Botanical Studies in the Genus *Oryza*

III. Embryo Transplantation

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Culture of *Gramineae* embryos have been studied and growth patterns and nutrient requirements were investigated in consideration of embryonal developmental phases and physico-chemical environment necessary for embryo growth and metabolic processes. Several cereals (1, 9, 12) have been used. Embryo culture in interspecific hybrids was successful in *Oryza* according to NAKAJIMA and MORISHIMA (10). Embryo culture of intergeneric hybrids in cereals was studied by ROMMEL (12). The effect of reciprocal grafting of embryo and endosperm development in crosses between *Corchorus* species was reported by SACHAR *et al.* (13). The technique of transplantation of immature embryos to their own and alien endosperms using several kinds of cereals was reported by KIKUCHI (7).

The aim of the present paper was to study the developmental patterns in grafting experiments, during from them clues to the evolution of *Oryza* species. Research of relationships between embryo and endosperm may help to detect affinities between those species. Preliminary reports had been published (3, 4, 5).

Materials and Method

In the present study two experiments with *Oryza* were carried out. In Experiment I, including two cultivated and four wild species, nine strains, belonging to six species having A genome, were used. In addition, other species some with other than A genome were used to study the phylogenetic differentiation of the genome. In this experiment (Experiment II) twenty-two strains belonging to fifteen species with genome constitution AA, CC, BBCC, CCDD, EE, FF, and others with unknown genomes were used, including two cultivated and twenty wild species. Most of the strains were collected by the members of the National Institute of Genetics and some were obtained from foreign workers.

Enumeration of the species, their distribution and chromosome numbers were given in Table 1 of the previous paper (6). In all experiments, the seeds were used at the time when the dormancy was terminated.

Seeds were all husked before the experiment in order to get uniform germination, soaked in water at 25°C for 3 to 5 hours, being changed to room temperature; they were then divided by a sharp knife into embryo and endosperm, sterilized in 70% ethyl alcohol for 5 minutes, sterilized in 10% chlorinated lime for 30 minutes and washed in sterilized water twice.

The respective embryo and endosperm were joined to each other by gelatine of a suitable concentration, which was changed by room temperature. The average optimum gelatine concentration is assumed to be 1.6%. The materials were placed on 10 cc of aseptic agar medium in 18 cm long glass test-tubes, which contained 0.5% agar; no other nutrition was added to avoid its being absorbed by developing roots. About one-fifth of the endosperm was buried diagonally in the agar medium, but no part of embryo was buried in it.

The embryos were directed horizontally upward and the endosperm downward (= attached to the medium) in all experiments. They were kept under continuous dark condition at 25°C to prevent photosynthesis. Observations were made during 30 to 60 days after incubation, unless otherwise stated. Ten to twenty seeds were used in each experimental plot, and each plot was replicated three times.

Four kinds of experimental materials were used, namely, intact seeds, detached embryos, embryos transplanted to their own endosperm (= homogeneous transplantation), and embryos transplanted to an alien endosperm (= heterogeneous transplantation). In the present paper, the following abbreviations are used;

1W ---------------- Whole grain of Code No.1 strain (intact)
1M ---------------- Embryo of Code No.1 strain (detached)
1D ---------------- Endosperm of Code No.1 strain (detached)
1M·1D ------------- Grafted embryo of Code No.1 strain on endosperm of Code No.1 strain (= homogeneous transplantation)
1M·2D ------------- Grafted embryo of Code No.1 strain on endosperm of Code No.2 strain (= heterogeneous transplantation)

Results and Discussion

Experiment I

In this experiment, at first, the technical possibility of embryo transplantation was explored and when found feasible the compatibility between embryo and endosperm was observed. Nine strains belonging to six *Oryza* species, including two cultivated and four wild species, were used in Experiment I. All of them have the A genome. In *O. sativa*, two subspecies, namely, *japonica* and *indica*, and two strains of each subspecies, were used.

Code number, species name, strain number and the origin of materials used are given in Table 1. Experimental plots of Experiment I are shown in Table 2, in which M and D mean embryo and endosperm, respectively.

Experimental results one month later in the case of intact seeds and detached embryo are given in Table 3. In the following three tables, Tables 3, 4 and 5, to the plumule, coleoptile, first foliage leaf, second foliage leaf, third foliage leaf and total length were recorded. As to the radicle, developed or not, number of roots and length of the longest root were recorded. In all data given in the tables, a figure indicates the average value.

Development of coleoptile, first leaf, second leaf and third leaf in the plumule and one to four roots in the radicle was observed in the seedlings from intact seeds (Abbr. W). This result indicated that coleoptile, first leaf, second leaf and third leaf have the capacity to develop from the plumule of cultivated as well as wild rice species nursed by their own endosperm without

Table 1. Materials used in Experiment I.

Code No.	Species	Strain	Origin
1	*O. sativa* (*japonica*)	Kyoto Asahi	Japan
2	〃 〃	Kinoshita Mochi	〃
3	〃 (*indica*)	108	Formosa
4	〃 〃	230	Philippines
5	*O. sativa* var. *spontanea*	W0106	India
6	*O. perennis*	W0120	〃
7	*O. barthii*	W1045	Africa
8	*O. glaberrima*	W0026	〃
9	*O. breviligulata*	W0831	〃

Table 2. Embryo transplantations made in Experiment I.

Endosperm / Embryo	Code No. 1	Code No. 2	Code No. 3	Code No. 4	Code No. 5	Code No. 6	Code No. 7	Code No. 8	Code No. 9	Intact seed	Detached embryo
Code No.1	◎	○	○	○	○	○	○	○	○	□	△
Code No.2	○		○							□	△
Code No.3	○	○		○	○	○	○	○	○	□	△
Code No.4	○		○							□	△
Code No.5	○		○							□	△
Code No.6	○		○							□	△
Code No.7	○		○							□	△
Code No.8	○		○							□	△
Code No.9	○		○							□	△

□, Intact seed; △, Detached embryo; ◎, Homogeneous transplantation;
○, Heterogeneous transplantation.

any external supply of nutrient. It was ascertained that endosperm became completely empty when the third leaf was fully developed.

In growth behaviour of the plumule, no obvious difference was recognized in all the strains used. They grew more than 100 mm in length. According to the previous observations, *japonica* type strains of *O. sativa*, grew faster than those of other species. However, no clear difference was observed one month after they were placed on the medium. A conspicuous growth of a very long seminal root of *O. barthii* was observed but the growth of the roots of *O. breviligulata* became slower than those of other species. "Kyoto Asahi", a *japonica* type of *O. sativa*, developed four vigorous roots. Roots of more than 100 mm in length were found in most cases.

Table 3. Growth of plumule and radicle in the case of intact seed and detached embryo in Experiment I.

Material	Plumule					Radicle		
	Coleoptile	1st[1)]	2nd	3rd	Length	Development	Number	Length
1W	+[2)]	+	+	+	116^{mm}	+	4	119^{mm}
2W	+	+	+	+	110	+	3	141
3W	+	+	+	+	126	+	3	157
4W	+	+	+	+	108	+	2	107
5W	+	+	+	+	121	+	2	118
6W	+	+	+	+	120	+	3	119
7W	+	+	+	+	109	+	1	184
8W	+	+	+	+	143	+	2	106
9W	+	+	+	+	138	+	3	82
1M	+	+	±[4)]	—	6	+	1	9
2M	+	—[3)]	—	—	1	—	0	0
3M	+	—	—	—	1	—	0	0
4M	+	—	—	—	1	—	0	0
5M	+	—	—	—	1	—	0	0
6M	+	—	—	—	1	—	0	0
7M	+	—	—	—	2	—	0	0
8M	+	—	—	—	2	—	0	0
9M	+	—	—	—	1	—	0	0

1) 1st, 2nd and 3rd foliage leaves from the base, 2) developed,
3) undeveloped, 4) sometimes developed and sometimes undeveloped.

Detached embryo (Abbr. M) without endosperm produced only a coleoptile with the exception in "Kyoto Asahi", a *japonica* type of *O. sativa*. Generally only a short coleoptile can develop in germination rice seed nourished by the embryo itself. In "Kyoto Asahi", the first leaf and sometimes the second leaf developed. In most species coleoptile developed less than 2 mm in length. Detached embryo did not produce roots except "Kyoto Asahi". In this strain the seminal root appeared in all seeds and developed 9 mm on the average. It is a specific characteristic of this strain.

The results mentioned above would may be considered as fundamental characteristics of growth pattern in intact seed and detached embryo. Anatomical studies of embryo in the resting stage would be requested to solve the cause of differences found in developments of the intact seed and the detached embryo, on the species level.

In general, embryo transplantation was successful not only in intraspecific, but also in interspecific combinations. Fusion between the tissues, the grafted embryo and those of endosperm was observed in some intraspecific combinations. A homogeneous transplantation (= embryo transplanted to its own endosperm) and sixteen heterogeneous transplantations (=embryo transplanted to alien endosperms) were made as shown in Table 4. Affinity of Code No.1 embryo to alien endosperms and alien embryos to Code No.1 endosperm were mainly studied. Seedlings from homogeneous transplantation (1M·1D) produced coleoptile, the first, the second and

Table 4. Results in the case of transplantation of Code No.1 embryos to endosperms of nine strains and its reciprocal combinations in Experiment I.

Combination	Plumule					Radicle		
	Coleoptile	1st	2nd	3rd	Length	Development	Number	Length
1M·1D	+	+	+	±	11^{mm}	+	2	13^{mm}
1M·2D	+	+	±	—	12	+	2	11
1M·3D	+	+	±	—	11	+	1	11
1M·4D	+	+	±	—	11	±	1	13
1M·5D	+	±	±	—	8	+	1	5
1M·6D	+	±	±	—	10	+	1	3
1M·7D	+	—	—	—	4	±	1	1
1M·8D	±	—	—	—	3	—	0	0
1M·9D	±	±	—	—	8	±	1	10
2M·1D	+	±	±	—	6	+	2	18
3M·1D	+	±	—	—	4	±	1	18
4M·1D	+	+	—	—	5	±	1	8
5M·1D	+	±	—	—	10	±	1	15
6M·1D	+	±	—	—	6	±	1	16
7M·1D	+	±	—	—	5	±	1	5
8M·1D	+	±	—	—	8	±	1	12
9M·1D	+	±	—	—	7	±	1	10

sometimes the third leaf, and two roots. In comparison of 1W with 1M·1D, it was clearly shown that plumule and radicle of 1W developed far better than those of 1M·1D. The lengths of plumule and radicle in 1M·1D were 9.5% and 10.9% of the values of 1W, respectively. Growth of intact seedlings was more vigorous than that of transplanted seedlings. An affinity relation between embryo and endosperm reduces more clearly even in the homogeneous transplantation than in the intact seed. These findings may be regarded as the fundamental characteristics for the grafting experiment.

Seedlings from heterogeneous transplantation between "Kyoto Asahi" (Code No.1) embryo and the endosperms of different Asian species, namely, 1M·2D, 1M·3D, 1M·4D, 1M·5D and 1M·6D, produced coleoptile, the first leaf, the second leaf and one root, except 1M·2D which produced two roots. The length of those produced organs was almost similar with that of 1M·1D. From this result it is assumed that all Asian species used have close physiological affinity with Code No.1, and they are phylogenetically very close to Code No.1, a *japonica* of *O. sativa*. On the contrary, seedlings obtained from other heterogeneous transplantation between Code No.1 embryo with different African species, namely, 1M·7D, 1M·8D and 1M·9D, produced coleoptile, sometimes the first leaf and zero to one root. Therefore, it is assumed that Code No.7, Code No.8 and Code No.9, all African species, are remotely related to Code No.1.

In the reciprocal heterogeneous transplantations using embryo of different species with Code No.1 endosperm, seedlings produced coleoptile, the first leaf, sometimes the second leaf and one to two roots. Development of plumule and radicle in the reciprocal combination quite similar to the above mentioned one was only to be found in 1M·2D and 2M·1D. However, on other combinations seedlings from 1M·xD (Code No.1 embryo with endosperms of different strains) produced more organs than in the case of xM·1D, the reciprocal combination.

Table 5. Results in the case of transplantation of Code No.3 embryos to endosperms of nine strains and its reciprocal combinations in Experiment I.

Combination	Plumule					Radicle		
	Coleoptile	1st	2nd	3rd	Length	Development	Number	Length
3M·1D	+	±	—	—	4mm	+	1	18mm
3M·2D	+	±	±	—	6	+	1	5
3M·4D	+	±	—	—	2	+	1	4
3M·5D	+	—	—	—	5	±	1	14
3M·6D	+	—	—	—	3	±	1	15
3M·7D	+	±	—	—	5	+	1	5
3M·8D	+	±	—	—	2	±	1	2
3M·9D	+	—	—	—	3	±	1	2
1M·3D	+	+	±	—	11	+	1	11
2M·3D	+	+	+	±	20	+	1	30
4M·3D	+	+	+	+	30	+	1	16
5M·3D	+	±	±	—	35	+	1	30
6M·3D	+	+	±	—	14	+	1	21
7M·3D	+	±	±	—	11	±	1	3
8M·3D	+	+	±	—	16	±	1	20
9M·3D	+	+	±	—	10	±	1	8

As shown in Table 5, when Code No.3 (*indica* of *O. sativa*) embryo was transplanted to the endosperms of other *O. sativa* strains, namely, 3M·1D, 3M·2D and 3M·4D, coleoptile, the first and sometimes the second leaf, and one root appeared. Among them the best development of plumule was found in 3M·2D. However, the same embryo grafted on the other species, namely, 3M·5D, 3M·6D, 3M·7D, 3M·8D and 3M·9D, coleoptile, sometimes the first leaf and one root appeared. The seminal roots of 3M·1D, 3M·5D and 3M·6D were remarkably longer than those of other combinations. In the reciprocal heterogeneous transplantations, 1M·3D, 2M·3D, 4M·3D, 5M·3D, 6M·3D, 7M·3D, 8M·3D and 9M·3D, coleoptile, the first, the second and sometimes the third leaf, and one root appeared. The best development was observed in 4M·3D, a combination between two strains of *indica* of *O. sativa*. Development of seedlings of a given embryo with Code No.3 endosperm was better than seedlings of Code No.3 embryo with a given endosperm.

The endosperm did not become empty even when the third leaf developed in some cases, because total length of seedlings obtained from transplantation was clearly smaller than that from the intact seeds.

Considering the results obtained in Experiment I, the following conclusions may be drawn. The growth pattern of embryo grafted upon endosperm differs due to the combinations. Growth of the embryo of *japonica* type of *O. sativa* grafted upon endosperms of Asian species was more vigorous than the same embryo upon endosperm of the African species. Such differences were not found when the embryo of *indica* type was used. Nutrient absorption from the endosperm of *indica* type by the grafted embryo of other species was more remarkable than that from the endosperm of *japonica* type.

Experiment II

From the results of Experiment I, it became clear that nutrient absorption from the endosperm of *indica* type of *O. sativa* by embryos of other closely related species was far better than that from the endosperm of *japonica* type strains. In order to study in more detail, eight strains of *O. sativa* (four *japonica* and four *indica*) and each one strain of five species, *i.e.*, *O. sativa* var. *spontanea*, *O. perennis*, *O. barthii*, *O. glaberrima* and *O. breviligulata*, were used.

Table 6. Materials used in Experiment II.

Code No.	Species	Strain	Origin	Genome
11	*O. sativa (japonica)*	Kyoto Asahi	Japan	AA
12	〃 〃	Kinoshita Mochi	〃	〃
13	〃 〃	Norin No.8	〃	〃
14	〃 〃	Norin Mochi No.26	〃	〃
15	〃 *(indica)*	108	Formosa	〃
16	〃 〃	230	Philippines	〃
17	〃 〃	C7754	Sikkim	〃
18	〃 〃	C8448	New Guinea	〃
19	*O. sativa* var. *spontanea*	W0106	India	AA
20	*O. perennis*	W0120	〃	〃
21	*O. barthii*	W1045	Africa	〃
22	*O. glaberrima*	W0026	〃	〃
23	*O. breviligulata*	W0831	〃	〃
24	*O. officinalis*	W1263	North Borneo	CC
25	*O. minuta*	W0016	Philippines	BBCC
26	*O. eichingeri*	W0015	Africa	〃
27	*O. latifolia*	W0019	South America	CCDD
28	*O. australiensis*	W0008	Australia	EE
29	*O. meyeriana* subsp. *granulata*	W0003	India	–
30	*O. ridleyi*	W0001	Thailand	–
31	*O. brachyantha*	W1405	Sierra Leone	FF
32	*O. subulata*	W0510	Argentina	–

Code number, species name, strain number and the origin and genome constitution of the materials used are given in Table 6. Combinations of transplantation between embryo and endosperm are given in Table 7. Results of intact seeds and detached embryos one month later are given in Table 8 and Table 9, respectively. Results of embryo transplantation are shown in eight tables, Tables 10 to 17. Development of coleoptile, the first, the second, and the third

Table 7. Embryo transplantations made in Experiment II.

Embryo (Code No.) \ Endosperm (Code No.)	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	Intact seed	Detached embryo
11	◎	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	□	△
12	○				○				○	○	○	○	○										□	△
13	○				○				○	○	○	○	○										□	△
14	○				○				○	○	○	○	○										□	△
15	○	○	○	○	◎	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	○	□	△
16	○				○				○	○	○	○	○										□	△
17	○				○				○	○	○	○	○										□	△
18	○				○				○	○	○	○	○										□	△
19	○	○	○	○	○	○	○	○															□	△
20	○	○	○	○	○	○	○	○															□	△
21	○	○	○	○	○	○	○	○															□	△
22	○	○	○	○	○	○	○	○															□	△
23	○	○	○	○	○	○	○	○															□	△
24	○				○																		□	△
25	○				○																		□	△
26	○				○																		□	△
27	○				○																		□	△
28	○				○																		□	△
29	○				○																		□	△
30	○				○																		□	△
31	○				○																		□	△
32	○				○																		□	△

□, Intact seed; △, Detached embryo; ◎, Homogeneous transplantation; ○, Heterogeneous transplantation.

leaf, their total length, number of roots, and length of the longest root were recorded. The values in those tables are the average of 30 to 60 seedlings.

Table 8. Results in the case of intact seeds in Experiment II.

Strain	Plumule					Radicle		
	Coleoptile	1st	2nd	3rd	Length	Development	Number	Length
11W	+	+	+	+	116 mm	+	4	119 mm
12W	+	+	+	+	110	+	3	141
13W	+	+	+	+	102	+	3	103
14W	+	+	+	+	105	+	3	110
15W	+	+	+	+	126	+	3	157
16W	+	+	+	+	108	+	2	107
17W	+	+	+	+	112	+	2	112
18W	+	+	+	+	131	+	3	132
19W	+	+	+	+	121	+	2	118
20W	+	+	+	+	120	+	3	119
21W	+	+	+	+	109	+	1	73
22W	+	+	+	+	143	+	2	106
23W	+	+	+	+	138	+	3	82
24W	+	+	+	+	98	+	1	54
25W	+	+	+	+	96	+	1	32
26W	+	+	+	+	73	+	1	30
27W	+	+	+	+	100	+	2	56
28W	+	+	+	+	75	+	1	20
29W	+	+	+	+	82	+	1	17
30W	+	+	+	+	80	+	1	15
31W	+	+	+	+	79	+	1	15
32W	+	+	+	+	53	+	1	18

(1) Seedlings from intact seeds of all strains had coleoptile, the first, the second, the third leaf, and one to four roots (Table 8). From these results, it was reconfirmed that the plumule of *Oryza* has the capacity to develop coleoptile, the first, the second and the third leaf, using only their own nutrient from endosperm without any external supply of additional nutrient. This capacity was observed only in *O. sativa*, cultivated species, by DANJO (2). In this experiment, however, it became clear that plumule of most species of *Oryza* produced coleoptile, the first, the second and the third leaf using only their own nutrient from endosperm. No obvious differences in growth behaviour of plumule were recognized among species having the A genome. Length of developed plumule was more than 100 mm. No obvious differences were observed among species having other genomes than the A genome. Their plumules developed 73 to 100 mm in length except *O. subulata*, which developed only 53 mm in length. The values were clearly smaller than those of species having the A genome. Seedlings of *O. sativa* developed two to four roots with their length more than 100 mm. Seedlings of five species having the A genome developed one to three roots with more than 73 mm in length. Seedlings of nine species having other genomes than the A developed one to two roots being less than 56 mm in length. It was ascertained that when the third leaf was full-developed, endosperm became emp-

ty. It is clear that radicle of the species having the A genome grows more vigorously than that of the species having other genomes than the A.

Table 9. Growth of plumule and radicle in the case of detached embryo in Experiment II.

Strain	Plumule					Radicle		
	Coleoptile	1st	2nd	3rd	Length	Development	Number	Length
11M	+	+	+	−	6 mm	+	4	9 mm
12M	+	−	−	−	1	−	0	0
13M	+	+	+	−	5	±	1	2
14M	+	+	−	−	2	±	1	3
15M	+	−	−	−	1	±	1	2
16M	+	−	−	−	1	−	0	0
17M	+	−	−	−	1	−	0	0
18M	+	−	−	−	2	−	0	0
19M	+	±	−	−	4	−	0	0
20M	+	±	−	−	3	±	2	3
21M	+	−	−	−	1	−	0	0
22M	+	±	−	−	3	±	1	2
23M	+	−	−	−	1	−	0	0
24M	+	−	−	−	2	−	0	0
25M	+	−	−	−	1	−	0	0
26M	+	−	−	−	1	−	0	0
27M	+	−	−	−	1	−	0	0
28M	+	−	−	−	1	−	0	0
29M	+	−	−	−	1	−	0	0
30M	+	−	−	−	1	−	0	0
31M	+	−	−	−	1	−	0	0
32M	+	−	−	−	1	−	0	0

(2) Seedlings from the detached embryo without endosperm of *O. sativa* (11M to 18M) produced only coleoptile but sometimes they developed the first and the second leaf, and zero to four roots. Detached embryo of *japonica* type (11M to 14M) grew more remarkably than those of *indica* type (15M to 18M). Detached embryo of Code No.11 produced constantly large plumule and long radicle. Detached embryo (19M to 23M) without endosperm of the A genome group except *O. sativa* produced coleoptile, sometimes the first leaf and zero to two roots. Therefore, their growth was clearly slower than that of *japonica* type of *O. sativa*. Detached embryo (24M to 32M) of the species having various genomes other than the A produced only coleoptile but no roots. From these results the degree of growth vigor was in the order of *japonica* type, *indica* type of *O. sativa*, wild species having the A genome and those species having other genomes than the A.

(3) The embryo transplantation was successful not only in intraspecific, but also in interspecific combinations. One homogeneous transplantation and twenty-one heterogeneous transplantations were made, using the embryo of Code No.11 and the endosperms of Code No.11 to Code No.32 as shown in Table 10.

Table 10. Results in the case of transplantation of Code No.11 embryos to endosperms of 22 different strains in Experiment II.

Combination	Plumule					Radicle		
	Coleoptile	1st	2nd	3rd	Length	Development	Number	Length
Homo. Trans.					mm			mm
11M·11D	+	+	+	±	11	+	2	13
Hetero. Trans.								
Groupe 1								
11M·12D	+	+	±	—	12	+	2	11
11M·13D	+	+	±	—	11	+	1	11
11M·14D	+	+	±	—	10	+	2	12
11M·15D	+	+	±	—	13	+	2	11
11M·16D	+	+	±	—	11	±	1	13
11M·17D	+	+	±	—	12	±	1	12
11M·18D	+	+	±	—	13	±	2	13
Group 2								
11M·19D	+	±	±	—	8	+	1	5
11M·20D	+	±	±	—	10	+	1	3
Group 3								
11M·21D	+	—	—	—	4	±	1	1
11M·22D	+	—	—	—	3	—	0	0
11M·23D	+	±	—	—	8	±	1	10
Group 4								
11M·24D	±	—	—	—	3	—	0	0
11M·25D	±	—	—	—	3	—	0	0
11M·26D	±	—	—	—	2	—	0	0
11M·27D	±	—	—	—	3	—	0	0
11M·28D	+	—	—	—	1	—	0	0
11M·29D	+	—	—	—	2	—	0	0
11M·30D	+	—	—	—	1	—	0	0
11M·31D	+	—	—	—	2	—	0	0
11M·32D	+	—	—	—	3	—	0	0

Seedlings from a homogeneous transplantation, 11M·11D, produced coleoptile, the first, the second and sometimes the third leaf, and two roots. Growth of plumule and radicle in 11M·11D was inferior to that of 11W (cf. Table 8 and Table 10). The lengths of plumule and radicle in 11M·11D were only 9.5% and 10.9% of those of 11W, respectively. Seedlings from the first seven heterogeneous transplantations (11M·12D, 11M·13D, 11M·14D, 11M·15D, 11M·16D, 11M·17D and 11M·18D) constantly produced coleoptile, the first and the second leaf, and one to two roots. The length of plumule and root in the heterogeneous transplantations was quite similar with that of homogeneous transplantation.

Seedlings from the second two heterogeneous transplantations (11M·19D and 11M·20D) showed almost the same degree of seedling growth as observed in the heterogeneous transplantation within *O. sativa*. However, their root length was clearly shorter than those observed within *O. sativa*. This indicates that *O. sativa* var. *spontanea* (Code No.19) and *O. perennis* (Code No.20) are related taxonomically and closely to *O. sativa*. This relationship was already recognized through statistical study by MORISHIMA and OKA (8). They indicated that *O. sativa*, *O. sativa* var. *spontanea* and *O. perennis* are included in the same cluster. Seedlings from the third three heterogeneous transplantations with the A genome group (11M·21D, 11M·22D and 11M·23D) produced coleoptile, sometimes the first leaf and zero to one root. From the results, it is assumed that *O. barthii* (Code No.21), *O. glaberrima* (Code No.22) and *O. breviligulata* (Code No.23) are distantly related to *O. sativa*, though they belong to the A genome group. This relationship was already confirmed from the genetical study by NEZU *et al.* (11). They indicated that two intra-fertile but inter-sterile groups were established among diploid species possessing the A genome. One group consists of three Asian species, namely, *O. sativa*, *O. sativa* var. *spontanea* and *O. perennis*, while to the other belong three African species, namely, *O. glaberrima*, *O. breviligulata* and *O. stapfii*. All hybrids between them, though highly sterile, have invariably 12 bivalents.

Seedlings from the remaining nine heterogeneous transplantations (11M·24D, 11M·25D, 11M·26D, 11M·27D, 11M·28D, 11M·29D, 11M·30D, 11M·31D and 11M·32D), having endosperms of several different genomes other than the A, produced only coleoptile but no roots. They showed grafting damage to some extent. As shown in Table 9, seedlings from detached embryo of Code No.11 produced even sometimes the second leaf. Therefore, in these nine heterogeneous transplantations, it is assumed that growth inhibition of embryo might be occurring without any addition coming from the absorption of nutrient from alien endosperms. It is clear that *O. officinalis* (Code No.24), *O. minuta* (Code No.25), *O. eichingeri* (Code No.26), *O. latifolia* (Code No.27), *O. australiensis* (Code No.28), *O. meyeriana* subsp. *granulata* (Code No.29), *O. ridleyi* (Code No.30), *O. brachyantha* (Code No.31) and *O. subulata* (Code No.32) are distantly related to *japonica* type of *O. sativa*. This finding is consistent with the results of taxonomical study of the genus *Oryza* (14).

(4) One homogeneous transplantation and twenty-one heterogeneous transplantations were made by embryos of Code No.11 to Code No.32 grafted on the endosperm of Code No.11 as shown in Table 11. These are reciprocal combinations of those shown in Table 10.

Seedlings from the first three heterogeneous transplantations between embryos of three strains of *japonica* and Code No.11 endosperm (12M·11D, 13M·11D and 14M·11D) produced coleoptile, the first and sometimes the second leaf, and two roots. The lengths of plumule and radicle were 6-10 mm and 17-18 mm, respectively. Growth behaviour of reciprocal combinations of those three was quite same. Seedlings from the second nine heterogeneous transplantations in combinations between embryo of *indica* type of *O. sativa*, *O. sativa* var. *spontanea*, *O. perennis*, *O. barthii*, *O. glaberrima* and *O. breviligulata* and Code No.11 (a *japonica* of *O. sativa*) endosperm (15M·11D, 16M·11D, 17M·11D, 18M·11D, 19M·11D, 20M·11D, 21M·11D, 22M·11D and 23M·11D) produced coleoptile, the first leaf and one root. The lengths of plumule and radicle were 4-10 mm and 7-18 mm, respectively. Growth rate of these combinations was same or a little smaller than that of the reciprocal combinations.

Seedlings from the remaining nine heterogeneous transplantations (24M·11D, 25M·11D, 26M·11D, 27M·11D, 28M·11D, 29M·11D, 30M·11D, 31M·11D and 32M·11D) produced only coleoptile with 1 mm length but no roots. Growth rate of these combinations was nearly same as those of the reciprocal combinations.

(5) In order to find the difference of endosperm affinity between *japonica* and *indica* with embryos of different species, one homogeneous and twenty-one heterogeneous transplantations

Table 11. Results in the case of transplantation of embryos of 22 different strains to endosperms of of Code No.11.

Combination	Plumule					Radicle		
	Coleoptile	1st	2nd	3rd	Length	Development	Number	Length
Homo. Trans.					mm			mm
11M·11D	+	+	+	±	11	+	2	13
Hetero. Trans.								
Group 1								
12M·11D	+	+	±	−	6	+	2	18
13M·11D	+	±	±	−	8	+	2	17
14M·11D	+	+	−	−	10	+	2	17
Group 2								
15M·11D	+	+	−	−	4	+	1	18
16M·11D	+	+	−	−	5	+	1	8
17M·11D	+	+	−	−	6	+	1	9
18M·11D	+	+	−	−	8	+	1	7
19M·11D	+	±	−	−	10	+	1	15
20M·11D	+	±	−	−	6	±	1	16
21M·11D	+	±	−	−	5	+	1	5
22M·11D	+	±	−	−	8	±	1	12
23M·11D	+	±	−	−	7	±	1	10
Group 3								
24M·11D	+	−	−	−	1	−	0	0
25M·11D	+	−	−	−	1	−	0	0
26M·11D	+	−	−	−	1	−	0	0
27M·11D	+	−	−	−	1	−	0	0
28M·11D	+	−	−	−	1	−	0	0
29M·11D	+	−	−	−	1	−	0	0
30M·11D	+	−	−	−	1	−	0	0
31M·11D	+	−	−	−	1	−	0	0
32M·11D	+	−	−	−	1	−	0	0

were made, using embryo of Code No.15, an *indica* type of *O. sativa*, and endosperms of Code No.11 to Code No.32 as shown in Table 12. Seedlings from a homogeneous transplantation (15M·15D) produced coleoptile, the first and the second leaf, and two roots. Growth of plumule and radicle in 15M·15D was inferior to that of 15W (cf. Table 8 and Table 12). The lengths of plumule and radicle in 15M·15D were only 15.4% and 12.1% of those of 15W, respectively. Seedlings from the first twelve heterogeneous transplantations (15M·11D, 15M·12D, 15M·13D, 15M·14D, 15M·16D, 15M·17D, 15M·18D, 15M·19D, 15M·20D, 15M·21D, 15M·22D and 15M·23D) produced coleoptile, sometimes the first leaf and one root. The lengths of plumule and radicle were 2-7 mm and 2-18 mm, respectively. These values were smaller than those of seedlings from xM·11D, combinations between embryos of different species and the endosperm of Code No.11 (a *japonica* type of *O. sativa*). Seedlings from the second nine heterogeneous transplantations (15M·24D, 15M·25D, 15M·26D, 15M·27D, 15M·28D, 15M·29D, 15M·30D, 15M·31D and 15M·32D) produced only coleoptile but no roots.

Table 12. Results in the case of transplantation of Code No.15 embryos to endosperms of 22 different strains in Experiment II.

Combination	Plumule					Radicle		
	Coleoptile	1st	2nd	3rd	Length	Development	Number	Length
Homo. Trans.					mm			mm
15M·15D	+	+	±	—	32	+	2	19
Hetero. Trans.								
Group 1								
15M·11D	+	±	—	—	4	+	1	18
15M·12D	+	±	±	—	6	±	1	5
15M·13D	+	—	—	—	5	+	1	8
15M·14D	+	—	—	—	6	+	1	9
15M·16D	+	—	—	—	2	+	1	4
15M·17D	+	—	—	—	4	+	1	7
15M·18D	+	—	—	—	7	+	1	6
15M·19D	+	—	—	—	5	±	1	14
15M·20D	+	—	—	—	3	±	1	15
15M·21D	+	±	—	—	5	±	1	5
15M·22D	+	±	—	—	2	±	1	2
15M·23D	+	—	—	—	3	±	1	2
Group 2								
15M·24D	+	—	—	—	2	—	0	0
15M·25D	+	—	—	—	1	—	0	0
15M·26D	+	—	—	—	1	—	0	0
15M·27D	+	—	—	—	1	—	0	0
15M·28D	+	—	—	—	1	—	0	0
15M·29D	+	—	—	—	1	—	0	0
15M·30D	+	—	—	—	1	—	0	0
15M·31D	+	—	—	—	1	—	0	0
15M·32D	+	—	—	—	1	—	0	0

The length of plumule was 1-2 mm and was clearly shorter than that of seedlings from xM·11D.

(6) One homogeneous transplantation and twenty-one heterogeneous transplantations were made, using the embryos of Code No.11 to Code No.32 and the endosperm of Code No.15 as shown in Table 13. These are reciprocal combinations of those shown in Table 12.

Seedlings from the first seven heterogeneous transplantations among *O. sativa* (11M·15D, 12M·15D, 13M·15D, 14M·15D, 16M·15D, 17M·15D and 18M·15D) produced coleoptile, the first, the second and sometimes the third leaf, and one root. The lengths of plumule and radicle were 13-30 mm and 11-30 mm, respectively. Those values were about four times in plumule and two times in radicle larger than those of the reciprocal combinations.

Seedlings from the second five heterogeneous transplantations (19M·15D, 20M·15D, 21M·15D, 22M·15D and 23M·15D) produced coleoptile, the first and the second leaf, and one root. The lengths of plumule and radicle were 10-35 mm and 3-30 mm, respectively. These values were about five times in plumule and two times in radicle larger than those of the reciprocal

Table 13. Results in the case of transplantation of embryos of 22 different strains to endosperms of Code No.15.

Combination	Plumule					Radicle		
	Coleoptile	1st	2nd	3rd	Length	Development	Number	Length
Homo. Trans.					mm			mm
15M·15D	+	+	±	−	32	+	2	19
Hetero. Trans.								
Group 1								
11M·15D	+	+	±	−	13	+	1	11
12M·15D	+	+	+	+	20	+	1	30
13M·15D	+	+	+	−	17	+	1	16
14M·15D	+	+	+	−	13	+	1	14
16M·15D	+	+	+	+	30	+	1	16
17M·15D	+	+	+	−	28	+	1	13
18M·15D	+	+	+	−	22	+	1	17
Group 2								
19M·15D	+	±	±	−	35	+	1	30
20M·15D	+	+	±	−	14	+	1	21
21M·15D	+	±	±	−	11	±	1	3
22M·15D	+	+	±	−	16	±	1	20
23M·15D	+	+	±	−	10	±	1	8
Group 3								
24M·15D	+	±	−	−	5	−	0	0
25M·15D	+	±	−	−	4	±	1	4
26M·15D	+	±	−	−	3	−	0	0
27M·15D	+	±	−	−	4	−	0	0
Group 4								
28M·15D	+	−	−	−	2	−	0	0
29M·15D	+	−	−	−	2	−	0	0
30M·15D	+	−	−	−	3	−	0	0
31M·15D	+	−	−	−	3	−	0	0
32M·15D	+	−	−	−	1	−	0	0

combinations. Seedlings from the third four heterogeneous transplantations (24M·15D, 25M·15D, 26M·15D and 27M·15D) produced coleoptile, the first leaf and zero to one root. The length of plumule was about three times larger than that of the reciprocal combinations. Seedlings from the remaining five heterogeneous transplantations (28M·15D, 29M·15D, 30M·15D, 31M·15D and 32M·15D) produced only coleoptile but no roots. The length of plumule was about two times longer than that of the reciprocal combinations.

It is very interesting that the growth patterns of the first group (24M·15D, 25M·15D, 26M·15D and 27M·15D) were clearly different from those of the second group (28M·15D, 29M·15D, 30M·15D, 31M·15D and 32M·15D). However, such clear difference was not found among 11M·xD, xM·11D and 15M·xD.

(7) In order to find the detailed affinity between embryo and endosperm among different strains of *O. sativa* and closely related species having the A genome, eighty heterogeneous trans-

plantations were made.

Twenty heterogeneous transplantations were made, using the embryos of Code No.11 to Code No.14 (all *japonica* type of *O. sativa*) and the endosperms of Code No.19 (*O. sativa* var. *spontanea*), Code No.20 (*O. perennis*), Code No.21 (*O. barthii*), Code No.22 (*O. glaberrima*), Code No.23 (*O. breviligulata*) as shown in Table 14.

Table 14. Transplantations made by embryos of *O. sativa*(*japonica* type) on the several endosperms of closely related species to *O. sativa* in Experiment II.

Combination	Plumule					Radicle		
	Coleoptile	1st	2nd	3rd	Length	Development	Number	Length
11M·19D	+	±	±	—	8 mm	+	1	5 mm
12M·19D	+	±	—	—	5	+	1	3
13M·19D	+	±	—	—	3	+	1	3
14M·19D	+	±	—	—	5	+	1	2
11M·20D	+	±	±	—	10	+	1	3
12M·20D	+	±	±	—	8	±	1	5
13M·20D	+	±	—	—	5	±	1	5
14M·20D	+	±	—	—	4	+	1	3
11M·21D	+	—	—	—	4	±	1	1
12M·21D	+	—	—	—	5	±	1	2
13M·21D	+	—	—	—	4	±	1	2
14M·21D	+	—	—	—	4	±	1	1
11M·22D	+	—	—	—	3	—	0	0
12M·22D	+	—	—	—	2	—	0	0
13M·22D	+	—	—	—	2	—	0	0
14M·22D	+	—	—	—	1	—	0	0
11M·23D	+	±	—	—	8	±	1	10
12M·23D	+	±	—	—	5	±	1	5
13M·23D	+	±	±	—	8	—	0	0
14M·23D	+	+	±	—	11	—	0	0

Seedlings from the first twelve transplantations in which four kinds of the embryos, 11M, 12M, 13M and 14M, were grafted on three kinds of the endosperms, 19D, 20D and 23D, produced coleoptile, the first and sometimes the second leaf, and one root. The lengths of plumule and radicle were 3-11 mm and 0-10 mm, respectively. However, seedlings from the second eight transplantations, namely, the embryos of 11M, 12M, 13M and 14M to the endosperms of 21D and 22D, produced only coleoptile and zero to one root. The lengths of plumule and that of radicle were 1-5 mm and 0-2 mm, respectively. Embryos of *japonica* type of *O. sativa* absorbed nutrient better from the endosperms of *O. sativa* var. *spontanea* (Code No.19), *O. perennis* (Code No.20) and *O. breviligulata* (Code No.23) than from the endosperms of *O. barthii* (Code No.21) and *O. glaberrima* (Code No.22).

(8) Twenty heterogeneous transplantations were made, using the embryos of *O. sativa* var. *spontanea* (Code No.19), *O. perennis* (Code No.20), *O. barthii* (Code No.21), *O. glaberrima* (Code No.22), *O. breviligulata* (Code No.23) and the endosperms of Code No.11 to Code No.

14 (*japonica* type of *O. sativa*) as shown in Table 15. These are reciprocal combinations of those shown in Table 14. Seedlings from all combinations produced coleoptile, the first leaf and one root. The lengths of plumule and radicle were 2-16 mm and 3-16 mm, respectively. Embryos of these four species absorbed nutrient almost uniformly from the endosperm of *japonica* type of *O. sativa*.

Table 15. Transplantations made by embryos of several species (closely related to *O. sativa*) to the endosperms of *O. sativa* (*japonica* type) in Experiment II.

Combination	Plumule					Radicle		
	Coleoptile	1st	2nd	3rd	Length	Development	Number	Length
19M·11D	+	±	−	−	10 mm	+	1	15 mm
20M·11D	+	±	−	−	6	±	1	16
21M·11D	+	±	−	−	5	+	1	5
22M·11D	+	±	−	−	8	±	1	12
23M·11D	+	±	−	−	7	±	1	10
19M·12D	+	±	−	−	11	±	1	11
20M·12D	+	±	−	−	13	±	1	8
21M·12D	+	±	−	−	10	±	1	9
22M·12D	+	+	−	−	5	±	1	7
23M·12D	+	±	−	−	7	±	1	18
19M·13D	+	+	−	−	10	±	1	11
20M·13D	+	±	−	−	12	±	1	8
21M·13D	+	±	−	−	10	±	1	10
22M·13D	+	±	−	−	6	±	1	9
23M·13D	+	±	−	−	2	±	1	16
19M·14D	+	±	−	−	2	+	1	3
20M·14D	+	+	−	−	2	+	1	6
21M·14D	+	+	−	−	3	+	1	6
22M·14D	+	±	−	−	16	±	1	6
23M·14D	+	−	−	−	3	±	1	3

(9) Twenty heterogeneous transplantations were made, using the embryos of Code No.15 to Code No.18 (all *indica* type of *O. sativa*) and the endosperms of Code No.19 to Code No.23 as shown in Table 16. Half of seedlings produced coleoptile, the first leaf and zero to one root. The lengths of plumule and radicle were 2-5 mm and 0-11 mm, respectively. The other half of seedlings produced only coleoptile and one root. The lengths of plumule and radicle were 1-5 mm and 2-15 mm, respectively. The second leaf was found only in 18M·23D combination.

(10) Twenty heterogeneous transplantations were made, using the embryos of Code No.19 to Code No.23 and the endosperms of Code No.15 to Code No.18 as shown in Table 17. Seedlings from three heterogeneous transplantations (23M·16D, 23M·17D and 23M·18D) (all with *O. breviligulata* embryo) produced coleoptile, the first leaf and one root. The lengths of plumule and radicle were 7-19 mm and 3-4 mm, respectively. Seedlings from eleven heterogeneous transplantations (19M·15D, 20M·15D, 21M·15D, 22M·15D, 23M·15D, 19M·16D, 22M·16D, 19M·17D, 22M·17D, 19M·18D and 22M·18D) produced coleoptile, the first and the

Table 16. Transplantations made by embryos of *O. sativa* (*indica* type) to several endosperms of closely related species to *O. sativa* in Experiment II.

Combination	Plumule					Radicle		
	Coleoptile	1st	2nd	3rd	Length	Development	Number	Length
15M·19D	+	–	–	–	5 mm	±	1	14 mm
16M·19D	+	±	–	–	3	±	1	11
17M·19D	+	–	–	–	2	±	1	5
18M·19D	+	–	–	–	2	±	1	7
15M·20D	+	–	–	–	3	±	1	15
16M·20D	+	–	–	–	3	±	1	3
17M·20D	+	±	–	–	2	–	0	0
18M·20D	+	±	–	–	3	±	1	3
15M·21D	+	±	–	–	5	±	1	5
16M·21D	+	±	–	–	4	±	1	4
17M·21D	+	±	–	–	4	±	1	3
18M·21D	+	±	–	–	5	±	1	3
15M·22D	+	±	–	–	2	±	1	2
16M·22D	+	–	–	–	3	±	1	3
17M·22D	+	–	–	–	1	±	1	3
18M·22D	+	±	–	–	3	–	0	0
15M·23D	+	–	–	–	3	±	1	2
16M·23D	+	–	–	–	2	±	1	3
17M·23D	+	–	–	–	3	±	1	2
18M·23D	+	+	±	–	5	+	1	3

second leaf, and one root. The lengths of plumule and radicle were 10-35 mm and 3-30 mm, respectively. Seedlings from six heterogeneous transplantations (20M·16D, 21M·16D, 20M·17D, 21M·17D, 20M·18D and 21M·18D) produced coleoptile, the first, the second and the third leaf, and one root. The lengths of plumule and radicle were 17-29 mm and 14-20 mm, respectively. It is interesting that only one root was appeared from the radicle even when the second leaf developed from plumule. It is assumed that growth of radicle is to be inhibited by grafting more easily than in those of plumule.

Conclusion

Growth lengths of the plumules in each plots of Experiment II are summarized in Table 18. In the table, j, i and r indicate the following strains:

j: *japonica* type of *O. sativa*; *i.e.*, Code No.11, Code No.12, Code No.13 and Code No.14

i: *indica* type of *O. sativa*; *i.e.*, Code No.15, Code No.16, Code No.17 and Code No.18

r: closely related wild and cultivated species to *O. sativa*; *i.e.*, Code No.19 (*O. sativa* var. *spontanea*), Code No.20 (*O. perennis*), Code No.21 (*O. barthii*), Code No.22 (*O. glaberrima*) and Code No.23 (*O. breviligulata*)

Table 17. Transplantations made by embryos of several sepcies (closely related to *O. sativa*) to the endosperms of *O. sativa* (*indica* type) in Experiment II.

Combination	Plumule					Radicle		
	Coleoptile	1st	2nd	3rd	Length	Development	Number	Length
Group 1					mm			mm
23M·16D	+	+	−	−	10	±	1	4
23M·17D	+	+	−	−	19	±	1	3
23M·18D	+	+	−	−	7	±	1	3
Group 2								
19M·15D	+	±	±	−	35	+	1	30
20M·15D	+	+	±	−	14	+	1	21
21M·15D	+	±	±	−	11	±	1	3
22M·15D	+	+	±	−	16	±	1	20
23M·15D	+	+	±	−	10	±	1	8
19M·16D	+	+	±	−	18	±	1	27
22M·16D	+	+	±	−	23	±	1	6
19M·17D	+	+	±	−	22	±	1	25
22M·17D	+	+	±	−	11	+	1	7
19M·18D	+	+	±	−	24	±	1	19
22M·18D	+	+	±	−	21	±	1	5
Group 3								
20M·16D	+	+	+	±	26	±	1	14
21M·16D	+	+	+	±	22	±	1	16
20M·17D	+	+	±	±	29	+	1	16
21M·17D	+	+	+	±	25	+	1	20
20M·18D	+	+	+	±	17	+	1	20
21M·18D	+	+	+	±	18	+	1	17

Table 18. Summarized growth lengths of the plumules in Experiment II.

jM·rD		rM·jD		iM·rD		rM·iD	
jM·19D	21 mm	19M·jD	33 mm	iM·19D	12 mm	19M·iD	99 mm
jM·20D	27	20M·jD	33	iM·20D	11	20M·iD	86
jM·21D	17	21M·jD	28	iM·21D	18	21M·iD	77
jM·22D	8	22M·jD	35	iM·22D	9	22M·iD	71
jM·23D	32	23M·jD	19	iM·23D	13	23M·iD	46
Total	105	Total	148	Total	63	Total	379
Average	21.0	Average	29.6	Average	12.6	Average	75.8
11M·rD	33	rM·11D	36	15M·rD	18	rM·15D	86
12M·rD	25	rM·12D	46	16M·rD	15	rM·16D	99
13M·rD	22	rM·13D	40	17M·rD	12	rM·17D	106
14M·rD	25	rM·14D	26	18M·rD	18	rM·18D	88
Total	105	Total	148	Total	63	Total	379
Average	26.3	Average	37.0	Average	15.8	Average	94.8

Table 19. Summarized growth lengths of the radicles in Experiment II.

jM·rD		rM·jD		iM·rD		rM·iD	
jM·19D	13 mm	19M·jD	40 mm	iM·19D	37 mm	19M·iD	101 mm
jM·20D	16	20M·jD	38	iM·20D	21	20M·iD	71
jM·21D	6	21M·jD	30	iM·21D	15	21M·iD	56
jM·22D	0	22M·jD	34	iM·22D	8	22M·iD	38
jM·23D	15	23M·jD	47	iM·23D	10	23M·iD	18
Total	50	Total	189	Total	91	Total	284
Average	10.0	Average	37.8	Average	18.2	Average	56.8
11M·rD	19	rM·11D	58	15M·rD	38	rM·15D	82
12M·rD	15	rM·12D	53	16M·rD	24	rM·16D	67
13M·rD	10	rM·13D	54	17M·rD	13	rM·17D	71
14M·rD	6	rM·14D	24	18M·rD	16	rM·18D	64
Total	50	Total	189	Total	91	Total	284
Average	12.5	Average	47.3	Average	22.8	Average	71.0

The order of seedling vigor was rM·iD, rM·jD, jM·rD and iM·rD. In the heterogeneous transplantations of rM·iD, the order of seedling vigor was 19M·iD, 20M·iD, 21M·iD, 22M·iD and 23M·iD.

The growth lengths of the radicles in each plots of Experiment II are summarized in Table 19. Seedlings from the rM·iD grew most vigorously and followed by rM·jD, iM·rD and jM·rD. In the heterogeneous transplantations of iM·rD and rM·iD, iM·19D and 19M·iD were most vigorous, iM·22D and 22M·iD were fairly vigorous, but iM·23D and 23M·iD were not vigorous.

From the results obtained, the following conclusions are drawn. The nutrient absorption from the endosperms of *indica* type strains by the embryos of the closely related wild species, is more striking than that from the endosperms of *japonica* type strains. It implies that *indica* type is taxonomically more closely related to wild relatives than *japonica* type. This relationship might have an important effect upon physiological affinity between embryo and endosperm in heterogeneous transplantation. In the course of phylogenetic differentiation of *O. sativa*, *indica* type might have occurred first from the putative ancestor, *O. sativa* var. *spontanea* or *O. perennis*, and afterward, *japonica* type might have derived from *indica* type.

Some embryos of *O. breviligulata* (Code No.23) and some seedlings grafted on endosperms of *O. breviligulata* grew vigorously than that of other seedlings. It might be due to the fact that embryo and endosperm of *O. breviligulata* are comparatively larger than that of the other wild species.

Sixty days after grafting, callus formation was found in the most combinations.

Summary

In order to investigate the compatibility relation between embryos and endosperms in the genus *Oryza*, embryo transplantations were made, using 22 strains belonging to 15 species, including 2 cultivated and 13 wild species.

It became clear that plumule of most species of *Oryza* used produced coleoptile, the first, the second and the third leaf, using only their own nutrient from endosperm. In general, the transplantations were successful not only in the intraspecific, but also in the interspecific combinations. Furthermore, fusion between tissues of embryo and that of endosperm was ascertained in some intraspecific combinations. The growth of seedlings obtained from homogeneous transplantations was more vigorous than seedlings obtained from heterogeneous transplantations. The growing rate of embryos grafted upon endosperms differs according to each combination.

The embryos of *japonica* type grafted upon the endosperms of Asian species were more vigorous than the same embryos grafted upon the endosperms of African species. Such differences were not found with the embryos of *indica* type strains. Nutrient absorption from the endosperms of *indica* type strains by embryos of the closely related wild species, was far better than from the endosperms of *japonica* type strains. From these results, it was strongly indicated that *indica* type might occurred first from the putative ancestor and afterward, *japonica* type might have derived from *indica* type. Several articles were discussed in taxonomical, physiological and phylogenetical viewpoint.

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Literature Cited

1. AMEMIYA, A., H. AKEMINE and K. TORIYAMA Cultural conditions and growth of immature embryo in rice plant (in Jap. with Eng. Sum.). *Bull. Nat. Inst. Agr. Sci. D* **6**:1-40, 1956.
2. DANJO, T. On the consumption of the reserve foods in endosperms of the cereal seeds during germination. (1) Relation to the growth of rice (in Jap. with Eng. Sum.). *Proc. Crop Sci. Soc., Japan* **19**:251-254, 1951.
3. KATAYAMA, T. C. Preliminary studies on embryo transplantation in the genus *Oryza*. *Ann. Rep. Nat. Inst. Genet.* **15**:82-83, 1965.
4. KATAYAMA, T. C. Embryo transplantation in the genus *Oryza*. *Ann. Rep. Nat. Inst. Genet.* **16**:65-66, 1966.
5. KATAYAMA, T. C. Further studies on embryo transplantation in the genus *Oryza*. *Ann. Rep. Nat. Inst. Genet.* **17**:57, 1967.
6. KATAYAMA, T. C. Botanical studies in the genus *Oryza*. I. *Memoirs Fac. Agr. Kagoshima Univ.* **7**:89-117, 1969.
7. KIKUCHI, M. Studies on the transplantation of immature embryo in cereals. *Special Bull. Coll. Agr. Utsunomiya Univ.* **21**:1-169, 1964.
8. MORISHIMA, H. and H. I. OKA The pattern of interspecific variation in the genus *Oryza*: its quantitative representation by statistical methods. *Evolution* **14**:153-165, 1960.
9. NAKAJIMA, T. Studies on embryo culture in plants. I. (in Jap. with Eng. Sum.). *Jap. Jour. Breed.* **7**:161-168, 1958.

10. NAKAJIMA, T. and H. MORISHIMA Studies on embryo culture interspecific hybrids in *Oryza* (in Jap. with Eng. Sum.). *Jap. Jour. Breed.* **8**:105-110, 1958.
11. NEZU, M., T. C. KATAYAMA and H. KIHARA Genetic study of the genus *Oryza*. I. *Seiken Zihô* **11**: 1-11, 1960.
12. ROMMEL, M. Eine vereinfachte Methode der Embryokultur bei Getreide. *Züchter* **28**:149-151, 1958.
13. SACHAR, K., M. S. SWAMINATHAN and R. D. IYER The effect of reciprocal grafting on embryo and endosperm development in crosses between *Corchorus olitorius* and *C. capsularia*. *Zeit. Pflanzenz.* **52**: 355-365, 1964.
14. TATEOKA, T. Taxonomic studies of *Oryza*. III. *Bot. Mag., Tokyo* **76**:165-173, 1963.