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1. Potassium content of opaque-2, floury-2, double mutant and normal versions of certain inbreds and hybrids.

Potassium was first reported to be higher in opaque-2 kernels by Goodsell (Crop Sci. 8:281-282). He used F_2 kernels from segregating ears to make his comparisons and found that opaque-2 kernels averaged 41% higher in potassium than normal siblings on the same ear. The data in Table 1 are from homozygous ears of inbred lines converted to opaque-2. Here again, o_2/o_2 segregates are higher in potassium than normal segregates in all cases. However, the level of potassium is a function of the inbred line and can vary quite extensively. Since the assay for potassium is simpler, cheaper, and more precise than that for lysine, it was proposed at one time that potassium level be used as a rough screening technique for lysine level. But this data would tend to suggest that in a heterogeneous opaque-2 population, the level of potassium would be independent of lysine level. The limited data for floury-2 and the double mutant are shown, but little should be made of them because they are in the initial stages of conversion to the inbred line.

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1. Chlorophyll-deficient mutants have differential capacities to accumulate assimilation starch.

Five chlorophyll-deficient mutants ($\underline{1}_3$, $\underline{1}_4$, $\underline{1}_7$, $\underline{1}_{4920}$, \underline{w}_{11}) out of 17 stocks tested (former plus \underline{w}_1 , \underline{w}_2 , \underline{w}_3 , \underline{yw}_{8896} , \underline{wh}_{8657} , \underline{cl}_1 , $\underline{1}_1\underline{w}_2$, $\underline{1}_6$, $\underline{1}_{4923}$, $\underline{1w}_1$, $\underline{1w}_2$, $\underline{1w}_3\underline{1w}_4$) accumulated significantly less assimilation starch from exogenous glucose than their normal sibs. Initial screening

Table 1
Lysine and potassium content of mature corn kernels

	Normal		$\frac{o_2}{o_2}$		$\frac{fl_2}{fl_2}$		$\frac{o_2}{o_2}$ $\frac{fl_2}{fl_2}$	
	lys	K	lys	K	lys	K	lys	K
R803	.35	.40	.56	.63				
R801	.36	.30	.51	.48				
R109	.35	.36	.60	.46				
WF9	.36	.36	.50	.47				
R75	.39	.45	.61	.58		.50		.50
o_7^N	.34	.32	.49	.44				
R802	.37	.34	.60	.46				
oh45	.31	.28	.58	.53	.42	.39	.55	.43
R801 x R75	.41	.40	.54	.52				
R802 x R803	.30	.41	.51	.60				

$r = .17$ for lysine and K within the opaques.

(See top of previous page for explanation of table.)

was accomplished by floating 1 cm leaf segments from etiolated seedlings on 0.5 M glucose for 24 hr in darkness at 27°C, killing in boiling 95% ethanol, and staining with an aqueous solution of 0.01 M I₂ + 0.03 M KI. Segments containing starch stained brown to blue-black whereas segments without starch and control segments incubated on distilled water stained light brown. The mutants varied in staining intensity from dark as normals to light as water treated controls.

The starch content of glucose incubated segments of the "starch-less" mutants, their normal sibs (homozygotes + heterozygotes), and a dent hybrid, WF9 x M14, was determined by the method of Hassid and Neufeld (Methods in Carbohydrate Chemistry, Vol. IV) as modified in our laboratory for small samples of fresh material. The starch content of the mutants varied over a five-fold range and their ranking on the basis of starch content was the inverse of their normal sibs. The normal sibs of l₄920 and w₁₁ contained significantly less starch than the normal sib of l₄, indicating the possibility of genetic variability for the capacity of "normal" leaves to accumulate starch from exogenous glucose. The starch data, given as ug/gfw ± std. error, are summarized in the table.

Table 1
Starch content (ug/gfw ± std error) of incubated leaf segments from five chlorophyll mutants, their normal sibs, and a dent hybrid.

Stock	<u>Water Treated</u>		<u>Glucose Treated</u>	
	Normal	Mutant	Normal	Mutant
WF9 x M14	72.1 ± 27.9	-	4,751 ± 384	-
<u>l</u> ₄	20.8 ± 2.1	87.4	3,506 ± 175	116 ± 6
<u>l</u> ₇	28.8 ± 4.8	-	2,732 ± 37	122 ± 6
<u>l</u> ₃	26.7 ± 5.4	35.6 ± 2.4	2,210 ± 60	406 ± 7
<u>w</u> ₁₁	32.9 ± 8.6	3.2 ± 0.4	1,713 ± 413	599 ± 82
<u>l</u> ₄ 920	34.4 ± 13.8	-	1,591 ± 90	464 ± 51

Cross sections of normal green leaves and treated segments were stained with IKI and examined under the microscope to determine the pattern of distribution of starch in the tissues. No starch was detectable in etiolated and water incubated tissues. Starch was present in normal green leaves in large amounts only in bundle sheath cells. Glucose incubation of etiolated segments resulted in accumulation of massive quantities of starch in both bundle sheath and mesophyll cells. The IKI stainable material which accumulated during glucose treatment was removed by incubating cross sections in α -amylase (11.1 units/ml) for two hr.

The nature and location of the mutations causing the observed effects are unknown. It is possible, but improbable, that the "starchless" phenotype is due to a separate mutation that is closely linked to the chlorophyll-deficiency factor in all five stocks. A more likely explanation invokes a single mutation that affects the synthesis or utilization of some essential amino acid, co-factor, or protein(s). Further studies to determine the activities of the enzymes of the starch biosynthetic pathway (ADP-glucose pyrophosphorylase, ADP-glucose glucosyltransferase, and starch phosphorylase) and of other chloroplast-specific enzymes are being initiated. These studies should provide much-needed information about the general scheme of formation of assimilation starch and some insight into the nature of these specific mutations.

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