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Iwata, Nobuo Plant Breeding Laboratory, Faculty of Agriculture, Kyushu University

Satoh, Hikaru Plant Breeding Laboratory, Faculty of Agriculture, Kyushu University

Omura, Takeshi Plant Breeding Laboratory, Faculty of Agriculture, Kyushu University

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Studies on the Trisomics in Rice Plants (*Oryza sativa* L.) VII. Some Marker Genes Located on Chromosome 2 and Their Sequence on the Linkage Map

Nobuo Iwata, Hikaru Satoh and Takeshi Omura

Plant Breeding Laboratory, Faculty of Agriculture, Kyushu University, Fukuoka 812, Japan (Received September 28, 1984)

Using trisomic method, some genes located on chromosome 2 were detected, and their gene sequence on the chromosome was inferred. A single recessive gene for a bright green leaf mutant, *bgl*, was newly described in this report.

INTRODUCTION

Nagao and Takahashi (1963) tentatively proposed twelve linkage groups which corresponded to a haploid chromosome number of rice (n=12).

By the use of trisomic method, Iwata and Omura (1976) suggested a possibility that three linkage groups VI, IX and XII were located on the same chromosome, chromosome 2, which is the extra chromosome of trisomic type L.

By the use of reciprocal translocations involving chromosome 2, Sato et al. (1982) found the linkage relations between nl-I, a marker gene of linkage group IX, and both d-l and gl-I, marker genes of linkage groups VI and XII, respectively.

Yoshimura *et al.* (1982) also confirmed by the reciprocal translocation method that d-l, marker gene of linkage group VI, and nl-I and ri, marker genes of IX, were located together on chromosome 2.

In the present study, a new marker gene located on chromosome 2 was found by trisomic analysis, and a sequence of genes on chromosome 2 was inferred.

MATERIALS AND METHOD

Marker genes used in this study were d-l, nl-2 and ops of linkage group VI, al-K-2, *al*-K-3, *nl*-1, *ri*, spl-7 and *spl*-8 of linkage group IX, *gl*-1 of linkage group XII, and *bgl* described newly in the study (Table 1). A mutant for bright green leaf was induced by treating fertilized egg cells with N-methyl-N-nitrosourea (MNU) solution and designated as *bgl*. It was single recessive to the normal. Trisomic type L (Iwata *et al.*, 1970) was used for the trisomic analysis to find the genes located on chromosome 2. Recombination values

Linkage ¹⁾ group	Gene ²⁾ symbol	Character	Reference		
VI	d - 1 nl-2 ops	daikoku dwarf neck leaf 2 open spikelet sterile	Nagao and Takahashi (1963) Iwata and Omura (1977) ″ (1971)		
IX	al-K-2 al-K-3 nl-1 ri spl-7 spl-8	albino Kyushu 2 albino Kuyshu 3 neck leaf 1 verticillate rachis spotted leaf 7 spotted leaf 8	Iwata and Omura (1978) " Nagao and Takahashi (1963) Iwata <i>et al. (1978)</i> "		
XII	gl-I	glabrous leaf and hull 1	Nagao and Takahashi (1963)		
	bgl	bright green leaf			

Table 1. List of marker genes used and their linkage groups.

1) By Nagao and Takahashi (1963)

2) Although the standardization of gene symbols in rice has been discussed and new symbols are recommended for some genes by Japanese Committee on Nomenclature and Linkage Group of Rice, original symbols are used in this study to avoid the confusion, because the symbols recommended are still not authorized.

were calculated in F₂ and F₃ by the mehtod of maximum likelihood.

RESULTS AND DISCUSSION

Segregation of marker genes in F_2 of the crosses with trisomic type L were shown in Table 2. Three marker genes of linkage group VI, d-l, nl-2 and ops, two of IX, nl-I and ri, and one of XII, gl-1, showed the trisomic segregation. Regarding bgl, the segregation mode of dominant 63: recessive 1 was observed in the F_2 with type L and it was also considered to be the trisomic segregation. It is reasonably expected, therefore, that these seven genes compose a linkage group located on chromosome 2.

Linkage relations were observed between bgl and some marker genes belonging to linkage groups VI and IX (Table 3). From F_2 and F_3 data, the weighted means of recombination values on bgl—d-l, bgl-ops, bgl—ri, bgl—spl-7and bgl-spl-8 were estimated at 34.2%, 12.6%, 2.9%, 16.1% and 15.4%, respectively. These facts show that the bgl links closely with ri of linkage group IX and also links with both d-l and ops of linkage group VI. It is obvious, therefore, that both linkage groups VI and IX which have been proposed by Nagao and Takahashi (1963) are combined in one group through the bgl gene.

The linkage relations among some marker genes belonging to the linkage groups VI or IX were investigated (Table 4). Recombination values of nl-2- d-l, nl-2—ops and d-l-ops were estimated at 22.3%, 44.0% and 30.7%, respectively, suggesting a sequence of nl-2-d-l—ops.

A recombination value of *spl-8*—*spl-7* was 29.6%. From this linkage intensity and the linkage intensities of *bgl-spl-7* (16.1%) and *bgl—spl-8*(15.4%)men-

Linkage group	Gene	O	χ^2		
		Dominant	Recessive	Total	for 3: 1
VI	d-1	390	48	438	46.05***
	nl-2	477	32	509	95.06***
	ops	402	36	438	65.78***
IX	nl–1	281	24	305	47, 74***
	ri	424	16	440	107.10***
XII	gl-1	410	24	434	87.75***
	bgl	63	1	64	18.75***

Table 2. Trisomic segregation of the marker genes belonging to three linkage groups and of a marker gene described newly, in F_2 of crosses with trisomic type L.

***: Significant at 0.1% level.

These data are partially quoted from Iwata and Omura (1976).

Table 3. Linkage relations between bgl and five genes belonging to the linkage groups Vi or IX.

Gene pair	Source of data E.	Phasen	Segregation*)	Total	Recombination value (%)	Weighted mean of recombination value (%)
bgld-1	$F_{3}(Ab)$	R	326: 130: 115: 1	5 586	34.7 ± 3.6	
0		R	47: 41	88	30.4 ± 4.5	
	F3(aB) F3	R	37: 28	65	39.8±6.0	34. 2±2.5
bgl—obs	F _a (Ab)	R	108: 49: 42: 0	199	≑ 0	
-8. 1	F	R	38: 11	49	12.6 ± 3.8	
bgl—ri	$F_{3}(Ab)$	R	228: 91: 61: 0	380	- <u>+</u> =0	
0	. ,	R	84: 4	88	2.3f1.2	
	F ₃ (aB)	R	53: 5	58	4.5 ± 2.0	2.9 ± 1.0
bgl—spl-7	F,	R	390: 194: 171: 5	760	16.5 ± 3.5	
0.	$F_3(Ab)$	R	68: 23	91	14.5 ± 3.0	
	F _a (aB)	R	63: 27	90	17.6±3.3	16.1±1.9
bgl—spl-8	F ₂	R	191: 94: 91: 3	379	17. 6f4.9	
	F ₃ (Ab)	R	70: 24	94	14.6 ± 3.0	15.4 ± 2.5

1) R=Repulsion phase

2) $F_2=AB$: Ab: aB: ab. $F_3(Ab)=AAbb$: Aabb. $F_3(aB)=aaBB$: aaBb.

tioned before, the sequence of *spl-8*, *spl-7* and **bgl** is considered as *spl-7*---bgl---spl-8.

Recombination values of *spl-7*—*nl-1*, *ri*—*spl-7* and *ri*—*nl-1* were estimated at 10.4%, 14.7% and 30.7%, respectively. The sequence of the three genes, thus, should be ri—*spl-7*—*nl-1*. The genes **bgl** and *ri* linked closely (2.9%) as mentioned above, therefore, a gene sequence of *spl-8*—*bgl*—*ri*—*spl-7*—*nl-1* or *spl-8*—*ri*—*bgl*—*spl-7*—*nl-1* is considered at this group. Moreover, based on the

Gene pai	Source r of data	Phase ¹⁾	Segregation ²⁾	Total	Recombination value (%)	Weighted mean of recombination value (%)
nl-2-d-l	F_2	R	398: 163: 194: 9	764	22.3 ± 3.0	
nl-2—ops	F ₂	R	212: 80: 77: 19	338	44. 0±4.1	
d–1–ops	F ₂	С	573: 142: 127: 104	946	34.1±2.0	
F F	F ₂ F ₃ (Ab)	R R	294: 27: 24 120: 112: 2	528 51	30.8±6.014.8±4.2	30.7fl.7
d-1-spl-7	F ₂	R	106: 47: 41: 9	203	40. lf5.8	
spl-8—spl-7	$F_2 F_2$	C R	224: 58: 50: 47 107: 49: 56: 1	379 213	33.0 ± 3.1 13.6 ± 6.7	29.6F2.8
ri-spl-7	F ₂ F ₃ (Ab) F ₃ (aB)	R R R	198: 91: 98: 1 53: 14 43: 28	388 67 71	10. 4 ± 5.0 11. 7 ± 3.1 24. 6 ± 4.5	14.7±2.3
ri-nl-1	F_2 F_2	<i>с</i> R	241: 48: 45: 54 220: 84: 93: 18	388 415	27.3 ± 2.7 40.5 ± 4.1	30. 7±2.1
spl-7-nl-1	F ₂ F ₃ (Ab) F ₃ (aB)	R R R	194: 102: 92: 0 34: 11 56: 11	-388 45 67	$0 \\ 13.9 \pm 4.2 \\ 8.9 \pm 2.7$	10.4 ± 2.3

Table 4. Linkage relations among the marker genes belonging to the linkage groups VI or IX.

1) R=Repulsion phase. C=Coupling phase.

2) F2=AB: Ab: aB: ab. F3(Ab)=AAbb: Aabb. F3(aB) =aaBB: aaBb.

Table 5. Linkage relations between two albino genes belonging to the linkage group IX and three marker genes locating on chromosome 2.

Gene pa	Source ir of data	Phase	Segregation')	Total	Recombination value (%)	Weighted mean of recombination value (%)
al-K-2—spl	$\begin{array}{c} -7 F_{3}(Ab) \\ F_{3}(AB) \end{array}$	R R	52: 14 17: 107	66 124	11.9 ± 3.1 15.0 ± 3.8	13.1±2.4
al–K–3—nl–	2 F ₃ (Ab) F ₃ (AB)	R R	46: 51 26: 90	97 116	35.7f4.7 26.8±6.0	32.3±3.7
al-K-3-d-l	F ₃ (Ab) F ₃ (AB)	R R	91: 38 52: 170	129 222	17.3±2.3 28.4±4.6	20.2±2.4

1) $F_3(Ab)=AAbb: Aabb. F_3(AB)=AAB_:AaB_.$

linkage intensities of bgl-d-1 (34.299, bgl-ops(12.6%), d-1-spl-7 (40.1%) and so on in Table 3 or 4, the gene sequence of d-I-spl-S-ops-bgl-spl-7 was tentatively proposed, although a three point analysis was essential to get the exact order of these genes on the map.

Two albino genes, *al-K-2* and al-K-3, which linked to *nl-1* with recombination values of 1.1% and 34.6% respectively have been reported by Iwata and

Omura (1978). In the present study, the linkage relations between the albino genes and some marker genes located on chromosome 2 were observed (Table 5). Weighted mean of recombination values of al-K-2-spl-7 was estimated at 13.1% and a gene sequence of either spl-7-nl-1-al-K-2 or spl-7-al-K-2 -nl-1 is considered. The al-K-3 was linked to nl-2 and d-1 with the recombination values of at 32.3% and 20.2%, respectively, suggesting the gene sequence of nl-2-d-I-al-K-3. The gene arrangement of al-K-3 and spl-8 at the map may be expected as d-I-al-K-3-spl-8-&l, however, further investigation will be necessary to confirm it.

Based on the results of the present study, the sequence of the ten genes on the chromosome 2 can be drawn tentatively as shown in Fig. 1.



Fig. 1. A linkage map of 10 genes on chromosome 2

Thus, the linkage group corresponded to the chromosome 2 was reconstructed with the marker genes of the two linkage groups which have been proposed tentatively by Nagao and Takahashi (1963).

Sato *et al.* (1982) estimated the gene sequence of d-l-nl-l-gl-l on the chromosome 2 from the facts that some interchanged points of reciprocal translocation lines involving chromosome 2 linked with both d-l and nl-l, and others linked with both nl-I and gl-l. Although ten genes were found to locate on the chromosome 2 in the present paper, none of the linkage relations between these genes and gl-l were found. To find out the genes linked with gl-l, trisomic analysis of mutant lines has been conducting by the use of type L.

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