

Homozygous Diploid Sugar Beets

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In June 1962, germinating seedlings of 2589C1, an isogenic annual line from the bolting resistant NB1 inbred (5)², were colchicine-treated for the purpose of producing a tetraploid line. From 25 surviving plants, the 5 best chimeras showing predominantly tetraploid vegetative tissue were selected for selfing. Forty-three C₁ seedlings were obtained from seed collected in January 1963. Of these, 39 were tetraploid, 2 were triploid, and 1 was a haploid (monoploid) with 9 chromosomes. The haploid was fairly vigorous, both as to foliage and root growth, and readily distinguished from the sister plants and untreated diploids of the same line by its narrower and more tapered leaves (Figure 1). It flowered profusely during the summer. The flowers were small as compared to those of the diploids. The anthers contained mostly empty pollen grains.



Figure 1.—A vigorous, haploid sugar beet with 9 chromosomes.

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² Numbers in parentheses refer to literature cited.

Because this plant was derived from a self-fertile line, a vegetative increase was made for the purpose of producing a homozygous diploid line by the use of colchicine. Cuttings were made from flower stalks (6) and, during the following winter, 17 grafts were made from root-crown cuttings. Thirteen flower-stalk cuttings survived, making a total of 30 haploid clonal plants. All were to be colchicine-treated except 5 which were produced from flower-stalk cuttings. These were reserved for maintaining the haploid line.

During June 1964, flower-stalk buds began to form in the leaf axils of the crowns where colchicine solutions were applied with a pipette. A 0.3% aqueous solution was chosen since this was the strength used for treating germinating sugar beet seedlings. On some of the plants a 0.3% colchicine solution in 10% glycerine was applied. Since the writer was unfamiliar with colchicine techniques involved in treating axillary buds in sugar beets, the number of applications of the solutions was varied from 1 (using in this case a 0.3% colchicine solution in 10% glycerine) to 7 times through an 8-hour period. All treatments proved equally effective.

A total of 68 gm of good seed was harvested. An initial seed increase was made, using 3 gm of seed which produced 183 seedlings. Cytological examinations showed 171 of these to be diploid; 10 were triploid and 2 were tetraploid. They were strikingly uniform in vigor and in leaf shape. The seed germinated promptly and uniformly. This homozygous line is now known as C5600.

Levan (4), apparently for the first time on record, found in his laboratory also a haploid sugar beet in the progeny of a colchicine-treated diploid. He attributed its origin to possible damage, by the colchicine treatment, of 1 of the gametes involved. He stated that "one pollen grain, for instance, may have been able to stimulate embryo development although incapable of fertilization."

Levan's haploid beet was in some respects similar in morphological characteristics to mine. Leaves were narrower and more numerous than those of the diploids. The flower stalks were more slender, and the floral parts decidedly smaller and more delicate, than in the diploid. The 2 plants differed in other respects. The top shoot of his plant gradually became fasciated. The numerous malformed flowers had 6 instead of the normal 5 petals and stamens.

Although Levan apparently made no effort to produce a homozygous diploid line (perhaps his original diploid line was self sterile), he did make some observations on the cytology of his haploid plant. He observed paired segments in the pachytene nuclei. Chiasmata were found at diakinesis and at metaphase I. Bivalents and trivalents (6 of 287 cells) were found in 40% of the cells. To account for pairing in the haploid when only 1 chromosome of each kind is present, he felt it reasonable to assume "that the main course of pairing is genetically fixed in the species, the haploid trying to imitate the diploid as closely as possible." He believed he had evidence that chiasmata or chromosome pairing in haploids are caused by pure chance and that these should not be taken as proof of homologies within the genome.

However, Kimber and Riley (3), in a recent monograph on haploid angiosperms, state that it is now widely assumed that chromosome pairing, when it occurs in monoploids (haploids), implies that these chromosomes possess homologous segments which originally arose through translocations. Homologies, also, may have originated through aneuploidy in the past, certain chromosomes having been duplicated either completely or in part. Also, pairing "may arise from homologies due to an archaic polyploid origin, of which pairing in the monoploid is the only trace, the remote diploid ancestors now being extinct."

Fischer (1), in his studies on twinning in sugar beets, found a haploid as a member of a set of twins—a fairly common occurrence in haploid angiosperms (3). All the haploids reported in species of cotton (*Gossypium*) arose in this manner. Less commonly, haploids may arise after experimental treatment, as did the sugar beet plants described in this paper. Fischer made no mention of having attempted to develop a homozygous diploid sugar beet line.

The value of homozygous material is reflected in work conducted to utilize vegetative increases for experimental work. Powers *et al.* (7) developed a technique for dividing sugar beet roots to preserve the genotype. Owen (6) succeeded in making crown-bud cuttings and cuttings from semi-vegetative flower stalks for use in asexual propagation. Harper and Tennant (2) devised a technique for bisecting young beet seedlings to form identical pairs, so that valid comparisons could be made with seedling beets. The use of homozygous seed should greatly facilitate basic experimental work with the physiology, chemistry and genetics of the sugar beet.

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