

3. Further evidence about the smoky modifier.

In the 1966 News Letter it was reported that when $\underline{R}^{\text{sk}} \underline{r}^{\text{g}}$ plants are crossed with $\underline{r}^{\text{g}} \underline{r}^{\text{g}}$ some of the resulting ears show, besides the expected colorless kernels (genotypically $\underline{r}^{\text{g}} \underline{r}^{\text{g}}$), two kinds of smoky, darker and lighter, often in equal frequency. Such results could be explained by assuming that the lighter smoky phenotype results from the interaction of $\underline{R}^{\text{sk}}$ with a Modifier of the smoky expression that assort independently of $\underline{R}^{\text{sk}}$.

The validity of this assumption can be tested by crossing plants derived from colorless kernels (obtained from the previously mentioned cross) with their dark smoky sibs. In fact, if the smoky Modifier assort independently of $\underline{R}^{\text{sk}}$, approximately one-half of the colorless kernels should carry it. Its presence can be proved by the appearance of two phenotypic classes of smoky, lighter and darker, in the ears obtained from the above mentioned cross. Twenty-four ears so obtained have been scored. Twelve of them segregate only dark smoky and colorless kernels, eight exhibit a clear segregation of dark and light smoky kernels, besides the expected 50% colorless, while the remaining four ears have been discarded because of scoring difficulties. These results clearly indicate that the two classes of smoky are due to the segregation of a smoky Modifier.

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4. Differential response of the R subunits to the paramutagenic action of \underline{Rst} .

Plant and seed pigments are controlled by the two subunits of the \underline{R} locus, respectively symbolized \underline{P} and \underline{S} . If paramutation is not confined to the \underline{S} component but affects the \underline{R} locus as a whole, its expression should be observable also in the sporophytic tissues. In a previous test (M.N.L. 1966) we tried to establish this point by comparing the concentration of pigment extracts of $\underline{r}^{\text{g}} \underline{R}^{\text{r}}$ and $\underline{r}^{\text{g}} \underline{R}^{\text{r}'}$ roots grown on filter paper. The spectrometric determination of root pigments failed to disclose a significant reduction in pigmentation level of paramutant $\underline{r}^{\text{g}} \underline{R}^{\text{r}'}$ roots. These data seemed to suggest that the \underline{R} component conditioning pigment formation in the roots is not significantly affected by its association with a paramutagenic allele. Alternatively, they could simply indicate that no suitable growing conditions for pigment formation were used in our test. In fact, it is likely that the establishment of growth conditions leading to an increased anthocyanin biosynthesis in the sporophytic tissues may make observable even small differences in pigment concentration between $\underline{r}^{\text{g}} \underline{R}^{\text{r}}$ and $\underline{r}^{\text{g}} \underline{R}^{\text{r}'}$ roots.

Such conditions are obtained by allowing seeds to germinate on a medium containing agar and sucrose (0.25%). Furthermore it is possible, using this medium, to extend the measurements of pigment concentration to other tissues like the coleoptile and mesocotyl. This medium has been used to obtain the data that are here presented.