A reappraisal of Kindiger and Hamann's 1993 protocol to produce large numbers of paternal haploids for cytoplasmic conversions

Kindiger and Hamann (1993, Crop Sci. 33:342-344) developed a novel system to efficiently produce paternal (androgenic) haploids in maize. They generated a maize tertiary trisomic plants with two normal chromosome 3s plus a B-3Ld chromosome in the inbred, W23. The two normal chromosome 3s carried the *ig1* allele and the dominant *Ig1* allele was present on the B3-Ld chromosome. The *ig1* locus in chromosome 3 is close to the breakpoint in B-3Ld, and in the inbred, W23, *ig1/ig1* plants are completely or almost completely male-sterile and plants with a dominant allele of the *ig1* locus are male-fertile.

They indicated that theoretically, transmission of the B-3Ld chromosome should be 50% through the female and only 2% through the pollen (Beckett 1983, Can. J Genet. Cytol. 25:346-353). Therefore, when a 3(*ig1*)/3(*ig1*) female parent is crossed by a 3(*ig1*)/3(*ig1*)/B-3Ld(*Ig1*) male parent, nearly all of the progeny would be *ig1/ig1* (and male-sterile in the inbred, W23) because the B-3Ld chromosome is rarely transmitted through the pollen. Progeny that received the B-3Ld (*Ig1*) chromosome would be male-fertile and could easily be recognized. Selfing of the male-fertile individuals 3(*ig1*)/3(*ig1*)/B-3Ld(*Ig1*) or crossing them to their male-sterile sibs 3(*ig1*)/3(*ig1*) can be used to maintain the 3(*ig1*)/3(*ig1*)/B-3Ld(*Ig1*) stock.

They also crossed 3(*ig1*)/3(*ig1*)/B-3Ld(*Ig1*) males by female parents with several different types of male-sterile cytoplasms and recovered 3(*ig1*)/3(*ig1*)/B-3Ld(*Ig1*) lines with the male-sterile cytoplasms.

Seed of selfed 3(*ig1*)/3(*ig1*)/B-3Ld(*Ig1*) plants and seed of 3(*ig1*)/3(*ig1*) plants with L, MY, ME, S, SD, VG, CA, C, and Q male-sterile cytoplasms crossed as female parents by 3(*ig1*)/3(*ig1*)/B-3Ld(*Ig1*) male parents were obtained from the Maize Genetics Stock Center, Univ. of IL and planted in field plantings in 1998. These stocks were originally provided to the Maize Genetics Stock Center by Kindiger's lab. A high frequency of distinctively smaller plants were observed in field-plantings of each of these stocks as shown in the following table:

| Cross | cytoplasm | # grew | # small | % small |
|---|---|---|---|---|
| 3-*ig1*/3-*ig1*/B-3Ld-*Ig1* self | N | 111 | 22 | 19.8 |
| 3-*ig1*/3-*ig1* x 3-*ig1*/3-*ig1*/B-3Ld-*Ig1* | L | 15 | 3 | 21.3 |
| 3-*ig1*/3-*ig1* x 3-*ig1*/3-*ig1*/B-3Ld-*Ig1* | MY | 17 | 5 | 30.7 |
| 3-*ig1*/3-*ig1* x 3-*ig1*/3-*ig1*/B-3Ld-*Ig1* | ME | 15 | 2 | 13.3 |
| 3-*ig1*/3-*ig1* x 3-*ig1*/3-*ig1*/B-3Ld-*Ig1* | S | 79 | 3 | 3.7 |
| 3-*ig1*/3-*ig1* x 3-*ig1*/3-*ig1*/B-3Ld-*Ig1* | SD | 19 | 8 | 29.6 |
| 3-*ig1*/3-*ig1* x 3-*ig1*/3-*ig1*/B-3Ld-*Ig1* | VG | 26 | 2 | 7.7 |
| 3-*ig1*/3-*ig1* x 3-*ig1*/3-*ig1*/B-3Ld-*Ig1* | CA | 22 | 6 | 21.4 |
| 3-*ig1*/3-*ig1* x 3-*ig1*/3-*ig1*/B-3Ld-*Ig1* | C | 120 | 22 | 14.0 |
| 3-*ig1*/3-*ig1* x 3-*ig1*/3-*ig1*/B-3Ld-*Ig1* | Q | 126 | 53 | 33.3 |
| Total | | 550 | 126 | 22.9 |

The smaller plants definitely did not have the morphology of a haploid plant; instead, they had a morphology that was typical of a monosomic-3 plant or a plant that was hypoploid for the long arm of chromosome 3. Monosomic-3 plants and plants hypoploid for 3L have a similar morphology. They are much smaller and have more upright leaves that are thicker, shinier, and narrower than those in their diploid siblings. Chromosome counts were carried out on several of the smaller plants, and they each had 20 chromosomes.

Therefore, the smaller plants produced by this system are not haploids; however, they appear to be hypoploids for the long arm of chromosome 3, and therefore appear to have one normal chromosome 3 and a 3B chromosome. Such

plants would be expected if the complete B-3Ld translocation was present in the male parent.

The Maize Genetics Stock Center (personal communication) confirms that the Kindiger *ig1* maintainer stock carries the complete TB-3Ld translocation—crosses of the maintainer stock to *a1* testers segregated for colorless kernels with colored plumules.