ORIGINAL RESEARCH ARTICLE

Genotype × environment interaction, genetic variability and inheritance pattern in breeding lines including varieties/cultivars of menthol mint (*Mentha arvensis* L.)

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Article History

Received: February 04th, 2020
Revised: March 18th, 2020
Accepted: April 15th, 2020

Key Words

G×E
Menthol-mint
Stability
Variability
Variety

ABSTRACT

Menthol mint which belongs to the genus Mentha of the *family-'Lamiaceae', is a valuable essential oil-yielding crop. Its essential oil, aroma compounds, menthol crystals are in trades worldwide. It is an aromatic herb, grown in sub-tropical parts of north India. The herbs upon hydro-distillation yield essential oil, containing high (75–80%) menthol content. The oil has a bitter cooling taste, odour and is the principal source of menthol. Natural menthol is preferred in the food and flavour industry. The literature about genotype × environment* (*G×E) is very meager on this crop. Therefore, the present investigation was carried out to determine the genetic stability and adaptability pattern among eight cultivars/varieties/breeding lines of menthol mint, namely, MAS-1, Kalka, Shivalik, Himalaya, Kosi, OPSP-33, OPSP-45, and OPSP-80, in eight environments. The morpho-metric data were recorded in terms of quantified genetic variability for nine different economic traits namely plant height, leaf numbers, leaf/stem ratio, leaf length, leaf width, herb yield, oil content were oil yield, and menthol content (%) in the essential oil. Further, stable genotypes for high oil yield and menthol content were identified over the years using G×E based on Eberhart and Russel's 1966 model. The varieties/lines, namely OPSP-80, followed by OPSP-33, OPSP-45, Kosi, Kalka, Himalaya, showed the widest stability due to their ability to tolerate a wide range of environmental conditions.*

INTRODUCTION

Menthol mint (*Mentha arvensis* of the family-'Lamiaceae') is a valuable essential oil-yielding crop. The essential oil, aroma compounds, and the menthol crystals are in the trade worldwide. It is an aromatic herb, grown in subtropical parts of north India. The herb on distillation yields the essential oil containing about 75–80% menthol, and is the principal source of natural menthol. Mint oil is used as an ingredient in cough drops, related

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Doi: https://doi.org/10.62029/jmaps.v42i2.Singh

pharmaceuticals, dentifrices, cosmetics, mouthwashes, scenting of tobacco products, and flavouring of beverages. Besides, natural menthol is preferred in the food and flavour industry (Patra and Kumar, 2004; Patra et al., 2000). Many species of mints are being cultivated all over the world, including India. Among them, only four species are predominantly grown in India. These include menthol mint (*Mentha arvensis* L.var. piperascens), peppermint (*M. piperita*), bergamot mint (*M. citrata*) and spearmint (*M. spicata*). India is the leading supplier of menthol mint oil in the world, and a large number of farmers in India are being benefitted through its cultivation (Lal et al., 2017a, 2017b, 2019).

The cultivation of menthol mint in this country dates back to about 48 years. Before the 1960s, the requirement of menthol in India was met by import. It was introduced as a crop in India through the efforts of CSIR's Central Indian Medicinal Plants Organization (now CSIR-CIMAP) and Regional Research Laboratory, Jammu (now CSIR-IIIM). The project on mint cultivation was taken up at the CSIR-CIMAP Research Centre, Pantnagar, which was established in 1962 near Haldwani in Uttarakhand state. As a result of the continuous efforts through this Research Center, large areas in the Tarai region of Uttarakhand and U.P, i.e., Kashipur, Moradabad, Rampur, etc. were brought under the organized cultivation and processing of the Japanese mint crop for its oil and menthol. The cultivation of mint has become progressively popular since then. It has spread gradually to vast areas of Uttar Pradesh, and small to large areas of Punjab, Haryana, Madhya Pradesh, and Bihar, etc.

About five years back, multinational companies (e.g., Symrise, BASF) also started production of menthol through synthetic routes posing a severe threat to the natural menthol. Besides this, climate changes that may affect the cropping pattern and yield potential of the existing cultivars are also a threat to mint farming. The diversified usage of menthol has shown that this commodity will be required consistently in large volumes to meet the domestic as well as global requirements. It is, therefore, imperative to steer the research for breeding improved and high yielding varieties which can be grown in adverse climatic conditions and improvised agro-technology for the cultivation of this industrial crop with minimum inputs. Recently, two new varieties, namely CIM-Kranti and CIM-Unnati of menthol mint, were developed and released by CSIR-CIMAP for sustainability of menthol production to compete with synthetic menthol. The literature about the genotype × environment (G×E) is very meager on this crop. Therefore, the present investigation was carried out to determine the genetic variability, stability, and adaptability pattern among eight cultivars/lines of *M. arvensis* for making proper recommendation of suitable lines/varieties for their large areas cultivation in India.

MATERIALS AND METHODS

This investigation was conducted in the main mint-cropping season (February to June) for four years at the CIMAP Research Centre Pantnagar, Uttarakhand, which is situated in foot-hills of Shivalik ranges of the Himalayas and falls in the humid subtropical climatic zone. It is located at the latitude of 29.5°N, the longitude of 79.3°E, and an altitude of 243.84m above the mean sea level. The weather conditions were entirely satisfactory for the normal growth of the mint crop. The soil texture of experimental plants was sandy clay loam.

Experimental layout

The study was initiated in the first week of December with the raising of nurseries for growing the open-pollinated seeds collected in bulk from the commercial plantation measuring one-hectare area for the popular variety Shivalik. The well grown 144 seedlings from the nurseries were transplanted in April in field progeny beds, each measuring 6 × 4 m in size and accommodating a total of 24 seedlings at 1.00 × 1.00 m spacing. These open-pollinated seed progenies (OPSPs) were maintained with frequent irrigations and other normal cultural practices. A total of 115 OPSPs which survived with proper plant growth served as the original gene pool for the investigation. On June 15, the 115 OPSPs (individual plants) were assessed for their morpho-physiological fitnesses' for three major plant characters: green herb weight, oil content (%), and estimated oil yield. Such assessment led to the identification of 30 elite OPSPs that could be carried forward to advance studies. These selected 30 OPSPs were frequently vegetatively multiplied through suckers in separate nurseries to raise their pure clonal populations for the preliminary and pilot-scale yield trials conducted in the field during two consequtive seasons.

For raising the 30 clonal populations for the trials, apical shoot portions of about 12-15cm length from each original OPSP plants were planted in sucker producing nurseries during rainy season every year and planting in the field. The trials were done during the first week of February in each year by the use of pure underground suckers collected from the sucker nurseries. As previously mentioned above, a total of 115 open-pollinated seed progenies OPSPs were developed in the popular variety Shivalik, which served as the original gene pool for the present study. These OPSPs could be assessed of their variability for three major economic traits: herbage weight, oil content (%), and estimated oil yield. Based on the performance, the 30 elite OPSPs were selected.

The essential oil ranged in the thirty selected OPSPs between 2.28-4.44 g v/s the single plant average of 1.20 g of the parent variety Shivalik (Table 1). Out of the 30 elite OPSPs, three promising OPSPS, namely, OPSPS -33, OPSPS -45, and OPSPS -50 selected and again evaluated in the eight years with varieties, namely MAS-1, Kalka, Shivalik, Himalaya, and Kosi. The mean squares due to treatments were highly significant for all the characters indicating thereby that the developed OPSPs had ample genetic variations usable in the selection program aimed at identifying among them top-ranking genotype(s). Further, the eight varieties/OPSPs/genotypes of menthol mint *(M. Arvensis* L.), Namely, MAS-1, Kalka, Shivalik, Himalaya, Kosi, OPSP-33, OPSP-45, and OPSP-80 were evaluated in the Initial Evaluation Trial (RBD, three replications plot size = 1.5m^2) over the eight years at the Research Farm of CSIR-Central institute of medicinal and aromatic plants, Research Centre Pantnagar, Uttarakhand, India.

Essential oil content and oil quality analysis

The essential oil content determination was done through the Clevenger apparatus (Clevenger, 1928) by distilling of 200gms of the fresh herb of the selected plants. Observation on the major quality constituents of the essential oil of the selected plants was recorded through gas-liquid chromatography on HP-5890 model using a DB-WAX capillary column (30m x 0.53mm x 0.2 mm film) with temperature programme from 60°C to 220°C @ 3°C/m, initial hold 4 minutes and hydrogen as carrier gas. Injector and FID temperatures were 220°C and 240°C, respectively. The data were processed on AIMIL chromatography data system.

Statistical analysis

Data were analyzed for stability parameters using Eberhart and Russel's 1966 (Eberhart and Russell, 1966) model for the stability by CSIR-CIMAP software version 4.0 available at the Department of Genetics and Plant Breeding and Genetics of the Institute (Lal et al., 2018b, 2020a,2020b; Singh and Chaudhury, 2014).

Analysis of variance

The data collected on various characters were analyzed separately using the conventional RBD analysis. The linear model was used to represent the mean performance of a genotype in a given plot (Singh and Chaudhury, 2014) as follows:

$Y_{ij} = m + t_i + r_i + e_{ij}$ (i = 1, 2, ...g ; j = 1, 2, ...r)

Where,

Y_{ij} = performance of ith treatment in the jth replication; m = general mean; t_i = effect of ith genotype; r_j = effect of jth replication; e_{ij} = random error associated with ith genotype in jth replication with 0 mean and variance 1.

The partitioning of total variance due to replications, genotypes, and error along with the degree of freedom, the sum of squares, and the mean sum of squares is given below:

Sources of variation	Degree of freedom	Mean squares	Expected mean squares	F-Test
Replication	(r-1)	μr	$\delta^2_e + g\delta^2_r$	Mg/Me
Genotype	(g-1)	μg	$\delta^2_e \quad r\delta^2_g$	
Error	(r-1) (g-1)	μe	δ^2_e	
Total	(rg-1)			

Where;

r = Number of replications; g = Number of genotypes; μr = Mean squares due to replications; μg = Mean squares due to genotypes; μe = Mean squares due to error; δ^2_{re} = Error variance; δ^2_{gr} = Replication variance; δ^2_g = Genotypic variance.

To compare the differences between means, the critical difference (CD) was calculated as follows:

CD = SE $_{diff}$ × t value at error degree of freedom

Where;

SE_{diff} (Standard error of the difference between two treatment) = $\sqrt{2 \text{ x error mean square}/r}$

r = number of replications

The significance of the difference among treatment means was tested by the 'F' test.

Comparison of means

To compare the difference between means, the critical difference (CD) was calculated as follows:

CD = SE $_{diff}$ ´ t value at error degree of freedom

Where;

SE_{diff} (Standard error of the difference between two treatment) = $\sqrt{2 \text{ x error mean square}/r}$

r = number of replications

If, $x_1 - x_2 \geq$ CD

or

$\leq$ CD

Where,

x = mean of variety/OSPS/genotypes

Then the difference between means was significant.

Regression and G × E interaction analysis

Regression analysis following Eberhart and Russell (1966) model (Eberhart and Russell, 1966) was carried out, which suggested three parameters.

1. Mean
2. Regression of individual mean performance on environment index, and
3. Deviation from regression

These parameters are defined by the following model:

$Y_{ij} = \mu_i + B_i I_j + d_{ij}$

Where,

Y_{ij} = mean performance of i^{th} genotype in j^{th} environment (i = 1, 2,....., v ; j = 1, 2,....., m); v = number of genotypes; m = number of environments; μ_i = mean performance of i^{th} genotype over all environments; B_i = regression coefficient of i^{th} individual mean performance environmental index I_j; I_j = j^{th} environmental index; δ_{Ij} = division from regression of the i^{th} genotype at j^{th} environment.

The environmental index, I, is estimated as the mean of j^{th} environment minus the grand mean, which may be expressed as follows:

$I = (\sum^v_{1=1} Y_{ij} / v) - (\sum^1_{1=1} \sum^n_{j=1} Y^{ij}) / mv$, with, $\sum^n_{j-1} I_j = 0$

The first parameter, the linear regression coefficient (bi) was estimated using the following formula.

$b_i = (\sum^n_{j=1} Y_{ij} \; I_j) / (\sum^n_{j=1} I^2_j)$

The second parameter, the deviation mean square (S^2di), was estimated using the following formula.

$S^2di = (\sum^n_{j=1} \delta^2_{ij}) / (n-2)$ – pooled error

Where,

$$\sum^n_j = 1\delta^2_{ij} = [\sum^n_{j=1} Y^2_{ij} - (Y_1)^2/n] - \frac{[\sum^n_{j=1} Yij \; Ij]^2}{\sum^n_{j=1} I^2}$$

The estimated pooled error was obtained by averaging error sum of squares over all the three environments the formula as follows

$S^2_e = \sum_{ni} Si^2 / \sum_{ni}$

Where,

ni = error degree of freedom,

S^2 di = error mean sum of squares.

The following test of significance was done.

1. The test of difference among the mean performance of genotype was done using the 'F' test.

$$F = M_{S1} / M_{S4}$$

Where,

SE (b) = MS due to pooled deviation

$\sum^n_{j=1} \; I^2 j$

5. Deviation from liner regression for each genotype was test using 'F' test:

$F = [\sum^n_{j=1} \; \delta^2_{ij} / (n\text{-}2)] / M_{es}$ (Pooled error).

Structure of combined regression analysis/ pooled analysis of variance (ANOVA) for estimation of stability parameters (Eberhart and Russell, 1966) is as below:

Source	d.f.	S.S.	M.S.
Total	nv-1	$\Sigma^v_{i=1} \Sigma^n_{i=j} Y^2_{ij}$ – CF = T.S.S	
Genotypes	V-1	$1/n\Sigma^v_{i=1} Y^2_i$ – CF = G.S.S	MS_1
Env. (E)	n-1	$1/v\Sigma^n_{j=1} Y^2_j$ – CF = E.S.S	
GxE (n-1)	(v-1)	T.S.S. – (G.S.S. + E.S.S.)	MS_2
E + GxE	V(n-1)	$\Sigma^v_{i=1} \Sigma^n_{j=i} Y^2_{ij} - \Sigma^v_{i=1} Y^2_i /_n$	
E (Linear)	1	$1/v(\Sigma^n_{j=1} Y_i I_j)^2 / \Sigma^n_{i=1} I^2_j$	
GxE (Linear)	v-1	$\Sigma^v_{i=1}[(\Sigma^n_{i=j} Y_{ij} I_j)^2 / \Sigma^n_{j=1} I^2_j]$ – E (Linear) S.S.	
Pooled deviation	V(n-2)	$\Sigma^v_{i=1} \Sigma^n_{j=1} \sigma^2_{ij}$	MS_3
Genotype 1	n-2	$\Sigma^n_{j=1} Y^2_{Ij} - (YI)^2 / n - [\Sigma^n_{j=1} Y_{Ij]}{}^2 / \Sigma^n_{j=1} I^2 j$	MS_4
Genotype V	n-2	$[\Sigma^n_{j=1} Y^2_{vj} - (Yv)^2/n] - [\Sigma^n_{j=1} Y_{vj} I_j]^2 / \Sigma^n_{j=1} I^2 \, \Sigma^n_{j=1} {}^S 2_{vj}$	
Pooled error	N(v-1) (v-1)	$\Sigma^n_{j=1}$ 1/n S^2_{ei}	MS_5

Where,

V= number of variance; n = number of environments; S^2e = estimates of error mean square at each environment; CF = Correction factor.

2. The genotype x environment interaction was tested using 'F' test

$$F = M_{S2} / M_{S5}$$

3. The genetic difference among genotypes for their regression on environmental index tested using 'F' test

$$F = M_{S3} / M_{S4}$$

4. The deviation on bi values from the unity was test using 't' test:

t = b-1 /SE (b) at V (n-2) degree of freedom

RESULTS AND DISCUSSION

The analysis of variance (ANOVA) revealed highly significant differences for genotypes and environment (linear) for three characters, namely fresh herb yield (Kg/plot), oil yield (g/plot) and menthol yield (g/plot) while, highly significant for the only two characters, namely fresh herb yield (Kg/ plot), and oil yield (g/plot) for the G× interactions (linear). For the G×E and E+ G×E were not significant for all the three characters, respectively (Table 1, Fig. 1-3). Therefore, the existing genetic variability in these crops was very high among genotypes. The existing genetic variability has been the primary source of diversity in the diverse germplasm useful for crop improvement. Menthol mint (*M. arvensis* L.) is propagated for its commercial cultivation entirely through vegetative means. Its existing genetic variability is much restricted. Hence, it needs to be supplemented with ample genetic variations for crop improvement via the deployment of systematic and powerful breeding approaches like induced mutagenesis, inter-specific hybridization, selection in clones raised from open-pollinated seed progenies and selection in its *in vitro* somaclones (Singh and Chaudhury, 2014; Tyagi, 1986). Because mint is a cross-pollinated crop and propagated through vegetative means by underground suckers. It is useful for plant breeding as after obtaining genetic variants in open-pollinated seed progenies it can be fixed through clonal selection (Patra et al., 2001a). Nevertheless, in peppermint (*M. piperita*) and menthol mint (*M. arvensis*), the remarkable genetic variability found, remains latent in the

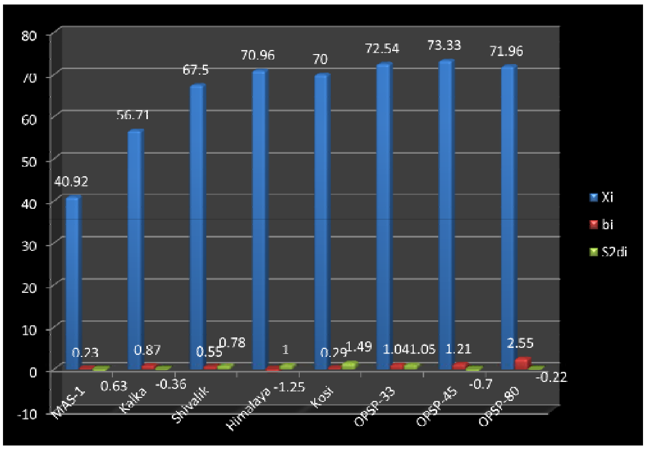

Figure 1: Stability parameters for herb yield

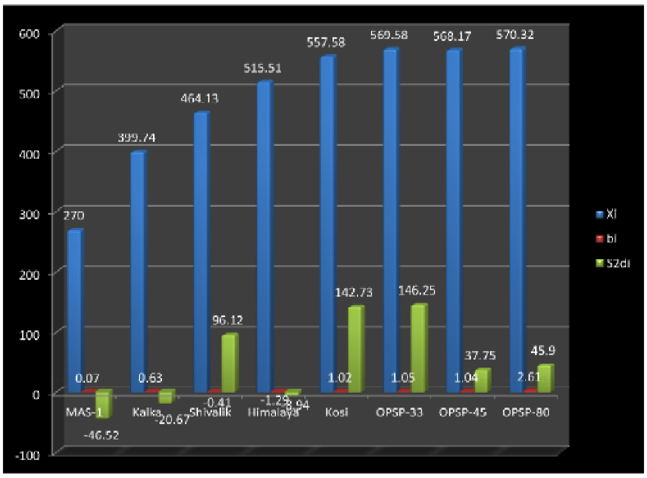

Figure 2: Stability parameters for oil yield

Figure 3: Stability parameters for menthol yield

existing vegetatively propagated/heterozygous clones of the varieties. (Khanuja et al., 2001; Kumar et al., 2004; Patra and Kumar, 2004; Patra et al., 2000). As reported by these researchers the oil content ranged between 0.37-1.08 in the OPSPs of M. *arvensis* cv. Shivalik. In agreement with these findings, the results of *M. arvensis* cv. Shivalik in the present study has also shown the existence of wide genetic variation in oil content range: 0.42 –

Table 1. ANOVA in regression analysis

Source of variance	d.f.	Mean sum of squares (mss) of the traits		
		Fresh herb yield (Kg/plot)	Oil yields (g/plot)	Menthol yield (g/plot)
Genotypes (G)	7	1015.00^{**++}	92490.20^{**++}	55632.16^{**++}
Environments (E)	7	6.26^{++}	405.26^{++}	185.99
G × E	49	3.20+	258.38	195.57
E+ (G×E)	56	3.58+	276.74^{+}	194.38
E (Linear)	1	43.93^{**++}	2837.11^{**++}	1304.51^{**++}
G × E (Linear)	7	11.33^{**++}	898.52^{**++}	128.52
Pooled deviation^{+}	48	1.61	132.72	151.69
Pooled error*	112	3.94	247.18	337.37

*P<0.05;**P<0.01; +P<0.05; ++ P<0.01

0.79% and the range of single plant oil yield: 0.52 – 4.4 g) in the derived 115 OPSPs, which has facilitated selecting 30 elite OPSPs having the single plant oil yield range of 2.28 – 4.40 g.

The developed clonal populations of the selected 30 OPSPs, when further assessed of their variability in morpho-physiological fitnesses for nine characters against the standard varieties including the so-far best variety Kosi in PYT and PST could reveal among them the presence of the three top-ranking genotypes: OPSP-33, OPSP-45, and OPSP-80 (Fig. 1-5). The per hectare oil yield in these three top-ranking OPSPs was 213.3, 211.3, and 209.8 kg, respectively, as against 191.7 kg of variety Kosi (Table 2). Thus, the three OPSPs could excel over the best control Kosi by giving 11.3, 10.2, and 9.5% additional oil yield, respectively (Table 1-3; Fig. 4-5).

The eight genotypes (three OPSPs + 5 control varieties) could constitute eight environments (i.e., seven different environments for seven genotypes + 1 self environment of each genotype). Stability and regression analysis of these for the three major characters herbage, oil, and menthol yield showed that the G×E mean squares being significant. All eight genotypes had intra- and inter-genotypic competitions, as also often suggested in menthol mint and other crops (Bahal et al., 2013; Gupta et al., 2016; Lal, 2013a, 2013b, 2015; Lal et al., 1999, 2001, 2010a; Mishra et al., 2017, 2018; Pandey, 1977; Patra et al., 2001b). Likewise, the values for the linear component being higher than the nonlinear components (pooled deviation squares for regression) for all the three traits. It was indicated that the performance of genotypes for the three characters could be reliably predicted in varying environments of mint genotypes. The results of the stability parameters (mean, regression coefficient, and deviation from the regression) have revealed that among all the genotypes, OPSP-33 and OPSP-45 were most stable in performance for the three major traits. Over different environments because of their high mean, almost unit regression and < 0 or non-significant deviation from regression. Among the five control varieties studied, Kosi has come out as the best genotype for stable performance for both oil and menthol yield (high mean, b » 1, and S^2di negligible/non-significant) with all other genotypes (Tables 2-3; Fig. 1-3).

Figure 4: Highly stable genotype OPSP-80

Figure 5: Field view of highly stable genotype OPSPs-80

As it is now widely documented and recognized (Lal et al., 2010b, 2010c; Patra and Kumar, 2004; Patra et al., 2000c) that the country's long effort for accelerating commercial production of menthol mint oil have found in the late nineties of the last country, a fascinating success which is essentially accredited to the development and release of the early maturing superior variety 'Kosi' endowed with the potentiality of giving doubled oil productivity. Whereas the per hectare oil yield of the previous popular variety Shivalik was 90 kg, the existing Kosi variety-based per hectare yield is more than 190 kg (Patra et al., 2001a,2001b; Rao and Prasad, 1984; Verma et al., 2010). To concede the real situation, after the development and extensive cultivation of the variety Kosi (estimated cultivated area for Kosi according to these authors is 1.5 lakhs

Table 2: Mean performances of the selected thirty OPSPs of menthol mint for the nine characters in preliminary yields trials (plot size 6 m × 4 m)

Varieties/ genotypes	Plant height (cm)	No. of leaves/aerial stem	L:S ratio	Leaf length (cm)	Leaf width (cm)	Herbage yield/plot (kg)	Oil content (%)	Oil yield/plot (g)	Menthol yield/plot (g)
MAS-1	75.33(±2.18)	94.33(±4.05)	0.83(±0.003)	6.80(±0.08)	3.35(±0.06)	30.54(±1.15)	0.67(±0.005)	204.61(±9.06)	165.44(±7.40)
Kalka	95.11(±2.62)	97.90(±7.31)	0.86(±0.006)	7.50(±0.08)	3.80(±0.08)	46.42(±1.76)	0.70(±0.011)	324.94(±6.87)	244.45(±4.12)
Shivalik	90.50(±3.54)	108.62(±8.51)	0.80(±0.006)	7.70(±0.05)	4.22(±0.09)	56.17(±1.06)	0.68(±0.006)	381.95(±7.66)	282.64(±5.60)
Himalaya	101.73(±3.41)	107.35(±5.85)	0.92(±0.006)	8.00(±0.14)	4.65(±0.06)	61.16(±1.90)	0.72(±0.003)	440.35(±2.11)	340.39(±7.83)
Kosi	98.50(±2.13)	130.18(±0.01)	1.15(±0.035)	7.60(±0.08)	3.92(±0.06)	57.90(±1.60)	0.79(±0.006)	457.41(±4.00)	359.34(±8.54)
OPSP-1	87.90(±3.17)	95.66 (±6.65)	0.81(±0.012)	7.10(±0.05)	3.80(±0.06)	47.77(±1.45)	0.73(±0.010)	348.72(±10.48)	263.03(±9.12)
OPSP-3	83.60(±4.48)	98.30 (±4.40)	0.78(±0.023)	7.20(±0.11)	3.90(±0.07)	45.67(±1.76)	0.68(±0.011)	310.55(±10.13)	239.43(±6.11)
OPSP-7	86.90(±9.21)	101.50(±9.49)	0.96(±0.003)	7.00(±0.05)	3.45(±0.12)	42.00(±1.17)	0.75(±0.008)	315.00(±11.08)	234.76(±7.69)
OPSP-9	78.80(±3.32)	95.35(±10.49)	0.77(±0.012)	7.20(±0.03)	4.00(±0.06)	46.92(±2.30)	0.70(±0.009)	328.44(±9.36)	253.42(±4.57)
OPSP10	85.50(±2.60)	99.80(±3.38)	0.81(±0.006)	7.30(±0.63)	4.10(±0.64)	48.30(±1.15)	0.69(±0.003)	333.27(±10.13)	254.81(±7.63)
OPSP-11	82.30 (±2.75)	97.49(±7.09)	0.80(±0.007)	7.00(±0.08)	4.20(±0.09)	51.54(±1.15)	0.71(±0.006)	365.93(±7.26)	273.20(±5.48)
OPSP-15	80.00 (±9.72)	99.43(±8.83)	0.78(±0.037)	7.40(±0.57)	4.10(±0.12)	50.70(±1.20)	0.77(±0.006)	390.39(±6.52)	294.97(±9.09)
OPSP-19	89.60(±2.39)	101.47(±7.50)	0.83(±0.007)	7.40(±0.03)	4.10(±0.06)	47.77(±0.57)	0.73(±0.063)	348.72(±4.30)	270.11(±2.42)
OPSP-25	81.50(±2.22)	93.55(±3.78)	0.78(±0.033)	7.50(±0.03)	4.20(±0.06)	42.54(±1.15)	0.71(±0.003)	302.03(±6.75)	230.93(±5.40)
OPSP-28	79.80(±2.38)	97.49(±6.35)	0.82(±0.033)	7.10(±0.05)	4.00(±0.63)	52.60(±0.88)	0.76(±0.003)	399.76(±8.60)	305.13(±4.42)
OPSP-33	98.50(±10.91)	112.30(±5.23)	0.98(±0.033)	7.50(±0.08)	4.12(±0.11)	65.00(±1.23)	0.78(±0.003)	507.00(±6.07)	402.20(±4.58)
OPSP-36	78.40(±4.18)	100.50(±10.78)	0.85(±0.008)	7.20(±0.05)	4.20(±0.06)	48.66(±0.88)	0.72(±0.006)	350.35(±2.08)	268.36(±1.56)
OPSP-39	86.50(±3.08)	98.73(±9.52)	0.79(±0.020)	7.30(±0.05)	4.30(±0.03)	51.49(±0.57)	0.75(±0.006)	386.17(±4.38)	295.14(±5.96)
OPSP-42	79.16(±4.27)	101.12(±6.88)	0.84(±0.006)	7.00(±0.03)	4.16(±0.08)	55.77(±1.20)	0.73(±0.060)	407.12(±8.65)	307.21(±6.26)
OPSP-45	100.50(±3.21)	116.25(±4.63)	1.12(±0.003)	4.50(±0.05)	2.0(±0.06)	65.40(±0.88)	0.77(±0.003)	503.58(±8.94)	398.98(±6.51)
OPSP-50	80.60(±1.89)	99.86(±2.84)	0.86(±0.014)	7.40(±0.05)	4.00(±0.06)	42.42(±1.20)	0.70(±0.003)	296.94(±7.50)	220.92(±5.33)
OPSP-55	85.45(±3.27)	94.20(±6.08)	0.93(±0.007)	7.00(±0.06)	4.30(±0.07)	43.26(±1.21)	0.77(±0.003)	333.10(±4.36)	250.69(±6.34)
OPSP-60	83.48(±1.97)	97.35(±5.60)	0.85(±0.038)	7.70(±0.54)	4.20(±0.06)	48.99(±1.52)	0.75(±0.006)	367.42(±5.78)	277.25(±8.06)
OPSP-64	90.43(±3.63)	89.38(±7.09)	0.81(±0.028)	7.10(±0.12)	4.10(±0.06)	45.05(±1.76)	0.71(±0.006)	319.78(±7.46)	230.88(±3.19)
OPSP-69	83.65(±3.61)	98.66(±5.81)	0.89(±0.037)	7.20(±0.06)	3.80(±0.03)	46.77(±1.20)	0.73(±0.003)	341.42(±6.76)	247.29(±4.93)
OPSP-72	86.33(±1.65)	91.45(±7.37)	0.94(±0.006)	7.30(±0.06)	4.00(±0.06)	54.89(±0.88)	0.74(±0.003)	406.18(±7.48)	289.85(±5.46)
OPSP-77	81.80(±2.06)	98.77(±8.98)	0.87(±0.21)	7.60(±0.05)	3.78(±0.05)	56.04(±1.45)	0.71(±0.003)	379.88(±8.04)	291.64(±5.43)
OPSP-80	97.65(±3.58)	110.14(±6.88)	1.16(±0.006)	7.70(±0.03)	3.83(±0.05)	62.40(±0.57)	0.79(±0.060)	492.96(±2.49)	383.81(±1.81)
OPSP-85	88.25(±1.97)	101.33(±8.19)	0.87(±0.033)	7.00(±0.06)	3.80(±0.07)	56.04(±0.88)	0.71(±0.003)	397.88(±6.67)	295.46(±4.73)
OPSP-89	84.53(±3.30)	98.66(±8.71)	0.83(±0.037)	7.60(±0.06)	3.97(±0.03)	47.10(±1.20)	0.76(±0.003)	357.96(±7.00)	258.44(±5.33)
OPSP-93	81.40(±2.13)	95.67(±3.17)	0.81(±0.008)	7.50(±0.09)	3.90(±0.03)	56.99(±1.06)	0.75(±0.003)	427.42(±5.53)	310.99(±4.04)
OPSP-95	87.50(±3.03)	100.45(±7.21)	0.97(±0.007)	7.40(±0.03)	4.11(±0.03)	53.89(±0.57)	0.74(±0.007)	398.78(±5.95)	304.26(±6.72)
OPSP-96	89.30(±2.34)	97.88(±6.93)	0.90(±0.035)	7.00(±0.08)	4.23(±0.05)	51.66(±0.88)	0.72(±0.006)	371.95(±8.92)	280.18(±6.28)
OPSP-103	90.15(±2.90)	91.70(±6.08)	0.83(±0.036)	6.93(±0.09)	3.80(±0.03)	50.77(±0.88)	0.73(±0.003)	370.62(±6.83)	260.28(±4.91)
OPSP-111	92.50(±3.39)	1.02.80(± 6.08)	0.86(±0.038)	6.90(±0.03)	3.60(±0.03)	54.99(±1.33)	0.75(±0.003)	412.42(±8.00)	298.59(±7.66)

ha), the development of further superior variety seems to be a significant challenge to the plant breeders. As also demonstrated by the results of the present study, a potent breeding technique like selection in open-pollinated seed progenies (the method which has also been used in developing the superior variety Kosi) has not been able to ensure improvement in yield level beyond 10%. In considering this aspect of OPSP selection *vis-à-vis* the recorded high potentiality (ensuring 27.6% yield improvement) of the other breeding approach of the present study: Mixed Varietal approach), it stands that resorting to the latter breeding approach in combination with the selection of OPSP would be a potent adjunct to the means of ensuring further yield improvement in menthol mint.

CONCLUSION

The present investigation was carried out at CIMAP Resource Centre, Pantnagar, Udham Singh Nagar, and Uttarakhand during 1998-2002. The experimental material comprised five menthol mint varieties MAS-1, Kalka, Shivalik, Himalaya and Kosi belonging to the *Mentha arvensis* L. The innovative plan was strategically made to conduct the study mainly in four steps: the first step concerned evolving new genotypes (open-pollinated seed progenies, i.e., OPSPs) from the variety Shivalik, the second step involved the clonal establishment of the top-ranking superior OPSPs (the outcomes of OPSPs selections) and their morphological characterization, the third step involved determining the level of genetic improvement for oil yield via OPSPs selections, and the fourth step concerned exploring the possibility of further upgrading the yield level of the genotypes. The success of this mostly depends upon the endowed properties of the genotypes to express their beneficial intra- and inter-genotypic competitions.

The developed new genotypes were established by their vegetative multiplications and repeated assessments in their morpho-physiological fitnesses for different plant characters including oil yield and quality potentials. All of them could be readily and firmly fixed for all plant characters through vegetative multiplications. The variability analysis for the developed new genotypes (OPSPs) and control varieties were performed by the use of standard statistical techniques essentially associated with Randomized Block Design. The analysis of inter genotypic competitions in the genotypic mixtures in the present study could be accomplished by following the sophisticated statistical methods used by (Singh and Chaudhury, 2014) for the analysis of five major parameters mean relative yield, relative yield total, adjusted mean, yield ability (over component lines, and overall varieties/OPSPs) and regression method based on Eberhart and Russell's (Eberhart and Russell, 1966) Model.

The study of G×E interaction in the eight environments for each variety (i.e., seven different environments of other varieties + oneself environment) revealed that all the studied eight genotypes had intra-and inter-genotypic competitions. The values for a linear component in the G×E interaction analysis being higher than the nonlinear components for the entire three traits studied namely, herbage, oil, and menthol yield. It has been suggested that performances of the genotypes in varying environments could be reliably predicted. The results of the stability parameters (mean, regression coefficient, and deviation from the regression) revealed that among all the genotypes OPSP-80, OPSP-33, and OPSP-45 are the most stable in yield performances.

Based on the information obtained from the present study, it is concluded that genetic selections in OPSPs, coupled with stability approach might ensure significant yield and quality augmentation in menthol mint.

Table 3: Regression coefficients (b_i), deviation from regression (S^2d_i) and mean performance (Xi) of different varieties/OPSP for the economic traits in eight environments

Genotypes	Stability parameters								
	Fresh herb yield (Kg/plot)			Oil yields (g/plot)			Menthol yield (g/plot)		
	Xi	b_i	S^2d_i	Xi	b_i	S^2d_i	Xi	b_i	S^2d_i
MAS-1	40.92	0.23	-0.63	270.00	0.07	-46.52**	22.41	-0.02	-63.56**
Kalka	56.71	0.87	-0.36	399.74	0.63	-20.67**	297.88	0.77	-73.75**
Shivalik	67.50	0.55	0.78	464.13	-0.41	96.12**	351.89	0.47	4.92**
Himalaya	70.96	-1.25*	1.00	515.51	-1.29*	-8.94**	392.67	-1.11	-85.28**
Kosi	70.00	0.29	1.49*	557.58	1.02	142.73**	428.92	1.06	226.24**
OPSP-33	72.54	1.04*	1.05	569.58	1.05	146.25**	451.08	1.01	14.27**
OPSP-45	73.33	1.21*	-0.70	568.17	1.04	37.75**	459.05	1.07	43.03**
OPSP-80	71.96	2.55**	-0.22	570.32	2.61**	45.90**	445.72	3.15**	334.05**
Mean ±	65.49	0.99	-	489.49	0.99	-	380.27	0.99	-
S.E.	0.48	0.54	-	4.35	0.61	-	4.65	0.96	-

*P<0.05, **P<0.01, x_i= mean; b_i = Regression coefficients S^2d_i - deviation from regression.

ACKNOWLEDGMENTS

The authors are thankful for the support and facilities provided by Director, CSIR-CIMAP, UP, Lucknow, India. This work supported by the Council of Scientific and Industrial Research, India. The second author is working as CSIR-Emeritus Scientist in the scheme No. 21 (1020)/16/EMR-II dated 18-11-2016.

Conflict of interest

The authors declare that there is no conflict of interest.

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