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Research Article

GENETIC VARIABILITY, GENETIC DIVERSITY, ASSOCIATION AND PATH ANALYSIS FOR ECONOMIC TRAITS IN INDIAN OREGANO (*Origanum vulgare* L)

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Abstract: To know the genetic variability, genetic divergence, the relationship between traits and path coefficient analysis of characters in Indian oregano accessions (*Origanum vulgare* L.). A total of fourteen Indian oregano accessions characterized for agro-morphological traits and chemical constituents of oil. Significant genetic variability noticed among the fourteen oregano accessions. The highest genotypic coefficient of variation (GCV)and phenotypic coefficient of variation (PCV) observed for herb yield (63.52, 64.22) respectively. Highest genetic advance(199.39) observed for herb yield. By D^2 values, all the fourteen oregano accessions grouped into two diverse clusters. The Cluster I was the largest group that comprised of ten oregano accessions. Maximum intracluster distance observed in cluster II (10344.76) and the minimum in cluster I (5720.50). A significant chemical variability also noticed among the fourteen oregano accessions. The genotypic correlation coefficient among the traits revealed that plant height significantly positively correlated with canopy diameter (0.80**) and internodal length (0.83**). Canopy diameter significantly positively correlated with herb yield - (0.72 **). The path coefficient analysis revealed that internode length (0.70) has a highest positive direct effect on oil yield.

Keywords: chemo-types, cluster, correlation, genotypic variance, heritability

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Introduction

Oregano is the sub-shrub or perennial herb of family *Lamiaceae*. The genus Origanum comprises 38 species, most of which are indigenous to the Mediterranean region. *Origanum vulgare* L. is the widest spread among all the species within the genus which distributed all over Europe, west and central Asia up to Taiwan [1]. In India, it widely distributed in sub-temperate/temperate Himalayas [2]. Oregano is a most precious culinary herb in world trade. It is widely used as a traditional remedy to treat various ailments such as whooping, digestive disorders and menstrual problems [3]. It is also used as raw material in the pharmaceutical, cosmetic, and food industries [4]. Recent findings revealed that antimicrobial, fungicidal, insecticidal, and antioxidant potential of the essential oil and extract of oregano [5]. Dried oregano leaves and essential oils are used largely by the flavoring industry in preparing various liqueur formulations, tomato sauces, condiments and in baked goods such as pizzas [6]. The oregano flowering in late summer and the plant height is ranged from 30 to 60 cm [7]. The glandular and non-glandular hairs covering the aerial parts and both types of hairs originate from epidermal cells [8]. The glandular hairs are more common on vegetative parts such as stems, leaves, and bracts, while their density lower on the reproductive organs such as calyxes and corollas [9]. The glandular hairs secrete an essential oil with a characteristic odor, mainly due to the presence of monoterpenes in the oil [10]. In India, till date, there was no study on genetic variability of oregano. So far, only a few studies have carried out, which demonstrated chemical diversity in Himalayan oregano [11, 12]. The major chemical components observed in the essential oil of oregano are Thymol and Carvacrol [13].

The availability of genetic information on the extent of genetic variations, heritability, relationship, path coefficient analysis of quantitative and qualitative traits is the basic requirement for planning the appropriate crop improvement programme in any crop. The correlation study helps in the understanding of yield contributing traits and assists in selection program [14,15]. Hence, the present investigation was planned to estimate the genetic variation, heritability, genetic advance, traits associations and path analysis for economic traits in oregano accessions. This study will help in genetic improvement of oregano.

Material and methods

1. Planting materials

The experimental planting materials for present study comprised of fourteen accessions of Oregano [Table-1].

2. Experimental site

This experiment conducted at the experimental field of CSIR-Central Institute of Medicinal and Aromatic Plants (CIMAP), Research Centre, Pantnagar, Uttarakhand, India. Located at coordinates of 29◦N, 79.38◦E and an altitude of 243.84 m.

3. Experimental design

All the fourteen accessions were planted in a randomized block design (RBD) with two replications in 3 m long rows with a row to row spacing of 60 cm and plant to plant spacing of 30 cm on raised beds (Bed size: 3 m x 3m) in the last week of November 2016. All required cultural practices followed throughout the crop season.

Table-1 Places of collection of fourteen accessions of Indian oregano

SN	Accessions	Places of collection/origin
1	OGP-1	Purara, Uttarakhand, India
2	OGP-2	Purara, Uttarakhand, India
3	OGP-3	Purara, Uttarakhand, India
4	OGP-4	Nandaprayag, Uttarakhand, India
5	OGP-5	Badrinath, Uttarakhand, India
6	OGP-6	Kapkote, Uttarakhand, India
7	OGP-7	Gopeshwar, Uttarakhand, India
8	OGP-8	Tharali, Uttarakhand, India
9	OGP-9	Mandal, Uttarakhand, India
10	OGP-10	Kausani, Uttarakhand, India
11	OGP-11	Chopta, Uttarakhand, India
12	OGP-12	Gwaldham, Uttarakhand, India
13	OGP-13	Karnaprayag, Uttarakhand, India
14	OGP-14	Purara, Uttarakhand, India

Table-2 Mean performance of Indian oregano accessions

SN	Accession No.	Plant height (cm)	Canopy diameter (cm)	Branches / plant	L:S ratio	Herb yield/plant	Internode length (cm)	Leaf petiole length (cm)	Leaf length (cm)	Leaf width (cm)	Oil yield (Ml)	Oil percent (%)	Thymol (%)	Carvacrol (%)	Days to harvest
1	OGP-1	83.00	143.50	19.00	1.02	257.00	4.00	0.80	1.10	0.70	0.30	0.30	23.12	0.80	124.00
2	OGP-2	25.00	63.50	21.00	0.81	60.00	2.00	1.28	1.60	1.20	0.35	0.58	1.20	53.83	130.00
3	OGP-3	42.00	91.50	56.00	1.45	360.00	1.50	0.97	1.90	1.30	0.15	0.15	9.77	0.97	110.00
4	OGP-4	44.00	84.00	66.00	1.98	259.00	2.00	1.85	2.15	1.35	0.11	0.11	0.20	0.83	100.00
5	OGP-5	35.00	84.00	97.00	2.05	246.00	1.50	0.82	1.15	0.90	0.15	0.15	23.54	1.22	136.00
6	OGP-6	47.00	80.00	68.00	1.64	196.00	2.50	0.98	1.25	0.85	0.10	0.10	1.54	1.23	128.00
7	OGP-7	38.00	98.50	52.00	2.60	173.00	2.25	1.02	1.70	1.25	0.15	0.15	17.64	0.86	118.00
8	OGP-8	34.00	46.50	24.00	2.54	58.00	2.20	1.28	1.75	1.30	0.10	0.17	1.88	0.40	148.00
9	OGP-9	34.50	62.50	37.00	2.49	97.00	2.00	0.92	1.40	0.85	0.20	0.21	0.50	0.50	134.00
10	OGP-10	46.50	55.50	22.00	2.36	67.00	2.70	1.28	1.55	1.00	0.15	0.15	22.24	2.10	129.00
11	OGP-11	40.50	73.00	39.00	1.50	73.00	2.10	1.15	1.45	1.05	0.15	0.14	12.54	42.51	132.00
12	OGP-12	37.43	51.00	46.00	3.25	91.00	1.80	1.35	1.80	1.20	0.16	0.16	19.24	15.42	136.00
13	OGP-13	24.60	51.50	56.00	3.81	93.00	2.00	1.13	1.50	1.40	0.11	0.11	15.32	1.05	105.00
14	OGP-14	33.80	51.50	70.00	2.15	96.00	2.00	1.05	1.50	1.45	0.16	0.16	14.24	1.24	123.00
	LSD(0.05%)	4.52	5.84	4.52	0.45	6.11	0.45	0.04	0.04	0.04	0.01	0.01	0.45	0.45	4.42

Table-3 Estimates of genetic parameters for economic traits of Indian oregano accessions

SN	Characters	Mean	V_g	V_P	GCV (%)	PCV (%)	h^2 (%)	GA	GAM	S.E.	C.D. (5%)	CV (%)
1	Plant height (cm)	40.38	199.38	198.96	34.34	34.31	99.79	29.03	70.6	0.46	1.39	1.57
2	Canopy diameter (cm)	74.04	674.93	674.52	34.91	34.90	99.94	53.48	71.88	0.45	1.39	0.86
3	Branches / plant	48.07	523.98	523.55	47.05	47.03	99.92	47.12	96.84	0.47	1.42	1.35
4	L:S ratio	2.12	0.63	0.62	37.28	36.93	98.12	1.61	75.36	0.08	0.24	5.10
5	Herb yield / plant (g)	151.86	9369.75	9369.36	63.52	64.22	97.98	199.39	130.84	0.44	1.34	0.41
6	Internode length (cm)	2.18	0.45	0.44	30.27	29.99	98.15	1.36	61.20	0.06	0.20	4.12
7	Leaf petiole length(cm)	1.13	0.07	0.07	23.50	23.48	99.83	0.55	48.33	0.01	0.02	0.96
8	Leaf length(cm)	1.56	0.08	0.08	18.50	18.49	99.83	0.60	38.05	0.01	0.03	0.77
9	Leaf width(cm)	1.13	0.06	0.06	20.72	20.66	99.47	0.48	42.45	0.01	0.04	1.51
10	Oil yield (ml)	0.17	0.01	0.01	41.79	41.65	99.33	0.15	85.52	0.00	0.01	3.42
11	Oil percent (%)	0.19	0.02	0.02	63.29	63.23	99.80	0.25	130.13	0.00	0.01	2.80
12	Thymol (%)	11.64	81.95	81.95	77.76	77.51	98.12	18.64	160.20	0.42	0.02	0.81
13	Carvacrol (%)	8.78	298.15	298.12	196.60	196.42	99.10	35.57	405.00	0.01	0.02	0.05

Table-4 Estimates of inter and intracluster distances

	Cluster -1	Cluster -2
Cluster- 1	5720.5	40765.81
Cluster-2	40765.81	10344.76

Table-5 Cluster means of thirteen economic traits of Indian oregano accessions

	Plant height	Canopy diameter	Branches plant^{-1}	L:S ratio	Herb yield / plant	Internode length	Leaf petiole length	Leaf length	Leaf width	Oil yield / plant	Oil percent	Thymol	Carvacrol
Cluster 1	36.13	63.35	43.5	2.32	100.4	2.16	1.14	1.55	1.16	0.16	0.19	10.90	14.63
Cluster 2	51	100.75	59.5	1.63	280.5	2.25	1.11	1.58	1.06	0.18	0.18	13.21	0.99

Table-6 Genotypic correlation coefficients among thirteen economic traits in Indian oregano accessions

Variables	Plant height	Canopy diameter	Branches plant-1	L:S ratio	Herb yield / plant	Internodes length	Leaf petiole length	Leaf length	Leaf width	Oil yield	Oil percent	Thymol	Carvacrol
Plant height	1.00												
Canopy diameter	0.80**	1.00											
Branches / plant	-0.26	-0.07	1.00										
L:S ratio	-0.44	-0.55**	0.19	1.00									
Herb yield / plant	0.47	0.72**	0.45	-0.36	1.00								
Internode length	0.83**	0.58**	-0.54**	-0.3	0.02	1.00							
Leaf petiole length	-0.24	-0.38	-0.12	0.16	-0.21	-0.2	1.00						
Leaf length	-0.33	-0.29	-0.05	0.21	0.07	-0.44	0.78**	1.00					
Leaf width	-0.61**	-0.5	0.17	0.39	-0.15	-0.54**	0.52	0.73**	1.00				
Oil yield	0.26	0.36	-0.51	-0.58**	-0.07	0.38	-0.23	-0.3	-0.33	1.00			
Oil percent	-0.03	0.11	-0.52	-0.55**	-0.22	0.18	-0.04	-0.13	-0.13	0.92**	1.00		
Thymol	0.42	0.65	0.21	0.14	0.38	0.14	-0.93	-0.98	-0.4	0.03	0.18	1.00	
Carvacrol	0.24	0.03	0.14	0.06	0.07	0.13	-0.15	0.14	0.04	0.03	0.14	0.01	1.00

Table-7 Direct (bold) and indirect effects on thirteen economic traits related to path analysis in Indian oregano accessions

Traits	Plant height	Canopy diameter	Branches / plant	L:S ratio	Herb yield / plant	Internode length	Leaf petiole length	Leaf length	Leaf width	Oil percent	Thymol	Carvacrol
Plant height	-0.82	-0.20	-0.02	0.16	0.08	0.58	0.02	-0.12	0.10	-0.02	0.34	-0.27
Canopy diameter	-0.66	-0.25	0.00	0.19	0.11	0.42	0.03	-0.11	0.08	0.10	0.28	-0.16
Branches / plant	0.22	0.00	0.07	-0.07	0.07	-0.37	0.01	-0.02	-0.03	-0.53	0.10	-0.36
L:S ratio	0.38	0.14	0.02	-0.34	-0.06	-0.23	-0.01	0.07	-0.07	-0.56	0.20	-0.43
Herb yield / plant	-0.40	-0.18	0.03	0.13	0.16	0.03	0.02	0.02	0.03	-0.22	0.10	-0.42
Internode length	-0.69	-0.15	-0.04	0.11	0.01	0.70	0.02	-0.16	0.08	0.20	0.25	-0.13
Leaf petiole length	0.21	0.09	-0.01	-0.05	-0.03	-0.14	-0.09	0.27	-0.09	-0.03	-0.38	0.19
Leaf length	0.27	0.07	0.00	-0.07	0.01	-0.31	-0.07	0.35	-0.12	-0.10	-0.37	0.02
Leaf width	0.49	0.13	0.01	-0.14	-0.03	-0.37	-0.05	0.26	-0.16	-0.13	-0.19	0.03
Oil percent	-0.20	-0.08	-0.04	0.20	-0.01	0.27	0.02	-0.10	0.05	0.92	0.06	0.54
Thymol	0.34	-0.16	-0.36	-0.43	-0.42	-0.13	0.19	0.02	0.03	-0.19	0.54	-0.19
Carvacrol	-0.27	0.11	-0.52	-0.55	-0.22	0.18	-0.04	-0.13	-0.13	-0.19	-0.13	0.66

Residual effect = 0.13

The Farm Yard Manure (FYM) at the rate of 8t / ha and 60, 30, 30 kg/ha of nitrogen (N), phosphorus (P) and potassium (K) applied at the time of planting. About 40 kg/ha of nitrogen was top dressed in two equal splits at 30 days and 60 days after planting. The experimental field irrigated as and when the crop was required. Observation on various morphological traits like plant height (cm), canopy diameter (cm), branches / plant, leaf to stem (L:S) ratio, herb yield / plant, intermodal length, leaf petiole length, leaf length, leaf width, dry herb yield / plant, oil yield /plant,, Thymol (%),Carvacrol(%) and days to harvest was recorded. The above observations recorded at the full blooming stage of the crop by selecting five plants in each accession.

4. Essential oil isolation and Gas chromatography (GC)

Herb from individual oregano accession harvested at blooming stage. Harvested herb was shade dried for about ten days, and the essential oil was isolated from individual oregano accessions by hydro-distillation for about 3-4 hrs using a Clevenger apparatus. The percentage of essential oil content (% v/w) estimated on a dry weight basis. The gas chromatography (GC) analysis of the oil sample was carried out on a Nucon Gas Chromatograph model 5765.

5. Statistical analysis

Statistical analysis was carried out by using Windostat statistical software 9.3 versions available at CSIR-CIMAP Research Centre, Pantnagar, based on the standard methods [15, 16]. The analysis of variation (ANOVA), genetic variability parameters like phenotypic variation (V_p), genotypic variation (V_g), the genotypic coefficient of variation (GCV), the phenotypic coefficient of variation (PCV), heritability (h^2) and genetic advance (GA) was estimated. Genetic divergence estimated using Mahalanobis D^2 statistics [17]. The clustering done by Torcher methods[18]. The mean values of all the traits subjected to correlation and path coefficient analysis [19].

Results and discussion

1. Genetic variability

The data in [Table-2] represent the means of all the traits. The plant height varied from 24.60 to 83.00 cm, herb yield ranged from 58.00 to 360.00 g/plant, and oil percent was ranged from 0.10 to 0.58%. The study of Analysis of Variation (ANOVA), standard errors (SE) and critical difference (CD) revealed highly significant differences among all the fourteen accessions of Oregano [Table-3]. The genotypic coefficient of variation (GCV) and phenotypic coefficient of variation (PCV) indicated a considerable amount of genetic variability, thereby emphasizing wide scope for selection for the improvement of economic traits of Oregano. The estimate of GCV and PCV was found to be highest for herb yield (63.52, 64.22) and essential oil content (63.29, 63.23), respectively. The estimates of PCV were almost equal to the corresponding GCV estimates for plant height, canopy diameter, branches per plant, L: S ratio, herb yields per plant and oil yield indicating that the environment less influenced the characters. Therefore, selection by phenotype alone can be effective for the improvement of these traits. A significant amount of genetic variation reported for herb yield, leaf: stem (L: S) ratio and other morphological characters. The heritability was highest for herb yield (99.98%), followed by canopy diameter (99.94%) and branches per plant (99.92%). The estimate of heritability alone is not very much use because it includes the effect of both additive and non-additive genes. The estimate of genetic advance was found to be highest for herb yield/plant (199.39) and lowest for oil yield/plant (0.15). The genetic advance as percent mean was highest for herb yield (130.84) and lowest for leaf length (38.05), this finding was in good agreement with earlier reports [20, 21, 22]. The maximum genotypic coefficient of variation and genetic advance as a percentage of the mean was reported for pulegone, followed by menthofuran and 1, 8-cineole in Peppermint (*Menthe piperita*) [23]. Significant genetic variability reported for agronomical and chemical constituents of oil in twenty-seven varieties of six Mentha species [24]. The herb yield per plant had the highest heritability with high genetic advance, and additive gene action predominantly controlled this character. Hence, genetic improvement through selection is most effective.

The GCV along with heritability estimates would provide a better idea of the amount of genetic gain expected through phenotypic selection [25]. High heritability coupled with high genetic advance observed for herb yield. This trait is under additive genetic control, and simple selection for this trait would be quite effective. By D^2 values, all the fourteen oregano accessions grouped into two diverse clusters [Fig-1]. Cluster I was the largest group that comprised ten accessions and Cluster II, consisted of four accessions. A set of thirty-eight genotypes of Japanese mint Mentha arvensis grouped into eight clusters based on essential oil yield and quality components. The maximum intracluster distance observed in cluster II (10344.76) and the minimum cluster distance for cluster I (5720.50). The Intercluster distance was 40765.81 [Table-4]. Highly divergent genotypes would produce a broad spectrum of variability in the subsequent generation for morphological and quality traits enabling further selection and improvement. The earlier studies reported that the parents for recombination breeding should be from two clusters with wider inter clusters distance to get maximum variability in the segregating generations [26]. Hybridization between genotypes of the different cluster required for the development of desirable genotypes. Based on the cluster means [Table-5] the important cluster for high oil yield was cluster II that possesses higher oil yield (0.18 %) along with high herb yield (280.50 %) and high plant height (51.00 %).

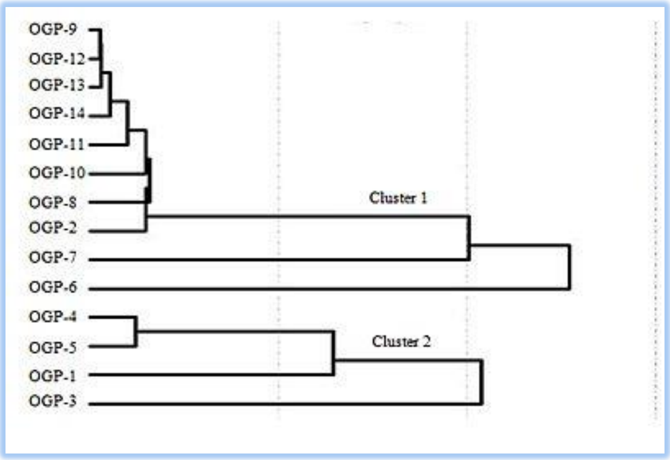

Fig-1 Dendrogram depicting the genetic variability in fourteen accessions of Indian Oregano

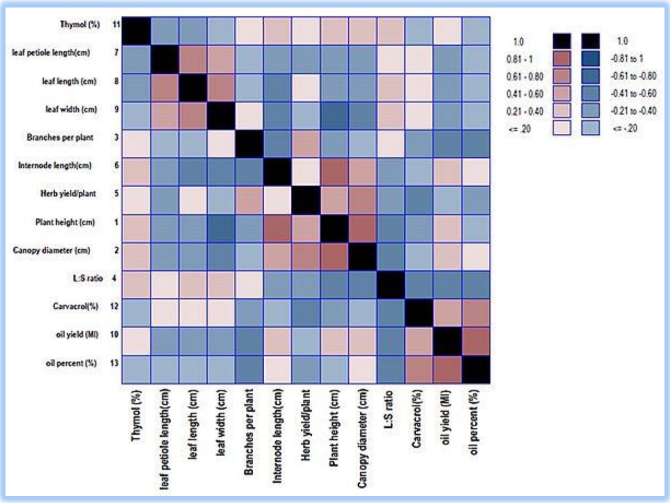

Fig-2 Shaded genotypic correlation matrix of thirteen economic traits in Indian oregano accessions

The maximum divergence observed in cluster II.

2. Phenotypic correlation

The genotypic correlation coefficient among thirteen traits presented in [Table-6]. The genotypic correlation coefficient among thirteen traits revealed that plant height significantly positively correlated with canopy diameter (0.80 **) and internode length (0.83**). The significant positive correlation observed between leaf petiole length (0.78**) and leaf length [Fig-2]. A similar type of results among different agro-morphological traits of Oregon reported earlier [22]. A significant positive correlation was reported for different chemical constituents in Peppermint (*Menthe piperita*) [23]. The significant and positive correlation reported between fresh herb yield with plant height, oil yield, and internodes in Mentha species [24].

3. Path coefficient analysis

The path coefficient analysis was worked out to know the direct and indirect effects of different traits on oil yield [Table-7]. The highest positive direct effect on oil yield was due to internodes length (0.70), leaf length (0.35), herb yield (0.16) and a number of branches (0.07). The plant height had a negative direct effect (-0.82) followed by L: S ratio (-0.34) and canopy diameter (-0.25). It showed that the improvement in herb yield would also lead to enhancement in oil yield. However, the other quantitative traits had a negative correlation, and negative direct effect indicates that care should be taken to consider these traits in selection.

Conclusion

The genetic improvement of any crop dependent upon the amount and nature of genetic variability present in that particular crop. In this study, we evaluated fourteen oregano accessions to understand genetic variability, genetic divergence, traits associations and direct and indirect contribution of various traits on oil yield. The genotypic and phenotypic coefficient of variation was largest for herb yield followed by oil percentage. All fourteen oregano accessions divided into two divergent clusters. It was evident from the path coefficient analysis that, the internode length (0.70) was the highest direct contributor to oil yield. All the above-studied traits showed high heritability (h^2) and medium to high genetic advance with positive genetic associations. The D^2 study revealed significant genetic diversity in the oregano accessions.

Application of research

Considering the huge scope of industrial uses of the Indian Oregano (*Origanum vulgare* L.) in pharmaceutical, food, flavor and fragrance industries. In India, commercial cultivation of oregano could not popularise till date due to lack of superior varieties for commercial cultivation. So, the present study will help in genetic improvement of oregano by understanding genetic variability, phenotypic associations and direct and indirect contribution of various yield components.

Research Category: Medicinal and Aromatic Plants

Abbreviations:
GCV: Genotypic Coefficient of Variation
PCV: Phenotypic Coefficient of Variation
GC: Gas Chromatography

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