Linkage Study of the Genes Belonging to the First Chromosome in Rice¹⁾

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Introduction

In rice, *Oryza sativa* L., several reports on the linkage relation between the gene for endosperm character and one for apiculus coloration have been published since TAKAHASHI (1923) first reported. NAGAO and TAKAHASHI (1948) proposed that apiculus coloration is dependent on the complementary genes, *C* and *Sp*, and concluded that *C* is the gene linked with *gl*, glutinous endosperm. On the other hand, MORINAGA et al. (1942, 1948) found the linkage relation between *C* and *pa*, depressed palea, with the recombination value of about 20%. Judging from these two reports, the linkage relation must exist between *gl* and *pa*, but it has not yet been confirmed.

We have dealt with this problem and found the linkage between these two genes, moreover, a fact that one of the genes for white stripe belongs to this linkage group. In the present paper the linkage relation of these genes is reported.

Description of Genes

Four genes taken up in this study and genetic characteristics revealed by them are as follows.

wx: Simple recessive gene for waxy endosperm. This gene has been described hitherto as *gl*, *gu*, *u*, etc., corresponding to glutinous endosperm.

C: Dominant gene for the formation of chromogen (NAGAO and TAKAHASHI, 1949).

dp: The gene for depressed palea. MORINAGA and KURIYAMA (1948) represented already this trait with the symbol *pa* but we designate it as *dp*, conforming to the rules of genetic nomenclature and symbolization. This character is seen in the mutant variety Hen-ei-to which has been preserved at our experimental farm since 1922. The palea of this mutant is undergrown and hence the shape of spikelet is abnormal as shown in Fig. 1, and the kernels are also abnormal in shape and size. Segregation of normal vs. depressed palea was investigated in 11 lines of F_2 generation. As seen in Table 1, it was found that depressed palea behaved simple recessive to normal type.

Fig. 1. Grain shape
A: depressed palea
B: normal palea

1) Received on April 16, 1962

Table 1. Segregation for normal vs. depressed palea in F_2

F_2 line no.	Normal palea	Depressed palea	Total	χ^2	P
32~27	378	132	510	0.212	0.75~0.50
43~24	276	90	366	0.033	0.90~0.75
46~20	122	43	165	0.099	0.90~0.75
51~63	199	68	267	0.031	0.90~0.75
55~95	152	40	192	1.778	0.25~0.10
58~588	226	74	300	0.018	>0.90
60~105	133	51	184	0.725	0.50~0.25
60~134	127	39	166	0.201	0.75~0.50
60~135	101	25	126	1.788	0.25~0.10
60~140	145	45	190	0.175	0.75~0.50
60~141	92	23	124	0.043	0.90~0.75
Total	1951	639	2590	0.132	0.75~0.50

Table 2. Segregation for green vs. white striped leaf in F_2

F_2 line no.	Green leaf	White striped leaf	Total	χ^2	P
44~56	318	93	411	1.234	0.50~0.25
46~6	95	26	121	0.796	0.50~0.25
46~77	222	70	292	0.644	0.50~0.25
47~33	240	65	305	2.213	0.25~0.10
48~30	317	121	438	1.610	0.25~0.10
52~40	273	71	344	3.488	0.10~0.05
52~41	208	56	264	2.020	0.25~0.10
55~79	163	69	232	2.782	0.10~0.05
55~134	116	36	152	0.140	0.75~0.50
58~558	220	80	300	0.444	0.75~0.50
Total	2172	687	2859	1.437	0.25~0.10

Results

Recombination values were calculated by the method of maximum likelihood given by FISHER et al. (1928). The results obtained are as follows. *dp-wx*: On the segregation of waxy vs. non-waxy endosperm, 3 of 5 lines tested were investigated in F_2 seeds in which the ratio of 3 non-waxy : 1 waxy was expected (Table 3-A). The remaining 2 lines were studied in F_3 in which the expected ratio of three genotypes, ++, +*wx* and *wxwx*, was 1:2:1 (Table 3-B). There combination values between *dp* and *wx* were 12.68% from the

ws: The gene for white striped leaf. Such Japanese rice varieties as Shima-ine, Shiroshima-ine etc. have this character. It appears from the seedling stage, but the manifestation of this character is remarkably clear in the tillering stage and mostly disappears by the heading time. Table 2 indicates that this character behaves simple recessive to green leaf blade, showing the tendency that the observed number of recessive individuals is fewer than the expected one. As to the stripe traits TAKAHASHI (1950) described two genes, *fs* and *gw*. We have not identified these two genes with *ws* by crossing but it was thought that *ws* was not the same as *fs* and *gw*, because of differences in the mode of manifestation of character and in the loci, namely *gw* and *fs* belong to the 6th and 7th linkage group respectively.

Table 3. Linkage relation between *dp* and *wx*A. F_2 segregation (3:1)(3:1)

Items	F_2 phenotype				Total
	++	+ <i>wx</i>	<i>dp</i> +	<i>dpwx</i>	
F_2 60~105	91	42	49	2	184
60~134	89	38	39	0	166
60~140	98	47	45	0	190
Total obs. no.	278	127	133	2	540
Calc. 12.68%	272.17	132.83	132.83	2.17	540

$$\chi^2=0.397 \quad P=0.95\sim0.90$$

B. F_2 segregation (3:1)(1:2:1)

Items	+			<i>dp</i>			Total
	++	+ <i>wx</i>	<i>wxwx</i>	++	+ <i>wx</i>	<i>wxwx</i>	
F_2 60~135	4	65	32	24	1	0	126
60~141	0	62	30	32	0	0	124
Total obs. no.	4	127	62	56	1	0	250
Calc. 2.05%	2.54	122.49	62.47	59.96	2.51	0.03	250

$$\chi^2=2.209 \quad P=0.75\sim0.50$$

former and 2.05 % from the latter. In order to estimate a more accurate distance between these two genes, F_3 seeds of single dominant, $dp+$, individuals obtained from the former 3 lines were grown to test their genotypes. The recombination value calculated from the data shown in Table 4 was 1.95 %, which closely corresponded to 2.05 % obtained from the data shown in Table 3-B. The weighted average calculated from these three recombination values was 2.23 ± 0.62 %, as shown in Table 5, by the method given by ROBERTSON et al. (1944). It is considered from these results that dp and wx are

Table 4. F_3 test of single dominant F_2 plants ($dp+$)

Items	F ₂ genotype for wx		Total
	++	$+wx$	
F ₂ 60~105	48	0	48
60~134	36	3	39
60~140	42	2	44
Total obs. no.	126	5	131
Calc. 1.95 %	126	5	131

Table 5. Weighted mean of p value between dp and wx

Source of data	p value	i	n	I=ni	pI
F ₂ (3:1)(3:1) rep.	0.1268	1.04066	540	561.9575	71.2562
F ₂ (3:1)(1:2:1) rep.	0.0205	50.24461	250	12561.1525	257.5036
F ₃ from $+dp$ plants	0.0195	100.64051	131	13183.9068	257.0862
Σ				26307.0168	585.8460

$$p_w = \frac{\Sigma pI}{\Sigma I} = 0.0223$$

located closely to each other on the first chromosome.

$dp-C$: This relation was tested in the F_2 data obtained from 4 lines, among which 3 lines were in coupling phase and 1 was in repulsion (Table 6). The recombination values were 21.10 % in coupling, 25.88 % in repulsion, and 21.33 ± 1.88 % for the weighted mean. This value corresponds to that obtained by MORINAGA et al. (1942, 1948).

$wx-C$: The recombination percentage between these genes were about 20 %, ranging from 19 to 24 % by different workers and materials. From our data presented in Table 7, it was calculated to be 23.10 ± 2.29 %, which agrees well with the above values.

$C-ws$: From the linkage relation between these two genes, it was found newly that ws belonged to this linkage group. Total observed numbers of four phenotypes, $+C$, $++$, wsC , and $ws+$, obtained from 9 F_2 lines were 1796, 260, 222, and 429, respectively (Table 8). The recombination value estimated from the data was 20.08 ± 0.88 %, and the fit to the

Table 6. Linkage relation between dp and C

Items	F ₂ phenotype				Total
	+C	++	dpC	dp+	
Coupling					
F ₂ 39~27	328	50	47	85	510
43~24	238	38	37	53	366
46~20	108	14	17	26	165
51~63	175	24	26	42	267
55~95	135	17	13	27	192
Total obs. no.	984	143	140	233	1500
Calc. 21.10%	983.44	141.56	141.56	233.44	1500
Repulsion					
F ₂ 58~558	145	81	68	6	300
Calc. 25.88%	155.02	69.98	69.98	5.02	300

Coupling : $\chi^2=0.033$ P 0.995, Repulsion : $\chi^2=2.646$
 $P=0.50 \sim 0.25$ $p_w=21.33 \pm 1.18$ %

Table 7. Linkage relation between *wx* and *C*

Items	F ₂ phenotype				Total
	+C	++	<i>wxC</i>	<i>wx+</i>	
F ₂ 40~58	123	20	17	20	180
40~126	201	23	30	39	293
55~115	127	16	15	24	182
56~57	126	25	21	27	199
Total obs. no.	577	84	83	110	854
Calc. 23.10%	553.26	87.24	87.24	126.26	854

$$\chi^2=3.437 \quad P=0.50\sim0.25$$

Table 8. Linkage relation between *ws* and *C*

Items	F ₂ phenotype				Total
	+C	++	<i>wsC</i>	<i>ws+</i>	
F ₂ 44~56	284	34	28	65	411
46~6	85	10	12	14	121
46~77	194	28	27	43	292
47~33	208	32	27	38	305
48~30	275	42	42	79	438
52~40	237	36	22	49	344
52~41	184	24	16	40	264
55~79	140	23	24	45	232
58~558	189	31	24	56	300
Total obs. no.	1796	260	222	429	2707
Calc. 20.08%	1785.75	244.50	244.50	432.25	2707

$$\chi^2=3.136 \quad P=0.50\sim0.25$$

Table 9. Linkage relation between *dp* and *ws*

Items	F ₂ phenotype				Total
	++	+ <i>ws</i>	<i>dp+</i>	<i>dpws</i>	
F ₂ 58~588	154	72	66	8	300
Calc. 31.62%	157.50	67.50	67.50	7.50	300

$$\chi^2=1.411 \quad P=0.75\sim0.50$$

Table 10. Linkage relation between *ws* and *wx*

Items	F ₂ phenotype				Total
	++	+ <i>wx</i>	<i>ws+</i>	<i>wswx</i>	
F ₂ 55~134	78	38	31	5	152
Calc. 33.83%	80.35	33.65	33.65	4.35	152

$$\chi^2=0.937 \quad P=0.90\sim0.75$$

Fig. 2. Provisional linkage map of 1st group

value expected on this basis proved to be good.

dp-ws: The data from this case are listed in Table 9 and the recombination percentage of *dp-ws* was calculated to be $31.62 \pm 5.12\%$. From this value and those of *dp-C* and *C-ws*, the linear order of the genes is inferred as *dp-C-ws*.

wx-ws: From the F₂ data presented in Table 10, $33.83 \pm 7.08\%$ was obtained as the recombination value. The accuracy of this value is reduced owing to a small number of lines and individuals tested, but it is sufficient to conclude that the three genes under consideration may be arranged in the order of *wx-C-ws*.

Discussion and Conclusion

The results mentioned above are summarized in Fig. 2. In this linkage map, the order of arrangement of the four genes on chromosome 1 is *wx-dp-C-ws*. As this order of arrangement was, however, presumed on the basis of recombination values obtained from different F₂ lines and moreover *wx* and *dp* were located closely, the more detailed data are needed to confirm the linear order of these four genes.

Several papers on the linkage relation between these genes and others have been presented. NAGAO and TAKAHASHI (1960) described the linkage map of chromosome 1, which consisted of three genes, *C*, *wx* and *d₄*. According to them, the recombination percentages of *C-wx*, *wx-d₄* and *C-d₄* were 22.8, 21.5 and 38.2%, respectively, and the linear order of the genes were proposed to be *d₄-wx-C*. Unfortunately,

we have not yet examine the relation between d_4 and others mentioned above, but it is inferred from our results and theirs that the five genes under consideration may be arranged in the order of d_4 - wx - dp - C - ws . They also reported that *Pla*, the gene for purple leaf apex and margin, belonged to this linkage group, but its locus was not shown in their map (1947, 1960). According to them, the recombination values between *Pla* and wx , and *Pla* and C were calculated to be about 45 and 21 % respectively, and the linear order of these genes was wx - C -*Pla*. Comparing with our results, it is inferred that *Pla* may be located near to ws .

JODON (1940) showed the linkage relation among v (virescent), Cl (Clusted spikelets), gu (glutinous endosperm, the same as wx) and As (C), and arranged them in the order of gu - As - v - Cl . The recombination value between As and v was found to be 24.05 %, which is almost equal to that of C and ws obtained by us. This fact suggests either ws and v are located near to each other or they identical.

According to TAKAHASHI (1936), d , the gene for sterility is linked with u , glutinous endosperm, with the recombination value of 2.2 %. HARA (1947) also presented that sf , the gene for male sterile mutant derived from the variety Fukukame No. 92, was linked with C and am , glutinous endosperm, and that the recombination values of sf - am , sf - C and am - C were 5.3, 18.9 and 21.7 %, respectively. From these data he concluded that the three genes might be arranged in the order of am - sf - C , and sf was perhaps identical with d symbolized by TAKAHASHI. Above two reports show the possibility that one of the genes for male sterility may be located near to wx and dp .

Studies are being continued to determine the loci of some of these genes and to make up this linkage map in detail.

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イネの第1連鎖群に属する遺伝子の連鎖研究

永松土巳・大村 武

長尾・高橋は C , wx , d_4 の3遺伝子からなる第1連鎖地図を作成している。筆者らは、これら3遺伝子の他に偏顕遺伝子 dp , 縞遺伝子 ws が本群に属することを明らかにした。結果の概要は次の通りである。

1. wx と dp の組換え価は 2.23% で、両遺伝子が極めて近接していることを示している。

2. $dp-C$, $wx-C$ の組換え価は、それぞれ 21.33, 23.17% であつた。これらの結果から3遺伝子の配列順序を $wx-dp-C$ と推定したが、 $wx dp$ との組換え価が小である

ため $dp-wx-C$ の可能性もあるので、この点に関しては目下検討中である。

3. ws は C , dp および wx とそれぞれ 20.08, 31.62 および 33.83% の組換え価を示した。したがって、4遺伝子の配列順序は $wx-dp-C-ws$ と考えられる。

4. 以上の成績を、本連鎖群に関する既報の成績と比較し、若干の考察を試みた結果、さらに2,3の遺伝子が本群に所属すると考えられるので、研究を続行し改めて報告する。