

| Cross             | Number of progeny tested | Number of translocations confirmed cytologically | Frequency of translocations |
|-------------------|--------------------------|--|-----------------------------|
| 2N X 2N           | 6980                     | 1  | 0.01%                       |
| Monosomic 2 X 2N  | 241                      | 1  | 0.41%                       |
| 2N X Monosomic 7  | 4083                     | 2*   | 0.05%                       |
| 2N X Monosomic 8  | 2787                     | 6  | 0.22%                       |
| 2N X Monosomic 10 | 3268                     | 6  | 0.18%                       |

\*One additional line which carries female and male semisterility from a 2N X monosomic 7 cross has not yet been analyzed cytologically; thus, it is likely that another member is in this class.

It is significant that a much higher frequency of reciprocal translocations is found in progeny of all monosomic X diploid crosses tested than in progeny of diploid X diploid crosses (control population). This strongly suggests that the unpaired monosomic chromosome can occasionally pair with homologous segments found in other regions of the genome, and recombination can occur between the paired regions.

18,992 additional progeny from monosomic by diploid crosses (monosomics 6, 8, and 10) as well as crosses involving a diploid control were screened in the summer of 1971, and 322 ears expressing some degree of semisterility were recovered. 12,203 progeny of monosomic X diploid crosses were screened this past summer.

David F. Weber

## 2. Fatty acid profiles from maize scutella: a new genetic tool.\*

Introduction: Most research involving the genetics of fatty acids in Zea mays has been conducted by extracting the lipids from either whole kernels or entire embryos. Since the kernels are destroyed by these methods, siblings must be used in subsequent crosses and their fatty acid profiles can only be inferred. This article describes a technique to

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analyze the fatty acid profiles from a single kernel without greatly reducing the viability of that kernel. We are currently employing this technique in our genetic studies on maize lipids (Plewa and Weber, 1972, Maize Genet. Coop. News Letter 46:46-48).

Materials and Methods: All kernels analyzed came from a single sib of inbred W22 carrying  $R^F$ . Separate fatty acid profiles from five whole embryos (group A), five scutella samples from the right side (group B), and five scutella samples from the left side of embryonic axes (group C), were compared.

Lipid extraction for group A was conducted by removing the pericarp covering the embryonic tissue and hand dissecting the entire embryo. The isolated embryos were placed into separate, numbered testtubes.

Lipid extraction for groups B and C were conducted as follows. The pericarp covering the scutellum area to be sampled was removed. Approximately a 1.5 mg scutellum sample, distal and parallel to the embryonic axis, was hand dissected, removed, placed in a vial, coded, and stored at  $-22^{\circ}\text{C}$ . Sampled kernels were saved for planting.

Each sample was macerated with a glass rod in 5 ml of lipid extraction solvent (redistilled petroleum ether and absolute methanol, 2:1 v/v) and the lipids were extracted overnight. Ten ml of methylating reagent (1%  $\text{H}_2\text{SO}_4$  in absolute methanol) were added to each sample and kept at room temperature for 1 hr. The volume was reduced to 5 ml by passing a stream of dry nitrogen ( $\text{N}_2$ ) over the liquid. Each sample was refluxed for 1 hr. at  $63^{\circ}\text{C}$  under an atmosphere of  $\text{N}_2$ . After cooling to room temperature, 5 ml of redistilled petroleum ether was added and the sample was poured into a separatory funnel. Each sample tube was quantitatively rinsed with 5 ml of redistilled petroleum ether and the rinse was added to the separatory funnel. The petroleum ether fraction with the dissolved fatty acid methyl esters was separated from the methanol fraction. The volume of the petroleum ether fraction was reduced under  $\text{N}_2$  and the sample transferred to a vial. Each sample was cleaned and all traces of the reagent were removed. The resulting sample contained the methyl esters of the maize fatty acids. The volume was reduced to approximately 0.5 ml; the vial was wiffed with  $\text{N}_2$ , sealed, and stored at  $-22^{\circ}\text{C}$ .

The methylated fatty acids were analyzed with a Hewlett-Packard model 5750 gas-liquid chromatograph. The column was 6 ft x  $\frac{1}{8}$  in OD stainless steel packed with 10% EGSS-X 100/120 gas chromatography-Q (Applied Science Laboratories). The column temperature was 180°C, injection port temperature was 250°C, and the flame detector temperature was 235°C. The carrier gas was N<sub>2</sub> and the flow rates for the gases were: N<sub>2</sub>, 20 ml/min; H<sub>2</sub>, 42 ml/min.; and compressed air 470 ml/min.

The relative amounts of the fatty acids were determined by using a Dietzgen Compensating Polar Planimeter. Two GLC runs were conducted per sample and the relative fatty acid concentrations presented in Table 1 are averages of the runs.

The mean of each individual fatty acid from each group (A, B, and C) was analyzed for departures from the null hypothesis ( $H_0 = u_A = u_B = u_C$ ) by Analysis of Variance,  $\alpha = 0.05$ . Four statistical tests were conducted, one for each fatty acid (palmitic acid, 16:0; stearic acid, 18:0; oleic acid, 18:1; and linoleic acid 18:2).

Results and Discussion: The relative individual fatty acid concentrations for each kernel of the three groups are presented in Table 1. The F values computed from the means of each separate fatty acid in the three groups did not indicate a departure from the null hypothesis (Table 1). There is no significant difference in the individual fatty acid profiles among the three groups. Therefore, a fatty acid profile from a scutellum sample is representative of the fatty acid profile of the entire embryo. Thus, data from whole embryo studies can be compared with the data from scutella samples.

Although there are differences in the relative concentrations of fatty acids in the embryonic axis as compared to the whole embryo or scutellum (E. Weber, personal communication; Plewa, unpublished), these differences are diluted by the larger mass of the scutellum.

This technique has obvious advantages for studying the genetics of fatty acid biosynthesis in maize. The sampled kernels may be planted and crosses can be performed. During the summer of 1972, over 1,500 kernels were sampled and field planted. Approximately 8% of the kernels germinated and it was possible to use them for crosses, and have fatty

Table 1  
Relative amounts of fatty acids in groups A, B, and C.

| Sample No.        | Fatty acids |         |        |          | Groups                             |
|-------------------|-------------|---------|--------|----------|------------------------------------|
|                   | Palmitic    | Stearic | Oleic  | Linoleic |                                    |
| MP234-21          | 0.1448      | 0.0259  | 0.2986 | 0.5290   | A<br>(Whole embryos)               |
| MP234-22          | 0.1692      | 0.0306  | 0.2898 | 0.5108   |                                    |
| MP234-23          | 0.2114      | 0.0298  | 0.2763 | 0.4826   |                                    |
| MP234-24          | 0.1843      | 0.0301  | 0.2584 | 0.5273   |                                    |
| MP234-25          | 0.1632      | 0.0421  | 0.2391 | 0.5558   |                                    |
| Means             | 0.1746      | 0.0317  | 0.2724 | 0.5211   |                                    |
| MP234-1           | 0.1971      | 0.0464  | 0.3333 | 0.4232   | B<br>(Scutella samples right side) |
| MP234-2           | 0.1402      | 0.0449  | 0.2876 | 0.5274   |                                    |
| MP234-3           | 0.2010      | 0.0300  | 0.2637 | 0.5054   |                                    |
| MP234-4           | 0.1391      | 0.0487  | 0.2383 | 0.5739   |                                    |
| MP234-5           | 0.1741      | 0.0278  | 0.2235 | 0.5747   |                                    |
| Means             | 0.1703      | 0.0396  | 0.2693 | 0.5209   |                                    |
| MP234-6           | 0.2037      | 0.0201  | 0.2361 | 0.5417   | C<br>(Scutella samples left side)  |
| MP234-7           | 0.1827      | 0.0543  | 0.2641 | 0.4989   |                                    |
| MP234-8           | 0.1545      | 0.0320  | 0.3045 | 0.5091   |                                    |
| MP234-9           | 0.1458      | 0.0292  | 0.2915 | 0.5335   |                                    |
| MP234-10          | 0.2041      | 0.0204  | 0.3010 | 0.4745   |                                    |
| Means             | 0.1782      | 0.0312  | 0.2794 | 0.5115   |                                    |
| <u>F</u> * values | 0.0714      | 1.0890  | 0.1341 | 0.0913   |                                    |

\* $F_{95}(2,13) = 3.89$ ; retain  $H_0$

acid profiles from embryos of the corresponding sporophytes. Under greenhouse conditions, germination rates of sampled kernels are over 90%. We believe that this technique is a valuable tool and we are currently employing it in our studies in mapping genes involved in fatty acid biosynthesis utilizing maize monosomics.

Michael J. Plewa  
David F. Weber