Incidence and frequency of desynapsis in *Eremurus persicus* (Jaub. & Spach) Boiss. (Asphodelaceae) – A native and important medicinal plant species of Western Himalaya

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Abstract. *Eremurus persicus* (Jaub. & Spach) Boiss. (Asphodelaceae); a little known species of genus *Eremurus* grows in arid and semi-arid regions, on rocky mountains in Central Asia and Middle East. The species is native and an important medicinal plant used to treat various diseases. In the current study, we examined male meiosis, karyotypic variability, pollen viability and reproductive output of three populations of the target species. All the studied populations have diploid chromosome count of 2n = 2x =14. Meiotic course in majority of the pollen mother cells (PMCs) in all the three populations is not normal due to the presence of desynapsis at diplotene and metaphase-I. Instead of 7IIs most of the PMCs examined reveal varying number of bivalents and univalents. Anaphasic segregation is also affected in few PMCs. Present study also revealed that the species has asymmetric karyotype of 10 long and 4 short chromosomes. Species exhibits fruit abortion that results in extremely low fruit/seed set. The meiotic abnormalities and low fruit/seed set may be attributed to restricted distribution of this important plant species. Present study may prove helpful in devising the conservation and management strategies for this prized plant species.

Keywords: desynapsis, native, diplotene, metaphase, karyotype, fruit abortion, conservation.

INTRODUCTION

Pairing of chromosomes is the first step that leads to the organized segregation of chromosomes during meiosis and is the basis of continuity of life (Fabig *et al.*, 2020). In addition being the first step of reduction division; this is also important as it leads to crossing over and reshuffling of alleles and contributes to the generation of genetic variations (Cai & Xu, 2007). Though it is religiously followed in stable diploid species during zygotene, instances where the deviation from norms occurs are not rare. Chromosomes in some

taxa fail to pair (asynapsis) (Jakson *et al.,* 2002; Ansari *et al.,* 2022) or their pairing breaks precociously (desynapsis) (Golubovskaya,1979; Wani & Bhat, 2017). These anomalies arerecognized when most or all the chromosomes remain as univalents at diakinesis and metaphase-I (Visser *et al.,*1999). Although theoretically the phenomenon of asynapsis & desynapsis are distinct from each other, it is difficult to distinguish them in practice. This is true for those plants where chromosomes are long and pairing cannot be followed during earlier stages (Koul, 1962; Wani & Bhat, 2017).

As a characteristic of a particular variety or ecotype, desynapsis has been reported in several plant genera viz *Zea* (Beadle, 1933); *Nicotiana* (Clausen, 1931); *Datura* (Bergner *et al.*, 1934); *Gossypium* (Beasely and Brown, 1942); *Triticum* (Li *et al.,*1945), *Alopecurus* (Jhonson, 1944) etc. But as an inherent feature of a taxa as a whole; it is a rare occurrence. Here we report this phenomenon in *Eremurus persicus* (Jaub. & Spach) Boiss. Described as a feature of meiosis in few plants of this species by Verma (2001) as well, it was found to be a regular feature of this plant species across all the plants in all populations scanned in present study.

Genus *Eremurus* M. Bieb. is commonly known as Foxtail lilies/ Desert candles of family Asphodelaceae comprises of more than 60 species that are native to Central Asia and Caucasia (Kamenetsky & Akhmetova, 1994). The species of the genus can be easily recognized by their long scapes bearing numerous colourful flowers along with rosette of leaves. Most of the species of this genus are important commercially as ornamental plants for landscaping and cut flower markets (Schiappacasse *et al.*, 2013). In addition to their ornamental value, *Eremurus* sp. have been used in traditional medicine and are potential sources for anti-inflammatory, anti-bacterial and anti protozoal drugs (Hashemi *et al.*, 2014 ; Mati *et al.*, 2011). Also bio-oil and adhesives prepared from this genus play an important role in industrial applications (Vala *et al.*, 2011). In India Genus *Eremurus* is represented by only two species *viz Eremurus himalaicus* Baker and *Eremurus persicus* (Jaub. & Spach) Boiss.

Eremurus persicus is a perennial herb which perennates through underground fleshy tuberous rootstocks that sprout every year in the month of February- March in the sub-tropical areas of Jammu, UT of J&K, India. The above ground portion of the plant appears in the form of rosettes of leaves which are light to dark green in colour, radical, thick and lanceolate. The inflorescence is raceme which bears flowers in an acropetal manner. Flowers are creamish to light pink in colour, pedicellate, bracteate, bisexual and glabrous. Species holds immense value for its medicinal attributes in addition to being used as a vegetable for its edible leaves. The species is widely distributed in South, East and West of Iran where it is locally known as Sarish (Karl, 1982; Safar *et al.*, 2009; Vala *et al.*, 2011). It is also reported from Kibri Kuch Ziarat of Pakistan (Khan *et al.*, 2011).

In India there are scanty reports of the distribution of the species, however; the species is growing in hilly terrains of Jammu province (Hamal & Karihaloo, 1983). Extensive field surveys, perusal of relevant literature and herbarium studies revealed that the plant species has a restricted distribution in the study area. Only few populations (7 populations) of the species have been located from district Reasi of J&K. Keeping in view the rare distribution and medicinal importance of the target plant species the present study was carried out to provide detailed meiotic, mitotic account, pollen viability and fruit/seed set of *E. persicus*. The knowledge generated in present study may prove helpful in devising conservation strategies for this prized plant species.

MATERIAL AND METHODS

A total of 7 populations of *E. persicus* were located during present investigation, which include: Slal, Kharjala, Slal Kotli, Sarmega I, Sarmega II, Ponsli I and Ponsli II (Figure 1 and 2). Of these three populations *viz.* Slal (Population-I), Sarmega I (Population-II) and Ponsli I (Population-III) were selected for detailed meiotic, mitotic investigation, pollen viability and seed set in the present study. Materials for male meiotic studies were collected from wild plants growing on rocky slopes of hilly areas of Trikuta hills of Reasi. Young unopened flower buds of suitable sizes were collected randomly from different plants of each population during the peak flowering period, i.e., March-April, when minimum and maximum temperature of this area averages 13-25°C. The floral buds were fixed in Carnoy's fixative for 24 h. Subsequently, the materials were transferred in 70% ethanol and stored under refrigerator at about 4°C until analysis. For meiotic preparations, smears of the fixed anthers were made in 1% propiocarmine. Photomicrographs of Pollen Mother Cells (PMCs) for chromosomal counts at different stages and meiotic irregularities were made from the freshly prepared slides using EVOS XL microscope. Chiasmata number was counted for cells at diplotene. Data was used to calculate Recombination Index using the following formulae

RI= Chiasmata frequency per cell + haploid chromosome number of the species

Figure 1. *Eremurus persicus*(a) Habitat, (b&c) A mature flowering plant, (d) Ripen globular fruits.

For Karyological studies, healthy root tips from seedlings were collected and washed with distilled water. These were pre-treated with 0.3% colchicines solution for 4 hours and fixed in acetic acid: ethanol (1:3) for 24 hours. Thereafter, the root tips were transferred to 70% ethanol for further use. For mitotic chromosomal preparations, preserved root tips were washed with water and hydrolyzed in 1 drop of 1N HCL and 9 drops of 1% aceto-orcein at 60°C for 12-15 min. Thereafter these root tips were squashed in 1% acetocarmine. Karyotype formula was determined according to Levan *et al.*, 1964. Karyotypes were compared using Stebbins classification, karyotype asymmetry index (As.K %) (Arano, 1963), total form percent (TF %) (Huziwara,1962), Rec and Syi indices (Venora *et al.*, 2002), intrachromosomal symmetry index (A1) and interchromosomal asymmetry index (A2) (Romeo-Zarco, 1986), dispersion index (DI) (Lavania & Srivastava, 1999), degree of asymmetry of karyotype (A index) (Watanabe *et al.*,1999), asymmetry index (AI) (Paszko, 2006), the coefficient of variation of chromosome length (CV_{CL}) and the coefficient of variation of centromeric index (CV_{CI}) (Paszko, 2006) and mean centromeric asymmetry (M_{CA}) (Peruzzi *et al.*, 2009). The strength of the association between karyotype asymmetry indices was tested using Pearson correlation analysis using PAST software.

To calculate number of pollen grains per anther and/ flower, 30 flowers ready to open with intact anthers were selected. Pollen quantity was estimated by squashing one anther (several times) in 10 drops of distilled water in a cavity block and shaken with a glass rod.

The following equations were used to calculate the number of pollen per flower:

$$r = p \times q \text{ and } t = r \times s,$$

where p is the mean pollen count per drop of water; q is the number of water drops taken initially in which one anther was squashed; r is the mean number of pollen per
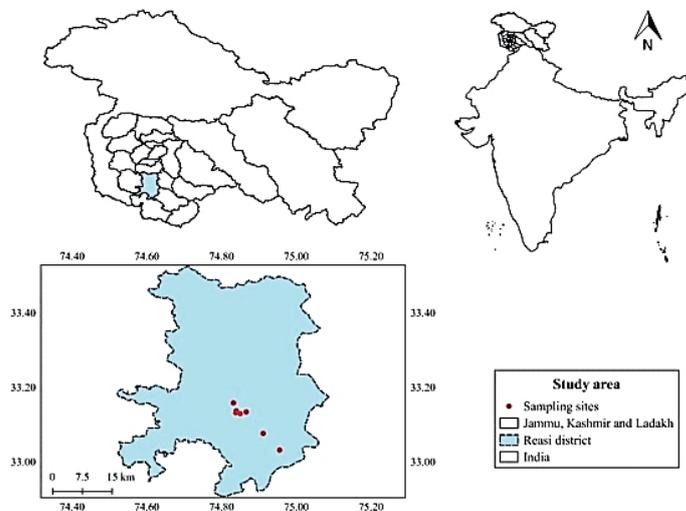
Figure 2. Map showing sampling sites of *E. persicus*.

anther; *s* is the mean number of anthers per flower; *t* is the total count per flower (Ganie *et al.,* 2021).

For calculation of pollen count per flower the average pollen number in one anther was multiplied by total number of anther per flower

Average ovule number per pistil was counted using dissection microscope (Ganie *et al.,* 2021).

Pollen viability was determined by subjecting the pollen grains to 1% acetocarmine, TTC and FDA. In first method, mature anthers were squashed in 1% acetocarime and kept for 10 minutes and then slides were scanned under microscope, well stained and plump pollen grains were considered viable while as lightly stained and shriveled pollen grains were considered as non-viable. In second method, the ready-to-dehisce mature anthers were placed in 1% tetrazolium chloride for one hour and squashed to check for viability (Rashid *et al.* 2023) and in third method pollen grains were mounted in Fluorescein diacetate (FDA) solution and incubated for 3-5 min. The pollens with fluorescent and non-fluorescent cytoplasm were treated as viable and nonviable, respectively (Rashid *et al.* 2023). In all the three methods, percentage of pollen viability was determined by following formula:

$$\text{Pollen viability} = \frac{\text{Number of pollengrains stained}}{\substack{\text{Total number of} \\ \text{observed pollen grains}}} \times 100$$

To check the reproductive efficiency on open pollination, 20 different inflorescences were tagged and kept for pollination to take place as it does in nature. These were monitored regularly from the time of flower opening to that of fruit maturation.These flowers were observed after some days to check the percentage fruit set. Percentage fruit set on unassisted selfing was performed by bagging large number of buds from 10 different inflorescences in transparent butter paper bags. Total number of fruits as an estimate of female success was calculated. Mature fruits were harvested shortly before dehiscence and scored for the presence of seeds. Percentage fruit set and seed set was then calculated by using Bharti *et al.,* 2021 method.

RESULT AND DISCUSSION

PMC meiosis

All the three populations of *E. persicus* matched in having 14 chromosomes in their PMCs revealing 2n= 2x = 14. PMCs were analysed at diplotene, diakinesis, metaphase-I and anaphase-I. Desynapsis was observed as a regular feature in majority of the cells at diplotene and all the cells at Metaphase-I. Meiotic behavior of three populations differed slightly and the meiotic behavior of the studied populations is given as under:

Population-I (P-I)

In P-1, a total of 97 cells were scanned at different stages of meiosis with 26 cells (26.80%) at diplotene, 15 cells (15.46%) at diakinesis, 39 cells (40.20%) at metaphase-I and 17 cells (17.52%) at anaphase-I (Figure 3 a-e). Out of 26 cells at diplotene only 2 cells were having perfect 7IIs (Figure 3 a). Of observed bivalents, 5 bivalents/chromosomes were large and 2 were small. Interestingly in these bivalents, chromosomes were mostly held by terminal chiasmata only. At metaphase-I, cells were observed with different configurations and with varying number of univalent and bivalents in all PMCs. It has also been observed that 38.46% of cells showed all the 14 chromosomes as univalents (Figure 3 b) while 25.64% of cells showed the presence of 2IIs along with 10Is. Other configurations like 1,3,4 & 5 IIs were also observed. The univalents at this stage are arranged close to each other. At anaphase-I, PMCs usually showed normal segregation of 7:7 chromosomes at each pole (Figure 3 c-e). Chiasmata frequency per PMC calculated at diplotene in this population is 9.15 while RI calculated is 16.5.
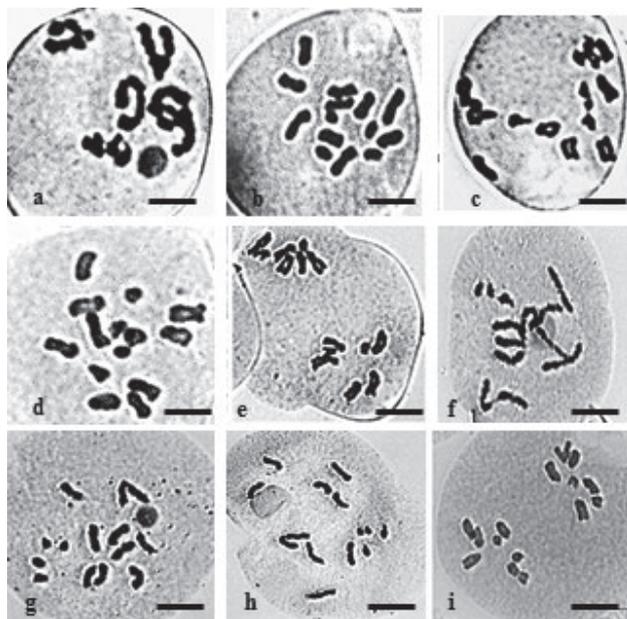
Figure 3. Population I (a-e): (a)A PMC at diakinesis with 7IIs, (b) A PMC at metaphase-I with univalents, (c&d) PMCs at early anaphase stage, (e) A PMC at anaphase-I. **Population II (f-i):** (f) A PMC at diplotene showing terminal chiasma, (g&h) PMCs at metaphase-I with 14 univalents, (i) A PMC at anaphase-I showing normal segregation of 7:7 chromosomes.

Population-II (P-II)

In P-II, 91cells were observed, out of which 33 cells (36.26%) were at diplotene, 37 cells (40.65%) were at metaphase-I and 21cells (23.07%) were at anaphase-I (Figure 3 f-i). In this population, formation of 7IIs was lacking even at the diplotene stage. At this stage only few II's were held by terminal chiasmata. At metaphase-I varied number of IIs and their configurations were observed, in addition varied number of Is were also observed in all the scanned PMCs. At this stage also chromosomes in IIs were associated terminally only. In 51.35% of cells all the 14 chromosomes as Is were observed (Figure 3 g-h) followed by 3,2&1 IIs along with Is. However, at anaphase-I usually normal segregation of 7:7 chromosomes was observed (Figure 3 i). Chiasmata frequency per PMC calculated at diplotene in this population is 6.9 and RI thus comes out to be 13.9.

Population-III (P-III)

In P-III, a total of 318 cells were observed at different stages, 45 cells (14.15%) at diplotene (Figure 4 a, b), 26 cells (8.17%) at diakinesis (Figure 4 c), 91 cells (28.61%) at metaphase-I, 79 cells (24.84%) at anaphase-I, 21 cells (6.60%) at telophase-I, 23 cells (7.23%) at metaphase-II and 33 cells (10.37%) at anaphase-II. In this population maximum frequency of PMCs with Is was observed (Figure 4 d), i.e., 61.53% followed by 2,3 & 5 II's in association with I's respectively. In present study 7.54% of cells scanned at anaphase-I revealed normal segregation of chromosomes with 7:7 distribution at each pole (Figure 4 e). Erratic distribution of 7:6, 6:6 and 6:5 chromosomes at each pole was observed in 7 cells (2.20%), 8 cells (2.51%) and 13 cells (4.08%) respectively. Also 27 cells (8.49%) showed presence of laggards and chromosomal bridges along with clumping of chromosomes (Figure 4 f-m). At anaphase-I and anaphase-II the formation of laggards and bridges was observed as a common feature. Chromosomal bridges were observed at metaphase-II also. All the cells scanned at metaphase-II and anaphase-II showed the presence of laggards and chromosomal bridges (Figure 5 a-d). Therefore the second meiotic division exhibited drastic irregularities in this population. Chiasmata frequency per PMC calculated at diplotene in this population is 5.3 and RI is 12.3.

Detailed studies on meiotic system revealed that in the species, majority of the PMCs at diplotene, diakinesis and metaphase consist of univalents ranging from 2-14 in all the three studied populations. At diplotene loosely bound IIs are seen mostly with terminal and
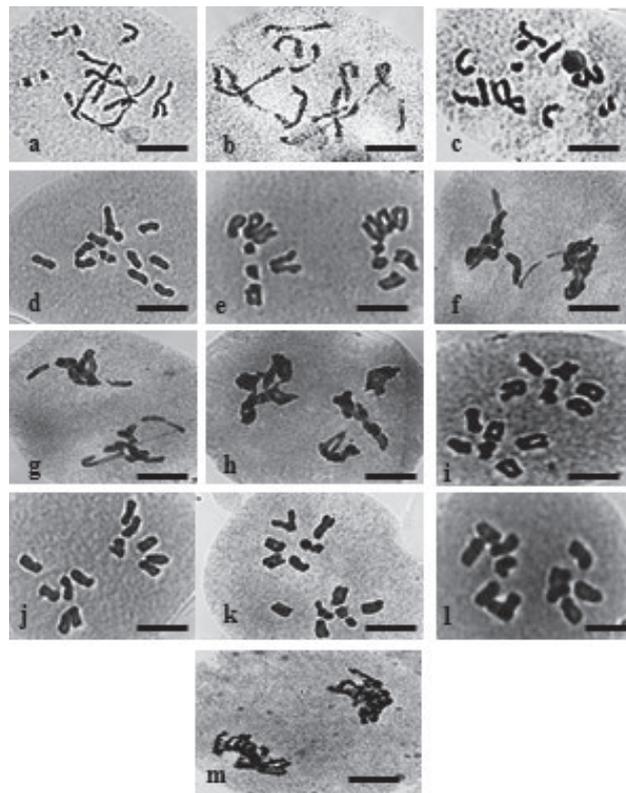
Figure 4. Population III (a-m): (a, b) PMCs at diplotene,(c) PMC at diakinesis(d) PMC at metaphase-I showing univalents,(e) PMC at anaphase-I showing normal distribution of 7:7 chromosomes,(f-h) PMCs at anaphase-I showing laggards and bridges along with clumping of chromosomes, (i) A PMC at anaphase-I showing 7:6 distribution, (j) A PMC with 6:6 distribution at anaphase-I,(k) A PMC showing 6:5 distribution with laggard, (l) A PMC with 6:5 distribution of chromosomes at anaphase-I, (m) A PMC at anaphase-I showing clumping.

rarely with intercalary chiasmata. As meiosis proceeds, intercalary chiasmata are not seen in any of the bivalent and the frequency of univalents goes on increasing. At later stage few bivalents and more frequency of univalents are observed. Subsequently the chromosomes fall apart and remain as univalents during rest of the cell cycle till metaphase-I. In most of the PMCs univalents were found to lie in pairs (distant pairing) were also observed in the present study. The frequency of univalent and bivalents observed in three populations are depicted in Figure 6.

SOMATIC CHROMOSOME COMPLIMENT

All the three populations studied show chromosomes in their somatic compliment confirming 2n=
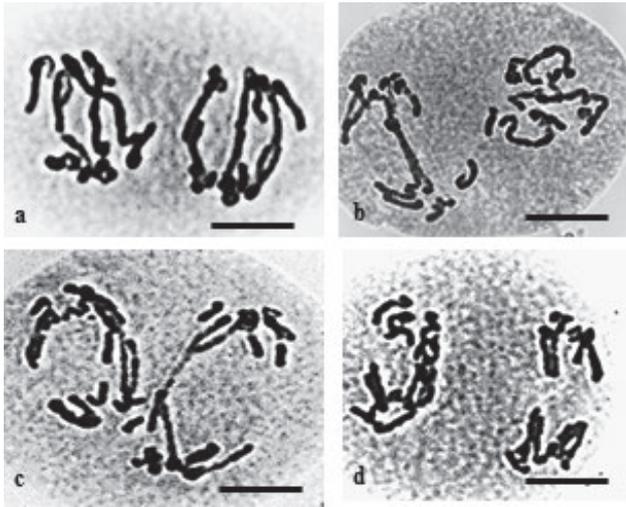
Figure 5. Population III (a-d): (a) A PMC at telophase-I, (b) APMC at metaphase –II with chromosomal bridges, (c, d) PMCsat anaphase-II with laggards and bridges.
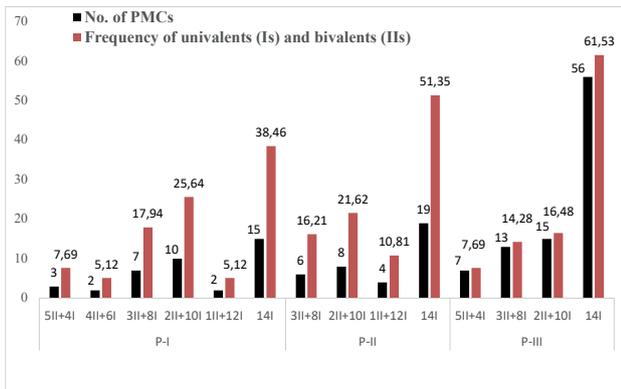
Figure 6. Graphical representation of configurations of univalents and bivalents.

Figure 7. Somatic metaphasic spread of P-I, P-II and P-III showing 14 chromosomes and the karyogram of the same. (Scale = 10μm).

μm) possessed the lowest value for the same. The highest and lowest values for the smallest chromosome occurred in P-III (4.4μm) and P-II (4.95μm) respectively (Table 1). The most asymmetrical karyotype was observed in P-III (Table 2)

P-III possessed the highest value for CV showing the highest variation among its chromosomes compared to the other populations.Pearson coefficient of correlation determined for karyotypic parameters among the populations studied revealed a high correlation significant at p≤0.05 (Table 3). The AI value and scatter diagram based on the CV_{CL} and CV_{CI} seem best suited to assess overall classification strength and display relationships among *E.persicus* populations with respect to karyotype asymmetry (Table 4; Figure 8 and 9).

Cluster analysis of the populations of *E. persicus* based on relative karyotypic data revealed that P-I and P-II are placed close to each other while P-III joined them with a distance (Figure 10). However, there is morphological similarity between them but due to karyotypic difference it stands separate from the rest of the two populations. This is also in accordance with their meiotic studies where P-III showed high variation in comparison to P-I and P-II.

2x = 14 as the diploid chromosome number of the species. These fall into two size groups. Group I includes 10 long chromosomes, all of them being subterminal while group II includes 4 small chromosomes which are submedian and subterminal. The total chromatin length of the somatic compliment for population I, II and III is 160.2, 158.4 and 168.3 μm. The kayotype formula for the three populations is 10ST+2ST+2SM, 10ST+3SM+1ST and 10ST+1ST+3SM respectively (Figure 7). These populations thus varied slightly in their karyotypic formulae too, although the chromosomes were of subtelocentric and submetacentric type only. These fallin 3C category of the Stebbin's (1971) chart of asymmetry. Among all the populations studied, P-III possessed the highest value (17.6 μm) of longest chromosome while P-I (15.54

Table 1. Karyomorphometric analysis of somatic metaphasic chromosomes complement of three populations of *E. persicus*.

| | Populations | | | | | | | | | | | | | | |
| | P-I | | | | | P-II | | | | | P-III | | | | |
S. No.	SA (µm)	LA (µm)	TL (µm)	AR	Type	SA (µm)	LA (µm)	TL (µm)	AR	Type	SA (µm)	LA (µm)	TL (µm)	AR	Type
1	3.33	12.21	15.54	3.66	ST	3.85	12.1	15.95	2.49	ST	3.3	14.3	17.6	2.88	ST
2	3.88	10.54	14.42	2.71	ST	3.85	11.55	15.4	2.75	ST	3.3	13.75	17.05	4.16	ST
3	3.33	10.54	13.87	3.16	ST	3.85	11	14.85	2.87	ST	3.85	13.2	17.05	3.42	ST
4	2.77	10.54	13.31	3.80	ST	3.3	11	14.3	3.14	ST	3.3	12.65	15.95	3.83	ST
5	3.33	11.65	14.98	3.49	ST	3.3	11	14.3	2.62	ST	2.75	11.55	14.3	4.2	ST
6	2.77	11.65	14.42	4.20	ST	2.75	12.1	14.85	2.86	ST	1.65	11	12.65	6.66	ST
7	3.33	9.99	13.32	3	ST	3.3	9.35	12.65	4.00	ST	2.2	11	13.2	5	ST
8	3.33	9.43	12.76	2.83	ST	2.75	9.9	12.65	1.71	ST	2.2	11	13.2	5	ST
9	2.22	8.88	11.1	4	ST	2.2	7.7	9.9	1.85	ST	2.2	12.1	14.3	5.5	ST
10	2.77	8.32	11.09	3.00	ST	2.75	8.8	11.55	3.24	ST	2.2	6.05	8.25	1.13	ST
11	1.65	6.05	7.7	3.66	ST	2.2	3.85	6.05	3.5	SM	2.2	6.05	8.25	2.75	ST
12	1.1	6.05	7.15	5.5	ST	1.65	3.3	4.95	3	SM	2.2	3.3	5.5	1.5	SM
13	2.22	3.33	5.55	1.5	SM	1.65	3.3	4.95	3.67	SM	1.65	2.75	4.4	1.66	SM
14	1.66	3.33	4.99	2.00	SM	1.65	4.4	6.05	1.40	ST	1.65	3.85	5.5	2.33	SM
Total	37.69	122.51	160.2			39.05	119.35	158.4			34.65	132.55	168.3		

Table 2. Karyotype formula according to Levan et al. 1964 and characteristics of the studied species SC –the shortest chromosome length; LC – the longest chromosome length; p – mean length of long arm; q – mean of short arm; CL – mean of chromosome; CI – mean centromeric index; ST-subtelocentric; SM- submetacentric; SD- standard deviation.

Population	Range SC-LC (µm)	Ratio LC/SC	p (µm) mean ± SD	q (µm) mean ± SD	CL (µm)	CI (µm)	Karyotype formula
P-I	1.1-12.21	11.1	8.75±2.85	2.69±0.78	11.44±3.49	0.24±0.06	10ST+2ST+2SM
P-II	1.65-12.1	7.33	8.52±27	2.78±0.79	11.31±3.99	0.26±0.06	10ST+3SM+1ST
P-III	1.65-14.3	8.66	9.46±3.98	2.47±0.68	11.94±4.48	0.22±0.07	10ST+1ST+3SM

According to the As. K %, TF% and Syi and Rec indices P- III is more asymmetric than P-I and P-II.

POLLEN COUNT AND VIABILITY

Pollen grains of *E. persicus* are ovate, smooth walled. These are shed at two celled stage. Highest pollen production per anther is observed for POP III ; 6619.4±381.46. Different anthers in a flower display some difference in pollen content, so that the average pollen output per flower comes out to be 38506.6±2298.49 (Table 5). In POP II, pollen production per anther is averaging 6252.9±361.03 and total pollen production per flower comes out to be 37517.4±2166.22. Pollen production per anther averages 5428.36±351.13 in POP I and total pollen production per flower comes out to be 32570.2±2106.79.

Highest pollen stainability by 1% acetocarmine was observed in population II followed by population I and III. However pollen viability by TTC and FDA averages 89.61±0.26 and 90.78±0.53 in population I, 90.53±0.17 and 92.61±0.82 in population II and 90.40±0.33 and 91.74±0.61 in population III by TTC and FDA respectively(Table 5 and Figure 11).

REPRODUCTIVE OUTPUT

All the three populations have very low reproductive output in terms of % fruit set. The plants in their natural populations (open pollination) showed 19.00±3.19, 20.21±2.45 and 18.62±3.46 % of fruit set in P-I, P-II and P-III respectively. Bagged inflorescences do not produce seeds in all the selected populations.Also

Table 3. Pearson correlations for asymmetry indices.

	As.K%	TF%	Rec	SYi	A1	A2	DI	A	AI	CVCI	CVCL	MCA
As.K%	1											
TF%	**-0.99896**											
Rec	-0.93746	**0.952374**										
SYi	**-0.99888**	**0.999999**	**0.952899**									
A1	0.945621	**-0.95949**	**-0.99971**	**-0.95997**								
A2	0.455858	-0.49602	-0.73717	-0.49752	0.720577							
DI	-0.83294	0.806804	0.588219	0.805783	-0.60765	0.112823						
A	**0.996066**	**-0.99907**	**-0.96462**	**-0.99915**	**0.970725**	0.532939	-0.78062					
AI	0.898327	-0.91745	**-0.99507**	-0.91813	**0.992377**	0.800534	-0.50514	0.933725				
CVCI	**0.992863**	**-0.99727**	**-0.97228**	**-0.9974**	**0.977664**	0.558753	-0.761	**0.999525**	0.94431			
CVCL	0.455858	-0.49602	-0.73717	-0.49752	0.720577	**1**	0.112823	0.532939	0.800534	0.558753		
MCA	**0.996066**	**-0.99907**	**-0.96462**	**-0.99915**	**0.970725**	0.532939	-0.78062	**1**	0.933725	**0.999525**	0.532939	1

Significant correlations (p<0.05) are in boldface.

Table 4. Karyotypes of *E. persicus* using different methods of evaluating karyotype asymmetry.

Population	ST class	As. K%	TF%	Rec	SYi	A1	A2	DI	A	AI	CVCI	CVCL	MCA
P-I	3C	76.47	23.47	93.28	30.74	0.97	0.30	6.36	0.52	7.5	25	30	52
P-II	3C	75.34	24.65	93	32.62	0.97	0.35	7.59	0.50	8.07	23.07	35	50
P-III	3C	78.75	20.58	83.14	26.10	0.98	0.37	6.16	0.58	11.76	31.81	37	58
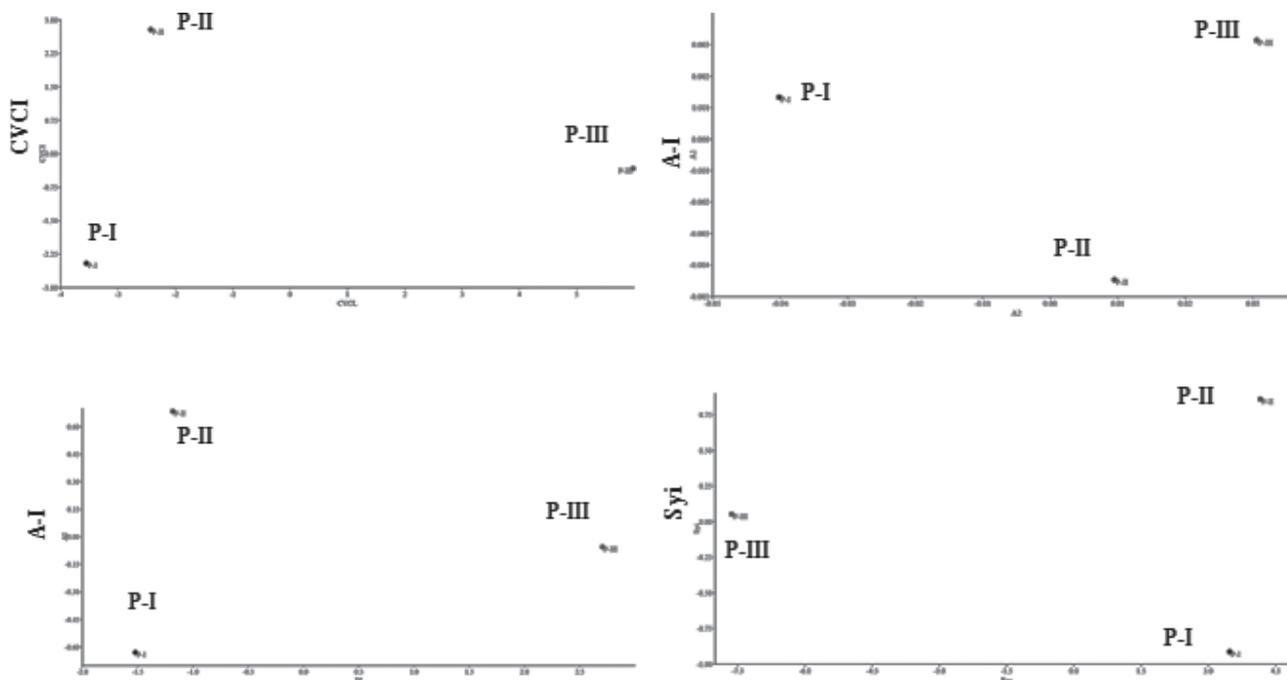
Figure 8-9. Scatter diagrams for P-I, P-II and P-III, (8) A1 against A2 & Rec index against Syi index, (9) CV$_{CL}$ againstCV$_{CI}$ & DI against AI.Degrees of asymmetry according to Stebbins.
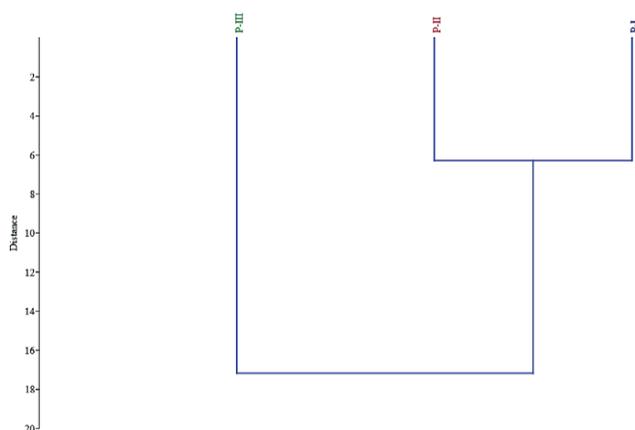
Figure 10. Dendrogram for P-I, P-II and P-III.

the percentage seed set calculated for these populations is very low i.e., 25.79±1.83, 14.88±3.20 and 17.48±3.14in P-I, P-II and P-III respectively. It was observed that fruits which start developing on a scape do not attain maturity and a large amount of the fruits abort during the course of development, in P-I (80.99±3.19%), in P-II (79.50±2.49%) and maximum fruit abortion (81.37±3.46%) was observed in P-III.

The present study revealed that *Eremurus persicus* is diploid with chromosome number of 2n = 2x = 14. The karyotype analysis revealed that various species of the genus *Eremurus* have 2n chromosome counts of 14 and 28 (Hadizadeh *et al.,* 2020). Somatic chromosome analysis of eight species of *Aloe* (Asphodelaceae) showed diploid chromosome number of 2n = 14 (Sánchez-G*et al.* 2018). The other species of the genus growing in India, i.e., *E. himalaicus* is also diploid with chromosome num-

ber of 2n = 14 (Kumari *et al.,* 2016). In present study meiotic abnormalities were found in the target species, similarly meiotic abnormalities were also found in *E, himalaicus* growing in Indian Himalayan region (Kumari *et al.,* 2016). The abnormalities are attributed to pervading environmental conditions in the Himalaya (Wani *et al.* 2023). The phenomenon of desynapsis was also found in the PMCs of presently studied plant species, both extrinsic conditions such as temperature and fertilizer quality (Dhesi *et al.*, 1975; Rao, 1975) and intrinsic conditions like gene action, loss of chromosome pair, apomixis and structural or numeral changes of chromosomes (Praaken, 1943) are responsible for the desynaptic behavior of plants. Over the years desynapsis has been established as a gene mediated phenomenon in a large number of plant species (Gottschalk & Baquar, 1971), however; the precise mode of action of these genes is not fully understood. Of the three types viz weak, medium and strong or complete desynapsis (Prakken, 1943); in present study medium desynaptic type was found in the target species because of the presence of many univalents and few loose bivalents. In spite of the lack of pairing and the formation of univalents as a consequence of desynapsis, anaphase segregation is normal in majority of cells which needs further investigation. The P-III displays irregular distribution of chromosomes with maximum frequency. Other meiotic irregularities include formation of laggards and bridges at anaphase-I, metaphase-II and anaphase-II. An interesting observation made in the present study revealed the presence of univalents of a pair close to each other, as also the regular anaphasic segregation point towards the species showing "distance pairing" in which the homologs lie

Table 5. Data on Pollen count and pollen viability.

S. No.	Characters (n=30)	POP-I	POP-II	POP-III	p-value
1.	No. of pollen grains per anther	5428.36±351.13* (2896-8983)**	6252.9±361.03 (3202-9883)	6619.4±381.46 (3268-9962)	0.07
2.	No. of pollen grains per flower	32570.2±2106.79 (17376-53898)	37517.4±2166.22 (19212-59298)	38506.6±2298.49 (19608-59772)	0.13
3.	No. of ovules per flower	18.33±0.41 (17-22)	20.13±0.30 (17-22)	20.3±0.33 (17-22)	0.0002***
4.	Pollen viability **a.** (1% acetocarmine)	93.18±0.22* (90-95)**	95.25±0.34 (92-98.78)	91.74±0.27 (89-94)	0.001**
	b. TTC	90.40±0.33 (88-94)	90.53±0.17 (89-93)	89.61±0.26 (88-92.56)	0.04
5.	**c.** FDA	91.74±0.61 (83.6-95)	92.61±0.82 (82-98.7)	90.78±0.53 (82.01-94)	0.16

*Mean± standard error.

**Range.

*Significant at p<0.05 (Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1).
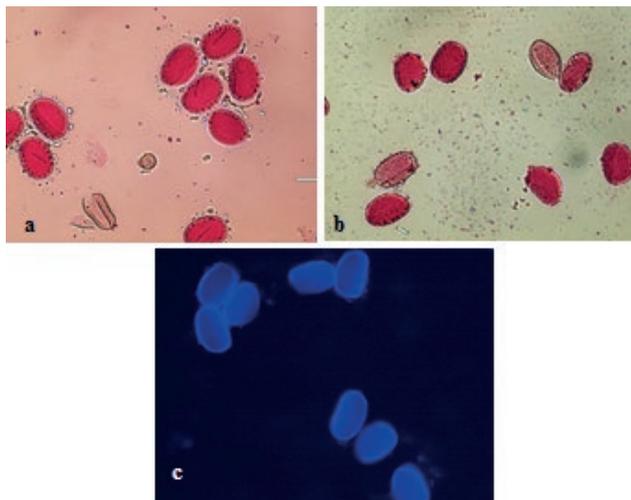
Figure 11. Pollen viability in *Eremurus persicus* (a) 1% acetocarmine, (b) TTC, (c)FDA.

side by side but apparently do not touch. Same has been observed in Human oocytes also (Thermann & Sarto, 1977). Low recombination index of the species due to lack of adequate crossing over can limit genetic variation which may ultimately compromise with the survival of the species (Szczecińska *et al.,* 2016).

The species has asymmetric karyotype of 10 long and 4 short chromosomes. The TF% and Syi and Rec values decreases with increasing asymmetry, while values of others i.e A1 and A2, A and CV_{CL} increase with increasing asymmetry (Zuo& Yuan, 2011) as observed in the present study.

The meiotic abnormalities observed in *E. persicus* may be attributed to high fruit and seed abortion rates. Most conservation biologists believe that before setting up a conservation program for plant species it is better to know about its intrinsic reproductive constraints (Friedman & Ryerson, 2009; Wani *et al,.* 2023). The presently investigated plant species are already facing threats such as habitat fragmentation, overexploitation, and grazing, therefore, the presence of meiotic bottlenecks can add to the factors that lead to a decrease in its population size. Therefore, keeping in view the meiotic abnormalities and anthropogenic threats sustainable conservation strategies are needed to devise for this medicinally important plant species.

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