

If the total population of kernels (16,798) is adjusted for the proportion of dark stippled kernels scored for seedling color, 3211/3836, and for the proportion of red seedlings verified, 7/16, the frequency of female gametes carrying both chromosomes 10 was seven in 6,152 gametes tested, a rate of 11.4×10^{-4} .

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2. The regulatory nature of the waxy locus.

Evidence accumulated over the past year makes it probable that the waxy locus is regulatory in nature and not the structural locus for the nucleotide transferase. The most important evidence is the finding that starch granule preparations from the embryos of developing waxy seeds (16 day) have as high or higher transferase activity than similar preparations from non-waxy seeds. The low level of activity shown by starch granule preparations from whole seeds of waxy stocks when ADPG is used as a substrate is not due entirely or even largely to the presence of starch granules from the embryo. Starch granule preparations from waxy endosperms alone still have low activity. Comparative activities are given in Table 1. At 16 days, the embryos were 1.5 and 1.1 percent of the wet weight of the waxy and non-waxy seeds, respectively.

Table 1
 μM ADP Released Per Mg. of Starch Under Standard Assay Conditions.*
 All Preparations from Developing Seeds Frozen 16 Days After Pollination.
 1962 Collections.

	\pm	WX
Starch granules from embryos alone	114	142
Starch granules from endosperms alone	30	3.3
Starch granules from whole seeds	35	3.7

*To 2.5 mg. of starch granule preparations (1 mg. if embryo is source) is added 25 microliters of a solution that contains $0.31 \mu\text{M}$ ADPG, $0.17 \mu\text{M}$ EDTA, $6.85 \mu\text{M}$ glycine, and is buffered at pH 8.4. After 15 minutes at 37°C , 25 microliters of 0.01M phosphoenolpyruvate solution and 25 microliters of a pyruvate kinase solution containing about 26 enzyme units per ml. are added, and reaction allowed to proceed for 15 minutes more before being stopped by the addition of dinitrophenyl hydrazine. Thus total reaction time is 30 minutes at 37°C .

It is clear from Table 1 that the low nucleotide transferase activity of the waxy endosperms is not characteristic of waxy embryos. Yet the evidence at hand points clearly to the dependence of endosperm transferase activity on the allelic state at the waxy locus. Thus it is improbable that the waxy locus is the structural locus for transferase and likely that it is regulatory in nature.

Corroborative evidence for this conclusion comes from a study of the transferase activity of 17 waxy mutants that occurred as separate mutational events. The results are given in Table 2. All the mutants have

Table 2

μM ADP Released Per Mg. of Starch Under Standard Assay Conditions.
All Preparations from Developing Seeds Frozen 16 Days After Pollination.
1963 Collections. Whole Seed Preparations.

Mutant	Origin	μM ADP
C	Spontaneous	5.4
90	Spontaneous	3.8
Bear G	Spontaneous	6.0
Brink 1	Spontaneous	3.6
Brink 2	Spontaneous	4.2
Brink 4	Spontaneous	4.8
Brink 6	Spontaneous	3.8
Brink 8	Spontaneous	3.2
H21	Spontaneous	4.4
T4B	Presumptive Irradiation*	6.6
Q1R	Presumptive Irradiation*	3.4
S3G	Presumptive Irradiation*	6.6
N1R	Presumptive Irradiation*	4.8
N3Y	Presumptive Irradiation*	7.4
wx^{m-1}	<u>Ds</u>	5.0
wx^{m-6}	<u>Ds</u>	6.4
wx^{m-8}	<u>Spm System</u>	4.8
M14 (Non-waxy)		62

*From Dr. Caspar, Blandy Experimental Farm.

measurable activity and in a rather restricted range. To find that 17 different mutants (spontaneous, irradiation, and controlling element), were all hypomorphic and to about the same degree would be improbable if the waxy locus were the structural locus for transferase unless a special set of assumptions was invoked.

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3. Differential crossing over in male and female gametes of plants heterozygous for Dp 9.

Rhoades (MNL, 32) first reported reduced recombination between markers on the short arm of chromosome 9 in plants heterozygous for Dp 9. In attempting in 1961 to evaluate the effect of heterozygosity for Dp 9 on recombination within the wx locus, we observed a pronounced difference between male and female gametes in crossing over between markers on the short arm of 9.

The genotype of the heterozygous plants (15562) was C Sh Dp + wx⁹⁰ Gl₁₅/c sh N wx^c + gl₁₅. Randomly selected plants were crossed as male parents onto a c sh N wx^c + gl₁₅ tester and as females by the same tester stock. Table 1 presents the data for the c sh interval and Table 2 for the sh gl₁₅ interval.