Three classes of spots (A, B and C) were compared on the basis of the location of breaks (distal or proximal to wx):

Class	Number of cells/spot	Number of spots examined	Loss of C-I Bz (break distal)	Loss of C-I Bz Wx (break proximal)
A	9-25	100	39	61
B	30-100	100	27	73
C	over 400	90	29	61

The spot size apparently was not affected by break location. When the phenotype of the spots was recorded for the three markers, spontaneous breakage frequencies were found to correspond satisfactorily to the physical distance of the relevant loci as indicated by the cytological map reported by Fabergé (1956). The data are summarized as follows:

Spontaneous breakage events in the short arm of chromosome 9
(SL = single losses; Cy = B-F-B cycles)

Region	I distal to C		II C-I-Bz		III Bz-Wx		IV proximal to Wx		
Breakage event	SL	Cy	SL	Cy	SL	Cy	SL	Cy	Total
Observed frequencies	—	14	6	0	34	15	143		212
Totals in regions II to IV			6		49		143		198
Expectations on the basis of physical distances			11.6		55.3		131.1		198

A comparison of the frequency of isolated spots (single losses) with the frequency of clusters of spots (B-F-B cycles) shows that a high proportion of spontaneous breaks can yield stable ends. Breaks induced in this region by UV and X-rays, according to Fabergé (1956), yielded no stable terminal deficiencies. The same author (Genetics 44:280-285, 1959), found that α particles could yield about 35% stable terminal deficiencies.

A. Ghidoni and N. Pogna

Giemsa staining of heteropycnotic regions in maize chromosomes — The study of longitudinal differentiation of chromosome regions has made considerable progress with the employment of denaturation-renaturation-Giemsa staining techniques (Pardue and Gall, Science 168:1356, 1970; Arrighi and Hsu, Cytogenetics 10:81-86, 1971). When these techniques were applied to maize chromosomes (Vosa et al., MNL 46:165-167, 1972; Sartori and Ting, Amer. J. Bot. 61,5 suppl.:63, 1974, abst.)

a correspondence was found between main Giemsa bands and knob locations. Centromeric bands were also observed with one of these methods (Filion, MNL 48:150-152, 1974).

The present investigation concerns a comparative study of Giemsa bands in mitotic metaphase chromosomes, and of chromocenters (condensed chromatin bodies) in resting nuclei, and the relation of these to known knob locations and to the presence of B chromosomes, in different maize stocks. The material is described below:

Stock 1: the inbred Black Mexican sweet corn, with and without B chromosomes. Plants with 0-4 B's were analyzed in the present study. This stock is nearly knobless, except for a medium-small size knob in position 6Ld.

Stock 2: a tester, outcrossed to related material and backcrossed to the same tester. Homozygous knobs were present in 4L and 8L and a heterozygous knob in 6L.

Stock 3: F_1 between a K10 stock (marked R-st) and a r tester. Homozygous knobs 4L, 6L; heterozygous knobs 2L, 8L, 10L (abn.).

Stock 4: inbred line KYS. All knobs homozygous in the following positions: 5L, 6L, 7L and 9St.

Stock 5: a r tester with the background of inbred line W22. All knobs homozygous in the following positions: 2L, 4L, 7L, 8L and 9St.

All of these stocks carry a knob-like heteropycnotic structure adjacent to the Nucleolus Organizer region (N.O.) in the short arm of chromosome 6.

Root tips were pre-treated with a saturated solution of α-bromonaphthalene or with .6% colchicine aqueous solution, for 5 to 6 hrs, and fixed overnight in 3:1 ethanol-acetic mixture. Then, a modified BSG (Barium hydroxide/Saline/Giemsa) technique described by Sumner et al. (Nature New Biol. 232:31-32, 1971) and by Vosa and Marchi (Nature New Biol. 237:191-192, 1972) was applied for the present study.

Chromocenters stained by Giemsa were observed in resting nuclei, and Giemsa bands in well-condensed metaphase chromosomes of root tips. The results are summarized on the following page.

The number and size of Giemsa bands and the number and size of chromocenters are correlated. Giemsa bands and chromocenters on one side, and knob numbers on the other side are also correlated. Some distortion was caused in the morphology of metaphase chromosomes by the denaturation treatment, but when individual chromosomes could be reasonably identified, Giemsa bands were found consistently at knob locations. Larger knobs were always observed as thick bands, and smaller

knobs as thinner bands, sometimes reduced to single granules. K10 invariably showed, besides a large chromocenter in resting nuclei, a thick band in metaphase chromosomes at the expected location. Occasionally, thin bands were observed in other locations such as telomeres, or close to centromeric regions.

Material	No. of B's	Average No. of chromocenters large or medium size	small size	Average No. of Giemsa bands thick	thin	No. of knobs* large or medium size	small size
Stock 1 (Black Mex.)	0	0	1-2	0	2	0	2
Stock 1 (")	1	0	1-3	0	2	0	2
"	2	0	2-4	0	2	0	2
"	3	0-1	2-3	0	2	0	2
"	4	0-1	2-3	0	2	0	2
Stock 2 (a/)	0	4	3-4	5	3-4	4	1-2
Stock 3 (K10/)	0	5-6	3-4	6	3-5	6	4
Stock 4 (KYS)	0	4	3-4	4	2-4	4	4
Stock 5 (W22)	0	8	2-4	8	2-4	8	4

*The knob-like structure adjacent to the N.O. is not included.

No large chromocenters were observed in the knobless stock with 0-2 B's. Occasionally, these were found in presence of 3-4 B's. The absence of large chromocenters in the presence of B chromosomes was also noted by D. T. Morgan, Jr. (J. Hered. 34: 194-198, 1943).

B chromosomes did not show prominent bands as a rule. A thin band was often observed near the centromeric region of the B chromosome; two more thin bands were occasionally observed in the median and/or in the distal portion of this chromosome.

A thin band was often observed in the distal tip of the satellite region of chromosome 6. Occasionally, a thin band was observed close to the N.O. region.

These observations suggest that the heterochromatin of knobs may differ from the heterochromatin of distal portions of the B chromosomes, although the two heterochromatin types appear indistinguishable by the conventional staining techniques. In this regard, the knob-like structure at the N.O. region also seems to differ from typical knobs of other chromosome regions.

A. Ghidoni, E. Sparvoli,* and G. Broggio

*Istituto di Botanica, Università di Milano