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# KARYOLOGY OF AMMODENDRON PERSICUM BUNGE EX BOISS-AN ENDEMIC PSAMMOPHYTE TO IRAN

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## ABSTRACT

Vast areas of the world are covered by sand dunes and shifting sands in which the living of inhabitants is being adversely affected. Biological control is an appropriate method for sand dune stabilization. Plant species of *Ammodendron persicum* Bunge ex Boiss. belonging to tribe Sophoreae from family Papilionaceae is an endemic psammophyte shrub to Iran which is distributed mainly in the eastern and southern east border regions of Iran. The species is suitable for sand dune stabilization. For studying the morphology of chromosomes in *Ammodendron persicum*, 50 slide samples were prepared and studied using their root meristems after fixation, hydrolysis and staining stages. Cytological study showed that *Ammodendron persicum* had 2n = 18 chromosomes with a karyotypic formula of 3M + 2 SM + 4T. Total length of chromosomes was 13.3 µm and total length of long and short arms was 9 µm and 4.3 µm, respectively. Maximum and minimum length of the chromosomes was 2.4 and 0.8 µm, respectively, and variation of the relative length range was 12.03µm.

KEYWORDS: Ammodendron persicum, cytogenetic, desert plant, sand dunes.

#### **INTRODUCTION**

Plant species of *Ammodendron persicum* Bunge ex Boiss. belonging to tribe Sophoreae from family Papilionaceae is an endemic psammophyte shrub to Iran which is distributed mainly in the eastern and southern east border regions of Iran including Ghaen in South Khorassan province and Khash in Sistan and Baluchistan province (Abbassi and Amirabadizadeh, 2000).

In view of the great importance of the plant species as a conservator against soil erosion and windbreak in desertification projects and rehabilitation of sandy desert areas of Iran, it appeared necessary to pay more attention on studying such plants and to acquire data on their different characteristics related to karyology, cytology, phenology, physiology, morphology, etc. as well as edaphic and climatic conditions for cultivation and plant breeding programs that can improve certain characteristics such as root length, tolerance to drought and salinity etc. for better utilization of natural resources for attaining sustainable development (Tavakoli *et al.* 2006).

Ammodendron persicum, is a late growing shrub up to 2.5 m high, covered with silvery hairs when juvenile and subglabrous when adult; leaves 10-45 cm long, ending to spine, having mostly 2 and rarely 1-3 pairs of leaflets; leaflets linear-acicular, covered with silky-tomentose hairs on both sides; inflorescence raceme, 3-7 cm long; calyx campanulate and hairy; corolla glabrous with viola color; pods  $20-30 \times 5-15$  mm in dimension, flat, long elliptic, spiral, winged, yellowish brown, bearing 1-3 seeds; seeds oval to elliptic, asymmetric, smooth, light brown; flowering season early to mid spring (Figs. 1, 2,3).

The purpose of this study was to introduce plant species of *Ammodendron persicum* as an endemic psammophyte shrub of Iran for sand dunes fixation and also to

investigate the karyotypic characteristics, finally using this data, the plant karyotype was drawn as an ideogram.

## **MATERIALS & METHODS**

Considering hard cover of the seeds of Ammodendron persicum, scratching method was made to accelerate seed germination before cultivating then, the scratched seeds incubated at 18°C in Petri plates. The treated seeds germinated after a week. The mitotic study of caryotype and morphological characteristic of chromosomes were done on the meristematic cells of root tips, from seed germination. The root tips were pretreated with 8for hydroxyquinoline 1, 2 and 4h and alphabromonaphtaline for 1.5, 2, 3.5h. (Meric and Dane, 1999) in which the best results were obtained from alphabromonaphtaline for 2h. The roots tips were removed from the pretreatment solution and rinsed thoroughly with distilled water so as to remove pretreatment solution from the root surface. In this experiment, Farmer solution was used as pretreatment. The root tips were mounted in this solution for 2.5, 3.5, 4, 20 and 24h and the best results were obtained at 24h. Root tips were hydrolyzed in hydrolic acid 1N at the temperature condition of 60°C. Selected times for the treatments were different according to root thickness as well as the results of the previous trials that ranged 10, 15, and 20 minutes then incubated in an oven at 60°C. After hydrolyzing, the root tips were rinsed with distilled water.

Staining was carried out with aceto-orcein for 20, 30, 60, 75, 120 min and then squashed in 45% acetic-acid. The best results were obtained at 60 min.

The prepared slides were observed under an optical microscope (Zeiss, 1.25X) by magnification of 10X and 40X to determine the presence of metaphasic cells. After

finding metaphasic or prometaphasic cells, they were followed under magnification of 100X. In the case of appropriate and distinct specimens of chromosomes, photography was done by an optical microscope equipped with camera (Contax, 167Mt).

Five metaphase cells were studied for several karyotypic parameters. The chromosome studying include to determine of chromosomes number, ploidy level and to provide of karyotype was been carried out. For the karyotype studying were been determined total length of chromosome (Valles *et al.*, 2001), long arm length, short arm length, relative length of chromosome (Romero-Zarco,1968). The chromosomes type was determined by Levania *et al.* table (16). Using this data, the plant karyotype was drawn as an ideogram.

## **RESULTS AND DISCUSSION**

The results showed that Ammodendron persicum had 2n =18 chromosomes (Fig. 4) with karyotypic formula of 3M + 2 SM + 4T including 22.23% submetacentric, 33.3% metacentric and 44.45% telocentric. The ideogram of Ammodendron persicum was shown in Fig 5. Total length of chromosomes was 13.3 µm and total length of long and short arms were 9 µm and 4.3 µm respectively. Maximum and minimum length of chromosomes was 2.4 µm and 0.8 µm, respectively (Table 1). According to a number of reports on plant karyology (Huziwara, 1962, Meric and Dane, 1999, Stace and Misra, 1994 ), the best part of a plant for studying mitotic division and identification of chromosomal arrangement and preparation of karyotype is apical meristems of roots, because root induction occurs rapidly, the mitotic division is very fast in this area and lack of chlorophyll in the root makes it easy for studying the cytogenetic characteristics (Safarnejad and Marvian Hosaini, 2005).

Chromosomal information is used in plant classification as two distinct methods including karyotype studies and chromosome pairing in mitotic division (Skula, 1984, Stace and Misra, 1994). Huziwara (1962) used from general karyotype form as a classification factor for expression of symmetry conditions.

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 Table 1- Chromosome number, length and type in Ammodendron persicum.

Chromosome	Total	Long	Short	Long/short	Short/long	Relative	Chromosome
pair	length	arm	arm	arms	arms	length	type
	(µm)	length	length	length	length		
		(µm)	(µm)	ratio	ratio		
1	2.4	1.20	1.20	1.00	1.00	18.04	М
2	2.0	1.33	0.67	1.99	0.51	15.03	SM
3	1.6	0.80	0.80	1.00	1.00	12.03	Μ
4	1.6	1.43	0.17	8.41	0.11	12.03	Т
5	1.6	1.07	0.53	2.01	0.49	12.03	SM
6	1.3	1.23	0.12	10.25	0.09	10.22	Т
7	1.2	0.60	0.60	1.00	1.00	9.02	М
8	0.8	0.67	0.13	5.16	0.194	6.01	Т
9	0.8	0.67	0.13	5.16	0.194	6.01	Т

M: metacentric, SM: submetacentric, T: telocentric



Fig. 1. Ammodendron persicum Bunge ex Boiss. plant growing on sand dunes.



Fig.2. Herbarium specimen of Ammodendron persicum Bunge ex Boiss. at the flowering stage

Karyology of Ammodendron persicum bunge of Iran



Fig.3. Herbarium specimen of Ammodendron persicum Bunge ex Boiss. at the fruiting stage



**Fig.4**. Chromosome number of *Ammodendron persicum*, 2n = 18 (×100)



Fig. 5 Ideogram of Ammodendron persicum chromosomes