# FERTILITY RESTORATION RESPONSE OF VARIOUS SUNFLOWER CYTOPLASMS

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## Summary

Male-sterile plants from three sunflower sources, CMG-1, CMG-2, and CMG-3, were selected and standard backcross procedures were utilized to incorporate the genome of HA 89 into the cytoplasm of each source. Male-fertile plants within each CMG germplasm were sibmated two generations to overcome self-incompatibility and then self-pollinated to produce S2 lines. The three cms CMG BC3 lines and cms HA 89 were then crossed to the S2 CMG restorer lines and RHA 274. Male-fertility restoration was observed and found to differ among the hybrid combinations. Cms HA 89 was restored by RHA 274 and RCMG-1. Cms CMG-1 was restored by RCMG-1 and RCMG-2. Cms CMG-2 was restored by only RCMG-2 while cms CMG-3 was restored by all restorers. The pollen fertility restoration pattern showed that each cytoplasm source was different from each other and that different gene actions were observed for pollen fertility restoration.

## Introduction

The discovery of cytoplasmic male-sterility (cms) in sunflower (Helianthus annuus L.) by Leclercq (1969) and the subsequent identification of restorer genes for pollen fertility by Kinman (1970), Enns et al. (1970), Leclercq (1971), and Vranceanu and Stoenescu (1971) has lead to the widespread use of cms in the production of sunflower hybrids. All sunflower hybrids are currently produced from the source of cms discovered by Leclercq (1969). The use of this single source of cms may limit the genetic base of the parental lines used in hybrid production. Hybrids with this cytoplasm may become vulnerable to new strains of disease. An example of this possibility is the southern corn leaf blight (Helminthosporium maydis, Nisikado and Miyade) epidemic on corn with Texas cytoplasm in 1970 (Tatum, 1971).

Recently, potential new sources of cms in sunflower were found, and germplasms with these cytoplasms were released (Whelan, 1980; Whelan and Dedio, 1980). These germplasms CMG-1, CMG-2, and CMG-3, are open-pollinated composites of partial interspecific substitutions of the nucleus of cultivated sunflower (H. annuus L.) cv. 'Saturn' into the cytoplasms of the species H. petiolaris Nutt., H. gigantetus L., and H. maximilliani Schrad., respectively. The purpose of this study was to purify these composite germplasms and produce hybrids using cms and restorer lines from each germplasm and a standard cms and restorer line to determine pollen restoration and differences due to cytoplasm interaction.

## Materials and Methods

Three sunflower germplasm composite crosses CMG-1, CMG-2, and CMG-3 were obtained from Whelan and Dedio (1980) in 1981. Sterile plants were selected and standard backcross procedures were utilized to incorporate the genome of HA 89 into the cytoplasms of each CMG germplasm source to produce cms BC3 lines. The BC3 lines, cms CMG-1/HA 89, cms CMG-2/HA 89, and cms CMG-3/HA 89 are referred to as cms CMG-1, cms CMG-2, and cms CMG-3, respec-

tively, in this paper. Pollen fertile (restorer) plants within each germplasm were sibmated two generations to overcome self-incompatibility, then self-pollinated for two generations producing S2 lines. One cms BC3 line and one S2 restorer line of each CMG germplasm source was selected for hybrid crossing. Each cms BC3 line including cms HA 89 was crossed by each of the CMG restorer lines and RHA 274. Separate randomized complete block designs were utilized to test for pollen restoration, pollen shed, height, size of leaves, and days to anthesis for the hybrids and the cms inbred lines. Pollen shed was rated on a scale 0-5 with 0 = no pollen and 5 = profuse pollen. Leaf width was measured on the widest part of the 12th leaf.

## Results and Discussion

RHA 274 fully restored male-fertility on cms HA 89 and partially restored cms CMG-3, but did not restore cms CMG-1 and cms CMG-2 (Table 1). RCMG-1 partially restored cms HA 89, cms CMG-1, and cms CMG-3, but did not restore cms CMG-2. RCMG-2 partially restored all the cms CMG germplasm lines, but did not restore cms HA 89. RCMG-3 restored male-fertility to only 2 percent of the plants on cms CMG-3 and no restoration was evident on any of the other cms lines. The partial pollen fertility restoration could be explained by the fact that the male fertile plants of the original composite germplasms were sibmated within each germplasm for two generations. This may have lost restorer genes effective for restoring fertility to this cytoplasm. The two generations of selfing were not sufficient to produce homozygous lines for restoration and also, the restorer lines developed were not tested for pollen restoration on their own cytoplasm. Further research needs to be conducted to determine these conclusions.

Cms HA 89/RHA 274 produced profuse pollen (Table 1). Cms CMG-1/RCMG-2 and cms CMG-3/RCMG-3 shed only a few pollen grains which was enough to indicate male fertility restoration was achieved. Hybrids cms HA 89/RCMG-1, cms CMG-2/RCMG-2, cms CMG-3/RHA 274, cms CMG-3/RCMG-1, and cms CMG-3/RCMG-2 produced near adequate to adequate amounts of pollen. All other hybrids did not produce any pollen and were thus classified as being male sterile.

Hybrids with cms HA 89 as the female parent and RCMG-1 as the male parent produced the tallest hybrids with cms HA 89/RCMG-1 as the tallest (Table 1). Hybrids with cms CMG-3 as the female parent and RMCG-3 as the male parent produced shorter hybrids with cms CMG-3/RCMG-3 being the shortest hybrid.

Cms CMG-3 produced hybrid plants with the smallest leaf width and the most days to anthesis (Table 1). All other cytoplasms were similar in leaf width and days to anthesis. The male parents had no influence on either leaf width or days to anthesis.

The cytoplasmic male sterile lines, cms CMG-1, cms CMG-2, and cms CMG-3 all produced stable cms lines (Table 2). Cms CMG-1 and cms CMG-2 were similar to HA 89 in days to anthesis, whereas, cms CMG-3 had later anthesis. Cms CMG-3 had less vigor than cms HA 89. All sources had smaller leaves and were taller than cms HA 89.

**Table 1.** Percent male fertile plants; mean pollen shed, days to anthesis, height, and number of leaves of hybrids for cytoplasmic male sterile sources crossed with restorers developed from each cytoplasmic source.

| Cytoplasmic Source | Percent Male Fertile Plants | | | | Pollen Shed* | | | | Height (cm) | | | | Leaf Width (cm)** | | | | Days to 50% Anthesis | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| cms HA 89 | 100 | 57 | 0 | 0 | 5 | 3.3 | 0 | 0 | 155 | 153 | 153 | 138 | 20 | 22 | 23 | 22 | 69 | 73 | 72 | 70 |
| cms CMG-1 | 0 | 10 | 38 | 0 | 0 | 0 | 1 | 0 | 148 | 157 | 150 | 134 | 21 | 20 | 21 | 23 | 71 | 71 | 71 | 69 |
| cms CMG-2 | 0 | 0 | 42 | 0 | 0 | 0 | 2 | 0 | 144 | 151 | 150 | 130 | 21 | 24 | 22 | 22 | 70 | 71 | 71 | 69 |
| cms CMG-3 | 74 | 88 | 87 | 2 | 2 | 2.4 | 2 | 1 | 142 | 142 | 133 | 132 | 17 | 19 | 15 | 15 | 72 | 73 | 73 | 72 |

* Pollen shed rated on the scale 0-5, with 0 = no pollen, 5 = profuse pollen
** Leaf measured was 12th from cotylenonary node



Table 2. Percent male fertile plants; mean pollen shed, days to anthesis, height, and number of leaves of cytoplasmic male sterile sources.

| Cytoplasmic source | Percent Male Fertile Plants | Pollen Shed* | Days to 50% Anthesis | Height (cm) | Leaf Width** |
|---|---|---|---|---|---|
| cms CMG-1 | 0 | 0 | 72 | 128 | 18 |
| cms CMG-2 | 0 | 0 | 72 | 123 | 20 |
| cms CMG-3 | 0 | 0 | 74 | 115 | 19 |
| cms HA 89 | 0 | 0 | 70 | 105 | 20 |
| HA 89 | 100 | 4.2 | 72 | 103 | 21 |

* Pollen shed rated on the scale 0-5, with 0 = no pollen, 5 = profuse pollen
** Leaf measured was 12th from cotylenonary node

## Conclusion

All three cms germplasms, cms CMG-1, cms CMG-2, and cms CMG-3 are different from the Leclercq cytoplasm and from each other as shown by their pattern of male-fertility restoration when crossed to restorers from each of the CMG cytoplasm sources and RHA 274. RCMG-2 restored all the cytoplasms except cms HA 89 indicating it may have many genes for restoration. Cms CMG-2 was the most specific in restoration as it was restored only by RCMG-2. Cms CMG-3 was the least specific for restoration as it was restored by all restorer lines used. Cms CMG-3 was the shortest, had the latest anthesis, and was the least vigorous of all the cytoplasms. All three cms CMG germplasms produced stable cms lines in the BC3 generation.

## References


Enns, H., D. G. Dorrell, J. A. Hoes, and W. O. Chubb. 1970. Sunflower research, a progress report. Proceedings 4th International Sunflower Conference. (Memphis, Tenn., 1970): 162-167.

Kinman, M. L. 1970. New developments in the USDA and State Experiment Station sunflower breeding programs. Proceedings 4th International Sunflower Conference. (Memphis, Tenn., 1970): 181-183.

Leclercq, P. 1969. Une sterilite male cytoplasmique chez le tournesol. Ann. Amelior. Plant. 19:99-106.

_____ 1971. La sterelite male cytoplasmique du tournesol. I. Premieres etudes sur la restauration de la fertilite. Ann. Amerior. Plant 21:45-54.

Tatum, L. A. 1971. The southern corn leaf blight epidemic. Science 171:1113-1116.

Vranceanu, V. A., and F. M. Stoenescu. 1971. Pollen fertility restorer gene from cultivated sunflower (Helianthus annuus L.). Euphytica 20:536-541.

Whelan, Ernest D. P. 1980. A new source of cytoplasmic male sterility in sunflower. Euphytica 29:33-46.

_____ and W. Dedio. 1980. Registration of sunflower germplasm composite crosses CMG-1, CMG-2, and CMG-3. Crop Science 20:832.