

nomically, other than in the commercial breeding program for which I was personally responsible, little use has been made of the [doubled haploid] method in the development of homozygous diploids. Increased interest in and use of such radical techniques is likely in the future as the challenges of intensified commercial maize culture and of the highly competitive hybrid seedcorn market necessitate an increasingly high degree of responsiveness on the part of the maize breeder.”

The practicality of the method today certainly owes much to Coe’s “stock 6” and its derivatives, to improvements and techniques for genome doubling, and to the higher technical skills of maize breeders.

Firsts (achieved by SSC):

1) First haploid to doubled haploid. The first homozygous diploid derived from a haploid was out of sweet corn (Golden Cross Bantam).

2) First substantial confirmation of different rates of parthenogenesis among female parents. (Stadler obtained a frequency of about 1:100 in a diploid multiple recessive tester; most prior information suggested rates of about 1:1000.)

3) First recognition that rates of parthenogenesis were influenced by the male parent.

4) First haploids in quantity.

5) First observation of high rates of somatic chromosome complement doubling in haploids.

6) First doubled haploids in quantity.

7) First to use “embryo markers” for dry seed haploid selection (Pu, etc.).

(A major disadvantage of Pu, purple embryo marker, is — was — that it occurs widely in Corn Belt maize; Stadler told me that 15% of the then available inbreds in use had the purple plumule phenotype, hence Pu was not a “clean” marker for my purposes.)

8) First doubled haploid line(s) in successful commercial hybrid(s). (Example: DeKalb 640)

9) First “second generation” doubled haploid lines in commercial hybrid (H2386 and H2398, both ex H73xH225).

10) First cloning of haploid through reproductive process (W23 haploid).

11) First, with Sam Goodsell, to demonstrate cytoplasmic transfer through androgenesis.

12) First to demonstrate (in W22) the possibility of “fixing” high-performing substrains of long established inbreds through haploidy.

SIoux FALLS, SD

Quantitative trait loci for leaf angle, leaf width, leaf length, and plant height in IBM-94

— Wassom, J

Modern maize varieties are more productive than varieties of a few decades ago, partly due to higher population densities and adaptations that permit vigorous growth at high densities. Plant forms that enable efficient light interception at high popula-

tion densities will increase yield production under modern field conditions. Leaf angle has been shown to affect yield especially at high densities. To determine the QTL affecting leaf traits and plant height in maize I experimented with IBM-94, a B73 x Mo17 recombinant inbred line (RIL) population developed by other researchers for genetic studies (Coe et al., *Plant Physiol* 128:9-12, 2002; Cone et al., *Plant Physiol* 130:1598-1605, 2002; Lee et al., *Plant Mol Biol* 48:453-461, 2002).

Seed for the 93 RIL constituting IBM-94 was obtained from the Maize Genetics Cooperative Stock Center (<http://maizecoop.cropsci.uiuc.edu>). In year 2006 the original seed was grown and plants were self-pollinated to produce enough RIL seed to plant replicated experimental plots at Sioux Falls, South Dakota, USA. In years 2007, 2008, and 2009 the 93 RIL were grown in randomized complete blocks with three replicates each year. Space was limited, so each plot included four plants spaced 279 mm apart in rows 76 mm apart. There were no extra spaces between plots in rows. At anthesis or soon after, the total plant height to the tassel tip and the leaf at the uppermost ear shoot were measured on the two center plants in each plot. Leaf measurements included the leaf angle from vertical, maximum leaf width, and distance from the ligule to the tip of the straightened leaf.

Statistical analysis, including analysis of variance and heritability was performed with PLABSTAT (University of Hohenheim, Germany, <https://plant-breeding.uni-hohenheim.de/-ipspwww/soft.html>). Genetic map distances of markers and molecular marker genotypes of each RIL were obtained from the Maize Genetics and Genomics Database (MaizeGDB) (<http://www.maizegdb.org>). Phenotype data for the RILs was combined with marker genotypic information and map distances from the MaizeGDB IBM2 map to analyze for QTL using PLABMQTL (University of Hohenheim, Germany, <https://plant-breeding.uni-hohenheim.de/-ipspwww/soft.html>).

There was significant variation among genotypes (Table 1; opposite page) and 1 to 3 QTL identified for each of the measured traits (Table 2). Larger plots and the larger IBM-302 population might have improved precision and enabled detection of more QTL. The total area taken up by nursery rows and experimental plots was about 50 by 75 feet, illustrating that even with limited resources the IBM-94 population and MaizeGDB data can be used for QTL mapping.

ST. PAUL, MINNESOTA

Adapted *Zea diploperennis*: *Zea diploperennis*-maize hybrid adapted to the U.S. corn belt

— Carlson, LA; Price, SC

Experiment 1. 1979, a cross between *Zea diploperennis* No. 1190 as female parent and Minhybrid 8201 (A641 x W182B) as male was made in St. Paul (materials were furnished by John Doebley of the University of Wisconsin and Jon Geadelmann of the University of Minnesota). Approximately 6,000 F2 seeds were collected from an isolation plot of the F1 plants. In 1982

Table 1. Characterization of IBM-94 RIL grown at Sioux Falls, SD, in 2007, 2008, and 2009.

	Leaf Angle		Leaf Width		Leaf Length		Plant Height	
	Degrees		mm					
RIL Mean †	26 ± 2.6		91.0 ± 2.95		764.6 ± 20.76		2231.3 ± 101.24	
Range among RILs ‡	9 to 53		71.2 to 114.5		641.7 to 907.0		1323.2 to 2796.8	
ANOVA								
Source of Variation	MS, F							
Genotypes	623.8	10.49**	576.6	7.24**	26374	6.80**	362149	3.93**
Years	1026.0	9.54*	990.6	3.31	582080	35.41**	18780568	71.16**
Genotype × Year	59.5	1.68**	79.6	1.52**	3878	1.86**	92242	4.27**
Replications in Years	107.5	3.04**	299.2	5.72**	16436	7.87**	263924	12.21**
Error	35.3		52.3		2089		21617	
Variance Components and H ²								
σ ^{2g}	62.7 ± 10.13		55.2 ± 9.40		2500 ± 429.8		29990 ± 5965.4	
σ ^{2e}	3.3 ± 2.61		2.5 ± 2.57		2027 ± 1475.5		66368 ± 47600.4	
σ ^{2ge}	8.1 ± 2.19		9.1 ± 2.96		596 ± 141.2		23542 ± 3226.4	
σ ^{2rep(e)}	0.8 ± 0.58		2.7 ± 1.61		154 ± 88.4		2605 ± 1419.0	
σ ^{2error}	35.3 ± 2.14		52.3 ± 3.16		2089 ± 126.2		1617 ± 1304.8	
H ² , %	90.47		86.20		85.30		74.53	
H ² , 90% confidence int.	86.24, 93.26		80.08, 90.23		78.78, 89.59		63.25, 81.97	

*,** Significant at α = 0.05, 0.01, respectively.

† ± Standard error

Table 2. Regression models with QTL for leaf angle, leaf width, leaf length, and plant height in IBM94 RILs. The QTL were detected and included in regression models if LOD values in scans were greater than the LOD threshold corresponding to á = 0.05, by permutation test. Effects that are positive in sign are favored by the Mo17 allele.

Bin†	Marker interval	Chrom. and position	CV‡	Support Interval§	LOD	Partial R ²	Effect¶	R ² # _{adj}	ˆp ††
Leaf Angle									
1.05–1.05	umc1603–uaz273	1/480	59.8%	465–495	4.27	16.4%	-3.802	27.1%	32.5%
5.04–5.05	csu308–umc1482	5/375	44.6	345–390	4.80	13.9	3.084		
9.01–9.01	umc1867–lim343	9/30	62.9	15–45	4.76	10.4	-2.760		
Leaf Width									
2.04–2.04	umc2088–umc2250	2/320	86.5	300–340	5.20	21.6	4.076	25.0	31.2
8.03–8.03	umc1735–php20714	8/280	85.9	260–300	5.28	15.2	-3.080		
Leaf Length									
2.09–2.09	bnlg1893–AY110389	2/660	54.4	640–680	4.08	11.1	-21.531	9.1	11.9
Plant Height									
4.06–4.06	umc2027–AY110310	4/350	40.3	330–360	4.01	10.4	-26.64	8.4	32.1

*, ** α = 0.05 or 0.01, respectively, for the probability that this QTL affected the trait independently of other QTL.

† Bins where the flanking markers are located and positions are the coordinate values on the MaizeGDB IBM2 map (<http://www.maizegdb.org>).

‡ Frequency of detection within a 1-LOD support interval in 1000 CV runs with families randomly divided for detection and validation.

§ Interval with LOD scores within 1 LOD of the QTL peak

¶ Effects were determined in a simultaneous multiple regression that included factors with LODs ≥ the α = 0.05 threshold.

R² adjusted for the number of terms in the multiple regression models.

††The proportion of genotypic variance explained by all QTL in the models.

380 seeds of the 1980 F2's were planted. Only two plants flowered at the normal (110-day maturity) time. These were observed and crossed with each other, and a satisfactory quantity of good seed was saved. Each following year the resulting seed (17LD) has been planted and has bloomed and produced seed. A minority (less than 5% of the plants) continue to bloom too late in Minnesota to mature seed.

In summation: The plants have a very pronounced teosinte growth habit, with 3 to 30 tillers; 10 to 200 silking locations; 10 to 100 viable seeds per ear; numerous brace roots extending from the 1st to the 7th node; some plants regenerate from planted nodes with brace roots. Seed is available on a limited basis from 1988 by contacting LAC.

Experiment 2. During 1985 in St. Paul, B73 was crossed with *Zea diploperennis* originating from Laventana, Jalisco, Mexico. The female parent was B73. A large amount of F1 seed was obtained, of which eight plants were grown in 1986 in St. Paul. The F1 plants were planted in May, and were "short-dayed" at the three leaf stage by covering them with 30 gallon trash barrels from 6 p.m. until 8 a.m. The short-day treatment was discontinued after 24 days. Three of the plants tasseled 11 days later. The eight F1 plants were grown in isolation to obtain as much F2 seed as possible. More than 600 seeds were obtained. In 1987 597 F2 plants were observed at the University of Minnesota. Seven plants flowered without the short-day treatment during the period July 28 through Aug. 15. Sibs were made between these plants. The balance of the plants did not mature. In 1988 about 35 F3 plants were grown in isolation without the short-day treatment in St. Paul. Open pollination with no selection was used, and a reasonable amount of seed was saved. All of the plants set seed with a three-week range in the time of maturity.

In summation: The plants have a very pronounced teosinte growth habit, with 1 to 10 tillers; 4 to 30 silking locations; 10 to 150 viable seeds per ear; numerous brace roots extending from the 1st to the 10th node; some plants regenerate from planted nodes with brace roots; new plant regeneration takes place when a tiller is held to the ground with the new roots growing from the node.

Experiment 3. The above seeds from 1979 and 1985 were combined and backcrossed two times to *Zea diploperennis* in the greenhouse. These plants continued to bloom in the long days of the Northern Corn belt. They continue to have multiple tillers and multiple ears but the ears returned to, for the most part, *Zea dip.*-like morphology. Most seeds were enclosed in a cupule. The progeny have been replicated several times (4) over the years in isolation. Seed from these plants, which I named "87½ *Zea dip.*," is available from the author (LAC) or the North Central Regional Plant introduction Station at Ames, Iowa.

In 2011 a single plant among 186 seemed to develop a cob-like structure. 51 plants from this selection were grown in isolation in 2012. All plants were harvested, but 20 plants were harvested individually. Of these, 13 plants yielded 1,032 grams of seed. 311 grams remain as ear or ear segments as of June 1, 2013. 721 grams are disarticulated. No attempt was made to preserve the ear structure. Without counting or weighing, at least 600 grams of these seeds had a pronounced extruded endosperm. The



Figure 1. Adapted *Zea diploperennis*, example ear.

other seven plants, without any articulated ears, yielded 1,179 grams of seed. All seeds from these seven plants were enclosed by the cupule.

URBANA, ILLINOIS

USDA/ARS/MWA

Maize Genetics Cooperation • Stock Center

2012/2013 allele tests at the Maize Genetics Stock Center

— Stinard, PS; Sachs, MM

During the past year, we obtained positive allele tests for the following previously uncharacterized mutants: *te**-87-2490-22 is allelic to *te1* and is now called *te1*-87-2490-22. *stb**-N938C is allelic to *oro1* and is now called *oro1*-N938C. *vp**-UFMu-03777 is allelic to *vp5* and is now called *vp5*-UFMu-03777.

We obtained negative allele tests for the following mutants: *clpp1-ys* is not allelic to *oro1*. *l3*, a poorly characterized pale luteus seedling mutant on 6L, is not allelic to *l10*. Previous tests showed that *l3* is not allelic to *l12* or *l15*. *l3* still remains to be tested against the 6L mutants *w1* and *w15*, which also have a pale luteus phenotype.

zebra7* and *luteus17* are allelic to *lemon white1

— Stinard, PS and Sachs, MM

Based on map location, function, and predicted phenotype, Stinard (MNL 86:29-31, 2013) hypothesized that the maize *lemon white1* (*lw1*) locus encodes the plastidial (MEP pathway) isopren-