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RESEARCH ARTICLE

Genetic Diversity in Natural Populations of Seabuckthorn (*Hippophae* L.) Growing in Cold Desert of Ladakh, India

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Abstract

Detailed surveys were carried out to study the wild germplasm of seabuckthorn (*Hippophae* L.) growing at high altitudes in Ladakh. Different regions of this cold desert were scanned to document the degree of variation in the naturally growing populations of this economically important taxon. Observations recorded on 26 quantitative traits of vegetative and reproductive nature were compiled from 122 plants belonging to 16 populations, and the mean values were subjected to Principal Component Analysis. This was done to determine and assess the extent of genetic diversity existing in the germplasm. Extensive variability at the intra- and inter-population levels was seen across the four major areas of Leh district in the Union Territory of Ladakh. The variation seems to be influenced largely by the local environmental factors and micro-habitat conditions.

Keywords: Cold desert, Genetic diversity, Germplasm, Heritability, *Hippophae*, Ladakh, Variability

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Introduction

The genus *Hippophae* L. (Seabuckthorn) is a drought-resistant, hardy deciduous, largely dioecious shrub/treelet (Ali, 2013; Tamchos, 2018). The genus is represented by 7 species and 11 subspecies distributed throughout the arid, semi-arid and high-altitude regions of Asia and Europe. Various geographical localities between 27˚N and 69˚N and 7˚W to 122˚E at altitudes ranging from 0 to 5000 masl harbour this species in abundance (Rousi, 1971; Bartish *et al*., 2000). The taxon assumes great importance because of its medicinal, ecological and nutritional values and, therefore, is being explored across the world. Recognizing the immense potential and applications of this plant, many countries like China, Russia, Germany, etc. have brought the species under cultivation (Utioh *et al*., 2007). It has been domesticated in the USA and Canada (Li and Beveridge, 2003). Those found in Asia are reported to exhibit a wide range of variation in almost every morphological feature, particularly in the size of leaves, thorns and fruits and also in fruit colour and shape (Rousi, 1971).

Four species of *Hippophae* L. (*H. rhamnoides* ssp. *turkestanica*, *H. tibetana*, *H. salicifolia* and *H. gyantsensis*) abound in four Himalayan states and one Union Territory (UT) of India (Rousi, 1971; Clement, 1991; Naithani, 2004; Tamchos, 2018). Himachal Pradesh, Uttarakhand, Sikkim, Arunachal Pradesh and the UT of Ladakh of the erstwhile state of Jammu and Kashmir have these taxa distributed between 3,000 and

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5,000 masl (Naithani, 2004; Dwivedi *et al*., 2006; Acharya *et al*., 2010). Two of the 4 namely *H. rhamnoides* ssp. *turkestanica* and *H. tibetana* are found in Ladakh. The former is well adapted to almost the entire Ladakh, whereas the latter is restricted to only some pockets of Leh and Kargil districts (Stobdan *et al*., 2011; Abassi and Kaul, 2015; Tamchos and Kaul, 2016). Locally, Seabuckthorn is known by different names like "*Tsermang*", "*Tses-ta-lulu*", "*sTarbu*", "Nak-tser", "Tser-*Nak*", "*Charma*", etc. (Stobdan *et al*., 2011; Tamchos and Kaul, 2019). The locals generally use the plants for bio-fencing, as fodder and firewood, in jam, juice, jellies and local bakeries (Abassi and Kaul, 2015; Kaul and Tamchos, 2021).

Extensively worked out by Rousi (1971), the species in Asia is homoplastic in nature. Thereafter, many researchers have attempted to quantify the magnitude of genetic diversity within and between the species using different molecular markers (Bartish *et al*., 2000; Ruan, 2006; Chunfang, 2007; Raina *et al*., 2012). Higher levels of phenotypic and genotypic variability found in all species of Seabuckthorn have been attributed to long-term evolution of the genus (Truţă *et al*., 2010). Perusal of the published literature brings to the fore the limited documentation of the germplasm availability in India. Similarly, data on genetic diversity is neither consolidated nor validated.

To tap the potential of this taxon, it is, therefore, quite imperative to document and characterize the germplasm at morphological, cytological, molecular and biochemical levels. The present investigation is a step towards assessing the extent of genetic diversity in the taxon in Ladakh, which represents ca. 67% of the Indian germplasm (Abassi and Kaul, 2015; Tashi and Sudan, 2022). It also aims to consolidate the diversity in the morphological traits of *Hippophae rhamnoides*. The diversity has been critically analyzed and re-validated over a period of 4 years (2012–2015). The plant species grows naturally in geographically isolated valleys of Ladakh, each characterized by its own topography and eco-climate.

Materials and Methods

The present investigation is based on 122 plants of *H. rhamnoides* (L.) both male and female, belonging to 16 populations from Leh, Nubra, Sham and Changthang valleys of the Leh district in the Union Territory of Ladakh, India (Fig. 1). These populations are located between 32˚to 34˚N and 76˚to 78˚E at altitudes ranging between 2732 and 4377 masl (Figs. 1 and 2, and Table 1). Specimens of each plant have been deposited in the Herbarium of the Department of Botany, University of Jammu under their respective voucher numbers (Table 1). The plants were critically scanned for qualitative and quantitative characters as per the descriptor given by various workers (Hyvönen 1996; Mathew *et al*., 2007).

Table 1: Area-wise details of wild populations of Seabuckthorn collected and studied during the present investigation

Valley	Population		Herbarium voucher	Altitude	Elevation	
	Name	Designated as	No. HBJU	(masl)	Latitude N	Longitude E
Leh	Choglamsar	CHGS	15767 – 15776	3229.2 (3176-3248)	34° (05.279"-05.295")	77° (36.404"-36.455")
	Shey I	SHY I	15777 – 15786	3259 (3258-3262)	34° (05.289"-05.295")	77° (37.188"-37.254")
	Shey II	SHY II	15787 – 15789	3275 (3274-3276)	34° (05.239"-05.242")	77° (36.182"-37.191")
	Matho I	MT	15806 – 15813	3480	34°	77°
	Matho II	MT Hr	15814 – 15816	(3474-3492)	(00.056"-00.067")	(38.322"-38.362")
	Spituk	SPT	15790 – 157925	3205 (3202-3210)	34° (07.498"-07.541")	77° (30.914"-31.009")
Sham	Sanjak	SNJ	15817 – 15824	2732 (2731-2734)	34° (34.383"-34.397")	76° (31.371"-31.396")
	Skurbuchan	SKB	15825 – 15828	2884.25 (2862-2904)	34° (27.005"-28.233")	76° (45.141"-45.667")
	Nurla	NL	15829 – 15834	3049.17 (3031-3062)	34° (17.948"-17.961")	76° (59.220"-59.277")
	Basgo	BSG	15835 – 15840	3302 (3232-3357)	34° (13.192"-13.722")	77° (16.233"-16.647")
Chang-thang	Chumur	CHU	15841– 15848	4376.78 (4368-4380)	32° (31.195"-31.297")	78° (16.00"-16.4")

Nubra	Partapur	PTP	15849 – 15858	3111 (3102-3123)	34° (36.567"-36.675")	77° (26.883"-26.904")
	Yakfarm	YKF	15859 – 15868	3115 (3109-3121)	34° (33.743"-33.767")	77° (32.402"-32.512")
	Tsaking	TSK	15869 – 15878	3117.8 (3115-3124)	34° (33.153V-33.210")	77° (31.912"-32.011")
	Lakjung	LKJ	15879 – 15888	3143 (3138-3148)	34° (33.207"-33.269")	77° (37.761"-37.809")
	Trith	TRT	15889 – 15898	3173 (3170-3176)	34° (31.998"-32.032")	77° (39.338"-39.467")

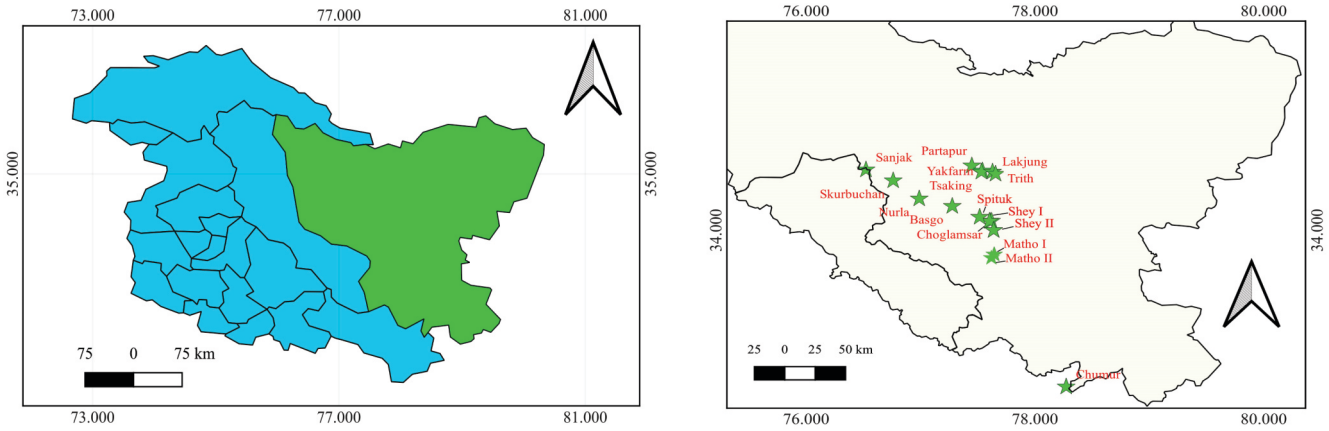


Fig. 1: Study area of Ladakh showing the distribution of natural populations. a) UT of Jammu and Kashmir and Ladakh; b) specific sampling localities in Ladakh.

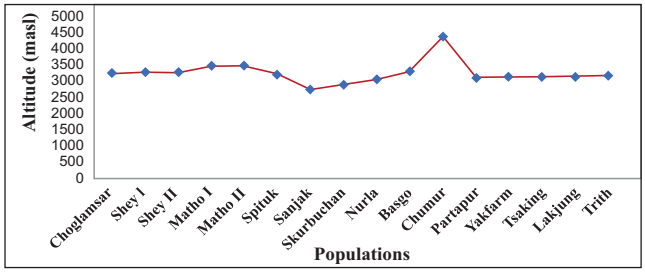


Fig. 2: Graphical representation of the altitudinal gradient of different populations

Observations and data collection were carried out in the field from live plants as well as in the laboratory (Biosystematics Lab., Department of Botany, University of Jammu), from their herbarium specimens prepared and deposited by us (Table 1). Data on quantitative characters like numbers of thorns, leaves, inflorescences and fruits were recorded per unit length of fruiting branch (1 unit length has been taken equivalent to 6 inches of any branch, fruiting or non-fruiting) and weights of fruits and seeds measured as g/100. Measurements were carried out with a scale or by using digital vernier callipers (Precise Measure). Observations were recorded for 26 morphological traits of vegetative and reproductive nature and compiled plant- and population-wise. The mean values of the recorded data of quantitative traits were statistically reviewed and subjected to principal component and cluster analyses to determine the significance of differences among the populations. UPGMA (Unweighted Pair Group Method with Arithmetic Mean) was done using PAST software (version 4.11) to draw phylogenetic relationships among the various populations. The degree of variability was estimated among the collections using Shannon-Weaver and Simpson diversity indices (Singh and Kaul, 2014; Singh *et al.*, 2014). Greater values of these indices indicate greater levels of genetic diversity. The values lie between 0 and 1, where 0 indicates no diversity, and 1 represents maximum genetic diversity possible (Singh *et al.*, 2014). The genotypic and phenotypic coefficients of variance, along with broad-sense heritability (h^2), were calculated as per the methods described by Lush (1940), Burton (1952) and Ahsan *et al.*, (2015) to assess the extent of genetic and environmental influences on phenotypic variation within the population.

The phenotypic (PCV) and genotypic (GCV) coefficients of variance are crucial in evaluating variation in plant species of economic importance. PCV measures the total observable variability in a trait, which includes both genetic and environmental influences, while GCV measures the variation due to

genetic factors alone.

In the case of PCV ≈ GCV, most of the variation is assumed to be genetic, and thus less influenced by the environment. In the case of PCV > GCV, the environment significantly influences the trait. Accordingly, the values of PCV and GCV prove useful in selecting traits important from the perspective of crop breeding. Traits showing high GCV are desirable as they have more genetic variability, are heritable, and thus have greater potential for improvement through selection. High heritability and high GCV indicate that selection would be effective and gains would be stable across environments. In short, these help in predicting breeding success; the higher the GCV, the more likely a breeder can achieve genetic improvement in future generations.

Calculations

Genotypic and phenotypic variances: These were calculated as:

(i) Genotypic variance

$$V_g = \frac{MSG - EMS}{\text{Number of replications}}$$

where, V_g = Genotypic variance; MSG = Mean square due to germplasm; EMS = Error mean square.

(ii) Phenotypic variance

$$V_p = \frac{V_g + V_e}{\text{Number of replications}}$$

where, V_p = Phenotypic variance; V_g = Genotypic variance; V_e = Error variance, *i.e.* EMS

The respective coefficients of variance, GCV and PCV, were calculated as per Burton (1952) and Ahsan *et al*., (2015).

$$GCV = \sqrt{\frac{V_g}{\bar{X}}} \times 100$$

where, V_g = Genotypic variance;

$$PCV = \sqrt{\frac{V_p}{\bar{X}}} \times 100$$

where, V_p = phenotypic variance;

Broad-sense Heritability (h^2): It was estimated as Lush (1940):

$$h^2 = \frac{V_g}{V_p}$$

where, h^2 = Heritability in broad sense; V_g = Genotypic variance; V_P = Phenotypic variance.

Results and Discussion

The present study encompasses four geographically different regions of only one district of Ladakh. Leh, Nubra, Sham and Changthang differ in their gross environmental conditions. Leh, at 3176 to 3492 masl, is the center of Ladakh, located along the Indus, whereas Sham (lower Ladakh) is relatively low-lying (2731 to 3357 masl) and warmer than other parts of Ladakh and is located towards Leh. Changthang, between 4368 and 4380 masl, is the remotest of the four, occurring in the high-altitude Tibetan plateau where nomadic people live. Nubra Valley at 3102-3176 masl is a desert area of Ladakh. Not with standing the geographical disparity, the plants of *Hippophae* L. are well established across this region, with a decrease in their density towards Changthang and Sham valleys.

Phenotypic variation was quantified for all the plants of these regions of Ladakh. Data for 26 quantitative morphological traits of vegetative and reproductive were generated for 122 plants in the present study. Plants tend to differ from each other at various levels, i.e., within and between populations, and among the sexes. On average, plants of Nubra Valley except Skurbuchan are the tallest (6.94±1.15 m) across the populations investigated (Fig. 3a). In contrast, those of Chumur are the smallest, attaining a maximum height of 30 cm irrespective of the sexes, while those of Leh and Sham fall in between. Similarly, thorn number per unit length of branch is maximum in plants of Leh, and minimum in Skurbuchan (Fig. 3a). Plants of Skurbuchan are also characterized by the smallest thorns (Fig. 3a). Like thorn number, the leaf number and size are also greater in plants of Leh as compared to those of Nubra. Thorns and leaves attain maximum size in plants of the Spituk population (Fig. 3a).

Differences in morphological attributes are manifested in reproductive traits also. As far as the sex expression is concerned, the plants are strictly dioecious with the male: female ratio biased towards males in each population. However, exceptions existed in that plants of three populations, namely Shey II and Skurbuchan, were only female and those of Matho II were only hermaphrodite. In this way, the overall sex expression is not typically dioecy but polygamodioecy (Sen *et al*., 2014). The plants are also facultatively apomictic (Mangla *et al*., 2015; Ali and Kaul, 2017).

Irrespective of sex, flowers consisting of two tepals are colourless, nectarless, and odourless. Reproductive features like the number of inflorescences per unit length of branch and flowers per inflorescence are the traits that directly influence pollen production as well as fruit set. These two vary significantly among populations. The average number of inflorescences per branch is highest in plants of Choglamsar and Shey I and lowest in those of Chumur (Fig. 3b). However, flower number in a single inflorescence is highest in Nurla followed by Sanjak and Skurbuchan. It is lowest in

plants of Shey II (Fig. 3b). Like these, significant variations also exist in the sizes of inflorescence, flower and its parts, fruit and seed. The highest number of fruits is produced by plants of Basgo (Fig. 3b). Significant variation also exists in fruit weight; the heaviest is produced by plants of Chumur and the lightest by those of Spituk. The average fresh weight of fruits ranges between 10 g and 35.6 g per 100 fruits.

The seed weight varies slightly both within and between plants of a population and among populations. However, seed size (length and width) is the only trait that does not differ significantly among different populations, thereby indicating it to be a conserved attribute (Fig. 3b).

To interpret variability of this magnitude and convert it into a wide usable base for selection of important traits, diversity indices were calculated. Values of Shannon-Weaver and Simpson diversity indices for all the characters were quite high, pointing towards greater diversity in the germplasm at the genetic level. Except for the number of inflorescences/unit length and size of anther, the Shannon-Weaver index for the reproductive traits approaches 1 (Table 2). The values for the former two are 0.96 and 0.94, respectively. For vegetative traits, however, the values fall between 0.93 and 0.99. The Simpson's indices, on the other hand, are 0.91, 0.92 and 0.93, shown by 10, 4 and 12 traits, respectively. Of the 12 in the latter, 6 traits are vegetative and reproductive (Table 2). The four, namely number of inflorescences/ unit length, size of anther, length of fruit and plant height, were uniform at 0.92, and the remaining 10 at 0.91. Similar studies have also been conducted on morphological and molecular data to quantify the magnitude of genetic diversity in chickpea, garlic and brinjal (Upadhyaya, 2003; Singh and Kaul, 2014; Singh *et al*., 2014; Singh *et al*., 2026). Diversity of this degree in seabuckthorn is quite obvious, characterized as it is by dioecism, sexual polymorphism, facultative apomixis and asexual mode of propagation (Sen *et al*., 2014; Mangla *et al*., 2015; Ali and Kaul, 2017). According to Singh and Kaul (2014), a high magnitude of diversity helps the species to sustain itself over diverse ecological conditions encountered over its large geographical range. Similar results were also obtained in *Cleome viscosa* L. (Saroop and Kaul, 2020). Such variables as per these authors strongly support the hypothesis that the species is moving towards diversifying selection via phenotypic polymorphism. This could eventually lead to speciation within a taxon. According to Draghi (2019), phenotypic variability can promote the evolution of adaptive plasticity.

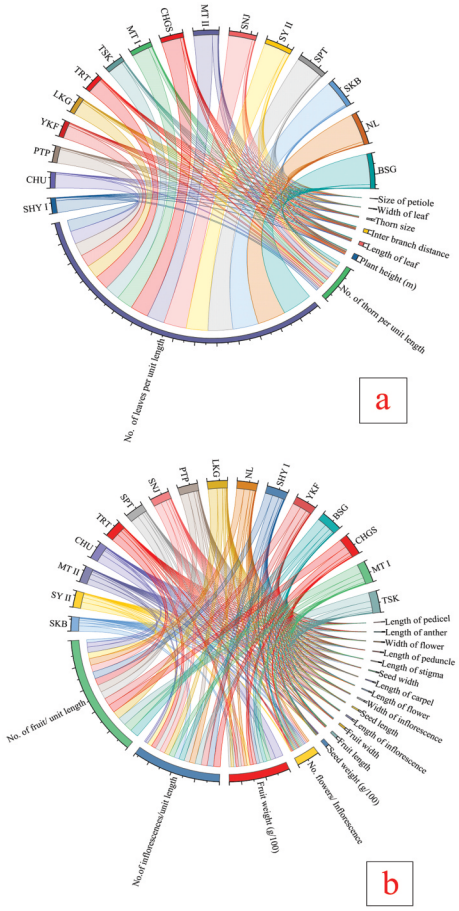


Fig. 3: Chord diagrams consolidating the extent of variation in vegetative (a); and reproductive (b) traits in *Hippophae rhamnoides*.
The lower half of each chord represents the traits and the upper half the populations.

According to Allard (2010), the coefficients of phenotypic and genotypic variance are reliable tools for determining the extent of variation within the germplasm. The two coefficients are influenced by environmental and genetic factors, respectively; their values in the present study range from 36.8 to 140.4 and 6.9 to 131.7. Clearly, the estimates of the former exceed those of the latter, indicating greater influence of the environment on the expression of characters. Interestingly, the lowest values of both PCV and GCV were recorded for seed width (36.8 and 6.9) and the highest for plant height (140.4 and 131.7). These correspond to the lowest and highest estimates of broad-sense heritability (h^2). Based on h^2 values of $\geqslant 0.5$, <0.5, and <0.2, the traits were classified into three different groups to gain insights into the role of genetic and environmental factors in their expression (Table 3). The lowest h^2 was recorded for seed width, followed by that for fruit length (Table 2). Similar findings have been reported in sorghum (Median and Chaudhary, 1980; Godbharle *et al*., 2010) and brinjal (Singh *et al*., 2026).

Table 2: Diversity indices, PCV, GCV and broad-sense heritability of quantitative traits of Seabuckthorn growing in Ladakh

S. No.	Characters	Indices		GCV	PCV	h^2
		Shannon Weaver's	Simpson's			
A. Vegetative traits						
1	Plant height	0.93	0.91	131.7	140.4	0.87
2	Inter-branch distance	0.98	0.93	89.4	104.0	0.73
3	No. of thorns/unit length	0.96	0.92	102.2	119.5	0.73
4	Thorn length	0.99	0.93	42.8	64.6	0.43
5	No. of leaves/unit length	0.98	0.93	86.4	100.9	0.73
6	Leaf length	0.98	0.93	71.3	84.7	0.70
7	Leaf width	0.99	0.93	22.3	48.1	0.21
8	Length of petiole	0.98	0.93	72.6	84.0	0.74
B. Reproductive traits						
9	No. of inflorescences/unit length	0.96	0.92	115.9	126.8	0.83
10	No. of flowers/inflorescence	0.99	0.93	42.8	57.4	0.55
11	Length of inflorescence	0.99	0.93	55.5	67.4	0.67
12	Width of inflorescence	0.99	0.93	45.0	59.5	0.57
13	Flower length	0.99	0.93	28.7	51.4	0.31
14	Flower width	0.99	0.93	29.8	52.8	0.31
15	Pedicel	0.99	0.93	51.8	72.7	0.50
16	Anther	0.94	0.92	42.3	61.9	0.46
17	Length Carpel	1	0.91	32.2	50.3	0.41
18	Stigma	1	0.91	92.5	113.6	0.66
19	Fruit	1	0.92	10.9	38.7	0.08
20	Fruit width	1	0.91	21.1	42.3	0.24
21	Peduncle length	1	0.91	22.6	44.3	0.26
22	No. of fruits/unit length	1	0.91	39	54	0.5
23	Seed length	1	0.91	22.9	43.2	0.28
24	Seed width	1	0.91	6.9	36.8	0.03
25	Fruit weight	1	0.91	78.0	87.1	0.80
26	Seed weight	1	0.91	18.0	45.6	0.15

Table 3 summarizes the categorization of 26 traits (8 vegetative and 18 reproductive) into three groups on the basis of their broad-sense heritability (h^2) estimates. Traits in Group-I, amounting to 53.8% of the total, exhibit higher h^2 values, which indicate the significant role of genetic factors in the expression of these traits. Many of these, such as plant height; number of thorns, leaves, inflorescences and fruits/unit length; number of flowers/inflorescences; and fruit weight, are economically very important. This will help in selection of the choicest parents and their subsequent utilization in a breeding program for further improvement of the germplasm. The remaining (34.6%) traits in Group-II are influenced more by environmental factors, while 11.5% in Group-III seem almost exclusively under the control of the environment.

From the breeding perspective, the high broad-sense heritability (h^2) of Group-I traits indicates that phenotypic selection for these characters may be effective in identifying promising plants within the Ladakh germplasm. Agriculture in cold-arid regions has its own constraints, so selection could help prioritize traits relevant to architecture and productivity of plants. Features like plant height, number and size of thorns and leaves, number of inflorescences per branch and flowers per inflorescence, and fruits per unit length and their weight require an appropriate selection strategy keeping in view their direct bearing on the economic importance of the species. However, h^2 includes both additive and non-additive genetic effects and does not by itself predict the response to sexual breeding. Selected plants should therefore be evaluated across

Table 3: Summary of broad-sense heritability (h^2) based on morpho-quantitative traits

Group	h^2 value	No.	Traits Description	Major role played by
I	⩾ 0.5 (0.5-0.87)	14	Plant height, inter-branch distance, No. of thorns and leaves/unit length, length of leaf and petiole, No. of inflorescences/unit length, No. of flowers/inflorescence, length and width of inflorescence, pedicel and stigma lengths, No. of fruits/unit length and fruit weight.	Genetic factors
II	< 0.5 (0.21-0.46)	9	Leaf and fruit widths, peduncle and seed lengths, flower length and width, lengths of anther, carpel and thorn	Environmental factors
III	< 0.2 (0.03-0.15)	3	Seed width and weight, fruit length	Environmental factors (almost entirely)

years and representative cold-arid sites to confirm the stability of their performance. Further, aspects of their reproductive biology including sex expression (dioecy, hermaphroditism), facultative apomixis and vegetative propagation by runners need to be considered when planning controlled crosses, parent combinations and propagation schemes. Group-II and Group-III traits would require more careful evaluation under replicated environments because environmental effects may contribute substantially to their observed variation.

Principal component analysis (PCA)

Results of PCA applied to the morphological traits of wild seabuckthorn germplasm yielded 15 principal components. Of the 15, the first 7 had eigenvalues greater than 1. The highest eigenvalue was, however, observed in PC1 and the lowest in PC15 (Fig. 4; Table 4). The cumulative proportion of total variance explained by the 7 consecutive principal components is 87.67%. That explained by the first four is 66.94%; it increases to 74.76% if the 5th one is included, pointing towards the germplasm being genetically variable (Reim *et al*., 2012) (Table 4; Fig. 4).

PC1 and PC2 explained 44.2% of the total observed variation. PC1, represented by 13 traits, contributed 26.25% to this variation. PC1 increases with 8 [plant height, petiole size, leaf width; inflorescence number, length and width; flower length and width] variables

Table 4: Eigenvalues and their contribution to the total variation-obtained from the PCA and significance parameters in each component

		Principal Component Analysis (PCA)														
S. No.	**Morphological traits**	**PC 1**	**PC 2**	**PC 3**	**PC 4**	**PC 5**	**PC 6**	**PC 7**	**PC 8**	**PC 9**	**PC 10**	**PC 11**	**PC 12**	**PC 13**	**PC 14**	**PC 15**
1	Plant height (m)	**0.67474***	-0.18004	0.29186	-0.39948	0.091574	-0.017714	-0.16825	0.22553	0.29288	0.010697	-0.046008	0.25987	0.094148	0.11887	-0.03363
2	Inter branch distance (cm)	0.33422	-0.47325	-0.024443	0.0012348	0.45075	0.38539	0.10177	0.47675	-0.15691	0.1756	0.11007	-0.035461	0.072647	-0.014529	-0.008149
3	No. of thorn per unit length	-0.25886	-0.035125	-0.218	**0.66725***	-0.070221	0.47669	0.18523	-0.30301	0.088171	0.048926	0.12958	0.22271	-0.064204	0.0046989	-0.0051712
4	Number of leaves per unit length	0.41429	0.2359	**-0.55079***	0.34207	0.13072	-0.31303	0.38166	-0.068989	0.13736	0.14958	0.15252	-0.12502	0.052129	0.059034	-0.022304
5	Thorn size (cm)	**-0.51413***	-0.40347	0.31438	0.48025	0.068044	0.18274	0.20645	0.079878	-0.22475	-0.26944	0.053941	0.062769	0.13069	0.036099	-0.089663
6	Length of leaf (cm)	0.37902	0.4815	-0.2876	**0.57985***	-0.24488	0.10702	-0.10155	0.2456	-0.17728	-0.029168	-0.01213	0.12906	0.082974	-0.0098103	0.086349
7	Size of petiole (cm)	**0.59349***	**0.67649***	0.016676	-0.015054	0.25361	0.20056	-0.09859	0.037922	-0.029337	-0.20633	-0.094003	0.10867	-0.076497	0.030965	0.055167
8	Width of leaf (cm)	**0.64988***	0.3201	-0.36898	0.28554	-0.26476	0.1187	0.17899	0.20166	0.09349	0.26066	-0.1331	-0.059531	-0.017166	0.00030414	-0.048781
9	No.of inflorescences/unit length	**0.69911***	**-0.52745***	0.2235	-0.011502	-0.14492	-0.17786	0.12481	-0.094712	-0.11125	0.087661	-0.06034	0.18143	-0.18256	-0.1244	0.018799
10	No. flowers/ Inflorescence	0.23812	**0.60202***	0.28984	0.059083	0.058304	-0.44272	0.25229	0.34068	0.0024748	-0.21476	0.24096	-0.075042	-0.048748	-0.039881	0.021775
11	No. of fruit/ unit length	0.061972	**-0.77927***	0.23107	0.35216	-0.39033	-0.055163	0.099555	0.12125	0.027297	0.045383	0.033098	-0.14171	-0.06514	0.036893	0.043459
12	Length of inflorescence (cm)	**0.64044***	0.43195	0.41182	0.093354	0.4137	-0.11051	-0.015329	0.033177	0.073138	0.14133	0.057774	0.072118	0.004174	-0.07395	-0.030228
13	Width of inflorescence (cm)	**0.54639***	-0.0063048	**0.643***	0.39663	0.057409	0.002594	-0.099575	-0.16753	0.18896	-0.071596	0.1917	0.0022208	-0.049354	-0.081352	-0.052161
14	Length of pedicel (cm)	-0.11939	**0.80963***	0.33632	0.043417	-0.021889	0.32387	-0.10844	-0.069146	-0.045607	0.088749	0.11797	-0.069813	-0.1784	0.181	0.00073902
15	Length of flower (cm)	**0.52333***	0.34626	**0.60583***	-0.069782	-0.19094	0.068147	-0.02635	-0.20529	-0.32335	0.14735	-0.031242	0.0029826	0.14627	-0.0069814	-0.041212
16	Width of flower (cm)	**0.65569***	0.11468	0.48335	-0.12342	-0.4368	0.1177	-0.15011	-0.14398	-0.050386	0.039209	0.0048426	-0.22011	0.085806	-0.007416	0.01356
17	Length of anther (cm)	-0.32281	-0.12864	**0.54454***	0.10049	**-0.65626***	-0.063687	0.056621	0.30858	0.14464	-0.072143	-0.04305	0.04831	-0.064297	0.067237	-0.022293
18	Length of carpel (cm)	0.20756	-0.13306	0.38488	-0.21112	0.49038	0.27175	**0.60334***	-0.14533	-0.012343	-0.1005	-0.13776	-0.10434	-0.040621	0.068335	0.024636
19	Length of stigma (cm)	-0.088127	-0.079008	0.33235	**0.73154***	0.33127	-0.1589	-0.39238	-0.025813	0.024014	0.10757	-0.16203	-0.033718	0.073638	0.065711	0.038648
20	Length of peduncle (cm)	**-0.70892***	0.2304	0.00066942	-0.047521	0.17856	-0.42942	-0.27741	0.096256	-0.30727	0.12594	0.073608	0.051328	-0.13709	0.020692	-0.045077
21	Fruit length (cm)	-0.46557	-0.048506	0.3448	0.12188	0.11301	**-0.69845**	0.25311	-0.17436	0.052129	0.13097	-0.014015	0.10614	0.11616	0.067433	0.052083
22	Fruit width (cm)	-0.6042	0.22302	**0.56635***	0.0068375	0.0088982	0.16325	0.4056	0.14943	-0.02885	0.16796	-0.095891	0.052407	-0.024646	-0.060784	0.074497
23	Fruit weight (g/100)	**-0.80869***	0.4537	0.14473	0.021078	0.021931	0.08171	0.11768	0.12906	0.093044	0.15874	-0.18524	-0.0073489	-0.056522	-0.050711	-0.084516
24	Seed weight (g/100)	-0.4242	**0.55865***	-0.062211	-0.4391	-0.38446	0.14442	0.23301	-0.067047	0.0098291	0.076399	0.18999	0.1377	0.14945	-0.013377	0.01791
25	Seed length (cm)	**-0.67703***	**0.56347***	0.13272	0.20411	0.10057	0.12265	-0.15968	0.028555	0.22718	-0.12296	-0.10335	-0.10973	0.09134	-0.12451	0.0088724
26	Seed width (cm)	**-0.57407**	-0.4427	0.27189	-0.18431	0.19775	0.3305	-0.30789	0.044223	0.13092	0.1914	0.25005	-0.03529	0.0079595	-0.022456	0.057987
27	**Eigenvalue**	**6.82553**	**4.66812**	**3.34992**	**2.56197**	**2.03466**	**1.89336**	**1.46275**	**0.923024**	**0.589054**	**0.522663**	**0.407208**	**0.350442**	**0.232116**	**0.124522**	**0.0546707**
28	**% variance**	**26.252**	**17.954**	**12.884**	**9.8537**	**7.8256**	**7.2822**	**5.626**	**3.5501**	**2.2656**	**2.0102**	**1.5662**	**1.3479**	**0.89275**	**0.47893**	**0.21027**
29	**% Cumulative**	**26.252**	**44.206**	**57.09**	**66.9437**	**74.7693**	**82.0515**	**87.6775**	**91.2276**	**93.4932**	**95.5034**	**97.0696**	**98.4175**	**99.31025**	**99.78918**	**99.99945**

* Eigenvalues significant >0.50

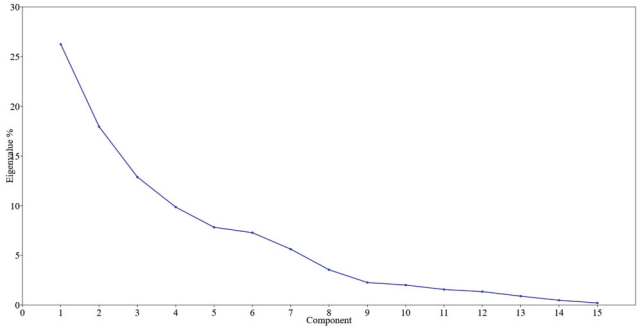


Fig. 4: Scree plot of the 15 PCs versus their % eigenvalues observed in the present investigation.

and decreases with 5 [thorn size, peduncle length, fruit weight, seed length and width] variables. That means PC1 increases 51.91% with these 8 traits and decreases 48.08% with the latter five. Together, therefore, this component focuses on the long-term survival, adaptability and stability of the species in the harsh environmental conditions of its native habitat. PC2 increases with 5 [petiole size, flower number per inflorescence, pedicel length, seed weight and length] traits and decreases with 2 [inflorescence and fruit numbers per unit length]. The influence of the former five is 72% against only ~28% of the latter two. This component primarily measures the extent of floral display (Fig. 5, Table 4). The traits such as leaf number per unit length, leaf size (length and width), size of petiole, flower number/inflorescence, inflorescence length, and flower size (length and width) showed positive correlation in both PC1 and PC2 (Figs. 5 and 6, and Table 4). However, the significant ones shared by the two PCs are only three: petiole size, inflorescence number and seed length. While petiole size is positively correlated in both, the latter two show a positive correlation in PC1 and a negative correlation in PC2.

High morphological and genetic diversity of *Hippophae rhamnoides* observed in Ladakh has been reported previously also. Using RAPD markers on 17 morphotypes from Nubra, Singh *et al.* (2006) found <25% similarity among them. Similarly, morphological and AFLP analyses of 17 populations of Leh and Nubra confirmed a strong correlation between traits and genetic variation (Korekar, 2013). Subsequent investigation by Dolkar (2018) revealed significant phenotypic plasticity across populations and sexes of seabuckthorn. These reports are in agreement with the conclusions made in the current investigation, as corroborated by the statistical analyses. In particular, the low similarity reported among RAPD-defined morphotypes provides molecular evidence (Rana and Shirkot, 2012; Srihari *et al*., 2013) consistent with the substantial differentiation observed among the populations in the present morphological analysis. The two data types are complementary: morphological traits describe expressed phenotypic differences, which may reflect genetic variation as well as environmental influence, whereas molecular markers provide an independent estimate of genetic relationships. Korekar

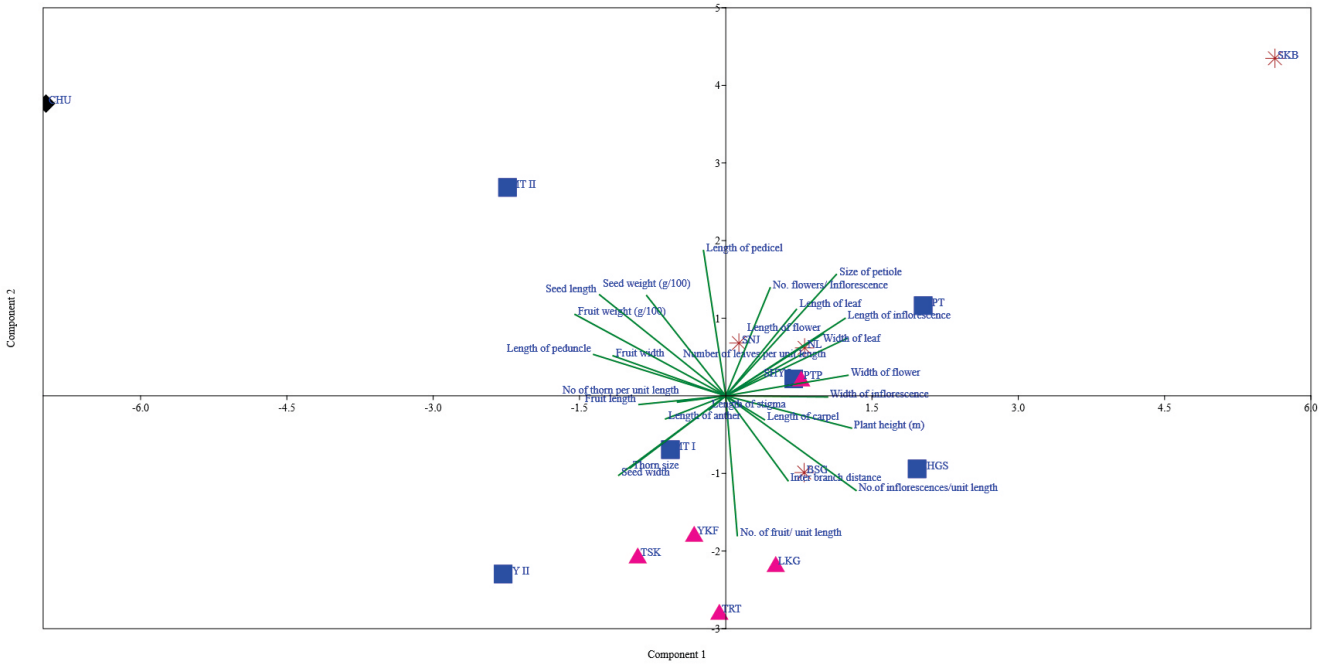


Fig. 5: PCA plot based on the first two components of morphological traits (using PAST software version 4.11) of wild seabuckthorn populations collected from four different areas of Leh district.

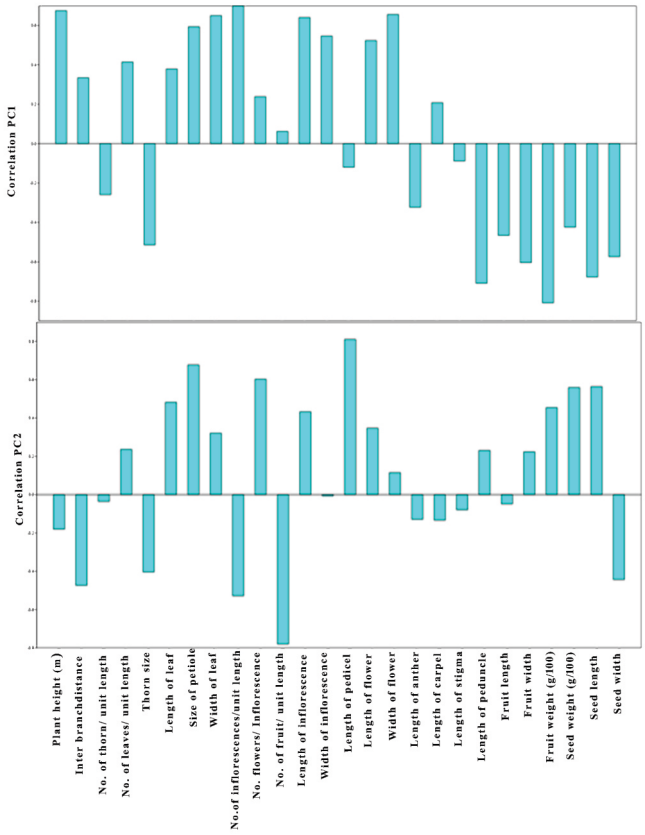


Fig. 6: Bar graph showing trait-wise positive and negative correlations in PC1 and PC2.

Cluster analysis

The dendrogram using the UPGMA method was prepared using the mean values of various morphological traits of both vegetative and reproductive nature. The resulting dendrogram separated the 16 populations into a single cluster and one outlier. The latter, represented by a single population (CHU), belongs to an altogether different species, *H. tibetana* Schltdl. (Tamchos and Kaul, 2016) and that is the reason for its segregation from the rest. The distinct taxonomic identity and morphological separation of the Chumur population make it a high-priority genetic resource for targeted conservation. Its occurrence should be documented further and monitored *in situ*, while representative germplasm and, where feasible, seeds or other suitable propagules need to be conserved *ex situ* with verified taxonomic identity and collection metadata. The remaining 15 populations again split into one major cluster and an outlier represented by (MT II), where the plants are uniformly hermaphrodite. The distinctive reproductive phenotype of Matho II (MT II) also warrants priority attention in conservation planning. Maintaining this population *in situ* and securing representative material *ex situ* would help preserve its unusual sex-expression pattern and also pave the way for future research on its reproductive biology and possible improvement. The only cluster obtained was further subdivided into two sub-clusters, A and B. Sub-cluster A is represented by six populations: one (SKB) cut off from the rest, four (NL, BSG, SNJ and SPT) assembled into one group, and one remained separate (SY II). Similarly, sub-cluster B is separated into two groups, one smaller with two (YKF and SHY I) and the second with five (PTP, TSK, LKG, CHGS, TRT) populations and one (MT-I) outlier (Fig. 7).

The heat map illustrated that 5 of the 26 quantitative traits segregate the populations. Interestingly, all are important from the commercial perspective. These are; numbers of a) leaves b) fruits c) inflorescences, and d) thorns per unit length and e) fruit weight. Of these, the number of leaves per unit length greatly affects the phenotypic diversity of seabuckthorn in the wild. It is also clearly evident from Figs. 3a and 3b.

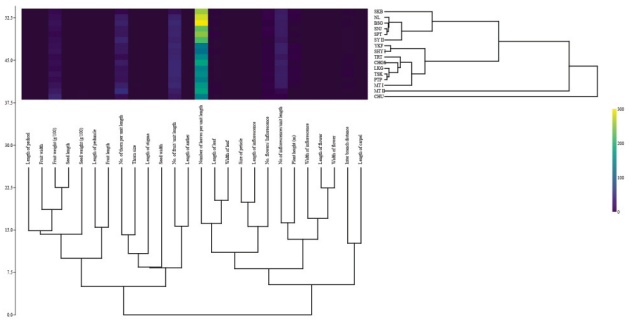


Fig. 7: Dendrogram based on morphological characters (using PAST software version 4.11) of 16 wild populations of *Hippophae rhamnoides* L. collected from Leh, Changthang, Sham and Nubra valleys.

Conclusion

The present investigation consolidates the information on the quantitative aspects of wild germplasm of Seabuckthorn in only one district of Ladakh, i.e., Leh. Based on 122 individuals of 16 different populations located in diverse habitats of varying topographic conditions such as Leh, Sham, Changthang and Nubra valleys, the present study provides the baseline data for subsequent improvement of the species. Each individual was critically examined for 26 quantitative traits of vegetative and reproductive nature, and a welter of variations was found in all of them across populations. On account of its medicinal, ecological, economic, and nutritional values, various traits like plant height, number of thorns, leaves, inflorescences and fruits/unit length, number of flowers/inflorescence and fruit weight can be used for selection-based

improvement of seabuckthorn. The higher value of diversity indices indicates that enormous variations have been maintained over years among varied ecological conditions of the vast geographical range it occupies. Broad-sense heritability values are a pointer towards these; the higher values indicate the major role of genetic factors in controlling the expression. The better genotypes can also be used for conservation and utilization of this wonder plant for various purposes.

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References

Abassi AA and V Kaul (2015) tSermang; a priceless gift to lackadaisical Ladakhis. *Curr. Sci.* 108(2): 163-164.

Acharya S, T Stobdan and SB Singh (2010) Seabuckthorn (*Hippophae* sp. L.); new crop opportunity for biodiversity conservation in cold arid Trans-Himalayas. *J. Soil Water Conserv.* 9(3): 201-204.

Ahsan MZ, MS Majidano, H Bhutto, AW Soomro, FZ Panhwar, AR Channa and KB Sial (2015) Genetic variability, coefficient of variance, heritability and genetic advance of some *Gossypium hirsutum* L. accessions. *J. Agric. Sci.* 7(2): 147-151.

Ali A (2013) Assessment of morphological and cytological variability in seabuckthorn (*Hippophae rhamnoides* L.) from some areas of Ladakh. PhD thesis, University of Jammu, Jammu, India.

Ali A and V Kaul (2017) Incidence of apomixis in sea buckthorn (*Hippophae rhamnoides* L.), a dioecious taxon of immense economic importance. *Curr. Sci.* 112(10): 1994-1996.

Allard RW (2010) *Principles of Plant Breeding*, 2nd edn. Wiley India Pvt. Ltd.

Asma BM and K Ozturk (2005) Analysis of morphological, pomological and yield characteristics of some apricot germplasm in Turkey. *Genet. Resour. Crop Evol.* 52: 305-313.

Bartish IV, N Jeppsson, GI Bartish, R Lu and H Nybom (2000) Inter- and intraspecific genetic variation in *Hippophae* (Elaeagnaceae) investigated by RAPD markers. *Plant Syst. Evol.* 225: 85-101.

Burton GW (1952) Quantitative inheritance in grasses. *Proceedings of the 6th International Grassland Congress*, Vol. 1, pp 227-283.

Cheikh-Affene ZB, F Haouala and F Harzallah-Skhiri (2015) Morphometric variation and taxonomic identification of thirteen wild rose populations from Tunisia. *Acta Bot. Croat.* 74(1): 1-17. https://doi.org/10.1515/botcro-2015-0008

Chunfang Z, C Guojuan, W Yuhua, K Helena and L Chunyang (2007) Genetic variation of *Hippophae rhamnoides* population at different altitudes in the Wolong Nature Reserve based on RAPDs. *Chin. J. Appl. Environ. Biol.* 13(6): 753-758.

Clement RA (1991) In: Grierson AJC and Long DG (eds) *Flora of Bhutan, Including a Record of Plants from Sikkim*, Vol. 2, Part 1. Royal Botanic Garden Edinburgh, Edinburgh, pp 213-216.

Dolkar P (2018) Gender differences in antioxidant properties, phenotypic plasticity and freeze tolerance in seabuckthorn (*Hippophae rhamnoides* L.) along an altitudinal gradient in Trans-Himalayan Ladakh, India. PhD thesis, Jaypee University of Information Technology, Solan, India.

Draghi J (2019) Phenotypic variability can promote the evolution of adaptive plasticity by reducing the stringency of natural selection. *J. Evol. Biol.* 32: 1274-1289. https://doi.org/10.1111/jeb.13527

Dwivedi SK, R Singh and Z Ahmad (2006) *The Seabuckthorn*. Field Research Ladakh, DRDO, Leh, Ladakh.

Godbharle AR, AW More and SS Ambekar (2010) Genetic variability and correlation studies in elite 'B' and 'R' lines in kharif *sorghum*. *Electron. J. Plant Breed.* 1: 98-993.

Hyvönen J (1996) On phylogeny of *Hippophae* (Elaeagnaceae). *Nord. J. Bot.* 16: 51-62.

Kaul V and S Tamchos (2021) Ethnobotany and tribals of Ladakh-need for scientific initiatives and interventions. In: B Singh and YP Sharma (eds) *Plants for Novel Drug Molecules: Ethnobotany to Ethnopharmacology*, pp 303-330. New India Publishing Agency, India.

Korekar G (2013) Genetic diversity of seabuckthorn (*Hippophae rhamnoides* L.) in trans-Himalayan Ladakh region, India. PhD thesis, Jaypee University of Information Technology, Solan, India.

Li TSC and J Beveridge (2003) *Sea Buckthorn* (*Hippophae rhamnoides* L.); *Production and Utilizatio*n. NRC Research Press. https://doi.org/10.1139/9780660190075

Lush JL (1940) Intra-sire correlation or regression of offspring on dam as a method of estimating heritability of characteristics. *33rd Annual Proceedings of the American Society of Animal Production*, pp 293-301.

Mangla Y, M Chaudhary, H Gupta, R Thakur, S Goel, SN Raina and R Tandon (2015) Facultative apomixis and development of fruit in a deciduous shrub with medicinal and nutritional uses. *AoB Plants* 7: plv098. https://doi.org/10.1093/aobpla/plv098

Mathew D, T Parimelazhagan, S Gomez and Z Ahmed (2007) Characterization of sea buckthorn (*Hippophae* spp.) genetic resources in India using morphological descriptors. *Biodivers. Int. Newsl. Portal* 149: 22-26.

Median FP and HR Chaudhary (1980) Variability parameters in *sorghum*. *Agron. Abstr.*, 62 p. American Society of Agronomy, Madison, Wisconsin, USA.

Naithani HB (2004) *Hippophae* L. (seabuckthorn) in India; a review. *Indian For.* 130: 1045-1056.

Rana R and P Shirkot (2012) Relationships between morphological descriptors and RAPD markers for assessing genetic variability in *Hippophae rhamnoides* L. *For. Stud. China* 14(2): 137-144.

Raina SN, S Jain, D Sehgal, A Kumar, TH Dar, V Bhat, V Panday, S Vaishnavi, A Bhargav, V Singh, V Rani, R Tandon, M Tewari and A Mahmoudi (2012) Diversity and relationships of multipurpose seabuckthorn (*Hippophae* L.) germplasm from Indian Himalayas as assessed by AFLP and SAMPL markers. *Genet. Resour. Crop Evol.* 59: 1033-1053.

Reim S, A Proft, S Heinz and M Hofer (2012) Diversity of the

European indigenous wild apple *Malus sylvestris* (L.) Mill. in the East Ore Mountains (Osterzgebirge), Germany: I. Morphological characterization. *Genet. Resour. Crop Evol.* 59: 1101-1114.

Rousi A (1971) The genus *Hippophae* L.; a taxonomic study. *Ann. Bot. Fenn.* 8: 177-227.

Ruan CJ (2006) Genetic relationship among some seabuckthorn cultivars from China, Russia and Mongolia using AFLP markers. *J. Hortic. Sci. Biotechnol.* 81: 409-414.

Ruiz D and J Egea (2008) Phenotypic diversity and relationships of fruit quality traits in apricot (*Prunus armeniaca* L.) germplasm. *Euphytica* 163: 143-158.

Saroop S and V Kaul (2020) Phenotypic variability in *Cleome viscosa* L. growing in Jammu region (J&K); some interesting insights in this prospective crop. *Genet. Resour. Crop Evol.* 67: 59-71.

Sen R, S Tamchos and V Kaul (2014) Sexual variants in a dioecious taxon, *Hippophae rhamnoides* L. *Int. J. Plant Reprod. Biol.* 6(1): 41-43.

Singh K, YP Sharma and S Gairola (2020) Morphological characterization of wild *Rosa* L. germplasm from the Western Himalaya, India. *Euphytica* 216(3): 41.

Singh L and V Kaul (2014) Quantification of phenotypic diversity in *Allium sativum* L. over a period of three years. *Int. J. Plant Reprod. Biol.* 6(1): 35-39.

Singh L, V Kaul and RN Gohil (2014) Analysis of morphological variability in the Indian germplasm of *Allium sativum* L. *Plant Syst. Evol.* 300: 245-254.

Singh R, SN Mishra, SK Dwivedi and Z Ahmed (2006) Genetic variation in seabuckthorn (*Hippophae rhamnoides* L.) populations of cold arid Ladakh (India) using RAPD markers. *Curr. Sci.* 91(10): 1321-1322.

Singh TH, S Pandey, CD Pandey and SR Yerasu (2026) morphological diversity analysis in brinjal (*Solanum melongena*) germplasm. *Plant Genet. Resour.* 39(2): 74-82.

Srihari JM, B Verma, N Kumar, RK Chahota, V Singh, R Rathour, SK Singh, SK Sharma and TR Sharma (2013) Analysis of molecular genetic diversity and population structure in sea buckthorn (*Hippophae* spp. L.) from north-western Himalayan region of India. *J. Med. Plants Res.* 7(43): 3183-3196. https://doi.org/10.5897/JMPR12.1112

Stobdan T, A Yadav, GP Mishra, OP Chaurasia and RB Srivastava (2011) Seabuckthorn: The Super Plant (Production, Characterization, Postharvest & Health Applications). Defence Institute of High-Altitude Research, Defence Research and Development Organisation, Leh, Ladakh.

Tamchos S (2018) Studies on morpho-taxonomic evaluation of seabuckthorn (*Hippophae* L.) growing in Leh and Nubra Valleys of Ladakh. PhD thesis, University of Jammu, Jammu, India.

Tamchos S and V Kaul (2016) Distributional status of *Hippophae tibetana*; a new report from Changthang Valley of Ladakh. *Natl. Acad. Sci. Lett.* 39(1): 59-62.

Tamchos S and V Kaul (2019) Seabuckthorn; opportunities and challenges in Ladakh. *Natl. Acad. Sci. Lett.* 42(2): 175-178.

Tashi T and FK Sudan (2022) Economic opportunities brought by sea buckthorn in the cold desert area of Nubra Valley, Ladakh. *Contrib. Indian Sociol. Sci.* 41(1–2): 184-195.

Truță E, G Căpraru, S Surdu, MM Zamfirache, Z Olteanu, CM Roșu and L Oprică (2010) Karyotypic studies in ecotypes of *Hippophae rhamnoides* L. from Romania. *Silvae Genet.* 59(4): 175-182.

Upadhyaya HD (2003) Geographical patterns of variation for morphological and agronomic characteristics in the chickpea germplasm collection. *Euphytica* 132(3): 343-352.

Utioh A, J Meseyton, N d'Avila and H Wang (2007) Seabuckthorn (*Hippophae rhamnoides* L.) products overview. *Proceedings of the 3rd International Seabuckthorn Association Conference*, Institute of Nutraceuticals and Functional Foods, Laval University, Canada, pp 177-184.