イネ属植物の細胞遺伝学的研究 (6)

誌名	The Japanese journal of genetics
ISSN	0021504X
巻/号	483
掲載ページ	p. 159-165
発行年月	1973年6月

JAPAN. J. GENETICS Vol. 48, No. 3: 159-165 (1973)

CYTOGENETICAL STUDIES ON THE GENUS *ORYZA* VI. CHROMOSOME PAIRING IN THE INTERSPECIFIC HYBRIDS BETWEEN O. *OFFICINALIS* AND ITS RELATED DIPLOID SPECIES

TSUGUFUMI OGAWA AND TAIRA KATAYAMA

Faculty of Agriculture, Kyushu University, Fukuoka 812

Received January 9, 1973

There are three species in diploid *O. officinalis* complex, that is, *O. officinalis* in Asia and *O. punctata* and *O. eichingeri* in Africa. Sharma *et al.* (1965) raised Ceylonese *officinalis* strain to specific level as *O. collina* (Trimen) Sharma et Shastry comb. nov., while Tateoka (1962) listed it up as a synonym of *O. eichingeri*. Furthermore, Tateoka (1965a, b) found clones showing intermediate features of *O. eichingeri* and diploid *O. punctata*, and described them as Intermediate of the both species.

Based on the morphological standpoint, Sampath (1962) assumed that B genome is present in Ceylonese *officinalis* and that diploid *O. punctata* has C genome of *O. officinalis*, but Gopalakrishnam (1966) enumerated Ceylonese *officinalis* has C genome of *O. officinalis*. From the observation of F_1 hybrid between diploid *O. punctata* and its related species, Katayama (1967) postulated that the genome of the diploid strain of *O. punctata* should be safely fixed as BB, while Hu (1970) showed that the genome of *O. eichingeri* (2n) is homologous with that of *O. officinalis*, and considered that the genome of diploid *O. punctata* can not be definitely postulated as A, B or C.

From the taxonomical and cytogenetical standpoints, it is important and interesting to make clear the relationship among the diploid species and strains in the *O. officinalis* complex. The present paper deals with the cytogenetical results obtained from the F_1 hybrids among the diploid *officinalis* complex.

MATERIALS AND METHODS

Twenty-five hybrids between strains of *O. officinalis* and its related species or strains (Intermediate, Ceylonese *officinalis* and *O. eichingeri*) were employed as materials (Table 1). All of the species and strains used in the present experiment were kindly furnished by the National Institute of Genetics at Misima except one strain (*O. collina*). Crosses and culture of the F_1 seeds were made by using the methods described already by Katayama (1966). After fixation of the PMCs with acetic alcohol (1: 3) for 24 hours or longer at low temperature of about 5°−8°C, the fixation solution was substituted by 70 percent ethyl alcohol. Preparations were made by usual acetocarmine squash method.

Table 1. Cross-combinations between *O. officinalis* and its related species used in this experiment

F_1 plant no.	Cross-combinations
NF_1 171	*O. officinalis* (W1291) × Intermediate (W1525)
175, 178	〃 (W1251) × 〃 〃
179	〃 (W0065) × 〃 〃
210	〃 (W1281) × 〃 〃
292	〃 (W1282) × 〃 〃
293	〃 (W0002) × 〃 〃
23	〃 (W1269) × 〃 (W1527)
24	〃 (W1302) × 〃 〃
37	〃 (W1203) × 〃 〃
38	〃 (W1282) × 〃 〃
86	〃 (W1278) × 〃 〃
87, 169	〃 (W1267) × 〃 〃
90	〃 (W1304) × 〃 〃
172	〃 (W1291) × 〃 〃
174	〃 (W1249) × 〃 〃
295	〃 (W0065) × 〃 〃
297	〃 (W1267) × *O. eichingeri* (W1522)
298	〃 (W1282) × 〃 〃
299	〃 (W0065) × 〃 〃
300	〃 (W1278) × 〃 〃
302	〃 (W1282) × Ceylonese *officinalis* (*collina*)
303	〃 (W1282) × 〃 〃 (W0006)
304	〃 (W0002) × 〃 〃 〃

RESULTS

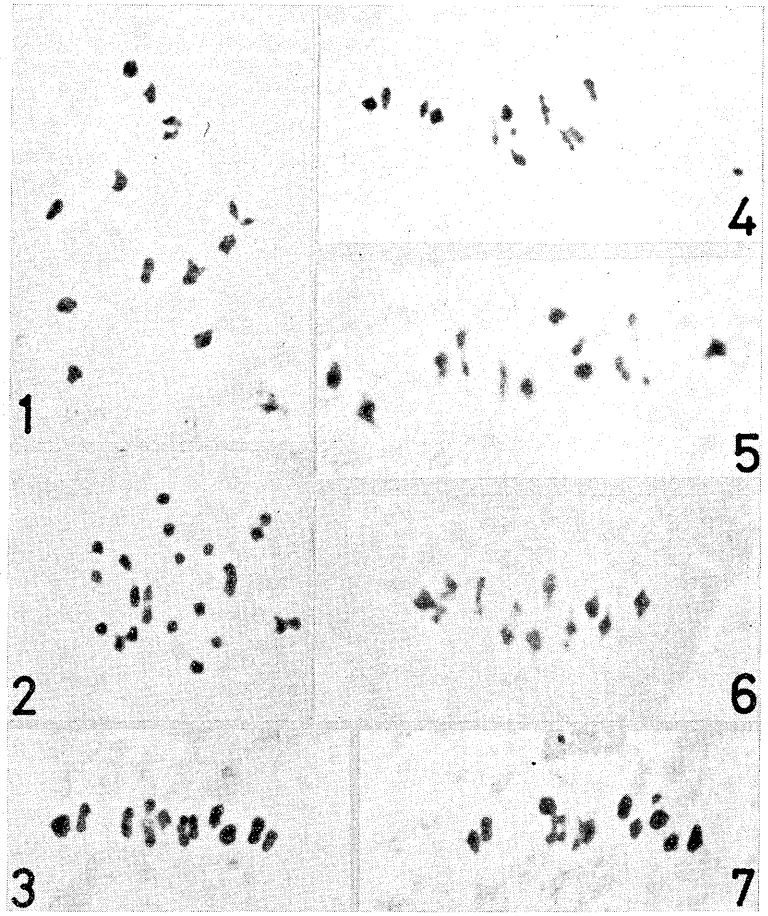
1. *O. officinalis*×Intermediate

In 1969, 1970 and 1971, from 360 spikelets crossed in 16 cross-combinations, 59 seeds were obtained. F_1 hybrid plants presented intermediate morphology of both parents. Pollen was completely sterile except for some F_1 hybrids having a few fertile pollen grains (NF_1 24, 38, 178, 293 and 295). As shown in Table 3, pollen fertility varied from 0 to 2.4%. Pollen showed nearly normal characteristics, though its size was variable.

Chromosome pairing in the F_1 hybrids was almost complete as shown in Table 2. At diakinesis and MI of some F_1 hybrids (NF_1 293, 86, 90 for the former: NF_1 210, 174, 295 for the latter), a few univalents and tetravalents were observed, but the mean of chromosome pairing was 12 or slightly less than 12 (Figs. 1-4). Some irregularities of chromosome behaviors were observed (Fig. 7).

2. *O. officinalis*×*O. eichingeri*

In 1971, of 25 seeds obtained from 100 spikelets crossed in four cross-combinations,



Figs. 1-7. Diakinesis and MI stages of PMCs of the F_1 hybrids between *O. officinalis* and its related species or strains.
1. Diakinesis stage, showing 12II (NF_1 86). 2. MI, showing 3II+18I (NF_1 38). 3. MI, showing 12II (NF_1 86). 4. MI, showing 11II+2I (NF_1 210). 5. MI, showing 12II (NF_1 297). 6. MI, showing 12II (NF_1 303). 7. MI, showing 12II +1f (NF_1 86).

21 F_1 hybrids raised and 3 F_1 hybrids of different cross-combination were observed cytologically (Table 2).

Their morphologies showed intermediate features of both parents. Their pollens were completely sterile (Table 3), and showed irregularities in shape as compared with that of F_1 hybrids from the other cross-combinations, *O. officinalis* × Intermediate and *O. officinalis* × Ceylonese *officinalis*.

At diakinesis and MI of the F_1 hybrids, number of bivalents ranged from 10 to 12 except for MI of NF_1 297, and showed almost 12 in mean (Table 2 and Fig. 5). The most bivalents observed showed compactly pairing shape, such as diamond-, rod- or dumbbell-shape (Table 3).

Table 2. Chromosome pairing at diakinesis and MI in the F_1 hybrids between *O. officinalis* and its related species

F_1 plant no.	diakinesis					MI				
	No. cells observed	IV		II		No. cells observed	IV		II	
		range	mean	range	mean		range	mean	range	mean
NF_1 171						34			11-12	11.9
210						48	0 - 1	0.02	10-12	11.9
292	15			11-12	11.8	13			12	12.0
293	42	0 - 1	0.05	10-12	11.9	33			11-12	12.0
23						10			12	12.0
24	14			12	12.0	50			12	12.0
37	9			12	12.0	52			3-12	11.8
38						32			2-12	10.7
86	30	0 - 1	0.07	10-12	11.9	72			11-12	12.0
87						23			6-12	11.7
90	92	0 - 1	0.05	10-12	11.8	28			12	12.0
169						52			11-12	11.9
172	21			11-12	12.0	50			11-12	12.0
174						55	0 - 1	0.02	10-12	12.0
295						46	0 - 1	0.04	10-12	11.6
297	29	0 - 1	0.03	10-12	11.8	50			5-12	11.9
298						52			11-12	12.0
300						15			12	12.0
302						21			11-12	11.9
303						31			10-12	11.9

3. *O. officinalis* × Ceylonese *officinalis*

In 1971, 16 seeds were obtained from 187 cross-pollinations with 2 Ceylonese *officinalis* strains as paternal parent. Pollen grain of NF_1 303 and 304 was almost normal in shape but that of NF_1 302 was scarcely formed.

At MI of PMCs of both F_1 hybrids, chromosome pairing was almost complete (Table 2 and Fig. 6), and the bivalents were also of compact type (Table 3).

Contrary to those crosses, no true F_1 hybrid could be obtained from the interspecific cross-combinations between *O. officinalis* and diploid *O. punctata*, though more than 2,000 spikelets were crossed.

DISCUSSION

The genome of *O. officinalis* was firstly designated as C by Morinaga and Kuriyama (1959) from cytogenetical studies. Furthermore, they (1960) observed 12 bivalents at MI of the F_1 hybrid between Ceylonese *officinalis* and *O. officinalis*, though there were the recognizable differences in those outward appearance and raised the sterility in its F_1 hybrid. Similar result on the Ceylonese *officinalis* was described from taxonomic status by Gopalakrishnam (1966). Tateoka (1962, 1963) thought from the morphological

Table 3. Frequency of loose-pairing bivalents at MI and pollen fertility of F_1 hybrids

F_1 plant no.	No. of cells observed	No. loose-pair. IIs/cell		No. of pollen		% pollen fertility
		range	mean	observed	fertile	
NF_1 171	24	0-5	1.8	553	0	0
175				465	0	0
178				559	3	0.5
179				546	0	0
210	40	0-2	0.6	496	0	0
292	13	0-5	2.2	307	0	0
293	31	0-7	2.2	824	20	2.4
23	7	0-2	0.9	505	0	0
24	50	0-4	1.0	1001	19	1.9
37	49	0-4	1.5	519	0	0
38	31	0-8	1.9	515	2	0.4
86	71	0-6	1.9			
87	20	0-5	2.1	515	0	0
90	26	0-6	1.4	515	0	0
169	34	0-3	1.1	509	0	0
172	41	0-5	1.0	531	0	0
174	30	0-3	0.8	527	0	0
295	44	0-9	2.1	958	6	0.6
297	49	0-4	0.9	520	0	0
298	51	0-3	0.5	496	0	0
299				111	0	0
300	15	0-3	0.6	539	0	0
302	20	0-3	1.2			
303	30	0-4	1.3	995	5	0.5
304				648	33	5.1

feature that the Ceylonese *officinalis* is very similar to *O. eichingeri*. Sharma and Shastry (1965) also considered the Ceylonese *officinalis* to be a distinct species from the morphological observation, and named it as *O. collina* (Trimen) Sharma et Shastry comb. nov. They proposed that *O. collina* is a diminutive form of *O. eichingeri* because it bears little resemblance with *O. officinalis*. In the present experiments, chromosome pairing at MI of PMCs in the F_1 hybrid between *O. officinalis* and Ceylonese *officinalis* was complete (Table 2, Fig. 6). This fact agrees with the results obtained by Morinaga and Kuriyama (1959, 1960) and Gopalakrishnam (1966), in which Ceylonese *officinalis* has similar genome with *O. officinalis* (CC), but differs from the assumption of Sampath (1962), in which Ceylonese *officinalis* may have B genome based on the morphological standpoint.

Judging from the results obtained cytogenetical observations at diakinesis and MI of PMCs in the F_1 hybrids of the cross, *O. officicinalis* × *O. eichingeri*, it is clear that the genome constitution of the both species is homologous (Table 2, Fig. 5), though

the pollen and the seed of the F_1 hybrids are completely sterile (Table 3). Based on the observations of chromosome number in somatic cells of the parents and of PMCs in the F_1 hybrids of the crosses, W 002 (*officinalis*, CC)×101424 (*eichingeri*, 2X), 101425×W 002, CR-2 (*latifolia*, CCDD)×101424, W 1524 (*eichingeri*, 4X)×W 0045 (*minuta*, BBCC), Hu (1970) proposed that there are 2 strains in *O. eichingeri*, diploid *O. eichingeri* with the C genome and tetraploid *O. eichingeri* with the BC genomes. The morphological characters of W 1524 employed by Hu, however, differ from the original plant (2n) which recognized firstly as *O. eichingeri* by Tateoka (1965b) (Tateoka, private communications; Watanabe and Sampath, 1972; Katayama and Ogawa, 1973). It may result from contamination of seeds. From the reason, no tetraploid strain in *O. eichingeri* exists in natural. But the result obtained by Hu (1970) on the genome constitution of *O. eichingeri* (101424, 2n) is in accordance with that observed in the present experiment.

So far, no cytogenetical investigation of the relationship between the Intermediate and its related species was reported. Based on the chromosome configulations at diakinesis and MI of PMCs in the F_1 hybrids between *O. officinalis* and Intermediate, the authors considered that both of them have similar genome.

In the present experiment, at diakinesis and MI of PMCs in the F_1 hybrids between *O. officinalis* and its related species and/or strains, 12 bivalents with almost compact pairing were observed. However, reciprocal crosses between diploid *O. punctata* and *O. officinalis* could not get success in spite of a fact that more than two thousand spikelets were cross-pollinated. In the same cross-combination, no successful result was obtained by two previous workers (Katayama, 1967; Hu, 1970).

From the results mentioned above, the authors conclude that the genome of Intermediate, *O. eichingeri* and the Ceylonese *officinalis* is homologous with that of *O. officinalis*, and that there are distinct reproductive barriers between these species or strains and diploid *O. punctata*. The present results are well in accord with those of zymographic studies obtained from matured leaf of those species and strains (Katayama and Chern, unpublished).

SUMMARY

In order to elucidate the relationship among diploid species in the *O. officinalis* complex, 25 F_1 hybrids between *O. officinalis* and its related species of strains (Intermediate, Ceylonese *officinalis* and *O. eichingeri*) were studied cytologically.

Pollen was almost completely sterile in these hybrids, but occasionally some fertile pollens were produced. At diakinesis and MI of PMCs in all the F_1 hybrids, 12 compactly paired bivalents were observed in most cases. On the other hand, crosses between *O. officinalis* and diploid *O. punctata* were unsuccessful, though more than two thousand spikelets were cross-pollinated.

Based on these results, the authors postulated that the genome of Intermediate, *O. eichingeri* and the Ceylonese *officinalis* is homologous with that of *O. officinalis* and that there may be distinct reproductive barriers between diploid *O. punctata* and its related species or strains.

ACKNOWLEDGMENTS

We wish to express our sincere thanks to Drs. H. I. Oka and H. Morishima for supplying the materials.

LITERATURE CITED

Gopalakrishnam, R., 1966 Taxonomic status of *Oryza collina* (Trimen) Sharma *et* Shastry. Ind. J. Genet. Plant Breed. **26**: 98-100.

Hu, C. H., 1970 Cytogenetic studies of *Oryza officinalis* complex. III. The genomic constitution of *O. punctata* and *O. eichingeri*. Cytologia **35**: 304-318.

Katayama, T., 1966 Cytogenetical studies on the genus *Oryza*. 2. Chromosome pairing in the interspecific hybrid with the ABC genomes. Japan. J. Genetics **41**: 309-316.

Katayama. T., 1967 Cytogenetical studies on *Oryza*. — F_1 hybrids of the crosses BBCC×CC, BBCC ×a diploid strain of *O. punctata* and CC×a diploid strain of *O. punctata*. Proc. Japan Acad. **43**: 327-331.

Katayama, T., and T. Ogawa, 1973 Studies on diploid *O. punctata* and its related species. Japan. J. Breed. 23, Supp. **1**: 315-316 (Abst. in Japan.).

Morinaga, T., 1964 Cytogenetical investigations on *Oryza* species. Symp. Rice Genet. Cytogenet., Int. Rice Res. Inst. (1963): 91-104.

Morinaga, T., and T. Kuriyama, 1959 Genomic constitution of *Oryza officinalis*. Japan. J. Breed. **9**: 259 (Abst. in Japan.).

Morinaga, T., and T. Kuriyama, 1960 Interspecific hybrids in the genus *Oryza* and its genomic constitutions. Agr. Hort. **35**: 935-938, 1091-1094, 1245-1247 (in Japan.).

Richharia, R. H., and C. V. Seshu, 1961 Cytogenetics of an interspecific hybrid in *Oryza*. Curr. Sci. **30**: 189-190.

Sampath, S., 1962 The genus *Oryza*: its taxonomy and species interrelationships. Oryza **1**: 1-29.

Sampath, S., 1966 The genus *Oryza* an evolutionary perspective. Oryza **3**: 30-34.

Sharma, S. D., and S. V. S. Shastry, 1965 Taxonomic studies in genus *Oryza* L. IV. The Ceylonese *Oryza* spp. affin. *O. officinalis* WALL. ex WATT. Ind. J. Genet. Plant Breed. **25**: 168-171.

Tateoka, T., 1962 Taxonomic studies of *Oryza*. I. *O. latifolia* complex. Bot. Mag. Tokyo **75**: 418-427.

Tateoka, T., 1963 Taxonomic studies of *Oryza*. III. Key to the species and their enumeration. Bot. Mag. Tokyo **76**: 165-173.

Tateoka, T., 1965a A Taxonomic study of *Oryza eichingeri* and *O. punctata*. Bot. Mag. Tokyo **78**: 156-163.

Tateoka, T., 1965b Taxonomy and chromosome numbers of African representatives of the *Oryza officinalis* complex. Bot. Mag. Tokyo **78**: 198-201.

Watanabe, Y., and S. Sampath., 1972. Cytotaxonomic considerations on African tetraploid species, *Oryza punctata* Kotschy ex Steud. Japan. J. Breed. 22, Supp. **2**: 220-221 (Abst. in Japan.).