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Karyotype Analysis of Some Legume Species (*Vicia noeana* Boiss. and *Lathyrus sativus* L.) Collected From Native Vegetation

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Abstract: In this study, morphology of chromosomes of some legume species (*Vicia noeana* Boiss., *Lathyrus sativus* L.) collected from the native vegetation in central province of Tokat in Turkey was determined by karyotype and idiogram analysis. The cytological studies showed that the chromosome numbers of *Vicia noeana* Boiss. and *Lathyrus sativus* L. were $2n=12$ and $2n=14$, respectively. It was determined that all of the chromosomes of *Vicia noeana* Boiss. were submedian. Of the 14 chromosomes present in *Lathyrus sativus* L. karyotype, 4 were submedian and 10 were median. In addition to this, chromosome length, arm ratio and relative chromosome length were between 5.75-7.77 μm , 2.43-2.96 and 7.21-9.72 for *Vicia noeana* Boiss. and between 4.75-6.36 μm , 1.31-1.82 and 6.16-8.26 for *Lathyrus sativus* L., respectively.

Key words: *Vicia noeana* Boiss., *Lathyrus sativus* L., karyogram, idiogram

Introduction

The family Leguminosae comprises of about 690 genera and more than 17,000 species (Hutchinson, 1964). *Vicia noeana* Boiss. and *Lathyrus sativus* L., two of the legumes, belongs to the tribe *Vicieae* of the sub-family Papilionaceae (Davis, 1970). Among the legumes, *Vicia* and *Lathyrus* have several interesting taxonomic, evolutionary and genetic problems, besides their well-known forage potentials (Schifino-Wittmann *et al.*, 1993). The genus *Vicia* has 180-200 species which are mostly temperate annual and perennial, auto and allogamous, erect or climbing plants (Kupicha, 1976). *Lathyrus* has 120-170 species, distributed through temperate Northern Hemisphere and South America, with herbaceous, climbing, annual and perennial, auto and allogamous plants (Jackson and Yunus, 1984). In Turkey, this genus includes 58 species (Davis, 1970). All genera (*Vicia* and *Lathyrus*) have their centres of origin in the Old World. These species (*Vicia* and *Lathyrus*) are used for human and animal nutrition, ornamentation and green manure.

Elci (1965) reported that *Vicia noeana* Boiss. has 12 chromosomes with submedian centromeres. He also obtained that the chromosome length, the arm ratio and the relative chromosome length varied from 5.22 to 7.10 μm , 0.348 to 0.441 and 7.030 to 9.573, respectively. Schifino-Wittmann *et al.* (1993) found that *Vicia hirsuta* L., *V. villosa* Roth., *V. nana* Vog., *V. epetalaris* Burk., and *V. macrograminea* Burk. have 14 chromosomes and *V. sativa* L. and *V. pannonica* L. have 12 chromosomes. Moreover, *Lathyrus sativus* L. has $2n=14$ chromosomes (Yamamoto *et al.*, 1984; Zhao *et al.*, 1984; Kumar and

Sinha, 1989; Kar and Sen, 1991; Campbell *et al.*, 1994; Karadag and Buyukburc, 2000). The cytological experiments conducted with five grasspea lines by Karadag and Buyukburc (2000) showed that four grasspea lines (452, 439, 455, 463) have sat-chromosomes but one grasspea line (38) have no sat-chromosome. Karadag and Buyukburc (2000) also reported that the chromosome length, the arm ratio and the relative chromosome length in grasspea line 38 varied from 4.57 to 6.54 μm , 1.21 to 1.50 and 5.92 to 8.50, respectively. In addition, they reported that the chromosome length, the arm ratio and the relative chromosome length in grasspea line 463 varied from 4.93 to 6.18 μm , 1.25 to 2.13 and 6.40 to 8.01, respectively. Kumar and Sinha (1989) reported that 10 grasspea varieties have no sat-chromosomes. Kar and Sen (1991) measured that the chromosome lengths of *Lathyrus sativus* L. varied from 3.5 to 5.5 μm .

The present work aimed to gather basic information on these species for their better characterisation, and especially to establish the basis for future breeding projects.

Materials and Methods

The materials (*Vicia noeana* Boiss., *Lathyrus sativus* L.) used in this study were collected from the native vegetation in central province of Tokat in Turkey. The seeds of *Vicia noeana* Boiss. and *Lathyrus sativus* L. were then germinated on moist filter paper in the laboratory at 25°C. The growing root tips, 1.0 to 1.5 mm long, were excised and pretreated in a saturated solution of α -bromonaphthalene for 3 hours. After the

pretreatment, the root tips were fixed in glacial acetic acid for ½ h. The chromosomes were stained according to Feulgen method after hydrolysis in 1 N HCl for 12 min at 60°C (Sharma and Gupta, 1982; Karadag and Buyukburc, 2000). Root tips were then squashed in 1% acetocarmine (Mannan *et al.*, 1991).

The photomicrographs were taken with a Zeiss research microscope fitted with a microphotographic attachment, using an oil immersion objective (100 X). The karyotypes were determined by examining about a well spread metaphases for each species. In each of these two species, 10 cells which were flat and the chromosomes well spread were photographed and arm lengths were measured on prints enlarged to a total magnification of 2720. Measurements were reported in μm . Each chromosome was identified on the basis of its total chromosome length, arm ratio and relative chromosome length. Analysis and identification of each chromosome were made according to the following parameters (Krikorian *et al.*, 1983):

Total chromosome length (μm)= the lengths of the chromosome arms (length of long arm + length of short arm)

$$\text{Relative chromosome length} = \frac{\text{Chromosome length}}{\text{Total of the chromosome lengths}} \times 100$$

$$\text{Arm ratio} = \frac{\text{Length of long arm}}{\text{Length of short arm}}$$

In the karyotypes, the chromosomes were arranged and numbered in order of decreasing length. The position of centromeric constriction was recorded as median (m: 1.0-1.7) and submedian (sm: 1.7-3.0) by the arm ratio (Levan *et al.*, 1964).

Results and Discussion

Morphological properties of mitosis chromosomes for *Vicia noeana* Boiss: The diploid chromosome number of *Vicia noeana* Boiss. was $2n=12$ (Fig. 1). The karyogram and idiogram of this species are shown in Fig. 2 and 3. Measurements of the chromosome complement are presented in Table 1. Some of the morphological features useful in identifying each individual chromosome of the *Vicia noeana* Boiss. complement are presented below:

Chromosome I: This is the longest chromosome of the complement; the centromere is in the submedian region. The chromosome length, the arm ratio and the relative chromosome length were $7.77 \mu\text{m}$, 2.68 and 9.72, respectively.

Chromosome II: It is a submetacentric chromosome. The relative chromosome length was 9.72. The chromosome length and the arm ratio were $7.21 \mu\text{m}$ and 2.43, respectively. This is the most symmetrical chromosome of the complement.

Chromosome III: The chromosome length was $6.76 \mu\text{m}$. The centromere is in the submedian region of the chromosome. The arm ratio and the relative chromosome length were 2.87 and 8.45, respectively.

Chromosome IV: This chromosome has a submedian centromere (arm ratio 2.96). The chromosome length was $6.40 \mu\text{m}$ and 7.95 in the relative chromosome length. Also, this is the most asymmetrical chromosome of the complement.

Chromosome V: The arm ratio of this chromosome was 2.55, the relative chromosome length was 7.61 and the chromosome length was $6.07 \mu\text{m}$. This chromosome is submetacentric.

Chromosome VI: This is the shortest chromosome of the complement (the chromosome length $5.75 \mu\text{m}$). The position of the centromere is submedian. The relative chromosome length and the arm ratio were 7.21 and 2.72, respectively.

The results of the cytological studies showed that the chromosome number of *Vicia noeana* Boiss. was $2n=12$. No satellite is present on the karyotype of the species. Similar results were reported by Elçi (1965). In addition, It was determined that all of the chromosomes of *Vicia noeana* Boiss. were submedian. The chromosome length, the arm ratio and the relative chromosome length were $5.75-7.77 \mu\text{m}$, 2.43-2.96 and 7.21-9.72, respectively. In looking at the species previously karyotyped by Elçi (1965) and the species karyotyped in this study, *Vicia noeana* Boiss. was determined to have similarity in their chromosomes.

Morphological properties of mitosis chromosomes for *Lathyrus sativus* L.: The somatic chromosome number of *Lathyrus sativus* L. was $2n=14$ (Fig. 4). The karyogram and idiogram of this species are presented in Fig. 5 and 6. Measurements of the chromosome complements are shown in Table 2. Some of the morphological features that are useful in identifying each individual chromosome of the *Lathyrus sativus* L. complement are presented below:

Chromosome I: The longest chromosome of the complement and has a median centromere. The

Table 1: Somatic chromosome measurements of *Vicia noeana* Boiss

Chromosome number	Chromosome length (μm)			Arm ratio			Relative chromosome length		
	Mean	Min.	Max.	Mean	Min.	Max.	Mean	Min.	Max.
I	7.77	6.37	8.92	2.68	1.55	3.43	9.72	9.00	10.55
II	7.21	6.07	8.67	2.43	1.73	2.95	9.01	8.46	9.66
III	6.76	5.87	7.77	2.87	1.95	3.65	8.45	8.00	8.66
IV	6.40	5.65	7.31	2.96	2.30	3.63	7.95	6.98	8.26
V	6.07	5.25	7.08	2.55	1.63	3.14	7.61	7.14	8.00
VI	5.75	4.96	6.19	2.72	1.46	3.69	7.21	6.79	8.03

Table 2: Somatic chromosome measurements of *Lathyrus sativus* L.

Chromosome number	Chromosome length (μm)			Arm ratio			Relative chromosome length		
	Mean	Min.	Max.	Mean	Min.	Max.	Mean	Min.	Max.
I	6.36	5.39	7.15	1.31	1.15	1.77	8.26	7.51	8.67
II	5.94	5.56	6.38	1.76	1.34	2.20	7.70	7.30	8.19
III	5.66	5.39	6.02	1.38	1.16	1.75	7.34	7.20	7.53
IV	5.47	4.96	5.81	1.82	1.61	2.33	7.09	6.62	7.39
V	5.31	4.96	5.75	1.38	1.24	1.81	6.89	6.61	7.15
VI	5.08	4.72	5.50	1.32	1.15	1.51	6.59	6.30	6.82
VII	4.75	4.46	5.28	1.35	1.19	1.61	6.16	5.76	6.48



Fig. 1: Chromosomes of root tip cells of *Vicia noeana* Boiss. (X 2720)

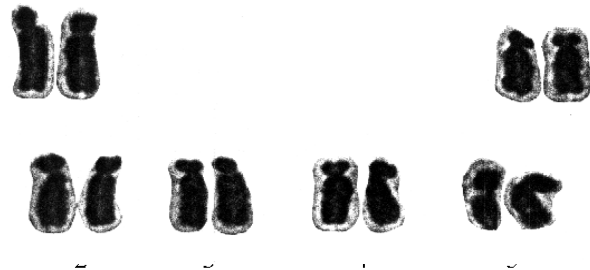


Fig. 2: Karyotype of *Vicia noeana* Boiss. (X 2720)

chromosome length was $6.36 \mu\text{m}$. The relative chromosome length and the arm ratio were 8.26 and 1.31, respectively. This is the most symmetrical chromosome of the complement.

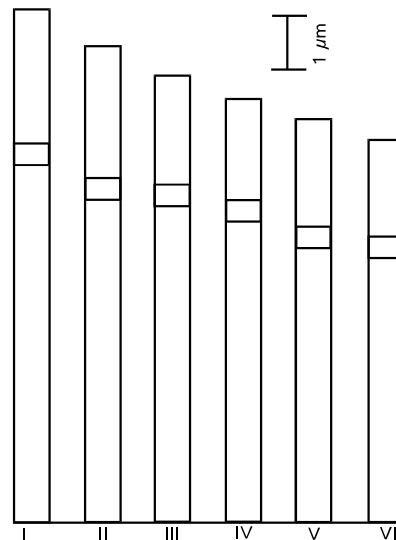


Fig. 3: Idiogram of somatic chromosomes of *Vicia noeana* Boiss.

Chromosome II: The centromere is in the submedian region of the chromosome (arm ratio 1.76). The chromosome length and the relative chromosome length were $5.94 \mu\text{m}$ and 7.70, respectively.

Chromosome III: The arm ratio and the relative chromosome length were 1.38 and 7.34, respectively. The chromosome length was $5.66 \mu\text{m}$. The centromere is located in the median region.

Chromosome IV: This is the most asymmetrical chromosome of the complement (arm ratio 1.82). The

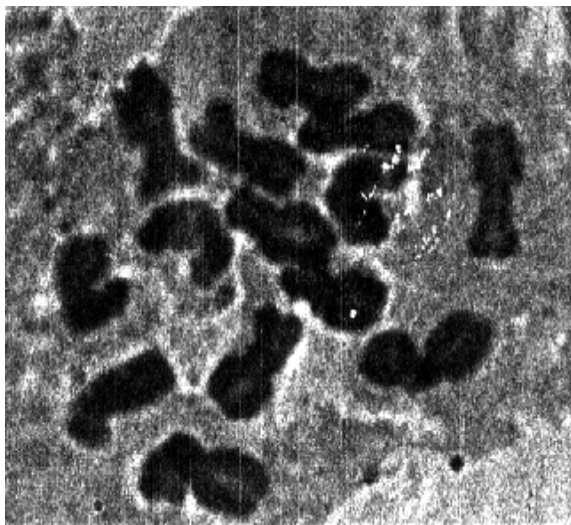


Fig. 4: Chromosomes of root tip cells of *Lathyrus sativus* L. (X 2720)



Fig. 5: Karyotype of *Lathyrus sativus* L. (X 2720)

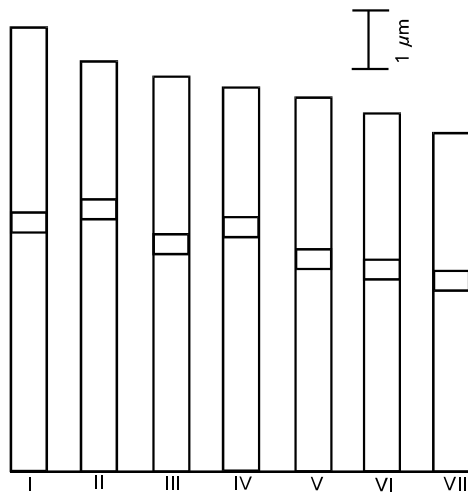


Fig. 6: Idiogram of somatic chromosomes of *Lathyrus sativus* L.

chromosome length and the relative chromosome length were $5.47 \mu\text{m}$ and 7.09 , respectively. The centromere is in the submedian region of the chromosome.

Chromosome V: It is a metacentric chromosome. The chromosome length and the arm ratio were $5.31 \mu\text{m}$ and 1.38 , respectively. Also, the relative chromosome length was 6.89 .

Chromosome VI: The chromosome length and the relative chromosome length were $5.08 \mu\text{m}$ and 6.59 , respectively. The position of the centromer is median on chromosome VI. The arm ratio was 1.32 .

Chromosome VII: This is the shortest chromosome of the complement. The chromosome length and the relative chromosome length were $4.75 \mu\text{m}$ and 6.16 , respectively. The centromere is in the median region (arm ratio 1.35).

The results of the cytological studies showed that the chromosome number of *Lathyrus sativus* L. was $2n=14$. Similar results were reported by some other researchers (Yamamoto *et al.*, 1984; Zhao *et al.*, 1984; Kumar and Sinha, 1989; Campbell *et al.*, 1994; Karadag and Buyukburc, 2000). Of the 14 chromosome present in *Lathyrus sativus* L. karyotype, 4 were submedian and 10 were median. In addition, the chromosome length, the arm ratio and the relative chromosome length were 4.75 - $6.36 \mu\text{m}$, 1.31 - 1.82 and 6.16 - 8.26 , respectively. No satellite is present on the karyotype of this species. Our karyotype determination of *Lathyrus sativus* L. was in agreement with results reported by Karadag and Buyukburc's (2000). Chromosome VI of *Lathyrus sativus* L. (Table 2) was very similar to chromosome VI of *Lathyrus sativus* L. as karyotyped by Karadag and Buyukburc (2000) (arm ratio: 1.32 ; chromosome length: $4.98 \mu\text{m}$; relative chromosome length: 6.46). On the other hand, our karyotype analysis of *Lathyrus sativus* L. was different from that of Kumar and Sinha (1989). For example, Kumar and Sinha (1989) reported that the KH-5 and KH-15 varieties of *Lathyrus sativus* L were 10 submetacentric and 4 metacentric. In addition, the KH-4 and KH-8 varieties were 6 submetacentric, 6 metacentric and 2 subtelo-centric. Kumar and Sinha (1989) also reported that the lengths of longest and shortest chromosome of *Lathyrus sativus* L. ranged between 5.44 to $4.06 \mu\text{m}$ for KH-5, 5.39 to $3.40 \mu\text{m}$ for KH-15, 4.83 to $2.13 \mu\text{m}$ for KH-4 and 4.16 to $2.96 \mu\text{m}$ for KH-8. Research findings showed that different forms within *Lathyrus sativus* L. species had different chromosome morphology. These differences might be due to evolution of this species. Chandola and Jain (1979) reported that as the evolution proceeds, the chromosome length of a species could decrease and turn from symmetrical to asymmetrical form during the course of evolution.

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