## OAK RIDGE NATIONAL LABORATORY<sup>1</sup> Oak Ridge, Tennessee Biology Division

## 1. Genetic studies on developing endosperm proteins.

Over the past two years we have been studying protein differences in developing endosperm tissue of mutants which affect starch synthesis and composition. We have used the technique of "substrate electrophoresis" separating the endosperm proteins on starch gel which is substrate for the enzymes involved in starch synthesis and degradation. In this technique, the separation of the proteins depends upon enzyme substrate affinity as well as charge. It is possible to directly test for the enzymes which alter the structure of starch, by staining the gel with iodine following the electrophoretic separation. Those enzymes which reduce the size of the starch polymer by digestion or branching appear as red bands against the blue background of the stained starch gel.

We have found that the  $\underline{Sh}_1$  gene controls the synthesis of a major protein component which is present in the highest concentration in the immature endosperm. This protein is completely lacking in  $\underline{sh}_1/\underline{sh}_1/\underline{sh}_1$  endosperm. The concentration of this protein (designated  $Sh_1$ ) is dose dependent increasing with increasing doses of the  $\underline{Sh}_1$  allele. This has been corroborated by immunochemical studies as well as moving boundary electrophoresis. Suppression of  $\underline{Sh}_1$  by  $\underline{Ds}$  also results in the complete absence of the  $\underline{Sh}_1$  protein, however, in the presence of  $\underline{Ac}$  the  $\underline{Sh}_1$  protein reappears but in reduced amount as is expected. We have not been able to detect the presence of a new protein band in  $\underline{sh}_1/\underline{sh}_1$  material; however, the intensity of the band which occupies a position just ahead of the  $\underline{Sh}_1$  protein is increased in this mutant. This band (designated  $\underline{G}$ ) shows a faster migration rate in the  $\underline{Sh}_1$  protein is increased in this mutant. This band (designated  $\underline{G}$ ) shows a faster migration rate in the  $\underline{Sh}_1$  mutant material and appears to be missing in  $\underline{su}_1$  and its allele  $\underline{su}_1$  am,  $\underline{bt}_1$  and its allele  $\underline{sh}_3$ ,  $\underline{sh}_2$ , and  $\underline{du}_1$ . That different genes are responsible for the absence of this protein band in these mutants is indicated by the presence of the band in the  $\underline{F}_1$  hybrid between  $\underline{su}_1$  and  $\underline{sh}_2$ , each of which lacks the band. Analysis of double mutant combinations points to gene interaction in the control of the synthesis of this protein.

<sup>1</sup> Operated by Union Carbide Corporation for the U. S. Atomic Energy Commission.

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## Investigations on induced polygenic mutability in maize.

An experiment was started in 1957 with the goal of studying polygene mutability in maize. A monoploid stock, HD 73, 1375/11 kindly supplied by Professor G. F. Sprague was used. 8 lines were established in 1957 by selfing as many plants obtained from seed of a single selfed ear. In 1958 three groups of about 5 plants were selfed within each line, one to be treated with 3,000 r applied to the tassel 2 days before using the pollen for self-fertilization, one to be treated with 1,500 r and one receiving no treatment. Treatments were given using an X-ray machine operated at 230 KV, 12 mA, ceiving no treatment. In Winter 1958-1959, the R<sub>1</sub> generation was obtained growing the plants in Somali-

land, with the cooperation of the Afgoi Agricultural Center, Mogadiscio. From each of the treated and untreated selfed ears obtained in the previous generation, from 5 to 10 plants were selfed establishing sublines.  $R_1$  and  $R_2$  seeds were grown in the same plot in Summer 1959 at the Agricultural Experiment Station in S. Angelo Lodigiano, in the Padana Plain, near Pavia, and observations were made for the following traits:

- 1) flowering time of the tassel determined as the number of days beginning with July 1st; on  ${\rm R}_1^{}$  and  ${\rm R}_2^{}$ 
  - 2) number of simple lateral branches of the tassel; on  ${\sf R}_2$
  - 3) number of composite lateral branches of the tassel; on R2
  - 4) total number of lateral branches of the tassel, simple and composite; on Ro
  - 5) number of spikelets per mm. of the rachis length in the central axis; on R<sub>2</sub>
  - 6) number of spikelets per mm. of the rachis length in the upper lateral branch; on R2.

It is expected that, as all the plants originated from a single selfed ear of a monoploid stock, any significant increase of variability observed for the treated lines is due to the effect of mutations induced by X-ray treatment, and any significant increase of variability observed for the untreated line in different generations is due to the effect of spontaneous mutations.

As the material used is represented by plants of lines and sublines differentiated by selfing, an increase of variability can be detected:

- a) by comparing the distribution range of variability of lines for the various treatments:
- b) by the analysis of variance within the three treatment groups in order to distinguish the contribution to variability of lines (genetic source), of sublines within lines (genetic source), and within lines (environmental component).

In this preliminary report data for comparison (a) are summarized in the following table, which gives: the mean for lines, their observed range, the estimated variances between lines within treatments and  $X^2$  estimates in the Bartlett test for heterogeneity between treatments.

The P values given in the table show that variances for line means within treatments are significantly heterogeneous for flowering time in  $R_1$  and  $R_2$ , for the number of composite lateral branches of the tassel and probably for the number of spikelets per mm. in the upper lateral branch of the tassel.

-- R. E. Scossiroli

Table 1.

		Table 1.			1
	Means( $\overline{x}$ ), range of line means and estimated variances (s <sup>2</sup> )				X <sup>2</sup> of the Bartlett test, 2 d.f.
i		control	1,500 r	3,000 r	
1. flowering time, days	range	20.99 20.65-21.57 0.1022	21.24 20.16-23.36 0.9824	22.13 20.57-23.63 0.8877	9.28 P = 0.01
R <sub>2</sub>	x range s <sup>2</sup>	20.68 19.62-21.49 0.4009	21.16 19.89-22.38 0.5918	21.47 20.75-22.63 0.4194	0.37 P = 0.98-0.99
2. no. simple lateral branches of the tassel, R <sub>2</sub>	x range s <sup>2</sup>	9.68 8.88-10.44 0.3683	9,59 8,54-10,48 0,4033	10.29 8.92-12.00 0.8644	1.87 P = 0.30-0.50
3. no composite lateral branches of the tassel, R <sub>2</sub>	x range s <sup>2</sup>	2.13 2.08-2.20 0.0014	2.10 1.92-2.31 0.0151	2.24 2.09-2.45 0.0195	11.46 P = 0.01
4. total no. lateral branches of the tassel,	x range s <sup>2</sup>	11.82 10.96-12.63 0.3862	11.70 10.65-12.72 0.4598	12.53 • 11.23-14.25 0.9057	1.71 P = 0.30-0.5
5. no spikelet/mm, central axis of the tassel, $R_2$	x range s <sup>2</sup>	1.10 1.07-1.15 0.0006	1.11 1.07-1.16 0.0010	1, 23 1, 09-1, 15 0,0005	1.07 P = 0.50-0.7
6. no. spikelet/mm. upper lateral branch of the tassel, R <sub>2</sub>	x range s <sup>2</sup>	0.26 0.25-0.26 0.00002	0.25 0.24-0.26 0.00012	0.25 0.23-0.27 0.00022	$\frac{3.65}{P = 0.10-0}$