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T. Tsuchiya
Colorado State University, Fort Collins, CO

W. McGinnies
ARS, Crops Research Laboratory, USDA

A. Shahla
Colorado State University, Fort Collins, CO

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A CHROMOSOME STUDY OF BLUE GRAMA (*BOUTELOUA GRACILIS*) IN NORTHERN COLORADO

T. Tsuchiya

Department of Agronomy, Colorado State University
Fort Collins, CO 80523

W. McGinnies

ARS, Crops Research Laboratory, USDA

A. Shahla

Department of Agronomy, Colorado State University
Fort Collins, CO 80523

Professor T. Tsuchiya was professor of Agronomy at Colorado State University at the time of his death in May 1992. This essay is being published posthumously in his memory. The editors especially want to thank Kenneth P. Vogel, Professor of Agronomy, University of Nebraska-Lincoln, for his editorial advice in the final preparation of this essay.

Abstract. *Chromosomes were studied in somatic cells of 60 plants of blue grama,* Bouteloua gracilis *(Willd. ex Kunth) Lag ex Griffiths, collected from a 7-hectare field of native range at the Central Plains Experiment Range (CPER), Nunn, Colorado, to determine chromosome constitution in relation to the plant characteristics. Somatic chromosomes were studied in root tips collected from vigorously growing plants in the greenhouse. Acetocarmine squash method was used to make slide preparation. The majority of the plants were 2n=40. However, three plants were 2n=50 and two were 2n=60. The pentaploid plants (2n=50) were recorded for the first time in this report. Additional 94 plants collected from entire CPER were all 2n=40. The basic chromosome number of blue grama was determined to be x=10. There was no significant relation between chromosome number and morphological traits: culm height and number, basal diameter, and plant dry weight.*

Figure 1. Blue grama. From *Grass: The Yearbook of Agriculture* 1948, published by the Government Printing Office.

Blue grama [(*Bouteloua gracilis* (Willd. ex Kunth) Lag ex Grifiths)] is one of the most widespread and valuable native forage grass species (Fig. 1) on the western Great Plains (Avdulov 1931). Morphological characteristics of blue grama plants are variable within and among populations. Chromosomes have been studied to some extent in various populations together with several other species in the genus *Bouteloua* (Gould 1979), but primary emphasis was on comparisons among populations. Some of the cytological results from the early work may not be completely reliable because of the techniques used. Fults (1942), who studied some blue grama materials in Colorado and other states, used a paraffin section technique for root tip chromosome studies. Judging from the photomicrographs shown, this method of slide preparation might have been inadequate for making accurate chromosome counts. Based on his results, Fults (1942) proposed that the basic chromosome number was x=7 for *Bouteloua*, and all the materials he studied were polyploids (2n=21, 28, 35, and 42). However, he could not find diploid plants with 2n=14. Snyder and Harlan (1953) studied meiotic chromosomes in the materials collected from New Mexico, Texas, and Oklahoma and provided reliable results with good photomicrographs of meiotic cells. No reliable report (other than Fults 1942) on chromosomes in blue grama in Colorado exists.

This paper is a brief report on the results of somatic chromosome studies in blue grama from northern Colorado in relation with morphological variations (McGinnies et al. 1988). Reliable photomicrographs of the somatic chromosomes of blue grama are presented for the first time in this paper (Fig. 2). Also, pentaploid blue grama (2n=50) is reported for the first time in this paper. The results obtained suggest that the basic chromosome number is x=10, rather than x=7, in blue grama.

Materials and Methods

Morphological and ecological studies of blue grama have been conducted for several years at the Central Plains Experiment Range (CPER), Nunn, Colorado. Sixty individual blue grama plants were collected from a 7-hectare field and transplanted into a spaced-plant nursery at the CPER for detailed observations. These data showed that the local blue grama population at CPER consists of plants with considerable variability in growth characteristics such as plant height, vigor, and number of tillers (McGinnies et al. 1988). Cytological information was not available for these original plants, so chromosome numbers of 60 plants from this nursery were studied

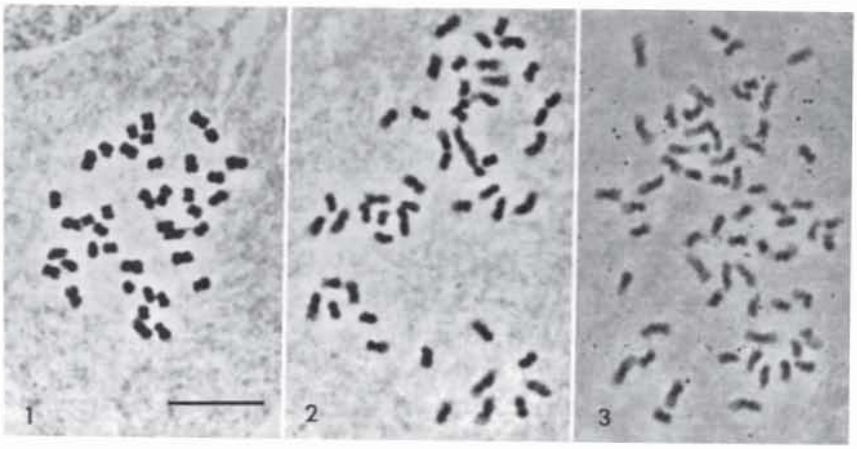

Figure 2. Somatic chromosomes of 1) tetraploid (2n=40), 2) pentaploid (2n=50), and 3) hexaploid (2n=60) plants of blue grama. Scale bar is 10 μ long.

in root-tip mitosis. An additional 94 plants collected from the entire CPER were also studied in root-tip mitosis.

Root tips were collected from all plants from the nursery that were grown in the USDA/ARS Crops Research Laboratory greenhouse at Fort Collins. Root tips were pretreated in tap water at approximately ±0°C in vials for 16 to 18 hours. The pretreated materials were then transferred to 0.7% acetocarmine solution and stored for several days. A squash preparation was made in 45% acetic acid (Tsuchiya 1971; Tsuchiya and Nakamura 1979).

The plants were observed in both the nursery and the greenhouse for two years, and characteristics were recorded with measurements of various plant organs (McGinnies et al. 1988). All plants were scored for maximum culm height, number of reproductive culms, basal diameter, and weight of the individual plants. A combined score was then calculated based on a summation of the measurements for the four traits. Because of a great variation in morphology among different plants in their population, the entire population was subdivided into four classes from the smallest least vigorous plants (1) to the largest, most vigorous plants (4) (Table 1). No information on the plant characteristics is available for the additional 94 plants collected from the entire CPER.

TABLE 1
CHROMOSOME NUMBERS OF BLUE GRAMA IN CPER, COLORADO

Class	Plant size	40	approx. 40	50	60	Total
			Nursery Plants			
1	Minute	9	1	0	1	11
2	Small	18		3	0	21
3	Medium	12	1	0	0	13
4	Large	14		0	1	15
Total		53	2	3	2	6
			Field Sample			
		29		3	2	94

Results

Chromosome numbers were definitely determined for 58 of the 60 nursery plants studied. Approximate chromosome numbers were determined for the two remaining plants (Table 1). Diploids were not found in this 60-plant sample. Fifty-three plants had 2n=40 and two had approximately 40 chromosomes. Chromosome numbers of the 94 additional plants collected from the entire CPER were mostly 2n=40, with a few plants having 2n=50 and or 2n=60 chromosomes (Table 1).

Preliminary observations of meiosis in a limited number of plants with 2n=40 indicated an autotetraploid nature. Meiosis in two 50-chromosome plants and one 60-chromosome plant indicated an autopentaploid and autohexaploid nature, respectively. (Tsuchiya et al. 1986).

The relationship between morphological characteristics and chromosome numbers was evaluated. The three 2n=50 plants were small (class 2), whereas one each of the two 2n=60 plants was minute (class 1) or large (class 4).

The results obtained from this limited material from the Central Plains Experimental Range, Colorado, suggest that there is no correlation between chromosome number and morphological traits such as plant height, number of reproductive culms, and weight of individual plants. The observed polymorphism in plant characteristics may be ascribed to the genetic variation within this autopolyploid population.

Discussion

Chromosome numbers in blue grama were studied by several previous workers with different results (Gould 1979). Fults (1942) reported a wide range of chromosome numbers, 2n = 21, 28, 35, 42, 61, and 77 in somatic cells which were studied by the classical paraffin-section technique. Avdulov (1931) reported 2n=40 and Nielsen and Humphrey (1937) posted 2n=40 and 2n=42. Snyder and Harlan (1953), working with meiotic cells in various blue grama materials collected in the southern Great Plains (New Mexico, Texas, and Oklahoma), observed chromosome numbers of 2n=20, 40, 42, 60, and 84. In all of these studies, most frequently observed plants were tetraploids with 2n=40, followed by diploids (2n=20). The least frequent were 2n=42 and 84.

Fults (1942) interpreted his results with the assumption that the basic chromosome number of the genus *Bouteloua*, including blue grama, was x=7. Judging from the photomicrographs in his paper (Fults 1942, p. 48) it is possible that his chromosome counts of paraffin-sectioned preparations could have been inaccurate. Inaccurate counts of chromosomes in *Bouteloua* are always possible because of the small size and different sizes in a complement, especially with inadequately prepared slides. The possibility of different basic numbers (x=7 and 10) in different populations cannot be completely ruled out. Brown (1950, 1951) studied several *Bouteloua* species (but not including blue grama) and found the basic number was x=7. However, Brown also used the paraffin section method and did not show photomicrographs. It should be pointed out that Brown (1950, 1951) and Fults (1942) always showed the chromosome numbers to be multiples of x=7, in many species, while other researchers found the chromosome number to be multiples of x=10.

Meiotic configurations in materials studied by Snyder and Harlan (1953) provided conclusive photographic evidence to support their interpretation that the basic number of their blue grama population was x=10, not x=7. The results of the present study confirm the basic number of x=10 for the materials collected in a rather limited area in Colorado. The additional 94 plants randomly collected from entire CPER were all 2n=40. This result also supports the conclusion that basic number of blue grama is x=10. A preliminary experiment of meiosis in plants with 2n=40, 50, and 60 also indicated that the basic number is x=10 in blue grama (Tsuchiya et al., 1986). In order to determine the real basic chromosome number of blue grama, it may be necessary to study meiotic chromosomes in many more species in the genus *Bouteloua*.

It is interesting that three definite cases of pentaploid (2n=5x=50) and two cases of hexaploid (2n=6x=60) were found, when the majority of plants in this population were tetraploid with 2n=40 or near 40. Snyder and Harlan (1953) and other researchers have never found any pentaploids (2n=50) in their studies, although they found hexaploid (2n=60) and other numbers (2n=42, 84)(Gould, 1979).

The lack of relationship between plant characteristics and chromosome complement found in this study (Table 1) may or may not be conclusive, and it will be necessary to conduct systematic studies with a large number of plants collected from different localities. The study of the relationship between morphology and chromosome complement in polyploids, especially the autopolyploids such as blue grama, may not be easy because of possible compensation by more than two homologous chromosomes. More research is necessary, however, to reach a final conclusion on this matter, because the number of pentaploid and hexaploid plants were too small to permit reliable comparison between chromosome number and plant characteristics. The wide range of variability in blue grama populations may be more genetic than cytological. Diploid populations (2n=20) would probably be better suited for genetic studies than the polyploids available to us.

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