

# CYTOGENETICAL STUDIES IN RANGE GRASSES OF IRAN

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## ABSTRACT

Cytogenetical and breeding studies of range grasses are in hand. Present article describes preliminary cytogenetical analysis of wheatgrass (*Agropyron*) and brome grass (*Bromus*) taxa (*Agropyron trichophorum* n = 21, *A. pectiniflorum* n = 21), two different populations of *A. repens* (n = 28) and *Bromus stenostachyus* (n = 14). The species varied with regard to chiasma number and distribution. The highest value occurred in *A. repens* (Mako population). Heterogeneity test for the paired samples showed lack of heterogeneity for chiasmata indicating homogeneity of *Agropyron* taxa. However a test for ring and rod bivalents showed significant differences between the two populations of *A. repens* indicating genomic differences. Multivalents occurred regularly except in *A. pectiniflorum* showing diplontic behaviour. B-chromosomes occurred in *Bromus stenostachyus* which moved to the poles, but were seen as laggard too. UPGMA cluster analysis separated two populations of *A. repens*, indicating their genomic differences. Other meiotic variations noticed were: occurrence of a synezetic knot and cytotoxicity causing aneuploidy in *B.stenostachyus*; clumping and laggards occurred frequently, indicating heterozygosity of the taxa due to cross pollination.

## KEYWORDS

Cytogenetics, wheatgrass, brome grass, meiosis

## INTRODUCTION

Knowledge of cytogenetics, breeding behaviour and genomic variations are important in planning breeding programmes of the plants. Range grasses of Iran grow widely in different regions of the country, therefore a cytogenetical survey of these plants was undertaken. The present article describes a preliminary cytogenetical study of some wheatgrass (*Agropyron*) and brome grass (*Bromus*) species of Iran.

## MATERIALS AND METHODS

Flower buds were collected from three species of wheatgrass (*Agropyron*) [*Agropyron trichophorum* (Link) Richt., *A. pectiniflorum* Romer & Schult., *A. repens* (L) Beauv (Two populations)] and one species of brome grass (*Bromus*) (*Bromus stenostachyus* Boiss.) from different regions of the country (Table 1). Flower buds were collected and fixed in fixative for 24 hours and then washed thoroughly. They were then kept in 70% alcohol until used (Sheidai and Inamdar, 1992a). A minimum of fifty and a maximum of seventy PMCs were analysed. Meiotic analysis was carried out from the earliest stages of division (Sheidai, 1992). Heterogeneity tests were performed on chromosome behaviour such as chiasma formation and chromosomal association (Sheidai and Inamdar, 1992 b). Cluster analysis was performed on cytological data (Sheidai et al., 1996). Pollen fertility was checked using acetocarmine and 50% glycerin (1:1) (Sheidai and Inamdar, 1991).

## RESULTS AND DISCUSSION

**Meiotic variations.** The following variations were observed in meiotic behaviour of the chromosomes: 1- Prophase-I substages of meiosis: In all the species studied synezetic knot stage was observed instead of leptotene and zygotene. In the early knot stage thin chromatin strands were surrounding the nucleolus which later on were drawn towards the nucleolus (Fig.I.1). Unravelling of chromosomes occurred in the next step. Now the chromatin strands are thicker due to active synapsis of homologues in the knot substage (Fig. I. 2). Pachytene followed this stage showing end to end attachment of chromosomes and occurrence of inter-chromosomal attachment. Presence of such substages of prophase - I has been reported in several plant species (John et al., 1987, Klastereska and Kaul, 1984, Sheidai and Inamdar, 1991). The pollen fertility observed

indicated that these substages are not abnormal and can be considered as meiotic variants. This is the first report of a synezetic knot substage in wheatgrass (*Agropyron*) and brome grass (*Bromus*).

Chromatin extrusion and cytotoxicity was noticed to occur among meiocytes at different stages of meiosis from the knot substage onwards (Fig.I.2,3). Migration of the chromatin occurred in a different direction. Several PMCs were observed to possess extra chromosomes in *Bromus stenostachyus* (Fig. I. 6). Occurrence of cytotoxicity and chromatin migration has been reported in many taxa including gramineae (Bedi et al., 1985; Patra, 1986). In rye (*Hordeum*) cytotoxicity resulted in an increase in chromosome number (Kamra, 1960). It has also been reported in *Triticum Agropyron* (Gaul, 1954).

**Chromosomal associations and segregation.** Chromosomal number and chiasma formation are presented in Table 1 (Fig. I.5, Fig. II.1-4). The highest mean total chiasma occurred in *A.repens* growing at 1825 m altitude at Mako followed by the second population of the same species growing at 1100 m at Uromieh. No intercalary chiasma occurred in Uromieh population (Table 1). The species studied also varied with regard to mean chiasma per bivalent. Heterogeneity test for chiasma distribution showed lack of heterogeneity among the species. However a difference was significant mean seen in populations of *A. repens* with regard to number of bivalents indicating genomic differences. Cluster analysis of chromosomal data using UPGMA, single linkage and complete linkage methods produced the same results separating these two populations (Fig. II. 8), indicating distinctness of the clusters produced. Fit of the clusters to the original data was confirmed by obtaining a cophenetic correlation ( $r > 0.70$  (Stanton et al., 1994). Multivalents occurred regularly in the species studied (Fig. II. 4,6) except *A. trichophorum* which showed only bivalent formation (Table 1). Trivalents and quadrivalents only occurred in *A. repens* growing in Uromieh. Chromosome segregation occurred normally, however in many cells laggard chromosomes were seen (Fig.I. 7-9). Such chromosomes would make micronuclei (Fig. I. 10, Fig. II. 4, 5). Reduction observed in pollen fertility of these species may be due to such anomalies. B-chromosomes were observed in *B. stenostachyus* (Fig. I. 5). Their number varied from one to two and could arrange themselves on a metaphase plane and move to the poles. However, they were seen as laggards too. Other cytogenetical anomalies observed include clumping of chromosomes and stickiness. Occurrence of such anomalies may indicate presence of a heterogenous background created due to cross pollination. Genomic differences present in two populations of *A. repens* can be used for breeding purposes.

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**Table 1**

Chromosome behavior with regard to chiasma number and distribution and chromosomal association

SPECIES	n	T-X	I-X	TO-X	X/B	RB	RDB	II%	III%	IV%	VI%
<i>A.repense</i> (Mako)	28	49.47 ±0.1 45-52	0.32 ±0.08 0-1	49.79 ±0.1 45-53	1.77 ±0.1 20-25	22.5 ±0.1 20-25	5.28 ±0.08 0-8	99	-	1	-
<i>A.repense</i> (Uromieh)	28	55.80 ±0.01 52-56		55.8 ±0.01 52-56	1.99 ±0.1 23-28	27.8 ±0.1 23-28	-	99.4	0.4	-	0.19
<i>A.trichophorum</i>	21	41.80 ±0.08 35-42	2.58 ±0.1 1-10	44.41 ±0.1 38-51	2.10 ±0.03 17-21	20.4 ±0.03 17-21	0.07 ±0.0 0-2	96.8	-	3.2	-
<i>A.pectiniforome</i>	21	41.88 ±0.02 41-42	-	41.88 ±0.02 41-42	1.99 ±0.02 20-21	20.9 ±0.02 20-21	0.10 ±0.20 0-2	100	-	-	-
<i>B.stenostachyus</i>	14	23.33 ±0.07 18-28	0.21 ±0.01 0-2	23.54 ±0.06 19-27	1.68 ±0.07 4-14	9.87 ±0.07 4-14	4.0 ±0.07 1-10	97.0	-	3.0	-

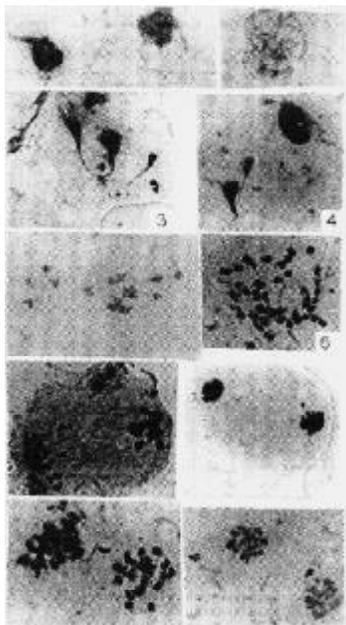
T-X = Terminal chiasma I-X = Intercalary chiasma TO-X = Total chiasma

X/B = Mean chiasma / Bivalent RB = Ring bivalent RDB = Rod bivalent

II = Bivalent III = Trivalent IV = Quadivalent VI = Hexavalent

**Figure I**

- 1 - Early synezetic knot in *A. repens*.
- 2 - Unravelling of the chromosomes from the knot in *Bromus stenostachyus*.
- 3 - Cytomixis in *B. stenostachyus*.
- 4 - Chromatin migration in *B. stenostachyus*.
- 5 - B-Chromosome in *B. stenostachyus*.
- 6 - Presence of extra chromosomes in *B. stenostachyus*.
- 7 - Laggard chromosome in *A. repens*.
- 8 - Laggard chromosome in *B. stenostachyus*.
- 9 - Laggard chromosome in *A. trichophorum*.
- 10 - Micronuclei formation in telophase-I in *A. pectiniforome*.



**Figure II**

- 1 - Bivalents in *A. repens*.
- 2 - Bivalents in *A. trichophorum*.
- 3 - Cytomixis in *B. stenostachyus*.
- 4 - Quadivalent in *B. stenostachyus* (Arrow).
- 5 - Micronuclei formation in *A. repens*.
- 6 - Quadivalent in *A. repens*.
- 7 - Bivalents in *B. stenostachyus*.
- 8 - Dendrogram produced from UPGMA cluster analysis : 1 = *A. repens* (Mako) 2 = *A. trichophorum*, 3 = *A. pectiniforome*, 4 = *A. repens* (Uromieh).

