ADDENDUM

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The effect of intermittent light on chlorophyll content of selected virescent mutants

Selective accumulation of chlorophyll a in dark-grown seedlings of bean (J. H. Argyroudi-Akoyunoglou and G. Akoyunoglou, 1970, Plant Physiol. 46:247) and pea (C. J. Arntzen, personal communication) subsequently exposed to intermittent light-dark cycles has also been shown in young maize seedlings (Hopkins, unpublished observations). In our laboratory, this phenomenon is being employed to produce chlorophyll b-deficient maize chloroplasts for the purpose of studying functional organization of chloroplast membranes. As an adjunct to this study, selected virescent mutants of maize were surveyed to learn whether these mutants respond in a similar fashion to intermittent light.

Seedlings were grown at 25 C under either 16 hr photoperiods (Controls) or intermittent light-dark cycles consisting of 2 min light and 58 min darkness (ImL); light intensity of approximately 800 ft-c was provided by a combination of cool white fluorescent and incandescent lamps. Seedlings were harvested at ten days and the leaf tissue extracted into 80% acetone. The first leaf was excluded, since earlier experiments had indicated that chl a/chl b ratios were significantly lower in the first leaf than in subsequent leaves. Chlorophyll content was calculated from absorbance values according to Arnon (1949, Plant Physiol. 24:1). The results of the survey are shown in Table 1.

Table 1. Effect of an intermittent light regime on chlorophyll composition of selected virescent mutants.

Genotype	Total Chl (mg/g f.w.)		Chl a/b Ratio		ImL-Cont.
	Controls	ImL	Controls	ImL	Cont.
+/+	1.52	0.17	2.6	9.1	2.5
Group I					
v/v	0.79	0.04	2.8	9.4	2.4
v2/v2	1.55	0.20	3.1	9.3	2.0
v3/v3	1.22	0.06	1.6	7.8	3.9
v4/v4	1.07	0.13	2.7	9.6	2.6
v5/v5	1.18	0.16	2.6	8.9	2.4
v16/v16	0.70	0.07	1.9	6.2	2.3
v17/v17	1.90	0.15	3.0	8.0	1.7
Group II					
v12/v12	0.83	0.12	2.1	3.3	0.6
v18/v18	0.68	0.09	3.4	4.8	0.4

Several observations may be drawn from the data:

1. Under control conditions of low light (ca. 800 ft-c) and 25 C, the virescent character was not strongly expressed. In fact, only v, v12, v16 and v18 showed any signs of virescence, as suggested by the slightly lowered chlorophyll contents. In our experience, pronounced phenotypic expression of the virescent character is obtained only under conditions of high light intensity (>1500 ft-c) and/or lower temperature.
2. In all genotypes surveyed, intermittent light conditions brought about a severe reduction in the total chlorophyll content of the seedlings; these reductions ranged between 85% and 95%.

3. There is evident some variation in the normal chl a/chl b ratios under control conditions, with ratios ranging from a low of 1.6 to a high of 3.1.
4. The virescent stocks may be placed in one of two groups on the basis of their response to intermittent light. In the first group are those in which, like the wild-type, the chl a/chl b ratio increases some 2-3 times under the intermittent light regime. The second group, comprised of only two of the mutants surveyed thus far, is characterized by a minimal enrichment in chlorophyll a content under intermittent light.

This latter observation is perhaps the most interesting. The basis for the absence of any selective accumulation of chlorophyll a in these two genotypes is not known at this time. Previous work in this laboratory has shown that wild-type maize seedlings produced under intermittent light are characterized by mesophyll chloroplasts lacking both grana and a specific chl a-b/protein complex, but possessing fully active photosystems I and II (unpublished observations). Whether either the v12 or v18 mutants will provide further insights into the role of chlorophyll b in the organization of chloroplast membranes remains to be seen.

On the other hand, it becomes increasingly clear that the traditionally vague description of virescence simply according to color and rate of greening belies the complexity of the system(s). The grouping of the virescents into two distinct classes according to their response to intermittent light may prove to be a useful first step toward the eventual biochemical characterization of the phenomenon.

W. G. Hopkins

Possibilities for gibberellin-male sterile relationships in corn--a proposal

Alternatives to cms for breeding programs have been proposed. Two possibilities, not yet fully explored, include nuclear-based male steriles which might be chemically reverted and the chemically induced sterility of normal plants.

In many species, various aspects of stamen growth and pollen production involve giberellin-dependent or -sensitive steps. In some, male sterility can be completely reverted with the application of gibberellins (Can. J. Bot. 51:2473; Euphytica 18:106). In others, it is known that normal stamen development is correlated with high endogenous levels of gibberellin (Plant Cell Physiol. 16:337; J. Am. Soc. Hort. Sci. 97:189 and J. Exp. Bot. 25:1004). The normal growth of filaments involves stamen-produced gibberellin (Amer. J. Bot. 54:971).

In corn, to our knowledge, no study of gibberellin-dependent stamen development has been reported, though the observations of Sladky (Biol. Plantarum 11:208) and Scheverbecke (C. R. Acad. Sci. Paris 260:5085) implicate gibberellins in tassel development and anther extrusion. Two reports (Ann. Mo. Bot. Gard. 46:19; Pl. Physiol. 56 Suppl.:44) of the exogenous application of gibberellin describe an inhibition of stamens of normal plants and therefore suggest a possible alternative interpretation of the role of gibberellin and stamen development.

However, in view of the large number of maize genes which are known to lead to reduced pollen fertility and the clear implication from other species that gibberellins could be involved, we suggest that the fertility of some male-sterile maize mutants might be restored by the administration of a gibberellin. This genetic material, which would respond to an exogenous application, could be a powerful adjunct to the breeder's arsenal for producing hybrid corn.

The opposite approach should also be attempted. Cultivars and procedures should be researched to ascertain if an application of anti-gibberellins could be used to induce male sterility. From a practical standpoint this procedure is less desirable than the first, but might be of value in certain situations.

If one accepts the above line of thought, it follows that:

a) A search for GA sensitive and/or reversible male-sterile mutants in corn should be started as soon as possible;