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PROCESSING AND ANALYSIS OF PLANT CHROMOSOME IMAGES

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Abstract

In this paper we present the application of a system for human chromosome image analysis and classification CAIS to describe the *N*-banded chromosome images of the wild cereal species *Aegilops geniculata* Roth. In addition, a modified system CHROMOS for processing and analysis of chromosome images from different species is described. The applicability of the system CHROMOS to establish cytological markers for plant chromosomes identification is assessed.

Key words: Aegilops geniculata, CAIS, CHROMOS, chromosome image, markers, plant chromosomes

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Introduction. Karyotyping of plant chromosomes aims at constructing the standard karyogram and idiogram of the species and includes the analysis of chromosome morphological parameters (length, arm ratio, position of the secondary constriction) and the analysis of polymorphisms in the chromosome C- or N-banding pattern (number, size, position and staining intensity of heterochromatin bands). The process of karyotyping requires analysis of a great number of complete metaphase spreads, counting of chromosomes, pairing of homologous chromosomes and classification of chromosomes according to their type (metacentric, submetacentric, acrocentric, telocentric) and banding pattern. The described process is time-consuming and laborious. In addition, the subjectivity due to the individual experience, skills and sensitivity of the researcher often makes the description of chromosomes inconsistent. In order to facilitate and speed up this process, different systems for chromosome image analysis have been developed and successfully applied to describe chromosomes of plant species: rye [1], wheat [2], *Crepis capillaris* [3], rice [4], alfalfa [5], cotton [6], sugarcane [7] and other plants.

The wild cereal grass, Aegilops geniculata Roth (2n = 28, genome formula UM), is of breeding value as a donor of important genes for improvement of the cultivated wheat. Its chromosomes have been studied for polymorphisms in the N-banding profiles [8] aiming at the construction of the standard banded karyogram of this species. In this paper, we discuss the application of the system for human chromosome image analysis and classification CAIS [9,10] to describe the N-banded chromosome images of Ae. geniculata. The development of the system CHROMOS for processing and analysis of different species chromosome images is described. Its applicability to establish

1387

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Fig. 1. Image of *Ae. geniculata* chromosomes shown by system CAIS

cytological markers for identification of the alien chromosomes in wheat background is assessed.

Methods used for image processing and analysis. Chromosomes of 7 Ae. geniculata accessions were stained differentially by Giemsa N-banding method [¹¹] and were analysed using the system CAIS (Fig. 1). The following methods from the system CAIS were successfully implemented: threshold calculation, chromosome isolation, counting, arranging by area, calculation of chromosome length and centromere index, bands detection, construction of bands descriptions (Fig. 2). As a result, the following characteristics of chromosomes were obtained: area; length; centromeric index; bands vector; bands descriptions.

Discussion. The analysis of plant chromosomes using the system CAIS showed that certain methods for processing and analysis of human chromosome images can be





Fig. 2. Chromosomes of Ae. geniculata isolated and arranged by system CAIS

E. D. Detcheva, S. P. Landjeva



Fig. 3. Image of Ae. geniculata chromosomes shown by system CHROMOS



Fig. 4. Brightness, contrast modification and threshold determination of *Ae. geniculata* chromosome image

successfully applied to plant chromosome images. To achieve more sophisticated image editing and automation of bands matching new methods were required. On this basis, the system CHROMOS for processing and analysis of chromosome images of different species was developed (Fig. 3). In the new system, CAIS-methods for correction of centromere position and possible chromosome rotation according to the new centromere position and the methods for comparison and bands matching were improved. In addition, the following new methods for image processing were implemented: brightness and contrast modification (Fig. 4); median smoothing; threshold determination (Fig. 4); chromosome editing tools; centromere position correction. The new methods make the system more robust and flexible and more comfortable to work with. Homologous chromosomes from different accessions of *Ae. geniculata* were analysed using the system CHROMOS. This comparison allowed precise description of monomorphic and polymorphic *N*-bands. As a result, a generalized idiogram of *Ae. geniculata* is proposed (Fig. 5). The established monomorphic bands can be used as cytological markers for discrimination of the individual alien chromosomes among the set of 14 chromosome pairs as well as from the wheat chromosomes in wheat *Ae. geniculata* hybrids.

Conclusions. The new system CHROMOS gives more clear description of the chromosome N-bands (position, intensity and size) and allows more accurate comparing of banding profiles of homologous chromosomes from different accessions. This

Compt. rend. Acad. bulg. Sci., 58, No 12, 2005

1389



Fig. 5. Generalized N-banded idiogram of the wild cereal grass Aegilops geniculata Roth (2n = 28, genome formula UM) (monomorphic bands are given in black, polymorphic bands are hatched)

makes the system CHROMOS appropriate to be used for establishing of cytological markers for precise chromosome identification and detection when introduced into another genetic background.

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E. D. Detcheva, S. P. Landjeva

1390