M. Feldman
Submitted to American Journal of Botany
1980

# THE RELATIONSHIPS BETWEEN MALE AND FEMALE FERTILITY AND AMONG TAXA IN DIPLOID WHEATS[1]

HARI C. SHARMA AND J. GILES WAINES

Department of Botany and Plant Sciences

University of California, Riverside, California 92521

## ABSTRACT

Pollen stainability appears to be a reliable indication of the ultimate seed set in diploid interspecific hybrid and backcross populations in *Triticum* L. The correlation between percent pollen stained and number of seeds set is positive and highly significant (r = 0.92). Such a significant correlation is of value in selecting among large hybrid and backcross progenies for fertility at an early stage without cytological preparations. Estimates of male and female fertility in the hybrids and backcrosses are interpreted to indicate that the domesticated diploid *Triticum monococcum* L. is genetically closer to wild *T. boeoticum* Boiss. em. Schiem. than to wild *T. urartu* Tum., and that *urartu* is not a variety of *monococcum* or *boeoticum*, but rather a separate species. The $F_1$ hybrids and backcrosses between *monococcum* and *boeoticum* are

---

[1] Received for publication ______________________.

This paper is part of the Ph.D. thesis of Hari C. Sharma, to be submitted to the University of California, Riverside. We express gratitude to Dr. F. C. Vasek and Dr. K. W. Foster for valuable suggestions in improving the manuscript.

This research was supported in part by the California Agricultural Experiment Station, USDA Hatch Funds, and SG 616-15-59.

normally male and female fertile. The $F_1$ hybrids between T. monococcum and T. urartu are completely sterile. Backcross 1 and 2 populations are completely to partially sterile. Partial sterility exists even in backcross 3. Thus, there appears to be a strong reproductive isolation between monococcum and urartu.

In cross and backcross populations of interspecific Triticum hybrids where some proportion of the gametes is imbalanced, pollen stainability has been used to estimate male-fertility (Vardi, 1974; Johnson and Dhaliwal, 1976; Schultz-Shaeffer and McNeal, 1977, and many others). However, the relationship between pollen stainability and seed set is unclear. If this relationship is established, pollen stainability can be used as a simple and inexpensive criterion to estimate ultimate fertility. This is especially important in screening large interspecific hybrid and backcross populations for fertility in biosystematic investigations, or in breeding programs where genes are being transferred from one species to another, in that at any backcross stage, individuals with a higher percentage of stainable pollen can be selfed to get enough seed to recover homozygotes for the gene under transfer. This relationship has been investigated in Crepis (Poole, 1931, 1932), Silene (Kruckeberg, 1955), Bromegrass (Bromus inermis Leyss.) (Jalal and Nielsen, 1965 and Pattanayak, 1968), timothy (Phleum pratense L.) (Nath and Nielsen, 1962 and Franckowiak, 1970) and Solanum nigrum (Schilling, unpublished), but with differing opinions.

Triticum monococcum L., T. boeoticum Boiss. em. Schiem. and T. urartu tum. are three diploid (2n = 14) wheat taxa. T. monococcum is domesticated, T. boeoticum and T. urartu are wild. Vilmorin (1880, a, b) and Blaringhem (1927) first reported fertile hybrids between monococcum and boeoticum. Many taxonomists regard these two taxa as a single biological species. In spite of this, T. monococcum resembles T. urartu rather than T. boeoticum in several characters which include seed color, spike density, and absence of hairs on the leaf blade. In Feldman's (1977) view, urartu should also be considered a variety of monococcum.

The present study was undertaken to answer two questions: 1) What is the relationship between pollen stainability and seed set in hybrid and backcross populations between monococcum and boeoticum and between monococcum and urartu?

2) On the basis of male and female fertility estimates in hybrid and backcross populations, what is the taxonomic relationship between these three diploid wheats?

MATERIALS AND METHODS--Materials comprised different accessions of Triticum monococcum, T. boeoticum (Table 1) and T. urartu (Table 2). T. monococcum x T. boeoticum hybrids and their reciprocals were made between different accessions of the parents. $BC_1$ seed was produced using boeoticum as the recurrent parent. Similarly, monococcum x urartu hybrids and their reciprocals were made and $BC_1$, $BC_2$, and $BC_3$ seed was produced using urartu as the recurrent parent.

$F_1$ hybrids and backcross plants were grown in the greenhouse along with the parents. The male fertility of the individual hybrid and backcross plants was measured in terms of percentage of pollen stained in 1% KI solution in 50% ethanol based on about 300 pollen grains. Female fertility was measured in terms of total number of selfed seeds obtained. These data were taken only on those plants that were uniform in general growth and tillering. Data from hybrids and backcrosses were included to cover a whole range of values for the two variables for the purpose of meaningful correlation. Correlation coefficients between percent pollen stained and number of seeds set were calculated in respect of monococcum x boeoticum hybrids and backcrosses, and in respect of monococcum x urartu hybrids, their reciprocals, and backcrosses separately as well as for both groups together. The significance of each correlation coefficient was tested by a t-test.

RESULTS AND DISCUSSION--A correlation between pollen stainability and seed set on selfing is apparent. All the parents involved had more than 92% pollen stainability and were fully fertile. The correlation coefficient between pollen stainability and seed set among 9 $F_1$ hybrid and backcross plants involving monococcum and boeoticum (Table 1) is positive (0.89) and significant at the 1% level. $F_1$ hybrids between monococcum and urartu as well as between

urartu and monococcum had zero pollen stainability and did not set any seed on selfing. Four of the five $BC_1$ plants involving urartu and monococcum (Table 2) also had zero pollen stainability and zero seed set. The correlation coefficient between these two traits among all 26 $BC_1$, $BC_2$, and $BC_3$ plants (Table 2) is positive (0.94) and highly significant. The overall correlation for all the 35 pairs in Tables 1 and 2 is 0.92, which is again significant at the 1% level. These results are interpreted to indicate that pollen stainability is a good measure of pollen viability and potential seed set in diploid/diploid wheat hybrids. Nath and Nielsen (1962) in timothy and Jalal and Nielsen (1965) and Pattanayak (1968) in bromegrass have reported low relationships between pollen stainability and seet set. However, Poole (1931, 1932) in Crepis and Franckowiak (1970) in timothy observed significant correlations between male and female fertility. The low pollen fertility in both the homoploid and heteroploid crosses was found to be correlated with low seed set in Silene (Kruckeberg, 1955). Preliminary studies of Schilling (unpublished) revealed that seed set is highly correlated with pollen stainability in Solanum nigrum. In diploid wheats we also report a highly significant correlation between these two traits.

It was not the intention here to emphasize the production of bad pollen through hybridization, but rather the use of good pollen percentages for quick and early identification of more fertile hybrids and their backcrosses.
An estimate of the fertility can be made from cytological studies, but this method is very laborious, and it may not be practicable for large populations in certain biosystematic investigations or breeding programs. Our results indicate that if the plants differ in their genetic constitution, the more fertile ones can be identified by the study of their pollen stainability.
The information is of special interest to plant biosystematists and to breeders who screen large hybrid and backcross progenies for fertility in interspecific

hybridization programs aimed at transferring genes from one species to another.

Variability for pollen stainability and seed set occurred among backcross individuals involving monococcum and urartu (Table 2). These data were, therefore, suitable for correlation calculations between these two traits. This variation is largely attributed to random elimination of the chromosomes of non-recurrent species (monococcum) such that different backcross individuals are at different degrees of return to the parental chromosomal constitution.

Fertility estimates indicate that the $F_1$ plants of monococcum x boeoticum have high seed set and high pollen stainability. Actual data were not taken in respect of reciprocals, but they also had high seed set. $BC_1$ plants involving these species have normal pollen stainability and normal seed set (Table 1) just like the parents. In addition to the 5 $BC_1$ plants of Table 1, 15 other $BC_1$ plants were scored and were found to be equally fertile. The high pollen stainability, high seed set and ring bivalents in meiotic metaphase I in hybrids between monococcum and boeoticum found by us support the conclusion of Vilmorin (1880a, b) and Blaringhem (1927) that these two taxa can readily be crossed to produce fertile hybrids and that these hybrids exhibit normal meiosis. In the light of these facts, monococcum and boeoticum should be considered as one and the same species.

In contrast, hybrids between monococcum and urartu, or between urartu and monococcum had zero pollen stainability and did not set any seed on selfing. Only 2 $BC_1$ individuals, (G3371m x G1828u) x G2961u-1 and (G1841u x G3373m) x G3248u-1 had 12.5% and 50.0% pollen stainability and bore 42 and 202 seeds respectively. The remainder of the $BC_1$ plants had 0.0-0.5% pollen stainability and zero seed set (Table 2). There was evidence of increase in fertility in $BC_2$, but only very few plants had good seed set like the parents. Out of 83 $BC_2$ plants, including 7 in Table 2, 48 plants did not set any seed. Most of the remaining 35 plants also had very low seed set. Fourteen of these had

only 2-17 seeds. In $BC_3$ plants (Table 2) there was again improvement in both male and female fertility. However, out of 11 plants, only two had pollen stainability like the parents; most had fertility levels much lower than normal.

The zero pollen stainability and zero seed set in *monococcum* x *urartu* and *urartu* x *monococcum* hybrids and zero to only partial fertility in backcross generations does not support the view that *urartu* is a variety of *monococcum* (Feldman, 1977), but rather that it is a separate species, which exhibits considerable reproductive isolation. *Triticum urartu* is reproductively isolated from *T. boeoticum* also (Johnson and Dhaliwal, 1976).

Backcrossing the interspecific hybrid to one of the parents leads to the elimination of chromosomes of the other parent, less abnormal meiosis and improvement in fertility. Fertility tends to increase directly with the degree of return of the hybrids to the parental chromosome constitution. The larger the number of genetic factors which contribute to the original sterility, the smaller is the number of fertile lines which can be extracted in a given number of generations (Stebbins, 1969). Seventy-five percent of the $BC_1$ plants and 59% of the $BC_2$ plants involving *monococcum* and *urartu* were completely sterile. Such a high proportion of sterile plants cannot be due to environmental factors. Failure of seed set was not because of stunted growth or any other noticeable abnormality. These plants were equally vigorous and healthy. Such a low fertility in *monococcum* x *urartu* and *urartu* x *monococcum* backcrosses compared to those between *monococcum* and *boeoticum* can be interpreted to indicate that *monococcum* is genetically closer to *boeoticum* than to *urartu* and that the reproductive barrier between *monococcum* and *urartu* is strong.

## LITERATURE CITED

BLARINGHEM, L. 1927. Afinités des blés sauvages *Triticum aegilopoides* Balansa et *Tr. monococcum* L., demontrees par leurs hybrides reciproques. Compt. Rend. Acad. Sci., Paris 184:225-227.

FELDMAN, M. 1977. Historical aspects and significance of the discovery of wild wheats. Stadler Symp. 9:121-146. Univ. of Missouri, Columbia.

FRANCKOWIAK, J. D. 1970. Cytological and morphological evaluations of back-cross progenies from tetraploid x hexaploid forms of *Phleum pratense* L. Ph.D. Thesis, Univ. of Wisconsin.

JALAL, S. M., AND E. L. NIELSEN. 1965. Interrelations among reproductive stages of *Bromus inermis* Leyss. Corp Sci. 5:401-403.

JOHNSON, B. L., AND H. S. DHALIWAL. 1976. Reproductive isolation of *Triticum boeoticum* and *T. urartu* and the origin of the tetraploid wheats. Amer. J. Bot. 63:1088-1094.

KRUCKEBERG, A. R. 1955. Interspecific hybridization of *Silene*. Amer. J. Bot. 42:373-378.

NATH, J. AND E. L. NIELSEN. 1962. Interrelations of reproductive stages of timothy. Crop Sci. 2:49-51.

PATTANAYAK, C. M. 1968. Studies on inter- and intra-plant compatibility for seed set and certain reciprocal cross effects for forage yield and plant height in *Bromus inermis* Leyss. Ph.D. Thesis, Univ. of Wisconsin.

POOLE, C. F. 1931. The interspecific hybrid *Crepis rubra* x *C. foetida* and some of its derivatives. I. Univ. of California Publ. Agric. Sci. 6:169-200.

POOLE, C. F. 1932. Pollen grain studies as an indication of fertility in hybrids. Genetics 17:125-136.

SCHULTZ-SHAEFFER, J. AND F. H. McNEAL. 1977. Alien chromosome addition in wheat. Crop Sci. 17:891-896.

STEBBINS, G. L. 1969. Developmental genetics and plant evolution. Jap. J. Genet:44(Suppl.):344-350.

VARDI, A. 1974. Introgression from tetraploid _durum_ wheat to diploid _Ae. longissima_ and _Ae. speltoides_. Heredity 32:171-181.

VILMORIN, H. De. 1880a. Note sur un croisement entre deux espéces de blés. Bull. Soc. Bot., France 27:73-74.

__________ 1880b. Essais du croisement entre blés différents. Bull. Soc. Bot., France 27:356-361.

TABLE 1. Pollen stainability and seed set on selfing in $F_1$ hybrids and $BC_1$ individuals between *monococcum* (m) and *boeoticum* (b)

| Cross/ backcross | Particulars of cross/backcross | Pollen Stainability % | No. of Seeds |
|---|---|---|---|
| $F_1$ | | | |
| m x b | G3315m x G2358b | 81.9 | 105 |
| | G3361m x G2576b | 84.4 | 152 |
| | G1560m x G2960b | 83.0 | 195 |
| | G2908m x G641b | 87.7 | 250 |
| $BC_1$ | | | |
| m x b x b | (G1560m x G2960b) x G2960b-1 | 84.0 | 180 |
| | (G1560m x G2960b) x G2960b-2 | 87.0 | 185 |
| | (G1560m x G2960b) x G2960b-3) | 96.2 | 295 |
| | (G2908m x G641b) x G1916b-1 | 87.2 | 235 |
| | (G2908m x G641b) x G1916b-2 | 93.0 | 290 |

TABLE 2. Pollen stainability and seed set on selfing in $BC_1$, $BC_2$, and $BC_3$ individuals between monococcum (m) and urartu (u) and between urartu and monococcum

| Backcross | Particulars of Backcross | Pollen Stainability % | No. of Seeds |
|---|---|---|---|
| $BC_1$ | | | |
| m x u x u | (G3371m x G3227u) x G3248u | 0.5 | 0 |
| | (G3371 m x G1828u) x G2961u-1 | 12.5 | 42 |
| | (G3371m x G1828u) x G2961u-2 | 0.5 | 0 |
| u x m x u | (G1834u x G3371m) x G3248u-1 | 0.0 | 0 |
| | (G1834u x G3371m) x G3248u-4 | 0.0 | 0 |
| | (G1841u x G3373m) x G3248u-1 | 50.0 | 202 |
| | (G1841u x G3373m) x G3248u-2 | 0.0 | 0 |
| | (G1841u x G3372m) x G3248u | 0.0 | 0 |
| $BC_2$ | | | |
| m x u x u x u | (G3371m x G3227u) x G3248u x G3248u | 5.0 | 0 |
| | (G3371m x G1828u) x G2961u-1 x G2961u | 25.0 | 72 |
| | (G3371m x G1828u) x G2961u-2 x G2961u | 10.0 | 17 |
| u x m x u x u | (G1834u x G3371m) x G3248u-1 x G3248u-1 | 40.0 | 50 |
| | (G1834u x G3371m) x G3248u-1 x G3248u-2 | 45.0 | 160 |
| | (G1834u x G3371m) x G3248u-1 x G1834u | 85.0 | 301 |
| | (G1834u x G3371m) x G3248u-4 x G3248u | 15.0 | 4 |

TABLE 2 (cont)

| Backcross | Particulars of backcross | Pollen stainability % | No. of Seeds |
|---|---|---|---|
| $BC_3$ | | | |
| m x u x u x u x u | [ (G3371m x G3227u) x G3248u x G3248u] x u-1* | 12.0 | 15 |
| | [ (G3371m x G3227u) x G3248u x G3248u] x u-2* | 20.0 | 20 |
| | [ (G3371m x G1828u) x G2961u-1 x G2961u] x u-1* | 35.5 | 70 |
| | [ (G3371m x G1828u) x G2961u-1 x G2961u] x u-2* | 20.0 | 15 |
| | [ (G3371m x G1828u) x G2961u-1 x G2961u] x u-3* | 60.5 | 70 |
| u x m x u x u x u | [ (G1834u x G3371m) x G3248u-1 x G3248u-1] x G3248u-1 | 95.0 | 310 |
| | [ (G1834u x G3371m) x G3248u-1 x G3248u-1] x G3248u-2 | 81.0 | 195 |
| | [ (G1834u x G3371m) x G3248u-1 x G3248u-2] x u-1* | 80.0 | 275 |
| | [ (G1834u x G3371m) x G3248u-1 x G3248u-2] x u-2* | 95.0 | 280 |
| | [ (G1834u x G3371m) x G3248u-1 x G3248u-4] x u-1* | 25.0 | 18 |
| | [ (G1834u x G3371m) x G3248u-1 x G3248u-4] x u-2* | 30.0 | 25 |

* Record of accession used as pollen source not kept.