Theor Appl Genet (1985) 70: 542–547

Genetic variations in the hybrids of rice (*Oryza sativa*) and sorghum (*Sorghum vulgare*)

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Received August 30, 1984; Accepted February 18, 1985
Communicated by Hu Han

Summary. Some of results from morphological and cytological observations and esterase-isozyme studies of a rice-sorghum hybrid are presented in this paper. There is a great diversity of morphological characters and some special characteristics in the progenies of the hybrids of rice with sorghum. The meiosis of pollen mother cells in the early generations of the hybrid was found to be abnormal. One main band coinciding with one found in sorghum but lacking in rice appeared in the majority of the hybrid lines. This band is characteristic of a are the specificities of the distant hybridization of rice and sorghum, and is rarely observed in the intervariety hybrids or hybrids between subspecies of *Oryza sativa, indica* and *japonica*. On the basis of these facts we concluded that the hybrids obtained are true hybrids of rice and sorghum.

Key words: Rice – Sorghum – Distant hybridization – Morphological characteristics – PMCs – Esterase isozymes – Abnormals

Introduction

There are only a few references in the literature to distant hybridization using rice as a maternal.

Since the 1950s many farmer-technicians and researchers in China have used a dozen different kinds of plants from various genera, e.g. Sorghum, maize, wheat, wild rice, yeed, green foxtail, reed mace, panic grass, bamboo etc. as the paternal parent for crossing with rice. Some morphological, anatomical, cytological, and biochemical studies were carried out and interesting results were obtained (Wu Suhsuen et al. 1963, 1965; Hsia Yung-shen et al. 1964; Zu Deming et al. 1979; Zhou Guangyu et al. 1979, 1980; Chen Jingquan et al. 1980; Shao Qiquan et al. 1980; Tand Xihua et al. 1981; Wang Chengping et al. 1982; Zhu Fengsui et al. 1983; Shen Jianhua et al. 1983).

In 1960 we began to cross rice with sorghum and this hybridization has been repeated many times. In this paper we would like to introduce some of the results of morphological and cytological observations and esterase-isozyme studies in the 1st–5th generation of a rice-sorghum hybrid.

Materials and methods

Rice A7705, a stable line from progenies obtained from the combination of 5466 (rice) × 3197B (sorghum), was used as the maternal parent (5466 is another stable line of 'Yinfang' (rice) × 'Henjiali' (sorghum)). Beginning in 1978 stigmas were emasculated by treatment with hot water and then pollinated with fresh pollen of 'Henjiali' (sorghum). F_1 seeds were planted in 1979 and F_5 plants were obtained until 1983. A7705 was used as a control.

The hybrid progenies were planted by individually. Observations on growth characteristics and individual plant selection were carried out in the field. Meiosis of pollen mother cells in F_2 and F_3 was studied by making acetic acid eosin smear slides. The analysis of esterase isozymes from different tissues in the F_2–F_5 was made by polyacrylamide gel electrophoresis. Gels were stained with α- and β-naphthyl-acetate and fast blue RR.

Results

1 Morphological characteristics

Data on the morphological characteristics of the F_1–F_5 plants is shown in Table 1. The date of heading of the F_1 hybrid is later than that of maternal, and its sprouting is not uniform; it is strong and compact; the plant height is over 30 cm higher than that of its maternal and its grain is larger although fertility is lower. The percentage of seedsetting is only 16.69% and grains drop down easily (Fig. 1).

Table 1. Plant character segregation in F_1–F_5 hybrid plants compared to that of the maternal parent (1979–1983)

Maternal parent & the hybrid generation	Date of heading	Plant height (cm)	Length of spike (cm)	Awnness	No. of grains in main panicle	Percentage of seed set (%)	1,000-seed weight (g)	Other variations
♀	mid-Aug.	100		no	198	81.82		
F_1	mid Aug.–early-Sept.	134.1		Sort tip	202	16.69		Grains are larger than those of maternal plant
♀	Aug. 6	110	23.5	no	178	96.6	27.15	
F_2	July 26–the last ten days of Aug.	100–160	19.5–32.8	no-long, violet tip	64–352	30.7–98.5	24.5–37.5	The spikelets grow on the neck of normal panicle
♀	Aug. 16–18	90	21.7	no	128	96.9	28.67	
F_3	Aug. 2–Sept. 10	75–140	18.4–28.7	no-long, violet awn	105–289	2.5–98.1	17.67–42	Tillers grow on the node above ground, tiller without ear
♀	Aug. 17–20	110	22	no	192	97.0	28.5	
F_4	Aug. 10–Sept. 5	85–115	13.5–33.3	no-long, violent	41–346	0–99.0	20.1–54	Size of grains varies in different panicles of the same plant, double flag leaf
♀	Aug. 6–8	105	22.5	no	137	98.5	28.1	
F_5	July 27–Aug. 31	90–157	14–33.7	no-long, violent awn	48–416	23–100	19.0–58.0	

Fig. 1. *a* The maternal parent (A7705); *b* F_1 hybrid plant

F_2 characters segregate to a considerable degree. There are obvious differences among characters of individual plants; date of heading, plant height, main panicle length, number of seeds per panicle, 1,000-seed weight and the percentage of seedsetting. In most cases, the number of seeds per panicle is higher and 1,000-seed weight is heavier than that of the maternal. There are both awnless and with long awn among them. Some new characters appear which were absent in the maternal – violet gluncetip, well-developed bracteal leaves in some plants, etc. Furthermore, some abnormal developed organs are observed, such as spikelets grown on the neck of panicle, two panicles grown on the same node of the flag leaf (one larger, another smaller). Grains on the larger panicle are usually bigger than that of the smaller one. The smaller panicle is often tightly wrapped by the flag leaf sheath and can hardly sprout out (Fig. 2).

Segregation of characters in the F_3 occurs continuously. There were still some abnormally developed

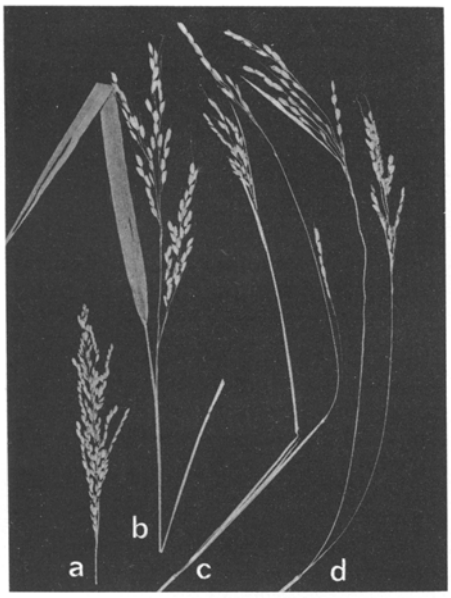

Fig. 2. *a* The panicle of A7705. *b–d* The panicles of the hybrid: *b* The spikelet grown on the neck of the normal panicle whose grains are not the same in size as those of the normal panicle; *c* The tiller grown on the node above-ground whose graine size varies markedly one of the panicles possesses well-developed bracteal leaves. *d* Two panicles grown on the same node of the flag leaf, of which, the left one possesses well-developed bracteal leaves and larger grains

Fig. 3. The tillers grown on the noder above-ground, one of them does not possess a panicle

organs. For instance, no sprouting was observed on the tiller at an early stage of development but it appeared on the secondery tillers. There were some branches on the node upground and ears bear on them (Fig. 3). A few red awn, violet-tipped glume and long-shaped

Fig. 4. *a* The panicle of A7705; *b* and *c* Two panicles from the same plant of the hybrid, showing the difference between the two panicles: *b* The panicle with smaller grains: *c* The panicle with larger grains

Fig. 5. Differently-shaped panicles with larger grains, most of them do not have main panicle axises

grains were also observed. The spike types show a great diversity and the range of 1,000-seed weight was increased and variation much wider.

Some new variations of characters were observed in the F_4 generation. The most prominent one was that the grain size of different spikes on the same plant varies. The average of 1,000-seed weight of larger-grain spikes is 45.5 g and the smaller one is 36.7 g (Fig. 4). Most of the spikes with larger grains do not have main panicle axes (Fig. 5) and are usually wrapped in the flag leaf sheath unable even to sprout out completely. Their seedsetting was normal. Another abnormal trait observed in the F_4 generation was the double flag

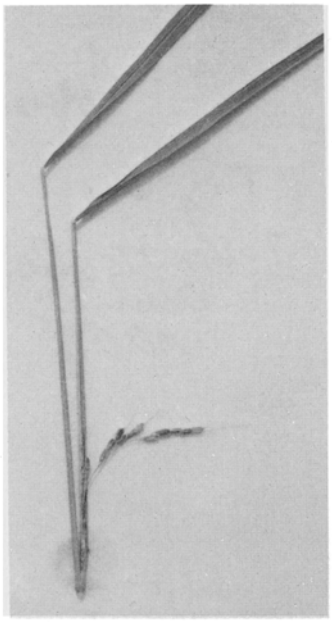

Fig. 6. Double flag leaves

Fig. 7. Two panicles grown on the same node of flag leaf and two leaf-blades bearing side by side on the same leaf sheath, the two leaf-blades possess their own main vein respectively

leaves (Fig. 6). Two leaf-blades bear side by side on the same leaf sheath, each with its own main vein (Fig. 7). Sometimes the number of flag leaf-blades may be 4 (Fig. 8). There are plants with double bracteal leaves and plants with smooth leaves and smooth glumes. Seeds being without embryos and the husks of sterile flowers being developed into glume can also be observed.

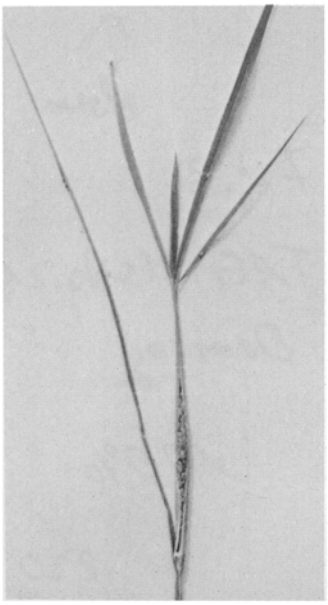

Fig. 8. Four leaf-blades bearing on the same leaf sheath, each with its own main vein

Variation and segregation of characters in the F_5 generation observed in 1983 were even much wider than those of former generations. The abnormal traits found in the previous four generations still appeared at even higher frequencies among the F_5 population, even though this population consisted of a lot of different lines. These abnormal traits can therefore be inherited.

2 Cytological observations

A The abnormal traits seen during the meiosis of the pollen mother cells in the F_2 are as follows:

1) The number of satellite chromosomes was usually two pairs. This is one pair more than the maternal parent and shows the dominance of the paternal parent.

2) The number of bivalents at diakinesis varied. In most cases it was 13, but 12, 14 or 15 were also observed. The size of the chromosomes was also unequal. The chromosome segments and chromatin granule also appeared in varying numbers and different sizes and shapes.

3) Some chromosomes lagged behind in metaphase I and anaphase I.

4) The meiosis of the pollen mother cells in the same single spikelet was out of synchronism: pachynema, diakinesis, metaphase, anaphase, first division, second division or even tetraspores may be observed at the same time.

B More complicated abnormal traits were found during meiosis of the pollen mother cells in the F_3

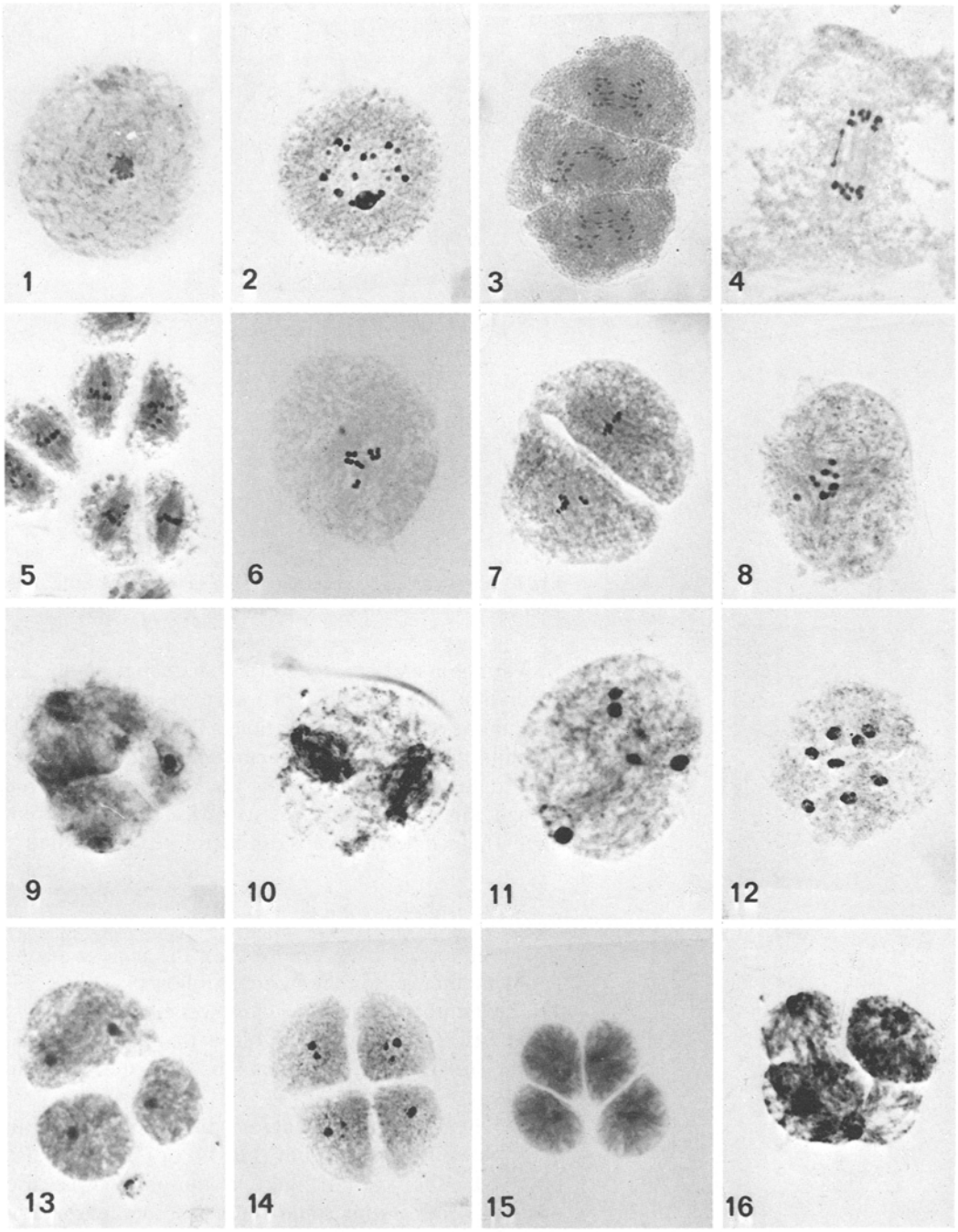

Fig. 9. *1* The PMC with many chromosomes attaching to the nucleolus at diakinesis; *2* The cell having over 12II, SAT=2; *3* Lagging chromosome in AI; *4* Chromosome bridge in AI; *5* The late conducing chromosome in MII, unsynchronized division, multipolar spindle; *6* Secondary association formed with bivalents in MI; *7* Grouping bivalents in MII; *8* The tripolar spindle; *9* The tritet; *10* Vertical two spindle in a PMC; *11* Unequal division; *12* The cell with 8 nuclei; *13* Unsynchronized tetrad; *14* Quartet with micronuclei; *15* Abnormal quartet arrangement; *16* Uneaqual size of the quartet

generation. In addition to those appearing in the F_2, the following abnormal features were observed (Fig. 9):

1) The number of satellite chromosomes varied from 1–4 pairs in different cells of the same anther. The majority of the chromosomes were connected to the nucleoli.

2) Lagging chromosomes and chromatin bridges were found in metaphase I, II and anaphase I, II.

3) Chromosome grouping in metaphase I and II and bivalent secondary association were observed.

4) Double spindles and vertically arranged spindles were also found.

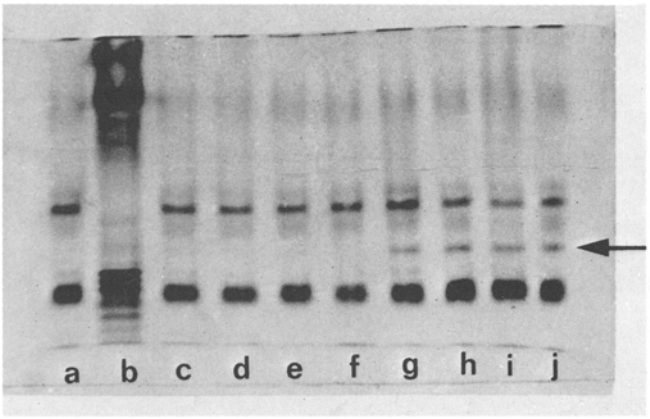

Fig. 10. Esterase isoenzyme patterns. *Arrow:* enzyme bands (band II) of sorghum and hybrid. *a* The maternal parent (A7705), without band II, *b* Sorghum ('Hengjiali'), with band II; *c–j* some lines from A7705 × 'Hengjiali': *c d e* and *f* without band II; *g h i* and *j* with band II

5) In anaphase abnormal states such as tripolarity, multipolarity, unequal division and multi-nucleus cells were appeared.
6) Microspores with 1–3 nuclei, asynchronized formation of tetrads and varying sizes of tetrads were observed.
7) Microspores without viability and sterile pollen were formed.

3 Analysis of esterase isozymes

The basic pattern of the zymograms of the hybrids was similar to that obtained from the maternal parent. The essential difference consisted of one main band (band II) coinciding with the sorghum but lacking in rice, which appeared in the majority of the hybrid lines (Fig. 10). The main genetic behavior of this band in the F_2–F_5 generations is as follows:
1) In the F_2–F_5 generations, the percentage of plants with band II was 81.1%, 65.2%, 68.0% and 55.5%, respectively. This means that the zymogram patterns of hybrids also segregated. The percentage of plants having band II in the hybrid population decreased gradually respective to the generation.
2) Segregation mainly occurred in those lines or plants which had band II. In progeny populations, the lines with band II comprised the majority. Segregation was rarely observed in the lines or plants lacking band II. For the few plants with band II in the F_2, this band was absent in their F_3 but reappeared in their F_4.

Discussion

1 A great diversity of morphological characters and some special characteristics present in the progenies of the hybrids of rice with sorghum, as mentioned above, are rarely observed in the intervarietal hybrids or hybrids between subspecies of *Oryza sativa, indica* and *japonica.* Such diversity did not occur by chance – it was seen repeatedly in other combinations. On the basis of our longtime observations, we believe that the great diversity of the morphological characteristics in the hybrid progenies is a specific of the distant hybridization of rice and sorghum.

2 Exclusive also to the hybrid progenies of rice and sorghum was the behavior at meiosis. Abnormal features appearing in the meiosis of pollen mother cells seldom happened in the intervariety hybrids or hybrids between *Oryza sativa indica* and *japonica.* There is very little information to be found in the literature.

Based on these two specificities and considering the additional fact that enzyme band II of sorghum appeared in the majority of the hybrid plant lines we concluded that hybrids obtained in the experiment described above are true hybrids of rice and sorghum.

References

Wu Suhsuen, Wang Jingju (1963) Cytological studies on the intergeneric F_1 hybrid between *Oryza sativa* L. × *Pennisetum* sp. Acta Bot Sin 11:293–307
Wu SH, Wang Jingju, Sun Jingsan, Sai Qigui (1965) On the fertilization of *Oryza sativa* L. × *Sorghum valgare* pers. Acta Bot Sin 13:297–313
Hsia Yungshen, Kao Yucheng, Chiang Yucheng (1964) A preliminary report on the intergeneric crosses between paddy rice and gu. Genet Sin 5:41–58
Zu deming, Dai Lanfang, Chen Shanbao, Song Xianbin, Duan Xiaolan (1979) Diversity and specific performance of progenies from distant hybridization between rice and sorghum. Acta Genet Sin 6:414–420
Zhou Guangyu, Gong Znenznen, Wang Zifen (1979) The molecular basis of remote hybridization – An evidence of the hypothesis that DNA segments of distantly related plants may be hybridized. Acta Genet Sin 6:405–413
Zhou Guangyu, Zeng Yishen, Yang Wanxia (1980) Molecular basis of remote hybridization – The recombination of sorghum DNA sequences with rice genomes during remote hybridization. Acta Genet Sin 7:119–122
Chen Jingquan, Zhang Yaolong (1980) The breeding and utilization of naked seed rice. Acta Genet Sin 7:185–188
Shao Qiquan, Wu Chunfa, Jiang Xingcun (1980) Cytological study on naked seed rice. Acta Genet Sin 7:366–369
Tang Xihua, Zhu Zhiping, Huang Qingliu (1981) Some aspects of morphological and physiological characters of "naked-seed" rice. Acta Genet Sin 8:350–355
Wang Chengping, Tang Xihua (1982) A new hybrid genus of Graminae (Poaceae). Acta Phytotax Sin 20:179–181
Zhu Fengsui, Fu Junhua, Li Liancheng, Chen Shanbao (1983) A cytological study on the intergeneric hybrid between rice and sorghum. Sci Agric Sin 4:26–29
Shen Jianhua, Zhou Guangyu (1983) The reassociation kinetic analyses of the repeated sequence DNA of rice, sorghum and their hybrid. Acta Genet Sin 10:28–35