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# FAO Progress Report 1999-2001

**Working Group :** Identification, Study, and Utilization in Breeding Programs of New Cms Sources.

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**1. Identification of rf genes in perennial *H. mollis* and *H. orgyalis* species.**

Numerous interspecific crosses were performed between female genotypes on Cms-PET1 or CMS-PEF1 cytoplasms and two diploid perennial species (*H. mollis* and *H. orgyalis*) and an outgroup species V*erbesina enceloides* from *helianthineae*.

**CMS-PET1 restoration**

The different situations observed (restored/ segregating/not restored) suggest that *H. mollis, H. orgyalis* and *Verbesina enceloides* wild plants contain homozygous or heterozygous alleles for fertility restoration of CMS PET1, at high frequency. The mean values of male fertility restoration were 53.5 %, 47.8 and 40.9 %, respectively for *H. mollis, H. orgyalis* and *Verbesina*

**CMS-PEF1 restoration**

Rf genes were identified at lower frequency in *H. mollis* and *H. orgyalis* (mean values for fertility restoration were 22.2 % and 16.7 % respectively). The unique hybrid plant obtained with *Verbesina* was not restored on that cytoplasm.

These results confirm that Rf genes for PET1 or PEF1 are rather common in perennial *Helianthus* species as well as in the *Helianthineae* tribe.

| Cytoplasm | Female parent | Male parent | MF | MS | % MF | % MF (average |
|---|---|---|---|---|---|---|
| CMS PET1 | HA89 x AA7-2-4 | H. mollis 230-9 | 25 | 16 | 60.98 | 53.1 |
| | HA99 | H. mollis 230-9 | 16 | 15 | 51.61 | |
| | HA300 | H. mollis 230-9 | 1 | 7 | 12.50 | |
| | HA89 x 89B2 | H.. mollis 230-9 | 2 | 1 | 66.67 | |
| | HA89 x AA7-2-4 | H. mollis 600-8 | 9 | 0 | 100.00 | 100 |
| | HA99 | H. mollis 230-2 | 0 | 7 | 0.00 | 0 |
| | HA89 x AA7-2-4 | H. orgyalis 108 | 8 | 5 | 61.54 | 47.8 |
| | HA99 | H. orgyalis 108 | 1 | 0 | 100.00 | |
| | HA300 | H. Orgyalis 108-8 | 0 | 5 | 0.00 | |
| | HA89 x 89B2 | H. orgyalis | 2 | 2 | 50.00 | |
| | HA89 x AA7-2-4 | Verbesina enceloides 1086 | 2 | 5 | 28.57 | 40.9 |
| | HA99 | Verbesina enceloides 1086-1 | 4 | 4 | 50.00 | |
| | 2603 | Verbesina enceloides 1086-1 | 1 | 1 | 50.00 | |
| | HA89 x 89B2 | Verbesina enceloides 1086-1 | 2 | 3 | 40.00 | |
| CMS PEF1 | D34 x 90HR15 | H. mollis 230-9 | 0 | 3 | 0.00 | 22.2 |
| | 85B3 x 83HR4 | H. mollis 230-9 | 0 | 1 | 0.00 | |
| | 85B3 x 89B2 | H. mollis 230-9 | 2 | 3 | 40.00 | |
| | 85B3 x 83HR4 | H. mollis 230-10 | 0 | 2 | 0.00 | 16.7 |
| | RHA274 | H. orgyalis 108-4 | 1 | 0 | 100.00 | |
| | D34 x 90HR15 | H. orgyalis 108-9 | 0 | 4 | 0.00 | |

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| | | | | | | |
|---|---|---|---|---|---|---|
| | D34 x 90HR15 | H. orgyalis 108 | 0 | 2 | 0.00 | |
| | D34 x 90HR15 | Verbesina encceloides 1086 | 0 | 1 | 0.00 | 0 |

## 2. RECIPROCAL CROSS AND CYTOPLASMIC EFFECTS ON AGRONOMIC TRAITS MEASURED ON ALLOPLASMIC HYBRIDS OF SUNFLOWER (*H. annuus* L.).

Serieys H *et al*. Proc.15th Int. Sunfl. Conf. 12-15 June 2000, Toulouse(Fra.) Vol II E-36, E-41H.

In sunflower, it was shown that the level of several agronomic quantitative traits was governed both by nuclear and cytoplasmic genetic components (Serieys, 1992; Petrov, 1992, Marinkovic *et al*, 1996). Another important aspect, is the study of the cross direction effect on the characters, independently of the nuclear genotype or cytoplasmic background. The purpose of this study was to quantify the level of cytoplasmic and reciprocal cross effects related to important agronomic traits such as flowering period, plant height, oil content and seed yield.

### MATERIAL AND METHODS

Hybrid combinations were investigated in a multilocal design to estimate , the effects of the cross direction, cytoplasmic background and location on agronomic traits. We used three hybrid combinations of the four inbred lines [RHA265, WG], [HA89, WG] and [HA89, RHA274] and nine cytoplasms originated from the following sources: PET1 (Leclercq, 1969), PET2 (Whelan and Dedio, 1980), PEF1 (Serieys et Vincourt, 1987); GIG1 (Whelan, 1981), ANN1, ANN2, ANN3, ANN4 (Serieys et Vincourt, 1987) and ANL2 (Heiser, 1982).

**Tab 1. Studied cytoplasm x hybrid combinations.**

| Cytoplasm | Hybrid combinations | | |
|---|---|---|---|
| | RHA265 /WG | HA89 / WG | HA89/ RHA274 |
| ANL2 | X | X | |
| ANN1 | X | | X |
| ANN2 | X | X | X |
| ANN3 | | X | X |
| ANN4 | X | | X |
| GIG1 | X | | |
| PET1 | | X | |
| PET2 | X | | |
| PEF1 | | | X |

Cytoplasms were compared through series of alloplasmic hybrids as indicated in Table 1. The parental lines of hybrids existed either under male-sterile or male-fertile forms allowing the creation of reciprocal hybrids. F1 hybrids onto ANN1, ANN2, ANN3 and ANN4 cytoplasms did not display any male-fertility restoration, whereas male-fertile plants were found in F1 hybrids with ANL2. Similarly the hybrid combination [WG, RHA265] expressed 13.5 % and 9.4 % of male fertile plants on GIG1 and PET2, respectively.

The experiments, using complete balanced-block design were performed at Montpellier and Toulouse and consisted of three to four replications with 60 to 80 plants each.

### RESULTS (Tables 2 & 3)

Significant effects were registered in most of traits for location, cytoplasmic, croos direction and interaction between cytoplasm and cross direction. But we did not observe cytoplasmic effect for seed moisture, reciprocal effects for plant height or location effects for seed yield.

**Table 2. Variance analysis. Cytoplasmic, reciprocal cross and location effects.**

| Hybrid combination | Trait | Effects | | | |
|---|---|---|---|---|---|
| | | Location | Cytoplasm | Cross direction | Cyto * Cross direction. |
| RHA265 + WG | Height | *** | | | * |
| | Days to flowering | *** | *** | *** | *** |
| | Yield | | *** | *** | ** |
| | Oil | *** | | ** | |
| HA89 + WG | Height | *** | ** | | *** |
| | Days to flowering | | *** | *** | *** |
| | Yield | | *** | ** | *** |
| | Oil | *** | *** | | |
| HA89 + RHA274 | Height | *** | | | |
| | Days to flowering | ** | *** | *** | *** |
| | Yield | | | | |
| | Oil | *** | | | |

(*) $P < 0.05$, (**) $P < 0.01$, (***) $p < 0.001$

**1) Cytoplasmic effects**

**Seed yield**

Two hybrid combinations [WG ,RHA265] and [WG, HA89] expressed significant cytoplasmic effects for seed yield, where the largest differences reached respectively 4.7 and 6.1 $q.ha^{-1}$ (*ie* 13 to 18 % of the mean yield). The ANN2 cytoplasm was outstanding for this trait.

**Flowering time**

The flowering date was significantly affected by the cytoplasmic component in the three hybrids. The largest differences in flowering time varied from 2.4 to 2.7 days. ANN2 induced either significant greater lateness (+ 2.6 and +2.7 days) compared to ANN3) respectively in the [RHA274, HA89] and [WG, HA89] hybrid combination, or the earliest flowering date in the [RHA265, WG] hybrid. These results suggest that flowering time is strongly interacting with the cytoplasmic background of the hybrid.

**Plant height**

Significant cytoplasmic effects were observed only in the [WG, HA89] hybrid. The ANN2 cytoplasm gave the highest plants.

· **Oil content**

Significant cytoplasmic effects were observed only in the [HA89, WG] hybrid. Variation in oil content reached 4.6 % (*ie* 10 % of total content), the highest value was found on ANN2 and the lowest on ANL2 cytoplasms.

**2) Reciprocal cross effects** (Tab 2)

· **Seed yield**

In the hybrids [WG, RHA265] and [WG, HA89], the direction of the cross has significant effect on seed yield. Seed yield was increased by 3.2 and 3.0 $q.ha^{-1}$ respectively when the inbred line WG was used as female parent.

· **Flowering date**

The direction of the cross exerted a significant effect on the flowering time, in all three F1 hybrids. The flowering date was increased by of 2.1 and 2.6 days when WG line was used as

Table 3. Cytoplasmic and reciprocal cross effects.

| Hybrid | Effects | | Yield (q/ha) | Height (cm) | Flowering (days) | Oil content (%) |
|---|---|---|---|---|---|---|
| 1 | Cytoplasm effect | ANL2 | 30.44 b | 159.75 | 7.23 a | 40.70 |
| RHA265 | | ANN1 | 30.54 b | 170.83 | 7.09 a | 40.32 |
| | | ANN2 | 34.99 a | 166.54 | 4.80 d | 39.72 |
| + | | ANN4 | 32.49 b | 163.33 | 5.16 d | 40.48 |
| | | PET2 | 30.28 b | 164.40 | 5.87 c | 39.88 |
| WG | | GIG1 | 31.75 b | 170.47 | 6.45 b | 39.77 |
| | | Sign. Pr > F | **0.001** | **0.008** | **0.001** | **0.002** |
| | Cross direction effect | RHA265 x WG | 30.13 | 167.25 | 7.11 | 39.58 |
| | | WG x RHA265 | 33.37 | 164.52 | 5.09 | 40.71 |
| | | Sign. Pr > F | **0.004** | 0.320 | **0.001** | 0.080 |
| 2 | Cytoplasm effect | ANL2 | 26.89 c* | 133.87 b | 4.13 b | 41.52 b |
| | | ANN2 | 33.01 a | 142.44 a | 5.70 a | 46.12 a |
| WG | | ANN3 | 27.71 bc | 135.65 b | 2.99 c | 44.30 c |
| | | PET1 | 30.53 ab | 135.65 b | 5.27 a | 45.84 a |
| + | | Sign. Pr > F | **0.001** | **0.008** | **0.001** | **0.002** |
| HA89 | Cross direction effect | HA89 x WG | 28.03 | 137.79 | 5.81 | 45.12 |
| | | WG x HA89 | 31.04 | 136.01 | 3.24 | 43.78 |
| | | Sign. Pr > F | **0.004** | 0.320 | **0.001** | 0.080 |
| 3. | Cytoplasm effect | ANN1 | 32.30 | 143.13 | 5.62 a | 43.52 |
| | | ANN2 | 30.66 | 144.91 | 6.20 a | 43.60 |
| | | ANN3 | 35.15 | 145.27 | 3.62 b | 43.43 |
| RHA274 | | ANN4 | 33.99 | 142.77 | 3.91 b | 44.50 |
| | | PEF1 | 33.84 | 150.24 | 6.14 a | 44.24 |
| + | | Sign. Pr > F | 0.061 | 0.230 | **0.001** | 0.450 |
| HA89 | Cross direction effect | HA89 x RHA274 | 32.54 | 145.06 | 4.51 | 44.03 |
| | | RHA274 x HA89 | 33.84 | 145.47 | 5.69 | 43.69 |
| | | Sign. Pr > F | 0.190 | 0.850 | **0.001** | 0.440 |

(*) Newman & Keuls test (0.05 level)

female parent in the crosses [WG, RHA265] and [WGHA89], respectively; and by 1.2 days when the RHA274 inbred line was used as female in the [RHA274, HA89] hybrid.

**Oil content**

Oil content was significantly modified (+ 1.1 %) by the direction of the cross in [WG, RHA265], when the WG inbred line was used as female parent.

## CONCLUSIONS

The level of agronomic traits in alloplasmic hybrids frequently appeared under dependance either of the cytoplamic background and / or the direction of the cross.

· **Significant cytoplasmic effects were registered for flowering period, plant height, seed moisture, oil content and seed yield.** In the three hybrid combinations the largest effects on seed yield due to cytoplasm varied from 13.5 % to 18.2 %. For oil content, the range of CMS effect reached 10 % (between ANN2 and ANL2 CMS sources).

· **Reciprocal effects were clearly identified for important agronomic traits** The most striking effects are linked to changes in flowering date, seed yield and oil content So, days to flowering varied in the range of 1.2 to 2.6 days, seed yield in the range 3.0 - 3.3 q. ha $^{-1}$ and oil content in the range 0.4 to 1.3 to %. In our experiment, the magnitude of cross direction effect appeared always lower than cytoplasmic effects.

These results underline the importance of CMS background and direction of the cross in the performance of sunflower hybrids, suggesting that these parameters should be taken in consideration in the breeding programmes.

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