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## 1. The effects of anthocyanin (cyanidin di-glucoside) on root growth.

Snythetic anthocyanidins, especially 3,7,4 trihydroxy 2-phenyl benzopyrilium chloride and 7,4 dihydroxy 2-phenyl benzopyrilium chloride, have been reported to stimulate the growth of wheat roots (Stenlid, Physiol. Plant. 15: 1962). To determine whether or not anthocyanin is also a growth stimulator, cyanidin di-glucoside was extracted from 'cy' husks of maize, and tested for biological activity on seedling roots of wheat, oats, and maize; all three types of roots showed a statistically significant increase in length over the controls. The maize seedlings used for assay were homozygous for the a gene. These data are being extended to include relative activities of pure preparations of cyanin, chrysanthemin and the aglycone, cyanidin. Responses to exogenous anthocyanin are also being determined for maize seedlings homozygous for a and seedlings homozygous for A.

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## 2. Leucoanthocyanidin accumulation by maize.

Inter-tissue complementation studies with fresh aleurone tissue have suggested the following gene action sequence for the control of steps along a pathway leading to anthocyanin synthesis (Reddy and Coe, Science 1962):

(C<sup>I</sup>)-C - (C<sub>2</sub>) - R - (In) - A - A - Bz - Bz - Anthocyanin.

In this sequence the homozygous a mutant accumulates a leucoanthocyanidin which can be converted to anthocyanidin by heating with acidified-alcohol (Coe, Genetics 1955). If the gene order is true, mutants which block steps prior to the a gene should lack leucoanthocyanidin when in combination with a. Those which block steps after the a gene should have leucoanthocyanidin when in combination with a. Homozygous double mutant stocks in several combinations were kindly supplied by Dr. Coe. Extracts of the aleurone were tested for the presence or absence of leucoanthocyanidin. Pericarps were peeled from 10-30 mature seed of each genotype after soaking in water for one hour. The exposed endosperms were then extracted with acidified-alcohol for 24-48 hours at room temperature. Each extract was heated 2-5 minutes to detect visible evidence of conversion to pigment. All tests turned out as predicted (Table 1). Recessive intensifier (in) increased the

amount of leucoanthocyanidin about five times. The purple pigments obtained by heating extracts from aleurone homozygous for in  $\underline{a}_2$ ,  $\underline{a}_2$ , and  $\underline{a}_2$ , had absorption spectra identical to cyanidin. Pigmentation could be visibly observed from the heated extract of a

Pigmentation could be visibly observed from the heated extract of a single  $\underline{a}_2$  mutant.

Table 1. Presence (+) or absence (-) of leucoanthocyanidin based on visible appearance of pigment, two to five minutes after heating an acidic-alcoholic extract of aleurone.

Mutant combination	CI a2	c <sub>1</sub> a <sub>2</sub>	e a 2 2	ra 2	in a 2	a a 1 2	a 2	a <sub>2</sub> bz	a <sub>2</sub> bz <sub>2</sub>	A <sub>2</sub> bz <sub>1</sub>
Color	-		1	1	+	-	+	+	+	-

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## 3. Double mutants for dwarfing genes of Zea mays.

Four of the five gibberellin-responding dwarf mutants of maize have been intercrossed in all possible combinations and the F1 selfed to give F2's which segregate for the double mutant. These presumptive double mutants can be identified in the early seedling stage in some cases, or in other cases as the plants become older. The d - and double mutant has been backcrossed to both d and an plants; all progeny were dwarf, confirming the presumptive genotype of the double mutant. All presumptive double mutants (seven different combinations tested) respond to exogenous gibberellin by increased growth to give a phenotype which approached that of the normals.

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