5. Seed irradiation studies.

Seed irradiation studies are continuing. In 1967, seeds of the B14 inbred were treated by Dr. Robert Briggs at the Brookhaven National Laboratory, and an isolated open pollinated field grown at the National Colonial Farm in Accokeek, Maryland. Many self-pollinations will be made in 1968 to test the effectiveness of inducing mutations in maize seed. This method should be applicable to all open pollinated crops.

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1. A dosage effect at the Ht locus.

The Ht gene conditions chlorotic lesion resistance to northern corn leaf blight. In a program directed toward the study of gene dosage at the Ht locus, we were able to obtain monoploid, triploid, and tetraploid seed from diploid Ht Ht and ht ht stocks. As a result, we were able to test one, two, three, and four doses of both the Ht and ht alleles for disease reaction. The diploid stocks were secured from Dr. Albert L. Hooker of the University of Illinois in Urbana. Lines R223 and 65:225-1 are homozygous Ht. W153R is homozygous ht. Lines 65:225-1 and W153R are isogenic. Most of our data were obtained from R223 and W153R. Experiments now in progress are aimed at testing the dosage levels in the isogenic material.

Putative monoploids were detected by crossing purple embryo marker as pollen parent to the diploid lines. The kernels with purple endosperm and non-purple embryos were saved. Monoploids were confirmed by chromosome counts on growing root tips. Tetraploid seedlings were obtained by means of Shaver's "decapitated root" technique (Maize Genetics Newsletter 38: 21-22). Triploids were obtained from tetraploid x diploid crosses.

Plants on the four dosage levels were inoculated at the three-four leaf seedling stage with spore suspensions of the pathogen, Helminthosporium turcicum. Seedlings were incubated for 18 hours at 20°C and 100% humidity. The degree of infection was determined by measuring the total area of the fourth leaf, and then the area of that leaf covered with lesions. The per cent infection was calculated as follows:

$$\frac{\text{Infected area of leaf}}{\text{Total area of leaf}} \times 100$$

Monoploid and diploid seedlings, carrying one and two doses of Ht, showed no significant difference in degree of resistance. Triploid and tetraploid seedlings, carrying three and four doses of Ht, did not differ significantly. However, three and four doses of Ht conferred a significantly higher level of resistance than did one or two doses. The heterozygote (Ht ht) was less resistant than the n-2n class, although this difference was not statistically significant. However, heterozygous seedlings were significantly less resistant than the 3n-4n class.

Monoploid ht seedlings were significantly more susceptible to leaf blight than were the diploid (ht ht) seedlings. There was no significant difference in susceptibility between 2n, 3n, and 4n seedlings carrying two, three, and four doses, respectively, of ht.

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2. The use of purple embryo marker in screening for twin embryo seeds.

We have been screening large populations of corn seeds for monoploids by crossing our test lines, as female parent, to purple embryo marker. In a population of about 83,000 kernels, we found four seeds, each of which possessed two distinct embryos. On all four kernels, each of the two embryos showed the purple color. It was apparent in two of these kernels that the twin embryo could not have been detected unless the pigment was present. The female parent was line 65:225-1, which is homozygous Ht.

We suggest the use of purple embryo marker to facilitate the detection of such seeds in programs where the primary purpose of research is to uncover kernels with twin embryos.

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1. Linkage intensities of lutescent-1.

The lutescent mutant in maize has been characterized (MNL 39:146-147). Its expression has been found to result from two recessive genes, lutescent-1 (lu_1) and lutescent-2 (lu_2) (MNL 41:150-152). One of these genes, lu_1, has been located on chromosome 5, and preliminary F_2 studies indicated close linkage with a_2.

A testcross was carried out between plants hybrid for lu_1, lu_2, a_2, bm_1, bt_1, and pr, and one homozygous for lu_1, lu_2, and pr. The latter plant was heterozygous for bm_1 and bt_1, but did not carry a_2. The seeds with colorless aleurone resulted from the segregation of a gene other than a_2 since no linkage could be demonstrated between the aleurone color gene and any of the other chromosome 5 markers. The results of this testcross are presented in Table 1. A high percentage of inviability was encountered among seeds homozygous for bt_1, and the values shown have been corrected to allow for this inviability.

Acceptable 3:1 monohybrid ratios were observed for colored vs. white seeds, full vs. brittle seeds, green vs. brown midrib, and green vs. lutescent leaf. It will be noted that 3:1 represents a testcross ratio for green and lutescent since duplicate genes are involved. A 1:1 ratio was observed for purple vs. red seeds. Dihybrid segregations involving lutescent plant, and brittle seed, brown midrib, or red aleurone all