Theor Appl Genet (1985) 70:349–354

Conditional lethality involving nuclear and cytoplasmic chlorophyll mutants in soybeans*

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Received April 22, 1984; Accepted January 25, 1985
Communicated by P. L. Pfahler

Summary. A conditionally lethal phenotype occurred when a nuclear chlorophyll mutant (y_{20}-k_2) was present with a cytoplasmic chlorophyll mutant (*cyt*-Y_2) in soybean (*Glycine max* [L.] Merr.). Nuclear mutant y_{20}-k_2, Genetic Type Collection Number T253, has yellow foliage, tan-saddle-pattern seed and is viable. The y_{20}-k_2 mutant cannot be separated by classical genetic tests into two separate components, y_{20} (yellow foliage) and k_2 (tan-saddle-pattern seed). Mutant *cyt*-Y_2, T275, is inherited cytoplasmically, has yellow foliage, and is viable. The genotype *cyt*-Y_2 y_{20}-k_2/y_{20}-k_2 is a conditional lethal; the genotype is lethal under field conditions, but plants survive under greenhouse conditions. This interaction is unique to y_{20}-k_2. This conditionally lethal genotype may be useful in molecular studies on the interaction between nuclear and plastid genomes.

Key words: Cytoplasm – *Glycine max* – Chloroplast – Interaction

Introduction

The interplay between gene expressions of the mitochondrial and plastid genomes with that of the nuclear genome is of major interest to biologists (Parthier 1982; Leaver and Gray 1982). In eukaryotes, mitochondria and plastids are the predominant carriers of extrachromosomal genetic information.

The biogenesis and function of chloroplasts result from a complex interaction between nuclear and plastid genomes. Our understanding of this cooperation can be enhanced through the study of nuclear mutants and cytoplasmic mutants and their interaction (Schotz 1970; Vedel and Remy 1983; von Wettstein 1981).

Three cytoplasmically inherited chlorophyll mutants are known in soybeans (*Glycine max* [L.] Merr.). Mutant *cyt*-G_1 has green cotyledons and seed coat (Terao 1918); mutants *cyt*-Y_2, (Genetic Type Collection Number T275 [Palmer and Mascia 1980]) and *cyt*-Y_3 (Shoemaker et al. 1985) affect chlorophyll levels in foliage and are viable. About 20 nuclearly inherited chlorophyll-deficient soybean mutants have been described (Bernard and Weiss 1973).

A nuclear-cytoplasmic interaction has been observed when *cyt*-G_1 is present with nuclear genes gy_3 (g determines yellow seed coat, and y_3 determines yellow foliage). Plants that are *cyt*-$G_1ggy_3y_3$ have yellow foliage and yellow cotyledons; *cyt*-G_1 is not expressed. In crosses of these plants as female parent with either G_1 or Y_3 as the male parent, the resultant hybrid seeds have green cotyledons, whereas the self-pollinated seeds of the female plant have yellow cotyledons (Palmer, unpublished).

There are no reports about nuclear-cytoplasmic interactions between cytoplasmic mutant *cyt*-Y_2 and the nuclearly inherited chlorophyll-deficient mutants. Reciprocal crosses between *cyt*-Y_2 and four of these nuclear mutants, y_{10}, y_{12}, y_{13}, and y_{18}, showed no interaction; all effects were independent (Palmer and Mascia 1980). In crosses with a fifth nuclear mutant, y_{20}-k_2, Genetic Type Collection Number T253, some anomalies were observed. This mutant is a chlorophyll-deficient plant with tan-saddle-pattern seeds (y_{20}-k_2). Saddle pattern refers to pigmentation on the seed of the area extending on either side of the hilum region, covering approximately half of the seed coat. Gene symbol k_2 has been assigned to the tan-saddle-pattern on the basis of allelism tests conducted with other saddle-pattern mutants (Rode and Bernard 1975). Palmer (1984) has shown that the yellow factor of T253 is not allelic to y_3, y_5, y_9, y_{11}, y_{13}, *or* y_{18}. The yellow factor was assigned gene symbol y_{20}. Furthermore, the y_{20}-k_2 mutant cannot be separated by classical genetic tests into two

* This is a joint contribution of North Central Region, USDA ARS, and Journal Paper No. J-11429 of the Iowa Agriculture and Home Economics Experiment Station, Ames, Iowa, and the Agriculture Experiment Station, Univ. of Puerto Rico, Mayaguez Campus, Mayaguez, Puerto Rico 00708. Projects 2471 and 2475. The research was supported in part by the Iowa Soybean Promotion Board.

separate components, y_{20} and k_2. T253 is written y_{20}-k_2 to indicate very close linkage or pleiotropy (Palmer 1984). In crosses of *cyt*-Y_2 as female parent with y_{20}-k_2, tan-saddle-pattern seeds were not observed on F_2 plants. In the reciprocal cross, however, seeds with the tan-saddle-pattern were found.

Our objective, therefore, was to characterize the type of nuclear-cytoplasmic interaction, genetically or environmentally induced, between y_{20}-k_2 and *cyt*-Y_2.

Materials and methods

Hybrid seeds were obtained following the procedure described by Walker et al. (1979). Crossing and progeny testing were done at the Bruner Farm, Ames, Iowa, and at the Iowa State University – University of Puerto Rico Soybean Breeding Nursery, at the Isabela Substation, Isabela, Puerto Rico, from 1979–1982. To verify success of cross-pollinations, nuclear mutants for flower color and pubescence color were used as marker genes.

Plants of *cyt*-Y_2 were crossed with plants homozygous for the saddle-pattern seed mutants k_1, k_2, k_3, i^k, and with y_{20}-k_2 (Table 1). The normal green sib line of *cyt*-Y_2, *cyt*-G_2 (Palmer and Mascia 1980), also was crossed with plants homozygous for k_2 and y_{20}-k_2. Reciprocal crosses were obtained for each of these combinations.

To study the nature of the nuclear-cytoplasmic interaction three experiments were designed to identify the genotype *cyt*-Y_2 y_{20}-k_2/y_{20}-k_2 if it were present but not expressed. In the first experiment, 19 F_2 plants from the self-pollination of *cyt*-Y_2Y_{20}-K_2/y_{20}-k_2 served as female parents in crosses with k_2 and y_{20}-k_2. The identity of the 19 F_2 plants was maintained, and at least 8 hybrid seeds per plant and combination were obtained to assure recovery of the heterozygotes (Y_{20}-K_2/y_{20}-k_2) at the 99.5% probability level (Mather 1951).

In the second experiment, F_1 plants that were either *cyt*-Y_2Y_{20}-K_2/y_{20}-k_2 or *cyt*-Y_2K_2/k_2 were used as female parents in crosses with k_2 and y_{20}-k_2. Ten F_1 plants were used, and a minimum of 21 hybrid seeeds of the four genetic combinations were obtained. Hybrid seeds of the combination *cyt*-Y_2Y_{20}-K_2/y_{20}-$k_2 \times y_{20}$-k_2 were germinated in a growth chamber, and seedlings were transplanted to the greenhouse.

In a third experiment, plants suspected to be of the genotype *cyt*-Y_2 y_{20}-k_2/y_{20}-k_2 (identified in the second experiment) were used as male parents in crosses with k_2 and y_{20}-k_2.

To test for gametophytic and sporophytic lethality, *cyt*-G_2 or *cyt*-Y_2 plants were crossed with K_2/k_2 and Y_{20}-K_2/y_{20}-k_2 plants. Pollen fertility was estimated on the basis of the number of plump pollen grains that stained red-brown with I_2KI solution. Ovule abortion was calculated as the percentage of unfertilized ovules, and seed abortion as the percentage of fertilized ovules that aborted at various stages of seed development, in relation to the total number of seeds per plant plus ovule abortions plus seed abortions.

Chi-square tests (Snedecor and Cochran 1980) were used to test for goodness of fit between observed and expected ratios for green : yellow plant color and absence : presence of tan-saddle-pattern seeds.

Table 1. Soybean genetic lines used in this study

Mutant	No.[a]	Description
cyt-Y_2	T275	cytoplasmic yellow, viable
k_1	T153	saddle-pattern seed
k_2	T239	tan-saddle-pattern seed
k_3	T238	saddle-pattern seed
i^k	L70-4204	saddle-pattern seed
y_{20}-k_2	T253	chlorophyll deficient tan-saddle-pattern seed, viable

[a] Genetic Type Collection Number except L70-4204, which is 'Clark' – *i* (L66-14)[6] × 'Black Eyebrow'

Results

In the first experiment the k_2 phenotype was not found on seeds from F_2 plants from crosses of *cyt*-$Y_2 \times y_{20}k_2$; however, tan-saddle-pattern seeds were observed in progeny from the reciprocal cross (Table 2). Expression of saddle color and F_2 Mendelian ratios were observed for reciprocal crosses of *cyt*-Y_2 with k_1, k_2, k_3, and i^k (Table 2). These data indicate that y_{20}-k_2 interacted with *cyt*-Y_2 but k_1, k_2, k_3, and i^k did not. The average germination percentage of F_2 seeds of the cross *cyt*-$Y_2 \times y_{20}$-k_2 was 68.7%, significantly lower than that of the reciprocal cross, which was 86.5% (Table 2). This interaction was confirmed by progeny tests of 170 F_2 plants from the cross *cyt*-$Y_2 \times y_{20}$-k_2. Germination percentages were between 60 and 70% in 109 of the 170 progeny rows, suggesting that 109 of 170 (ca. 2/3) F_2 plants were of the genotype *cyt*-Y_2Y_{20}-K_2/y_{20}-k_2. A total of 6,319 F_3 plants from these progeny rows were checked for the presence of saddle-pattern seeds, and no tan-saddle-pattern seeds were found.

To test the hypothesis that the effect between the nuclear and cytoplasmic mutants was unique to the combination of *cyt*-Y_2 with y_{20}-k_2, cytoplasmic normal green (*cyt*-G_2) sib plants of *cyt*-Y_2 were crossed recipro-

Table 2. F_2 data for various soybean saddle-pattern mutants crossed reciprocally to *cyt*-Y_2

Parents		No. of F_2 plants		% germination
♀	♂	No saddle	Saddle	
cyt-Y_2	k_1	105	38	83.7
cyt-Y_2	k_2	537	183	86.8
cyt-Y_2	k_3	141	56	90.1
cyt-Y_2	i^k	178	61	88.2
cyt-Y_2	y_{20}-k_2	1,353	0	68.7**
k_1	*cyt*-Y_2	436	184	88.5
k_2	*cyt*-Y_2	253	78	90.6
k_3	*cyt*-Y_2	184	50	86.4
i^k	*cyt*-Y_2	308	113	83.7
y_{20}-k_2	*cyt*-Y_2	787	238[a]	86.5**

[a] All plants were yellow (y_{20} y_{20})

** Significantly different at $P < 0.05$ level

cally with k_2 and y_{20}-k_2 (Table 3). In all cases, a good fit to a 3 : 1 ratio for absence : presence of saddle pattern was observed, with similar germination percentages for the progeny of the four combinations obtained. These data indicate that y_{20}-k_2 interacted with *cyt*-Y_2 but did not interact with *cyt*-G_2.

Three possible explanations for the apparent lack of the k_2 phenotype on seeds from F_2 and F_3 plants in the cross *cyt*-$Y_2 \times y_{20}$-k_2 are: 1) the male or female y_{20}-k_2 gametes are lethal in *cyt*-Y_2 cytoplasm (i.e., certation or gametophytic lethality); 2) *cyt*-$Y_2\, y_{20}$-k_2/y_{20}-k_2 genotype is a sporophytic lethal; or 3) the genotype *cyt*-$Y_2\, y_{20}$-k_2/y_{20}-k_2 occurs but k_2 is either altered or not expressed. To test these three hypotheses, F_1 plants of various genetic combinations involving *cyt*-G_2 and *cyt*-Y_2 as female parents with K_2/k_2 and Y_{20}-K_2/y_{20}-k_2 were obtained, and pollen, ovule, and seed abortions were recorded (Table 4). Results showed that gametophytic lethality, either pollen or ovule, is not the explanation for failure to find the k_2 phenotype on seeds from F_2 plants of the cross *cyt*-$Y_2 \times y_{20}$-k_2. In addition, sporophytic lethality at the developing-seed stage is not a plausible explanation. The seed-abortion percentage observed in these crosses was within the normal range for soybean (Palmer and Heer 1984).

Under the assumption of no interaction between *cyt*-Y_2 and y_{20}-k_2, self-pollination of *cyt*-$Y_2 Y_{20}$-K_2/y_{20}-k_2 plants is expected to produce 1 : 2 : 1 genotypic ratios in the F_2 generation of Y_{20}-K_2/Y_{20}-$K_2 : Y_{20}K_2/y_{20}$-$k_2 : y_{20}$-k_2/y_{20}-k_2. To test this assumption, the second experiment was conducted. F_2 plants from self-pollination of *cyt*-$Y_2 Y_{20}$-K_2/y_{20}-k_2 were crossed reciprocally with k_2 (Table 5) and y_{20}-k_2 (Table 6). In crosses with k_2 (Table 5), a significant phenotypic ratio of 7 : 12 : 0 was observed instead of the expected 4.75 : 9.50 : 4.75 and 3 : 1. Similarly, we observed a 7 : 12 : 0 ratio with y_{20}-k_2 as female parent but a 19 : 0 : 0 ratio with y_{20}-k_2 as male parent (Table 6). Both ratios are significantly different from the expected 4.75 : 9.50 : 4.75 and 3 : 1. Progenies of the plants suspected to be Y_{20}-K_2/y_{20}-k_2 differed when crossed as female parent with k_2 (Table 5) or with y_{20}-k_2 (Table 6). These tests failed to identify the *cyt*-$Y_2\, y_{20}$-k_2/y_{20}-k_2 genotype. Progeny with genotype *cyt*-$Y_2 Y_{20}$-K_2/y_{20}-k_2 were identified when y_{20}-k_2 was female parent but not when it was used as male parent.

In cross pollinations of *cyt*-$Y_2 K_2/k_2$, and of *cyt*-Y_2 Y_{20}-K_2/y_{20}-k_2 F_1 plants (♀) with k_2 or y_{20}-k_2, all hybrid plants were yellow (Table 7). The observed ratios for the absence : presence of tan-saddle were not significantly different from the expected 1 : 1 ratio,

Table 3. F_2 data of reciprocal soybean crosses of k_2 and y_{20}-k_2 with *cyt*-G_2

Parents		No. of F_2 plants		% germination
♀	♂	No saddle	Saddle	
cyt-G_2	k_2	493	163	91.9
cyt-G_2	y_{20}-k_2	516	160[a]	90.1
k_2	*cyt*-G_2	501	15	90.6
y_{20}-k_2	*cyt*-G_2	708	232[a]	93.3

[a] All plants were yellow ($y_{20}\, y_{20}$)

Table 4. Test for gametophytic and sporophytic lethality among various soybean genotypes

Genotype of F_1 plants	No. of F_1 plants	Abortion (%)		
		Pollen	Ovule	Seed
cyt-G_2 K_2/k_2	5	4.8	5.6	8.2
cyt-G_2 Y_{20}-K_2/y_{20}-k_2	6	4.3	3.0	9.6
cyt-$Y_2 K_2/k_2$	11	1.7	3.4	5.2
cyt-Y_2 Y_{20}-K_2/y_{20}-k_2	8	2.3	4.2	6.7

Table 5. Genetic tests with k_2 seeking to identify the F_2 genotype *cyt*-$Y_2\, y_{20}$-k_2/y_{20}-k_2 in soybean

Parents		Expected phenotypic ratios				Expected genotypic ratio	Observed no. of F_2 plants
F_2 plants		Plant color		Saddle pattern			
♀	♂	Green	Yellow	Absent	Present		
cyt-Y_2 Y_{20}-K_2/Y_{20}-K_2	k_2	0	1	1	0	1	7
cyt-Y_2 Y_{20}-K_2/y_{20}-k_2	k_2	0	1	1	1	2	12
cyt-$Y_2\, y_{20}$-k_2/y_{20}-k_2	k_2	0	1	0	1	1	0
	F_2 plants						
k_2	*cyt*-Y_2 Y_{20}-K_2/Y_{20}-K_2	1	0	1	0	1	7
k_2	*cyt*-Y_2 Y_{20}-K_2/y_{20}-k_2	1	0	1	1	2	12
k_2	*cyt*-Y_2 y_{20}-k_2/y_{20}-k_2	1	0	0	1	1	0

Table 6. Genetic tests with y_{20}-k_2 seeking to identify the F_2 genotype *cyt*-Y_2 y_{20}-k_2/y_{20}-k_2 in soybean

Parents		Expected phenotypic ratios				Expected genotypic ratio	Observed no. of F_2 plants
F_2 plants		Plant color		Saddle pattern			
♀	♂	Green	Yellow	Absent	Present		
cyt-Y_2 Y_{20}-K_2/Y_{20}-K_2	y_{20}-k_2	0	1	1	0	1	19
cyt-Y_2 Y_{20}-K_2/y_{20}-k_2	y_{20}-k_2	0	1	1	1	2	0
cyt-Y_2 y_{20}-k_2/y_{20}-k_2	y_{20}-k_2	0	1	0	1	1	0
	F_2 plants						
y_{20}-k_2	*cyt*-Y_2 Y_{20}-K_2/Y_{20}-K_2	1	0	1	0	1	7
y_{20}-k_2	*cyt*-Y_2 Y_{20}-K_2/y_{20}-k_2	1	1	1	1	2	12
y_{20}-k_2	*cyt*-Y_2 y_{20}-k_2/y_{20}-k_2	0	1	0	1	1	0

Table 7. Genetic tests with saddle pattern and plant color seeking to identify genotype *cyt*-Y_2 y_{20}-k_2/y_{20}-k_2 using F_1 soybean plants

Parents		Expected phenotypic ratios		Observed no. of plants	
F_1 plants		Saddle pattern			
♀	♂	Absent	Present	Absent	Present
cyt-Y_2 K_2/k_2	k_2	1	1	15	19
cyt-Y_2 K_2/k_2	y_{20}-k_2	1	1	9	12
cyt-Y_2 Y_{20}-K_2/y_{20}-k_2	k_2	1	1	19	16[a]
cyt-Y_2 Y_{20}-K_2/y_{20}-k_2	y_{20}-k_2	1	1	16[a]	2[b]
		Plant color			
		Green	Yellow	Green	Yellow
cyt-Y_2 K_2/k_2	k_2	0	1	0	34
cyt-Y_2 K_2/k_2	y_{20}-k_2	0	1	0	21
cyt-Y_2 Y_{20}-K_2/y_{20}-k_2	k_2	0	1	0	35
cyt-Y_2 Y_{20}-K_2/y_{20}-k_2	y_{20}-k_2	0	1	0	29[c]

[a] Plants progeny tested and data are presented in text

[b] Plants were yellow with tan saddle and were suspected to be *cyt*-$Y_2 y_{20}$-k_2/y_{20}-k_2. Data confirming this genotype are presented in Table 8

[c] Eleven yellow seedlings died and absence or presence of tan-saddle-pattern seed could not be determined

except in the cross of *cyt*-$Y_2 Y_{20}$-K_2/y_{20}-$k_2 \times y_{20}$-k_2. For this combination we observed 16 no saddle : 2 saddle plants, a ratio that was significantly different from the expected 9 : 9 (Table 7). Eleven F_1 seedlings from this cross died several weeks after being transplanted from the growth chamber to the greenhouse (Table 7). The two tan-saddle plants were suspected to be *cyt*-Y_2 y_{20}-k_2/y_{20}-k_2, and were used as male parents in crosses with k_2 and y_{20}-k_2 in a third experiment (Table 8). The expected and observed green : yellow plants and the absence : presence of tan-saddle fit perfectly for both crosses. We concluded, therefore, that the testcrosses had identified the genotype *cyt*-Y_2 y_{20}-k_2/y_{20}-k_2. We believe that the 11 seedlings that died (Table 7) also were of the genotype *cyt*-Y_2 y_{20}-k_2/y_{20}-k_2. The number of hybrid seeds we worked with in these crosses are normal for soybeans. Artificial hybridizations in this species are a laborious task, because flowers are very small. An experienced crosser may perform about 24 pollinations in 1 h (Walker et al. 1979) and about 50%

Table 8. Confirmation of genotype *cyt-*Y_2 y_{20}-k_2/y_{20}-k_2 by using saddle pattern and plant color in soybean

Parents		Expected phenotypic ratios		Observed no. of plants	
		Saddle pattern			
♀	♂	Absent	Present	Absent	Present
k_2	*cyt-*Y_2 y_{20}-k_2/y_{20}-k_2	0	1	0	2
y_{20}-k_2	*cyt-*Y_2 y_{20}-k_2/y_{20}-k_2	0	1	0	2[a]
		Plant color			
		Green	Yellow	Green	Yellow
k_2	*cyt-*Y_2 y_{20}-k_2/y_{20}-k_2	1	0	2	0
y_{20}-k_2	*cyt-*Y_2 y_{20}-k_2/y_{20}-k_2	0	1	0	2[a]

[a] Plants progeny tested and data are presented in text

of those will set seed. This number could vary, however, depending on environmental conditions and genotype of both male and female parents.

Forty seeds from the self-pollination of each of the two confirmed plants with genotype *cyt-*Y_2 y_{20}-k_2/y_{20}-k_2 (Table 8) were germinated in the growth chamber, transplanted to the greenhouse, and placed under shade cloth. Twenty plants were grown to maturity for seed increase, and 20 were transplanted to the field at the first trifoliolate stage. Even though these field transplants were shaded and given water, they died within 8–10 days. In addition, 20 seeds from self-pollination of each of the two *cyt-*Y_2 y_{20}-k_2/y_{20}-k_2 plants were planted directly in the field and watered. Seeds germinated, but the seedlings died before the unifoliolates were expanded. All 60 seedlings from each of the two plants had the expected yellow foliage phenotype and the 20 plants grown for seed increase produced all tan-saddle-pattern seeds.

Thirty seeds from self-pollination from each of the 16 plants expected to be *cyt-*Y_2 Y_{20}-k_2/y_{20}-k_2 (Table 7, saddle seed) from the cross *cyt-*Y_2 Y_{20}-K_2/y_{20}-$k_2 \times k_2$, and from each of the 16 plants expected to be *cyt-*Y_2 Y_{20}-K_2/y_{20}-k_2 (Table 7, non-saddle seed) from the cross *cyt-*Y_2 Y_{20}-K_2/y_{20}-$k_2 \times y_{20}$-k_2 were planted as progeny rows in the sandbench in the greenhouse. For the two genotypes, segregation for yellow viable : yellow lethal was 279 : 91, and 370 : 127, respectively. These results fit the expected 3 : 1 ratio and confirmed the heterozygous condition, Y_{20} y_{20}, of the plants.

Discussion

On the basis of our results, we conclude that a nuclear-cytoplasmic effect occurs between *cyt-*Y_2 and y_{20}-k_2. Our experiments with y_{20}-k_2 indicate that only with cytoplasmic mutant *cyt-*Y_2 did we have an interaction, i.e., y_{20}-k_2 per se cannot elicit the response. The *cyt-*Y_2 y_{20}-k_2/y_{20}-k_2 genotype is a conditional lethal. This is the first report of such a nuclear-cytoplasmic interaction in soybeans.

In field studies at Ames, Iowa, and Isabela, Puerto Rico, the genotype is lethal, plants carrying it often fail to emerge from the soil. With the moderate light and temperature conditions of the growth chamber, germination is normal. With special care, these genotypes flower and set seed in the greenhouse. This interaction is unique to the genotype *cyt-*Y_2 y_{20}-k_2/y_{20}-k_2. Reciprocal crosses of *cyt-*Y_2 with saddle-pattern mutants k_1, k_2, k_3, and i^k failed to elicit an interaction. Reciprocal crosses of *cyt-*G_2 with y_{20}-k_2 allowed expression of both y_{20} and k_2. Thus the interaction is specific to *cyt-*Y_2 y_{20}-k_2/y_{20}-k_2. With the *cyt-*Y_2 and y_{20}-k_2 mutants, the genotype *cyt-*Y_2 y_{20}-k_2/y_{20}-k_2 is impaired in coordination between cytoplasmic and nuclear components in a manner that is lethal in the field and sometimes correctable under growth-chamber and greenhouse conditions.

Epigenetic changes induced by nuclear-gene mutants may occur, causing heritable defective phenotypes. For example, abnormal plastid differentiation induced by iojap (*ij*) in maize seems to be such a phenomenon (Walbot and Coe 1979; Thompson et al. 1983). Another example is chloroplast mutator in maize (Stroup 1970). Chloroplast mutator-affected plants have a variety of phenotypes ranging from those containing very short stripes of affected tissue to those that are half affected. These two examples represent nuclear genes that not only affect plastid development, but also result in a heritable change in the potential of affected plastids to differentiate in subsequent generations.

Nuclear-cytoplasmic interactions form the basis for the use of cytoplasmic male sterility-restorer systems for the production of hybrid seed (Duvick 1965; Beckett 1971). The

nature of the cytoplasmic defects have been attributed to alterations in chloroplast or mitochondrial DNA (Frankel et al. 1979; Levings 1983). Interactions affecting plant color are also known. Stubbe (1964) and Schotz (1970) reported a broad range of phenotypes from yellow to almost green in certain *Oenothera* crosses.

Most reports detail the effect of nuclear gene mutations on plastid differentiation and subsequent effects on function. These examples are well documented in maize for aberrant differentiation of plastids (Robertson et al. 1978), lack of specific nuclear-coded gene products (Leto and Miles 1980), and impaired synthesis and function of the light-harvesting chlorophyll a/b protein (Schwarz and Kloppstech 1982). In soybeans the k_2 component of y_{20}-k_2 is independent of the effects of the loci *O, R, T,* and W_1 that control saddle color of mutants k_1, k_3, and i^k (Bernard and Weiss 1973). The chemistry for the tan-saddle-color mutant k_2 is not known. The y_{20} component of y_{20}-k_2 has not been studied chemically or ultrastructurally. The *cyt*-Y_2 mutant has normal-appearing mitochondria and chloroplasts with grana stacks, lamellae, and starch accumulation when grown under moderate light and temperature conditions of the growth chamber (Palmer and Mascia 1980). Dark-grown *cyt*-G_2 plants and *cyt*-Y_2 plants develop typical etioplasts. The amounts of protochlorophyllide accumulated by *cyt*-G_2 and *cyt*-Y_2 plants grown in the dark were similar. Under growth-chamber conditions, leaves from *cyt*-Y_2 plants accumulated about 77% of the normal level of chlorophyll, compared with about 38% of the normal in plants grown in the field (Palmer and Mascia 1980).

Additional information is needed on the ultrastructure and chemistry both of *cyt*-Y_2 and y_{20}-k_2 separately as well as of the *cyt*-Y_2 y_{20}-k_2/y_{20}-k_2 genotype. This conditional lethal could be used to study the non-additive effects that result in an interaction between nuclear and plastid genomes.

References

Beckett JB (1971) Classification of male-sterile cytoplasms in maize (*Zea mays* L.). Crop Sci 11:724–727

Bernard RL, Weiss MG (1973) Qualitative genetics. In: Caldwell BE (ed) Soybeans: improvement, production, and uses. Am Soc Agron, Madison Wis, pp 117–154

Duvick DN (1965) Cytoplasmic pollen sterility in corn. Adv Genet 13:1–56

Frankel R, Scowcroft WR, Whitfeld PR (1979) Chloroplast DNA variation in isonuclear male-sterile lines of *Nicotiana*. Mol Gen Genet 169:129–135

Leaver CJ, Gray MW (1982) Mitochondrial genome organization and expression in higher plants. Annu Rev Plant Physiol 33:373–402

Leto KJ, Miles CD (1980) Characterization of three photosystem II mutants of *Zea mays* L. lacking a 32,000 Dalton lamellar polypeptide. Plant Physiol 66:18–24

Levings CS III (1983) The plant mitochondrial genome and its mutants. Cell 32:659–661

Mather K (1951) The measurement of linkage in heredity. Methuen & Co, London

Palmer RG (1984) Pleiotropy or close linkage of two mutants in soybean. J Hered 75:445–447

Palmer RG, Heer HE (1984) Agronomic characteristics and genetics of a chromosome interchange in soybean. Euphytica 33:651–663

Palmer RG, Mascia PN (1980) Genetics and ultrastructure of a cytoplasmically inherited yellow mutant in soybeans. Genetics 95:985–1000

Parthier B (1982) The cooperation of nuclear and plastid genomes in plastid biogenesis and differentiation. Biochem Physiol Pflanz 177:283–317

Robertson DS, Anderson IC, Bachmann MD (1978) Pigment-deficient mutants: genetic, biochemical and developmental studies. In: Walden DB (ed) Maize breeding and genetics. Wiley & Sons, New York, pp 461–494

Rode MW, Bernard RL (1975) Inheritance of a tan-saddle mutant. Soybean Genet Newslett 2:39–42

Schotz F (1970) Effects of disharmony between genome and plastome on the differentiation of the thylakoid system in *Oenothera*. In: Miller PL (ed) Control of organelle development. Soc Exp Biol Symp 24:39–54

Schwarz HP, Kloppstech K (1982) Effects of nuclear gene mutations on the structure and function of plastids in pea. Planta 155:116–123

Shoemaker RC, Cody AM, Palmer RG (1985) Characterization of a cytoplasmically inherited yellow foliar mutant (*cyt*-Y_3) in soybean. Theor Appl Genet 69:279–284

Snedecor GW, Cochran WG (1980) Statistical methods. Iowa State University Press, Ames, Iowa

Stroup D (1970) Genic induction and maternal transmission of variegation in *Zea mays*. J Hered 61:139–141

Stubbe W (1964) The role of plastome in evolution of genus *Oenothera*. Genetica 35:28–33

Terao H (1918) Maternal inheritance in the soybean. Am Nat 52:51–56

Thompson D, Walbot V, Coe Jr EH (1983) Plastid development in *iojap*- and *chloroplast mutator*-affected maize plants. Am J Bot 70:940–950

Vedel F, Remy R (1983) Interaction des genomes nucleaire et chloroplastique. Bull Soc Bot Fr 130, Actual Bot 1:43–50

Walbot V, Coe Jr EH (1979) The nuclear gene *iojap* conditions a programmed change to ribosome-less plastids in *Zea mays*. Proc Natl Acad Sci USA 76:2760–2764

Walker AK, Cianzio SR, Bravo JA, Fehr WR (1979) Comparison of emasculation and nonemasculation for hybridization of soybeans. Crop Sci 19:285–286

Wettstein (von) D (1981) Chloroplast and nucleus: Concerted interplay between genomes of different cell organelles. In: Schweiger HG (ed) International cell biology. Springer, Berlin Heidelberg New York, pp 250–272