# Experimental study of gene flow between wild and cultivated *Pennisetum glaucum*

**J.-F. Renno, T. Winkel, F. Bonnefous, and G. Bezançon**

Abstract: Under natural conditions, wild and cultivated pearl millet, *Pennisetum glaucum* (L.) R.Br., exchanged genes for millenia and, nevertheless, maintain high morphological differentiation. Under experimental conditions in the Sahel, hybridization between wild and cultivated pearl millet was measured using isozymic markers and interpreted in relation to the phenology of the plants. Gene flows were asymmetric, engendering 8% of hybrids in the progeny of the wild phenotype, 45% in that of the cultivated phenotype, and 39% in that of the intermediate "shibra" phenotype; these last two phenotypes constitute the sample of cultivated pearl millet. The proportion of hybrids in the progeny of the wild sample was time dependent during the flowering phase of cultivated pearl millet. The proportion of hybrids produced by the cultivated pearl millet was not time dependent. In the seeds produced by the cultivated phenotype along its reproductive phase, the proportion of viable seeds was negatively correlated with the frequency of hybrids. Likewise, the speed of germination of seeds produced by the cultivated or the shibra phenotypes was negatively correlated with the frequency of the hybrids that they contained. The effects of balancing among genetic intermixing, isolation and reproduction barriers, and differential anthropic and natural selection pressures are discussed to better understand the evolution and the maintenance of the polymorphism of *Pennisetum glaucum*.

*Key words*: pearl millet, wild pearl millet, *Pennisetum glaucum*, gene flow, domestication, hybrid.

Résumé : Dans la nature, les formes sauvage et cultivée du mil (*Pennisetum glaucum* (L.) R.Br.) échangent des gènes depuis des millénaires et maintiennent cependant une forte différenciation morphologique. En conditions expérimentales au Sahel, les flux de gènes entre mils sauvage et cultivé ont été mesurés à l'aide de marqueurs isozymiques et interprétés en relation avec la phénologie des différentes formes de mil. Ces flux étaient dissymétriques, produisant 8% d'hybrides chez les descendants du phénotype sauvage, contre 45% chez ceux du phénotype cultivé et 39% chez ceux du phénotype intermédiaire « chibra », ces deux derniers phénotypes constituant l'échantillon de mil cultivé. Les effets des échanges géniques sur les variations temporelles de la proportion d'hybrides chez les descendants de l'échantillon de mil sauvage étaient modulés par le cycle de floraison des plantes du phénotype cultivé, tandis que leurs effets sur la formation d'hybrides dans l'échantillon de mil cultivé sont apparus indépendants du temps. Le taux de graines viables produites par le phénotype cultivé à différents stades de la phase reproductive, ainsi que la vitesse de germination des graines issues des phénotypes cultivé ou chibra, étaient corrélés négativement à la fréquence d'hybrides qu'elles contenaient. Les effets de l'équilibre entre brassage génétique, isolements et barrières à la production, pressions de sélection anthropique et naturelle, sont discutés en vue de mieux comprendre l'évolution et le maintien du polymorphisme chez *Pennisetum glaucum*.

*Mots clés* : mil, mil sauvage, *Pennisetum glaucum*, flux de gènes, domestication, hybrides.

## Introduction

Domesticated and wild pearl millet have coevolved in a large part of the Sahelian region from neolithic times. The most ancient archaeological records of cultivated pearl millet in sympatry with the wild relative dates to about 3000 years BP (Amblard and Pernès 1989), proving that domestication dates back millenia. Pearl millet is so polymorphic that it was initially considered to be a polyspecific taxon. Hutchinson and Dalziel (1936) distinguished a dozen species among wild and cultivated pearl millet, while Clayton (1972) recognized one cultivated and four wild species. More recently, these different "morphological species" were regrouped in *Pennisetum glaucum* (L.) R.Br., according to the biological species concept (Mayr 1974), and divided into three subspecies (Brunken 1977; Brunken et al. 1977). Then Van der Zon (1992) retained the nomenclature *P. glaucum* subsp. *glaucum* for the cultivated phenotype, *P. glaucum* subsp. *violaceum* (Lam.) A. Rich. for the wild phenotype, and *P. glaucum* subsp. *sieberianum* (Schlecht.) Stapf & Hubb. for the intermediate form. Currently the differentiation of *P. glaucum* into three principal morphological infraspecific taxa (cultivated, wild, and intermediate) is accepted.

Pearl millet is an annual species, diploid ($2n = 2x = 14$), sexual, hermaphrodite, and preferentially allogamic as a result of a strongly marked protogyny and an anemophilic pollination (Pernès 1986; Sandmeier 1993). These characteristics favour gene exchange over generations within and among wild and cultivated pearl millet forms that are geneti-

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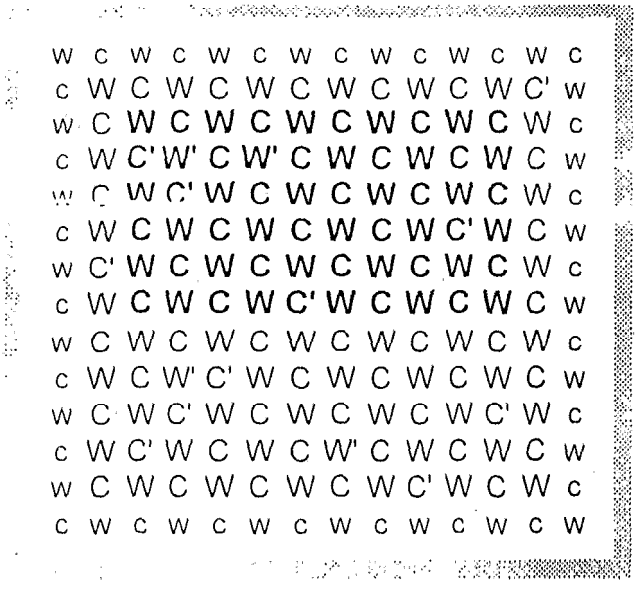
Fig. 1. Plan of the experimental plot. C, cultivated phenotype; C', shibra phenotype from the cultivar; W, wild phenotype; W', shibra phenotype from the wild pearl millet. Lowercase letters represent border plants; uppercase letters represent the plants used to measure the gene flow and the production of spikes and seeds; boldface uppercase letters represent the plants observed for male flowering (female flowering was observed on every plant, including the borders).




cally very close according to the isozymic studies (Tostain 1992). Consequently, pearl millet fields and the wild populations can be invaded by forms intermediate between the cultivated and wild phenotypes, called "shibras," a name derived from the Haoussa language of Niger. The shibras produce fertile progeny when crossed with other shibras or with wild or cultivated phenotypes. In spite of gene exchange in *P. glaucum*, the differentiation between wild and cultivated phenotypes has been maintained for millenia. To explain this phenomenon, two principal factors that limit gene exchange in *P. glaucum* ought to be considered: reproductive isolation and reproductive barriers. Reproductive isolation is the consequence either of spatial isolation due to allopatry or, in sympatric situations, of temporal isolation when phenological cycles of the different genotypes do not coincide entirely. This situation is observed in pearl millet, the wild form of which has a flowering period much more extended than that of the cultivated form (Renno and Winkel 1996). When there is no reproductive isolation, gene exchanges are possible, but reproductive barriers contribute to reduce them. First, prezygotic barriers come into play, such as pollenic competition (Sarr et al. 1988; Robert et al. 1991); then, postzygotic barriers result in reduced viability of hybrid seeds (Amoukou and Marchais 1993). However, when there is gene flow and hybridization between cultivated and wild pearl millet, the domestication syndrome may reappear in the next generations owing to genetic recombinations determined by a few closely linked recessive alleles (Pernès et al. 1984).

What is the proportion of exchange between wild and cultivated pearl millet? Are the gene flows symmetrical between wild and cultivated pearl millet? How are they influenced by the phenology of the plants? What are their effects on the frequency of hybrids likely to contribute to the following generations? This study addresses these questions by tracing under experimental conditions the gene flow between wild and cultivated pearl millet in relation to the reproductive processes and focuses on their consequences on the viability and germination of their progeny.

## Materials and methods

### Basic principles

The experimental protocol corresponds to a compromise between two partly conflicting requirements:

(1) Control of the spatial distribution of the plants to avoid bias originating from the distance between plants and from the variations in environmental conditions.

(2) Approximation of the natural conditions of growth and development of pearl millet plants so that the experimental results have a biological significance in accordance with a real situation.

Contrary to the use of fixed lines, the use of seeds taken from the agrosystem permits access to part of the genetic variability of pearl millet, as it is expressed in its Sahelian environment. During the rainy season in Niger, plants originating from cultivated and wild pearl millet populations were left to open pollination in a plot isolated from other crops. Planted out to a checkerboard plan (with 1 plant/m$^2$), each individual plant had similar neighbours: four cultivated and four wild plants (Fig. 1). Before setting them in place, the wild and cultivated samples were differentiated into two categories of homozygotes by two allozymes each corresponding to an allele of the same gene, which served as a marker. In the progeny of the plants of the experimental plot, the heterozygous individuals of the marker gene (F1 shibra) should be the product of a hybridization of wild and cultivated pearl millet. The frequency of these hybrids in the progeny of one sample thus accounted for the fertilizing gene flow emitted by the other sample. The gene used for sorting, that of the enzyme phosphoglucomutase (PGM, E.C. 5.4.2.2.), is not related to the genes of the domestication syndrome (Tostain 1993), which makes it an appropriate choice for a neutral marker for the phenotypes under study. Differences observed between cultivated and wild samples were tested by a $t$ test. Coefficients of correlation were tested at the 5% significance level.

### Origin of the material

The seeds used to set up the experimental plot originated from the region geographically defined by three towns in Niger: Tanout (14°58'N, 8°53'E), Belbeji (14°10'N, 8°00'E), and Bakin Birji (14°15'N, 8°47'E). The land race "Ankoutess" is typically found in this region (Clément 1985). The Ankoutess seeds were selected by farmers from several hundred cultivated plants. The wild seeds were collected from several hundred individuals displaying the wild phenotype in spontaneous populations near pearl millet fields.

### Experimental setup

The experiment was performed during the rainy season in Niamey (Niger) from July to November 1994. To avoid possible pollution by pollen from surrounding fields, planting was delayed by 1 month compared with the normal for the region, and the plot, located far from any agricultural zone, was surrounded by a protective border of pearl millet and walls 2.5 m high. Seeds were sown in sand in pots. To replicate natural conditions as closely as possible the cultivated pearl millet seeds were embedded 4 or 5 cm deep, and those of wild pearl millet were sown on the surface. Emergence was synchronous in both forms of pearl millet, and after 3 days no new plants emerged. A leaf fragment from each of the young plants was analyzed by starch gel electrophoresis to identify alleles at the *Pgm* locus according to the protocol described by Wendel and Weeden

(1989). At 7 days after sowing (DAS), all the young plants were sorted by the *Pgm* marker and transplanted to the experimental plot (Fig. 1). In total, 196 plants were put in place (98 wild and 98 cultivated). Plants on the border were not used in the analysis of gene flow, but since they participated in the fertilization of the plot, their flowering was observed. Thus, 144 plants (72 wild and 72 cultivated) were used to quantify gene flow and to measure the production of spikes and seeds.

### Observations on the phenology of flowering

Flowering was followed from the first to the last spike on each of the 196 plants for the female flowering and on each of 60 plants (30 cultivated and 30 wild) for the male flowering (Fig. 1). For each of the plants observed the number of spikes at the female stage and the number of spikes at the male stage were noted daily until 113 DAS, then every 2 days until the last spike had flowered. The date of the female stage of each spike was marked using a coloured label. A spike was considered to be female with the appearance of the first pistils and male with the appearance of the first anthers. In this way, the female stage corresponds to the beginning of a possible fertilization of the spike by the pollen cloud, while the male stage indicates that the spike is about to emit pollen.

In the experimental plot, as in nature, each of the cultivated and wild samples included a fraction of plants of the intermediate phenotype (shibra), distinguishable only at the moment of flowering. The cultivated sample was constituted of plants of the cultivated (CLT) phenotype and of plants of the shibra (SHB) phenotype. The wild sample was constituted of plants of the wild (WLD) phenotype and of SHB plants. This latter group was too small (four plants) to be significant for study. Spikes were harvested as they matured, grouped into WLD, CLT, or SHB, and subgrouped according to the date at which they had reached the female stage.

### Daily variations in the production of seeds and the number of fertilized ovules

For each day, the number of efficient fertilizations (i.e., producing seeds) was estimated by the number of seeds engendered by the spikes that had been noted as being in female flowering on that day, assuming that the ovules of a spike are fertile only on the day when it is considered to be female. This is an approximation because in reality a spike stays female for several days. The number of seeds produced per day was counted with an electric counter. In the case of the cultivated sample, all the spikes were threshed and all the seeds from each day were counted. In the case of the wild sample the number of seeds had to be estimated, the quantity of spikes being too great to apply the same method as for the cultivated pearl millet. From the beginning of the flowering period of the wild phenotype until the end of the flowering period of the cultivated one, six successive time intervals, each corresponding to the production of 20 female spikes by the mean wild plant, were distinguished a posteriori (at the end of the experiment). The remainder of the flowering period of the wild plants was considered as a seventh time interval. For each time interval, 10 harvested plants were taken at random (20 in the last time interval). The seeds of all the spikes produced by these plants on the date corresponding to the middle of the time interval were counted. For each time interval the mean number of seeds estimated per spike, multiplied by the total number of spikes produced by all the wild plants of the plot on each day, allows the estimation of the production of seeds for each day of the flowering period.

### Pollen cloud and time lapse between male and female flowering

For each phenotype, the time variations in the percentage of hybrids were considered in relation to the time variations in the pollen cloud of the other phenotype. The intensity of the pollen cloud was estimated from the daily variations in seed production, assuming that for any given spike the average length of receptivity and emission of pollen varies little during the flowering cycle and that seed production is highly correlated to the number of pollen receptors (ovules) and emitters (anthers). To relate the time variations in the percentage of hybrids and the pollen clouds, the time lapse between the female and the male stages should be taken into account. It was obtained by calculating the mean interval of time between successive steps of the cumulative curves of female and male flowering.

### Proportion of viable seeds

The proportion of germinating (thus viable) seeds produced in the trial was measured on the samples that were produced each day for WLD and CLT, and on those produced 1 of 2 days for SHB. For CLT and SHB, subsamples of 150 seeds were taken at random for each day. The same sampling method was used for the wild for each day until the end of the flowering period of CLT (83 DAS). Two more subsamples were taken at 88 and 95 DAS to characterize the end of the flowering period of WLD. The size of these two subsamples was 450 seeds to increase the number of hybrids observed and thus to compare them statistically. To promote germination, the seeds were placed for 15 days in an oven at 40°C. Then, they were placed in Petri dishes on blotting paper soaked in water and a fungicide. The germination of the seeds was observed to occur within a maximum of 3 days, and five distinct periods were distinguished as follows: period 1 (0–8 h), period 2 (8–16 h), period 3 (16–24 h), period 4 (24–48 h), and period 5 (48–72 h). There were no more germinations after 72 h.

### Isozyme analysis

The measurement of gene flow was obtained after enzymatic electrophoresis of the subsamples that had been used to determine the proportion of viable seeds. All the seedlings were analyzed as and when they germinated. In each group of pearl millet (WLD, CLT, SHB), the frequency of the heterozygotes *Pgm (100, 104)* accounted for the magnitude of the gene flow. For each phenotype the global percentage of viable hybrids formed during the entire growing season ($H_T$) was estimated by the weighted average:

$$[1]\quad H_T = \sum_{DAS=1}^{n} (H_{DAS} VS_{DAS}) / \sum_{DAS=1}^{n} VS_{DAS}$$

where $H_{DAS}$ is the percentage of hybrids formed on a given DAS, and $VS_{DAS}$ is the number of viable seeds produced on this DAS.

## Results

Of the 196 plants observed for their phenology, 144 plants were used to quantify the gene flow and to measure the production of spikes and seeds. They divide into two groups as follows (Fig. 1):

(1) 72 plants, homozygotes *Pgm (104, 104)*, constituting the wild sample, of which 68 were WLD; the 4 others of the shibra phenotype were not used for the gene flow analysis;

(2) 72 plants, homozygotes *Pgm (100, 100)*, constituting the cultivated sample, of which 60 were CLT and 11 were of SHB. One CLT plant did not produce spikes and consequently was not usable.

To describe the flowering phenology, 22 000 spikes were observed and labelled in the field. To measure the gene flow, 7 942 seedlings were analyzed: 2 597 produced by CLT, 1 503 by SHB, and 3 842 by WLD.

Fig. 2. Variations in the mean number of female spikes for cultivated, wild, and shibra pearl millet plants during the flowering cycle.

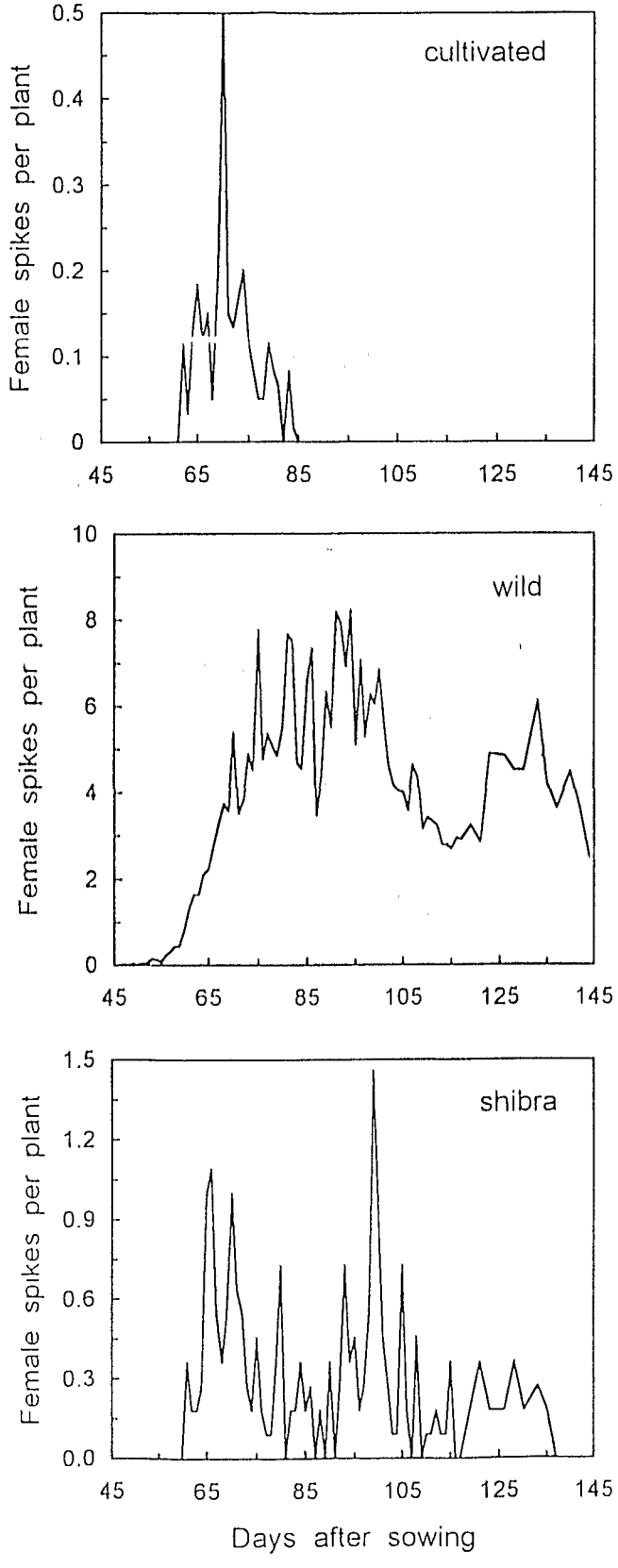



Table 1. Mean number of spikes and seeds per plant, percentage of seeds germinated, and percentage of hybrids in the production of seeds for each of the three pearl millet phenotypes.

| | Spikes per plant | Seeds per plant | Germinated (%) | Hybrids (%) |
|---|---|---|---|---|
| Cultivated | | | | |
| Mean | 2.8 | 7 861 | 80.1 | 50.2 |
| *N* | 60 | 60 | 22 | 22 |
| CV (%) | 50 | 55 | 16 | 21 |
| SE | 0.2 | 559 | 2.9 | 2.3 |
| Wild | | | | |
| Mean | 319 | 56 359 | 91.8 | 11.3 |
| *N* | 68 | 68 | 24 | 24 |
| CV (%) | 53 | 49 | 6 | 68 |
| SE | 20 | 3 365 | 1.2 | 1.6 |
| Shibra | | | | |
| Mean | 21.1 | 11 134 | 71.2 | 41.9 |
| *N* | 11 | 11 | 15 | 15 |
| CV (%) | 58 | 42 | 27 | 34 |
| SE | 3.7 | 1 420 | 5.0 | 3.7 |

## Phenology of flowering

For CLT and WLD, the distribution over time of the average number of spikes at the female stage followed as a whole a "bell curve," in contrast to the SHB for which it followed a "sawtooth curve" with no definite tendency (Fig. 2). The WLD began to flower at 47 DAS, with a maximum around 91 DAS (8 spikes/plant). Its flowering was not yet totally finished by the end of the experiment (144 DAS). The CLT began flowering at 62 DAS, with a maximum at 70 DAS (0.5 spikes/plant). Its flowering was finished at 84 DAS. The flowering of SHB began at 61 DAS. During the flowering period of CLT, its maximum was at 66 DAS (1 spike/plant); outside this period there was a peak at 99 DAS (1.5 spikes/plant), continuing up to 135 DAS. The "sawtooth" profile of the flowering curve of the SHB could be the consequence of the low number of observed plants ($n = 11$). The flowering period of the cultivated phenotype thus took place entirely within that of the wild phenotype and corresponded to only 25% of the flowering period of the wild phenotype.

The average time lapse estimated between the female and male stages of a spike was not significantly different ($P < 0.05$) between the wild (4.1 ± 0.31 days (mean ± SE), $n = 24$) and cultivated (3.6 ± 0.5 days, $n = 16$) phenotypes. Thus the variations of the pollen cloud for the wild and cultivated samples were assumed to follow the distribution of the average number of spikes at the female stage with a time lapse of 4 days.

## Production of seeds and proportion of viable seeds

The average total number of spikes per plant was significantly different among the three phenotypes ($P < 0.01$), and the average number of seeds per plant was significantly different between WLD and the others ($P < 0.01$) and between CLT and SHB ($P < 0.05$) (Table 1). A WLD plant produced on average 114 times more spikes than a CLT plant and 7 times more seeds. The seeds produced by the wild sample before and after the emission of the pollen cloud of CLT

(period extending from the first to the last cultivated spike at the male stage) represented 35% of its total production. A SHB plant produced on average 7.5 times more spikes than a CLT plant and 1.5 times more seeds per plant. Most of the seeds produced by SHB (91%) were produced during the flowering of CLT from 62 to 88 DAS.

Considering the entire seed production, 92% of the seeds produced by WLD germinated. This proportion was significantly higher ($P < 0.01$) than those of CLT and SHB, which were 80 and 71%, respectively, and not significantly different from one another ($P < 0.05$).

## Magnitude of gene flow according to the phenotypes and the date of fertilization

### *Gene flow from the wild sample (WLD) to the cultivated sample (CLT and SHB)*

The percentage of hybrids formed each day ranged between 35 and 66% for CLT and between 24 and 79% for SHB (Fig. 3), with an average value not significantly different between the CLT (50%) and the SHB (42%) ($P < 0.01$). The comparison of Fig. 2 (wild) and Fig. 3 (cultivated and shibra) suggests that the fertilization of the cultivated sample (CLT and SHB) was not related to the variations of female flowering of the wild sample and thus to those of its pollen cloud. There was no correlation between the variation in the percentage of hybrids in the progeny of CLT and SHB and the variation of pollen cloud emitted by WLD ($r = 0.30$, df = 20). The weighted average of the global percentage of viable hybrids formed during the entire growing season ($H_T$) was 45% for CLT and 39% for SHB.

### *Gene flow from the cultivated sample (CLT and SHB) to the wild sample (WLD)*

The percentage of hybrids formed in the WLD each day during the flowering of the cultivated millet varied between 0.5 and 29% with an average of 11% (Fig. 3). The fertilization of the wild sample appeared to be dependent on the variations in the intensity of the pollen cloud emitted by the cultivated sample ($r = 0.61$, df = 20, $P < 0.01$). The percentage of viable hybrids formed during the entire growing season was 8% as a result of the large proportion of seeds (35%) produced before and after the flowering of the CLT. It was not possible to differentiate between the relative contributions of the CLT and the SHB in the gene exchange with the WLD during the flowering of the CLT. However, the results obtained for the post-flowering period of the CLT show a low proportion of hybrids produced by fertilization of wild plants by the SHB (0.1%). The gene exchange by which the SHB participated thus appeared to be very residual. This is explained, at least in part, by the low proportion of seeds (9%), thus of flowers, produced by the SHB outside the flowering period of the CLT.

The gene flows between the wild and the cultivated pearl millet were very different: 45% of exchanges from the WLD to the CLT or 39% to the SHB, as opposed to 8% in the opposite direction. For the wild phenotype, in an average production of 51 738 progenies (viable seeds) per plant, 92% (viz. 47 599) were produced by crossing plants of the same sample. For the cultivated phenotype, in an average produc-

**Fig. 3.** Percentage of hybrids in the production of cultivated, wild, and shibra pearl millet seeds in relation to the day of fertilization. Error bars are SE.

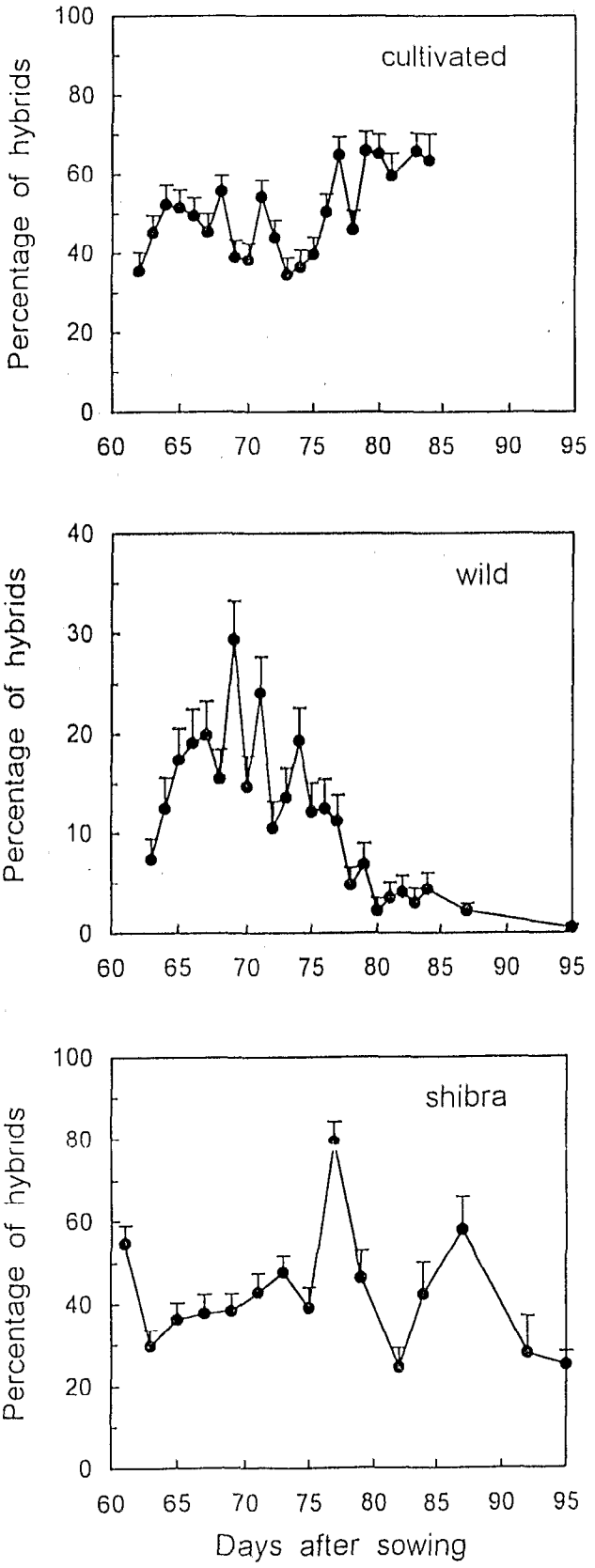
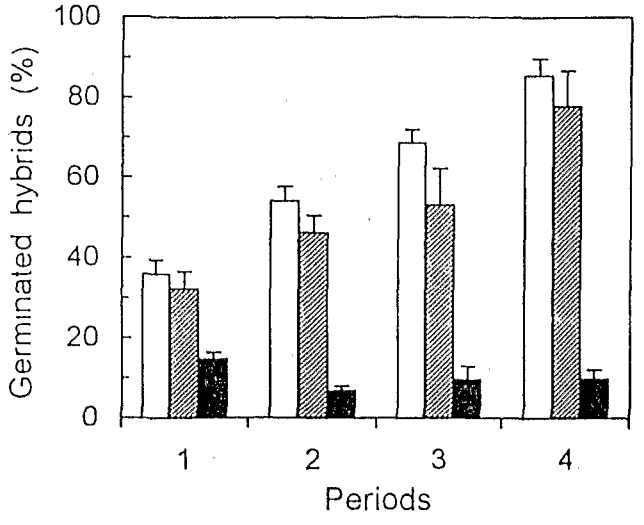


Fig. 4. Variation in the percentage of hybrids germinated between 0 and 48 h in the progeny of cultivated (open bars), shibra (hatched bars), and wild (solid bars) pearl millet according to four periods: 1 (0–8 h), 2 (8–16 h), 3 (16–24 h), and 4 (24–48 h). Error bars are SE.




tion of 6 297 progenies per plant, 55% (viz. 3 463) were so produced. Thus, in our experimental conditions, the probability of a wild plant engendering another plant of the same phenotype was 14 times higher than that for a plant of the cultivated phenotype.

### Viability of seeds and speed of germination according to their genotype

For the CLT, the proportion of viable seeds produced on a given day was inversely correlated to the percentage of hybrid seeds produced on this day ($r = -0.53$, df = 20, $P < 0.01$). This phenomenon was not observed with the seeds produced by the SHB and the WLD. The mean percentage of viable hybrids, considering all the germination dates, regularly increased from period 1 to period 4 by 36 to 85% for the CLT and by 32 to 78% for the SHB (Fig. 4). This phenomenon was not observed for the WLD. After 48 hours (period 5) the percentage of hybrids stayed high in the CLT and the SHB (82 and 67%, respectively), and low in the WLD (5%), but the number of seeds was too low to be significant.

## Discussion

In regions where cultivated pearl millet and its wild relative are sympatric, the frequency of intermediate phenotypes (shibra) in the fields varies between 5 and 30% (Rey-Herme 1982). In populations of wild pearl millet in Senegal, this frequency has been estimated at 31%; in Niger, 19% (Marchais and Tostain 1992). These estimations attest to gene flow in natural conditions but do not explain the intensity and asymmetry of gene flow between wild and cultivated pearl millet as measured in this study.

The flowering period of cultivated pearl millet took place entirely within that of wild pearl millet. By contrast the wild pearl millet is in temporal reproductive isolation during more than half of its reproduction period. These observations of the largely endogamic reproduction of wild pearl millet are corroborated by observations under natural conditions in Niger (Marchais 1994). The large number of spikes produced by one wild plant (319 compared with 3 per cultivated plant in this study) favours a staggered flowering. The proportion of hybrids formed on the cultivated phenotype does not appear to be time dependent, probably because the pollen cloud of the wild sample was never limiting to the fertilization of the cultivated one. On the contrary, the pollen cloud emitted at the beginning and at the end of flowering by the sample of cultivated plants was limiting to the fertilization of wild plants. When sympatric wild and cultivated populations are the same size, the preponderance of the wild pollen cloud over the cultivated one reinforces the endogamic reproduction in the wild population while diminishing the genetic influence of the cultivated field by competing with its pollen. The distribution of plants in the plot is evidently not the same as that observed in nature where plants of a same cultivated or wild phenotype are regrouped in fields or in spontaneous populations. Moreover, Burton (1974) has observed that the anemophile dispersion is not extensive. In one generation in natural conditions, the gene exchange between wild populations and cultivated fields would thus be less than under our experimental conditions.

The influence of prezygotic barriers on gene exchange was not perceptible in the context of this experiment, while the effect of postzygotic barriers was obvious. The loss of viability of the hybrid seeds produced by cultivated pearl millet plants has already been noted by Amoukou and Marchais (1993) in the case of controlled crossing with wild pearl millet. To this phenomenon is added the loss of viability of the hybrid seeds produced by the shibra plants and the slower germination of hybrid seeds produced by plants of the cultivated and shibra phenotypes. The more rapid germination of the cultivated phenotype could be a determinant in its selection by the traditional farmer, who eliminates the less vigourous plants at the time of thinning. In fact, the domesticated pearl millet is incapable of maintaining itself without Man, while the persistence of the wild populations is based only on the adjustment of the genome to the variations in natural selection pressures. Genetic exchange with wild pearl millet could favour the adaptability of the cultivated pearl millet to the ecological variations of the Sahelian environment (Pernes 1983).

Since the beginning of domestication a coadaptation has established between cultivated and wild pearl millet. The biotechnological risk in introducing genetically transformed plants in regions where they are able to exchange genes with wild relatives was raised by Rogers and Parkes (1995). Pearl millet belongs to the species likely to be improved by genetic transformation of its sexual reproduction system into an asexual system. Such a transformation could disturb the gene flow within *P. glaucum*. In an ideal situation presented by breeders (Savidan and Dujardin 1992), the farmer of an apomictic pearl millet, free of the vicissitudes of sexual reproduction, would have the guarantee of a harvest of plants that always conform to a chosen type. The genetic determinism of apomixy relies on few genes, and the pseudogamy described for most of the apomictic *Poaceae*, some *Pennisetum* species, implies the existence of fertile pollen (Asker and Jerling 1992). Even residually, sexuality is essential to

the maintenance of genetic diversity (Renno et al. 1995; Schmelzer and Renno 1997) and apomictic species evolve without the intervention of Man. Dumping apomictic pearl millet in Sahelian agrosystems could have serious consequences on the future of *P. glaucum*, the staple food of about 44 million people in this part of the world. At the risk of falling into an agronomic dead end, a real sustainable improvement of pearl millet should not be attempted without a thorough understanding of gene flow in relation to the mechanisms of evolution.

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