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Heterochromatic Knob Number and Karyotype in Korean Indigenous Maize

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韓國 在來種 옥수수 染色體의 Heterochromatic Knob 數와 核型

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ABSTRACT : A Giemsa C-banding method was used for the identification of somatic chromosomes and heterochromatic knob positions in Korean indigenous maize(*Zea mays* L.). Total of 10 inbred stocks were examined and their knob numbers ranged from 6 to 12. In comparison of homologous chromosomes of two stocks of Waesungri and PI213749, arm ratios and relative length of chromosomes were different between genotypes. In comparison of arm ratios, all the homologous chromosomes except chromosome 2 were different each other. In comparison of relative length of chromosomes, that of chromosome 1 in Waesungri and PI213749 was 223.22 and 192.03 respectively. The relative length of homologous chromosomes in Waesungri were generally lager than those of PI213749. A C-banded diagram showing knob positions, arm ratios and relative length of chromosome could be used as a good tool to compare the characteristics of chromosomes of Korean indigenous maize stocks.

Key words : Giemsa C-banding, Heterochromatic knob, Maize, Inbred stock.

The chromosomes of maize possess a number of distinguishing features used by cytologist to identify a chromosome to the other. Among them dark staining spot known as heterochromatic knob is one of the important features. These knobs have been shown to appear at certain points on certain chromosomes and knob number is constant for any individual plant. Therefore, it can be used as one criterion in determining the relationship of various kinds of maize.

Heterochromatin is cytologically recognizable in interphase and early stages of mitosis and meiosis as highly condensed regions of chromatin. Following certain pretreatments, heterochromatin can be visualized with Giemsa stain as knobs or bands in mitotic or meiotic metaphase preparations[2]. The preferential staining ability is probably due to a DNA-protein interaction involving protein that are specifically associated with the heterochromatin[7].

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Heterochromatin is known to consist of highly repetitive DNA, to replicate late, and to be genetically silent[10]. The presence of heterochromatic blocks in the genome has been shown to influence both chiasma formation and genetic recombinationin in *Zea mays* L.[14]. In *Zea*, furthermore, chromosome knob has been used to suggest evolutionary relationship between different taxa of this genus[12,16].

While C-banded somatic metaphases have revealed the presence of distal bands that correspond with knobs[8,11,15,17,18], cytogenetic studies such as chromosome knob number and position, diagramatic representation of the band position, and chromosome arm ratio on Korean indigenous maize have been few.

In the present study, to know the knob numbers of Korean indigenous maize, the chromosome morphology, 10 inbred stocks were investigated. Also arm ratio and relative length of chromosome of two representative stocks which had different knob positions on the chromosome were measured, and diagrams revealing the knob positions, arm ratio and relative length of chromosome were presented.

MATERIALS AND METHODS

Ten inbred stocks of Korean indigenous maize were used for this study. The stocks and collection sites are shown in Table 1.

A modified Leishman C-banding employed by Bennet et al.[2] on rye(*Secale cereale* L.) was adopted as the basic treatment. The C-banding procedure previously described by Lee et al.[11] was used. Chromosome were investigated with phase-contrast microscope (Olympus, B202). Identification of chromosomes was referred to Chen[5], Horn & Walden[9].

Chromosome length was determined from photographic prints magnified to ×5,000, with a pair of calipers. Arm ratio showing short arm versus long arm and chromosome relative length estimated by percentage of the length of chromosome 10 were calculated with 10 samples.

For the diagrammatic representation of each chromosome of the complement, homologous chromosome pairs were compared 10 times, respectively.

Table 1. Stocks used for the study and their collection sites

Stock	Collection site	Stock	Collection site
Waesungri	Nonsan	Won	Jewon
PI213749	Sanchung	Dangin	Dangin
IK-1	Euisung	Sung	Bosung
IK-2	Iri	Sinkihong	Nonsan
IK-3	Uleuing	Pop	Yangsan

RESULTS AND DISCUSSION

Ten stocks of Korean indigenous maize were stained by Giemsa C-banding method and heterochromatic knobs were investigated under phase-contrast microscope. The results are shown in Table 2.

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Table 2. Numbers of heterochromatic knob in 10 Korean indigenous maize stocks

Stock	Knob number	Stock	Knob number
Waesungri	10	Won	8
PI213749	10	Dangin	6
IK-1	12	Sung	8
IK-2	12	Sinkihong	8
IK-3	8	Pop	12

materials ranged from 6 to 12, and average number was 9.4. The knob numbers on the chromosome vary from stock to stock, and the results show the same tendency to Central and South American maize, the United States maize and Italian maize.

Reeves[13] collected maize seeds from Central and South America, and investigated chromosome knob. The knob numbers ranged from 0 to 9 with the greatest number in Guatemala, the lowest in Ecuador. The results showed that the knob numbers varied to the collection countries.

Brown[4] reported that the knob numbers of the United States maize were from 0 to 12 with the greatest number being concentrated in the Southern Dents, the lowest in the Northern Flints, and high numbers of knobs were positively correlated with the following external features of the ear and plant; high row numbers, denting, absense of husk leaves, many seminal roots, and irregular rows of kernels.

Bianch et al.[3] reported that the number of Italian maize ranged from 0 to 8(average number: 2.7) with the greatest number in Northern Italy(average number: 3.28), the lowest number in Southern Italy(average number: 1.7).

In this study, Korean indigenous maize had more knobs than those of the United States maize, Central and South American maize or Italian maize. The relationship between knob number and collection site is not clear. To find the relationship, more data are needed.

Giemsa C-banding metaphase plates of Waesungri and PI213749 are presented in Fig. 1. Five pairs of the chromosome component show dark and heterochromatic knobs and one pair has a satellite.

The chromosomes 3, 4, 5, 7 and 10 in

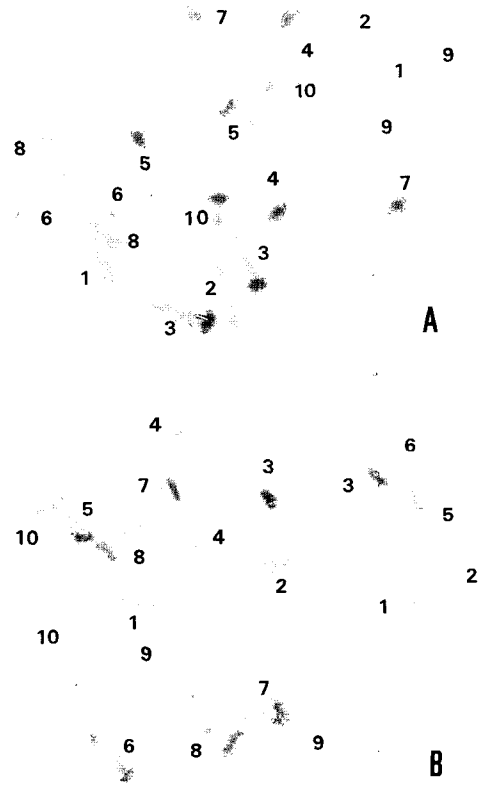

Fig. 1. Giemsa stained somatic metaphase of Waesungri(A) and PI213749(B).

Waesungri show terminal heterochromatic knobs, whereas the chromosome 3, 5, 6, 7 and 8 in PI213749 show terminal heterochromatic knobs. The knobs of the chromosome 4 and 10 in Waesungri and the knobs of the chromosome 6 and 8 in PI213749 are unique to each stock.

Table 3 shows the values of arm ratio and relative chromosome length expressed as a percentage of the length of chromosome 10 in Waesungri and PI213749. As shown in Table 3, the values of chromosome arm ratios vary from 1.21(chromosome 1, 9) to 1.72(chromosome 7) in Waesungri and from 1.25(chromosome 2) to 2.37(chromosome 5)

Table 3. Arm ratio and relative chromosome length expressed as percentage of chromosome 10 in Waesungri and PI213749

Chromosome #	Waesungri		PI213749	
	Arm ratio (n=10)	Chromosome length	Arm ratio (n=10)	Chromosome length
1	1.21	223	1.38	192
2	1.25	193	1.25	161
3	1.55	178	1.48	158
4	1.46	161	1.42	153
5	1.58	147	2.37	141
6	1.46	145	1.50	140
7	1.72	125	1.83	137
8	1.58	124	1.36	120
9	1.21	115	1.39	110
10	1.54	100	1.75	100

in PI213749. The comparisons of the values of arm ratios between homologous chromosomes in the two stocks do not show the same tendency.

In the values of relative length of chromosomes, those in Waesungri generally vary more than those in PI213749.

Above results show that the arm ratios and the relative length of homologous chromosomes are different between the genotypes, and show the same tendency to the report of Aguiar-Perecin and Vosa[1].

Idiograms representing the somatic maize karyotype of Waesungri and PI213749 are shown in Fig. 2. These idiograms show arm ratios, relative length of chromosomes and knob positions. As can be seen in these idiograms, 4 terminal knobs(4S, 5L, 7L and 10L), 1 subterminal knob(3L), 1 heterochromatic band of centromere(5) and 1 satellite (6) are shown in Waesungri(2A), whereas 4 terminal knobs(5L, 6L, 7L and 8L), 1

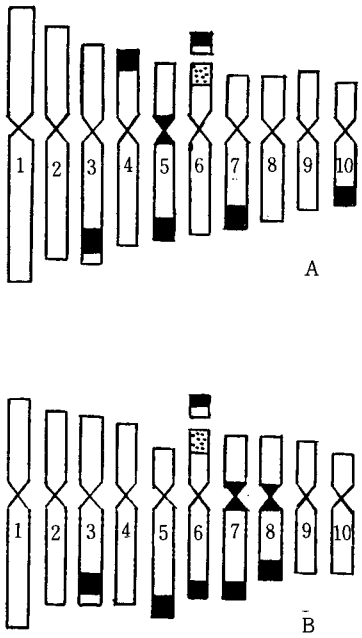

Fig. 2. Diagram of the C-banded maize somatic Karyotype of Waesungri(A) and PI213749(B).

subterminal knob(3L), 2 heterochromatic bands of centromere(7, 8) and 1 satellite(6) are shown in PI213749.

Filion & Walden[6] mentioned that variations of arm ratios could be observed in metaphase chromosomes of different maize stocks, but no evidence was shown of correlation between arm length and band width. Aguiar-Perecin and Vosa[1] mentioned that the C-banded chromosome diagram could be considered illustrative of band positions in maize somatic chromosomes and might be useful in the future standardization of maize somatic cytogenetics.

Above result shows that knob position and heterochromatic band of centromere as well as arm ratio and relative length of chromosome are different between the genotypes. And the result shows that the C-banded

chromosome diagram can be considered illustrative of band positions, arm ratios and relative chromosome length in maize somatic chromosomes.

摘 要

한국 재래종 옥수수의 염색체 특성을 알아보기 위하여 재래종 옥수수 10개의 自殖系統을 C-banding법으로 염색하고 염색체상에 존재하는 heterochromatic knob 수를 조사한 결과는 다음과 같았다.

1. Knob수는 6~12개이었고 평균 9.0개이었으며 계통별로 차이가 있었다.
2. 염색체의 장완과 단완의 비율, 상대적 길이 등을 비교해 보기 위하여 Waesungri와 PI213-749 두 계통을 조사해 본 결과 계통별로 차이가 있었다. 즉, 장완과 단완의 비율은 2번 염색체의 경우에만 1.25로서 동일하였고 다른 염색체의 경우는 모두가 다르게 나타났다. 염색체의 상대적 길이는 일반적으로 Waesungri에서 크게 나타났는데 1번 염색체의 경우 Waesungri에서는 223이었고 PI213749에서는 192이었다.
3. 염색체의 상대적 길이, 장완과 단완의 비율, 그리고 knob의 위치 등을 나타내는 모식도를 통하여 두 계통의 염색체 특성을 보다 명확하게 비교할 수 있었다.

LITERATURE CITED

1. Aguiar-Perecin de M.R.L and Vosa C.G. 1985. C-banding in maize; Ⅱ. Identification of somatic chromosomes. Heredity 54:37-42.
2. Bennet M.D, Gustafson J.P and Smith B. 1977. Variation in nuclear DNA in the genus *Secale*. Chromosoma 61:149-176.
3. Bianch A, Chatnekar M.V and Ghidoni A. 1963. Knobs in Italian maize. Chromosoma 14:601-617.
4. Brown W.L. 1949. Numbers and distribution of chromosome knobs in United States maize. Genetics 34:524-536.
5. Chen C.C. 1969. The somatic chromosomes of maize. Can. J. Genet. 11:752-754.
6. Filion W.G and Walden D.B. 1973. Karyotype analysis: The detection of chromosomal alternations in the somatic karyotypes of *Zea mays* L. Chromosoma 41:183-194.
7. Gendel S.M and Fosket D.E. 1979. The role of chromosomal proteins in the C-banding of *Allium cepa* chromosomes. Cytobios. 22:155-168.
8. Hadlaczky G.Y and Kálmán L. 1975. Discrimination of homologous chromosomes of maize with Giemsa staining. Heredity 35:317-374.
9. Horn J.D and Walden D.B. 1971. Fluorescent staining of euchromatin and heterochromatin in maize(*Zea mays*). Can. J. Genet. Cytol. 13:811-815.
10. Hsu T.C. 1975. A possible function of constitutive heterochromatin: the bodyguard hypothesis. Genetics 79(suppl) :137-150.
11. Lee I.S, Choe B.H and Gustafson J.P. 1996. C-banding pattern of mitotic chromosome in Korean indigenous maize. Korean J. Crop Sci. 41(4):429-433.
12. Mastenbroek I, Cohen C.C and de Wet J. M.J. 1981. Seed protein and seedling isozyme pattern of *Zea mays* and its closest relatives. Biochem. Syst. Ecol. 9:179-183.
13. Reeves R.G. 1944. Chromosome knobs in relation to the origin of maize. Genetics 29:141-147.
14. Rhoades M.M. 1978. Genetic effects of

heterochromatin in maize. *In* Maize breeding genetics. *Edited by* Balden D. B. Wiley. New York. pp. 641-671.
15. Sachan J.K.S and Tanaka R. 1977. Variation in pattern of C-banding in *Zea* chromosomes. Nucleus 20:61-62.
16. Smith J.S.C, Goodman M.M and Lester R.N. 1981. Variation within teosinte. I. Numerical analysis of morphological data. Econ. Bot. 35:187-203.
17. Vosa C.G and Aguiar-perecin M.L.R.de. 1972. New techniques for knob detection in mitotic chromosomes in maize and teosinte. Maize Genet. Coop. Newsl. 46: 165-167.
18. Ward E.J. 1980. Banding patterns in maize mitotic chromosomes. Can. J. Genet. 22:61-67.