

# Traditional Plant Breeding in *Ocimum*

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## Abstract

*Ocimum* spp. is an important medicinal and aromatic plant. It has many medicinal properties. It is a rich source of carbohydrates, fibers, iron,  $\beta$ -carotene, vitamins, phosphorous, calcium, protein, and in aromatic oils. It is also used for the treatment in stomach pain, cough and cold, diarrhea and indigestion. Asthma, ulcers, nausea, and ringworm can also be cured with *Ocimum*. It lowers the blood sugar level and increases lactation. CSIR-CIMAP is actively involved in genetic enhancement of the *Ocimum* species following with different breeding approach in view of traditional

importance. At CSIR-CIMAP, available genetic stocks are seven *Ocimum* species—*Ocimum sanctum*—Krishna and Shyam tulsi, *O. basilicum*, *O. kilimandscharicum*, *O. americanum*, *O. africanum*, *O. gratissimum*, *O. tenuiflorum*, and 100 genetic stocks of *O. basilicum* and nine varieties, namely CIM Ayu, CIM Angana, CIM Saumya, CIM Kanchan, Vikarsudha, CIM Jyoti, CIM Sharada, CIM Surabhi, and CIM Snigdha. In future, there will be possibility to develop varieties for high oil and herb yield with high specific needs chemical like high eugenol, methyl eugenol, methyl cinnamate, geraniol, germacrene A and D, linalool, elemicin,  $\beta$ -elmene, (Z)-ocimine content with some other herbal products.

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## 7.1 Introduction

*Ocimum* belongs to Lamiaceae family, and it is a genus of about 180–250 species of annual and perennial aromatic herbs and shrubs. Several species are native to the tropical and warm temperate regions of the old world, including India. The dry herb (leaves), *Ocimum* leaf tea, essential oil and its chemical derivatives (eugenol, methyl-eugenol, linalool, methyl chavicol, germacrene A and D, elemicin,  $\beta$ -elmene, (Z)-ocimine) are exported to European countries in extensive quantity every year (Simon et al. 1990). The



**Fig. 7.1** Released varieties/cultivars from CSIR-CIMAP of *Ocimum*

annual export of dry leaves herb, its products, essential oil, and its derivatives/chemical constituents of *Ocimum* are worth 5000 tons. People know the plant as Tulsi as *Surasah* in Sanskrit and *Tulsi* in Hindi. Due to antioxidant and anti-aging properties of Tulsi, Hindus use fresh leaves in the *Panchamrut/Charanamrut* drink after *puja*. Tulsi is religion. It is regarded not merely as a utilitarian Godsend, as most holy plants are viewed to be, but as an incarnation of the Goddess Herself. The classic Hindu myth, *Samudramathana*, the ‘Churning of the Cosmic Ocean,’ explains that *Vishnu* spawned Tulsi from the turbulent seas as a vital aid for all mankind. The Tulsi leaves, when eaten, can control thirst, and so was invaluable to tired travelers. The oil is used as antiperspirant and as fly and mosquito repellent. CSIR-CIMAP is actively involved in genetic enhancement of the *Ocimum* species by using different breeding approaches in view of traditional significance, together with the need for developing a better plant type having high herb, essential yield characters combined with a consistent high yield of phenylpropanoid eugenol and other economically important chemical constituents to formulate value-added products. By the application of very intensive plant breeding techniques, a number of varieties have been developed at the CSIR-Central Institute of Medicinal and Aromatic Plants, India, including: Khushmohak, CIM Saumya (Lal et al. 2003),

CIM Sharda of *Ocimum basilicum*, CIM Jyoti of *O. africanum*, CIM Ayu (Lal et al. 2004), CIMAngana of *Ocimum sanctum* (Lal et al. 2008), CIM Kanchan from *O. tenuiflorum*, CIM Surabhi, and CIM Snigdha (Fig. 7.1). These varieties/ cultivars are also useful for intercropping with aromatic grasses and other cereal crops. Some of the international popular varieties/ cultivars and released varieties/cultivars from CSIR-CIMAP of *Ocimum* are summarized in Tables 7.1 and 7.2.

By conventional breeding method, Lal et al. (2003) developed a high-yielding eugenol-rich oil producing variety of *O. sanctum*, CIM Ayu; Lal et al. in 2004 developed an early, short duration, high essential oil, methyl chavicol and linalool yielding variety of Indian basil (*O. basilicum*), CIM Saumya; Lal et al. (2008) also developed a high-yielding dark purple pigmented variety CIM Angana of Shyam tulsi (*Ocimum sanctum* L.); Singh and Sehgal (1999) chemically characterized and made selection of *Ocimum* genotypes and described their uses in traditional medicine. They studied the genetic variability, medicinal, and economic value of *Ocimum* germplasm. Lal (2014) developed new and stable chemotypes in *Ocimum*. Patel et al. (2015a, b) studied genetic variability pattern and possibility of its exploitation of *Ocimum* germplasm in Fig. 7.2. Patel et al. (2015a, b) also studied the phenotypic characterization and

**Table 7.1** International popular varieties/cultivars of *Ocimum* species

S. no.	Varieties/cultivars	Botanical identification	Origin of the plant material	Chemotypes	Breeding strategies
1.	Blue Spice	<i>Ocimum basilicum</i>	Czech Republic	Bisabolene	Asexual reproduction
2.	Fino Verde	<i>Ocimum basilicum</i>	Maine, USA	Linalool	Asexual reproduction
3.	Holandjanin	<i>Ocimum basilicum</i>	Istria	Linalool	Asexual reproduction
4.	Compact	<i>Ocimum basilicum</i>	Maine, USA	Linalool	Asexual reproduction
5.	Genovese	<i>Ocimum basilicum</i>	Slovenia	Linalool	Selection
6.	Purple Opal	<i>Ocimum basilicum</i>	Czech Republic	Linalool	Selection
7.	Lattuga	<i>Ocimum basilicum</i>	USA	Linalool	Selection
8.	Osmin	<i>Ocimum basilicum</i>	California	Linalool	Selection
9.	Cinnamon	<i>Ocimum basilicum</i>	Wroclaw, Poland	Linalool and methyl cinnamate	Selection
10.	Purple Ruffles	<i>Ocimum basilicum</i>	Germany	Linalool	Cross selection
11.	Lime	<i>Ocimum americanum</i>	Czech Republic	Geranial and neral	Selection
12.	Siam queen	<i>Ocimum basilicum</i>	USA	Methyl chavicol	Cross selection
13.	Dark Lady	<i>Ocimum basilicum</i>	Wolsier, Germany	Linalool	Asexual reproduction
14.	Dbasbloom	<i>Ocimum basilicum</i>	Israel	Linalool	Asexual reproduction
15.	Green Bell	<i>Ocimum basilicum</i>	Wolsier, Germany	Linalool	Asexual reproduction

stability analysis for biomass and essential oil yields of 15 genotypes of five *Ocimum* species. Patel et al. (2016) studied the differential response of genotype  $\times$  environment on phenology, essential oil yield, and quality of natural aroma chemicals of five *Ocimum* species. Srivastava et al. (2018) conducted germplasm characterization and correlation studies in *O. basilicum* for yield and related traits. Singh et al. (2004, 2014) chemically characterized and made selection in *Ocimum* genotypes.

## 7.2 Breeding for Disease Resistance

Breeding work for disease resistance in *Ocimum* is very meager as described below.

Mishra et al. (2016) reported the identification and functional characterization of an *O. basilicum* PR5 family member that is responsive to multiple stresses and hormonal elicitation. The results highlighted the role of a thaumatin-like protein, ObTLP1, in mediating tolerance to the

**Table 7.2** Released varieties/cultivars from CSIR-CIMAP of *Ocimum* species with their quality

S. no	Varieties/cultivars	Botanical identification	Common name	Herb yield (q/ha)	Oil content (%)	Oil yield (kg/ha)	Main essential oil constituents				Breeding strategy	
							Eugenol	Methyl eugenol	Methyl chavicol	Linalool		Methyl cinnamate
1.	Vikarsudha	<i>Ocimum basilicum</i>	French basil	335	0.5	167.5	0.62	–	78	0.16	–	Developed through intraspecific hybridization between exotic basil from Australia (EC-331886-CSIRO No. L6323) and local adaptive landrace, Badaun local
2.	Khushmohak	<i>Ocimum basilicum</i>	Sweet basil	391	0.4	134	–	–	37	45	–	Developed through selection in seed raised progeny of the introduced strain from Argentina
3.	CIM Ayu	<i>Ocimum sanctum</i>	Holy basil (Krishna)	200	0.72	110.95	83.56	–	–	0.05	–	Developed through mass selection
4.	CIM Angana	<i>Ocimum sanctum</i>	Holy basil (Shyama)	181	0.64	91.7	40.42	–	–	1.92	–	Developed through half-sib selection
5.	CIM Kanchan	<i>Ocimum teniflorum</i>	Holy basil	197	0.49	110.7	4.36	78.4	–	0.09	–	Developed through selection
6.	CIM Saumya	<i>Ocimum basilicum</i>	Indian basil	290	0.68	197.2	–	–	62.54	24.61	–	Developed through half-sib selection
7.	CIM Jyoti	<i>Ocimum africanum</i>	Lemon-scented basil	200	0.75	150	–	–	–	–	75	Developed through selection
8.	CIM Sharada	<i>Ocimum basilicum</i>	French basil	280	0.7	190	–	–	89.75	0.067	–	Developed through intensive breeding
9.	CIM Shurabhi	<i>Ocimum basilicum</i>	Sweet basil	200	0.75	166	–	–	0.44	75.71	–	Developed through half-sib selection
10.	CIM Snigdha	<i>Ocimum basilicum</i>	French basil	221	0.9	190	–	–	9.1	1.95	78.7	Developed through half-sib selection

**Fig. 7.2** Variability among leaves of the genetic stocks of *Ocimum*



fungal pathogens and to the abiotic stresses by hindering fungal colonization in host and by maintaining osmotic adjustment in cells, respectively, and ObTLP1 might be a useful candidate for providing tolerance in crops toward multiple stresses.

Zaim et al. (2016) studied the occurrence of yellow mosaic and leaf curl disease in *Ocimum* spp. and conducted screening for disease management. They investigated that yellow mosaic and leaf curl diseases affected *O. killimandscharium*,

*O. gratissimum* and very severely to *O. basilicum* var. CIM Saumya in non-homozygous population. Investigation has also been carried out to evaluate the available germplasm of *Ocimum* spp. for identifying resistance source. Since *O. basilicum* was found to be highly susceptible to these viral diseases, a few lines have been selected, which were found to be tolerant against the leaf curl disease. These studies will help in strategizing the effective management practices and developing resistant cultivars in *Ocimum*.

### 7.3 Polyploidy in Basil

Sobti and Pushpangadan (1982) presented in detail the cytogenetical and evolutionary relationship in *Ocimum americanum*, *O. canum*, and *O. basilicum*. *O. americanum* was found to be morphologically intermediate between the other two species in most characters like height, length of spike, calyx, corolla, stamen, style fruiting calyx. Crossability studies and induction of allopolyploidy were also attempted by above workers. Karyomorphological investigations were done by Sobti and Pushpangadan (1982), in five *Ocimum* species with their different races. Closer examination of the karyotypes revealed variation in the type and number of chromosomes and considered the presumption as mentioned in Chap. 6. Meiotic studies in five species of *Ocimum* from pollen mother cells by Khosla (1989) suggested that the base number  $x = 8$  represented Sanctum group while base number  $x = 12$  represented Basilicum group. Omidbaigi et al. (2010) studied the induction and identification of polyploidy in *O. basilicum* using colchicines in different concentrations (0.00, 0.05, 0.10, 0.20, 0.50, and 0.75%) and four treatments (seed, the growing point of seedlings at the emergence of cotyledon leaves stage and emergence of true two type leaves stage, and root treatment). The 0.5% dosage proved to be the most effective in producing autotetraploids having larger stomata and pollen grains, increased chloroplast number in guard cells, and decreased stomata density, compared to diploid control plants.

### 7.4 Linalool Rich, Evergreen, and Cold-Tolerant Interspecific Cross Hybrid

A cold-tolerant evergreen triploid and its amphidiploid also developed using interspecific cross between *O. basilicum* and *O. kilimandscharicum* are under field evaluation trial for their essential oil yield with good quality (Fig. 7.3). It is an evergreen aromatic perennial undershrub. Improving the cold tolerance and developing resistant varieties of *Ocimum* are very

important for the growing areas of the species (Dhawan et al. 2015). In relation to adaptive behavior, *O. basilicum* and other species are sensitive to cold stress except *O. kilimandscharicum*; therefore, an evergreen interspecific hybrid was developed by hybridization through breeding efforts from *O. basilicum* (CIM Surabhi) with *O. kilimandscharicum* for stress tolerance (Fig. 7.4). The developed evergreen interspecific hybrid may thus provide a base to various industries which are dependent upon the bioactive constituents of *Ocimum* species, and one another cold-tolerant evergreen interspecific cross between *O. basilicum* cultivar CIM Surabhi and *O. kilimandscharicum* (–) linalool 75–80% is also under pipeline and under field evaluation trial for their essential oil yield with good quality (–) linalool.

### 7.5 A Novel Source of Chavibetol Constituent in *Ocimum basilicum*

CSIR-CIMAP, Lucknow, has also developed a new line with chavibetol (8–10%) proportions and flavor of *Ocimum* leaves as paan/betel (*Piper betle*) (Fig. 7.6). Eugenol was recorded in this accession in trace amount. Structure of chavibetol was confirmed using NMR experiments (Fig. 7.5).

### 7.6 Basil as an Edible Ornamental Herb

#### 7.6.1 Commercial Cultivars

Several basil varieties, differing in the size, shape, aroma, and color of the leaves, exist. Commercial basil cultivars also display a wide diversity in growth habit, flower, leaf, and stem colors, and aroma. Many of the cultivars evaluated belong to the ‘sweet’ basil group with ‘Genovese,’ ‘Italian large leaf,’ ‘Mammoth,’ ‘Napoletano,’ and ‘Sweet’ dominating the American fresh and dry culinary herb markets. Several other basil like ‘Sweet Fine’ appear similar to ‘sweet’ basil



*O. basilicum* and *O. kilimandscharicum*

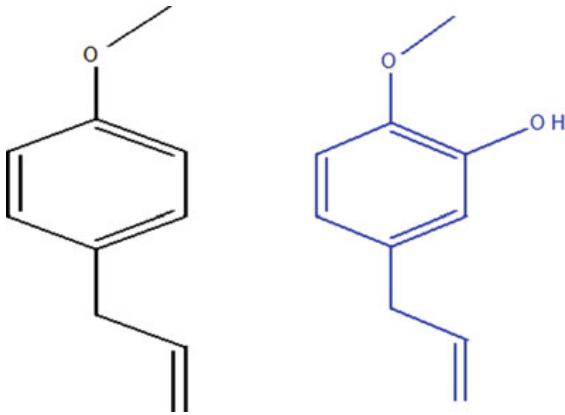
**Triploid**

**Amphidiploids**

**Fig. 7.3** A cold-tolerant evergreen triploid and its amphidiploid also developed using interspecific cross between *O. basilicum* and *O. kilimandscharicum* are under field evaluation trial for their essential oil yield with good quality

**Fig. 7.4** A (-) linalool-rich cold-tolerant evergreen interspecific cross between *O. basilicum* cultivar CIM Surabhi and *O. kilimandscharicum* (-) linalool 75–80%





**Fig. 7.5** Structure of chavibetol



**Fig. 7.6** OCL-32 chavibetol in *Ocimum*

though the leaves tend to be smaller (Simon et al. 1999). The lemon-scented cultivars ('Lemon' and 'Lemon Mrs. Burns') differ from each other in days to flower, and total oil content, but not in citral content. The 'Maenglak Thai Lemon' basil, which varies in appearance from the other lemon basil, is an attractive ornament. Among the purple basil, 'Osmin Purple' and 'Red Rubin Purple Leaf' are the most attractive and best retain their purple leaf color (Simon et al. 1999). Anthocyanins in purple basil are genetically unstable, leading to an undesirable random green sectoring and reversion over the growing season (Phippen and Simon 1998). Several basil with dwarf growth habit were developed as ornamental border plants including 'Bush,' 'Green Globe,'

'Dwarf Bush,' 'Spicy Globe,' and 'Purple Bush.' A group of ornamental basil was selected and named for their characteristic aroma including 'Anise' (methyl chavicol), 'Cinnamon' (methyl cinnamate), 'Licorice' (methyl chavicol), and 'Spice' (bisabolene) (Darrah 1972; Albuguerque 1996; Simon et al. 1999).

## 7.6.2 Sensoric Quality

Fresh basil leaves have a strong and characteristic aroma, not comparable to any other spice, although a traceable hint of cloves exists. In addition to the 'Mediterranean type' most common in the West, a plethora of other varieties or cultivars with different flavors, many of which are hybrids, is available. India has 'sacred basil' (*O. sanctum* = *O. tenuifolium*) with an intensive, somewhat pungent smell, and Thailand has a sweet basil with a licorice aroma (Singh and Sehgal 1999). Varieties sold to gardeners in the West include cinnamon basil, camphor basil, anise basil, and spice basil; the latter has a very pleasant, complex, and warm flavor (Darrah 1974; Albuguerque 1996; Morales and Simon 1996; Phippen and Simon 1998; Martins et al. 1999; Simon et al. 1999). A last group of cultivars, characterized by citrus aroma, are 'Thai Lemon basil' (*O. citriodorum*) which has a distinct balm-like flavor and lime basil and another lemon basil (*O. americanum*) which has an extraordinarily pure and fresh lemon aroma (Morales and Simon 1996). Perennial basil species from Africa (*O. kilimandscharicum*) and Asia (*O. canum*) have recently been introduced to the European herb and gardening market. These species have a strong, but less pleasant flavor, and hybrids between them and Mediterranean basil are a recent innovation with a novel appearance and flavor that are enjoying a growing popularity (Darrah 1972; Kanebo 1992; Grieve 1999). All basil varieties have in common that dried leaves are much less aromatic than fresh ones; deep freezing the herb is the best method of preservation.



### 7.6.3 Ecology

Sweet basil is cultivated in agro climates between 7 and 27 °C, with 0.6–4.2 m annual precipitation and soil pH 4.3–8.2. While susceptible to frost and cold temperature injury, the species develop best under long days, in full sun and on well-drained soils.

## 7.7 Future Prospects

The conventional function of plant breeding in the area of feed, food, fiber, and ornate will persist to be significant. In spite of this, new functions are progressively budding for plants. The skill of using plants as bioreactors for generating pharmaceuticals will expand; this knowledge is therefore over a decade. Approaches for using plants for generating medicinal antibodies, engineering antibody-mediated pathogen resistance, and altering plant phenotypes by immunomodulation are being refined. New skills are being expounded for plant breeders, particularly, in the areas of the biotechnological applications to plant breeding. New marker knowledge is going on to be refined, and older ones are progressive. Tools assisting the plant breeders to be more efficiently controlling the quantitative traits will be improved.

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