* **59**: 25-29.

HEGNAUER R.1973. *Chemotaxonomie der Pflanzen*, Vol. **6**, Birkhäuser Verlag, Basel and Stuttgart, Germany.

HENRY A. J. &.GRINDLEY D. N. 1944. The oils and seeds of *Ocimum kilimandscharicum*, *Euphorbia calycina*, *E. erythareae*, *Sterculia tomentosa* and *Trichilia emetica*. *J. Soc. Chem. Ind.* **63**: 188-194.

HUSSAIN A. I., ANWAR F., SHERAZI S. T. H. & PRZYBYLSKI R. 2008. Chemical composition, antioxidant and antimicrobial activities of basil (Ocimum basilicum) essential oils depends on seasonal variations. *Food Chem.* **108**: 986-995. doi:10.1016/j.foodchem.2007.12.010.

HUSSAIN A. I., ANWAR F., T. H. SHERAZI S. & PRZYBYLSKI R. 2008. Chemical composition, antioxidant and antimicrobial activities of basil (*Ocimum basilicum*) essential oils depends on seasonal variations. *Food Chem.* **108**: 986-995. doi:10.1016/j.foodchem.2007.12.010.

KEIVAN SAEB. & SARA GHOLAMREZAEE. 2012. Variation of essential oil composition of *Melissa officinalis* L. leaves during different stages of plant growth. *Asian Pacific Journal of Tropical Biomedicine.* **2**: 547-549.

KIZIL S., IPEK A., ARSLAN N. & KHAWAR K. M. 2008. Effect of different developing stages on some agronomical characteristics and essential oil composition of oregano (*Origanum onites*). *N. Z. J. Crop Hort. Sci.* **36**: 71-76.

KOUTSOUDAKI C., KRSEK M. & RODGER A. 2005. Chemical composition and antibacterial activity of the essential oil and the gum of Pistacia lentiscus Var. chia. *Journal of Agricultural and Food Chemistry.* **53**: 7681-7685.

LACHOWICZ K. J., JONES G. P., BRIGGS. D. R., BIENVENU. F. E., PALMER M. V., MISHRA V. & HUNTER. M. M. 1997. Characteristics of plants and plant extracts from five varieties of basil (*Ocimum basilicum* L.) grown in Australia. *J Agric Food Chem.* **45**: 2660-2665.

LAWRENCE B. M. 1993. *Labiatae oils-mother nature. Chemical factory, essential oils, allured, Carol Stream,* IL, 88-206.

LAWRENCE B.M. 1992. *Chemical components of Labiatae oils and their exploitation.* In: *Advances in Labiatae Science. Ed*. HARLEY R. M., REYNOLDS T. *Royal Botanical Gardens: Kew, UK*. 399-436.

LOPEZ P., SANCHEZ C., BATLLE R. & NERIN C. 2005. Solid and vaporphase antimicrobial activities of six essential oils: Susceptibility of selected food borne bacterial and fungal strains. *Journal of Agriculture and Food Chemistry*. **53**: 6939-6946.

MALLAVARAPU G. R., KULKARNI R. N., BASKARAN K., RAO L. & RAMESH S. 1999. Influence of plant growth stage on the essential oil content and composition in davana (*Artemisia pallens* Wall.). *J. Agric. Food Chem.* **47**: 254-258.

MAROTTI M., PICCAGLIA R. & GIVANELLI E. 1996. Differences in essential oil composition of basil (*Ocimum basilicum* L.) Italian cultivars related to morphological characteristics. *J Agric Food Chem.* **44**: 1033-1039.

NYKÄNEN I. 1989. The effect of cultivation conditions on the composition of basil oil. *Flav. Fragr. J.* **4**: 125-128.

OLIVEIRA M. J., CAMPOS I. F. P., OLIVEIRA C. B. A., SANTOS M. R., SOUZA P. S., SERAPHIN J. C. & FERI P. H. 2005. Influence of growth phase on essential oil composition of *Hyptis suaveolens. Biochem. Syst. Ecol.* **33**: 275-285.

OZGUVEN M. & TANSI S. 1998. Drug yield and essential oil of *Thymus vulgaris* L. as influenced by ecological and ontogenetical variation. *Turk. J. Agric. Forest.* **22**: 537-542.

PATWARDHAN V. A. 1940. *Thesis, University of Bombay, 1930 cited by Hilditch, T.P., in the chemical constitution of natural fats*, Wiley, New York.

ROHLOFF J., DRAGLAND S., MORDAL R. & HENNING IVERSEN T. 2005. Effect of harvest time and drying method on biomass production, essential oil yield and quality of peppermint (*Mentha×piperita* L.). *J. Agric. Food Chem*. **53**: 4143-4148.

SARTORATOTTO A., MACHADO A. L. M., DELARMELINA C., FIGUEIRA G. M., DUARTE M. C. T. & REHDER V. L. G. 2004. Composition and antimicrobial activity of essential oils from aromatic plants used in Brazil. *Brazilian Journal of Microbiology*. **35**: 275-280.

SEFIDKON F., ABBASI K., JAMZAD Z. & AHMADI S. 2007. The effect of distillation methods and stage of plant growth on the essential oil content and composition of Saturejare chingeri Jamzad. *Food Chem.* **100**: 1054-1058.

SEIDLER-ŁOŻYKOWSKA K. & KRÓL D. 2008. The content of essential oil in ten sweet basil (*Ocimum basilicum* L.) cultivars and its composition. *Herba Pol.* **54**(3): 7-12.

SIMON J. E., REISS-BUBENHEIM D., JOLY R. J. & CHARLES D. J. 1992. Water-stress induced alterations in essential oil content and composition of sweet basil. *J. Ess. Oil Res.* **4**: 71-75.

SOKOVIC M. & VAN GRIENSVEN L. J. L. D. 2006. Antimicrobial activity of essential oils and their components against the three major pathogens of the cultivated button mushroom, *Agaricus bisporus*. *European Journal Plant Pathology*. **116**: 211-224.

SÖRENSEN L. & HENRIKSEN K. 1992. Effects of seed rate, plastic covering, and harvest time on yield and quality of Danish grown basil (*Ocimum basilicum*). *Danish Journal of Plant and Soil Science.* **96**: 499-506.

SUCHORSKA K. & OSINSKA E. 1992. Variability of the purple and the lettuce-leaved sweet basil (*Ocimum basilicum* L.). *Herba Polonica*, **XXXVIII**: 175-181.

SUCHORSKA K. & OSINSKA E. 1992. Variability of the purple and the lettuce-leaved sweet basil (*Ocimum basilicum* L.). *Herba Polonica*, **XXXVIII**: 175-181.

SUCHORSKA K. & OSIŃSKA E. 2001. Morphological developmental and chemical analyses of 5 forms of sweet basil (*Ocimum basilicum* L.). *Ann Warsaw Agric Univ Hortic (Landscape Architecture)*. **9**: 17-22.

SUPPAKUL P., MILTZ J., SONNEVELD K. & BIGGER S. W. 2003. Antimicrobial properties of basil and its possible application in food packing. *Journal of Agriculture and Food Chemistry*. **51**: 3197-3207.

VAN VUUREN S. F., VILJOEN A. M., OZEK T., DEMIRICI B. & BASER K. H. C. 2007. Seasonal and geographical variation of *Heteropyxis natalensis* essential oil and the effect thereof on the antimicrobial activity. *South African Journal of Botany*. **73**(3): 441-448.

VIŇA A. & MURILLO E. 2003. Essential oil composition from twelve varieties of basil (*Ocimum* spp.) grown in Colombia. *J Braz Chem Soc*. **14**(5):744-749.

WANNISSORN B., JARIKASEM S., SIRIWANGCHAI T. & THUBTHIMTHED S. 2005. Antibacterial properties of essential oils from Thai medicinal plants. *Fitoterapia*. **76**: 233-236.

WETZEIL S. B., KRUGER H., HAMMER K. & BACHMANN K. 2002. Investigations on morphological and molecular variability of *Ocimum* L. species. *J Herb Spices Med Plants.* **9**: 8183-8187.

How to cite this article:

KAKARAPARTHI P. S., SATYA SRINIVAS K. V. N., KOTESH KUMAR J., NIRANJANA KUMAR A. & KUMAR A. 2015. Composition of herb and seed oil and antimicrobial activity of the essential oil of two varieties of *Ocimum basilicum* harvested at short time intervals. *J. Plant Develop*. **22**: 59-76.

***Received*: 17 April 2015 / *Accepted*: 16 November 2015**

Tab. 1. Morphological characters, oil content and essential oil yield /plant in of *Ocimum basilicum* varieties Vikarsudha and Kushmohak at different times of harvest during October 2012 to February, 2013

Harvest date	leaf wt,g	Leaf n o	leaf area, cm^2	Oil content	Oil yield, g/plant	leaf wt, g	Leaf no	leaf area, cm^2	Oil content	Oil yield, g/plant
	Vikarsudha					Kushmohak				
15	58.4	163.0	15.0	0.6	1.1	55.0	32.0	14.0	0.5	0.8
30	253.6	221.0	23.0	0.6	4.6	165.0	143.0	21.0	0.4	2.0
45	358.4	435.0	24.0	0.5	5.4	197.0	212.0	24.0	0.5	3.0
60	368.4	553.0	28.0	0.4	4.4	350.0	314.0	26.0	0.5	5.3
75	306.0	427.0	31.0	0.3	2.8	302.0	221.0	32.0	0.4	3.6
CD	13.8	10.75	2.47	0.12	0.04	5.8	7.5	1.91	0.04	0.07
CV	11.06	4.05	7.27	16.84	5.92	6.34	3.63	5.45	6.17	9.52
10	32.5	68.0	24.0	0.3	0.3	52.0	**25.0**	16.0	0.2	0.3
20	43.5	196.0	28.0	0.6	0.8	142.0	96.0	23.0	0.4	1.7
30	97.3	212.0	31.0	0.7	2.0	267.0	146.0	26.0	0.5	4.0
40	157.0	324.0	31.0	0.7	3.3	289.0	198.0	29.0	0.5	4.3
50	217.5	412.0	34.0	0.5	3.3	352.0	324.0	31.0	0.5	5.3
60	343.5	522.0	34.0	0.4	4.1	387.0	392.0	32.0	0.5	5.8
70	326.9	512.0	34.0	0.4	3.9	321.0	265.0	36.0	0.3	2.9
CD	12.89	0.04	0.04	0.09	0.04	12.18	0.04	0.04	0.05	0.09
CV	9.26	0.01	0.09	14.34	5.59	4.81	0.01	0.08	6.58	6.1

Tab 2. Chemical composition of the essential oil of *Ocimum basilicum* variety Vikarsudha at different times of harvest during October 2012 to February, 2013

Harvest date	Chemical constituents of essential oil, %																		
	Camphene	β myrcene	Limonene	1,8 Cineole	Ocimene	Linalool	Camphor	Terpinen-4-ol	Methyl chavicol	Neral	Geranial	Bornyl acetate	Eugenol	Methyl cinnamate	Methyl Eugenol	β Caryophyllene	α Humulene	γ Cadinene	Cadinol
15	0.2	0.9	0.2	5.2	0.3	28.1	0.9	0.2	45.0	0.8	0.9	0.1	1.9	0.8	5.1	1.5	0.3	0.5	1.5
30	0.1	0.6	0.1	4.7	0.2	35.5	0.8	0.4	36.4	1.0	1.1	0.4	1.1	2.3	2.9	1.8	0.4	0.6	1.6
45	0.1	0.6	0.1	4.5	0.1	40.1	0.8	0.8	32.7	0.8	0.9	0.1	2.8	0.8	5.1	1.6	0.2	0.5	1.3
60	0.1	0.7	0.2	4.3	0.2	26.6	1.1	1.0	51.9	0.3	0.2	0.2	2.7	0.4	1.9	1.3	0.2	0.4	1.2
75	0.3	0.5	0.1	2.7	0.1	23.5	1.5	1.0	25.4	1.4	0.4	2.6	4.2	1.6	6.0	1.1	0.3	0.5	0.9
CD	0.04	0.12	0.04	1.40	0.04	10.79	0.17	0.14	9.22	0.11	0.11	0.10	0.92	0.91	1.40	0.18	0.04	0.12	0.17
CV	15.33	11.59	17.55	22.41	16.01	24.12	11.38	13.98	16.82	9.10	10.73	32.68	24.92	35.42	22.87	8.42	10.38	15.11	9.34
10	0.2	0.8	0.2	4.2	0.2	33.7	0.7	1.0	40.0	1.3	1.4	0.3	1.7	1.7	1.9	1.2	0.2	0.4	1.0
20	0.2	0.8	0.1	5.1	0.1	29.5	0.8	0.5	48.4	0.4	0.4	0.2	2.1	1.1	4.1	0.9	0.2	0.3	0.9
30	0.2	1.0	0.2	5.9	0.1	22.9	0.6	0.7	52.3	0.4	0.4	0.3	2.6	0.8	4.1	1.3	0.2	0.5	1.1

40	0.2	0.8	0.2	4.8	0.2	22.8	0.6	0.7	52.7	0.6	0.7	0.3	1.5	1.8	3.7	1.4	0.2	0.6	1.5
50	0.2	1.2	0.2	5.0	0.2	25.8	1.2	0.6	47.9	0.5	0.6	0.5	2.9	0.5	2.3	1.3	0.2	0.6	1.8
60	0.2	0.9	0.2	4.8	0.2	26.0	0.8	1.1	47.6	0.6	0.5	0.6	1.6	1.7	4.7	1.2	0.2	0.6	1.4
70	0.3	1.3	0.3	5.2	0.2	30.6	0.7	0.8	39.0	0.4	0.3	0.5	3.2	0.5	6.0	2.0	0.3	0.9	1.9
CD	0.03	0.18	0.03	0.78	0.03	5.75	0.09	0.09	7.86	0.11	0.13	0.09	0.25	0.11	0.77	0.14	0.03	0.08	0.16
CV	11.73	12.34	11.33	10.61	13.58	13.73	7.74	7.66	11.90	12.70	15.04	15.07	7.69	6.43	13.73	7.02	11.43	10.35	7.86
RI Values	949	984	1030	1032	1034	1105	1130	1165	1186	1222	1240	1274	1340	1348	1376	1422	1456	1507	1650

Tab. 3. Chemical composition of the essential oil of *Ocimum basilicum* variety Kushmohak at different times of harvest during October 2012 to February, 2013

Harvest date	Chemical constituents of essential oil, %																		
	Camphene	β pinene	Limonene	1,8 Cineole	Ocimene	Linalool	Camphor	Terpinen-4-ol	Methyl chavicol	Bornyl acetate	Eugenol	Methyl Eugenol	βelemene	β Caryophyllene	α Humulene	γ Cadinene	Cadinol		
15	0.3	0.8	0.2	3.8	0.2	25.5	1.0	0.6	47.1	0.3	2.7	6.2	0.6	1.5	0.3	0.7	2.1		
30	0.3	0.6	0.2	3.5	0.2	25.4	0.8	0.5	47.7	0.2	2.8	4.5	0.9	2.5	0.4	0.7	1.8		
45	0.2	0.5	0.2	3.2	0.2	25.3	0.9	0.7	53.4	0.2	2.1	1.9	0.5	1.8	0.2	0.8	1.8		

	60	0.2	0.7	0.2	4.5	0.1	19.2	0.7	0.6	51.5	0.3	3.7	6.9	0.8	1.5	0.2	0.5	1.4
	75	0.1	0.9	0.2	3.6	0.2	20.8	1.1	1.0	34.7	1.2	5.4	10.1	0.8	1.0	0.2	0.5	1.4
CD		0.04	0.08	0.04	0.94	0.04	4.62	0.14	0.09	9.20	0.10	1.38	2.30	0.08	0.23	0.04	0.10	0.92
CV		13.09	7.91	16.31	17.25	16.91	13.66	10.26	9.61	13.49	16.65	28.44	26.59	8.05	9.47	10.51	11.04	37.13
	10	0.1	0.4	0.1	2.2	0.1	23.0	0.7	0.4	52.7	0.2	1.3	5.4	0.6	1.9	0.3	0.9	2.0
	20	0.2	0.9	0.2	4.9	0.2	16.1	0.8	0.7	52.9	0.2	3.2	7.4	0.6	1.3	0.3	0.6	1.6
	30	0.1	0.5	0.1	3.4	0.1	22.9	0.7	0.6	59.2	0.2	1.7	2.3	0.5	1.5	0.2	0.6	1.4
	40	0.2	1.2	0.2	5.8	0.2	24.9	0.8	0.8	51.2	0.3	2.2	3.2	0.5	1.2	0.2	0.5	1.2
	50	0.1	0.7	0.2	3.7	0.1	18.8	1.3	1.0	43.9	0.7	2.6	8.9	0.9	1.7	0.3	0.8	2.0
	60	0.2	1.2	0.2	5.2	0.1	21.9	1.0	0.6	45.0	0.7	2.3	8.3	0.8	1.2	0.2	0.6	1.7
	70	0.3	1.4	0.3	5.1	0.3	21.3	1.0	1.0	39.4	0.6	3.8	4.7	0.7	1.8	0.2	0.7	1.7
CD		0.03	0.15	0.05	0.78	0.04	7.78	0.13	0.11	11.65	0.12	0.78	1.94	0.12	0.21	0.03	0.15	0.37
CV		12.21	11.40	18.45	12.35	16.49	23.57	9.95	10.76	16.29	21.23	21.99	23.26	12.80	9.76	8.62	15.39	15.26
RI Values		949	984	1030	1032	1034	1105	1130	1165	1186	1274	1340	1376	1396	1422	1456	1501	1650

Tab. 4. Antimicrobial activity[§] of the essential oil of two varieties of *Ocimum basilicum* and one variety of *Ocimum tenuiflorum* at different harvest dates during December, 2012 to February, 2013

Variety	Harvest date	**Antibacterial activity**				**Antifungal activity**			
		Staphylococcus aureus		***Chromobacterium violaceum***		***Aspergillus niger***		***Candida albicans***	
		2 µl*	4 µl**	2 µl	4 µl	2 µl	4 µl	2 µl	4 µl
Ocimum basilicum variety Vikarsudha	10	14.5	16.5	22.5	40.0	0.0	0.0	5.5	12.5
	20	9.5	12.5	14.5	19.5	0.0	6.0	0.0	5.5
	30	7.5	13.0	12.0	22.0	0.0	7.0	0.0	5.5
	40	8.5	10.5	17.5	25.5	0.0	14.0	0.0	6.0
	50	6.5	11.5	17.5	25.5	0.0	5.5	0.0	6.0
	60	7.5	10.0	14.5	29.5	0.0	7.5	0.0	6.0
	70	11.0	13.0	12.0	19.0	0.0	16.5	0.0	7.0
Ocimum basilicum variety Kushmohak	10	11.0	12.5	21.0	30.0	0.0	7.0	5.5	12.5
	20	11.0	12.5	19.5	29.0	0.0	8.0	4.0	5.5
	30	7.5	11.5	16.0	21.5	0.0	7.0	5.5	11.5
	40	8.5	11.5	19.0	14.0	0.0	6.0	9.0	13.5
	50	8.5	10.0	20.5	24.5	0.0	8.0	7.5	11.0
	60	8.5	10.0	15.0	25.0	0.0	16.5	8.0	11.0
	70	9.0	11.0	14.5	30.5	0.0	16.0	9.0	12.0

Tab. 5. Antimicrobial activity§ of the essential oil components of *Ocimum* sps.

Components	Antibacterial activity				Antifungal activity			
	Staphylococcus aureus		***Chromobacterium violaceum***		***Aspergillus niger***		***Candida albicans***	
	2 µl	4 µl	2 µl	4 µl	2 µl	4 µl	2 µl	4 µl
Eugenol	23.0	25.0	24.0	28.5	20.0	30.0	20.0	35.0
Linalool	4.5	7.0	6.5	9.5				
Methyl Chavicol	18.0	30.0	27.0	34.0	14.0	18.0	9.0	9.0
Beta Caryophyllene	6.5	9.5	15.5	17.5				
Penicillin G	8.0	17.5	0.0	0.0	0.0	0.0	0.0	0.0
Streptomycin	0.0	0.0	12.5	15.0	0.0	0.0	0.0	0.0
Fluconazole	0.0	0.0	0.0	0.0	15.0	20.0	15.0	20.0

§: Inhibitory zone diameters in mm

Tab. 6. Fatty acid composition of fixed oil from seed

Variety	**Seed yield , kg/ha**	**Fatty acid composition of fixed oil from seed (%, w/w)**							
		16:0	**16:1**	**17:0**	**18:0**	**18:1**	**18:2**	**18:3**	**20:0**
Kushmohak	**230**	9.2	0.2	0.1	3.8	10.3	26.0	**49.3**	0.3
Vikarsudha	250	8.0	-	0.1	3.6	12.3	23.4	**52.4**	0.2