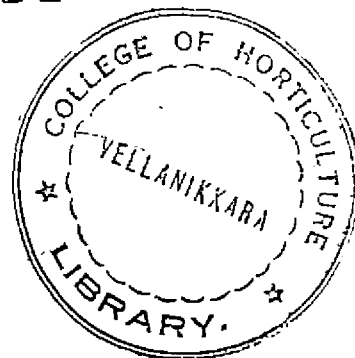


**QUALITY OF OIL OF CLOVE (Ocimum gratissimum  
Linn.) AS INFLUENCED BY STAGES OF  
HARVEST AND SHADE**



By

**REKHA R. PILLAI**

**THESIS**

Submitted in partial fulfilment of the  
requirement for the degree

**Master of Science in Agriculture**

Faculty of Agriculture  
Kerala Agricultural University

Department of Soil Science and Agricultural Chemistry

**COLLEGE OF HORTICULTURE**

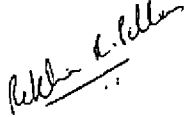
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## DECLARATION

I hereby declare that the thesis entitled "Quality of oil of clove (Ocimum gratissimum Linn.) as influenced by stages of harvest and shade" is a bonafide record of research work done by me during the course of research and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title of any other University or Society.

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CERTIFICATE

Certified that the thesis entitled "Quality of oil of clove (Ocimum gratissimum Linn.) as influenced by stages of harvest and shade" is a record of research work done independently by Ms.Rekha R. Pillai under my guidance and supervision and that it has not previously formed the basis for the award of any degree, fellowship or associateship to her.

Chinnamma.

N.P. CHINNAMMA  
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CERTIFICATE

We, the undersigned members of the Advisory Committee of Ms.Rekha R. Pillai, a candidate for the degree of Master of Science in Agriculture with major in Soil Science and Agricultural Chemistry, agree that the thesis entitled "Quality of oil of clocimum (Ocimum gratissimum Linn.) as influenced by stages of harvest and shade" may be submitted by Ms.Rekha R. Pillai, in partial fulfilment of the requirement for the degree.

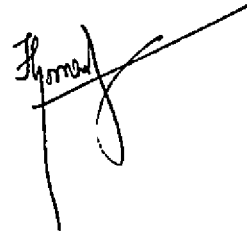
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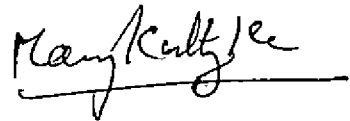
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## ACKNOWLEDGEMENT

As the author of the work, it gives me great pleasure to express my esteemed sense of gratitude to Dr.N.P. Chinnamma, Professor, Soil Science and Agricultural Chemistry, Chairperson of my Advisory Committee for her valuable and erudite guidance, never ending encouragement and perpetual support during the entire course of research work and I owe enormous debt to her for her immense help in the preparation of this manuscript.

I am fortunate in having Dr.A.I. Jose, Professor and Head of the Department of Soil Science and Agricultural Chemistry as member of my Advisory Committee. The valuable assistance and guidance received from him contributed largely to the early and successful completion of the programme.

I extend my sincere thanks and express my indebtedness to Dr.J. Thomas, Associate Professor and Head, Aromatic and Medicinal Plants Research Station, Odakkali, for all his help and suggestion during the course of this research work. In spite of his busy schedule, he had conscientiously gone through each and every bit of my work and offered searching comments and suggestions and I shall always owe a deep sense of gratitude for that.

I am obliged to Dr.K.C. Marykutty, Associate Professor, Department of Soil Science and Agricultural Chemistry for providing the required help during my work.

It is with pleasure that I express my heartfelt thanks to Dr.A.D. Damodaran, Director and Dr.J. Madhusudhana Rao,

Scientist, Regional Research Laboratory, C.S.I.R., Trivandrum for their estimable help and for providing all the necessary facilities for carrying out my research work. I also owe my sincere thanks to Mrs.Beena Simon and Mrs.Padmakumari in helping me out with GLC analysis.

My profound sense of gratitude is also due to Mr.K.P. Kuriakose, Assistant Professor and Mrs.Jayasree Sankar, Junior Assistant Professor, AMPRS, Odakkali for the sustained interest shown throughout the course of my research.

I am thankful to Dr.Saifuddin, Associate Professor, Department of Soil Science and Agricultural Chemistry, College of Agriculture, Vellayani for providing me the necessary facilities for carrying out the chemical analysis of my samples. I presume, this is the chance that I get to convey my thankfulness to Mr.Narayanan, S., my friend and classmate, who has helped me a lot in the timely completion of the analysis of my work.

The assistance and co-operation rendered to me by the Farm Assistants and Labourers of the AMPRS, Odakkali are appreciated very much. I thank each and every one of them profusely.

My gratefulness and personal obligation go without any reservation to each and every member of the Department of Soil Science and Agricultural Chemistry for extending all possible help in the proper conduct of my research work.

A deep sense of gratitude is owed to my friends for the innumerable acts of help and encouragement during the course.

I shall always remain beholden to my parents for their help, inspiration and tolerance throughout my course. At this juncture, I remember my brothers Ashok and Dina whose steadfast love and affection helped me a lot in the successful completion of this manuscript.

*Rekha R. Pillai*

REKHA R. PILLAI

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# *Introduction*

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## INTRODUCTION

Aromatic plants have long been exploited for the essential oils they produce in many parts of the tropical region. Essential oils are one of India's traditional items of trade in both exports and imports. They are the most vital constituents of spices and medicinal plants and form indispensable ingredients of many cosmetics, perfumery, soap and pharmaceutical products. In cosmetics essential oils are mainly responsible for fragrance and in spices they are the principles which contribute to flavour, easy digestibility and sometimes to durability of food. Essential oils in varying proportions directly enter into some perfumery compounds and flavouring essence.

India's exports show varying trends, exports of essential oils during 1987-'88 being valued at Rs.159.5 million as against Rs.236.9 million during 1986-'87 and Rs.156.6 million in 1985-'86 (Anon. 1989). At the same time India has also been importing huge quantities of essential oils like cinnamon bark and leaf oils, clove oil, nutmeg oil, pepper oil, etc. and aromatic chemicals like menthol, isoeugenol, linalool, geraniol, thymol, etc.

Clove oil and cinnamon oils are being imported into India mainly for their aroma chemical, eugenol.

According to the reports on the monthly statistics of the Foreign Trade of India 1985-'86 a quantity of 2,41,440 kg of clove

oil valued at 11,475.9 thousand rupees and 6490 kg of cinnamon leaf oil and 1220 kg of cinnamon bark oil valued at 418.9 thousand and 76.1 thousand rupees respectively were imported in India. So was 7186 kg of isoeugenol valued at 142.7 thousand rupees.

Eugenol is an important flavouring agent used in confectionary and food products. It was hitherto obtained from the clove buds of Eugenia caryophyllata containing 80-95 per cent eugenol and from leaves and bark of Cinnamomum zeylanicum containing only 50 to 80 per cent eugenol. These sources of eugenol are highly priced and therefore, the eugenol obtained from these sources is very costly (Sobti et al., 1979).

The working group on the status report to National Committee on Science and Technology has suggested that aromatic chemicals should be indigenously produced. From time immemorial Ocimum species have been recognised as potential source of aroma chemicals such as eugenol, citral, geraniol, linalool, etc.

Detailed investigations at the Regional Research Laboratory, Jammu, have shown that out of the many Ocimum species, O. sanctum and O. gratissimum containing good percentage of eugenol can serve as new potential sources of eugenol. A new strain of O. gratissimum which yields an oil similar to that of clove oil was evolved by Sobti et al. (1980) as a substitute to the eugenol yielding essential oils from trees, viz., cinnamon and clove and

was rightly named 'Clocimum'. This improved strain was found to contain on an average 70-80 per cent eugenol which makes it a very dependable alternative source of clove type oil having great potential not only for import substitution of clove oil of commerce but for export also.

Clocimum oil represents an excellent starting material for the isolation of eugenol, isoeugenol and for the synthesis of high grade vanillin, which together or individually find important use in pharmaceutical, perfumery and flavour industry. Besides eugenol, the oil contains 10 to 12 per cent myrcene which is a known flavouring agent having raw mango flavour, thus rendering the oil doubly useful to the flavour industry.

Considering the high cost of production of clove oil, if a strain of Ocimum spp. rich in eugenol suitable to Kerala home-stead conditions is identified, this probably can be used as an alternative source of eugenol due to its relatively easy cultivation, distillation and comparable yield. Clocimum was thus introduced to the Aromatic and Medicinal Plants Research Station (AMPRS), Odakkali during 1981-'82 and initial studies have revealed that it could be successfully cultivated under the climatic conditions of Kerala.

As the agronomic practices for clocimum were not standardised under Kerala conditions, two field experiments were laid out



simultaneously at the AMPRS, Odakkali with the following prime objectives.

1) To study the variations in quantity and quality of cocimium oil obtained from different intervals of harvest of the plant.

2) To study the changes in the yield and composition of oil under varying shade levels so as to decide the possibility of growing it on a commercial scale in the coconut gardens of Kerala.

3) To study the plant nutrient content and its uptake at different harvest intervals and under varying levels of shade.

# *Review of Literature*

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## REVIEW OF LITERATURE

Most of the available literature relating to the influence of stage of harvest of the crop and shade on oil yield, physico-chemical properties of oil, composition of the plant and uptake of nutrients relevant to the study are presented in this chapter.

### 1. Domestication of Ocimum gratissimum Linn. var. Clocimum

In a programme of finding out a new and cheap substitute for clove oil rich in eugenol, an extensive screening of Ocimum species collected from different parts of the world was carried out by Sobti et al. (1980) at the Regional Research Laboratory (RRL), Jammu. Well planned crossings of selected line of O. gratissimum evolved a strain with desirable oil composition similar to that of clove oil and therefore was named 'Clocimum' meaning clove scented Ocimum yielding on an average 75 per cent eugenol.

Clocimum was found to come up well under the agro-climatic conditions of Kerala having good rejuvenation capacity and it can definitely be an alternative and profitable source for eugenol (Kurian et al., 1984).

### 2. Soil and climate

An equitable annual rainfall of 85-130 cm is reported to be the ideal for the plant. It is usually raised as a rainfed crop. However, water logging should be avoided. It can be grown in

varied types of soil but well drained sandy loam soil is best suited for its growth (Balyan et al., 1982).

### 3. Yield attributes and yield

#### 3.1. Biometric parameters

In an experiment to study the inheritance pattern of certain phenolic and sesquiterpenic constituents of essential oils in the interspecific hybrids of Ocimum species, Sobti et al. (1978) revealed that O. gratissimum harvested just after 10-15 days of flowering had a plant height varying from 150 to 300 cm with a mean height of 240 cm. The size of the leaf blade varied from 5.5 x 2.5 cm<sup>2</sup> to 14.5 x 6.3 cm<sup>2</sup> with a mean value of 9.8 x 4.4 cm<sup>2</sup>. A quick and non-destructive formula,  $L \times W \times 0.612$  for finding out the leaf area of Ocimum has been reported (Balyan, 1981) and according to which the calculated leaf area of Ocimum plucked randomly at various growth stages was 18.781 cm<sup>2</sup>.

An experiment conducted by Trivedi et al. (1981) to study the performance of different varieties of Ocimum gratissimum (E.C. 11281, E.C.111091 and E.C.110959) harvested in the fifth month after planting in the Malwa region of Madhya Pradesh revealed that the plant height (cm), average number of tillers, length of inflorescence (cm) and length of leaves (cm) ranged from 67.6 - 71.2, 48.7 - 62.0, 6.6 - 21.2 and 3.3 - 4.9 respectively.

The average plant height and number of branches of O. gratissimum harvested at 60 days interval, four times in a year varied from 63.3 to 98.2 cm and from 18.2 to 57.3 respectively in the first year, both the parameters being the highest in the fourth cutting (Choudhury and Bordoloi, 1984).

Kurian et al. (1984) had shown that clocimum grown under Kerala conditions had 17.5, 18.6 and 16.7 number of branches and plant heights of 82, 119.4 and 94.7 cm at full flowering stage during three harvest seasons (July, October and January).

Choudhury et al. (1986) found that the mean plant height at different harvests of O. gratissimum in the first year ranged from 74.8 to 94.1 cm. The average number of branches per plant varied from 30.8 to 57.6.

In another experiment conducted at Jorhat, Assam to study the influence of micronutrients and harvesting time on foliage and oil quality of O. gratissimum, Choudhury et al. (1986) reported that the average height of plants showed an increasing trend from the first harvest (60.3 cm) recorded in June 1982 to the fourth harvest (114.5 cm) recorded in April 1983. The corresponding number of branches were 30.8 and 57.6 respectively.

From the above reference, it can be seen that the plant height, number of branches, length of inflorescence, and leaf area vary under different agro-climatic conditions and ranges from 60.3-300 cm, 16.7 - 57.6, 6.6 - 21.2 cm and 13.75 - 91.35 cm<sup>2</sup> respectively.

### 3.2. Yield of herbage and oil

In a study conducted to find out a new and cheap substitute for clove oil Sobti et al. (1980) reported that O. gratissimum (Race No.1), O. gratissimum (Race No.2) and the improved strain 'Clocimum' produced herbage yields of 250, 450 and 700 g/plant in one cutting and the corresponding oil yield on fresh weight basis were 0.15, 0.08 and 0.5 g respectively.

Trivedi et al. (1981) observed that the fresh herbage yield of O. gratissimum (E.C.112811), O. gratissimum (E.C.111091) and O. gratissimum (E.C.110959) grown under Malwa conditions of Madhya Pradesh were 643.9, 1025.3 and 383.7 g/plant respectively and the corresponding oil yields were 29.13, 63.47 and 27.73 l ha<sup>-1</sup>. The crop was harvested in the fourth month after transplanting. A fresh herb yield of 40 t ha<sup>-1</sup> year<sup>-1</sup> and oil yield of 160 kg ha<sup>-1</sup> for the improved strain of O. gratissimum (RRL-08) were observed from the studies conducted at RRL, Jammu (Sobti and Pushpan-gadan, 1982).

Kurian et al. (1984) found that clocimum harvested at full flowering stage gave herbage yields of only 1.3, 7.3 and 3.2 t ha<sup>-1</sup> and oil yields of 9.1, 40.5 and 15.9 kg ha<sup>-1</sup> respectively in the first (July), second (October) and third (January) harvest respectively.

According to Choudhury and Bordoloi (1986) four cuttings per year could be taken in O. gratissimum and the highest pooled yield of herb for two years ( $71.1 \text{ t ha}^{-1}$ ) and oil yield ( $387.2 \text{ kg ha}^{-1}$ ) were obtained when sown in the month of May followed by the sowing in April ( $69.1 \text{ t ha}^{-1}$  and  $376.1 \text{ kg ha}^{-1}$ ).

In the same year Choudhury et al. (1986) reported from another study that the herb yield and oil yield per plant of O. gratissimum varied between 166 and 213 g and from 0.95 to 1.46 g both the parameters being the highest in the third harvest among the four harvesting times taken in the year 1982-83.

From the review of the work done it can be seen that the herbage yield in O. gratissimum varied between  $39.8$  and  $74.0 \text{ t ha}^{-1} \text{ year}^{-1}$  and the oil yields between  $50.25$  and  $160.0 \text{ kg ha}^{-1} \text{ year}^{-1}$ . The number of harvests obtained in a year also showed wide variation due to the differences in the agro-climatic conditions and it was found to vary from 2 to 4.

### 3.2.1. Effect of stage of harvest on yield of herbage and oil

The optimum harvesting time in O. gratissimum was 10-15 days after flowering and the herbage yield per plant in one cutting was 575 g (Sobti et al., 1978).

Balyan et al. (1982) based on a study conducted to find out the optimum stage of harvest of the crop at flower initiation, full flowering and seed setting stages reported that maximum herb

yield of 590 q ha<sup>-1</sup> was obtained at flower initiation which was observed to decrease with increasing maturity of the crop. The herb yield obtained at full flowering and seed setting stages were 540 and 502 q ha<sup>-1</sup> respectively.

### 3.3. Oil content

The essential oil content of O. gratissimum var. Suavis Hook was 0.5 per cent on fresh weight basis, while the oil content on dry weight was found to vary between 1.8 to 2.0 per cent (Yeh, 1960 and Sobti et al., 1978).

The two parental strains of clocimum viz., O. gratissimum (Race No.1), O. gratissimum (Race No.2) and clocimum were found to yield 0.15, 0.08 and 0.5 per cent essential oil respectively on fresh weight basis (Sobti et al., 1980).

In a study to assess the performance of certain Ocimum species with respect to oil content under Malwa conditions of Madhya Pradesh at Indore, Trivedi et al. (1981) found that the oil content on fresh weight basis in the leaf, inflorescence and whole plant of O. gratissimum (E.C.112811) was 0.17, 0.13 and 0.068 per cent, of O. gratissimum (E.C.111091), 0.22, 0.23 and 0.093 per cent and of O. gratissimum (E.C.110959), 0.18, 0.18 and 0.108 per cent respectively.

Choudhury and Bordoloi (1984) had shown that the highest oil content in O. gratissimum was associated with the second cutting



among the four cuttings taken in the first and second years (0.64 and 0.63 per cent respectively). The least oil content was obtained in the fourth cutting of both the years (0.29 and 0.45 per cent respectively).

The oil recovery in clocimum grown under Odakkali conditions was found to be maximum for the first harvest taken in July (0.7 per cent) and the content went down in the next two harvests taken in October and January (0.55 and 0.5 per cent respectively) on fresh weight basis (Kurian et al., 1984).

Oil content in O. gratissimum (2 years) varying between 0.255 and 0.619 per cent on fresh weight basis has also been reported (Choudhury et al., 1986). All the harvests were made at 60 days interval.

### 3.3.1. Effect of stage of harvest on oil content

Based on the studies conducted on O. gratissimum in the Nikitin Botanical Garden, Knishevetskaya (1940) reported that the time of mass blossoming is the optimum time of harvest as the volatile oil is contained principally in the leaves and blossom.

According to Balyan et al. (1982) the oil content in clocimum was maximum at flower initiation and seed setting stages (0.66 and 0.65 per cent respectively) and the oil content went down to 0.53 per cent at full flowering stage. They were of the opinion that the harvesting of clocimum should be done without

including the woody stem portion which contained only negligible oil (0.05 to 0.07 per cent), almost all oil being stored in the leaves.

Review of the work done so far indicated that the oil recovery in O. gratissimum varied between 0.25 to 0.7 per cent on fresh weight basis the maximum content being recorded at flower initiation stages which decreased with increasing maturity of the crop.

#### 3.4. Physico-chemical properties of oil

##### 3.4.1. Physical properties of oil

Yeh (1960) reported that the essential oil of O. gratissimum var. Suavis Hook had density at 20°C ( $d^{20}$ ) 0.9989, refractive index at 20°C ( $n^{20}$ ) 1.5254 and optical rotation ( $L_D$ ) -17.8°.

O. gratissimum grown at Bangalore from seeds imported from U.S.S.R. had slightly varying values, specific gravity 0.9395, optical rotation -18° 1', refractive index 1.5132, saponification value 15.71 (Anon. 1966).

The oil of clocimum procured from Kerala were found to have refractive index 1.5268, specific gravity of 1.0067 at 30°C and soluble in 1.5 vols. of 70 per cent alcohol. The colour of the oil was observed to have more similarity to clove oil but with regard to other properties it had more resemblance to cinnamon leaf oil (Kurian et al., 1984).

### 3.4.2. Chemical composition of oil

Chiris (1929) described that oils of O. gratissimum Linn. distilled in Madagascar and the Comoro Islands contained ocimene 12.0%, polyterpene compounds and cadinene 3.0%, eugenol 89.0% and other phenols with an odour of cresol and guaiacol in traces.

The other oils of O. gratissimum L. distilled in Madagascar and the Comoros were found to have the following constituents in the oil (%): d- $\alpha$ -pinene (traces), ocimene (12.0), amyl alcohol (traces), linalool and terpineol (3.5 - 3.8), eugenol (62.5), other phenols (traces), methyl chavicol (small quantities), sesquiterpenes, mainly strongly laevorotatory cadinene (15.0), polyterpenes (3.0) (Glichitch and Naves, 1933).

The gas chromatographic analysis of the essential oil of O. gratissimum var. Suavis Hook was found to contain eugenol 62%, ocimene 18%, d-L phellandrene + L - cadinene 3% and L - perillyl alcohol 4% (Yeh, 1960), whereas O. gratissimum grown at Bangalore from seeds imported from U.S.S.R. gave a dark brown oil with the odour of cloves. It contained 61.8% eugenol, ocimene 15% and an unidentified alcohol 10% (Anon. 1966).

The chromatographic analysis of the leaf oil of O. gratissimum indigenous to Nigeria by Sainsbury and Sofowora (1971) however revealed more constituents (%):  $\alpha$ -pinene (2.6), camphene (4.0),  $\beta$ -pinene (0.6),  $\alpha$ -terpinene:  $\Delta^3$  carene (4.1), myrcene (1.4),

l, 8 cineole (1.1),  $\alpha$  - terpinene (6.2),  $p$  - cymene (16.2), limonene (1.8), camphor (0.6), linalool (0.2),  $\alpha$  - terpineol (2.4), thymol (47.6), methyl eugenol (1.7), methyl isoeugenol (trace), caryophyllene (2.1), humulene (0.5),  $\beta$  - selinene (1.6), longifolina (3.0), clovene (trace). Oil from the flowers were found to have essentially the same composition except that the proportion of camphene was reduced.

Sobti et al. (1978) reported that O. gratissimum cultivated at the Experimental Medicinal Plants Garden of the RRL, Jammu contained eugenol (70%), isoeugenol (13.9%), 4-terpineol +  $p$  - cymene (8.9%) as the major constituents in the oil when harvested after 10-15 days of flowering.

The improved strain of clocimum-grown under Jammu conditions had an average of 75 per cent eugenol content as compared to the 45 to 65 per cent in the original races of O. gratissimum (Race No.1) and 50-80 per cent in O. gratissimum (Race No.2) (Sobti et al., 1980).

Cheng et al. (1983) studied the composition of leaf, stem and flower oil from O. gratissimum L. grown in Taiwan. They could identify 26 compounds of which the major compounds reported were:

<u>Compounds</u>	<u>Leaf</u>	<u>Stem</u>	<u>Flower</u>
Eugenol	84.87	17.60	46.11
1,8 - cineole	2.93	40.20	23.04
Caryophyllene	3.19	4.18	4.40
∞ - Terpinyl acetate	2.18	2.52	6.34
4 - Terpineol	0.50	2.20	0.85
Myrcene	0.02	1.57	-
Limonene	0.02	6.49	-
Ocimene	0.03	1.48	0.03
Methyl eugenol	0.61	1.61	-

Choudhury and Bordoloi (1984) had shown that the content of eugenol in the essential oil of O. gratissimum grown at RRL, Jorhat, Assam was maximum (78.1%) in the first cutting of the first year and then progressively decreased upto the fourth cutting (61.4%). In the second year eugenol percentage was highest in the 2nd cutting (72.7%) which again decreased (61.4%) at the fourth cutting. The oil of clocimum grown under Odakkali conditions and harvested at full flowering stage was reported to contain 70 per cent eugenol and commented to be as good as that produced at Jammu by the RRL chemists (Kurian et al., 1984).

Gas liquid chromatographic analysis of the essential oils of O. gratissimum (2n = 40) harvested at the flowering stage by Khosla et al. (1985) showed the presence of 20 compounds of which

only fifteen could be identified. The essential oil analysis revealed eugenol as the major component (68.14%) followed by isoeugenol (13.88%) and myrcene (8.87%). The compounds identified were as follows:

<u>Compounds identified</u>	<u>Oil percentage (MFB)*</u>
α-pinene	0.21
Limonene	0.35
Phellandrene	0.62
Myrcene	8.87
4-terpineol	1.55
α-terpineol	0.51
Carveol	0.43
Carvone	1.19
Geranyl acetate	1.33
Caryophyllene	0.16
Eugenol	68.14
Isoeugenol	13.88
Methyl eugenol	1.74
Methyl isoeugenol	0.93
Caryophyllene-oxide	0.89

\*MFB = Moisture free basis

However oil of clove (O. gratissimum RRL-08) obtained from RRL, Jammu and reported to contain 70-75 per cent eugenol

and 10-15% myrcene on further investigation (GC-MS analysis) by Krishnamoorthy (1985) revealed only 60% eugenol and the major monoterpene hydrocarbon was identified as  $\beta$  - Ocimene and not myrcene as reported with strong herbal and terpenic by-odour and subdued spicy impact.

Choudhury et al. (1986) reported that O. gratissimum harvested during the warmer period of the year (June to October) recorded a higher eugenol per cent in the oil. Out of the four harvests in a year, eugenol content increased from the first harvest to second harvest and declined thereafter in both the years (1981-'83). The eugenol content ranged from 59.1 to 75.2 per cent.

Maheswari et al. (1988) could identify 40 compounds in clove oil covering more than 95 per cent composition of the oil. Initial GC studies revealed that the oil contained mainly eugenol (70-80%), and myrcene (10-15%). Further investigations by detailed GC/MS studies revealed the presence of eugenol (76.24%), geranyl acetate +  $\delta$  -cadinene (3.11%),  $\beta$ -caryophyllene (2.64%),  $\gamma$ -cadinene (1.76%);  $\beta$ -elemene (1.49%),  $\alpha$ -copaene (1.55%), tert-amorphol (1.25%), caryophyllene 4,5 - epoxide (1.10%) and vanillin (0.90%) in the essential oil. The other compounds identified in the oil had contents lower than 0.5 per cent.

In conclusion, it was observed that the essential oil of O. gratissimum exhibited refractive index, specific gravity, optical

rotation and solubility in 70% alcohol varying between 1.5132 - 1.5268, 0.9395 - 1.0067,  $-17.8^{\circ}$  to  $-18.1^{\circ}$  and 1.3 - 1.5 vols. respectively. As regards the chemical composition eugenol, isoeugenol, 4-terpineol, methyl eugenol, myrcene,  $\beta$ -caryophyllene have been identified as the major components. The eugenol content varied between 60-80 per cent, whereas the content of  $\beta$ -caryophyllene, myrcene, 4-terpineol, varied from 0.16-2.64, 0.02-15 and 0.5-1.55 per cent respectively. Besides the above components thirty other components have so far been identified in clocimum oil.

### 3.4.3. Physical properties of major components identified in clocimum oil

Perkin (1904) observed that the density of limonene ( $D_4^{20.84}$ ) was 1.4727 while the density, refractive index and optical activity at  $20^{\circ}\text{C}$  for  $\gamma$ -cadinene were 0.9125, 1.5075 and  $148^{\circ}$  respectively. Goulding and Roberts (1914) reported values of  $D^{15}$  0.8047,  $n_D^{20}$  1.4722 for myrcene. Ruzicka et al. (1939) opined that the chemical isolate eugenol had a specific gravity of 1.068, density at  $25^{\circ}\text{C}$ , 1.0620 and refractive index of 1.5439 while  $\beta$ -caryophyllene had density and refractive index of 0.9052 and 1.5009 respectively. Other workers have reported similar values for the physical constants of eugenol ( $d_4^{20}$  1.0664,  $n_D^{20}$  1.5410),  $\beta$ -caryophyllene ( $n_D^{17}$  1.5009,  $d_4^{17}$  0.9052,  $(\alpha)_D^{15}$   $-5.2^{\circ}$ ), limonene ( $d_4^{20.85}$  0.8402,  $n_D$  1.4744), myrcene ( $n_D^{25}$  1.4661,  $d_{25}^{25}$  0.7959) and 4-terpineol ( $d_4^{20}$  0.934,  $n_D^{20}$  1.4818,  $(\alpha)_D^{20}$   $+ 92.45^{\circ}$ ) (Anon. 1976).



3.4.4. Changes in the chemical constituents of oil with stage of growth of plant in other related Ocimum species

Dey and Choudhuri (1981) reported that the different stages of the reproductive development of the plant O. sanctum had profound modulating influence on the different biosynthetic pathways of secondary chemicals like eugenol and caryophyllene. They observed that eugenol was highest with the onset of reproductive phase, slightly decreased later followed by a sharp fall with the appearance of maximum inflorescence remaining more or less stable thereafter. They also observed that the decrease in eugenol content was compensated by a concomittant increase in caryophyllene upto the appearance of maximum inflorescence.

Gas chromatography analysis of the essential oil of O. sanctum revealed that eugenol, the principle aroma chemical showed a relative stability between initiation of flower bud (52.49%) to full bloom (53.53%) period but showed a sharp decline from early seeding (49.29%) to seed maturity stage (36.11%). There was corresponding gains in caryophyllene and unidentified sesquiterpene contents of the oil (Pareek et al., 1982).

The gas chromatographic analysis of the oils from O. sanctum conducted by Dey and Choudhuri (1985) revealed that there was a negative correlation between eugenol and caryophyllene contents. They also found that the major components of O. sanctum viz. eugenol and caryophyllene varied in different months and the

quantitative variation of each component of the oil was interdependent of each other though they were chemically of diverse nature.

#### 4. Physico-chemical properties of clove and cinnamon oils which are the other major sources of eugenol

Gas chromatography analysis of the relative constituents of leaf, stem bark and root bark oils of cinnamon (Cinnamomum zeylanicum) grown in Sri Lanka revealed that all three possessed the same array of monoterpene hydrocarbons though in different proportions. The main constituents of leaf-, bark-, and root oils were identified as eugenol, cinnamaldehyde and camphor respectively (Wijesekera et al., 1974).

Lawrence (1977) proposed the following physico-chemical properties of the essential oil obtained from clove and cinnamon.

Oil	Specific gravity	Specific rotation	Refractive index	Solubility in aqueous ethanol	Assay
Clove bud	1.036-1.060	-1° 30' to 0°	1.527 to 1.537	1.2 vols. 70%	85-95% phenols by vol.
Clove leaf	1.038-1.068	-2° to 0°	1.531 to 1.535	1.2 vols. 70%	84-88% phenols by vol.
Clove stem	1.048-1.056	-1° 30' to 0°	1.534 to 1.538	1.2 vols. 70%	88-95% phenols by vol.
Cinnamon leaf	1.030-1.060	-2° to 11°	1.529 to 1.540	1.2 vols. 70%	80 phenols by vol.
Cinnamon bark	1.010-1.030	-2° to 0°	1.573 to 1.591	1.3 vols. 70%	55-78% aldehydes calculated as cinnamaldehyde

Rabha et al. (1980) observed that Cinnamomum zeylanicum and O. gratissimum which are some of the important essential oil yielding plants of commercial value of North Eastern India possessed 94.5 and 74.6 per cent eugenol in the essential oils.

According to the Central Institute of Medicinal and Aromatic Plants reports, the essential oil of clove leaf procured from Kerala and examined by gas chromatography was found to contain  $\alpha$ -pinene 0.08%,  $\beta$ -pinene 0.05%, limonene 0.09%, caryophyllene 0.68%, eugenol 87.20%, eugenol acetate 9.43%, eugenol methyl ether 1.10% and isoeugenol methyl ether 1.07% (Anon. 1984).

The cinnamon leaf oil procured again from Kerala was identified as a dark brown liquid with refractive index at 30°C varying between 1.5223 to 1.5305, specific gravity at 30°C varying between 1.0443 to 1.0680, solubility in 3 volumes of 70 per cent alcohol and eugenol content of not more than 90%. The clove leaf oil had the properties of being a colourless to pale yellow liquid with refractive index and specific gravity measured at 30°C varying between 1.5270 to 1.5310 and 1.0330 to 1.0430 respectively, optical rotation of 0 to -2° and eugenol content of not more than 84 to 88 per cent (Kurian et al., 1984).

Zacharia and Gopalam (1987) reviewed the studies on clove and cinnamon and reported the following physico-chemical properties and chemical composition of clove buds, stem and leaf oil and of Ceylon cinnamon leaf and bark oils.

Physico-chemical properties	Clove			Cinnamon	
	bud oil	stem oil	leaf oil	leaf oil	bark oil
Specific gravity	1.051	1.050	1.054	1.037	1.023-1.040
Optical rotation	-0° 32'	-0° 36'	-1° 20'	-1° 96' to -0° 40'	Slightly laevorotatory
Refractive index	1.5318	1.5352	1.5379	1.5288	1.581-1.691
Solubility in 70% alcohol	1 vol.	1 vol.	1 vol.	1.5 vol.	2 to 3 vol.
Eugenol content	91%	91%	88.5%	77.3 to 90.5%	4 to 10%
Chemical Composition	Eugenol Eugenol acetate Caryophyllene Caryophyllene oxide Methyl salicylate Valeraldehyde Furfuryl alcohol Methyl furfuryl alcohol Vanillin	Eugenol Eugenol acetate β-caryophyllene Furfural Methyl alcohol Methylamyl Naphthalene	Same as stem oil	Dipentene Phellandrene Pinene Linalool Geraniol Terpineol Eugenol Cinnamyl alcohol  Benzyl benzoate Cinnamanol Folial Caryophyllene	Furfural Phellandrene p-Cymene Benzaldehyde Cinnamalhyde Linalool Eugenol Caryophyllene  Cinnamic aldehyde

The major components of Indian clove bud, stem and leaf oils were also identified as eugenol, eugenol acetate and  $\beta$ -caryophyllene by Gopalakrishnan and Narayanan (1988).

In comparison with clove and cinnamon oils to which clocimum was found to have a lot of similarities it was observed that while the specific gravity of clove and cinnamon oils varied between 1.023 - 1.054, the specific gravity of clocimum oil was very much less (0.9395 - 1.0067). The refractive index of clocimum oil was also found to be less when compared to that of clove and cinnamon oils. However, the oil of clocimum exhibited a higher optical activity upto  $-18.1^\circ$  when compared to clove and cinnamon oils where the optical rotation varied between  $-0.32^\circ$  to  $-1.96^\circ$ . The solubility of all the three oils in 70 per cent alcohol fell within the same range 1.0 - 3 vols., though clocimum had more resemblance to clove oil in this property. Regarding eugenol content clove oil possessed maximum eugenol (88.5 - 91.0%) followed by cinnamon leaf oil. However, the eugenol content of clocimum closely matched that of cinnamon leaf oil.

##### 5. Nutrient content of the plant

Investigations conducted by Balyan et al. (1988) to study the effect of spacing on dry matter accumulation and uptake of N, P and K in clocimum revealed that the total N, P and K content of plants varied from 0.845 - 0.890, 0.414 - 0.470 and

1.050 - 1.125 per cent respectively. The total N, P and K uptakes were 61.66, 33.61 and 82.91 kg ha<sup>-1</sup> year<sup>-1</sup> respectively.

## 6. Effect of shade on yield attributes and yield of other shade tolerant plants

Preliminary studies conducted at the AMPRS, Odakkali, have shown that clocimum comes up well under shaded condition than under open conditions. No other literature is available on the effect of shade on the growth, yield, quality of oil, nutrient contents and its uptake by clocimum. So effect of shade on the yield and quality of oil, content and uptake of nutrients of a few other crops under open and shaded conditions are reviewed.

### 6.1. Biometric parameters

Positive influence of shading on plant height was reported in ginger (Aclan and Quisumbing, 1976), cowpea (Tarila et al., 1977), coleus and sweet potato (Bai and Nair, 1982) groundnut (George, 1982), winged bean (Sorenson, 1984) and cassava (Ramanujam et al., 1984 and Sreekumari et al., 1988).

Cooper (1969) observed that in the case of tomato, shading either decreased or had no effect on mean stem extension rate. In Mentha arvensis, Duriyaprapan and Britten (1982) failed to record any influence of shade on plant height.

In general, the shade effect on branching is found to be adverse. The response to shade on number of branches produced per plant is negative as reported in cowpea (Tarila et al., 1977) and rice (Kemp and Whingwiri, 1980). Duggar (1903) also elucidated that plants under shaded conditions exhibited reduced number of branches.

In apple, tomato and many horticultural plants Clark (1905) reported an increase in total leaf area with shading. Duriyaprapan and Britten (1982) also noticed increased leaf area development in shaded Mentha arvensis plants.

However, decreased shading or increased light intensity resulted in greater leaf area in clove (Beinhart, 1963).

## 6.2. Yield

Sunlight being the source of energy for plants for photosynthesis, the rate and subsequent dry matter accumulation in general are found to be adversely affected by shading. But in ginger, coffee, etc. positive influence was reported. Still in some other crops like pineapple, there was no appreciable decrease in dry matter accumulation even upto 75 per cent shading.

Positive influence of partial shading on yield was reported in ginger which gave as much yield as that under full sunlight (Aclan and Quisumbing, 1976; Bai, 1981). In crops like tomato,

tea, chilli and chickpea also partial shading was found beneficial.

However, high light intensity was found to improve blossom, pod number and seed yield in cowpea (Tarila et al., 1977), black gram (Leelavathi, 1979) and pulse crops (George, 1982). In turmeric rhizome yield was significantly higher in the open than under shade (Ramadasan and Satheesan, 1980). In coleus, the yield of tubers was unaffected by shading (Bai, 1981).

### 6.3. Effect of shade on oil content

Experiments on shaded and unshaded plants indicated that light favours formation of oil (Lubimenko and Norvikoff, 1914; Rabak, 1916). Balyan et al. (1982) reported that the crop clocimum came up well under partially shaded conditions in Jammu, though the oil content was slightly low under shaded conditions. However, preliminary studies conducted at the Aromatic and Medicinal Plants Research Station, Odakkali have shown high vegetative growth and yield for clocimum under shaded conditions. So the present study was undertaken to study the suitability and adaptability of clocimum in intercropping situations in coconut gardens of Kerala.

In general, even though shading increased vegetative growth of the plants, its influence on yield and yield attributes were found to be adverse. Plants vary in their response to shade.



#### 6.4. Effect of shade on nutrient content

In general, the mineral nutrient status of plants have been found to improve under shading as in the case of apple, cocoa, spinach and tea. Potassium contents were approximately doubled by shading as observed in some grass species (Myhr and Saebo, 1969), spinach (Cantiliffe, 1972), cowpea and groundnut (George, 1982). Bai (1981) reported that in all the plant components of the different crops viz., coleus, colocasia, sweet potato, turmeric and ginger, contents of N, P and K increased with increasing intensities of shade.

On the contrary, nitrogen content was positively related to illumination levels in soyabean plants (Trang and Giddens, 1980). But no distinct trend on nutrient contents was recorded in sweet potato and colocasia (Bai, 1981) and Dracaena sanderiana (Rodriguez et al., 1973).

## *Materials and Methods*

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## MATERIALS AND METHODS

Two separate field experiments were conducted simultaneously in the Aromatic and Medicinal Plants Research Station, Odakkali during 1989-'90. In these experiments the growth and yield characters of clove (Ocimum gratissimum Linn.) and its composition as well as the quality parameters of the oil at different stages of growth of the crop and under varying degrees of shade were studied in detail.

The station is situated at an elevation of 66 m above MSL with an average annual rainfall of 3600 mm with around 160 rainy days in a year.

The materials used and methods adopted in the course of these investigations are described below.

### 1. Soil analysis

The soil of the experimental site was lateritic clay loam with the following characteristics.

#### 1.1. Particle size distribution

Coarse sand	..	25.6 per cent
Fine sand	..	20.2 "
Silt	..	17.6
Clay	..	33.9

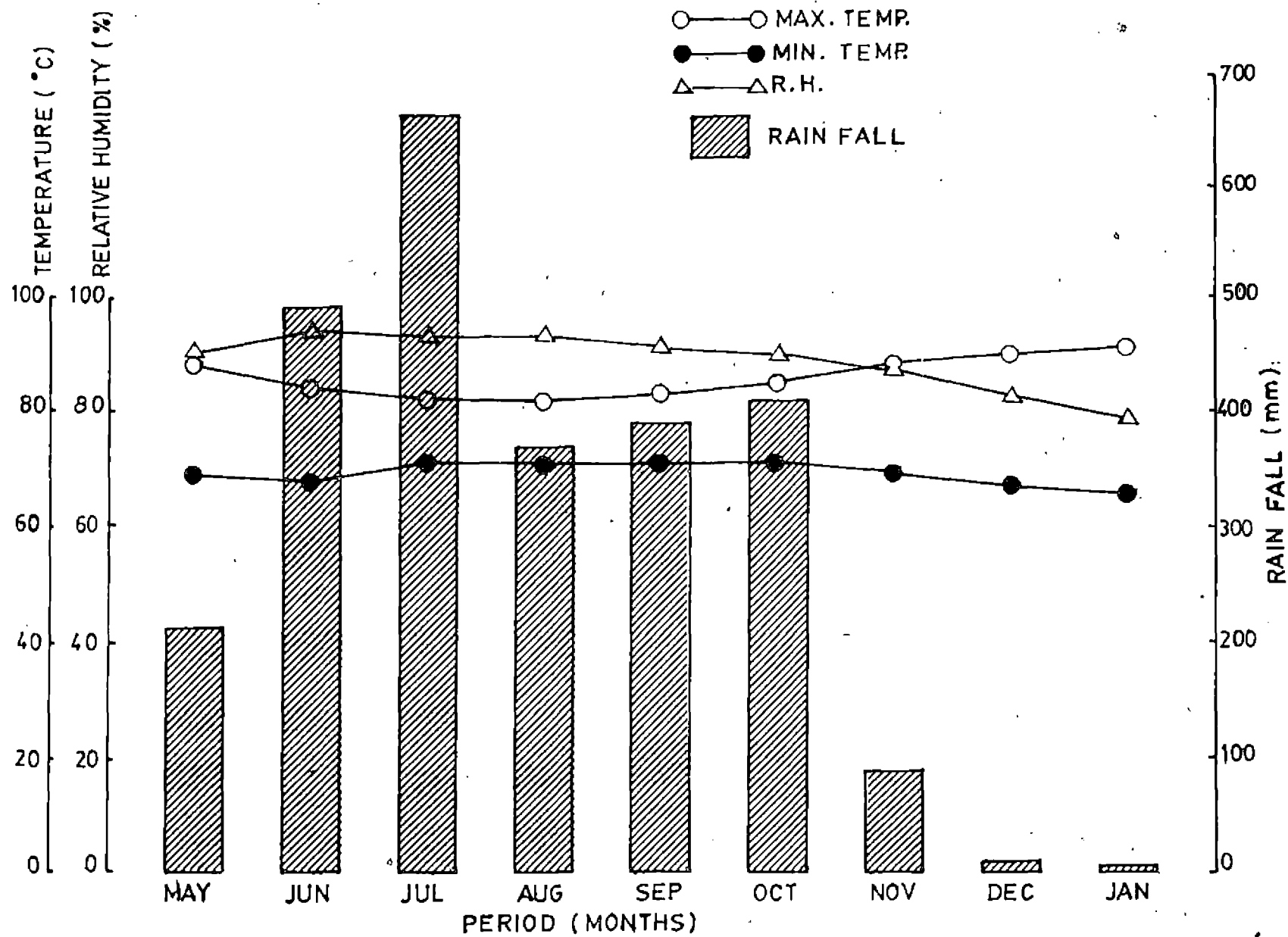


FIG.1- METEOROLOGICAL DATA DURING THE EXPERIMENTAL PERIOD OF 1989-'90



Plate 1. Clocinum plant in full bloom

## 1.2. Chemical characteristics

<u>Constituents</u>	<u>Contents</u>	<u>Rating</u>
Total N	0.102 per cent	
Total P <sub>2</sub> O <sub>5</sub>	0.082 "	
Total K <sub>2</sub> O	0.212 "	
Total CaO	0.208 "	
Total MgO	0.085 "	
Available N	274.4 kg ha <sup>-1</sup>	Medium
Available P <sub>2</sub> O <sub>5</sub>	3.36 kg ha <sup>-1</sup>	Low
Available K <sub>2</sub> O	61.6 kg ha <sup>-1</sup>	Low
Available Fe	245 ppm	
Exchangeable Mn	8 ppm	
Available Zn	4 ppm	
pH (Soil:Water ratio 1:2.5)	5.3	Acidic
Organic carbon	0.831 per cent	

## 2. Season and climate

The experiments were started in the month of May 1989 and continued upto February 1990. The meteorological data for the above period are presented in Fig. 1.

## 3. Planting material

Seeds of clocimum, a polycross hybrid strain developed at the Regional Research Laboratory, Jammu (Plate 1) and having

a germination percentage of 82 were used for the experiments. For the nursery, seeds were sown on the 12th May, 1989 in pots which contained loamy soil mixed with spent lemongrass compost. After sowing the seeds, a mixture of the lemongrass compost and soil was thinly spread over the seeds and then watered. BHC 10% was also sprinkled around the pots to prevent the attack of ants and termites. The seeds of clocimum germinated within 8-12 days of sowing which at 4-6 leaf stage were used for transplanting to the main field.

#### 4. Experimental details

##### 4.1. Trial No.1

An experiment was undertaken to study the changes in the quantity and quality of clocimum oil as influenced by the maturity of the crop and to arrive at the most suitable interval for harvest. The experiment was laid out in RBD with 4 replications and five harvesting intervals namely 60, 75, 90, 105 and 120 days as the treatments. After completing the first harvest in all the treatment plots, it was observed that the oil and eugenol content were higher at 60th day harvest interval which showed a tendency to decline thereafter. Therefore an additional observation was taken on the ratoon crop to assess the physico-chemical properties of the oil at 40 and 60 days.

#### 4.1.1. Preparatory cultivation

The experimental field was thoroughly prepared to get uniform condition and four blocks were formed each of 5 plots. Then the individual plots were once again dug and levelled. Spent lemongrass at the rate of  $5 \text{ t ha}^{-1}$  was applied before the final ploughing. Raised plots of  $5 \times 4 \text{ m}^2$  were prepared with an interspace of 1 m between plots. The N, P and K contents of spent lemongrass were 1.13, 0.088 and 1.01 per cent respectively.

#### 4.1.2. Fertilizers and fertilizer application

The fertilizers used in this experiment were urea (46 per cent N), single superphosphate (16 per cent  $\text{P}_2\text{O}_5$ ) and muriate of potash (60 per cent  $\text{K}_2\text{O}$ ). The fertilizers were applied at the rate of 60:60:60 kg N,  $\text{P}_2\text{O}_5$  and  $\text{K}_2\text{O}$ /ha respectively. Full doses of  $\text{P}_2\text{O}_5$  and  $\text{K}_2\text{O}$  and half dose of N were applied as basal dose. The remaining quantity of N was applied after the first uniform harvest in all plots.

#### 4.1.3. Planting

Healthy seedlings at 4-6 leaf stage were planted at the rate of two seedlings per hole. Planting of seedlings was done on 20th June, 1989 at a spacing of 50 x 50 cm. Gap filling was done after 3 weeks. The crop was raised under rainfed condition.



#### 4.1.4. Weeding

Weeds were removed before top dressing with N and earthing up was done twice during the course of the experiment. It was done in the months of September and November.

#### 4.1.5. Plant protection

The plant and soil were drenched with Emisan-6 at the rate of 0.5 per cent (80 l per plot) as the plants showed symptoms of bacterial wilt. This was done on 2nd September 1989. Another spraying with Streptocycline (200 ppm) was carried out on 15th September, 1989 to obtain immediate control of this disease.

#### 4.1.6. Harvest

The crop requires about 3 months for well establishment and if the crop is harvested before that period the mortality rate would be high. So a uniform harvest was made 90 days after transplanting with the onset of rains in September for all the treatments and thereafter the harvests were made at the specified intervals of 60, 75, 90, 105 and 120 days as per treatments.

The herbage was cut from all the plants in the plots except those left for collection of plant samples for chemical analysis. The fresh weight of the herbage was recorded immediately after harvest.

The plants tag-marked for chemical analysis were separately harvested initially before the plot-wise harvest.

The essential oil content was determined by bulk distillation conducted treatment wise in small stills of 25 kg capacity installed for the experimental purpose. Distillation was carried out for  $3\frac{1}{2}$  hours. The oil obtained after the distillations was further clarified to make it free of sediments and water drops before the quantity was measured. The oil from each plot was measured and kept in amber coloured bottles with tight fitting corks for physico-chemical analysis.

#### 4.2. Trial No.II

The objective of the second experiment was to study the changes in the quantity and quality of clove oil under varying degrees of shade. The experiment was laid out in RBD with 5 replications and 4 intensities of shade. The treatments were

##### Levels of shade

No shade	100 per cent light
Low shade	25 per cent shade
Medium shade	50 per cent shade
High shade	75 per cent shade

##### 4.2.1. Preparatory cultivation

The land was prepared as in trial I. Raised plots of 5 x 4 m<sup>2</sup> were prepared and the plots separated by distances of 1.5m.



Plate 2. General view of the shaded plots

#### 4.2.2. Fertilizer and fertilizer application

Was done as in trial I.

#### 4.2.3. Planting

Seedlings at 4-6 leaf stage were transplanted on 10th July at the rate of two seedlings per hole. They were planted at 50 cm spacing accommodating 80 plants per plot.

#### 4.2.4. Weeding

Weeding was carried out as done in trial I.

#### 4.2.5. Provision of shade

Artificial shading to the desired level was obtained by placing unplaited coconut leaves on erected pandals.

Pandals of size  $5 \times 4 \text{ m}^2$  were individually erected for each shade level by fixing wooden reapers on posts. Sufficient space (1.5 m) was provided between the treatments so that mutual shading of plots was avoided. Each pandal was covered on all sides with coconut leaves except for 60 cm from the ground level to avoid the direct entry of slant rays. An Aplab luxmeter was used for adjusting the shade intensities approximately to the desired levels. (There were, however, variations upto a maximum level of about 10 per cent at different locations within a pandal). Frequent checks were made throughout the course of the experiment to maintain the shade intensities to the desired level (Plate 2).

#### 4.2.6. Plant protection

Was carried out as in trial I.

#### 4.2.7. Harvests

The crop was harvested treatment-wise when 50 per cent of plants were in full bloom. Two harvests were obtained for the plants in the open since the crop flowered earlier under open condition. For all other treatments only one harvest was obtained. All the plants except those left for collection of plant samples were cut from the field at 10-15 cm height in the morning. Observations on the fresh herbage weight and the extraction of essential oil were carried out as in trial I.

### 5. Observations

#### 5.1. Biometric characters

For recording growth characters such as plant height, number of branches, number of flowered branches, leaf area, length of inflorescence and spread of the plant, 5 plants were marked out from each plot at random after leaving the border plants.

##### 5.1.1. Plant height

Height of the plant from ground level to the tip of the longest branch was measured (cm).

#### 5.1.2. Total number of branches

The total number of branches in the observation plants was counted and expressed in numbers.

#### 5.1.3. Flowered branches

The number of branches that had flowered at the time of harvest was recorded.

#### 5.1.4. Length of inflorescence

The length of the inflorescence was measured from the point of the inflorescence stalk union on the main branches to the tip of the inflorescence.

#### 5.1.5. Height at first branching

Height of the plant from ground level to the first branching point was measured (cm).

#### 5.1.6. Spread of the plant

Maximum horizontal extension of branches in the N-S and E-W direction was measured and multiplied to get a measure of spread of the plant ( $\text{cm}^2$ ).

#### 5.1.7. Leaf area

From each of the observation plants a leaf was selected and using the relationship between length and width and mean area of leaf a correction factor was worked out using the formula

$$\frac{EA}{L \times W}$$

Where EA is the area of leaf from leaf area meter

L is the maximum length of leaf

W is the maximum width of leaf

Then leaf area was calculated by multiplying the maximum length and width of leaves with the correction factor.

## 5.2. Yield characters

### 5.2.1. Herbage yield per hectare (Fresh weight basis)

The herbage yield from each plot was noted immediately after harvest and the yield per hectare calculated.

### 5.2.2. Dry matter yield

The samples from each harvest of tag-marked plants meant for chemical analysis of herbage were first sundried and then oven-dried at 80°C to constant weight. The dry matter yield from each plot was computed for each harvest using this dry matter percentage.

### 5.2.3. Oil yield

The oil yields in  $\text{kg ha}^{-1}$  were calculated from the yield of oil from each plot.

### 5.2.4. The essential oil content

The quantity of oil obtained from each treatment was used

for working out the oil content (volume of oil/weight of herbage) on fresh and dry weight basis of the herbage.

#### 5.2.5. Eugenol yield per hectare

Eugenol yield per hectare was calculated by multiplying eugenol percentage with oil yield per hectare.

### 5.3. Physico-chemical properties of the oil

The samples obtained from each treatment were examined for all important physical parameters by standard analytical procedures. The oil collected was analysed by GLC also.

#### 5.3.1. Physical properties

Physical properties such as specific gravity, refractive index, optical activity and solubility in 70 per cent alcohol were determined by the methods prescribed by ISI (Anon. 1974).

#### 5.3.2. Chemical constituents and chemical property

Eugenol percentage of the oil sample was determined by the method described by ISI (Anon. 1974).

Gas liquid chromatographic analysis of the oil obtained from all treatments was conducted using a Hewlett Packard 5840 gas chromatograph. The carrier gas used was nitrogen, flow rate 20-30 ml/min, column: OV-17, oven temperature 80°C and FID temperature 250°C or 270°C.



The compounds were identified by comparing their retention times with those of authentic reference samples. In the gas chromatograph used the percentage of eugenol and other constituents were recorded automatically.

#### 5.4. Analysis of soil samples

Particle size distribution of the soil was determined by the International Pipette Method (Piper, 1942). Total N was determined by the microkjeldahl method (Jackson, 1958) and available N by alkaline permanganate method (Subbiah and Asija, 1956).

Total  $P_2O_5$  and total  $K_2O$  in the soil were determined using standard procedures as outlined by Jackson (1958). Available  $P_2O_5$  was determined by Bray's No.1 method (Jackson, 1958) and available  $K_2O$  was extracted by neutral normal ammonium acetate and estimated by using EEL flame photometer. Total Ca and Mg were determined in diacid extract by EDTA titration method. For the determination of available micronutrients, the soil samples were extracted with DTPA extractant in the ratio of 1:2, shaken for 2 h and estimated with atomic absorption spectrophotometer (Lindsay and Norwell, 1978). Organic carbon was determined by Walkley and Black method (Piper, 1942) and the pH of the soil determined using a pH meter (Jackson, 1958).

### 5.5. Analysis of plant samples

The plant samples were dried in an oven at 80°C and ground in a Wiley mill.

The total N content of the samples was determined by the microkjeldahl method (Jackson, 1958).

Phosphorus, potassium, calcium and magnesium were determined in the diacid extract of the plant material. Phosphorus was determined by the vanado-molybdo phosphoric yellow colour method (Jackson, 1958). Potassium was determined by using EEL flame photometer and calcium and magnesium were estimated in the extract by the versenate titration method as given by Cheng and Bray (1951). Micronutrient elements in the plant material viz., Fe, Mn and Zn were determined in the diacid extract using an atomic absorption spectrophotometer.

### 5.6. Uptake of fertiliser nutrients

The total uptakes of N, P, K, Ca, Mg and micronutrients by the plant were calculated at different stages of growth and at different levels of shade by multiplying the dry matter of the crops with the respective nutrient content and expressed in  $\text{kg ha}^{-1}$ .

### 5.7. Statistical analysis

The data recorded for different characters were compiled and tabulated in proper form and were subjected to analysis of variance (Panse and Sukhatme, 1967).

## *Results and Discussion*

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## RESULTS AND DISCUSSION

The results obtained on the effect of intervals of harvest and shade on the yield and quality of oil of clove are presented and discussed in this chapter.

### 1. Experiment to study the effect of harvest intervals on yield and quality of oil, nutrient content and uptake

#### 1.1. Effect of intervals of harvest on biometric characters

Data presented in Table 1 represent the biometric characters recorded during the course of investigation.

There was no significant difference in the height of the plants due to the variations in the intervals of harvest and the values varied from 84.90 to 95.67 cm. However, the relative increase in height was greater during the second harvest interval. This may be due to the fact that the plant attained its full physiological growth only at this harvest interval. Kurian et al. (1984) reported plant height at full flowering stage for clove to be between 82.0 and 119.4 cm which is in agreement with the values obtained in the present study. However, the height of plants under Jammu conditions varied between 150 to 300 cm (Sobti et al., 1978) which is much higher than the values recorded in the present study.

Table 1. Effect of harvest intervals on the biometric characters

Harvest intervals	Plant height (cm)	Total number of branches	Number of flowered branches	Length of inflorescence (cm)	Height at first branching (cm)	Spread (cm <sup>2</sup> )	Leaf area (cm <sup>2</sup> )
60 days	89.52	14.90	5.85	10.35	1.20	3940.45	27.25
75 days	95.67	15.65	13.20	14.37	2.62	5400.40	30.54
90 days	91.30	14.60	10.55	15.35	1.30	5744.40	30.54
105 days	93.15	15.45	11.10	11.65	1.47	5593.20	15.56
120 days	84.90	13.75	10.75	10.97	1.75	3930.40	11.12
SEm $\pm$	4.66	1.58	1.52	1.11	0.53	523.60	2.14
CD (0.05)	NS	NS	4.582	3.409	NS	NS	6.588

NS - Not significant

There was also not much variation in the total number of branches due to the treatments. The maximum number of branches was observed at 75 days harvest interval which also indicated that the plant attained its prime period of growth at this harvest interval. The number of branches obtained in the present study is in conformity with the results reported by Kurian et al. (1984) under Kerala conditions.

Harvest of the crop at different intervals resulted in significant difference in the number of flowered branches. The number of flowered branches at the 60th day harvesting interval was significantly lower than all the other treatments. The number of flowering shoots almost increased by more than two fold when the harvest interval was increased from 60 to 75 days cutting interval. Maximum number of flowered branches was recorded by the crop harvested at 75 days interval and it was on par with all other longer harvest intervals namely 90, 105 and 120 days. This increase in the number of flowered branches at 75 days harvest interval might probably be due to the fact that this harvest interval synchronised with the maximum flowering stage of the plant and the harvesting interval of 60 days might be the early flowering stage of the crop.

The length of inflorescence under the influence of different harvest intervals ranged from 10.35 to 15.35 cm. The length of

inflorescence increased with increasing maturity of the crop upto the 90th day. The maximum length recorded at 90 days harvest interval was significantly superior to that recorded at 60, 105 and 120 days harvest intervals and was on par with that recorded at 75 days. Hence it is assumed that maximum flower development and elongation of inflorescence stalk took place at this interval (75 days) which extended over to the early seed setting stage (90 days cutting interval).

There was no difference in height due to the treatments at the first branching and the values varied from 1.20 cm of the 60th day to 2.62 cm of the 75th day cutting interval.

Spread of the plants was also not influenced by the variation in the harvest intervals and the values varied from 3930.40 to 5744.40 cm<sup>2</sup>. There was a progressive increase in plant spread upto 90th day with increase in the growth of the plant after which the plant spread decreased. This apparent decrease in spread with increase in harvest intervals after 90 days may be due to the decrease in growth rate and the destruction of unharvested inflorescence flower heads after they dried up.

Maximum leaf area of 30.54 cm<sup>2</sup> was exhibited at 75 days interval of harvest and it was on par with 60 days harvest interval. But with further increase in the maturity of the plant, the leaf area decreased. It can be presumably due to the maximum

photosynthesis and rate of growth occurring upto 75 days i.e., the maximum flowering stage as the crop had already entered the reproductive stage. The decline in leaf area at later stages may be attributed to the diversion of photosynthate for seed development which had so far been utilized for the production of leaves coinciding with the onset of leaf senescence.

The results thus indicated that the harvest interval of 60 days matched with the early flowering stage and that of 75 days with full flowering stage in which most of the biometric parameters like plant height, number of flowered branches and length of inflorescence were observed to be maximum. The cutting intervals of 90, 105 and 120 days were also seen to coincide with the early seeding, late seeding and seed ripening stages of the plant.

#### 1.2. Effect of intervals of harvest on herbage yield, oil content and oil yield

The herbage yield, oil yield and oil content as influenced by the treatments are presented in Table 2 and Fig. 2.

##### 1.2.1. Herbage yield

The yield of herbage varied from 8.43 to 12.94 and from 5.02 to 8.42 t ha<sup>-1</sup> on fresh and dry weight basis, respectively. Herbage yield was not influenced significantly due to treatments. The



Table 2. Effect of harvest intervals on herbage yield, oil content and oil yield

Harvest interval	Herbage yield, t ha <sup>-1</sup>		Essential oil content, per cent		Essential oil yield, kg ha <sup>-1</sup>
	FWB	DWB	FWB	DWB	
60 days	12.94	8.42	0.58	0.90	75.18
75 days	12.93	8.06	0.54	0.84	69.76
90 days	10.12	7.70	0.55	0.73	56.38
105 days	11.90	8.15	0.23	0.33	27.85
120 days	8.43	5.02	0.21	0.36	17.67
SEm ±	1.64	1.03	0.06	0.05	9.10
CD (0.05)	NS	NS	0.181	0.174	27.805

FWB = Fresh weight basis

DWB = Dry weight basis

NS = Not significant

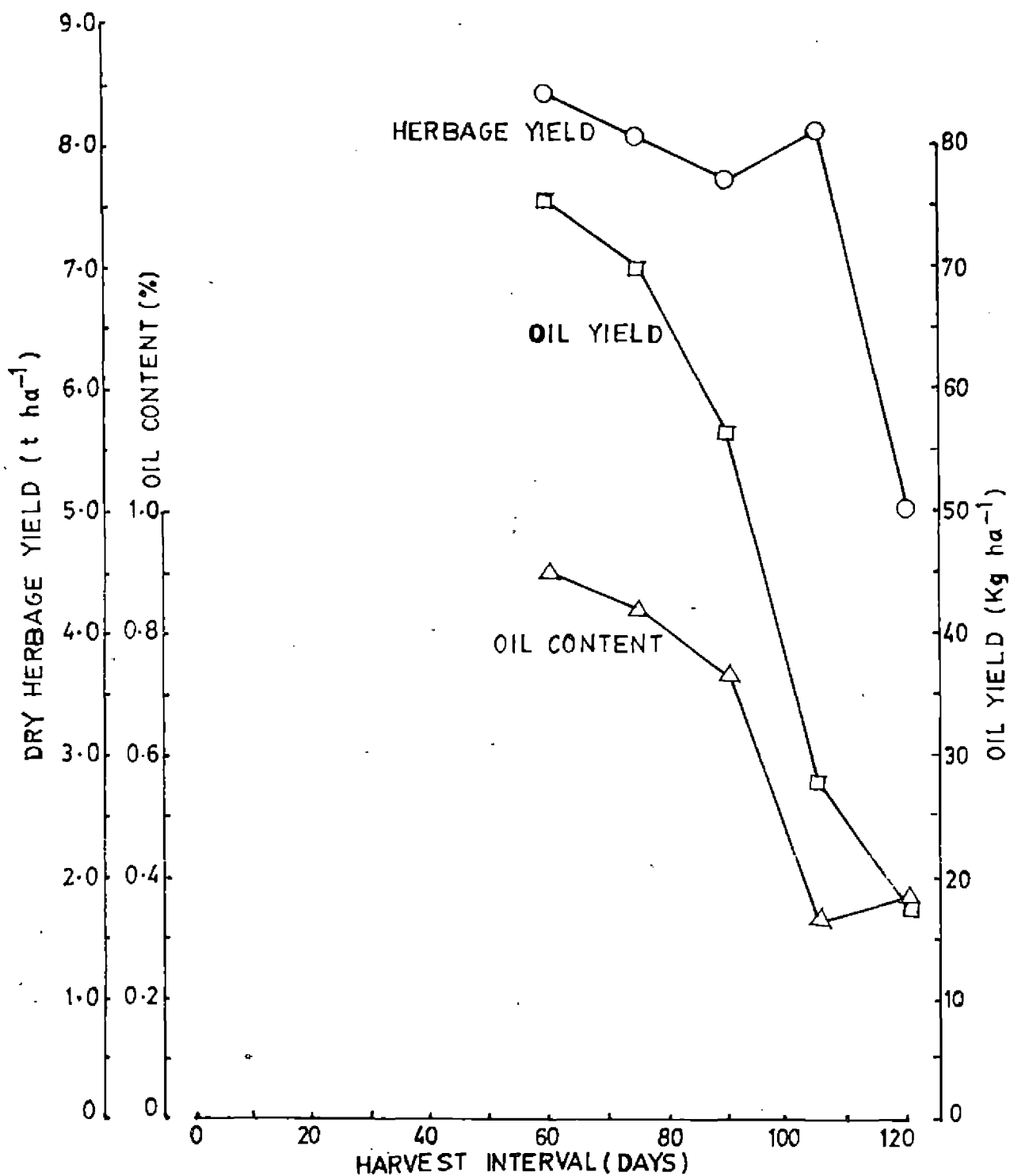


FIG. 2. EFFECT OF HARVEST INTERVALS ON HERBAGE YIELD, OIL CONTENT AND OIL YIELD

highest yield was registered at the shortest interval of harvest tried and a decreasing trend was observed with increase in the interval of harvest. Similar results have been reported by Balyan et al. (1982). One of the reasons for this may be that the maximum growth was attained in 60 to 75 days and as the growth advanced further, senescence had slowly set in and the assimilate produced was diverted for seed set (Cock and Yoshida, 1972 and Evans et al., 1973). The least herbage yield of  $5.02 \text{ t ha}^{-1}$  on dry weight basis was recorded at the last harvesting interval wherein the plants possessed only a few leaves and only the stalks persisted.

#### 1.2.2. Oil content

The results revealed that there was significant difference in oil content due to treatments. The oil contents at 60, 75 and 90 days harvest intervals, though on par among themselves were significantly superior to those at 105 and 120 days harvest intervals. The highest oil recovery of 0.90 per cent on dry weight basis was recorded at 60 days interval and with further increase in growth the oil content decreased. This is in conformity with the results obtained by Balyan et al. (1982). The decrease in oil content with increase in the interval of harvest may be due to the decrease in the quantity of leaves as the stage of growth of the crop advances, since the leaves

contain much higher oil content than the inflorescence and stem in O. gratissimum (Trivedi et al., 1981). Kurian et al. (1984) reported that the oil content in clocimum harvested at full flowering stage ranged between 0.5 and 0.7 per cent on fresh weight basis which is in accordance with that obtained in the present study for the corresponding stage, but disagrees with the reports of other workers (Sobti et al., 1979 and Trivedi et al., 1981). This variation in essential oil recovery can be attributed to the variation in agro-climatic conditions and the variation in proportion of flower, leaf and stalk in the herbage.

The results of the observation taken on the ratoon crop collected and distilled at earlier intervals viz., 40 and 60 days indicated that the oil content increased with the advancement of crop growth and the values obtained at 40 and 60 days interval are 1.10 and 1.48 per cent respectively. The higher content of oil recorded in this study at 60 days interval compared to that recorded for the corresponding period in the previous growth cycle may presumably be due the effect of the season as the oil content in the dry season is reported to be higher than that under humid conditions (Guenther, 1950 and Hotin, 1968).

### 1.2.3. Oil yield

The total oil yield varied from 17.67 to 75.18 kg ha<sup>-1</sup>. Highest yield was recorded by the crop harvested at 60 days

interval, though it was statistically not different from the crop harvested at 75 and 90 days interval. The harvest intervals of 105 and 120 days were found to be on par and were inferior to the earlier intervals of harvest. The main reason for this is that the oil yield is a function of the oil content and herbage yield. In the present study, both of them were the highest at the early flowering stage and naturally oil yield was also highest under this treatment.

In this context it must be emphasised that the crop in the present study was raised purely as a rainfed crop and hence the low yields compared to those crops raised with irrigation.

### 1.3. Effect of intervals of harvest on oil quality and eugenol yield

The data on the physico-chemical properties of the oil are presented in Tables 3 to 7.

#### 1.3.1. Physical properties of oil

The specific gravity of oil was significantly influenced due to treatments (Table 3). The oil obtained from the shortest harvest interval has recorded the highest specific gravity. Though there was no significant difference between the treatments of 60, 75, 90 and 105 days harvest intervals, specific gravity showed a tendency to decrease with increasing intervals of harvest and

Table 3. Effect of harvest intervals on the physical properties of oil

Harvest intervals	Specific gravity at 30°C	Refractive index at 30°C	Solubility in 70% alcohol (vol.)	Optical rotation
60 days	0.999	1.524	1.25	-9.15°
75 days	0.990	1.523	1.37	-9.77°
90 days	0.989	1.521	1.35	-12.75°
105 days	0.982	1.521	1.75	-14.47°
120 days	0.968	1.521	1.77	-9.47°
SEm ±	0.005	0.001	0.004	0.033
CD (0.05)	0.0190	NS	0.137	0.103

NS = Not significant

the longest harvest interval tried has recorded the lowest value and it was significantly lower than all other harvest intervals.

Data on the refractive index of the oil showed that there was no significant difference due to treatments. The highest value was recorded by the shortest interval of harvest i.e., 60 days and it was on par with the refractive index of the oil obtained at 75 days harvest interval. But with further increase in the growth of the crop, refractive index decreased and there was no difference between the remaining three longer harvest intervals.

The decrease in the specific gravity and refractive index with advancement of growth may be due to a decrease in the eugenol content. Eugenol has a higher specific gravity and refractive index when compared to the other chemical constituents identified in the oil such as  $\beta$ -caryophyllene, limonene, myrcene,  $\gamma$ -cadinene and 4-terpineol (Anon. 1965).

Solubility of the oil in 70 per cent alcohol decreased with increase in intervals of harvest. Highest solubility was recorded by the treatment harvested at 60 days. No significant difference in solubility of oil was noticed till the growth of the crop progressed upto 90 days, but with further increase in the growth of the crop, the solubility of the oil decreased and there was no significant difference in the solubility between the crop harvested at 105 and 120 days harvest intervals. Thus the

solubility of oil was found to decrease almost linearly with increase in the intervals of harvest. Eugenol being a polar compound will have a higher solubility in a polar solvent like alcohol. Hence the increase in solubility of oil at shorter intervals may be attributed to the higher eugenol content at the early growth stages.

The optical rotation of the oil was significantly influenced by the different intervals of harvest. The maximum and minimum values were observed at 105 and 60 days harvest intervals respectively. The higher optical activity of oil observed at the longer harvest intervals may be attributed to the increased concentration of the components such as limonene,  $\gamma$ -cadinene and 4-terpineol in the oil which are having higher optical activity than eugenol (Anon. 1965).

There was not much difference in the physical properties of oil due to harvesting the crop at 40 and 60 days interval (Table 4).

#### 1.3.2: Chemical composition of oil determined by Gas liquid chromatography

The gas liquid chromatographic analysis of oil (Table 5 and Fig. 3 to 8) revealed that eugenol content was highest with the onset of reproductive phase at 60 days harvest interval (75.14



Table 4. Effect of shorter harvest intervals on the physical properties of oil

Harvest intervals	Specific gravity at 30°C	Refractive index at 30°C	Solubility in 70% alcohol (vol.)	Optical rotation
40 days	0.989	1.524	1.2	-9.10°
60 days	0.996	1.525	1.3	-9.05°

Table 5. Effect of harvest intervals on chemical constituents of oil (determined by GLC)

Harvest intervals	Eugenol, %	$\beta$ -caryophyllene, %	$\gamma$ -cadinene, %	Myrcene/ Limonene %	4-terpineol, %
60 days	75.142	2.395	4.702	8.838	0.619
75 days	72.044	2.572	4.981	10.487	0.629
90 days	66.367	3.202	6.169	10.214	0.453
105 days	64.849	4.069	6.939	10.818	0.806
120 days	61.685	3.954	6.091	10.886	0.845

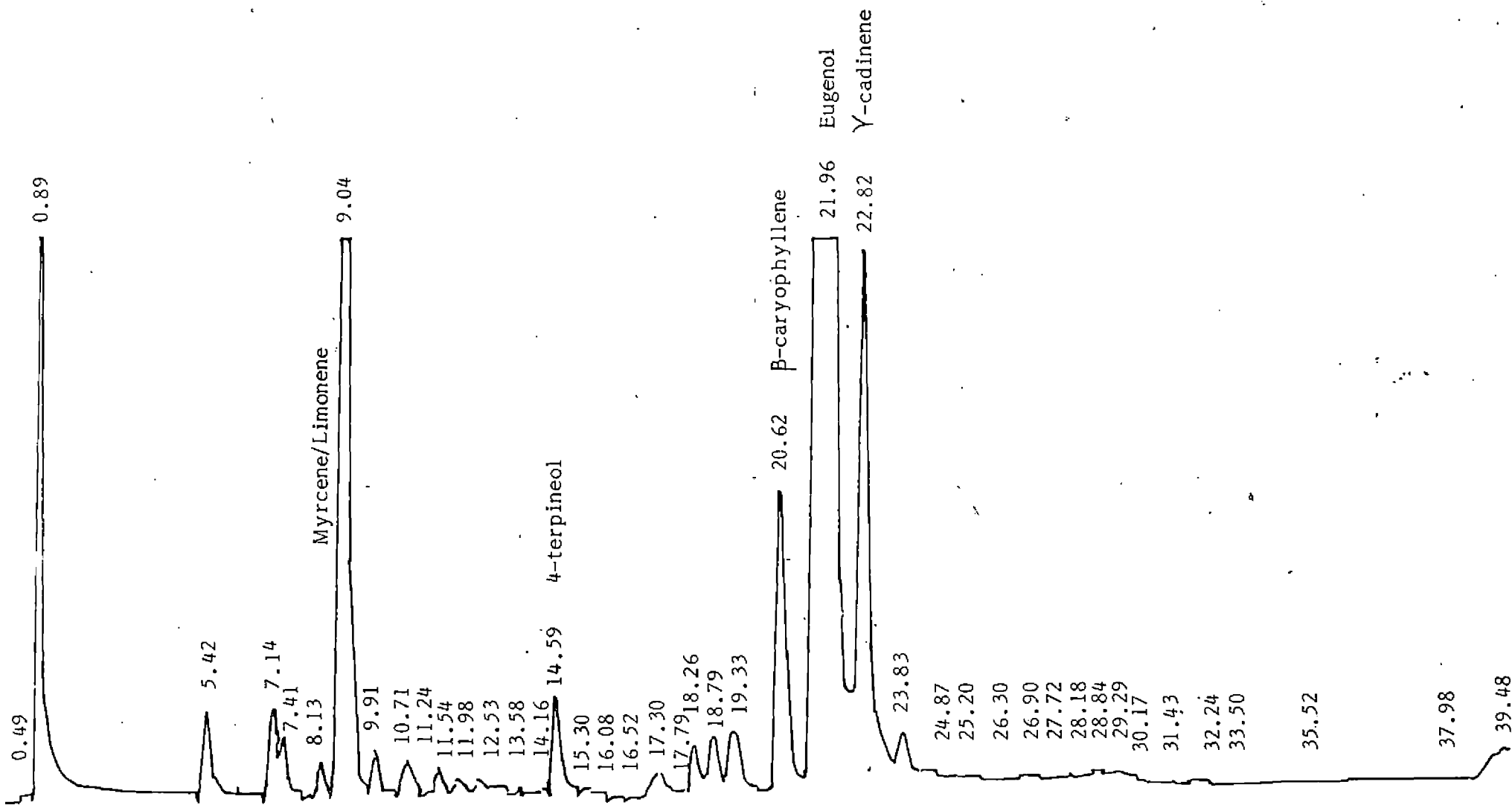


FIG. 3. GLC PROFILE OF OIL AT 60 DAYS HARVEST INTERVAL

RT	AREA	AREA %
5.42	254200	0.467
7.14	248800	0.457
7.41	181500	0.333
8.13	10070	0.018
8.43	93800	0.172
9.04	4815000	8.838
9.91	129300	0.237
10.71	126700	0.233
11.24	92600	0.017
11.54	67300	0.124
11.98	42500	0.078
12.53	40390	0.074
13.58	6990	0.013
14.16	34180	0.063
14.59	337500	0.619
15.30	25140	0.046
16.08	1170	0.002
16.52	923	0.002
17.30	118700	0.218
17.79	23820	0.044
18.26	179300	0.329
18.79	236600	0.434
19.33	331500	0.608
20.62	1305000	2.395
21.96	40940000	75.142
22.82	2562000	4.702
23.83	467500	0.858
24.87	79500	0.146
25.20	162700	0.299
26.30	124400	0.228
26.90	201300	0.369
27.72	106700	0.196
28.18	67540	0.124
28.84	209300	0.384
29.29	205300	0.377
30.17	130700	0.240
31.43	135500	0.249
32.24	126200	0.232
33.50	105200	0.193
35.52	93800	0.172
37.98	34930	0.064
39.48	111200	0.204

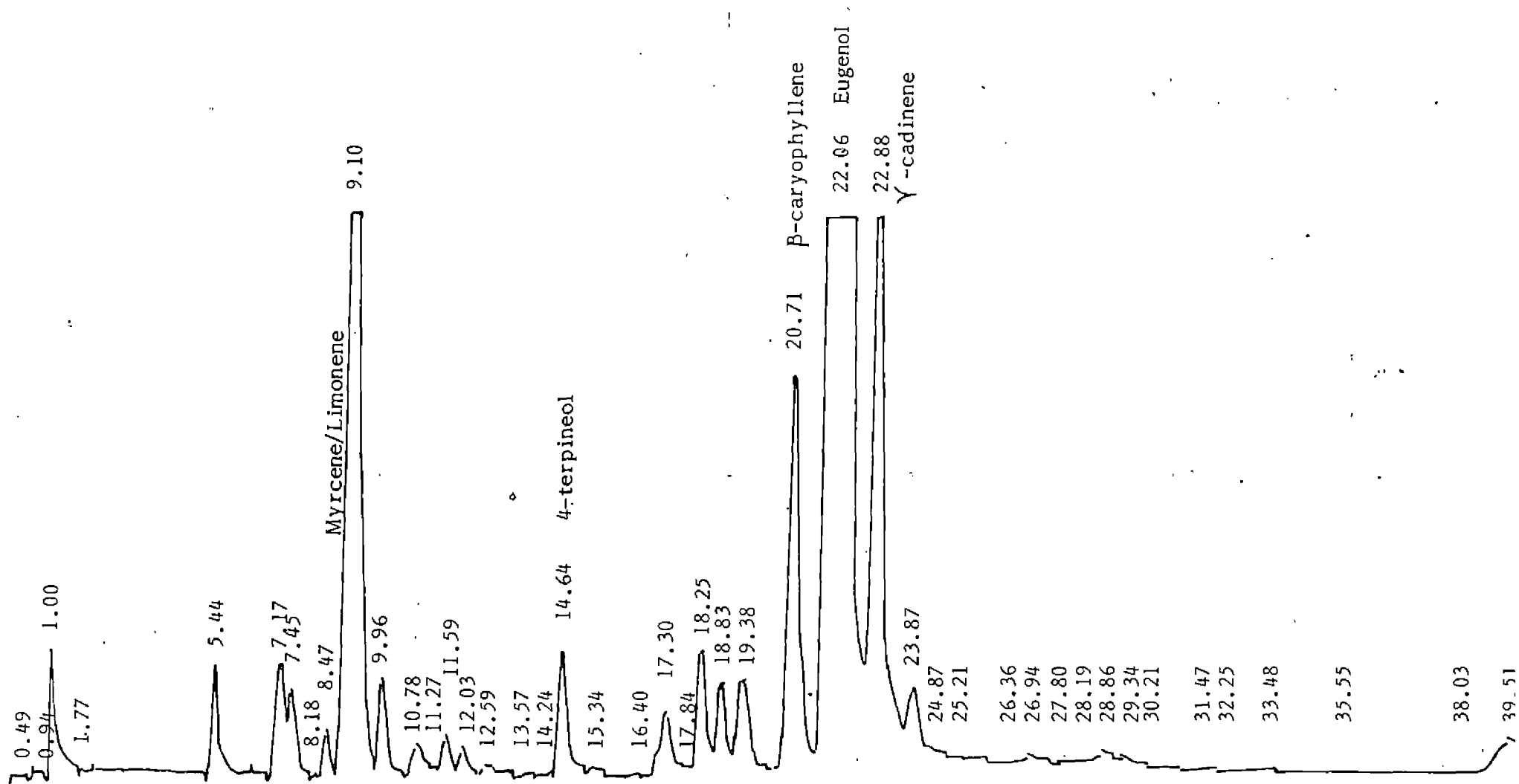


FIG. 4. GLC PROFILE OF OIL AT 75 DAYS HARVEST INTERVAL

RT	AREA	AREA %
1.00	305600	0.437
1.77	9242	0.013
5.44	346400	0.495
7.17	325300	0.465
7.45	254500	0.364
8.18	10330	0.015
8.47	134000	0.192
9.10	7338000	10.487
9.96	316600	0.452
10.78	143400	0.205
11.27	26130	0.037
11.59	119200	0.170
12.03	88840	0.127
12.59	38670	0.055
13.57	5373	0.008
14.24	17100	0.024
14.64	440200	0.629
15.34	32830	0.047
16.40	12570	0.018
17.30	307500	0.439
17.84	44930	0.064
18.25	453000	0.647
18.83	355900	0.509
19.38	529600	0.757
20.71	1800000	2.572
22.06	5041000	72.044
22.88	3485000	4.981
23.87	619000	0.885
24.87	103600	0.148
25.21	172700	0.247
26.36	132300	0.189
26.94	224100	0.320
27.80	114100	0.163
28.19	77080	0.110
28.86	223300	0.319
29.34	211700	0.303
30.21	130700	0.187
31.47	125900	0.180
32.25	102500	0.146
33.48	112900	0.161
35.55	70560	0.101
38.03	34680	0.050
39.51	166300	0.238

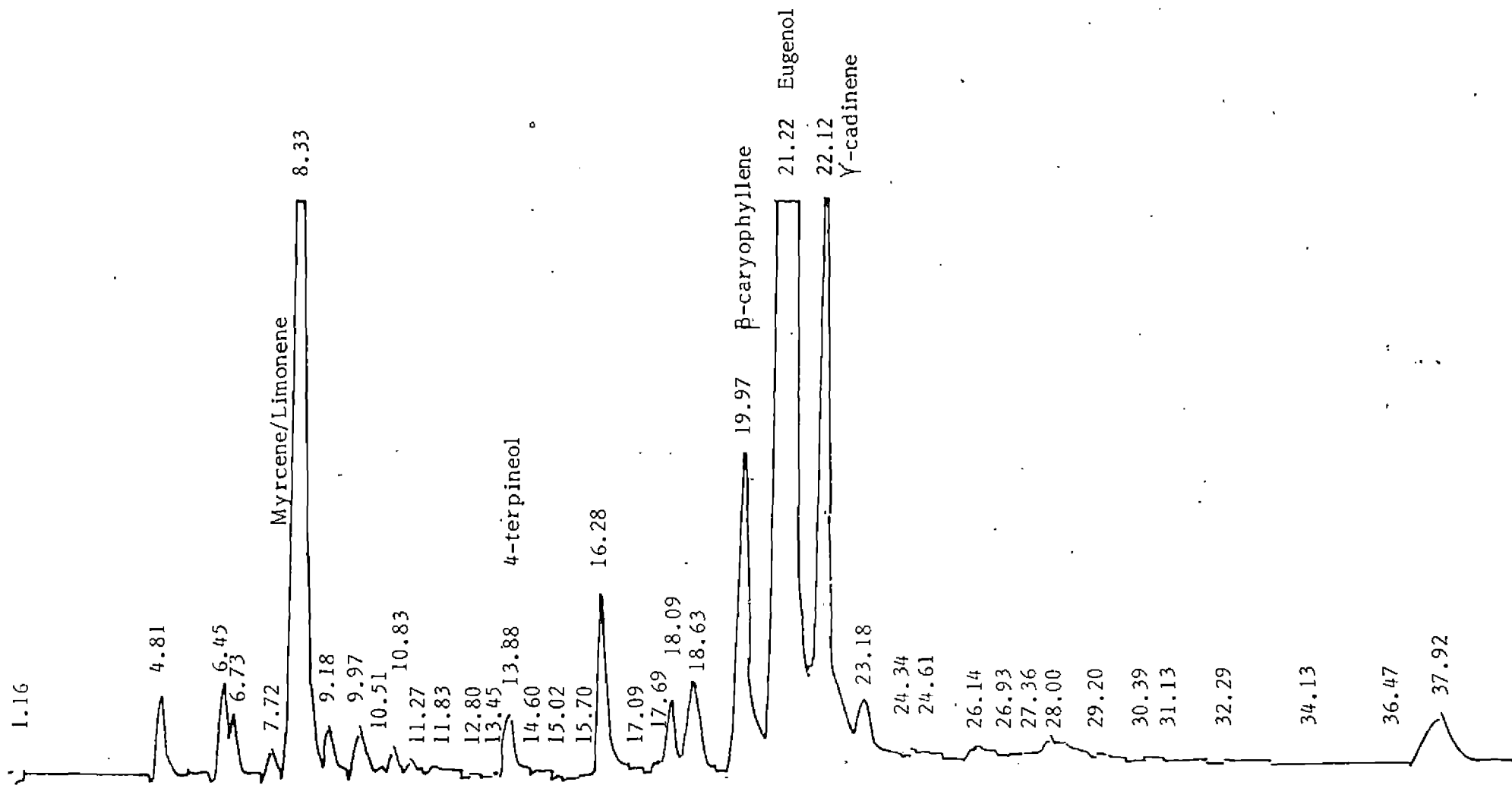


FIG. 5. GLC PROFILE OF OIL AT 90 DAYS HARVEST INTERVAL

RT	AREA	AREA %
1.16	633	0.001
4.81	241300	0.500
6.45	256700	0.532
6.73	171700	0.356
7.72	76500	0.158
8.33	4931000	10.214
9.18	175700	0.364
9.97	181500	0.376
10.51	33630	0.070
10.83	83800	0.174
11.27	56060	0.116
11.83	34190	0.071
12.80	4300	0.009
13.45	9150	0.019
13.88	218600	0.453
14.60	20070	0.042
15.02	581	0.001
15.70	5871	0.012
16.28	682200	1.413
17.09	34840	0.072
17.69	48500	0.100
18.09	290400	0.602
18.63	491000	1.017
19.97	1546000	3.202
21.22	32040000	66.367
21.73	133700	0.277
22.12	2978000	6.169
23.18	547300	1.134
24.34	134700	0.278
24.61	165200	0.342
26.14	405700	0.840
26.93	126500	0.262
27.36	104000	0.215
28.00	491800	1.019
29.20	164700	0.341
30.39	186900	0.387
31.13	151500	0.314
32.39	164100	0.340
34.13	152800	0.317
36.47	158200	0.328
37.92	578100	1.197



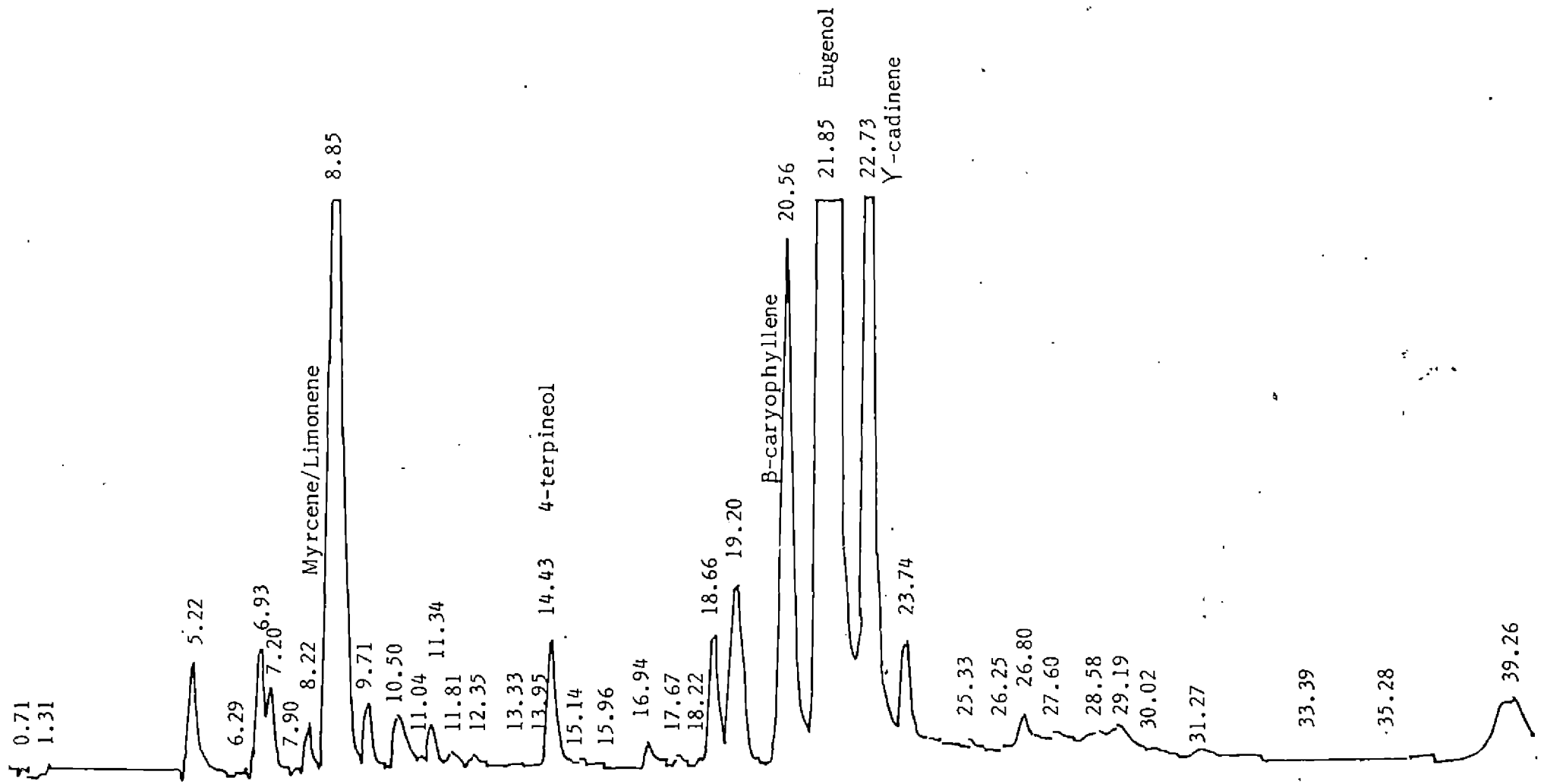


FIG. 6\_ GLC PROFILE OF OIL AT 105 DAYS HARVEST INTERVAL

RT	AREA	AREA %
1.31	1406	0.002
5.22	343700	0.553
6.29	5428	0.009
6.93	350000	0.564
7.20	246900	0.398
7.90	18330	0.030
8.22	145300	0.234
8.85	6718000	10.818
9.71	239900	0.386
10.50	270100	0.435
11.04	43430	0.070
11.34	162700	0.262
11.81	93640	0.151
12.35	85420	0.138
13.33	62070	0.100
13.95	51160	0.082
14.43	500700	0.806
15.14	67520	0.109
15.96	29960	0.048
16.94	150700	0.243
17.67	64740	0.104
18.22	59740	0.096
18.66	530900	0.855
19.20	906200	1.459
20.56	2527000	4.069
21.85	40270000	64.849
22.73	4309000	6.939
23.74	926200	1.492
25.33	217100	0.350
26.25	180400	0.291
26.80	475300	0.765
27.60	333500	0.537
28.58	363700	0.586
29.19	401300	0.646
30.02	155300	0.250
31.27	252800	0.407
33.39	68660	0.111
35.28	26790	0.043
39.26	442800	0.713

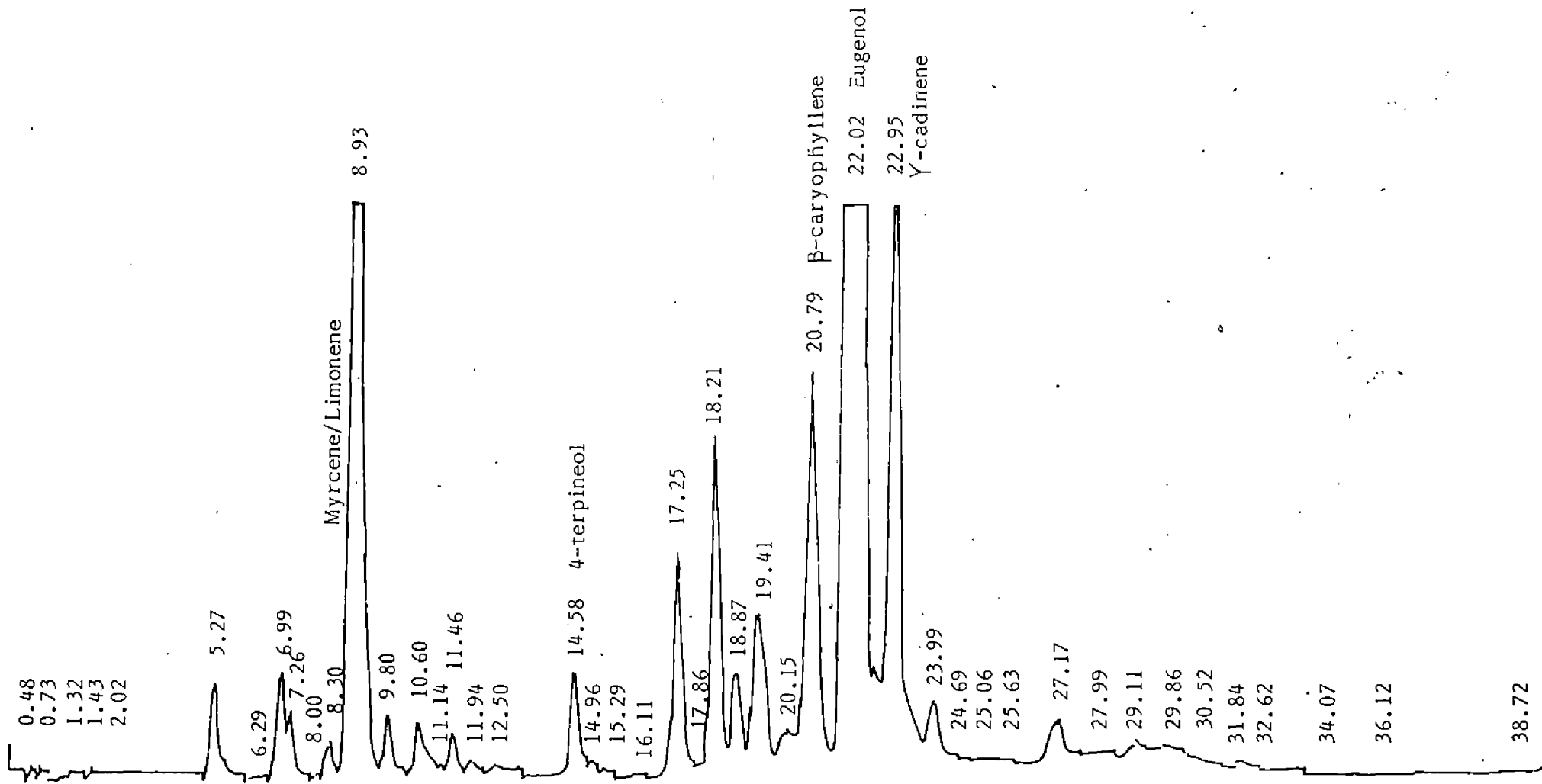


FIG. 7. GLC PROFILE OF OIL AT 120 DAYS HARVEST INTERVAL

RT	AREA	AREA %
1.32	1001	0.002
1.43	1560	0.003
2.02	187	0.000
5.27	284100	0.599
6.29	15610	0.033
6.99	302100	0.636
7.26	192600	0.406
8.00	13780	0.029
8.30	107000	0.225
8.93	5167000	10.886
9.80	208100	0.438
10.60	237500	0.500
11.14	28820	0.061
11.46	13400	0.277
11.94	57470	0.121
12.50	33100	0.070
14.58	401200	0.845
14.96	45340	0.096
15.29	26200	0.055
16.11	14020	0.030
17.25	86400	1.822
17.86	44640	0.094
18.21	1219000	2.568
18.87	388600	0.819
19.41	734400	1.547
20.15	252500	0.532
20.79	1877000	3.954
22.02	29280000	61.685
22.95	2891000	6.091
23.99	454000	0.956
24.69	65620	0.138
25.06	83840	0.177
25.63	157900	0.333
27.17	565600	1.192
27.99	236500	0.498
29.11	281200	0.592
29.86	337600	0.711
30.52	193700	0.408
31.84	133700	0.288
32.62	63220	0.133
34.07	36460	0.077
36.12	22620	0.048
38.72	14980	0.032

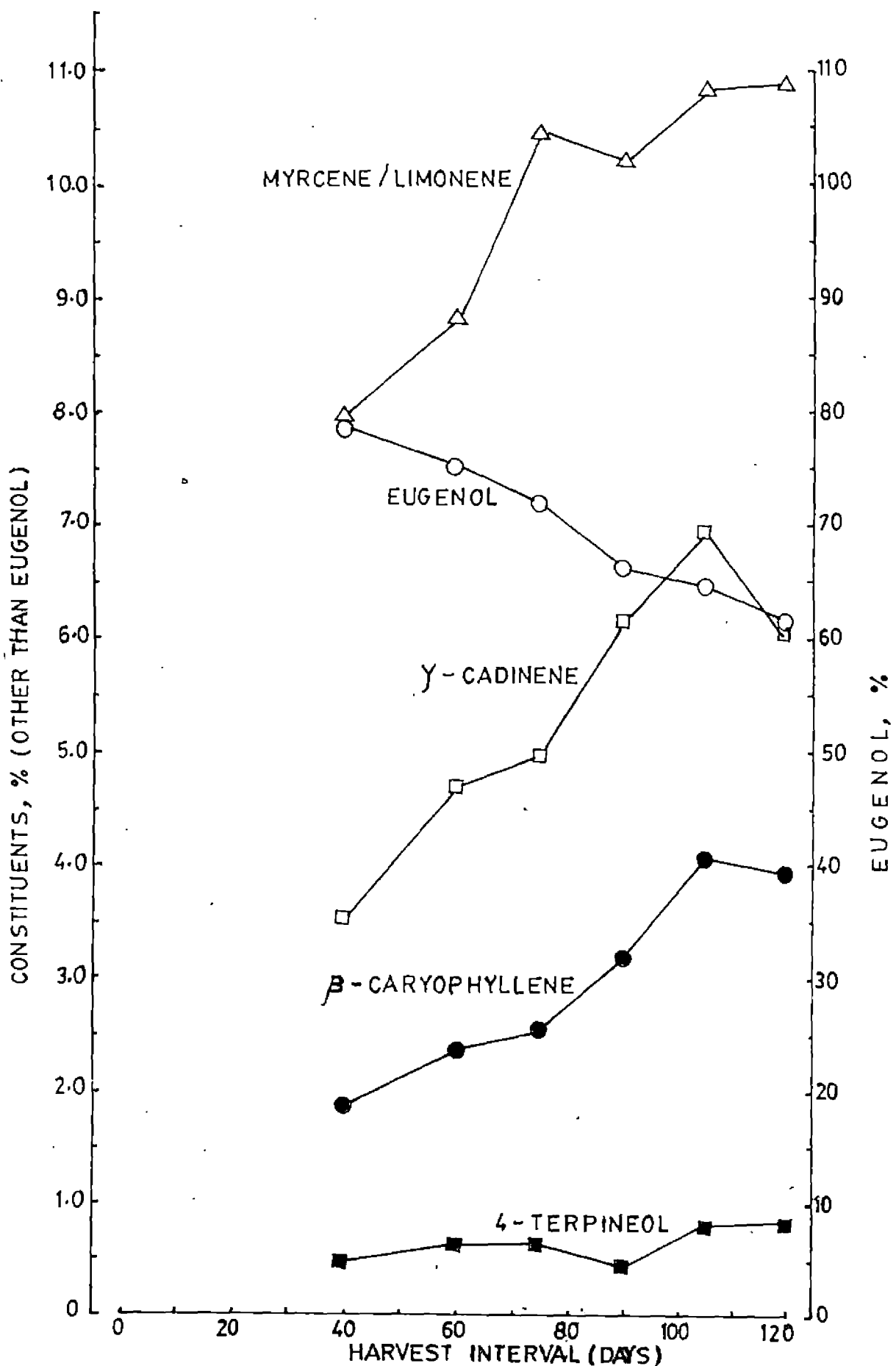


FIG. 8 - CHANGES IN THE CONSTITUENTS OF OIL AT DIFFERENT HARVEST INTERVALS AS DETERMINED BY GLC ANALYSIS

per cent), slightly decreased at 75 days followed by a sharp fall at 90 days which coincided with early seed setting stage and continued to decrease steadily with advancement of the growth stage (105 and 120 days). However the decrease in eugenol content was accompanied by a concomittant increase in the other components identified in the oil such as  $\beta$ -caryophyllene and  $\gamma$ -cadinene. In the case of myrcene, the content went on increasing with increase in the intervals of harvest. But the influence of the treatments on the content of 4-terpineol did not show any definite pattern. Similar changes in the oil composition with progressive maturity were recorded in O. sanctum (Dey and Choudhuri, 1981 and Pareek et al., 1982) and O. gratissimum (Cheng et al., 1983).

Analysis of the oil obtained from the ratoon crop harvested at 40 and 60 days revealed that the chemical composition of the oil obtained at 40 days was more or less similar to that obtained at 60 days (Table 6 and Fig. 9 and 10). This indicated that the composition of oil in the plant remained almost stable upto 60 days and reappropriation took place only after this period.

### 1.3.3. Eugenol content (determined by chemical method) and eugenol yield

Eugenol content is significantly influenced by different intervals of harvest (Table 7). The crop harvested at 60 days possessed maximum eugenol in the essential oil (82.5 per cent)

Table 6. Effect of shorter harvest intervals on the chemical constituents of oil (determined by GLC)

Harvest intervals	Eugenol, %	$\beta$ -caryophyllene, %	$\gamma$ -cadinene, %	Myrcene/ Limonene %	4-terpineol, %
40 days	78.632	1.886	3.514	7.992	0.462
60 days	78.498	2.003	3.684	7.907	0.476

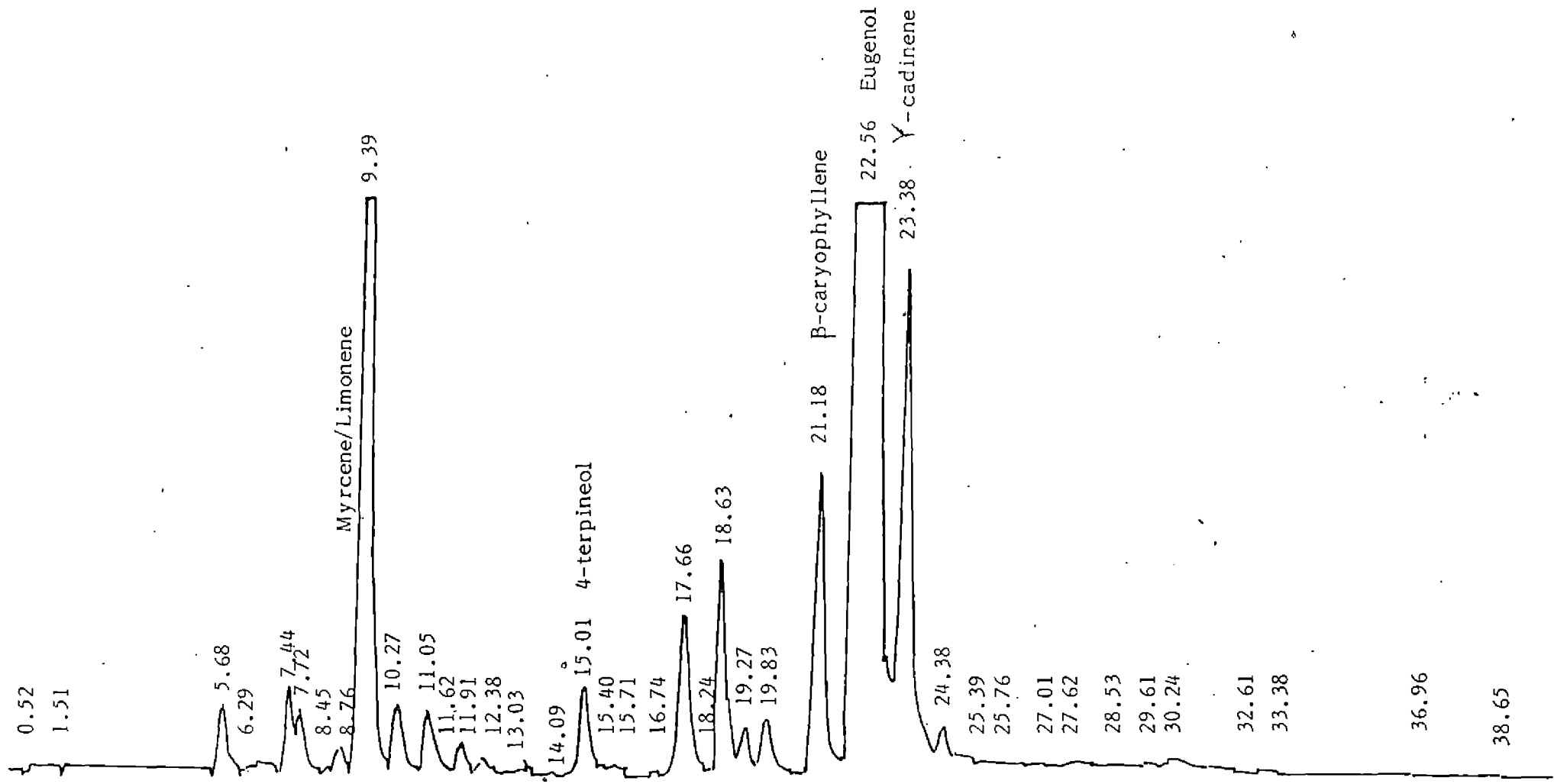
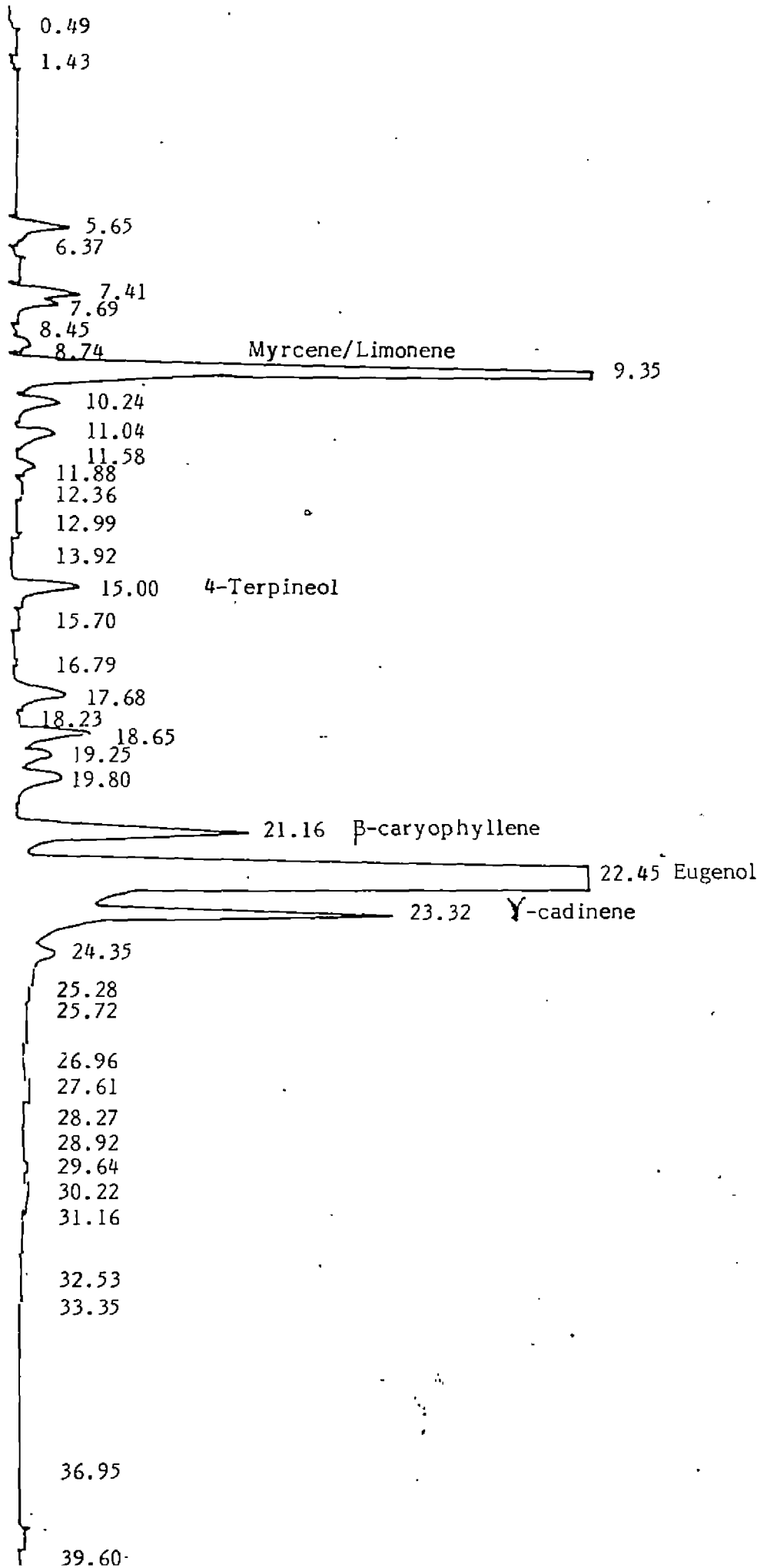


FIG. 9. GLC PROFILE OF OIL OF RATOON CROP AT 40 DAYS HARVEST INTERVAL



RT	AREA	AREA %
0.52	615	0.001
1.51	43	0.000
5.68	190300	0.288
6.29	9550	0.014
7.44	229200	0.347
7.72	162900	0.247
8.45	11420	0.017
8.76	70540	0.107
9.39	5277000	7.992
10.27	207300	0.314
11.05	221400	0.335
11.62	5262	0.008
11.91	90620	0.137
12.38	51660	0.078
13.03	18700	0.028
14.09	7168	0.011
15.01	305000	0.462
15.40	31750	0.048
15.71	33770	0.051
16.74	17600	0.027
17.66	606100	0.918
18.24	24450	0.037
18.63	768600	1.164
19.27	178100	0.270
19.83	276300	0.418
21.18	1245000	1.886
22.56	51920000	78.632
23.38	2320000	3.514
24.38	348500	0.528
25.39	82560	0.125
25.76	149700	0.227
27.01	120200	0.182
27.62	205600	0.311
28.53	143800	0.218
29.61	162300	0.246
30.24	276900	0.419
32.61	106500	0.161
33.38	133200	0.202
36.96	15280	0.023
38.65	4092	0.006

FIG.10 - GLC PROFILE OF OIL OF RAISON CROP AT 60 DAYS HARVEST INTERVAL



RT	AREA	AREA %
0.49	599	0.001
1.43	719	0.001
5.65	162800	0.333
6.37	4535	0.009
7.41	184100	0.377
7.69	123100	0.252
8.45	6086	0.012
8.74	53670	0.110
9.35	3863000	7.907
10.24	142200	0.291
11.04	151200	0.309
11.58	9250	0.019
11.88	67020	0.137
12.36	39590	0.081
12.99	13220	0.027
13.92	3295	0.007
15.00	232400	0.476
15.70	27750	0.057
16.79	28020	0.057
17.68	250700	0.513
18.23	31090	0.064
18.65	277700	0.568
19.25	159800	0.327
19.80	253400	0.519
21.16	991400	2.029
22.45	38350000	78.498
23.32	1800000	3.684
24.35	337400	0.691
25.28	71540	0.146
25.72	145900	0.299
26.96	97180	0.199
27.61	185300	0.379
28.27	99720	0.204
28.92	49110	0.101
29.64	149600	0.306
30.22	170900	0.350
31.16	88920	0.182
32.53	93660	0.192
33.35	118700	0.243
36.95	19340	0.040
39.60	687	0.001

Table 7. Effect of harvest intervals on eugenol content (determined by chemical analysis) and eugenol yield

Harvest intervals	Eugenol content, %	Eugenol yield, kg ha <sup>-1</sup>
60 days	82.50	60.54
75 days	77.00	52.62
90 days	74.50	42.00
105 days	68.25	21.98
120 days	68.50	11.57
S $\bar{E}$ m $\pm$	1.12	6.20
CD (0.05)	3.456	19.111

which was found to be significantly superior to all the other treatments. The eugenol content was found to decrease with increased intervals of harvest. The variation in eugenol content with harvest intervals showed a similar trend as in GLC analysis but the values obtained in the chemical method were higher perhaps due to the combined estimation of other phenols in the oil.

The eugenol yield for the different treatments varied from 11.57 to 60.54 kg ha<sup>-1</sup>. Maximum eugenol was produced by the crop harvested at 60 days followed by the next succeeding interval of harvest viz., 75th day and there was no significant difference between these two treatments. Eugenol yield was found to decrease with increase in the intervals of harvest. The higher eugenol yield recorded at 60 days and its decline thereafter may be due to the higher oil yield and eugenol content recorded at the earlier cutting interval which decreased later.

#### 1.4. Effect of intervals of harvest on plant nutrient content and uptake

Data on the nutrient content and uptake are presented in Tables 8 to 11.

##### 1.4.1. Nitrogen content

Nitrogen content of the plant samples showed a tendency to decrease with increase in growth of the plant (Table 8).

Table 8. Effect of harvest intervals on N, P, K, Ca and Mg content of clover, per cent

Harvest intervals	Nitrogen	Phosphorus	Potassium	Calcium	Magnesium
60 days	1.82	0.40	1.59	2.33	0.40
75 days	1.53	0.39	1.84	2.09	0.31
90 days	1.47	0.42	1.53	2.36	0.35
105 days	1.33	0.35	1.28	1.77	0.28
120 days	1.31	0.37	1.39	2.56	0.39
SEm $\pm$	0.06	0.01	0.03	0.09	0.01
CD (0.05)	0.195	0.030	0.097	0.304	0.020

Comparing between the harvest intervals, 60 days harvest interval showed the highest nitrogen content with 1.82 per cent. It was found to be statistically superior to the other four treatments tried. At early stages of growth of plant, carbohydrate utilization is more as more protein is formed from the manufactured carbohydrates. Since nitrogen is an invariable component of proteins and therefore of protoplasm which is higher in younger tissues, the increase in the nitrogen content at the early stages of growth may be due to this (Tisdale and Nelson, 1956). The decrease noticed in the nitrogen content in the later stages of growth may be due to the formation of fibres and distribution of so far accumulated proteins and protoplasm for seed set and oil synthesis. Guenther (1950) claimed that precursors of essential oil could be obtained through degradation of carbohydrates and proteins. Pridham (1967) has also reported that certain compounds like sugars and aminoacids are capable of being translocated into the sites of essential oil synthesis and ultimately used during synthesis of monoterpenes.

#### 1.4.2. Phosphorus content

Intervals of harvest influenced the phosphorus content of plants significantly. But no regular trend was noticed in the P content of the plant with change in the interval of harvest. The maximum (0.42 per cent) and minimum (0.35 per cent) values were recorded by the treatments harvested at 90 and 105 days intervals

respectively. The P content in the plant tissues reduced after 90 days probably due to breakdown of aminoacids and other phosphorus compounds at later stages. The high P content observed at initial seeding stage might probably be due to the higher requirement of the plant at the time of maturation as most of the P of plant is required for formation of seeds and fruit.

#### 1.4.3. Potassium content

Significant variation was observed in the potassium content of the plant samples, but as in the case of phosphorus, no regular trend was noticed in the potassium content of the plants with increase in the growth period. The values varied from 1.28 to 1.84 per cent and the maximum and minimum values were recorded at 75 and 105 days harvest intervals, respectively. Since potassium content is related to nitrogen metabolism and has a catalytic influence on protein synthesis, a decrease in K content with increase in maturity of the plant may be due to a decrease in the photosynthetic rate and a decrease in nitrogen content of the plant (Tisdale and Nelson, 1956).

#### 1.4.4. Calcium content

There was significant difference in the calcium content of the plants due to intervals of harvest. But no definite trend was noticed in the calcium content with change in the intervals of harvest.



#### 1.4.5. Magnesium content

The influence of harvest intervals on the magnesium content of plants was significant. The values varied from 0.28 per cent recorded at 105 days to 0.40 per cent at 60 days harvest interval. As in the case of calcium no definite trend was noticed in the content of magnesium with increase in the intervals of harvest.

#### 1.4.6. Iron content

The iron content of the plant samples though varied significantly did not show any particular trend for the harvest intervals (Table 9). The range in the iron content varied from 58.25 to 339.95 ppm.

#### 1.4.7. Manganese content

The manganese content of the plant samples did not show statistical difference between the treatments and the values ranged from 69.37 ppm at 120 days to 98.75 ppm at 60 days harvest interval.

#### 1.4.8. Zinc content

No significant variation was observed in the zinc content of the plant samples due to the intervals of harvest. The range in the zinc content varied from 16.25 to 53.12 ppm.

Table 9. Effect of harvest intervals on the Fe, Mn and Zn content of clove, ppm

Harvest intervals	Iron	Manganese	Zinc
60 days	276.00	98.75	16.25
75 days	133.12	75.62	16.25
90 days	339.95	89.37	49.45
105 days	58.25	84.37	16.25
120 days	102.50	69.37	53.12
SEm <u>+</u>	54.61	9.78	12.84
CD (0.05)	168.295	NS	NS

NS = Not significant

#### 1.4.9. Nitrogen uptake

The uptake of nitrogen by the crop did not differ significantly among the treatments (Table 10). But maximum uptake of nitrogen was recorded by the treatment harvested at 60 days harvest interval. General trend was a decrease in the uptake with increase in the harvest interval.

#### 1.4.10. Phosphorus uptake

The phosphorus uptake also showed no significant variation due to treatments. The uptake of phosphorus ranged from 26.60 kg ha<sup>-1</sup> to 41.50 kg ha<sup>-1</sup> and the maximum uptake was recorded by the treatment harvested at 60 days harvest interval.

#### 1.4.11. Potassium uptake

The potassium uptake was significant due to the effect of different intervals of harvest. Maximum uptake was recorded at 75 days interval and there was no significant difference between the treatments 60, 75, 90 and 105 days cutting intervals. The treatment harvested at 120 days recorded the lowest uptake and it was significantly lower than all other treatments.

In general the uptake of nitrogen, phosphorus and potassium decreased with increase in the intervals of harvest. The main reason for this decline may be due to a decrease in the dry

Table 10. Effect of harvest intervals on uptake of N, P, K, Ca and Mg, kg ha<sup>-1</sup>

Harvest intervals	Nitrogen	Phosphorus	Potassium	Calcium	Magnesium
60 days	172.56	41.50	160.26	218.01	35.13
75 days	147.84	40.14	163.11	189.64	28.37
90 days	129.80	29.24	115.49	148.46	25.13
105 days	147.40	39.96	117.06	202.95	28.36
120 days	107.12	26.60	81.57	132.73	21.82
SEm <u>±</u>	18.36	4.92	17.91	25.87	3.58
CD (0.05)	NS	NS	55.196	NS	NS

NS = Not significant

matter yield with increased intervals of harvest.

#### 1.4.12. Calcium uptake

Uptake of calcium was not influenced significantly due to various harvest intervals and the uptake varied from 132.73 to 218.01 kg ha<sup>-1</sup>. The maximum uptake was recorded by treatment interval of 60 days and the minimum by the 120 days harvest interval.

#### 1.4.13. Magnesium uptake

The uptake of magnesium was also not significant due to different intervals of harvest. But it showed a tendency to decrease with increase in the intervals of harvest and the magnesium uptake varied from 21.82 to 35.13 kg ha<sup>-1</sup>.

Thus the decline in uptake of calcium and magnesium at higher intervals of harvest may be due to decreased dry matter production.

#### 1.4.14. Iron uptake

There was significant difference in the uptake of iron due to different intervals of harvest. The maximum uptake was recorded by the shortest interval of harvest (2.29 kg ha<sup>-1</sup>) and the minimum uptake (0.51 kg ha<sup>-1</sup>) by the treatment harvested at the longest interval of 120 days.

Table 11. Effect of harvest intervals on uptake of Fe, Mn and Zn, kg ha<sup>-1</sup>

Harvest intervals	Iron	Manganese	Zinc
60 days	2.29	0.83	0.14
75 days	0.94	0.59	0.17
90 days	2.19	0.54	0.31
105 days	0.52	0.70	0.13
120 days	0.51	0.35	0.26
SEm ±	0.43	0.12°	0.08
CD (0.05)	1.319	NS	NS

NS = Not significant

#### 1.4.15. Manganese uptake

The influence of intervals of harvest on manganese uptake was not significant. Maximum uptake was recorded by the harvest interval of 60 days and the values ranged from 0.35 to 0.83 kg ha<sup>-1</sup>.

#### 1.4.16. Zinc uptake

In the case of zinc also there was no variation in the uptake. Maximum uptake was recorded by the longest interval of harvest tried. The uptake of zinc varied from 0.13 to 0.31 kg ha<sup>-1</sup>.

The maximum uptake recorded at 60 days may be due to the increased dry matter production at this stage as there was no significant difference in the contents of the above nutrients due to treatments.

## 2. Experiment to study the effect of varying levels of shade on yield and quality of clove oil and nutrient content and uptake

### 2.1. Effect of levels of shade on biometric characters

A perusal of the data (Table 12) suggests that the height of the plants under all the shade levels were significantly higher than that under open condition. The plant height was maximum under 50 per cent shade and it was statistically on par with the plant height at 25 per cent shade. The plants under shade

Table 12. Effect of levels of shade on the biometric characters

Levels of shade	Plant height (cm)	Total number of branches	Number of flowered branches	Length of inflorescence (cm)	Height at first branching (cm)	Spread (cm <sup>2</sup> )	Leaf area (cm <sup>2</sup> )
No shade (100 per cent light)	109.00	23.08	12.24	14.86	5.75	5054.0	37.61
Low shade (25 per cent shade)	155.00	18.48	8.16	16.86	4.60	6045.0	44.52
Medium shade (50 per cent shade)	163.32	8.72	4.88	15.22	4.10	7074.0	38.63
High shade (75 per cent shade)	140.00	13.04	4.96	10.34	2.18	2864.0	27.64
SEm ±	3.33	1.22	1.03	1.45	1.37	561.75	2.69
CD (0.05)	10.264	3.757	3.172	4.457	NS	1731.059	8.303

NS = Not significant



tended to grow longer and they apparently were slender and weak. The shade effect is believed to be due to auxin enhancement, probably acting synergistically with gibberillic acid (Leopold, 1964). Theoretically photodestruction of auxin is less in shaded stands; shading tends to increase auxin levels which could affect internode length (Evans, 1973).

Number of branches were influenced significantly due to different intensities of shade. Open condition recorded the maximum number of branches (23.08) and it was superior to all other treatments. Similar results have been reported in crops such as sweet potato, coleus, ginger and turmeric (Bai, 1981 and Varghese, 1989).

Highly significant variation was noticed between treatments with respect to number of flowered branches. A decrease in number of flowering shoots was observed due to shade. Maximum number of flowered branches (12.24) was recorded under open condition and it was significantly superior to all other treatments. The plants in the open flowered early by 43 days when compared to that under the shaded conditions. Hence two harvests could be obtained from the plants grown in the open, whereas only one harvest was obtained under shaded conditions. The decrease in the number of flowering branches under 50 and 75 per cent shade may be due to the fact that the optimum light which is required

for the production of flowers is above that required for vegetative development.

The length of inflorescence showed significant variation due to different intensities of shade. Maximum length of inflorescence (16.86 cm) recorded by the plants grown under 25 per cent shade was on par with plants grown under open and 50 per cent shade. Minimum value was recorded under 75 per cent shade and it was statistically inferior to the other treatments.

The influence of different levels of shade with regard to the height at the first branching was not significant and the values ranged from 2.18 to 5.17 cm.

Plants grown under 50 per cent shade exhibited the maximum spread ( $7074.0 \text{ cm}^2$ ) and it was statistically on par with the plants grown under 25 per cent shade ( $6045.0 \text{ cm}^2$ ). The least spread recorded by plants grown in the high shade ( $2864.0 \text{ cm}^2$ ) was found to be statistically inferior to the rest of the treatments. The primary reason for the decrease in the spread of plants under high shade is the reduction in the number of branches under shaded conditions.

Maximum leaf area of  $44.52 \text{ cm}^2$  was exhibited at the shade level of 25 per cent and it was on par with no shade and the medium shade level of 50 per cent. But with further increase in the intensity of shade, the leaf area decreased ( $27.64 \text{ cm}^2$ ).

The results reported by Duriyaprapan and Britten (1982) are not in agreement with the results of the present study as they have observed an increase in leaf area development in shaded Mentha arvensis.

## 2.2. Effect of levels of shade on herbage yield, oil content and oil yield

The results on the herbage yield, oil content and oil yield as influenced by the treatments are presented in Table 13 and Fig. 11.

### 2.2.1. Herbage yield

With increase in shading the fresh as well as dry herbage yield showed a decreasing trend. The yield under open condition was superior to the rest of the treatments. In spite of the lower plant height, spread and leaf area under full illumination compared to the low and medium shade levels, herbage yield was highest for the plants grown in the open. This may be due to an increase in the total number of branches and flowered branches. This may also be attributed to the fact that while two harvests were taken in the case of plants grown in the open only one harvest could be taken in the case of plants grown under shade.

A second harvest in the shaded plants was not possible due to delayed flowering. When the yield of only one harvest

Table 13. Effect of levels of shade on herbage yield, oil content and oil yield

Levels of shade	Herbage yield, t ha <sup>-1</sup>		Essential oil content, %		Essential oil yield, kg ha <sup>-1</sup>
	FWB	DWB	FWB	DWB	
No shade (100 per cent light)	20.17	15.33	0.44	0.58	89.63
Low shade (75 per cent shade)	12.49	9.29	0.33	0.45	40.45
Medium shade (50 per cent shade)	11.21	8.33	0.26	0.37	30.32
High shade (75 per cent shade)	8.08	5.99	0.25	0.35	20.99
SEm ±	0.96	0.73	0.11	0.05	5.21
CD (0.05)	2.964	2.251	0.040	0.157	16.053

FWB = Fresh weight basis  
DWB = Dry weight basis

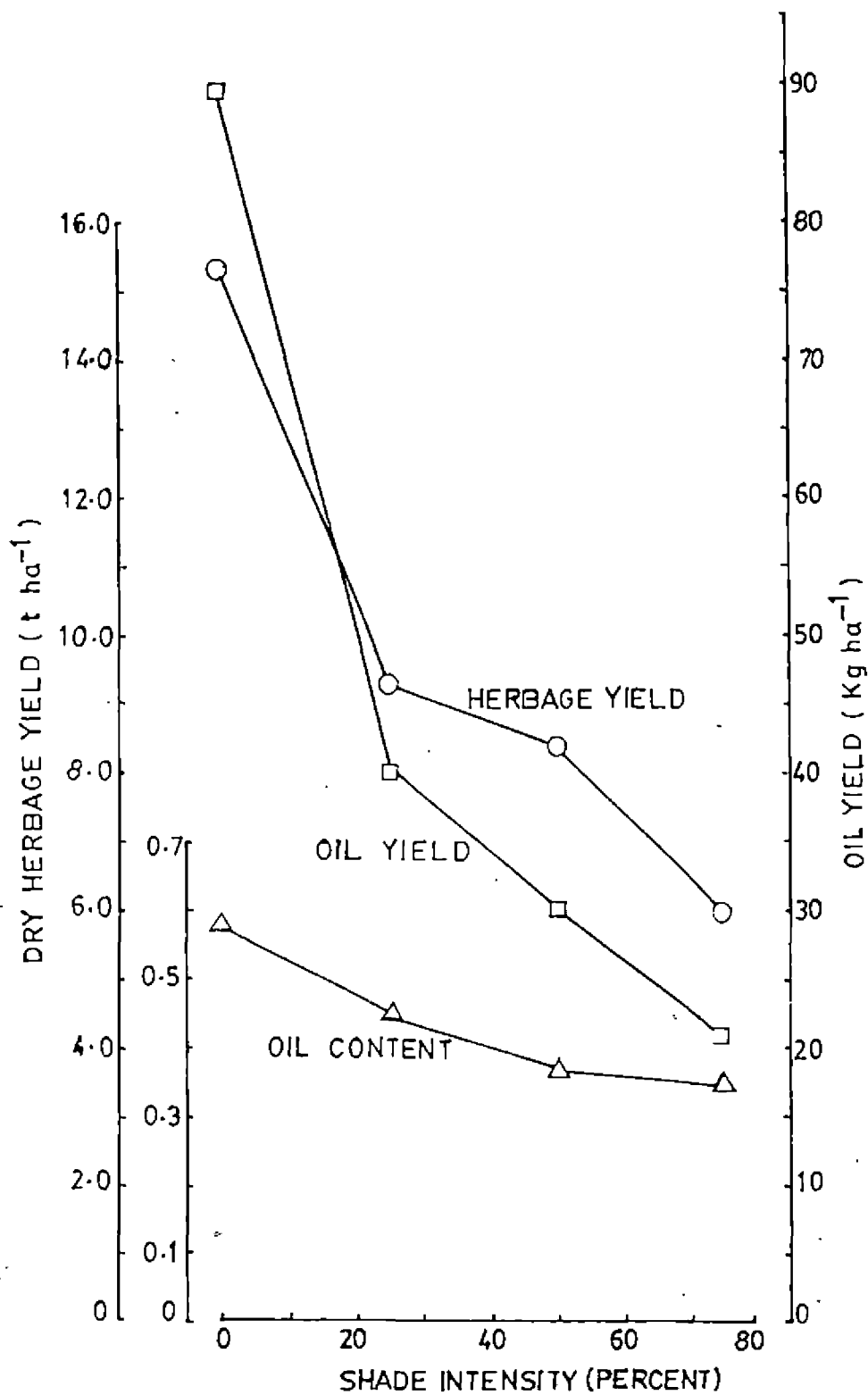


FIG.11- EFFECT OF DIFFERENT SHADE LEVELS ON HERB-  
AGE YIELD, OIL CONTENT AND OIL YIELD

was considered then low shaded plants were found to perform slightly better since the yield obtained from the open was less ( $11.5 \text{ t ha}^{-1}$ ) compared to 25 per cent shade ( $12.5 \text{ t ha}^{-1}$ ) on FWB and the corresponding figures on DWB were  $8.8$  and  $9.3 \text{ t ha}^{-1}$ .

#### 2.2.2. Oil content

The percentage of oil on fresh and dry weight basis decreased linearly with increase in the intensity of shade. The highest value was recorded with 100 per cent light (0.58 per cent) on dry weight basis and it was on par with 25 per cent shaded condition. Higher intensities of shade viz., 50 and 75 per cent recorded lower oil contents. Similar results in other essential oil yielding crops such as Japanese mint and peppermint have been reported by many workers (Handerson, 1911 and Dutta, 1971). Not much difference was observed in the oil content between open (0.46 per cent) and 25 per cent shade (0.45 per cent) when only a single harvest of the open was considered.

#### 2.2.3. Oil yield

The yield of oil obtained from plants grown under full light recorded the maximum value ( $89.63 \text{ kg ha}^{-1}$ ) and was significantly superior to all other shade levels. The increase in oil yield was almost linear with increase in light intensity. The highest oil yield under open condition may be attributed to the higher herbage yield and oil content recorded under this treatment

which are the contributing factors for oil yield. Again, the oil yield from a single harvest of the plants in the open (40.57 kg ha<sup>-1</sup>) was found to be almost equal to that under 25 per cent shade (40.45 kg ha<sup>-1</sup>).

### 2.3. Effect of levels of shade on oil quality and eugenol yield

The data obtained on the physico-chemical properties of the oil are presented from Table 14 to 16.

#### 2.3.1. Physical properties of oil

Specific gravity of oil was influenced significantly due to treatments (Table 14). The values varied from 0.965 to 0.993. The values obtained showed a tendency to decrease at higher levels of shade.

The effect of the treatments was not significant in the case of refractive index of oil and the values ranged from 1.518 to 1.524. As in the case of specific gravity of oil the values obtained at higher intensities of shade were lower than that under open condition. This may be due to the decrease in the eugenol content which exhibits higher values for the above two properties compared to the other components identified in the study (Anon. 1965).

Though there was significant difference in the solubility of the oil due to treatments the values varied from 1.26 to 1.54.

Table 14. Effect of levels of shade on the physical properties of oil

Levels of shade	Specific gravity at 30°C	Refractive index at 30°C	Solubility in 70% alcohol(vol.)	Optical rotation
No shade (100 per cent light)	0.990	1.524	1.32	-13.0°
Low shade (25 per cent shade)	0.993	1.524	1.26	-13.7°
Medium shade (50 per cent shade)	0.965	1.518	1.54	-18.5°
High shade (75 per cent shade)	0.975	1.521	1.44	-16.8°
SEm <sub>+</sub>	0.006	0.000	0.048	0.044
CD (0.05)	0.0191	NS	0.150	0.13

NS = Not significant



A decrease in the solubility of oil was noticed at higher levels of shade intensity and this may also be due to the decrease in the content of eugenol which is having a high solubility compared to the other components identified.

Optical rotation of the oil was also not influenced significantly by the different levels of shade. The values of optical rotation recorded for 0, 25, 50 and 75 per cent shade levels were  $-13.0^\circ$ ,  $-13.7^\circ$ ,  $-18.5^\circ$  and  $-16.8^\circ$  respectively. The higher optical activity recorded under higher intensities of shade may be due to the reduced content of eugenol in the oil which was found to possess no optical activity.

#### 2.3.2. Chemical composition of oil determined by Gas liquid chromatography

The results revealed high variation in the eugenol content of the oil due to different levels of shade (Table 15 and Fig.12 to 16). Eugenol content was found to decrease as the intensity of shade increased from 0 to 75 per cent. The highest value (72.0 per cent) was recorded for no shade and the lowest value (57.0 per cent) under 75 per cent shade. The low content of eugenol under shaded conditions may be due to the inhibition of eugenol synthesis under shaded conditions or the conversion of eugenol into other constituents. This explanation seems logical in that the percentage of other minor constituents in the oil viz.,

Table 15. Effect of levels of shade on chemical constituents of oil (determined by GLC)

Levels of shade	Eugenol, %	$\beta$ -caryophyllene, %	$\gamma$ -cadinene, %	Myrcene/ Limonene %	4-terpineol, %
No shade (100 per cent light)	72.03	2.62	6.10	9.53	0.55
Low shade (25 per cent shade)	69.12	3.11	7.12	8.47	0.60
Medium shade (50 per cent shade)	60.60	3.55	7.75	12.50	0.54
High shade (75 per cent shade)	57.0	2.90	7.18	9.74	0.57

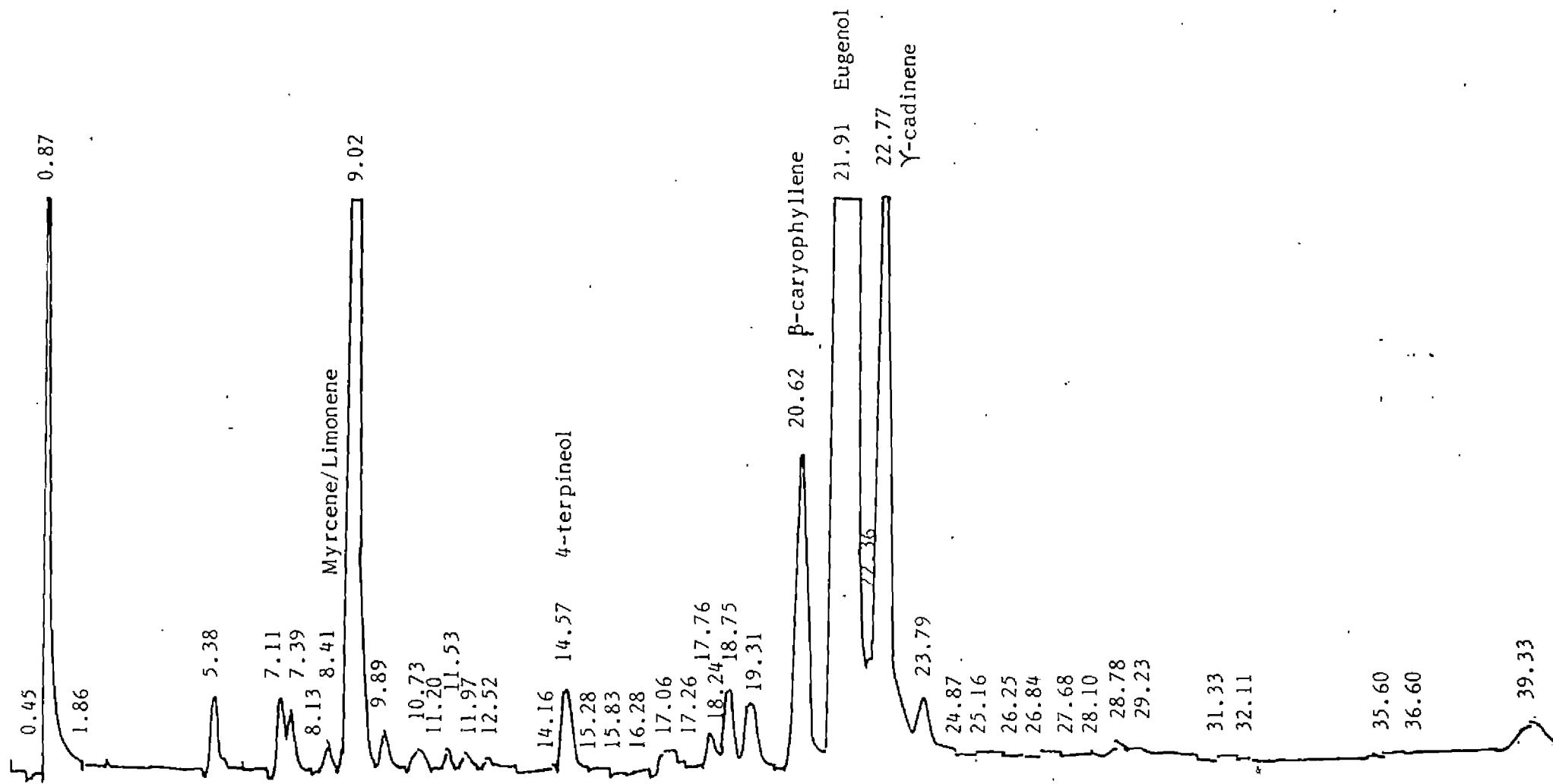


FIG.12. GLC PROFILE OF OIL AT 0 PER CENT SHADE LEVEL

RT	AREA	AREA %
1.86	29780	0.055
5.38	233700	0.432
7.11	214100	0.396
7.39	182100	0.337
8.13	9224	0.017
8.41	87060	0.161
9.02	5149000	9.527
9.89	140100	0.259
10.73	97060	0.180
11.20	13880	0.026
11.53	64960	0.120
11.97	54810	0.101
12.52	36120	0.067
14.16	37590	0.070
14.57	295500	0.547
15.28	22630	0.042
15.83	5722	0.011
16.28	11940	0.022
17.06	53830	0.100
17.26	95240	0.176
17.76	32820	0.061
18.24	144000	0.266
18.75	320500	0.593
19.31	376200	0.696
20.62	1414000	2.616
21.91	38930000	72.034
22.36	143600	0.266
22.77	3295000	6.097
23.79	522300	0.966
24.87	77080	0.143
25.16	188100	0.348
26.25	113400	0.210
26.84	200800	0.372
27.68	112000	0.207
28.10	83920	0.155
28.78	225200	0.417
29.23	299700	0.555
31.33	120400	0.223
32.11	89420	0.165
35.60	195900	0.362
36.60	85860	0.159
39.33	239100	0.442

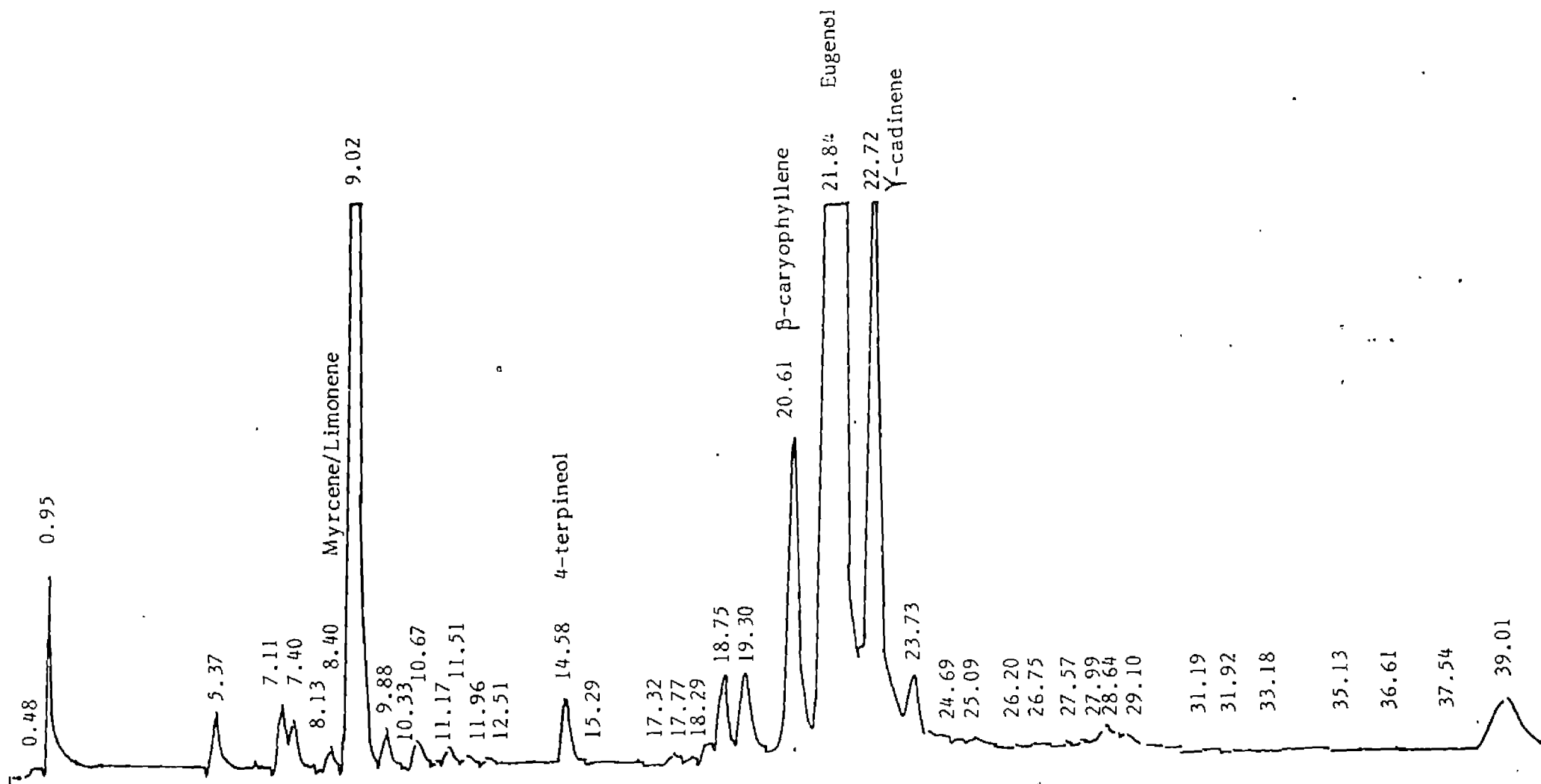


FIG.13 - GLC PROFILE OF OIL AT 25 PER CENT SHADE LEVEL

RT	AREA	AREA %
0.48	402	0.001
0.95	394100	0.776
5.37	175500	0.345
7.11	181100	0.356
7.40	141800	0.279
8.13	4829	0.010
8.40	66740	0.131
9.02	4304000	8.471
9.88	131800	0.259
10.33	1969	0.004
10.67	124300	0.245
11.17	28470	0.056
11.51	76440	0.150
11.96	56940	0.112
12.51	46220	0.091
14.58	306200	0.603
15.29	24580	0.048
17.32	58680	0.115
17.77	39690	0.078
18.29	94960	0.187
18.75	355200	0.699
19.30	507000	0.998
20.61	1578000	3.106
21.84	35120000	69.122
22.72	3615000	7.115
23.73	641000	1.262
24.69	99420	0.196
25.09	216000	0.425
26.20	146900	0.289
26.75	222900	0.439
27.57	142500	0.280
27.99	104200	0.205
28.64	308100	0.606
29.10	385400	0.759
31.19	176200	0.347
31.92	163100	0.321
33.18	158300	0.313
35.13	97540	0.192
36.61	24340	0.048
37.54	23720	0.047
39.01	465200	0.916

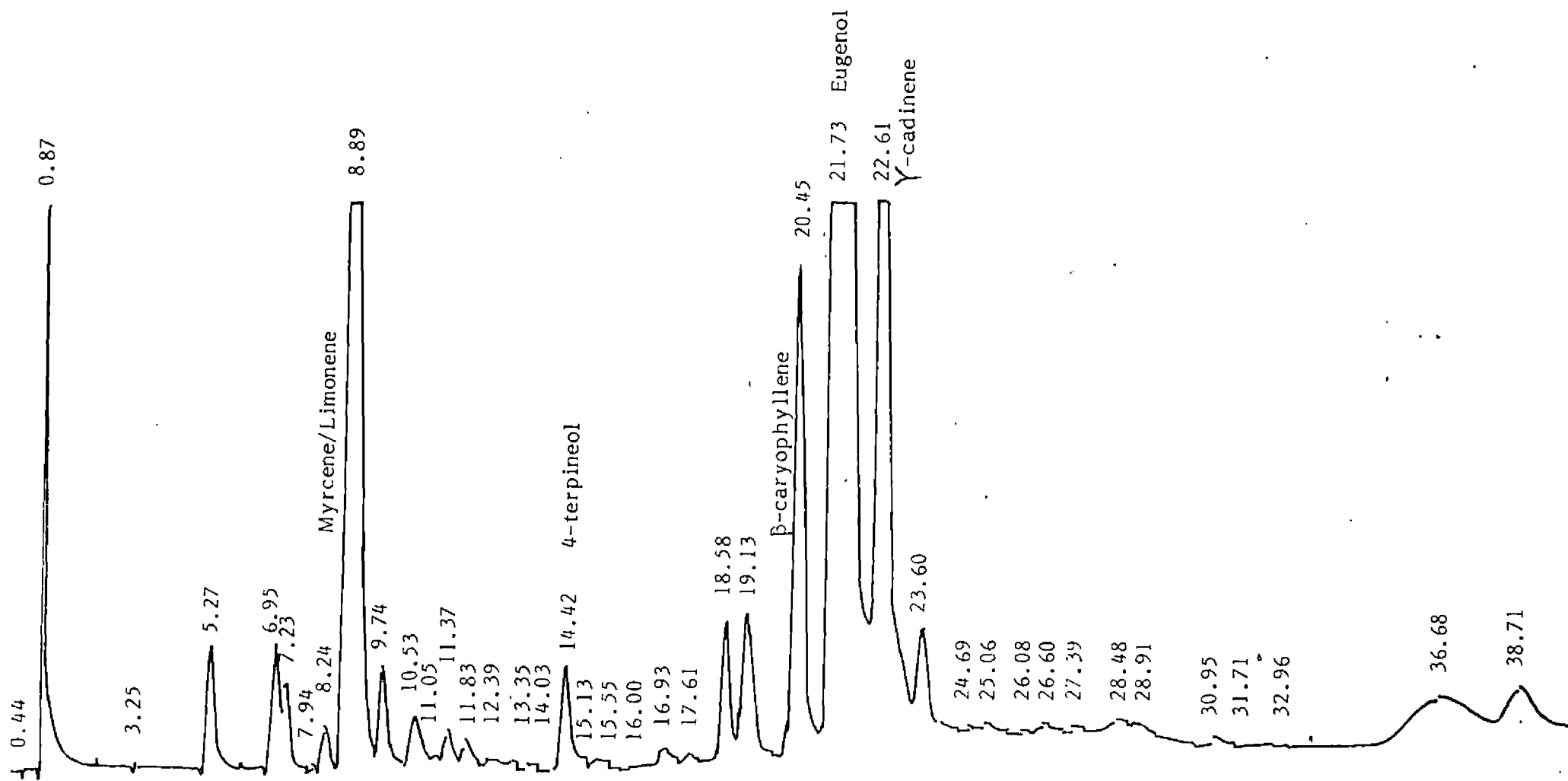


FIG.14. GLC PROFILE OF OIL AT 50 PER CENT SHADE LEVEL

RT	AREA	AREA %
0.44	50	0.000
0.87	1087000	1.668
3.25	289	0.000
5.27	347200	0.533
6.95	349400	0.536
7.23	258300	0.396
7.94	9846	0.015
8.24	123100	0.189
8.89	8182000	12.552
9.74	329100	0.505
10.53	227300	0.349
11.05	40090	0.062
11.37	118100	0.181
11.83	93720	0.144
12.39	32280	0.050
13.35	4278	0.007
14.03	7094	0.011
14.42	353300	0.542
15.13	32300	0.050
15.55	3124	0.005
16.00	6742	0.010
16.93	99840	0.153
17.61	60310	0.093
18.58	587000	0.901
19.13	764600	1.173
20.45	2312000	3.547
21.73	39500000	60.597
22.61	5049000	7.746
23.60	863800	1.325
24.69	193200	0.296
25.06	390200	0.599
26.08	228100	0.350
26.60	339100	0.520
27.39	290600	0.446
28.48	536800	0.824
28.91	551200	0.846
30.95	215700	0.331
31.71	127900	0.196
32.96	83360	0.128
36.68	872200	1.338
38.71	313100	0.790



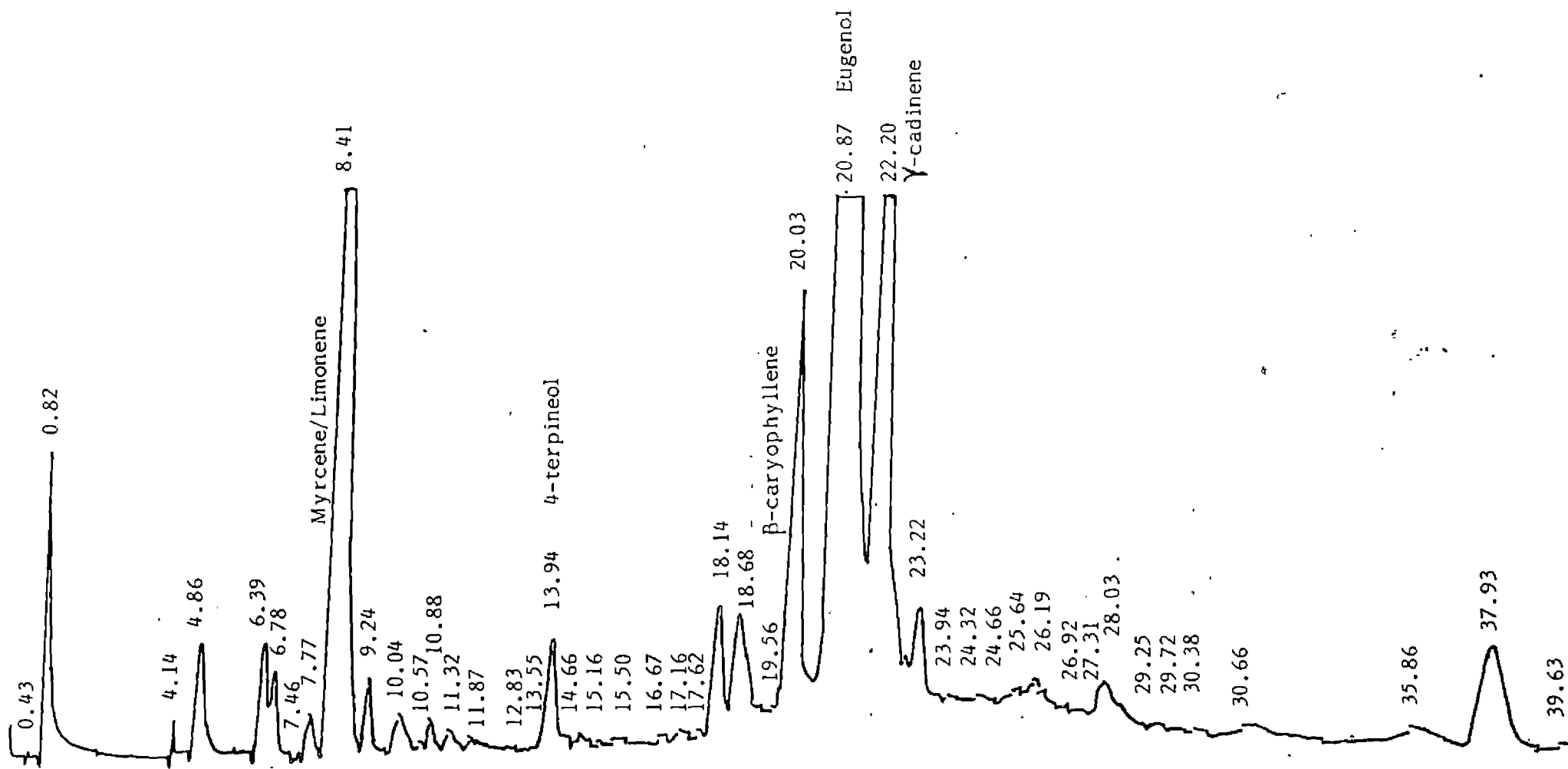


FIG.15\_ GLC PROFILE OF OIL AT 75 PER CENT SHADE LEVEL

RT	AREA	AREA %
0.43	62	0.000
0.82	522100	0.741
4.14	135800	0.019
4.86	445700	0.633
6.39	336000	0.477
6.78	239600	0.340
7.40	12650	0.018
7.77	131000	0.186
8.41	6862000	9.740
9.24	276000	0.392
10.04	200800	0.285
10.57	35060	0.050
10.88	113600	0.161
11.32	72960	0.104
11.87	35250	0.050
12.03	455200	0.646
12.83	8940	0.013
13.55	29230	0.041
13.66	22180	0.031
13.94	405300	0.575
14.66	68100	0.097
15.16	27990	0.040
15.50	60790	0.086
16.67	150700	0.214
17.16	105900	0.150
17.62	71000	0.101
18.14	636000	0.903
18.68	1237000	1.756
19.56	448100	0.636
19.75	406600	0.577
20.03	2104000	2.986
20.87	40830000	57.953
22.20	5060000	7.182
23.22	1146000	1.627
23.94	245900	0.349
24.32	259600	0.368
24.66	447900	0.650
25.64	541300	0.768
26.19	836400	1.187
26.92	313200	0.445
27.31	273300	0.388
28.03	998800	1.418
29.25	193400	0.275
29.72	207000	0.294
30.38	215900	0.306
31.66	951600	1.351
35.86	980800	1.392
37.93	1228000	1.743
39.63	181600	0.258

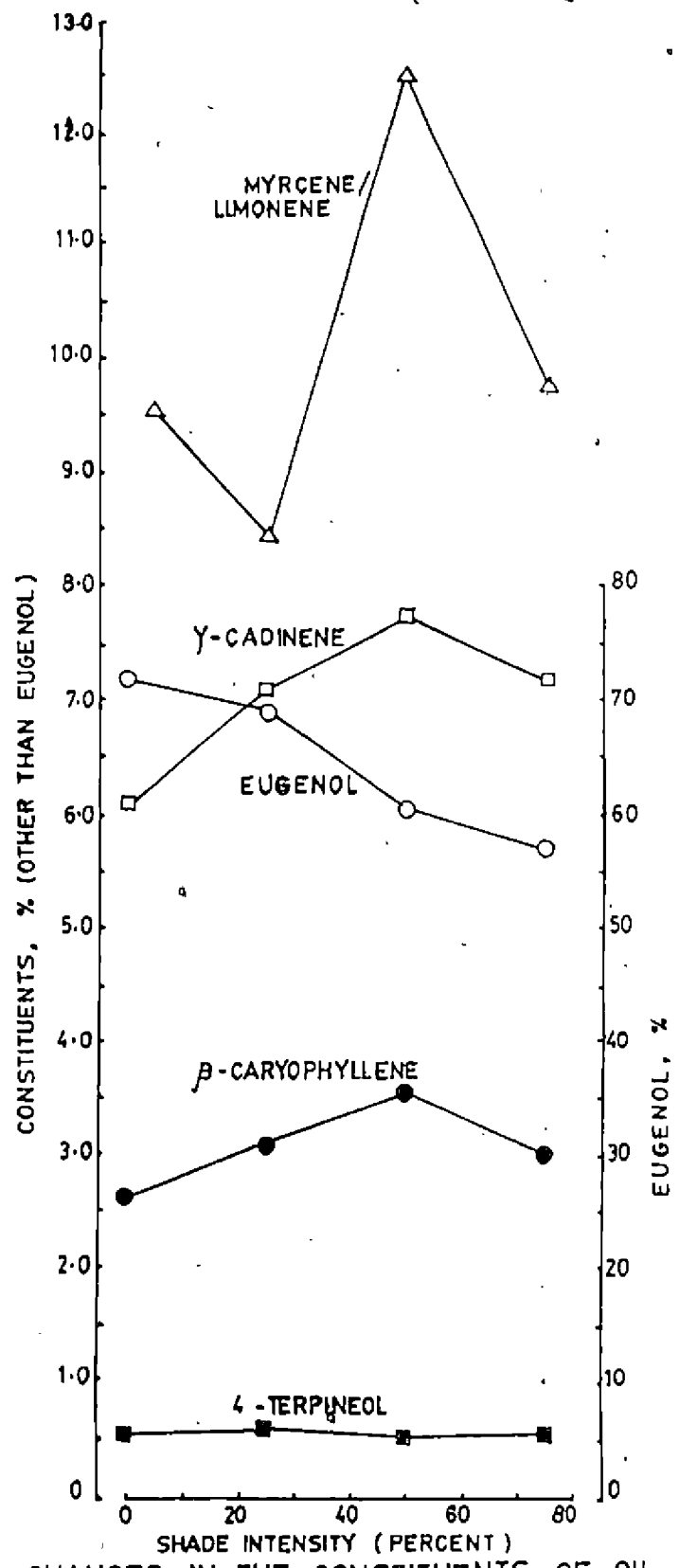


FIG. 16- CHANGES IN THE CONSTITUENTS OF OIL AT DIFFERENT SHADE LEVELS

Table 16. Effect of levels of shade on eugenol content (determined by chemical analysis) and eugenol yield

Levels of shade	Eugenol content, %	Eugenol yield, kg ha <sup>-1</sup>
No shade (100 per cent light)	80.60	72.24
Low shade (25 per cent shade)	79.12	32.00
Medium shade (50 per cent shade)	71.20	21.53
High shade (75 per cent shade)	70.40	14.61
SEm <u>+</u>	0.50	4.07
CD (0.05)	1.538	12.546

myrcene,  $\gamma$ -cadinene and  $\beta$ -caryophyllene were found to increase as the levels of shade increased.

### 2.3.3. Eugenol content (determined by chemical method) and eugenol yield

The results indicated that the eugenol content determined by chemical method manifested similar trend as in GLC analysis (Table 16). Highest eugenol content in the oil (80.60 per cent) was observed under open condition and was on par with low shade level. High intensities of shade recorded significantly lower eugenol content.

The eugenol yield for the different intensities of shade varied from 14.61 to 72.24 kg ha<sup>-1</sup>. Maximum eugenol was produced by the crop grown under full illumination and it was significantly superior to all other treatments. The eugenol yield decreased with increasing levels of shade and the lowest value was recorded at the highest shade level. The eugenol yield per unit area is determined by the oil yield and eugenol content of the oil. In the present study both of them were the highest in the crop grown under open condition and naturally the eugenol yield was also the highest under this treatment.

### 2.4. Effect of levels of shade on plant nutrient content and uptake

Data relating to the nutrient content and uptake are presented in Tables 17 to 20.

#### 2.4.1. Nitrogen content

Though there was significant difference due to treatments, varying levels of shade did not show any particular trend in respect to the N content of the herbage (Table 17). As the intensity of shade increased from full illumination to 25 per cent shade the N content also increased significantly. However, further increase in the intensity of shade significantly decreased the N content. Muramoto et al. (1967) also observed a deterioration in the photosynthetic mechanism of leaves due to shading and a consequent reduction in the nitrogen content of the leaves.

#### 2.4.2. Phosphorus content

The influence of different levels of shade on the P content of the herbage was significant. There was not much variation in the P content under the open condition and that under 25 and 50 per cent shade. At the highest shade intensity level tried the P content increased significantly and it was significantly higher than all the other treatments.

#### 2.4.3. Potassium content

With increasing levels of shade the K content of the herbage increased significantly. The highest K content of 2.82 per cent recorded under 50 per cent shade was on par with 75 per cent shade. The tendency of potassium content was to increase

Table 17. Effect of levels of shade on N, P, K, Ca and Mg content of 'clocimum, per cent

Levels of shade	Nitrogen	Phosphorus	Potassium	Calcium	Magnesium
No shade (100 per cent light)	1.91	0.39	2.03	3.47	0.49
Low shade (25 per cent shade)	2.23	0.40	2.23	3.37	0.52
Medium shade (50 per cent shade)	1.36	0.36	2.82	3.88	0.40
High shade (75 per cent shade)	1.47	0.69	2.81	2.84	0.48
SEm $\pm$	0.08	1.01	0.18	0.17	0.02
CD (0.05)	0.250	0.030	0.573	0.523	0.075

with increase in shade levels. The role of potassium ions under stress conditions is well established (Tisdale and Nelson, 1956). George (1982) also observed increased K content under high shade in cowpea and groundnut. Similar results of increase in content of K by shading have been widely reported in several crops (Myhr and Saebo, 1969; Cantilliffe, 1972 and Rodriguez et al., 1973).

#### 2.4.4. Calcium content

Though the calcium content of the plants was influenced significantly due to treatments no significant difference was recorded under 0, 25 and 50 per cent shade levels. The minimum value of 2.84 per cent observed under intense shade of 75 per cent was significantly lower than the values obtained in all the other treatments.

#### 2.4.5. Magnesium content

The magnesium content of the herbage was also influenced significantly with different levels of shade. But no definite trend was noticed with changes in the intensity of shade. The values varied from 0.40 to 0.52 per cent.

#### 2.4.6. Iron content

Maximum iron content was recorded by the treatment of low shade and it was on par with no shade. Higher intensities of shade tried significantly decreased the content (Table 18).



Table 18. Effect of levels of shade on Fe, Mn and Zn of clove, ppm

Levels of shade	Iron	Manganese	Zinc
No shade (100 per cent light)	411.06	92.46	17.98
Low shade (25 per cent shade)	521.42	98.00	25.40
Medium shade (50 per cent shade)	242.56	75.70	56.90
High shade (75 per cent shade)	267.50	72.00	26.90
SEm ±	18.73	6.54	7.91
CD (0.05)	150.155	20.155	24.388

#### 2.4.7. Manganese content

There was significant difference in the manganese content due to treatments. The manganese content showed a tendency to decrease under the high intensity of shade.

In general, the decrease in Fe and Mn contents at higher intensities of shade indicated the inability of the plants to absorb these micronutrients under intense shade.

#### 2.4.8. Zinc content

The zinc content of the herbage increased with increase in the intensity of shade from no shade to medium shade. The medium shade level recorded maximum value (56.90 ppm) and with further increase in the shade level the zinc content decreased.

#### 2.4.9. Nitrogen uptake

There was significant difference in the uptake of N with different levels of shade and the uptake decreased with increase in the intensity of shade. The maximum uptake was recorded under full illumination followed by the light shade level (25 per cent) and both these treatments were significantly higher than the treatments at higher shade intensities. The uptake of N followed the same trend as that of dry matter production (Table 19).

Table 19. Effect of varying levels of shade on uptake of N, P, K, Ca and Mg, kg ha<sup>-1</sup>

Levels of shade	Nitrogen	Phosphorus	Potassium	Calcium	Magnesium
No shade (100 per cent light)	296.34	58.54	312.64	529.59	74.43
Low shade (25 per cent shade)	207.23	37.16	206.68	315.12	48.38
Medium shade (50 per cent shade)	112.17	30.10	233.61	322.23	33.55
High shade (75 per cent shade)	88.55	41.48	165.08	170.35	28.57
SEm ±	18.90	2.42	19.36	28.47	3.44
CD (0.05)	58.239	7.468	59.649	87.736	10.592

#### 2.4.10. Phosphorus uptake

Different levels of shade influenced significantly the uptake of P. The highest uptake was recorded by the plants grown in the open and it was significantly superior to all other shade levels. The results revealed that as in the case of N the uptake was influenced mainly by the dry matter yield.

#### 2.4.11. Potassium uptake

Uptake of K was maximum and significantly higher under full illumination than all other treatments. The lowest uptake was recorded at the highest shade level and it was significantly lower than 0 and 50-per cent shade levels. The effect of the higher K content recorded at higher intensities of shade was not reflected in the uptake as the herbage yields obtained under the shaded condition was remarkably low.

#### 2.4.12. Calcium uptake

The calcium uptake decreased with increase in the intensity of shade. The maximum value was recorded at the open condition and it was significantly higher than that at all other shade levels. Though no definite trend was noticed in the calcium content of the plants due to shade levels, the uptake showed a definite trend and it again confirmed the influence of dry matter production in determining the uptake.

#### 2.4.13. Magnesium uptake

The uptake of Mg was influenced significantly by varying levels of shade and the uptake increased linearly with increasing intensity of light. The highest value recorded under full illumination was significantly higher with an uptake of  $74.43 \text{ kg ha}^{-1}$  followed by the next succeeding intensity of shade (25 per cent). The lowest value was recorded with 75 per cent shade level. The uptake of magnesium also followed the same pattern as that of the dry matter production.

#### 2.4.14. Iron uptake

The uptake of iron increased almost linearly with increasing intensity of illumination ( $6.18 \text{ kg ha}^{-1}$ ) and the minimum under 75 per cent shade ( $1.59 \text{ kg ha}^{-1}$ ) (Table 20).

#### 2.4.15. Manganese uptake

As in iron uptake similar trend was observed for Mn uptake also. Maximum uptake was recorded with 100 per cent light and it was statistically higher than the values obtained at the medium and intense shade levels. The uptake of Mn at different levels of shade ranged from  $0.43$  to  $1.40 \text{ kg ha}^{-1}$ .

#### 2.4.16. Zinc uptake

Different levels of shade showed no significant influence on the Zn uptake by the plants. The range in the values, however, varied between  $0.16$  and  $0.47 \text{ kg ha}^{-1}$ .

Table 20. Effect of levels of shade on uptake of Fe, Mn and Zn, kg ha<sup>-1</sup>

Levels of shade	Iron	Manganese	Zinc
No shade (100 per cent light)	6.18	1.40	0.28
Low shade (25 per cent shade)	4.73	0.90	0.23
Medium shade (50 per cent shade)	2.02	0.62	0.47
High shade (75 per cent shade)	1.59	0.43	0.16
SEm ±	0.49	0.09	0.08
CD (0.05)	1.497	0.272	NS

NS = Not significant

From the results of the present study it can be concluded that for obtaining maximum quantity of quality oil per unit area the crop has to be grown under open condition and has to be harvested at the early flowering stage (60 days after planting). If only one cycle of harvest is considered in the shade experiment then 25 per cent shaded plants can be considered as good as that under open. The physico-chemical properties of oil obtained showed that it can form a good substitute for the high priced clove and cinnamon oils for the production of eugenol. The clove oil has more similarities with cinnamon oil in respect to the physical properties and also the eugenol content but differed very much from clove oil with regard to these properties.

*Summary*

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## SUMMARY

An investigation was conducted at the Aromatic and Medicinal Plants Research Station, Odakkali to study the effect of five harvest intervals of 60, 75, 90, 105 and 120 days and four levels of shade viz., 0, 25, 50 and 75 per cent on the yield and quality of clove oil. Two field experiments were conducted separately in a randomised block design during 1988-90. In addition to the above harvest intervals, intervals less than 60 days were also tried as an observation study on the ratoon crop. This was necessitated since the oil obtained at the shortest harvest interval of 60 days contained highest oil and eugenol content. The results of the investigations are summarised below under two heads.

1. Effect of intervals of harvest on quantitative and qualitative parameters of clove oil, nutrient content and uptake.
2. Effect of different levels of shade on yield and quality of clove oil, nutrient content and uptake.

### **Effect of intervals of harvest on quantitative and qualitative parameters of clove oil, nutrient content and uptake**

Different intervals of harvest did not influence significantly growth and most of the yield attributes. The crop harvested at 75 days interval, however, recorded the maximum number of

flowered branches, length of inflorescence and leaf area.

Harvest intervals showed significant influence on herbage yield, oil yield and oil content and the maximum value for all these parameters was recorded at the 60 day interval which was on par with 75 day interval. In the observation study conducted on the ratoon crop, the oil content showed an increasing trend as the interval increased from 40 to 60 days and thereafter it showed a decreasing trend. Sixty days harvest interval coincided with early flowering stage of the crop.

All the physical properties of oil except refractive index was significantly influenced by harvest intervals. Increasing the interval of harvest from 60 to 120 days decreased the specific gravity, refractive index and solubility in 70 per cent alcohol and increased the optical rotation. But when the harvest interval was increased from 40 to 60 days, the specific gravity and refractive index of oil increased and solubility of oil decreased.

Analysis of the oil by GLC showed that eugenol content decreased with increase in the harvest intervals from 60 to 120 days. The contents of all the other components identified viz.,  $\beta$ -caryophyllene, limonene,  $\gamma$ -cadinene increased with increase in the harvest intervals. Analysis of the oil samples collected from the ratoon crop at 40 and 60 days harvest intervals showed relative stability in the eugenol content but the percentage of

the other components increased with increase in the harvest interval. The eugenol content of oil as determined by chemical method also showed similar trend as in GLC.

Eugenol yield was significantly influenced by the different harvest intervals. Harvest interval of 60 days gave the maximum value and it decreased with increase in the harvest intervals.

As the intervals of harvest increased, the N, P, K and Mg contents of the herbage decreased. The maximum content for all these nutrients recorded at the 60th day was on par with 75 days harvest interval except in the case of P. No significant difference in the concentration of the micronutrients Fe, Mn and Zn resulted with variation in the harvest intervals.

No significant variation in the uptake of N, P, Ca and Mg was noticed due to different intervals of harvest. But the K uptake was influenced significantly and it showed a tendency to decrease with increase in the intervals of harvest. Although no particular trend was observed in the uptake of micronutrients, the highest uptake was recorded for the 60th day interval for all the micronutrients.

#### **Effect of different levels of shade on the growth, yield attributes, yield and quality of clove oil**

The declining effect of shade on photosynthesis and translocation was reflected in all the primary yield components. The

effect of shade on plant height and spread was positive upto intermediate shade level whereas its effect on number of branches, number of flowering shoots, length of inflorescence and leaf area was negative.

The flowering and attainment of maturity were delayed progressively with increasing intensities of shade. Two harvests could be obtained under open condition whereas only a single harvest was possible in all the other treatments. Hence the total herbage yield was highest in the open which showed a decreasing trend with increase in the intensities of shade. The highest values of oil content and oil yield were also recorded by the plants grown in the open and it was significantly superior to the rest of the treatments. All the harvests were made at 50 per cent flowering stage of the crop. If only a single harvest of the crop in the open is considered the herbage yield, oil yield and oil content under open will be almost equal to that under 25 per cent shade.

The physical properties of oil were also significantly influenced by varying levels of shade. The values for all the parameters decreased with increase in the intensities of shade upto the medium shade level of 50 per cent and thereafter showed an increasing trend.

The GLC analysis of the oil showed that eugenol content of the oil decreased with increase in the levels of shade. The highest value for eugenol was recorded under the open condition. The contents of the other constituents of the oil viz.,  $\beta$ -caryophyllene,  $\gamma$ -cadinene and myrcene showed an increasing trend upto the intermediate shade level. The eugenol content determined by chemical method showed significant variation due to treatments and it decreased with increase in the intensities of shade. Maximum content was recorded under open condition and it was on par with that under 25 per cent shade.

Eugenol yield was also significantly influenced by the different shade levels. Open condition gave the maximum value which decreased progressively upto the highest shade intensity.

Though significant difference in the N, P, K, Ca and Mg contents were noticed with varying levels of shade only K content showed a persistent increase with shading of the crop. No definite pattern was noticed in the contents of other nutrients with change in the intensities of shade. Though the contents of micronutrients was also influenced significantly due to treatments no regular trend was observed.

The uptake of all the nutrients increased with increasing intensities of light. It was also noted that the dry matter production had the dominant role in deciding the total uptake and that the higher dry weight of the unshaded plots compensated for the higher contents of potassium in shaded plots.

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**QUALITY OF OIL OF CLOCIMUM (*Ocimum gratissimum*  
Linn.) AS INFLUENCED BY STAGES OF  
HARVEST AND SHADE**

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**ABSTRACT OF A THESIS**

Submitted in partial fulfilment of the  
requirement for the degree

**Master of Science in Agriculture**

Faculty of Agriculture  
Kerala Agricultural University

Department of Soil Science and Agricultural Chemistry

**COLLEGE OF HORTICULTURE**

Vellanikkara, Thrissur

1990



## ABSTRACT

Clocimum (Ocimum gratissimum Linn.), an essential oil crop introduced in Kerala from Jammu, nearly a decade ago, is slowly replacing the other eugenol yielding sources viz., clove and cinnamon oils. Several agro-techniques have yet to be standardised for the commercial cultivation of this crop in the State. The present studies were undertaken at the Aromatic and Medicinal Plants Research Station, Odakkali during 1988-90. Two field experiments were laid out separately one to obtain information on the optimum time of harvest for getting maximum yield of quality oil and the other to study the effect of shade on the production and quality of oil. The average removal of N, P, K, Ca, Mg, Fe, Mn and Zn from the soil by clocimum has also been worked out.

The treatments in one experiment consisted of five intervals of harvest (60, 75, 90, 105 and 120 days) and four levels of shade (0, 25, 50 and 75 per cent) were tried in the second experiment. Both experiments were laid out in randomised block design. The required intensity of shade was provided by erecting artificial pandals. A Aplab luxmeter was used for adjusting the shade intensities.

The investigations revealed that herbage yield, oil yield and oil content were maximum at 60 days harvest interval (early

flowering stage) and it was on par with 75 days harvest interval. With further increase in the harvest intervals these parameters were found to decrease. Sixty days harvest interval also recorded the maximum eugenol content and it was significantly superior to all other treatments. The concentration and uptake of macro and micronutrients were also maximum at this interval.

Among the shade levels tried the herbage yield was maximum under open condition as the plants flowered early and so two harvests could be taken under this treatment while only one harvest could be obtained for the other treatments. The oil yield, oil content, eugenol content were also maximum under open conditions. The content of almost all nutrients studied did not show any particular trend except in the case of K which showed a tendency to increase with shading. The uptake of all nutrients was maximum under full illumination which decreased with increasing shade intensity.

The results thus indicated that the optimum interval of harvest is between 60 and 75 days i.e., harvesting the crop between early and maximum flowering stages. The results also revealed that maximum yield and quality of oil in clove can be obtained only if there is ample light infiltration. But if only a single harvest is considered the crop under 25 per cent shade can be seen to give equally good quality oil but for a lesser number of harvest that those grown under full illumination.

A high eugenol content of eighty per cent obtained for clocimum in the present study also indicates that it can definitely be used as an alternative and cheap substitute to clove and cinnamon oils.