CHEMOTAXONOMY AND COMPARATIVE ANATOMY OF CERTAIN SPECIES OF OCIMUM OCCURRING IN INDIA

SYNOPSIS

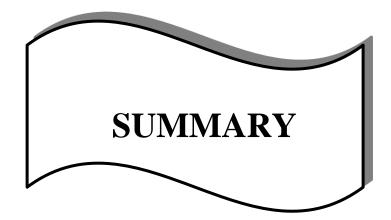
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CHAPTER - VII



Five species of **Ocimum** occurring in Southern India have been differentiated on the basis of their chemical constituents and anatomical characters. Chemical investigations were made with the help of chromatographic techniques. Distinct fingerprints were obtained for each species. The entire chemical study focused solely on the roots of the plants. Comparative anatomical studies were made from microscopic observations. Seed and pollen morphological studies have also been included. Detailed descriptions and indented keys have been provided. The inferences obtained from chemical and anatomical data have been compared with current taxonomic opinion. The role of phytochemistry and comparative anatomy in systematics has been highlighted. The results of the present study will help in the correct identification and standardization of these medicinally important species, besides offering immense scope for further research.

The highlights of the results of this study are listed below :

Section A: Chemotaxonomic studies

GC – MS profiles

- The GC MS chromatograms of the hexane extract of roots showed 24 peaks in O.americanum, 27 peaks in O.basilicum, 29 peaks in O.filamentosum, 28 peaks in O.gratissimum and 14 peaks in O.tenuiflorum.
- The compounds identified in the hexane extract of *O.americanum* roots were found to be benzaldehyde, 4-(1-methylethyl)- (aromatic), phenol, 5- methyl-2-(1-methylethyl)- (phenol), eugenol (phenylpropanoid), tetradecanoic acid, ethyl ester (saturated fatty acid ester) and butyl tetradecanoate (saturated fatty ester).

- The compounds identified in the hexane extract of **O.basilicum** roots were found to be acetic acid, 2-methylpropyl ester (aliphatic ester), benzene, ethyl-(aromatic), benzene, 1, 2-dimethyl- (aromatic), 1-methoxy-4-(1-E-propenyl) benzene (aromatic). beta-caryophyllene (sesquiterpene), 1. 2benzenedicarboxylic acid, dimethyl ester (aromatic ester), beta-bisabolene (sesquiterpene), dodecanoic acid, methyl ester (saturated fatty acid ester), gamma-cadinene (sesquiterpene), cyclohexadecane (fatty hydrocarbon), 1,2methylenedioxy-5,6-dimethoxy-4-allylbenzene (aromatic). pentadecanoic acid, 14-methyl-, methyl ester (saturated fatty acid ester). 8,11octadecadienoic acid, methyl ester, 9-octadecenoic acid (Z)- methyl ester and octadecanoic acid, butyl ester (unsaturated fatty acid esters).
- The compounds identified in the hexane extract of *O.filamentosum* roots were found to be camphene (monoterpene), eugenol (phenylpropanoid), 1- tetradecanol (fatty alcohol), trans-caryophyllene (sesquiterpene), pentadecane (saturated hydrocarbon), elemicin (aromatic), 1-hexadecene (fatty alkene), 1- octadecene (fatty alkene), hexadecanoic acid, methyl ester (saturated fatty acid ester), butyl tetradecanoate (saturated fatty acid ester) and 9,12- octadecadienoic acid (Z,Z)-,methyl ester (unsaturated fatty acid ester).
- The compounds identified in the hexane extract of *O.gratissimum* roots were found to be methyl-2-(1-methylethyl)-(phenol), alpha-copaene (sesquiterpene), trans-caryophyllene (sesquiterpene), alpha-humulene (sesquiterpene), benzene, 1-(1,5-dimethyl-4-hexenyl)-4-methyl- (aromatic), pentadecane (saturated hydrocarbon), beta-bisabolene (sesquiterpene), beta-cadinene (sesquiterpene), 1,6,10-dodecatrien-3-ol, 3,7,11-trimethyl-,(E)- (saturated fatty alcohol), spathulenol (monoterpene), (+)-aromadendrene (terpene), 1,2-methylenedioxy-5,6-dimethoxy-4-allylbenzene (aromatic), 8-heptadecene

(saturated hydrocarbon), **heptadecane** (saturated hydrocarbon) and **octadecanoic acid** (unsaturated fatty acid).

- The compounds identified in the hexane extract of *O.tenuiflorum* roots were found to be 2-pentanone,4-hydroxy-4-methyl- (fatty ketones), benzene,1,4-dimethyl- (aromatic), hexadecanoic acid, methyl ester (saturated fatty acid ester), 2-cyclopentene-1-undecanoic acid, methyl ester (unsaturated fatty acid ester), and 9-octadecenoic acid (Z) (unsaturated fatty acid).
- The chloroform extract of *O.americanum* roots showed the presence of benzaldehyde,4-(1-methylethyl)- (aromatic), thymol (monoterpene), isocaryophyllene (sesquiterpene), dodecanoic acid (saturated fatty acid), oxacyclotetradecane-2,11-dione,13-methyl (fatty hydrocarbon), caryophyllene oxide (sesquiterpene), tetradecane (saturated hydrocarbon), tetradecanoic acid (saturated fatty acid), 9,12-octadecadienoic acid (Z,Z) (unsaturated fatty acid), 9,17-octadecadienal, (Z) (unsaturated fatty aldehyde) and 1,e-11, Z-13-octadecatriene (unsaturated hydrocarbon).
- The chloroform extract of *O.basilicum* roots showed the present of tetradecanoic acid (saturated fatty acid), hexadecanoic acid (saturated fatty acid), 6-dodecenol (saturated fatty alcohol) and 9,12-octadecadienoic acid (Z,Z) (unsaturated fatty acid). The major compound was Peak 5 (9,12-octadecadienoic acid (Z,Z)) with M⁺ (m/z) 280.
- The chloroform extract of *O.filamentosum* roots showed the presence of benzaldehyde,4-(1-methylethyl)- (aromatic), eugenol (phenylpropanoid), trans-caryophyllene (sesquiterpene), pentadecane (saturated hydrocarbon), 2propenoic acid,3-(4-methoxyphenyl)-ethyl ester (aliphatic fatty acid ester),

hexadecanoic acid, methyl ester (saturated fatty acid ester) and 9,12octadecadienoic acid, methyl ester(E, E) - (unsaturated fatty acid ester).

- The chloroform extract of *O.gratissimum* roots showed the presence of alphacopaene (sesquiterpene), trans-caryophyllene (sesquiterpene), alphahumulene (sesquiterpene), pentadecane (saturated hydrocarbon) and betabisabolene (sesquiterpene).
- The chloroform extract of *O.tenuiflorum* roots showed the presence of tetradecanoic acid (saturated fatty acid), hexadecanoic acid (saturated fatty acid), 2-cyclopentene-1-undecanoic acid, methyl ester (unsaturated fatty acid ester) and 9,12-octadecadienoic acid (Z,Z) (unsaturated fatty acid ester).

HPLC Profiles

- All 5 species of **Ocimum** showed 1 peak in common viz. peak 4.
- The 5 species contained species specific compounds viz. peak 1 in *O.americanum*, peak 5 in *O.basilicum* and peak 3 in *O.gratissimum*.
- Peak 11 was common for both *O.filamentosum* and *O.tenuiflorum* while peak 13 was common for both *O.filamentosum* and *O.gratissimum*.
- **O.basilicum** showed the highest concentration of compounds while **O.filamentosum** showed the lowest.
- The UV spectra of peaks 1, 12,13, 14 and 15 resembled phenolice or benzenoids
- The UV spectra of peaks 2, 4 and 6 resembled that of phenylpropanoids or coumarins.
- The UV spectra of peaks 7 and 8 resembled that of chalcones.
- The UV spectra of peaks 9 and 10 resembled that of diterpenoids, which have so far not been reported in **Ocimum**.
- The UV spectra of peak 3 showed a maximum in visible light.
- The UV spectra of peak 11 resembled that of a phenylpropanoid.

HPTLC fingerprints

- **O.americanum** showed 12 peaks, **O.basilicum** 6 peaks, **O.filamentosum** 6 peaks, **O.gratissimum** 7 peaks and **O.tenuiflorum** 5 peaks.
- The chemical marker compound (a mixture of ursolic and oleanolic acids) was found to be present in all 5 species of **Ocimum** in varying proportions.

Column Chromatographic Studies

• The chemical constituents isolated from the roots of **O.americanum** included α – amyrin, β – sitosterol, ursolic acid, oleanolic acid and β - sitosterol- 3β - D – glucoside.

Section B: Comparative Anatomical Studies

- The root periderm was pericyclic in origin in *O.americanum* and *O.gratissimum*.
- The root periderm was cortical in origin in **O.basilicum**, **O.filamentosum** and **O.tenuiflorum**.
- In *O.gratissimum* the midrib had 2 adaxial vascular traces and the petiole had 2 wing strands for every wing.
- In *O.americanum* the midrib had no adaxial vascular traces and the petiole had only 1 wing strand for every wing.
- In *O.tenuiflorum* the midrib was flat on the adaxial side.
- In **O.basilicum** and **O.filamentosum** the midrib was convex on the adaxial side.
- In **O.basilicum** the stem was biconvex in cross-sectional view.
- In **O.filamentosum** the stem was bluntly four-angled in cross-sectional view.

POLLEN MORPHOLOGY

• In O.filamentosum the pollen was subprolate.

- In *O.americanum*, *O.basilicum*, *O.gratissimum* and *O.tenuiflorum* the pollen were suboblate.
- The ectoaperture was 1 µ wide in **O.basilicum**.
- The ectoaperture was > 1 μ wide in O.americanum, O.gratissimum and O.tenuiflorum.
- The Apocolpal distance was 10-15 µ.
- The Apocolpal distance was $15 25 \mu$ in *O.americanum* and *O.tenuiflorum*
- . The Polar diameter was 30 37.5 µ in *O.americanum*.
- The Polar diameter was 40 50 µ in *O.tenuiflorum*.

Seed Morphology

- In *O.americanum* the seeds were reddish brown in colour, elliptical in shape with surface smooth.
- In *O.basilicum* the seeds were black, oblong triquetrous in shape with pitted surface.
- In *O.filamentosum* the seeds were steel blue or black narrowly ovate in shape with smooth surface.
- In *O.gratissimum* the seeds were bright brown in colour broadly ovate or circular in shape with shallow pitted surface.
- In O.tenuiflorum the seeds were dark brown in colour, oblong with short apical beak and warty surface.