Meiotic Drive of Chromosomal Knobs Reshaped the Maize Genome

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ABSTRACT

Meiotic drive is the subversion of meiosis so that particular genes are preferentially transmitted to the progeny. Meiotic drive generally causes the preferential segregation of small regions of the genome; however, in maize we propose that meiotic drive is responsible for the evolution of large repetitive DNA arrays on all chromosomes. A maize meiotic drive locus found on an uncommon form of chromosome 10 [abnormal 10 (Ab10)] may be largely responsible for the evolution of heterochromatic chromosomal knobs, which can confer meiotic drive potential to every maize chromosome. Simulations were used to illustrate the dynamics of this meiotic drive model and suggest knobs might be deleterious in the absence of Ab10. Chromosomal knob data from maize's wild relatives (*Zea mays* ssp. *parviglumis* and *mexicana*) and phylogenetic comparisons demonstrated that the evolution of knob size, frequency, and chromosomal position agreed with the meiotic drive hypothesis. Knob chromosomal position was incompatible with the hypothesis that knob repetitive DNA is neutral or slightly deleterious to the genome. We also show that environmental factors and transposition may play a role in the evolution of knobs. Because knobs occur at multiple locations on all maize chromosomes, the combined effects of meiotic drive and genetic linkage may have reshaped genetic diversity throughout the maize genome in response to the presence of Ab10. Meiotic drive may be a major force of genome evolution, allowing revolutionary changes in genome structure and diversity over short evolutionary periods.

MEIOTIC drive is found in many taxa, where it ordinarily causes the preferential segregation of small regions of the genome (Lyttle 1991). Meiotic drive is normally portrayed as arising either from the evolution of a drive locus and linked modifiers or from "a direct consequence of intrinsic chromosome structure" (Lyttle 1991). Maize meiotic drive does not readily fall into either category. Our analysis of existing data suggests that meiotic drive is the result of rapid and extensive evolution of drive loci on the Abnormal 10 chromosome (Ab10) and multiple unlinked cytological features known as knobs. Instead of viewing knobs as an intrinsic structure of the genome, we test the view that knobs are the products of a genome evolving in response to meiotic drive.

Meiotic drive in maize results from an interaction between Ab10 and knobs (Rhoades 1942). Chromosomal knobs are large blocks of cytologically visible heterochromatin (McClintock 1929), which can be found at 34 distinct locations spread across all 10 chromosomes of maize and teosinte (Kato 1976; Figure 1). Knobs consist of thousands to millions of tandem 180- and 350-bp repeats, and they may account for as much as 8% of the genome (Peacock *et al.* 1981; Ananiev *et al.* 1998b). Most genetic and evolutionary data on knobs were collected with light microscopes, and a small knob as defined by this methodology may have roughly 20,000 repeats (Peacock *et al.* 1981). Most 180-bp repeats are found in visible knobs; however, some 180-bp repeats are distributed and isolated among all chromosomes (at least 100 per chromosome; Ananiev *et al.* 1998a). This research focuses on the evolution of visible knobs (>20,000 repeats) rather than the evolution of isolated knob repeats, as these visible knobs form the functional neocentromeres necessary for meiotic drive.

The 180-bp knob sequence has one 68-bp region with similarity to sequences mapping to maize centromeres (Burr *et al.* 1992; Alfenito and Birchler 1993). The 350-bp repeat has two short segments (<30 bp) with homology to the 180-bp sequence (Ananiev *et al.* 1998b). Neocentromeric activity is observed only when Ab10 is present and thousands of these repeats form a knob (Rhoades 1952; Dawe and Cande 1996). The origin and original function of the knob sequences is unclear. Centromeric origin of knobs is suggested because they contain one 68-bp motif characteristic of maize sequences that map to centromeres. However,

This manuscript is dedicated to Eleanore Small Buckler (1943–1998), who introduced her sons and thousands of Virginia school children to the wonders of science.

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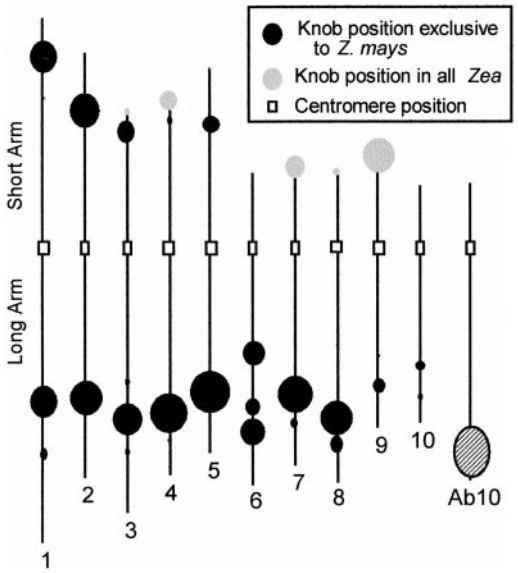

Figure 1.—Chromosomal knob positions in *Z. mays* ssp. *parviglumis* and ssp. *mexicana* (Kato 1976). The area of the circle is proportional to the size and frequency of the knob (knob index). The black internal knobs are unique to populations of *Z. mays* ssp. *parviglumis*, *mexicana*, and/or *mays*, while the gray terminal (telomeric) knobs are found in all Zea species. Ab10's chromosomal knob is larger than representation in this figure would allow.

subtelomeric origin is supported phylogenetically, because at least three consecutive outgroups to maize have visible knobs only at their telomeres (Randolph 1955; Kato 1976; Kato and Lopez 1990).

Abnormal 10 is an uncommon version of chromosome 10 that has an extended, rearranged long arm, a large knob, and tightly linked factors that cause segregation distortion (Rhoades and Dempsey 1985; Dawe and Cande 1996). When Ab10 is present, meiotic drive is observed in megasporogenesis for Ab10 and for any other chromosome heterozygous for a knob (Rhoades 1942). The average knobbed chromosome of the heterozygous pair preferentially segregates to 70% of the viable megaspores instead of the expected 50% (Rhoades 1942), although preferential segregation varies from 59 to 82% for different knob sizes and loci (Longley 1945). Preferential segregation occurs when crossing over between the knob and centromere produces a heteromorphic dyad (Rhoades 1952). The chromatids bearing the knobs are pulled toward the spindle poles in meiosis I and eventually toward the outermost megaspores (Rhoades 1952; Dawe and Cande 1996), which results in the two outermost megaspores having the knobbed chromosomes. The chalazal (basal) megaspore will become the gametophyte and produce gametes (Bedinger and Russell 1994). The degree of preferential segregation is positively associated with the size of the knob; *e.g.*, the knobs on 9S exhibit the following levels of preferential segregation: a large knob, 69%; a medium knob, 65%; and a small knob, 59% (Kikudome 1959). Additionally, when a locus is heterozygous for knobs of different sizes, the larger knob will exhibit preferential segregation over the smaller knob (Kikudome 1959). For example, when chromosome 9S is heterozygous for a small knob and a medium knob, the medium knob is preferentially segregated 65–70% of the time (Kikudome 1959).

On the basis of observations from a few maize races, it appears that races with Ab10 may have more knobs (Longley 1945) and that maize knobs are nonrandomly located along chromosomal arms (Longley 1939). These observations suggest that selection might play an important role in knob evolution. We hypothesize that chromosomal knobs are the result of meiotic drive and suggest the following model: (1) A rearrangement in chromosome 10 resulted in complete linkage between repetitive knob DNA and factors that ensure the segregation of the knob to the distal megaspores of the meiotic tetrad, creating a meiotic drive system that favored its own segregation (Ab10). (2) Ab10's frequency was determined by the balance between preferential segregation and the fitness of the permanently linked genes in Ab10's rearrangement (Rhoades and Dempsey 1985). (3) In populations where Ab10 reached modest frequencies, strong selection would favor knobs on multiple chromosomes. Because larger knobs outcompete smaller knobs for preferential segregation (Kikudome 1959), meiotic drive would favor ever larger knobs. (4) Knobs should also develop at chromosomal positions that allow high frequencies of preferential segregation. Two competing constraints could affect knob position. Recombination between the knob and the centromere is necessary to produce the heteromorphic dyad, which sets the stage for meiotic drive. This process favors knobs in telomeric positions. Counterbalancing this, microtubule interactions of centromeres with the knob neocentromeres must be coordinated so that both regions are pulled toward the same pole, a constraint that may favor knobs closer to the centromere (Yu *et al.* 1997; discussed below). (5) Once an efficient and functional knob developed on a chromosomal arm, it would be unlikely that another knob will develop on the same chromosomal arm. (6) Knobs must be slightly deleterious, which prevents them from being fixed in most populations.

This hypothesis conflicts with evidence that suggests most repetitive DNA is either neutral or slightly deleterious (Charlesworth *et al.* 1994). A balance between amplification mechanisms and losses through deletion, drift, and unequal crossing over dominates the population dynamics of most repetitive DNA. The permissive hypothesis posits that selection is not responsible for the accumulation of repetitive DNA; rather, repetitive and junk DNA accumulate in regions of low recombination as long as the fitness costs of replicating repetitive DNA are low. The permissive hypothesis therefore predicts that repetitive DNA should be prevalent in chromosomal regions with little recombination and in popula-

TABLE 1

Predictions and tests of the meiotic drive and permissive hypotheses for knob evolution

Tests	Knob evolution hypotheses	
	Meiotic drive	Permissive
Ab10 frequency and knob index correlation	+**	NP
Intraspecific variability in knob size	High*	Low
Knob index and population size correlation	+*	—
Knob position within chromosomal arm	Distal**	Regions with low recombination
Knob distribution among chromosomal arms	Repulsed**	Random

NP, the hypothesis makes no prediction for the test.
* Predictions consistent with observations from *Z. m.* ssp. *parviglumis* and *mexicana*.
** Predictions with statistical support ($P < 0.05$).

tions with small sizes (Charlesworth *et al.* 1986; Stephan 1987). The permissive hypothesis of knob evolution suggests that chromosomal knobs are the products of repetitive DNA dynamics without the positive selection of meiotic drive.

The fitness cost of repetitive DNA is an important consideration for either of these hypotheses. The meiotic drive hypothesis provides a positive selection regime (Ab10) that could favor knob DNA even if knob DNA were normally deleterious. The permissive hypothesis suggests repetitive DNA is either neutral or only slightly deleterious. Experimentally evaluating selection on maize knobs, while controlling for closely linked genes, will require high-resolution maps of maize chromosomes and knobs. These resources will only become available over the next 3 to 5 years. Instead, this study models how knob fitness reduction and meiotic drive could balance to produce knob polymorphisms. We also consider how the environment may modify knob and Ab10 fitness and may explain the prevalent knob-environment correlations (Poggio *et al.* 1998).

We examine the chromosomal evidence in light of both the meiotic drive and the permissive hypotheses (Table 1). The meiotic drive hypothesis predicts that increases in Ab10 frequency should result in more frequent and larger knobs, Ab10 frequency will be affected by the environment, knobs should be in optimal chromosomal positions for meiotic drive, and knobs should have a repulsed distribution caused by competition between knobs. The permissive hypothesis predicts that knob repetitive DNA will proliferate in regions with little recombination and in small populations. With chromosomal data from the maize's sister taxa, *Zea mays* ssp. *parviglumis* and *mexicana*, we used phylogenetic comparisons to show that changes in Ab10 frequency are probably the most important determinant of knob frequency and size, although environment may affect knob evolution. The cytological distribution of knobs supports the conclusion that knobs are the products of meiotic drive and not of permissive evolution.

METHODS

Model of meiotic drive hypothesis: We used two models of meiotic drive to illustrate the interactions between Ab10 and knobs. Viability selection, meiotic drive, recombination, and gametic selection are modeled without drift; mutation, unequal crossing over, and transposition were not modeled. Multiple starting allele frequencies were tested to avoid local minima or maxima, and recursions of the model were continued until allele frequencies changed $<1 \times 10^{-6}$ per generation.

Model 1 describes the meiotic drive of Ab10 in female gamete production, the meiotic drive effects on an unlinked knob, and the gametic selection against Ab10 in male pollen (Table 2). KA is a gamete with a knob and Ab10, while ka is a gamete with no knob and normal 10. *vKK* is viability reduction of knob homozygotes, *vAA* is the viability reduction of Ab10 homozygotes, *vA* is the viability reduction of individual Ab10 chromosomes, and *vK* is the viability reduction of individual knobbed chromosomes. *d* is the meiotic-drive-based segregation distortion of knobbed chromosomes in the presence of Ab10 ($d = 0.5$ is Mendelian segregation); *g* is the gametic selection against Ab10 in pollen.

Model 2 describes the competition of two linked chromosomal knobs (Table 3), where the chromosomal order is centromere, small knob, and then large knob. When both a large and a small knob are on the same heteromorphic dyad, only the large knob exhibits meiotic drive. The model assumes a constant frequency of Ab10 in the population. Table 3 only describes megasporogenesis when Ab10 is present, while megasporogenesis without Ab10 and microsporogenesis follow Mendelian segregation. *vLL* is viability reduction against large-knobbed homozygotes, *vL* is the viability reduction of individual large-knobbed chromosomes, *vSS* is viability reduction against small-knobbed homozygotes, *vS* is the viability reduction of individual small-knobbed chromosomes, r_1 is the recombination distance between the centromere and the small knob, r_2 is the recombination distance between the small knob and the large knob, and *m* is the probability that a given heteromorphic dyad will result in the knobbed chromosome ending up in the polar megaspore. The double-stranded crossovers resulting from single crossovers in r_1 and r_2 were modeled, but double crossovers within r_1 or r_2 were not modeled because of the minor effects on the total model and because chromosomal interference would reduce the likelihood of such a situation.

Sampling: Fifty-one populations of *Z. mays* ssp. *parviglumis* and *mexicana* were scored for Ab10, knobs, B chromosomes, and altitude by Kato (1976) using light microscopy. Knobs

TABLE 2

Model for Ab10 and an unlinked chromosomal knob

Genotype	Viability	Female gametes produced				Male gametes produced			
		KA	Ka	kA	ka	KA	Ka	kA	ka
KKAA	$(1 - vKK)(1 - vAA)\ (1 - 2vK)$	1	0	0	0	$1 - g$	0	0	0
KKAa	$(1 - vKK)(1 - 2vK)$	d	$1 - d$	0	0	$\frac{1}{2}(1 - g)$	$\frac{1}{2}$	0	0
KKaa	$(1 - vKK)(1 - 2vK)$	0	1	0	0	0	1	0	0
KkAA	$(1 - vAA)(1 - vK)$	d	0	$1 - d$	0	$\frac{1}{2}(1 - g)$	0	$\frac{1}{2}(1 - g)$	0
KkAa	$1 - vK$	d^2	$d(1 - d)$	$d(1 - d)$	$(1 - d)^2$	$\frac{1}{4}(1 - g)$	$\frac{1}{4}$	$\frac{1}{4}(1 - g)$	$\frac{1}{4}$
Kkaa	$1 - vK$	0	$\frac{1}{2}$	0	$\frac{1}{2}$	0	$\frac{1}{2}$	0	$\frac{1}{2}$
kkAA	$1 - vAA$	0	0	1	0	0	0	$1 - g$	0
kkAa	1	0	0	d	$(1 - d)$	0	0	$\frac{1}{2}(1 - g)$	$\frac{1}{2}$
kkaa	1	0	0	0	1	0	0	0	1

Entries in the table are the relative fitness for various gametes and plant genotypes.

in telomeric positions are classified as terminal, while all the other interstitial knobs are classified as internal. Because the two cytologically differentiable types of Ab10 (I and II) both produce meiotic drive, their frequencies were combined in this analysis (Rhoades and Dempsey 1985). Type I is found in maize, while Types I and II are found in *Z. mays* ssp. *parviglumis* and *mexicana*. A knob index was calculated to reflect the average number of knob repeats index at each of the 34 chromosomal positions in each population (Bretting *et al.* 1987). Knobs were cytologically scored as absent, small, medium, or large and given weightings of 0, $\frac{1}{3}$, $\frac{2}{3}$, and 1, respectively. The knob index for a population is the sum over all size classes of the knob frequency in a population times the weighting for the size of the knobs.

The environment may be related to repetitive DNA fitness (Poggio *et al.* 1998). Plants at high altitudes have short growing seasons, rapid developmental rates, and take fewer days to flower. Days to flowering and altitude have been measured for 15 central Mexican populations (Wilkes 1967), and regression between altitude and time to flowering explained much of the variation ($r^2 = 62\%$, $P < 0.001$). For the 49 populations found in central Mexico, altitude is closely related to the length of the growing season; two populations in northern Mexico (Nobogame) are outliers in this regression, which is otherwise based on central Mexican populations. To account for latitudinal effects on time to flowering, we used the regression equation and data on Nobogame population time to flowering (Wilkes 1967) to derive a central Mexican altitude proportional to their flowering time and environment (altitude is 3487 m).

Phylogenetic contrasts: The population frequencies of Ab10 and knobs should not be directly correlated, because the number of independent comparisons depends on the populations' shared evolutionary history (Felsenstein 1985). By using a phylogenetic tree and by calculating the differences in knob and Ab10 frequencies between the immediate descendants of an ancestor, we can compare the changes in knob and Ab10 frequencies that occurred during these periods of independent evolution. Phylogenetic independent contrasts (Purvis and Rambaut 1995) were used to evaluate correlations between Ab10, altitude, and knob indexes. This hierarchical phylogenetic approach is appropriate for these wild *Z. mays* populations because analysis of isozyme data suggested dispersal, rather than subsequent hybridization, was most important in determining the population structure of ssp. *parviglumis* and *mexicana* (E. S. Buckler, T. P. Holtsford and J. F. Doebley, unpublished results). The phylogeny [(Zmp-West Central Balsas [13], Zmp-East Central Balsas [5]), (Zmp-South Balsas [5], (Zmmx-Nobogame [2], (Zmmx-Chalco [14], Zmmx-Central Plateau [12]))))] was based on analyses of isozyme and ribosomal data (Doebley *et al.* 1984; Buckler and Holtsford 1996). Zmp is ssp. *parviglumis*, and Zmmx is ssp. *mexicana*, while [5] indicates that five accessions were sampled in that region. The branch lengths were assumed to be unknown. Within each of the six regions, a polytomy (a node with more than two descendants) was assumed between accessions, because gene flow has probably occurred between the multiple geographically close accessions. Eleven contrasts (one for each of the ancestral nodes) were calculated for each of the 34 knob loci. Because the contrasts were not normally distributed, Wilcoxon's signed-rank test was used to evaluate the associations between the contrasts (Sokal and Rohlf 1995). Sample sizes were sometimes small for individual accessions or individual knobs; therefore the overall strength of the Ab10-knob correlation was estimated by averaging the individual contrasts for all knob positions and by excluding the six within-region comparisons, where phylogenetic independence was dubious due to hybridization.

For comparison purposes, we also examined these correlations without accounting for phylogeny. This might be an acceptable model, because many of the populations probably diverged from one another at approximately the same time. Spearman's coefficient of rank correlation was used because some of the variables were not normally distributed (Sokal and Rohlf 1995).

RESULTS AND DISCUSSION

Ab10 and knobs in teosintes: Ab10 is found in 37% of the populations of *Z. mays* ssp. *parviglumis* and *mexicana*. Within those populations, Ab10 ranges in frequency up to 50% with an average frequency of 14%. Knobs were highly variable in frequency and size in all populations. The knob index ranged from 0.03 to 0.25 with an average of 0.15. For a scale of reference, a knob index of 0.15 is equivalent to being homozygous for small knobs at 15 of the 34 knob positions.

Why is Ab10 uncommon despite meiotic drive? Despite strong meiotic drive, Ab10 is uncommon in populations perhaps because of gametic selection against Ab10 pollen (Rhoades 1942). Model 1 (Table 2) shows

TABLE 3

Model of two linked chromosomal knobs during female meiosis with Ab10 present

Genotype	Viability	Female gametes produced			
		sL	Sl	sL	sl
SSLL	$(1 - vLL)(1 - vSS)(1 - 2vL)(1 - 2vS)$	1	0	0	0
SSLl	$(1 - vSS)(1 - vL)(1 - 2vS)$	$r_1(1 - r_2)m$ $+(1 - r_1)r_2m$ $+\frac{1}{4}r_1r_2(1 + 2m)$ $+\frac{1}{2}(1 - r_1)(1 - r_2)$	$r_1(1 - r_2)(1 - m)$ $+(1 - r_1)r_2(1 - m)$ $+\frac{1}{4}r_1r_2(3 - 2m)$ $+\frac{1}{2}(1 - r_1)(1 - r_2)$	0	0
SSll	$(1 - vSS)(1 - 2vS)$	0	1	0	0
SsLL	$(1 - vLL)(1 - 2vL)(1 - vS)$	$\frac{1}{2}$	0	$\frac{1}{2}$	0
SL / sl	$(1 - vL)(1 - vS)$	$r_1(1 - r_2)m$ $+\frac{1}{2}(1 - r_1)r_2m$ $+\frac{1}{8}r_1r_2(1 + 2m)$ $+\frac{1}{2}(1 - r_1)(1 - r_2)$	$\frac{1}{2}(1 - r_1)r_2(1 - m)$ $+\frac{1}{4}r_1r_2$	$\frac{1}{2}(1 - r_1)r_2m$ $+\frac{1}{8}r_1r_2(1 + 2m)$	$r_1(1 - r_2)(1 - m)$ $+\frac{1}{2}(r_1r_2)1 - m)$ $+\frac{1}{2}(r_1r_2)(1 - m)$ $+\frac{1}{2}(1 - r_1)(1 - r_2)$
Sl / sL	$(1 - vL)(1 - vS)$	$\frac{1}{2}r_2(1 - r_1)m$ $+\frac{1}{8}r_1r_2(1 + 2m)$	$r_1(1 - r_2)m$ $+\frac{1}{2}r_2(1 - r_1)(1 - m)$ $+\frac{1}{4}r_1r_2$ $+\frac{1}{2}(1 - r_1)(1 - r_2)$	$r_1(1 - r_2)m$ $+\frac{1}{2}r_2(1 - r_1)m$ $+\frac{1}{8}r_1r_2(1 + 2m)$ $+\frac{1}{2}(1 - r_1)(1 - r_2)$	$\frac{1}{2}r_2(1 - r_1)(1 - m)$ $+\frac{1}{2}r_1r_2(1 - m)$
Sll	$(1 - vS)$	0	$r_1(1 - r_2)m$ $+\frac{1}{2}(1 - r_1)r_2$ $+r_1r_2m$ $+\frac{1}{2}(1 - r_1)(1 - r_2)$	0	$r_1(1 - r_2)(1 - m)$ $+\frac{1}{2}(1 - r_1)r_2$ $+r_1r_2(1 - m)$ $+\frac{1}{2}(1 - r_1)(1 - r_2)$
ssLL	$(1 - vLL)(1 - 2vL)$	0	0	1	0
ssLl	$(1 - vL)$	0	0	$r_1(1 - r_2)m$ $+r_2(1 - r_1)m$ $+\frac{1}{4}r_1r_2(1 + 2m)$ $+\frac{1}{2}(1 - r_1)(1 - r_2)$	$r_1(1 - r_2)(1 - m)$ $+r_2(1 - r_1)(1 - m)$ $+\frac{1}{4}r_1r_2(3 - 2m)$ $+\frac{1}{2}(1 - r_1)(1 - r_2)$
ss11	1	0	0	0	1

Entries are the relative fitness for various gametes and plant genotypes.

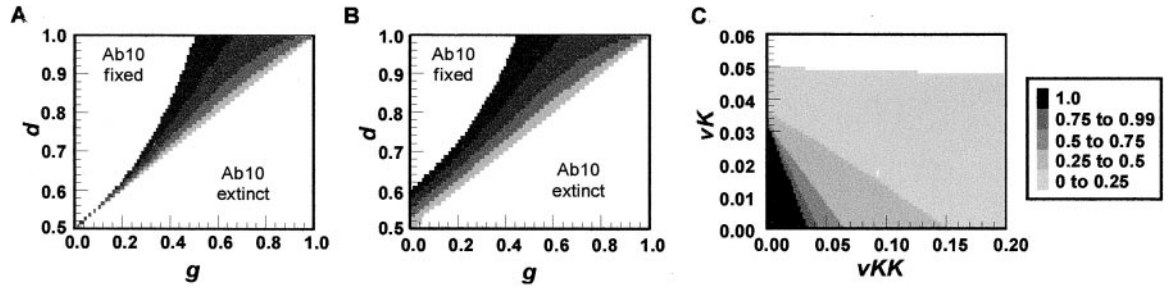

Figure 2.—Simulation results of Ab10 and an unlinked knob (model 1, Table 2). Frequency of the Ab10 or the knob is noted by shading. (A) Ab10 frequency with various meiotic drive (d) and gametic selection (g) values and no homozygous viability reduction; $vAA = 0$. (B) Ab10 frequency with various meiotic drive (d) and gametic selection (g) values and homozygous viability reduction; $vAA = 0.10$. (C) Unlinked knob frequency for various intensities of viability reduction by knob homozygotes (vKK) and viability reduction from individual knobbed chromosomes (vK); $d = 0.70$, $g = 0.37$, $vAA = 0.10$.

the interaction between meiotic drive, gametic selection, and viability reduction. Ab10 meiotic drive and gametic selection can result in an evolutionary stable polymorphism of Ab10 (Figure 2A). If Ab10 homozygosity reduces viability, then an even wider range of meiotic drive and gametic selection values can produce a stable Ab10 polymorphism (Figure 2B).

Ab10 meiotic drive varies depending on genetic background and growth conditions ($d = 0.50$ to 0.75; Rhoades 1942; Kikudome 1959). The theoretical estimates of male gametic selection fall near the range of field observations for Ab10 type I ($g = 0.09$ to 0.27; Rhoades 1942); however, the cytologically different type II version of Ab10 has full male gametic fitness in the one maize background tested (Rhoades and Dempsey 1988). We have no estimate of viability reduction by homozygous Ab10, but it is likely that the rearranged end on chromosome 10 carries fixed deleterious recessive alleles. These empirical estimates of Ab10 meiotic drive (s) and gametic selection (g) are in rough agreement with the values that produce polymorphic populations for Ab10 as predicted by Model 1 (Figure 2, A and B). This persistence of meiotic drive in maize sets the stage for knobs to develop on all chromosomes.

Is Ab10's meiotic drive responsible for the frequency and size of knobs in *Z. mays*? When Ab10 is present in a population, model 1 (Table 2) simulations indicate unlinked chromosomal knobs will obtain very high frequencies (Figure 2C). Over evolutionary time, the meiotic drive hypothesis suggests populations with high frequencies of Ab10 should develop higher frequencies of larger knobs. The best approach to evaluate this prediction of knobs and Ab10 evolution is to compare populations that have been separated long enough for differences in knob and Ab10 frequencies to have evolved. Maize's closest relatives (*Z. mays* ssp. *parviglumis* and ssp. *mexicana*) were used for the phylogenetic comparisons, because their populations have probably migrated and hybridized less than domesticated maize. The maize knob constitution is a subset of the variability found in these closest wild relatives, and all of the patterns described below can be seen in maize.

Although individual knob loci exhibited high variation in response to Ab10, the phylogenetic contrasts for the 34 loci showed a strong positive association between the knob index and Ab10 frequency (Table 4). The change in the genome-wide knob index was strongly correlated with the change in Ab10 frequency (Figure 3). The nonphylogenetic approach also indicated a significant correlation (Table 4). Hence, these significant associations and correlations between Ab10 frequency and the knob index supported the meiotic drive hypothesis.

Genetic experiments suggest multiple B (accessory) chromosomes can cause the loss of knobbed chromosomes (Rhoades and Dempsey 1972); therefore, we also considered whether B chromosomes had a confounding evolutionary effect on knobs. B chromosomes have no significant association with the knob index or Ab10 (Table 4), and therefore B chromosomes probably do not have a large influence on knob frequency or size over evolutionary time. The lack of correlation between B chromosomes and Ab10 was expected; all B chromosomes have the same knob, and therefore heteromorphic dyads are impossible.

Are knobs slightly deleterious? The simulation of meiotic drive model 1 indicates that knobs will rapidly go to fixation in Ab10 populations unless the knobs slightly reduce plant viability (Figure 2C). Viability reduction (vK or vKK) on the order of 0.03 is sufficient to maintain polymorphism when $d = 0.70$; however, lower levels of viability reduction ($vK = 0.01$) would be required to maintain polymorphism for small knobs exhibiting less drive ($d = 0.59$; Kikudome 1959). Twelve of the 34 knob loci are found to be fixed in some populations of *Z. mays* ssp. *parviglumis* or *mexicana*, but most knobs are not fixed within populations. Therefore, if the meiotic drive hypothesis is correct, the knobs of many loci must be slightly deleterious to prevent their fixation. The permissive hypothesis also suggests that large regions of repetitive DNA might be slightly deleterious.

In the absence of Ab10, knobs may be deleterious because of their effects on replication. Late replication and mitotic abnormalities have been associated with

TABLE 4

Correlations between the knob index, Ab10, B chromosomes, and altitude

	Phylogenetic independent contrasts				Nonphylogenetic	
	Pearson correlation		Signed-rank test of association[a]		Spearman correlation	
Knob index w/	R	P ($N = 5$)	Direction	P ($N = 374$)	R	P ($N = 52$)
Ab10 Freq.	0.896	<0.05	+	2.6×10^{-7}	0.366	<0.01
B Freq.	0.051	NS	+	0.520	−0.132	NS
Altitude	−0.796	NS	−	2.9×10^{-4}	0.247	NS
Ab10 Freq. w/	R	P ($N = 5$)	Direction	P ($N = 11$)	R	P ($N = 52$)
B Freq.	0.386	NS	+	NS	−0.166	NS
Altitude	−0.919	<0.05	−	NS	−0.114	NS

NS, not significant.

[a] Contrasts in which there was no difference between the independent variables were excluded from this test of association.

chromosomal knobs in maize (Pryor *et al.* 1980). Higher frequencies of multiple bridges have been found in plants with large numbers of chromosomal knobs (Fluminhan and Kameya 1997), and these bridges were especially common in plants that were germinated from seeds maintained in wild-like conditions (18 mo at 25°). Most of the chromosomal breaks appeared to occur adjacent to or proximal to chromosomal knobs. This could create somaclonal variants and inviable seedlings, which could directly reduce viability. These mitotic abnormalities may be related to the late replication of knob heterochromation relative to all other chromatin (Pryor *et al.* 1980; Lee and Phillips 1988; Fluminhan and Kameya 1997). Alternatively, individual knob loci may also be slightly deleterious because of their position relative to other genes. Knobs may be in linkage disequilibrium with deleterious neighboring genes and/or the heterochromatin of knobs might deleteriously affect the expression of neighboring genes.

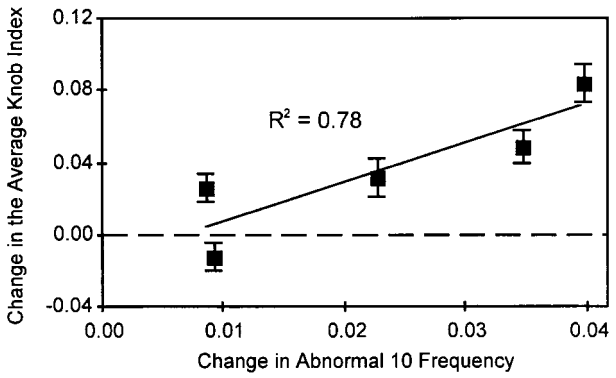

Figure 3.—Regression between the phylogenetic independent contrasts of knob indexes averaged over all loci and Ab10 frequency indicate the increases in Ab10 may have caused increases in knob frequency and size. The regression line is significant at $P < 0.05$; error bars indicate the standard error of the mean.

There are several avenues by which knob loci could be slightly deleterious, but this will remain unproven until experiments on natural populations examine knob fitness and control for linked loci.

Does the environment determine the distribution of chromosomal knobs? According to a recent review by Poggio *et al.* (1998), most studies have found negative correlations between genome size and altitude, between knobs and altitude, and between knobs and latitude. This suggests knobs might be more deleterious in certain environments. However, these studies have not considered Ab10 frequency or the phylogenetic history of the samples. Altitude and latitude are both related to the developmental rate of maize and teosinte. This selection for rapid mitotic cycles might favor smaller genome sizes and selection against repetitive DNA, while slower development would permit larger genome sizes. Evidence supporting this hypothesis has been found in animals (Pagel and Johnstone 1992). In plants there is far more intraspecific variation in genome size, and the proximate causes of associations between genome size, the environment, and development are unclear (Bennett 1985; Laurie and Bennett 1985; Porter and Rayburn 1990; Reeves *et al.* 1998). In addition, some of the claims for plant intraspecific genome size variation are being questioned (Greilhuber 1998).

To examine the connection between the environment and knobs, altitude and the knob index were correlated (Table 2). There was a significant negative rank association between altitude and the knob index but no significant linear correlation (Table 2). This suggests that there is a possible connection between the knobs and the environment. Alternatively, altitude may modify Ab10's distribution, and in turn, the Ab10 distribution is responsible for the correlation between altitude and the knob index. Ab10 frequency was significantly negatively correlated with altitude in one but not all the tests of association (Table 2). Therefore, the connection

between altitude and knob size and frequency may result from selection on knobs or on Ab10.

The distribution of knobs and/or Ab10 appears to vary with environment. The environmental connection could be explained by at least two alternatives: selection on knobs themselves or on loci linked to Ab10's rearrangement.

1. Knob DNA replicates very late relative to euchromatin and most heterochromatin, and it is likely that knobs lengthen the S phase (Pryor *et al.* 1980). This slow replication could slow cell division and plant development, selecting against knobs at high altitudes or latitudes. This effect could be even more substantial for Ab10, which has 1 million knob repeats (Peacock *et al.* 1981).
2. The associations between Ab10 and altitude could also be directly produced by the fitness consequences of the many genes permanently linked to the knob and meiotic drive factor through Ab10's rearrangement. One gene that is probably in Ab10's rearrangement is a maize quantitative trait locus (QTL) for flowering time (Koester *et al.* 1993). Further research is needed to refine this QTL's exact position. This QTL is probably in other Zea taxa, because an orthologous QTL is also present in Sorghum (Lin *et al.* 1995).

Observations from domesticated maize suggest Ab10's environmentally related distribution, rather than developmental selection, produces the associations between knob frequency and altitude. Ab10 is found extensively throughout maize races of Mexico, and these populations have the greatest numbers of knobs (McClintock *et al.* 1981). In contrast, many equatorial races have lower knob and Ab10 frequencies (McClintock *et al.* 1981). The lowland-Mexican maize race Zapalote Chico has the largest measured genome size and a rapid developmental rate, which argues against genome size being directly related to development (Wellhausen *et al.* 1952; Laurie and Bennett 1985). Zapalote Chico also has an extremely high knob index (0.25) and very high Ab10 frequencies (Ab10 frequency = 0.25; McClintock *et al.* 1981). This suggests that Ab10, rather than developmental rate, is more important for the evolution of internal knobs.

Environmental conditions are related to the distribution of knobs, which suggests knob fitness is somehow related to the environment. We cannot rule out that selection on repetitive copy number may produce this pattern, but a more likely possibility is that Ab10's fitness is directly related to the environment, and the knob association is indirect.

Does population size predict the knob index? The permissive hypothesis predicts that repetitive DNA should be most prevalent in small populations where drift is more important than selection against repetitive DNA (Stephan 1986), while the meiotic drive hypothesis suggests knobs should be favored most effectively when drift is small relative to the selection of meiotic drive; *i.e.*, in large populations. Genetic diversity determined from isozymes is the best available estimate of effective population size for these taxa (Doebley *et al.* 1984). Only 12 populations have both isozyme estimates of genetic diversity and knob estimates. For those populations, there is a nonsignificant positive correlation between the knob index and genetic diversity (Spearman $r = 0.447$, $n = 12$, $P > 0.10$). At a higher taxonomic level, the internal knobs are prevalent in the high diversity (large population size) taxa of *Z. mays* ssp. *parviglumis* ($H_T = 0.311$) and *mexicana* ($H_T = 0.287$), while they are nonexistent in the low diversity (small population size) taxa of *Z. mays* ssp. *huehuetenangensis* ($H_T = 0.173$) and *Z. luxurians* ($H_T = 0.155$). Both of these observations are more consistent with the predictions of the meiotic drive hypothesis rather than the permissive hypothesis.

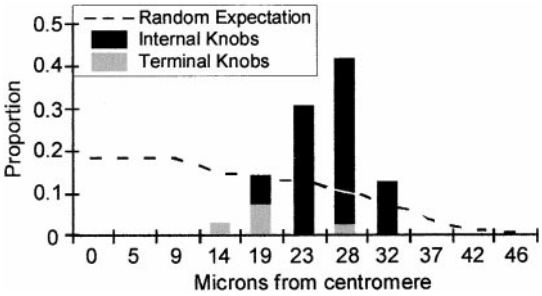

Figure 4.—The chromosomal position of knobs relative to the centromere. Distances from the centromere are approximate positions measured by Kato (1976) during the pachytene stage. Frequencies of knobs were based on the knob index averaged for all populations of *Z. mays* ssp. *parviglumis* and *mexicana*. Terminal knobs are found at the telomeres, while internal knobs are interstitial. The frequency of internal plus terminal knobs sums to one. The random expectation is the expected distribution of knobs if they were randomly distributed along chromosomal arms.

Does the permissive hypothesis predict the chromosomal position of knobs? Originally the nonrandom distribution of knobs was thought to result from certain chromosomal regions being able to efficiently "collect knob material" (Longley 1939). We now know knobs are heritable blocks of repetitive DNA. The chromosomal distribution of knobs was studied by plotting the proportion of knobs at various distances from the centromere (Figure 4). The proportion of knobs was based on the knob index for each locus relative to the total knob index for all loci averaged for all 51 populations. Distances from the centromere are approximate positions measured by Kato (1976) during the pachytene stage and were scaled to the average length of 1L (46.32 μm) measured by Longley (1939). These data allow us to compare the different predictions of the meiotic drive and permissive hypotheses for the optimal location of knob repetitive DNA evolution.

Cytologically visible knob positions appear to have dramatically changed during the evolution of Zea. Most Zea species and the sister genus Tripsacum have exclusively telomeric knobs on almost every chromosome arm, but they have no Ab10 (Kato 1976; Kato and Lopez 1990). However, the taxa with Ab10 (maize, ssp. *mexicana* and *parviglumis*) have 28 of their 34 knob positions in distal, but not telomeric, positions. Phylogenetic analysis with Zea taxa and Tripsacum indicated that these 28 internal knob positions in maize and *Z. mays* ssp. *parviglumis* and *mexicana* represent the derived or advanced state. *Z. diploperennis* is an exception because it has only terminal knobs and populations have been identified with Ab10 (Kato and Lopez 1990); however, the Type I Ab10 in *Z. diploperennis* might result from the extensive recent introgressions that occur between *Z. diploperennis* and Type I Ab10 containing maize lines (Buckler and Holtsford 1996).

Unequal crossing over can produce variation in repetitive DNA array size, such that drift or selection can easily eliminate neutral or slightly deleterious repetitive DNA in regions with high levels of recombination (Charlesworth *et al.* 1986). The permissive hypothesis predicts that most repetitive DNA should be in regions of low recombination and little unequal crossing over such as telomeres and centromeres. The positions of Zea telomeric knobs agree with this permissive hypothesis prediction. In addition, the absence of frequent unequal crossing over should result in repetitive DNA arrays that exhibit little intraspecific size variation (Charlesworth *et al.* 1994). However, the 28 internal knob positions are unlikely to be the product of the permissive process, as two points suggest there is no general reduction in levels of recombination.

1. Genetic experiments using flanking markers indicate that knobs affect recombination, but they can either decrease or increase recombination depending on the position of the knob, the size of the knob, and heterozygosity or homozygosity (Kikudome 1959; Rhoades and Dempsey 1966). The knobs can increase recombination by 36% or reduce it by 63% depending on the exact case (Kikudome 1959; Rhoades and Dempsey 1966). There does not appear to be any general pattern of suppression of recombination by knobs themselves; however, Ab10 does consistently increase recombination proximal to knobs (see below).
2. The tremendous within-population variation in knob size (Kato 1976) suggests recombination and unequal crossing over is frequent within knobs, although direct molecular evidence of recombination within a knob is not available. Thus, there is no general evidence of recombination suppression by knobs, which could support the permissive hypothesis.

Does the meiotic drive hypothesis predict the chromosomal position of knobs? The positions of knobs in maize, ssp. *mexicana* and *parviglumis* contradict the permissive hypothesis, but do they agree with the meiotic drive hypothesis? The meiotic drive hypothesis predicts that knobs will form in positions that maximize their chances of being transmitted to progeny through meiotic drive. First, crossovers must occur between the knob and centromere to produce the heteromorphic dyad necessary for preferential segregation (Rhoades and Dempsey 1966). One crossover is necessary between the centromere and knob; however, a large proportion of higher order crossovers will also produce heteromorphic dyads. Assuming the location of crossovers is binomially distributed, a knob would end up in a heteromorphic dyad 40% of the time at 30 cM from the centromere, 59% at 70 cM, and 66% at an infinite distance. The optimal efficiency for this preferential segregation system could be 83%, if heteromorphic dyads can be formed 66% of the time and the knobbed chromosomes experience 100% preferential segregation ($m = 1.0$) and homomorphic dyads exhibit normal segregation [$(1.0 \times 0.66) + (0.5 \times 0.33) = 0.83$]. Therefore, the meiotic drive hypothesis predicts that knobs will be preferentially found on the distal halves of chromosomes (Figure 4). Second, it has been suggested that during chromosome alignment, neocentromeres must apply tension to linked centromeres to ensure the coorientation of the two structures at metaphase (Yu *et al.* 1997). The effect of a poleward-moving neocentromere on the orientation of a linked centromere would be most pronounced when the neocentromere and centromere were in close proximity. Our expectation is that coorientation is under strong selection. In the absence of coorientation, the dicentric (centric-neocentric) chromosomes would be subject to chromosome bridging, breakage, and loss (*e.g.*, McClintock 1943).

The idea that there is an optimum knob position for meiotic drive is supported by a reevaluation of experimental data for the 3L knob (Rhoades and Dempsey 1966). Rhoades and Dempsey's (1966) work used inversions on chromosome 3L to examine preferential segregation. The normal knob is at a physical position of 0.6 on 3L. Inversion In3a results in the knob being positioned distally at 0.75, and inversion In3b moves the knob proximally to 0.45. We compared the frequency of preferential segregation among testcrosses with knobs at these three positions, while Ab10 and 3L knobs were heterozygous. Preferential segregation of the 3L knob is most efficient near its present position (recovered in 71% of testcross progeny; crosses 4, 5, and 11). Inversion 3a brings the 3L knob to a more distal position and strongly reduced preferential segregation (62% in cross 20; compared to normal, test of independence indicated $G = 57.6$, d.f. $= 1$, $P = 3 \times 10^{-14}$). In contrast, inversion 3b brings the 3L knob to a more proximal position and also slightly reduces preferential segregation (67% in

cross 14; compared to normal, $G = 12.0$, d.f. = 1, $P = 0.0005$).

This balance between selection for increased recombination and for coorientation of knobs and centromeres should result in knobs being clumped at an optimal distance if meiotic drive is responsible for their evolution. To test whether knob chromosomal position within the arms was nonrandom, we compared the observed distribution of 34 knob positions to a distribution that assumed knobs should be uniformly distributed across the length of all the chromosomal arms using a Kolmogorov-Smirnov test for goodness of fit (Sokal and Rohlf 1995). Knob positions were not uniformly distributed along the chromosomal arms (Kolmogorov-Smirnov test; $D = 0.468$, $n = 34$, $P < 0.001$). To determine whether the knobs were segregated to telomeric or centromeric ends of the chromosomal arm, we compared the positions of the 34 knobs to the mean chromosomal distance from the centromere with a *t*-test (Sokal and Rohlf 1995). Half of the genome is within 14 μm of the centromere, but the knobs were significantly distal to this midpoint (based on Student's *t*-test; $t = 10.26$, $n = 34$, $P = 5.9 \times 10^{-12}$), with a distinct modal distance of ~25 μm from the centromere (Figure 3). The six terminal knob positions were found only on short chromosomal arms with lengths <28 μm (Figure 1). The inversions on 3L suggested physical boundaries between which high levels of preferential segregation were found (In3a knob ≈31 μm from the centromere; In3b knob ≈19 μm). Only 28% of the genome is between 19 and 31 μm from the centromere; however, 82% of the knob index and 71% of the knob sites fall between these boundaries. Knob sites were significantly overrepresented in this physical region, with high preferential segregation potential ($G = 25.97$, d.f. = 1, $P = 3.4 \times 10^{-7}$). These observations agree with the meiotic drive hypothesis. One alternative to explain this positional bias is that centromeric heterochromatin obscures the cytological identification of the knobs near the centromere. This possibility is refuted by *in situ* hybridization experiments that demonstrate the vast majority of knob repeats are restricted to the cytologically observed positions (Peacock *et al.* 1981; R. K. Dawe, unpublished results).

Unfortunately, the genetic positions of most knobs are unknown, but we can make minimum estimates for the terminal knobs and rough estimates for a few internal knobs based on translocation breakpoints (Maize genome database, www.agron.missouri.edu). The six terminal knobs average 68 cM from the centromere; the internal knobs (1L1, 1S2, 2S1, 3L1) have a minimum distance of 37 cM and a rough average of 55 cM. In addition, Ab10 encodes a function that strongly increases recombination up to twofold in the regions proximal to the knobs (Kikudome 1959; Rhoades and Dempsey 1966), which may partially result from increased recombination in regions of centromeric heterochromatin (*e.g.*, Robertson 1968; Rhoades and Dempsey 1970; Nel 1973). This magnification of recombination by Ab10 permits high frequency heteromorphic dyad formation even for the internal knobs.

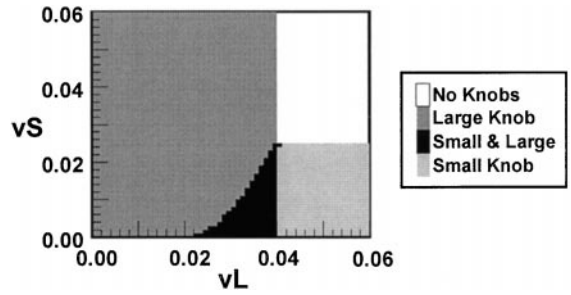

Figure 5.—Simulation results of linked large and small knobs (model 2, Table 3) for various levels of viability reduction against the large knob (*vL*) and the small knob (*vS*). Only in the black region did both the large and small knobs segregate together in the same population, otherwise either one or both of the knobs became extinct. The fixed parameters are as follows: $r_1 = 0.3$, $r_2 = 0.3$, $vLL = vSS = 0.02$, $m = 0.9$, freq(Ab10) = 0.12.

Do chromosomal knobs compete within a chromosomal arm? Meiotic drive predicts that chromosomal knobs may compete against one another on the same chromosomal arm, because only the largest knob is preferentially segregated (Kikudome 1959). Simulations of model 2 (Table 3) indicate that only if the viability reductions of both knobs are carefully balanced do they allow for multiple knobs segregating per chromosomal arm (Figure 5). This occurs because the smaller knob reduces viability constantly; however, the smaller knob experiences meiotic drive only in the uncommon situation when the large knob is absent. If the small knob were closer to the large knob (*e.g.*, $r_1 = 0.5$, $r_2 = 0.1$), then the parameter space of both knobs segregating slightly increases. Even when two knobs are segregating on a single arm, the smaller knob increasing in size could quickly result in only one knob present on the arm.

Thus the first functional knob to develop on a chromosome in a favorable position is likely to outcompete other linked knobs. Therefore, knobs should have a uniform or repulsed distribution among arms. The observed distribution indicates there is generally one frequent position per arm (Figure 1), while only the shortest chromosomal arms have no knobs. To test whether knobs were repulsed from each other, we calculated a coefficient of dispersion (CD) for the number of knob positions per arm (Sokal and Rohlf 1995). The significance of the CD was evaluated by randomly placing 34 knobs on 20 arms with a uniform probability per arm (1000 randomizations). The actual distribution of knobs was significantly repulsed relative to the random distribution according to the CD (CD = 0.50, $n = 34$, $P = 0.039$). Only chromosome 6 violates this pattern. 6L has multiple common knobs, while 6S has the nucleolar

organizing region that appears to preclude knobs or knob repeats from this arm (Kato 1976; Peacock *et al.* 1981). The knobs are spread out on 6L, which may have allowed more knobs to evolve; however, repulsion still appeared to occur between these knobs. We used phylogenetic independent contrasts between the knobs to test for repulsion. Knob 6L1 is negatively associated with knob 6L2 (Wilcoxon's signed-rank test $T_s = 0$, $n = 7$, two-tailed $P = 0.016$), while the neighboring knobs 6L2 and 6L3 do not show a consistent association. The meiotic drive hypothesis predicts this consistent pattern of repulsion, while neutral theories might expect a more random distribution.

Other genetically untested theories could also contribute to knob competition. First, the knob closest to the centromere could dominate the coorientation of the centromere, and this could favor internal knobs. Second, depending on the timing of crossovers and the microtubule attachment to knobs, multiple knobs on an arm could result in a chromosomal arm being pulled apart.

Transposition of knob sequences: Internal knobs appear to be a derived state, so how did the knobs get to these internal positions? Translocations are probably not responsible for the movement of knobs, because chromosome structure and linkage relationships are similar among Zea species (Randolph 1955; Kato 1976). Transposition of individual repeats or large tandem arrays seems most likely, and they have been characterized as megatransposons (Ananiev *et al.* 1998b). Knobs have several transposon-like characteristics. Inverted 350-bp repeats have been found, which could form fold-back DNA elements (Ananiev *et al.* 1998b). Full-length retroposons have been found interspersed within chromosomal knobs (Ananiev *et al.* 1998a). Additionally, we discovered that individual 180-bp repeats can form highly stable fold-back-like structures that are significantly more stable than random sequences with the same composition (E. S. Buckler, unpublished results).

These fold-back characteristics of knob repeats may have facilitated the transposition of knob repeats throughout the genome. Additionally the associated retroposons may have moved knob repeats throughout the genome. The initial movement of knob repeats probably has little to do with meiotic drive, but it established knob repeats throughout the genome for meiotic drive to work upon. After transposition, an increase of knob repeats in favorable chromosomal positions could be accomplished via unequal crossing over or *in situ* amplification, eventually giving rise to a cytologically visible knob with thousands of repeats. Although size variability normally leads to stochastic loss of repeats, the combination of size variability with positive selection from meiotic drive can lead to larger arrays of repetitive DNA.

Genomic implications of knob evolution: Chromosomal meiotic drive systems may be a powerful force that can rapidly and extensively modify the genome and even create novel genomic structures. Genome-wide meiotic drive can develop rapidly; for example, cytologically visible internal knobs have evolved subsequent to the *Z. mays* divergences within the last 100,000 years. Meiotic drive may be an ephemeral system, which rapidly results in either fixation or extinction of the "driver" genes. However, while meiotic drive persists, strong selection for ever better knobs (larger knobs with better positions) has probably affected thousands of knob-linked loci. The combination of selection and linkage disequilibrium between knobs and tightly linked genes may have caused linked genes to lose diversity. Future large-scale diversity surveys will allow this prediction to be tested. The powerful effect of Ab10 on knobs calls into question the phylogenetic utility of knobs for examining the divergence patterns of maize races, as the spread of knobs would depend tremendously on Ab10. The presence of the knob heterochromatin throughout the genome may also modify the expression of genes neighboring knobs. Furthermore, meiotic drive may have retarded selection from effectively evaluating the organismal-level fitness of the knob-linked genes. In deep time, meiotic drive systems that interact with the cytoskeleton may be very important in creating genomic structures such as centromeres and dispersed tandem arrays (satellite DNA); *i.e.*, satellite DNA may be the remnant of ancient episodes of meiotic drive.

Conclusions: The existence, frequency, size, and position of knobs agree with the theory that Ab10 is responsible for their evolution (Table 1). The permissive theory of repetitive DNA evolution cannot explain the rapid evolution or the position of the 28 cytologically visible internal knob positions. The environment may affect the evolution of knobs but it is unclear whether this occurs through selection against knobs or through selection on Ab10. Knobs are also an important example of repetitive DNA being favored by selection. Meiotic drive appears to have played an important role in structuring the maize genome.

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LITERATURE CITED

Alfenito, M. R., and J. A. Birchler, 1993 Molecular characterization of a maize B chromosome centric sequence. Genetics **135:** 589–597.

Ananiev, E. V., R. L. Phillips and H. W. Rines, 1998a Complex structure of knob DNA on maize chromosome 9: retrotransposon invasion into heterochromatin. Genetics **149:** 2025–2037.

Ananiev, E. V., R. L. Phillips and H. W. Rines, 1998b A knob-associated tandem repeat in maize capable of forming fold-back

DNA segments: are chromosome knobs megatransposons? Proc. Natl. Acad. Sci. USA **95:** 10785–10790.

Bedinger, P., and S. D. Russell, 1994 Gametogenesis in maize, pp. 48–61 in *The Maize Handbook*, edited by M. Freeling and V. Walbot. Springer-Verlag, New York.

Bennett, M. D., 1985 Intraspecific variation in DNA amount and the nucleotypic dimension in plant genetics, pp. 283–302 in *Plant Genetics*, edited by M. Freeling. Alan R. Liss, New York.

Bretting, P. K., M. M. Goodman and C. W. Stuber, 1987 Karyological and isozyme variation in West Indian and allied American mainland races of maize. Am. J. Bot. **74:** 1601–1613.

Buckler, E. S., IV, and T. P. Holtsford, 1996 *Zea* systematics: ribosomal ITS evidence. Mol. Biol. Evol. **13:** 612–622.

Burr, B., F. A. Burr, E. C. Matz and S. J. Romero, 1992 Pinning down loose ends mapping telomeres and factors affecting their length. Plant Cell **4:** 953–960.

Charlesworth, B., C. H. Langley and W. Stephan, 1986 The evolution of restricted recombination and the accumulation of repeated DNA sequences. Genetics **112:** 947–962.

Charlesworth, B., P. Sniegowski and W. Stephan, 1994 The evolutionary dynamics of repetitive DNA in eukaryotes. Nature **371:** 215–220.

Dawe, R. K., and W. Z. Cande, 1996 Induction of centromeric activity in maize by *suppressor of meiotic drive 1*. Proc. Natl. Acad. Sci. USA **93:** 8512–8517.

Doebley, J. F., M. M. Goodman and C. W. Stuber, 1984 Isoenzymatic variation in *Zea* (Gramineae). Syst. Bot. **9:** 203–218.

Felsenstein, J., 1985 Phylogenies and the comparative method. Am. Nat. **125:** 1–15.

Fluminhan, A., and T. Kameya, 1997 Involvement of knob heterochromatin in mitotic abnormalities in germinating aged seeds of maize. Genome **40:** 91–98.

Greilhuber, J., 1998 Intraspecific variation in genome size: a critical reassessment. Ann. Bot. **82:** 27–35.

Kato Y., T. A., 1976 Cytological studies of maize (*Zea mays* L.) and teosinte (*Zea mexicana* Schrader Kuntze) in relation to their origin and evolution. Mass. Agric. Exp. Stn. Bull. **635:** 1–185.

Kato Y., T. A., and A. R. Lopez, 1990 Chromosome knobs of the perennial teosintes. Maydica **35:** 125–141.

Kikudome, G. Y., 1959 Studies on the phenomenon of preferential segregation in maize. Genetics **44:** 815–831.

Koester, R. P., P. H. Sisco and C. W. Stuber, 1993 Identification of quantitative trait loci controlling days to flowering and plant height in two near isogenic lines of maize. Crop Sci. **33:** 1209–1216.

Laurie, D. A., and M. D. Bennett, 1985 Nuclear DNA content in the genera *Zea* and *Sorghum*. Intergeneric, interspecific and intraspecific variation. Heredity **55:** 307–313.

Lee, M., and R. L. Phillips, 1988 The chromosomal basis of somaclonal variation. Annu. Rev. Plant Physiol. Plant Mol. Biol. **39:** 413–437.

Lin, Y. R., K. F. Schertz and A. H. Paterson, 1995 Comparative analysis of QTLs affecting plant height and maturity across the Poaceae, in reference to an interspecific sorghum population. Genetics **141:** 391–411.

Longley, A. E., 1939 Knob position on corn chromosomes. J. Agric. Res. **59:** 475–490.

Longley, A. E., 1945 Abnormal segregation during megasporogenesis in maize. Genetics **30:** 100–113.

Lyttle, T. W., 1991 Segregation distorters. Annu. Rev. Genet. **25:** 511–557.

McClintock, B., 1929 Chromosome morphology in *Zea mays*. Science **69:** 629.

McClintock, B., 1943 Maize Genetics. Yearb. Carnegie Inst. Wash. **42:** 148–152.

McClintock, B., T. A. Kato Y. and A. Blumenschein, 1981 *Chromosome Constitution of Races of Maize*. Colegio de Postgraduados, Chapingo, Mexico.

Nel, P. M., 1973 The modification of crossing over in maize by extraneous chromosomal elements. Theor. Appl. Genet. **43:** 196–202.

Pagel, M., and R. A. Johnstone, 1992 Variation across species in the size of the nuclear genome supports the junk dna explanation for the c-value paradox. Proc. R. Soc. Lond. Ser. B Biol. Sci. **249:** 119–124.

Peacock, W. J., E. S. Dennis, M. M. Rhoades and A. J. Pryor, 1981 Highly repeated DNA sequence limited to knob heterochomatin in maize. Proc. Natl. Acad. Sci. USA **78:** 4490–4494.

Poggio, L., M. Rosato, A. M. Chiavarino and C. A. Naranjo, 1998 Genome size and environmental correlations in maize (Zea mays ssp. mays, Poaceae). Ann. Bot. **82:** 107–115.

Porter, H. L., and K. L. Rayburn, 1990 B-chromosome and c-band heterochromatin variation in Arizona maize populations adapted to different altitudes. Genome **33:** 659–662.

Pryor, A., K. Faulkner, M. M. Rhoades and W. J. Peacock, 1980 Asynchronous replication of heterochromatin of maize. Proc. Natl. Acad. Sci. USA **77:** 6705–6709.

Purvis, A., and A. Rambaut, 1995 Comparative analysis by independent contrasts (CAIC): an Apple Macintosh application for analysing comparative data. Comput. Appl. Biosci. **11:** 247–251.

Randolph, L. F., 1955 Cytogenetic aspects of the origin and evolutionary history of corn, pp. 16–61 in *Corn and Corn Improvement*, edited by G. F. Sprague. Academic Press, New York.

Reeves, G., D. Francis, M. S. Davies, H. J. Rogers and T. R. Hodkinson, 1998 Genome size is negatively correlated with altitude in natural populations of Dactylis glomerata. Ann. Bot. **82:** 99–105.

Rhoades, M. M., 1942 Preferential segregation in maize. Genetics **27:** 395–407.

Rhoades, M. M., 1952 Preferential segregation in maize, pp. 66–80 in *Heterosis: A Record of Researches Directed Toward Explaining and Utilizing the Vigor of Hybrids*, edited by J. W. Gowen. Hafner Publishing Co., New York.

Rhoades, M. M., and E. Dempsey, 1966 The effect of abnormal 10 on preferential segregation and crossing over in maize. Genetics **53:** 989–1020.

Rhoades, M. M., and E. Dempsey, 1970 Crossing over and preferential segregation in chromosome 10 induced by abnormal 10. Maize Genet. Coop. Newsl. **44:** 57.

Rhoades, M. M., and E. Dempsey, 1972 On the mechanism of chromatin loss induced by the B chromosome of maize. Genetics **71:** 73–96.

Rhoades, M. M., and E. Dempsey, 1985 Structural heterogeneity of chromosome 10 in races of maize and teosinte, pp. 1–18 in *Plant Genetics*, edited by M. Freeling. Alan R. Liss, New York.

Rhoades, M. M., and E. Dempsey, 1988 Structure of K10-II chromosome and comparison with K10-I. Maize Genet. Coop. Newsl. **62:** 33.

Robertson, D. S., 1968 Increased crossing over in chromosome 5 in the presence of abnormal 10. Maize Genet. Coop. Newsl. **42:** 89.

Sokal, R. R., and F. J. Rohlf, 1995 *Biometry*. W. H. Freeman, New York.

Stephan, W., 1986 Recombination and the evolution of satellite DNA. Genet. Res. **47:** 167–174.

Stephan, W., 1987 Quantitative variation and chromosomal location of satellite dnas. Genet. Res. **50:** 41–52.

Wellhausen, E. J., L. M. Roberts, E. Hernandez X. and P. C. Mangelsdorf, 1952 *Races of Maize in Mexico: Their Origin, Characteristics and Distribution*. Bussey Institution of Harvard University, Boston.

Wilkes, H. G., 1967 *Teosinte: the Closest Relative of Maize*. Bussey Institution of Harvard University, Cambridge, MA.

Yu, H. G., E. N. Hiatt, A. Chan, M. Sweeney and R. K. Dawe, 1997 Neocentromere-mediated chromosome movement in maize. J. Cell Biol. **139:** 831–840.

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