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Research Notes: University of Nevada

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with some III's and V's. Seed will be available for distribution after the 1974 harvest from R. L. Bernard, U.S. Regional Soybean Laboratory, Urbana, Illinois 61801.

1. Exploitation of leaf mosaicism for determination of allelic relationship in Glycine max.

_Glycine max_ (L.) Merrill (soybean) is said to have at least 18 loci responsible for the development of chlorophyl (Bernard and Cremeens, 1970). One of these, _Y_11_, discovered by Weber and Weiss (1959), is characterized by the development of golden yellow color of the leaves and stem in homozygous (_Y_11Y_11_) combination. The heterozygous plants are light green and differ from the _Y_11Y_11 homozygotes which have normal, dark green color. The two simple and the first compound leaves of the heterozygous plants are dotted with dark green, yellow and twin or double (dark green-yellow) spots. Origin of some of these spots, particularly those of twins, has been attributed to the process of somatic crossing over (see Vig and Paddock, 1968; Vig, 1971, 1972, 1973a,b). Also, the frequency of these spots can be increased several fold by treating the seed with chemicals, e.g. caffeine, mitomycin C, etc.

Another gene, _Y_9, in homozygous recessive _Y_9Y_9_ combination produces a bright greenish yellow leaf and stem color (Probst, 1950). _Y_9Y_9_ or _Y_9Y_9_ plants are dark green. Considering the two genes and their alleles so far discussed, one gets the following combinations: _Y_9Y_9_ - _Y_11Y_11_ = dark green; _Y_9Y_9_ - _Y_11Y_11_ = light green; _Y_9Y_9_ - _Y_11Y_11_ = yellow, lethal; _Y_9Y_9_ - _Y_11Y_11_ = bright greenish yellow; _Y_9Y_9_ - _Y_11Y_11_ = not known.

Traditionally, relationships between genes (or alleles) can be studied by raising hybrids like _Y_9Y_9_ - _Y_11Y_11_ and analyzing the segregating populations. However, we decided to make use of induced somatic mosaicism for such a study. The reasoning is as follows: if _Y_9Y_9_ - _Y_11Y_11_ plants are treated with a known recombinogen or mutagen (say caffeine or mitomycin C) one should...
expect a mutation of the gene \( y_9 \) to \( y_g \), thus changing the color of the affected colony of cells to dark green. Another possible mutation of \( y_{11} \) to \( y_{11} \) will not alter the phenotype of the resulting \( y_g y_g y_{11} y_{11} \) colony. If, on the other hand, genes \( y_9 \) and \( y_{11} \) are alleles, rather than occupying two loci, one may also expect some dark green sectors \( (y_g y_{11}) \), considering that genotype of bright greenish yellow plant is \( y_g y_{11} \) \((y_g, y_{11} \) are alleles) and not \( y_g y_g y_{11} y_{11} \). In the latter case, however, one may also expect a few yellow \( (y_g y_{11}) \) spots if \( y_{11} \) mutates to \( y_{11} \) (and if \( y_{11} > y_g \)). In case of \( y_g y_g y_{11} y_{11} \) genotype, no yellow spots are possible unless one postulates an unexpected double mutational event involving both \( y_{11} \)’s.

In one series of experiments, the seeds of Clark x T135 (segregating for \( y_g y_g \)), T135 \((y_g y_g)\), L65-1237 \((Y_{11} y_{11}) \) segregant) and T219 \((y_{11} y_{11}) \) segregant) were planted without pretreatment. In all cases, all three types of spots previously seen on \( y_g y_{11} \) plants were observed. The color of the spots on the \( y_g y_g \) plants resembled closely the color of \( y_{11} y_{11} \) leaves. In another experiment, seeds of T219, L65-1237, T135, Clark x T135, T136 \((Y_g y_g)\) and Clark x T136 (segregating for \( Y_g y_g \)) were planted with and without treatment with mitomycin C. The control \( y_g y_g \) had a few spots of all three types found on \( y_{11} y_{11} \) leaves. The frequency in the treated material went up by 2 to 6 times in case of both \( y_g y_g \) and \( y_{11} y_{11} \) plants. No spots, however, were observed on the \( Y_g y_g \) plants.

A third set of experiments was performed using \( y_g y_g \) (T135) seed soaked in water, or solution of mitomycin C, or caffeine. Spot frequencies per leaf ranged from 0.56 for water-soaked (control) material, to 0.94 for 0.002% mitomycin (18 hr) and 9.49 for 0.1% caffeine (18 hr). All three types of spots were found in almost equal frequencies. Several more experiments gave parallel results.

The data reported above raise some interesting points. First, the appearance of yellow spots on the T135 leaves indicates a relationship between \( Y_{11}, y_{11} \) and \( y_g, y_g \). The most convincing solution to this puzzle appears to consider \( Y_{11} \) and \( y_g \) as the same gene and \( y_g \) \((or \ y_{11})\), \( y_g, y_{11} \) as alleles. Thus \( y_g \) in \( y_g y_g \) can mutate to either \( y_g (= Y_{11}) \) or to \( y_{11} \). This explains the origin of two types of single spots on T135 leaves. Thus \( y_g (= Y_{11}) \) and \( y_{11} > y_g \), but \( y_g (= Y_{11}) \) is incompletely dominant over \( y_{11} \).
Another problem is the explanation of the origin of twin spots on the \( Y_gY_g \) leaves. It requires a complementary exchange between the two \( Y_g \) genes producing \( Y_g (= Y_{11}) \) and \( Y_{11} \) in a single step. I wish to advance the following hypothesis: the gene \( Y_g \) is composed of a DNA segment which carries two tandem repeats. An addition of another repeat causes the partial loss of activity of the final product (the protein produced). This was \( Y_g \). A loss of one of the repeats in \( Y_g \) causes almost the total loss of activity of the product. This is \( Y_{11} \). In a somatic cell with some arrangement for interaction between homologous chromosomes, there is every chance of a given repeat segment loosely pairing with any other similar segment found in recombination required by somatic crossing over. Such unequal crossing over in \( Y_gY_g \) homozygote will produce \( Y_gY_g - Y_{11}Y_{11} \) cells which will express as a double spot colony. This hypothesis is also compatible with spots on \( Y_{11}Y_{11} \) plants.

In view of the above, I suggest that possible allelic relationship between \( Y_g \) and \( Y_{11} \) be explored, but the question of redesignation of \( Y_g \) and \( Y_{11} \) be postponed until further evidence is available from crosses and segregating populations.

The lack of yellow spots on other genes tested may indicate non-allelic relationship with \( Y_g \) series.

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References


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