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Chromosome Identification of Durum Wheat by Acetocarmine Wright C-banding Technicque.

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C-banding 法에 의한 Macaroni Wheat의 염색체동정

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Abstract

A combination of acetocarmine-Wright C-banding technique was utilized to identify each chromosomes in durum wheat, *Triticum durum* var. Hordeiforme (2n=4x=28 AABB). This technique elucidated qualitative and quantitative traits of the individual chromosomes in complement. Most conspicuous bands were observed at the centromere of B-genome chromosomes. Each chromosomes of A-genome had somewhat weak centromeric, proximal and terminal bands. Chromosomes 2A and 4A has a small subterminal bands. 6A is smallest and metacentric chromosome and, has two faint interstitial band. Chromosomes 1B and 6B showed satellite and constriction lage band. Short arm of 3B has three heavily interstitial bands. Both arms of chromosome 4B has a lage centromeric band and a very lage proximal band. 5B had heavily centromeric band and the long arm showed prominent two interstitial bands. Chromosomes 2B and 7B has a small terminal band of both arms.

Introduction

The introduction of the C-banding technique to plant material has allowed, in some species, identification of individual chromosomes with on unprecedented precision. Using this technique, Gill and Kimber (1974) demonstrated that the C-banding technique can be used to identify individual common wheat chromosomes. Since, by many geneticist have been studies the C-banding technique open the way for the characterization of the chromosomes and the chromosome identification of aneuploids.

The C-banding karyotype of tetraplid wheat was described by Zurabishvili et al. (1978), Seal (1982), Bebeli and Kaltsikes (1985) and Simeone, perrone and Blanco (1988). Different authors have used different materials and have designated the chromosome in their own way. The recently developed Giemsa and Leishman staining method have advantages and disadvantages in chromosome analysis. However, if the two methods are combained, the results could be more precise and dependable than those obtained from either method alone. Nakata et al. (1977) and Fujigaki and Tsuchiya (1985) demonstrated the possibility of combining acetocarmine-Giemsa staining technique in the rye chromosomes. The present study attempts to investigate the acetocarmine-Wright C-banding patterns of 14 pairs chromosomes for identification of durum wheat trisomics. Each chromosomes of A and B genome was identified correctly. The result are presented in this paper.

Materials and Methods

Seeds of durum wheat *Triticum durum* var. Hordeiforme (2n=4x=28 AABB) have been maintained by selfing in laboratory of genetics and plant Breeding, Tokyo university of agriculture.

Seeds were germinated in petri dishes at 25°C incubator. Root tips 0.5 to 1.5cm long were excised and pretreated in ice water for 16 to 18 hours at 0°C before being fixed in 3:1 alcohol-acetic acid. The C-banding technique was carried out to modification the cytological method of Fujigaki and Tsuchiya (1990). That technique was follows:

1. Stain the fixed materials in 0.6% acetocarmine for 1 to 3 hours.
2. Make squash preparations in 45% acetic acid.
3. Remove cover glass after dry ice freezing.
4. 5% Barium hydroxide treatment at 56°C for 3 minutes.
5. Rinse the slides in distilled water (pH7.0).
6. 2 X SSC treatment at 56°C for two hours.
7. Rinse the slides in distilled water.

8. Stain in 20% Wright solution diluted with phosphate buffer (pH6.8).
9. Rinse the in distilled water, air dry and mount by Permount.
10. Take photographs of the same cells as the one taken before.

Karyotype analysis were contructed from complete chromosomes showed the maximum posible banding paterns in at least 10 different cells.

Result

The C-banding karyotype of durum wheat, *Triticum durum* var. Hordeiforme is showed in Figure 1. In agreement with earier observations (Seal 1982; Bebeli and Kaltsikes 1985; Someone, Perrone and Blanco 1988) the distinctive C-banding patterns allowed the durum wheat chromosome to bear-ranged in 14 pairs. Individual A-and B-genome chromosomes were identified by comparison with their C-banding patterns in *Triticum durum* cv. Cappelli as described by Simeone et al. (1988). Dvorak (1983) concluded that chromosome 4A should be plased in the B-genome and chromosome 4B belongs to the A-genome. Naranjo, Roca, Goicoechea and Giraldez (1988) reported the genome reassignment of chromosomes 4A and 4B in common wheat. In this paper, chromosomes 4A and 4B should be designated 4B and 4A, respectively. A-genome chromosomes were less heterochromatin than B-genome chromosomes. Most conspicuous bands were observed at the centromere of B-genome chromosomes. The bands showed some variation in intensity between preparations and within same preparation. The basic banding patterns of the fourteen chromosomes are shown in figure 1 and, are described below:

1 A: No bands were observed in this chromosome.

2 A: Has a small centromeric band and one very faint proximal band and small subterminal band in the long arm.

3 A: The short arm has one proximal band. The long arm was no banded.

4 A: This chromosome has a small proximal band, a subterminal band and faint terminal band in the long arm. No bands were observed in the short arm.

5 A: This chromosome have a small proximal band in the long arm.

6 A: This chromosome is metacentric and smallest of the complement chromosomé, in one arm one interstitial band and close to it very faint band is evident in some preparations.

7 A: This metacentric chromosome has a faint terminal band in one arm.

1 B: The short arm has a lage centromeric band and one interstitial band. The satellite chromosome has one constriction lage band. In the long arm fairly lage centromeric band, a proximal band, one small interstitial band and prominent terminal band have been observed.

2 B: This chromosome has a small centromeric band. The short arm has a lage proximal band, one subterminal band and a prominent terminal band. The long arm has one faint interstitial band and a small subterminal band.

3 B: The short arm has a prominent proximal band and two prominent interstitial band. The long arm has two proximal bands, one small interstitial band and a small subterminal band.

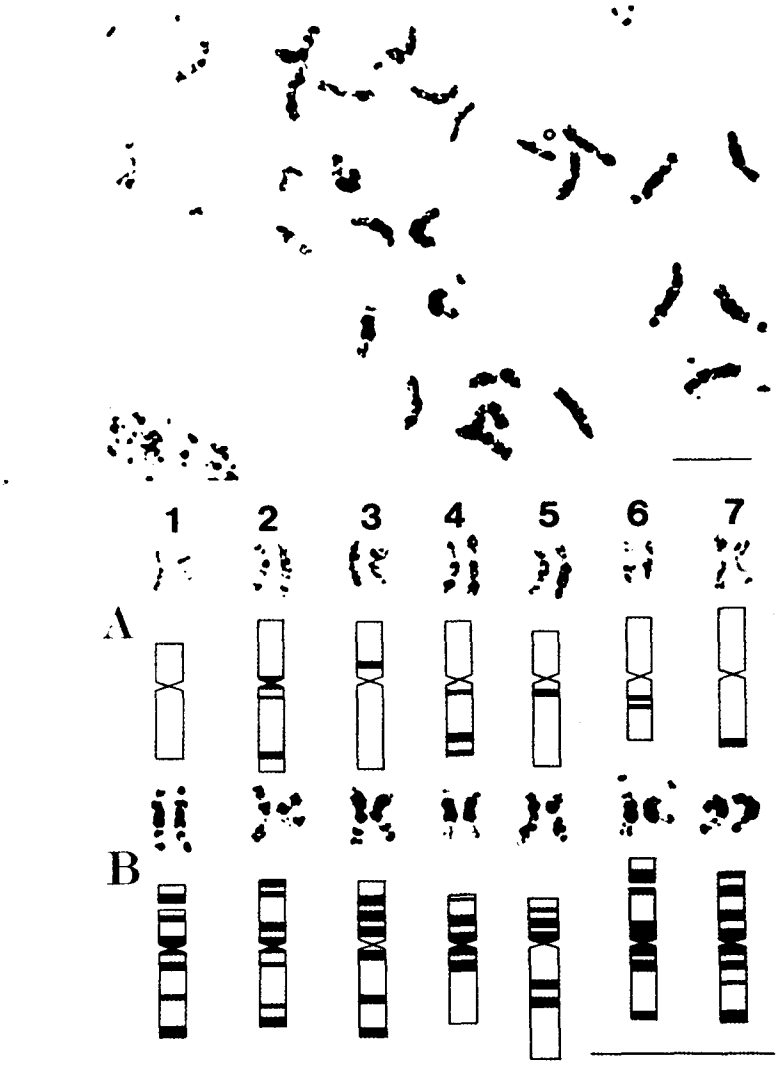

Fig. 1. The acetocarmine Wright C-banding patterns of durum wheat, *Triticum durum* var. Hordeiforme. Bar is 10 μ .

4 B: The both arms has a lage centromeric bands and a very lage proximal band and a faint subterminal band of short arm.

5 B: The short arm has a prominent proximal band, a lage centromeric band and a small subterminal band. The long arm has two prominent interstitial bands.

6 B: The short arm has a lage centromeric band, one lage proximal band and a faint constriction band. The satellite chromosome has a secondary constriction band. The long arm presents a lage centromeric band and a lage proximal band and a small terminal band.

7 B: This chromosome has centromeric bands in the both arms. The short arm has a lage proximal band, one interstitial band and one a small subterminal band. The long arm has a lage proximal band, one faint interstitial band and a prominent terminal band.

Relative lengths and arm ratio of tetraploid wheat chromosomes were reported by many authors (Chen and Gill 1983; Noda 1983; Bebeli and Kaltsikes 1985; Simeone, Perrone and Blanco 1988). According to Bebeli and Kaltsikes (1985) reported that based on the centromere position, the chromosomes were divided in to the following three categories, metacentric (arm ratio between 1 and 1.35), sub metacentric (arm ratio between 1.36 and 1.75) and subterminal (arm ratio greater than 1.76).

Discussion

Individual of the 14 chromosomes of durum wheat, *Triticum durum* var. Hordeiforme (2n=4x= 28 AABB) has its distinctive acetocarmine-Wright C-banding patterns thus allong identification. This is in agreement with earlier obserbations (Seal 1982; Simeone et al 1988) the distinctive C-banding patterns in tetraploid chromosomes to be arranged in 14 pairs. Simeone, Perrone and Blanco (1988) were reported similar to this results.

The designation of chromosome arms and bands followed the rules of the international system of human cytogenetics nomenclature (1978) and proposal made by Biekerk and Pienaar (1983) and Schlegel and Gill (1987) for C-banded chromosomes of *Triticum aestivum* cv. Chinese Spring. Chromosome banding paterns of *Aegilops* specis of A-genome have been studied by Kimber (1974), Natarazan and Sarma (1974) and Gerlach (1977). Chromosomes 2A and 3A were identified according to the classification of Endo and Gill (1984). The present results showed that chromosomes 1A, 3A, 5A, 6A and 7A into A- genome of *Triticum durum* var. Hordeiforme is contribute very little to the total amount of heterochromation. Similar results have been obtained by other author in tetarploid wheat (Simeone et al 1988). Chromosome 2A were differently from reports of other author (Seal 1982, Bebeli and Kaltsikes

1985 and Simeone et al 1988), had centromeric band and very faint proximal and a small subterminal bands of long arm. The highly heterocromatic chromosome 4A into A-genome was a small proximal band, conspicuous subterminal band and very faint terminal band in the long arm. This were same with C-banding patterns of *durum* cv. Mexicali (Bebeli and Kaltsikes 1985), cv. Cappelli (Simeone 1988), URSS-3310 of triticale (Seal 1982) and *turgidum* cv. 34 of tetraploid wheat (Hainer and Hesemann 1988). Noda (1983) was presented the paper about to chromosome mosaic of the B-genome in tetraploid wheat. Comparison of chromosome banding patterns of B-genome carry out in accordance with the karyotypic analysis of tetraploid wheat (Bebeli and Kaltsikes 1985; Simeone et al. 1988). The satellite chromosome 1B of the present material was similar to the 1B satellite of Chinese Spring (C-banding paterns by means of Seal 1982). The subterminal and terminal bands in short arm of 2B was almost the same with *durum* cv. Cappelli (Simeone et al. 1988) but different with the results of Noda (1983), Seal (1983) and Hainer and Hesemann (1988). The banding patterns of 3BS was identical to other papers, but long arm of 3B has conspicuous terminal band but differed in Chinese Spring and other tetraploid wheat where no band was found. Chromosome 4B was smallest in B-genome and had the highly centromeric band that corresponding to banding patterns of Hairynecked Viking (Seal 1982). The long arm of 4B chromosome had occasionally the faint subterminal band. The 5B of subterminal centromeric chromosome was the most intensely stained centromeric resion and have a small subterminal band of short arm, and has two conspicuous intersitial bands in long arm. The present result was the similar to banding patterns of *durum* cv. Cappelli of 5B chromosome (Simeone et al. 1988). Chromosome 6B has usually C-band on the short arm near the secondary constriction and small constriction band of satalite chromosome. Similar to results were reported in C-banded B-genome chromosomes of Cocorit and Chinese Spring of bread wheat and 6TB-059 of triticale (Seal 1982). According to Seal (1982) were chromosome arms 6BS and 7BS also showed some correspondence in C-banding patterns but the present result showed by different banding paterns. However, chromosomes 1BL and 7BL had similar to banding patterns in *Triticum durum* var. Hordeiforme.

The variation found in C-banding patterns may be useful in determining the chromosomal location of cross-over events in A and B genome chromosomes in hibrids between the various genotypes. And also, that's necessary for discrimination of chromosome in aneuploids and addition lines. Such studies may also permit a comparison of genetic map of these chromosomes with their physical dimensions (Linde-Laursen 1979).

This acetocarmine-Wright C-banding technique established the banding patterns of each chromosomes in durum wheat.

摘 要

Acetocarmire-Wright C-banding 法을 이용하여 *Triticum durum* var. Hordeiforme (2n=4X=28 AABB)의 각염색체들을 동정하였다. 또한 각각의 염색체의 성질과 특성을 해명하였다. 가장 눈에 뜨는 band는 B genome의 動原體부근에서 관찰되었다. A genome의 각 염색체는 동원체에 인접한 부분과 염색체의 말단에서 약한 band가 보여졌다. 염색체 2A와 4A는 말단 가까운곳에 band가 있으며, 6A의 염색체는 가장 작고 중부 동원체적 염색체이며, 두개의 약한 개재성 band를 갖는다. 1B와 6B염색체는 부수체가 있으며 협착이 큰 band를 보였다. 3B의 短腕은 3개의 짙은 개재성 band를 가지며, 4B 염색체의 短腕은 동원체부근과 동원체에 가까운 부분에 band가 있었다. 5B는 동원체에 짙은 band가 보이며, 長腕에서 두개의 짙은 개재성 band를 갖는다. 2B와 7B염색체는 長腕과 短腕의 말단에 band가 있었다.

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