

11. Restoration of fertility to cytoplasmic male-sterile corn.

The cytoplasmic male-sterile inbreds C106-sterile and Tx203Ms used in these studies derive the male-sterile character from the Texas variety Golden June, a derivative of Mexican June.

In contrast to the condition reported by Bauman (1952) in which each inbred of the 250 tested restored fertility completely to the KyS cytoplasmic male-sterile line, few inbreds so far tested restore fertility to the Texas cytoplasmic male-sterile lines. Forty-two inbreds have been tested for genes restoring fertility to Tx203Ms and C106-sterile. The F_1 progenies of Tx203Ms and C106-sterile x Inbreds were classified as: sterile - plants which rarely exerted anthers and dehisced no viable pollen; partially sterile - plants containing a few spikelets with exerted anthers dehiscing viable pollen in variable amounts; fertile - plants all of whose spikelets contained normally exerted anthers dehiscing abundant viable pollen.

Of the inbreds tested four (9.5%) restored fertility completely in the F_1 and four (9.5%) produced F_1 progenies segregating fertile plants. It should be possible to isolate lines homozygous for fertility-restoring genes from inbreds whose F_1 progenies segregate fertile plants, and these could be used in single crosses as effectively as lines already homozygous for fertility-restoring genes. As more inbreds are tested, additional lines will undoubtedly be found which carry fertility-restoring genes, however, the indications now are that the majority of U.S. inbreds lack such genes. It is desirable, therefore, to find other sources of fertility-restoring genes. One such source is to be found among Latin American maize varieties.

A total of 124 varieties of Mexican, Central and South American maize varieties have been tested for fertility-restoring genes. Of the 124 F_1 populations resulting from crossing C106-sterile by various Latin American varieties, 96 were tested in both 1951 and 1952. In only one of these was there a significant variation in classification, probably due to sampling errors. A larger percentage of lines containing fertility-restoring genes was found among the Latin American varieties than among U.S. inbred lines. 13.7% of the varieties tested restored fertility completely in the F_1 , while 45.9% produced F_1 's segregating fertile plants. Besides serving as an additional source of fertility-restoring genes, some of these varieties may also prove to be valuable sources of germplasm for improving corn in the U.S. (Wellhausen et al., 1952).

Data from F_2 and F_3 populations indicate that a single gene is responsible for restoration of fertility in two U.S. inbreds tested, while two genes are indicated in a third inbred. Chi-square tests for goodness of fit to an assumed ratio of 3 fertile: 1 sterile were applied to six F_3 progenies from the cross Tx203Ms x K55. Chi-square values for all of these progenies were low and equivalent P. values ranged from P.10 to P.90. The Chi-square value obtained for an assumed ratio of 3 fertile: 1 sterile from one F_2 progeny of the cross Tx203MS x 127C lay between P.30 and .50. The Chi-square value obtained for an assumed ratio of 9 fertile: 7 sterile in one F_2 progeny of the cross Tx203Ms x K64 lay between P.80 and .90. In the two F_2 progenies, partially sterile and sterile plants were grouped and treated as

steriles. This was done on the assumption that the partially sterile plants do not carry fertility-restoring genes but, due to the action of segregating modifying genes and/or environment, produce some viable pollen. In addition two F₃ populations from the cross Tx203MS x K55 contained only fertile plants, 20 and 23 in the respective populations. One F₃ population of the cross Tx203Ms x 127C contained 56 plants all of which were fertile. These three F₃ populations containing only fertile plants must have resulted from the unconscious selection of F₂ plants homozygous for the fertility-restoring factor. That two factors are involved in fertility-restoration in K64 is indicated by the marked deviation from a 3:1 ratio and by the high P value obtained for the assumed 9:7 ratio.

An investigation of the number of factors involved in fertility-restoration by Latin American lines has been begun. Fertile plants among the F₁ populations of one Mexican and two Bolivian varieties (C106-sterile x Latin American Variety) were backcrossed to C106-sterile. The Chi-square values obtained for the assumed ratio of 1 fertile: 1 sterile in the three backcross progenies were low. These ratios indicate that a single gene is responsible for fertility-restoration.

Knob counts from 58 of the Latin American varieties tested for fertility-restoring genes were obtained from Table 17 of Races of Maize in Mexico (Wellhausen et al., 1952) and from Mangelsdorf and Paxson (this issue of the News Letter). Forty-five varieties from ten countries had average knob numbers ranging from 4.5 to 9.5. Among these, four (16%) of the 25 varieties producing completely fertile or segregating-fertile F₁ progenies had fewer than 5.5 knobs, while nine (45%) of the 20 varieties producing completely sterile or segregating-partially sterile F₁ progenies had fewer than 5.5 knobs. These figures indicate a correlation between high knob number and fertility restoration but do not show whether knobs restore fertility or are associated with genes which do so. Thirteen varieties from Ecuador, Peru, and Bolivia had low average knob numbers, 1.5, 2.5 and 0.57 respectively. No correlation has been found between knob number and fertility restoration in the varieties from these countries.

Meiosis in male-sterile plants of the stocks studied appear to be normal. This is in agreement with the condition reported by Rhoades (1933) in another male sterile. Degeneration of the microspore in sterile plants occurs after the formation of the quartet of pollen grains. Degeneration varies within individual anthers from completely empty pollen grain walls to pollen grains containing varying quantities of protoplasm.

Gabelman (1949) has postulated certain particles one or more of which when present in a pollen grain produce sterility. Rhoades (1950) has suggested that differences in mitochondria might be responsible for cytoplasmic male-sterile plants arising from male-fertile progenies containing the gene *iojap* in the homozygous condition. Pollen mother cells as well as somatic tissues from leaf and glume epidermis, root tip, scutellar and endosperm tissue from C106-sterile and C106-fertile have been examined in an effort to determine whether any visible differences exist between the cytoplasms or cytoplasmic organelles [sic] of fertile and sterile plants. Janus Green B, Pinacyanol, and Neo-tetrazolium chloride have been used to

stain living tissue, while Zirkle's modification of Erliki's fixative and Heidenhains' hematoxylin have been used in sectional material. Neo-tetrazolium chloride seems to be the most promising of the stains used thus far. It permits utilization of the squash technique on both living and fixed material. Since nuclear structure is not brought out with Neo-tetrazolium chloride, young tassels have been divided, half being stained with Neo-tetrazolium chloride, half fixed in alcohol:acetic acid. Aceto-carmine staining of pollen mother cells indicates the meiotic stage of the nuclei in corresponding areas of the other half of the tassel. No differences in the appearance or number of mitochondria have been observed in either the somatic or germinal tissue so far examined. Since pollen degeneration occurs after the completion of meiosis, continued observations of the later stages of pollen formation may uncover some differences between mitochondria in C106-sterile and C106-fertile.

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