# Wild marigold (Tagetes minuta) variety 

## Him Swarnima (CSIR-IHBT-TM-09)

Introduction:
Wild marigold (Tagetes minuta L.) is a plant native to South America (subtropical and tropical America). It belongs to Asteraceae family and has spread throughout the world. It is an erect, annual, hermaphrodite herb possessing green stem up to 1-2 m in height, compound leaves with narrow serrated leaflets. Wild marigold is an annual crop suitable for cultivation in the plain and hilly areas, as a monocrop or intercrop. In India, wild marigold is found naturally in the western Himalayas between altitude range of $1000 \mathrm{~m}-2500 \mathrm{~m}$. Himachal Pradesh and the hills of Uttarakhand are the regions where wild marigold occurs in its natural habitat. The wild growth of this plant in these regions of India forms the source of essential oil commonly known as "Tagetes oil".

## Uses

Volatile oil of wild marigold is used in perfumery and as a flavor component in food products, and has suppressive biological activity against different pathogens and insects. The essential oil has been reported to have antimicrobial, insecticidal, repellent, antioxidant, antifungal, nematicidal and allelopathic properties.


## 'Him Swarnima' (CSIR-IHBT-TM-09)

The variety 'Him Swarnima' (CSIR-IHBT-TM-09) of Tagetes minuta has been developed by CSIRInstitute of Himalayan Bioresource Technology, Palampur through half-sib progeny selection approach. The variety has high biomass yield (18.0-23.0 tonnes/ha) and essential oil content of 0.30 to $0.34 \%$ and was selected from advanced breeding lines developed from germplasm core collections through halfsib family selection. The variety was evaluated in multi-location trials and found to be vigorous in growth with good adaptability.

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## Breeding methodology

Tagetes minuta is an aromatic herb, commercially cultivated for its essential oil present in aerial parts of the plant. The floral biology of wild marigold suggests often cross pollinated breeding behaviour. Accordingly for varietal development, selective breeding of wild marigold was done using half-sib progeny selection approach. Out of 334 diverse accessions, 18 accessions with high biomass were identified. Preliminary yield trials of 18 accessions were carried out by raising progeny rows of each accession with three replications. Nine potential selections of Tagetes minuta identified on the basis of biomass yield were evaluated in multi-location trials along with check variety (Himgold) in a Randomized Block Design (RBD) with three replications at four different locations in Himachal Pradesh. CSIR-IHBT-TM-09 has a biomass yield of 18.0-23.0 tonnes/ha and essential oil content ranges from 0.30 to $0.34 \%$ at crop maturity.

## Agrotechnology

The crop is most suitable for the production of essential oil in the mid-hills of Western Himalayas. The crop duration is from June to November and cultivation is done by either direct sowing or transplanting of seedlings. The crop is harvested at seed maturity stage during October-November. Removal of the apical shoots during August is essential to check the erect growth and promote lateral branches. Nipping at early stages decreases the ratio of stem to leaf and flower parts and enhances the biomass yield and oil yield per unit area. This practice also reduces the risk of crop logging. Proper drainage of access rain water is necessary to avoid water logging in the crop field. Wild marigold grows slowly during initial growth period and may be suppressed by weeds, therefore, weeding and hoeing are necessary during first 60 days to reduce competition with weeds.

## Propagation

Tagetes minuta is an annual crop and is propagated by seeds. Crop can be raised by direct seed sowing or through transplanting. Seeds of the Tagetes minuta are light and small in size weighing 1000 seeds per gram. About one kg seed is sufficient for one ha when crop is raised through transplanting. Direct sowing requires 3 kg seeds $/ \mathrm{ha}$. Seeds should be sown at soil surface and evenly distributed and covered with thin layer of soil mixture. Seed germination starts after about ten days of sowing. In case of transplantation, spacing of $30 \times 45 \mathrm{~cm}$ is maintained for proper growth of the plants.


Field view of Tagetes minuta during vegetative phase

## Harvesting, distillation and storage

Essential oil is present in leaf and flower parts of the crop, while stem is devoid of any oil. Therefore, as a common practice, crop is harvested about two feet above the ground level, leaving the stem portion without leaves. For higher oil recovery, the proportion of inflorescence and leaves should be more than the stem. Crop should be harvested at seed maturity stage for good essential oil content and quality. The essential oil extraction from the crop is done through steam distillation. The produce should be distilled within 2-3 days of harvesting. Stored biomass or essential oil should not be exposed to sunlight, moisture and high temperature at any stage of distillation as these factors deteriorate the oil quality. Wild marigold oil is pale yellow to dark yellow coloured and moisture should be removed immediately after distillation. The oil should be stored in stainless steel, amber colour glass container or aluminium containers filled up to the brim to avoid auto-oxidation and stored in cool and dark place.


Field view of Tagetes minuta at pre-flowering stage Statement of distinction
Variety 'Him Swarnima' is about two meters in height with compact plant stature and multiple branches which are erect and upright. It has large leaves, dark green in colour with multiple clusters of flower heads.


Field performance of TM-09 in comparison to check over different locations


Close-up view of Tagetes minuta inflorescence


Field view of Tagetes minuta during reproductive phase

## DNA Fingerprinting of improved cultivar 'Him Swarnima' using SSR markers

Genetic distinctness of selection 'Him Swarnima' (CSIR-IHBT-TM-09) was established using 10 SSR markers recently reported by our group. Ten selections namely, TM-01 to TM-09 and TM-10 as check variety (Himgold) of Tagetes minuta (wild marigold) were characterized using molecular markers. In total, 30 alleles were detected ranging from 2 to 4 with an average of 3.0 alleles per SSR locus. Ten SSR markers evincing reproducible polymorphic loci among the CSIR-IHBT-TM-09 (Him Swarnima) and other selections were used for development of fingerprints. Based on the SSR data, consolidated DNA fingerprints were developed with rare or unique marker loci. Cluster analysis of ten selections based on 30 polymorphic loci grouped in three major groups. Improved selection TM-09 captured significant diversity and clustered with selection TM-08 and Himgold. Pair-wise genetic similarity (GS) of TM-09 varied from a minimum of $36 \%$ (TM-05) to maximum of $70 \%$ (TM-08). In conclusion, genetic similarity data based on 30 polymorphic loci suggested that improved selection TM-09 has captured high level of genetic diversity and can be potentially used as promising parental group for future genetic improvement programme of Tagetes minuta.


Representative SSR profile of Tagetes minuta selections using SSR primers

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Table represents Jaccard's dissimilarity coefficients among the Tagetes minuta selections

|  | $\mathbf{1}$ | $\mathbf{2}$ | $\mathbf{3}$ | $\mathbf{4}$ | $\mathbf{5}$ | $\mathbf{6}$ | $\mathbf{7}$ | $\mathbf{8}$ | $\mathbf{9}$ | $\mathbf{1 0}$ |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| $\mathbf{1}$ | $\mathbf{0}$ |  |  |  |  |  |  |  |  |  |
| $\mathbf{2}$ | 0.57 | $\mathbf{0}$ |  |  |  |  |  |  |  |  |
| $\mathbf{3}$ | 0.56 | 0.54 | 0 |  |  |  |  |  |  |  |
| $\mathbf{4}$ | 0.44 | 0.54 | 0.24 | 0 |  |  |  |  |  |  |
| $\mathbf{5}$ | 0.52 | 0.50 | 0.43 | 0.50 | 0 |  |  |  |  |  |
| $\mathbf{6}$ | 0.56 | 0.48 | 0.29 | 0.19 | 0.50 | 0 |  |  |  |  |
| $\mathbf{7}$ | 0.48 | 0.46 | 0.26 | 0.23 | 0.42 | 0.35 | 0 |  |  |  |
| $\mathbf{8}$ | 0.45 | 0.50 | 0.50 | 0.44 | 0.58 | 0.50 | 0.35 | 0 |  |  |
| $\mathbf{9}$ | 0.45 | 0.43 | 0.56 | 0.38 | 0.64 | 0.44 | 0.42 | 0.30 | 0 |  |
| $\mathbf{1 0}$ | 0.57 | 0.55 | 0.67 | 0.59 | 0.75 | 0.65 | 0.52 | 0.24 | 0.33 | 0 |

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