Gas Chromatographic-Mass Spectrometric Determination of the Flavour Compositions of *Ocimum basilicum, Myrtus communis* and *Mentha arvensis*

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Summary: *Ocimum basilicum, Myrtus communis* and *Mentha arvensis* were subjected to steam distillation for the extraction of essential oils. For quantitative and qualitative analysis, GC-MS techniques were used. In case of *Ocimum basilicum*, 13 compounds were identified, while in *Myrtus communis* and *Mentha arvensis* 15 and 12 compounds were characterized, respectively.

Introduction

Ocimum basilicum (Basil) is an annual herbaceous plant belonging to family Labiatae [1]. It is native to Asia and other tropical regions cultivated as a culinary herb throughout Europe [2]. It is also found in Kashmir, Gilgit and Chitral, areas of Pakistan [3]. The plant possesses a clove like scanty aromatic odour, somewhat saline taste, used as a flavouring agent and in perfumery formulations. Universally four types of basils have been recognized [4]. European or methyl chavicol-type basil contains methyl chavicol and camphor but not linalool as the main constituents. Reunion or Comoro-type is of doubtful nomenclature which is now produced in Comoro found to contains methyl chavicol and camphor but not linalool. Methyl cinnamate-type which is distilled in Bulgaria, Sicily, Egypt and India contains methyl chavicol, linalool and substantial amount of methyl cinnamate. Eugenol-type which is distilled in Java, Seychelles, Samoa and Russia contains eugenol as the principal constituent. A lot of work has been published on the chemical constituents of basil oils [5-6], yet no work has been reported on the oils of different species of basils available in Pakistan, except of a paper reported by Riaz *et al.* on the GC-MS analysis of basil oil (Comoro-type).[7].

Myrtus communis L. (Black Berry) is an evergreen shrub belonging to family Myrtaceae [8]. It is found in Sindh, Baluchistan, North Waziristan, Kurrum valley, Panjkora valley, Peshawar and lower Swat [9]. An infusion or tincture of leaves found to be useful for the treatment of prolapsus and leucorrahea. The powdered leaves are also used in some pulmonary disorders [10]. Many papers have been published on the essential oils of this plant. The more recent work have been reported by Fujata [11] who investigated the essential oil by GC-MS technique.

Mentha arvensis L. is a perennial herb spread by roots, belonging to family Labiatae [12]. It is also called Japanese Mint. This plant has been known and grown in Japan since antiquity. The oil of Japanese mint was first analyzed by Shinozaki [13-14]. Handa and co-workers [15] and later on by Lawrence and co-worker [16-18] extensively analyzed this oil.

In Pakistan, it is cultivated in Hazara, Peshawar, Chitral, Swat and Baluchistan. In present paper GC-MS techniques have been used to analyze the flavour compositions of the essential oils of these plants grown in Pakistan.

Results and Discussion

Gas chromatographic-mass spectromtric results of the volatile components of *Ocimum basilium, Myrtus communis* and *Mentha arvensis* along with the retention time and percent compositions are summarized in Tables **1-3**. The compounds were identified on the basis of their EI-MS (electron-impact mass spectrum) [19], mass spectral database libraries, spectra of authentic compounds reported in literature [20], fragmentation pattern and retention time. Comparing with the mass spectral database is an

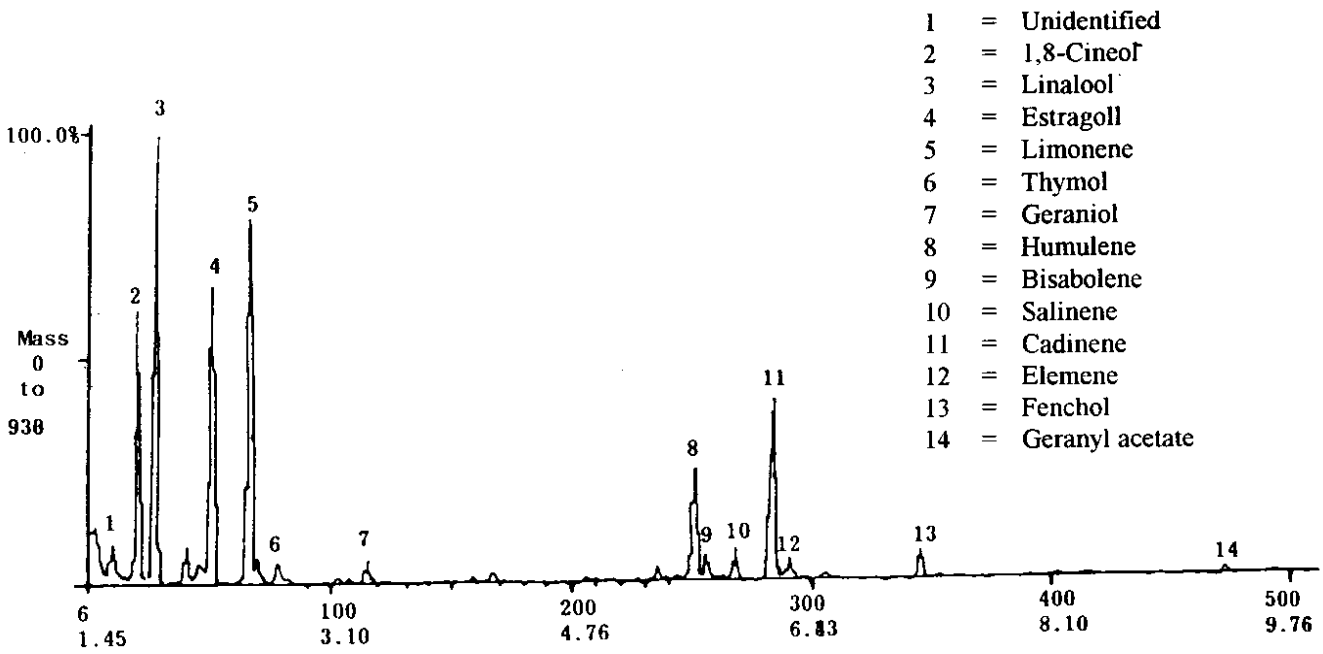

Fig. 1: Gas Chromatogram of Essential oil Constituents of *Ocimum basilicum*.

Table - 1 :Compounds Identified from *Ocimum basilicum* as Determined by GC-Mass spectrum along with their Peak Areas and Percent Concentrations.

Peak No.	Retention Time (min.)	Identity	Molecular Weight	Peak Area	Percent Concentration
1	1.46	Unidentified	126	8.55	7.60
2	1.75	1,8-Cineol	154	20.60	18.31
3	1.88	Linalool	154	19.6	17.42
4	2.26	Estragol	148	16.77	14.91
5	2.51	Limonene	136	17.12	15.22
6	2.71	Thymol	150	1.12	0.99
7	3.35	Geraniol	154	1.50	1.33
8	5.60	Humulene	204	5.35	4.75
9	5.68	Bisabolene	204	1.47	1.30
10	5.88	Selinene	204	1.83	1.62
11	6.15	Cadinene	204	13.90	12.35
12	6.26	Elemene	204	1.80	1.60
13	7.18	Fenchol	154	1.60	1.42
14	9.30	Geranyl acetate	196	1.25	1.11

excellent method for the identification of organic compounds [20-21].

In case of *Ocimum basilicum*, 13 volatile compounds were identified. Compound at retention time 2.71 minutes was identified as thymol for the first time from this plant. From the gas chromatogram it appeared that 1,8-cineol, linalool, limonene and estragol or methyl chavicol are in higher concentrations, while thymol have low percent concentration (0.99%). The variety of *Ocimum* under investigation for essential oil cannot be identified on the basis of chemical constituents and morphological examination. The presence of thymol alone is very unusual feature which indicate that the variety under investigation could very well be a new variety or hybrid. There are other sub species such as *Ocimum gratissum* found in India contains both thymol and eugenol as the principal constituents, while the present Ocimum species contains only small amount of thymol 0.99% but no eugenol.

The GC-Mass spectra of *Myrtus communis* revealed the presence of 15 known components and 5 unknown components. The names of known constituents along with their molecular weights and retention times are given in Table-2. The gas chromatogram of *M. communis* revealed that 1,8-cineol, pinene, limonene and geraniol are in higher concentrations while myretenol, terpenyl acetate and myretnyl acetate concentrations were 7.78%, 4.65% and 0.17% respectively. The sesquiterpenes and fenchol were found to be in low concentrations.

In the case of *Mentha arvensis* L. the GC-mass spectra confirmed the presence of 13 identified and one unidentified components. The

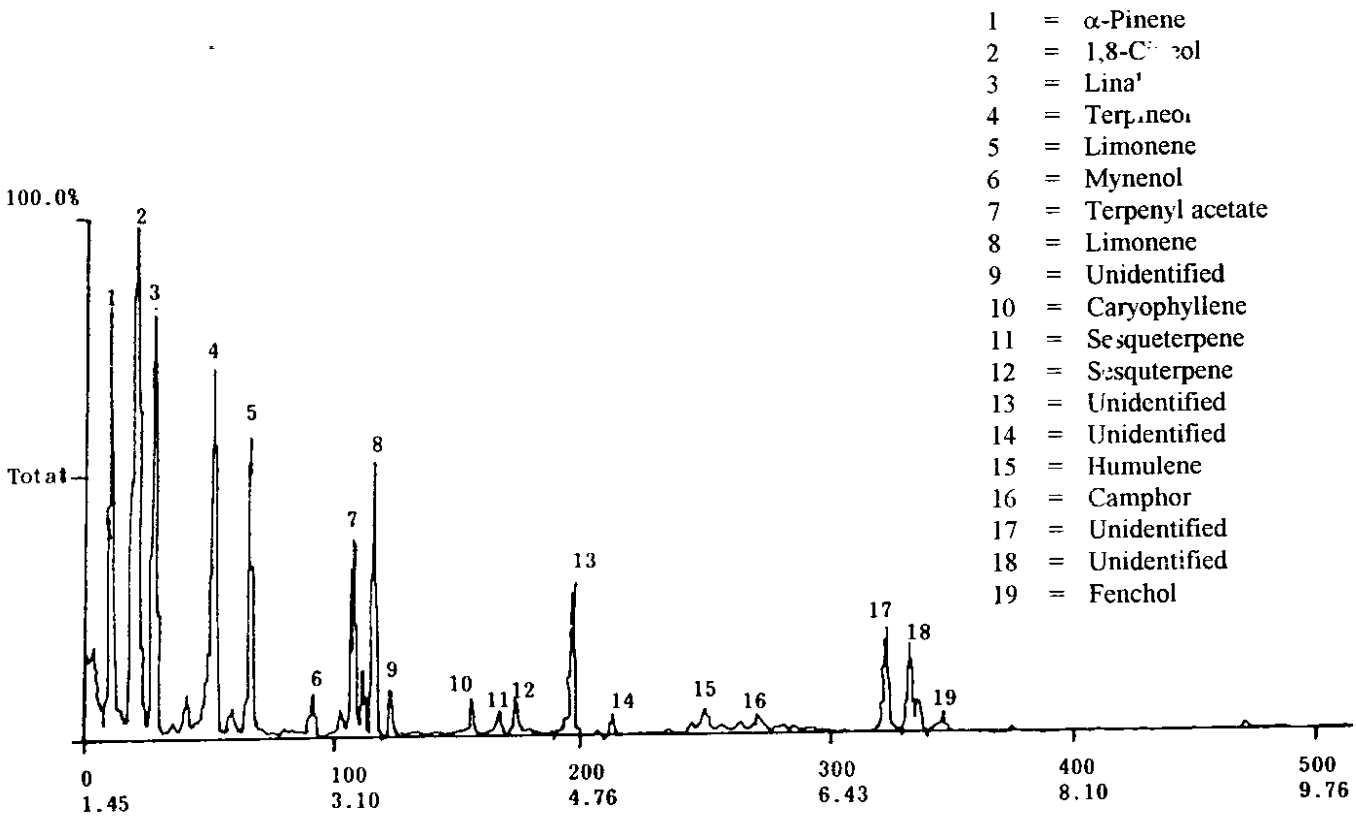

Fig. 2: Gas Chromatogram of Essential oil Constituents of *Myrtus communis*.

Table - 2 : Compounds Identified from *Myrtus communis* as Determined by GC-Mass spectrum along with their Peak Areas and Percent Concen-rations.

Peak No.	Retention Time (min.)	Identity	Molecular Weight	Peak Area	Percent Concen-tration
1	1.58	α-Pinene	136	38.45	16.18
2	1.76	1,8-Cineol	154	45.55	19.91
3	1.88	Linalool	154	26.67	11.66
4	2.28	Terpineol	154	22.92	10.02
5	2.53	Limonene	136	29.15	12.74
5	2.95	Myretenol	152	17.08	7.78
7	3.23	Terpenyl Acetate	196	10.65	4.65
8	3.30	Myretnyl Acetate	196	0.39	0.17
9	3.38	Geraniol	154	24.25	10.60
10	3.48	Unidentified	191	2.54	1.11
11	4.03	β-Caryophyllene	204	2.08	1.22
12	4.21	Sesquiterpene	204	1.75	0.76
13	4.33	Sesquiterpene	204	1.77	0.77
14	4.73	Unidentified	236	1.04	0.61
15	4.98	Unidentified	252	0.22	0.09
16	5.61	Humulene	204	0.37	0.16
17	5.95	Camphor	152	0.3	0.13
18	6.83	Unidentified	298	0.92	0.4
19	6.98	Unidentified	296	0.69	0.3
20	7.20	Fenchol	154	0.1	0.04

compound at retention time 1.61 minutes which have the molecular weight of m/z 129 and concentration 3.93% could not be identified, while compound eluted at retention time 2.31 minutes have the molecular weight m/z 154 was characterized as menthol has the highest concentration 71.78% in its essential oil. Other compounds such as pipertone, pulegone and sesquiterpenes were in lower concentrations (Table-3).

Table - 3 : Compounds Identified from *Mentha arvensis* as Determined by GC-Mass spectrum along with their Peak Areas and Percent Concentrations.

Peak No.	Retention Time (min.)	Identity	Molecular Weight	Peak Area	Percent Concen-tration
1	1.61	3-Octanol	129	11.5	3.69
2	1.75	1,8-Cineol	136	10.00	3.21
3	1.88	Linalool	154	10.44	3.35
4	2.31	Menthol	156	223.40	71.78
5	2.60	Piperitone	152	17.85	5.73
6	2.83	Pulegone	152	5.16	1.65
7	2.73	Unidentified	-	4.00	1.28
8	4.03	Caryophyllene	204	8.22	2.84
9	5.50	Sesquiterpene	204	2.50	0.80
10	5.56	Sesquiterpene	204	6.00	1.92
11	5.80	Phellendrene	136	4.00	1.28
12	6.13	Sesquiterpene	204	2.25	0.72
13	6.26	Humulene	204	3.56	1.14
14	6.43	Sesquiterpene	222	2.36	0.75

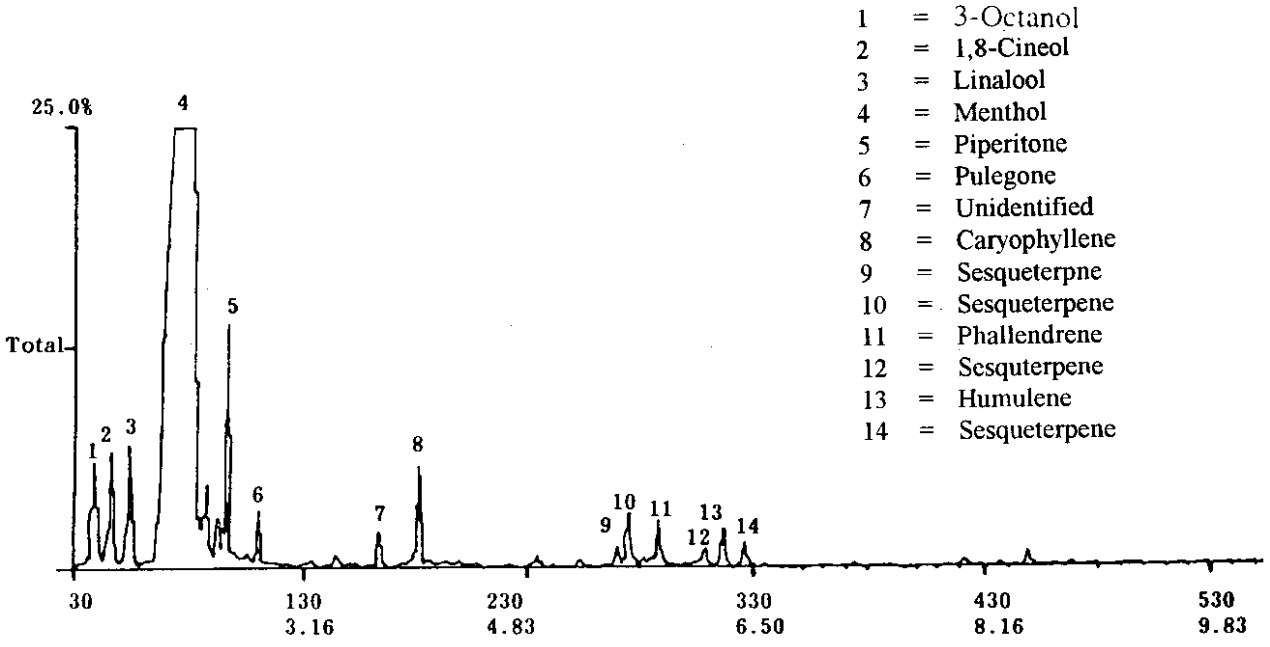

Fig. 3: Gas Chromatogram of Essential oil Constituents of *Mentha arvensis*.

Experimental

Sample Analysis

GC-MS spectra were recorded on GC-model 14A Shimadzu, Japan, GC-mass spectrometer model QP 2000A (70 ev) in EI mode using capillary column (fused silica) 101, CBP1 (25 m x 0.25 mm and film thickness 0.25 μm) using Helium as a carrier gas at the rate of 1 ml/min. The injection temperature was 250 °C while the column temperature was 150 °C, increased at the rate of 10 °C/min. to reach the final temperature 250 °C. The FID temperature was 250 °C. Injected volume was 0.2 μl, while split ratio was 1:100.

Sample Collection

Ocimum basilicum (unknown var.), *Myrtus communis* (leaves) and *Mentha arvensis* were collected for NWFP area (North-West, Pakistan) during the winter of 1993. Plants were identified by the Plant Taxonomist, Department of Botany, University of Peshawar and voucher specimens were deposited in the herbarium of the same department.

Isolation of Essential Oils

The essential oils of each plant material were extracted through steam distillation methods after a week of drying in dark. Plants material were subjected to steam distillation for two hours. The distillate were collected in n-hexane and dried over Na_2SO_4.

Identification of Compounds

The identification of the compounds were achieved on the basis of computerized NBS/NIH/EPA mass spectral database, present at H.E.J. Research Institute of Chemistry, University of Karachi.

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