INHERITANCE OF PURPLE LEAF COLOR FOUND IN INDICA RICE

—Genetical studies on rice plant, XCIV[1]—

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Received January 17 1986

Introduction

Inheritance studies of anthocyanin coloration was initiated at the dawn of rice genetics and the data has been accumulated by many workers[2,7,10,17,21]. An elaborate genic system was first established by NAGAO and TAKAHASHI[11, 13] based on comprehensive crossing experiments using Japonica rice.

On the other hand, Indian workers advocated many coloration genes on the respective organs and the linkage relationships were considered among them. As a result, there is some discrepancy of the genes postulated between them and further research is needed to confirm the particular gene by the actual crossings even if the same gene symbol is assigned to the coloration.[5]

In this paper, the authors intended to carry out the genic identification of the coloration of leaf blade and sheath by using the fully purple strain in Indica which were introduced from the International Rice Research Institute in the Philippines.

Before going further, the authors wish to extend their sincere appreciation to Dr. T. T. CHANG, Geneticist, IRRI for the seeds of the purple leaf strain.

Materials and Methods

For the genic identification, three types of purple leaf color were used in the experiments and the pattern of their colorations are shown in Table 1 and Plate I.

These colorations are dependent on the pleiotropic actions of the distri-

[J. Fac. Agr. Hokkaido Univ., Vol. 62, Pt. 4. 1985]

1) Contribution from the Plant Breeding Institute, Faculty of Agriculture, Hokkaido University, Sapporo, Japan

buting genes under the co-existence of higher rank of alleles at *C* and *A* loci. *Pl* type is a popular purple leaf type called as "Murasaki-ine" in Japonica. Pl^i type closely resembles the *Pl* type except for non-coloration of leaf collar (juncture between leaf blade and sheath) and pulvinus (stem node). Pl^w type is characterized by a purple wash leaf blade accompanied by purple pericarp

TABLE 1. List of the strains used in the experiment

Strain	Leaf color	Genotype (leaf color)	Source
A-5 Akamuro	Green	$C^{Br}A$	Hokkaido variety
A-31 Fukoku	Green	$C^{Bm}A^+$	do.
A-32 Furenbozu	Green	$C^{Bm}A^+$	do.
A-58 Kokushokuto-2	Green and purple margin	$C^BA\ Pn$	do.
A-133 Norin 9 go	Green	$C^{Bm}A^d$	do.
Taichung 65	Green	C^BA^+	Introduced from Taiwan
T65 *lg*	Green	C^BA^+	Isogenic line of Taichung 65
N-70 Toyohikari bunwai	Green	$C^{Bm}A^+$	Mutant from Toyohikari
H-21	Green	$C^{Bm}A^+$	Linkage tester
L-34 Intermediate dwarf	Green		Jodon's tester 7975
I-9 Yakei-mochi	Green		Introduced from China
I-32 Karalath	Green	$C^{Bk}A$	Introduced from India
[*ms-bo*] A-133 Norin 9 go	Green	$C^{Bm}A^d$	Cytoplasmic male sterile line
[*ms-bo*] A-136 Shiokari	Green	$C^{Bm}A^d$	do.
H-100	Purple	$C^{Bp}A\ Pl$	Linkage tester
H-126	Purple	$C^{Bp}A\ Pl$	do.
H-127	Purple	$C^{Bp}A\ Pl$	do.
H-406	Purple wash	$C^{Bp}A\ Pl^w$	do.
H-409	Purple wash	$C^{Bp}A\ Pl^w$	do.
I-92 Wu-no-tao	Green	$C^{Bp}A\ Pl^w$ *I-Pl*	Introduced from Taiwan
I-102 Fully purple	Purple		IRRI Acs. No. 100835
H-478	Purple		Progeny of the cross, A-58×I-102
MA1-96	Purple		Progeny of the cross, H-79×H-478

TABLE 2. Pattern of anthocyanin coloration in the three purple leaf types, *Pl*, Pl^w and Pl^i

Type	Leaf						Stem		Pericarp
	Blade		Sheath	Collar	Auricle	Ligule	Pulvinus (node)	Internode	
	Early	Late							
Pl-type	+	++	++	+	+	+	+	±	
Pl^w- »	++	+	++		++	+		++	+
Pl^i- »	+	++	++			+		±	

and deeply colored internodes. According to NAGAO *et al*,[15] the genes Pl^w and *Pl* together with Pl^+ consist of multiple alleles showing codominancy between *Pl* and Pl^w.

The strains used in the crossing experiments are shown in Table 2. H-478 and MA1-96 were produced by cross breeding using I-102 or H-478 and belong to Pl^i type. Two cytoplasmic male sterile strains were used for backcrossing of F_1s from the crosses involving Pl^i type. For the distinct expression of coloration, most of the F_2 and B_1 populations were grown in the paddy field located at Sapporo and Kanagi-machi in Aomori prefecture.

Results

1. Mode of inheritance

I-102 Fully purple was crossed with several strains of both Japonica and Indica. Based on the assumption by NAGAO and TAKAHASHI,[11] the genotype of apiculus color of I-102 was estimated as $C^{Bp}AP$. As shown in Table 3, F_2 segregations from the crosses satisfied the expected ratios due to the segregation at *C* and *A* loci. Although it is known that the

TABLE 3. F_2 segregations of apiculus color in the crosses involving I-102 Fully purple

Cross	Gene concerned	Apiculus color		Total	Goodness of fit (3:1 or 9:7)		
		Purple	White or pink		χ^2	d. f.	P
A- 5×I-102	*C*	233	69	302	0.746	1	0.3-0.5
N- 70×H-476	*C*	192	60	252	0.190	1	0.5-0.7
I- 9×I-102	*A*	185	67	252	0.339	1	0.5-0.7
I-102×A- 32	*C*, *A*	205	150	355	0.326	1	0.5-0.7
H- 21×I-102	*C*, *A*	261	155	416	0.006	1	0.9-0.95

F_2 segregation from Japonica × Indica cross shows some distortion due to hybrid sterility[16] and gametophytic selection,[4,9] no-irregularity was found in the F_2 segregation regardless of the high sterility in some F_1s.

As for the leaf color, most of the F_1s from the crosses indicated green leaf blade and sheath except for the pleiotropic effects of the gene *Pn* which expresses purple tip, margin and collar of the leaf in F_1 of the cross, A-58 × I-102. In F_2, only colored apiculus plants due to $C^BA\,P$ or $C^{Bp}A\,P$ were extracted and classified into green and fully purple leaves. F_2 segregations were examined depending on the genic postulations, namely 1 : 3 in the monogenic ratio and 3 : 13 in the inhibitory ratio. Eight crosses fitted to 3 : 13 of purple : green indicating the interaction of a distributing gene and an inhibitor, while I-32 × I-102 showed a better fit to 1 : 3 showing monogenic segregation (Table 4). Although the results of genic identification and allelism will be explained in the next section, the new color-distributing gene and the inhibiting gene are temporarily designated as Pl^i and *I-Pl-6*, or *I-Pl*$_6$, respectively.

TABLE 4. F_2 segregations of leaf color in the crosses involving I-102 Fully purple

Cross	Leaf blade and sheath		Total	Goodness of fit			
				1 : 3		3 : 13	
	Purple	Green		χ^2	P	χ^2	P
A- 5 × I-102	30	203	233	18.268	<0.001	5.278	0.02-0.05
A- 31 × I-102	46	221	267	8.601	<0.01	0.406	0.5 -0.7
A- 58 × I-102	72	318	390	8.892	<0.01	0.021	0.8 -0.9
N- 70 × I-102	35	157	192	4.694	0.02-0.05	0.034	0.8 -0.9
H- 21 × I-102	41	220	261	12.017	<0.001	1.585	0.2 -0.3
L- 34 × I-102	81	303	384	3.125	0.05-0.10	1.385	0.2 -0.3
I- 9 × I-102	30	155	185	7.613	<0.01	0.780	0.3 -0.5
I-102 × A- 32	30	175	205	11.748	<0.001	2.280	0.1 -0.2
I- 32 × I-102	136	382	518	0.435	0.5 -0.7	19.151	<0.001

The genic assumption was tested by raising F_3 families from both purple and green plants in F_2 (Table 5). In A-58 × I-102, the justification of the inhibiory ratio was demonstrated from the fact that the progeny of some of the purple plants segregated into 3 purple : 1 green due to the heterozygosity of Pl^i. A segregation of F_3 families was close to the expected ratio. On the other hand, a monogenic segregation of F_3 families was confirmed

in the cross, I-32×I-102. Thus the new genic scheme of the purple leaf color was dependent on the interaction of the distributing gene *Pl*^{i} and its inhibitor *I-Pl-6*. It is reasonable that the purple leaf color is expressed by *Pl*^{i} in co-existence with high rank of alleles at *C* and *A* loci and *I-Pl-6* derived from both Japonica and Indica strains suppresses completely the color expression due to *Pl*^{i}.

TABLE 5. Progeny test in F_3 families depending on the genic postulation on the inheritance of leaf color in the crosses, A-58×I-102 and I-32×I-102

a. A-58 (*++ I-Pl$_6$I-Pl$_6$*)×I-102 (*Pl^{i}Pl^{i} ++*) F_3

F_2		F_3 segregation observed		Theoretical	
Phenotype	Genotype	Mode	Obs.	Ratio	Number
Purple	*Pl^{i}Pl^{i} ++*	Bred true to purple	3	1	2.7
	Pl^{i} + ++	Segregation for 3:1 (purple:green)	4	2	5.4
Green	*Pl^{i}Pl^{i} I-Pl$_6$+* or *Pl^{i}+ I-Pl$_6$+*	Segregation for 1:3 or 3:13 (purple:green)	13	6	16.1
	Pl^{i}Pl^{i} I-Pl$_6$I-Pl$_6$ *Pl^{i}+ I-Pl$_6$I-Pl$_6$* *++ I-Pl$_6$I-Pl$_6$* *++ I-Pl$_6$+* *++ + +*	Bred true to green	23	7	18.8
		Total	43		
		χ^2	1.926		
		d.f.	3		
		P	0.5–0.7		

b. I-32 (*Pl^{i}Pl^{i} I-Pl$_6$I-Pl$_6$*)×I-102 (*Pl^{i}Pl^{i} ++*) F_3

F_2		F_3 segregation observed		Theoretical	
Phenotype	Genotype	Mode	Obs.	Ratio	Number
Purple	*Pl^{i}Pl^{i} ++*	Bred true to purple	12	1	11.75
Green	*Pl^{i}Pl^{i} I-Pl$_6$+*	Segregation for 1:3 (purple:green)	20	2	23.50
	Pl^{i}Pl^{i} I-Pl$_6$I-Pl$_6$	Bred true to green	15	1	11.75
		Total	47		
		χ^2	1.426		
		d.f.	2		
		P	0.2–0.3		

2. Identification of purple leaf color genes

a. *Pl* and Pl^i

According to NAGAO *et al.*,[14] *Pl* and Pl^w are responsible for the various coloration of leaf blade and sheath, through co-existence with the basic color-

TABLE 6. F_2 segregations of the crosses involving *Pl* and Pl^i types

Phenotype	Leaf blade	Patrially colored	Fully colored	Fully colored	Total	Goodness of fit		
	Leaf sheath	Colored	Colored	Colored				
	Collar and pulvinus	Colored	Colored	Non-colored		χ^2	d.f.	P
	Color type	Suppressed *Pl*	*Pl*	Pl^i				
Genotype		*Pl I-Pl*	*Pl* +	Pl^i, *I-Pl* or +				
H-126 (*PlPl* + +) × H-478 (Pl^iPl^i *I-PlI-Pl*)	Obs.	166	42	49	257	7.593	2	0.02-0.05
	C. R.	9	3	4				
	Cal.	144.56	48.19	64.25				
MA1-96 (Pl^iPl^i + +) × H126 (*PlPl* + +)	Obs.		269	98	367	0.568	1	0.3 -0.5
	C. R.		3	1				
	Cal.		275.25	91.75				

TABLE 7. Progeny test of the allelism between *Pl* and Pl^i in the crosses, H-126×H-478 and MA1-96×H-126

F_2		Segregation of F_3 families				
			H-126×H-478		MA1-96×H-126	
Color type	Genotype	Mode	Obs.	Ratio	Obs.	Ratio
Pl	*Pl Pl* + +	Bred true to *Pl*	11	1	13	1
	Pl Pl^i + +	*Pl* : Pl^i = 3 : 1	27	2	46	2
Suppressed *Pl*	*Pl Pl I-Pl I-Pl*	Bred true to S-*Pl*	29	1		
	Pl Pl I-Pl +	*Pl* : S-*Pl* = 1 : 3	28	2		
	Pl Pl^i *I-Pl I-Pl*	S-*Pl* : Pl^i = 3 : 1	49	2		
	Pl Pl^i *I-Pl* +	S-*Pl* : *Pl* : Pl^i = 9 : 3 : 4	55	4		
Pl^i	Pl^i Pl^i *I-Pl I-Pl*	Bred true to Pl^i	43	4	20	1
	Pl^i Pl^i *I-Pl* +					
	Pl^i Pl^i + +					
		Total	242		79	
		χ^2	31.554		3.381	
		P	<0.001		0.1-0.2	

producing genes. Besides that, three inhibitors *I-Pl-1*, *I-Pl-2* and *I-Pl-3* depress partly the coloration of leaf blade due to *Pl* and Pl^w genes.

Two Pl^i type testers were crossed with H-126 having the genotype, $C^{Bp}A$ *Pl*. F_1 of the cross, H-126×H-478 showed partial coloration of leaf blade and sheath which corresponds to the genotype *Pl I-Pl*, while F_1 of the cross, MA1-96×H-126 remained in purple leaf color, suggesting the difference of genotypes between H-478 and MA1-96.

As shown in Table 6, no plant with colorless leaf is segregated in F_2s. This indicates that the locus of Pl^i may be identical with that of *Pl*. An inhibitor *I-Pl* derived from H-478, depressed the coloration of *Pl* and was identified as one of the three inhibitors, *I-Pl-1*, *I-Pl-2* and *I-Pl-3*. In contrast with this, the coloration of Pl^i was not affected by *I-Pl*. Through the segregations, *Pl* behaved as dominant over Pl^i. As shown in Table 7, the results in F_3 of the two crosses, H-126×H-478 and MA1-96×H-126 were in conformity with the expectations and the allelic relation was also sustained except a poor fitness in the F_3 ratio. In B_1 generation of the cross, (I-102×H-127) F_1×A-133 the monogenic segregation due to $Pl : Pl^i$ was confirmed under the presence of inhibitors for *Pl* and Pl^i (Table 8).

TABLE 8. Segregation of leaf color in B_1 generation of the cross (I-102×H-127) F_1×A-133

Color type	Suppressed *Pl*	Green	Total	Goodness of fit		
Genotype	*Pl*+ *I-Pl*[1)]	+Pl^i *I-Pl*$_0$[2)]		χ^2	d. f.	P
Obs.	252	61	313	0.259	1	0.5-0.7
C. R.	3	1				
Cal.	234.75	78.25				

1) *I-Pl I-Pl* or *I-Pl* +.
2) *I-Pl*$_0$ +.
3) Genotype: I-102 $Pl^i Pl^i$ *I-Pl I-Pl* ++
H-127 *Pl Pl* ++ ++
A-133 ++ *I-Pl I-Pl I-Pl*$_0$ *I-Pl*$_0$.

b. Pl^w and Pl^i

In the previous paper, Pl^w behaved as an allele of the *Pl* locus and the histological locations of anthocyanin pigment due to the both genes were different showing co-dominancy in F_1.[15)] The pleiotropic action of Pl^w was suppressed by the cooperative action of three genes *I-Pl-1*, *I-Pl-2* and *I-Pl-3* for leaf color and the other inhibitors, *I-Pl-4* and *I-Pl-5* affected the coloration of pericarp.[4)]

TABLE 9. F_2 segregations of leaf color in the crosses involving Pl^w and Pl^i types

Phenotype		Leaf blade	Partially colored	Purple wash	Fully colored	Total	Goodness of fit		
		Leaf sheath	Colored	Colored	Colored				
		Collar	Non-colored	Non-colored	Non-colored		χ^2	d.f.	P
		Color type	Suppressed Pl^w	Pl^w	Pl^i				
Genotype			$Pl^w I$-Pl	Pl^w +	Pl^i, I-Pl or +				
H-478 ($Pl^i Pl^i$ I-$Pl I$-Pl) × H-406 ($Pl^w Pl^w$ + +)	Obs.	135	26	51	212	6.912	2	0.02–0.05	
	C. R.	9	3	4					
	Cal.	119.25	39.75	53.00					
H-406 ($Pl^w Pl^w$ + +) × MAl-96 ($Pl^i Pl^i$ + +)	Obs.		204	59	263	0.924	1	0.3 –0.5	
	C. R.		3	1					
	Cal.		197.25	65.75					

TABLE 10. Progeny test of the allelism between Pl^w and Pl^i in the cross, H-406 × MAl-96

F_2		Segregation of F_3 families		
Color type	Genotype	Mode	Obs.	Ratio
Pl^w	$Pl^w Pl^w$	Bred true to Pl^w	68	1
	$Pl^w Pl^i$	$Pl^w : Pl^i = 3 : 1$	115	2
Pl^i	$Pl^i Pl^i$	Bred true to Pl^i	51	1
		Total	234	
		χ^2	2.538	
		d. f.	1	
		P	0.2–0.3	

As mentioned in the previous section, it was disclosed that H-478 possesses an inhibitor for Pl^w and MA1-96 has no inhibitor regardless of the same coloration. As shown in Table 9, the allelic relation between Pl^w and Pl^i was supported by the non-appearance of green leaf plant. Only the coloration due to Pl^w was partly suppressed leaving the coloration of tip of blade, auricle, ligule and both fringes (upper and lower margin) of the collar in H-478 × H-406. The segregation ratio, 9 : 3 : 4 due to the segregations of both Pl^w and I-Pl was supported though fitness was not adequate in one of the crosses. In F_3 families, allelic relation between Pl^w and Pl^i was supported as shown in Table 10.

TABLE 11. Segregations of leaf color in B_1 generation of the crosses [*ms-bo*] Shiokari×(H-478×H-406) F_1 and [*ms-bo*] Norin 9 go×(H-478×H-406) F_1

Color type / Genotype		Suppressed *Pl*^{w} *Pl*^{w}+, *I-Pl* + *I-Pl I-Pl*	Green *Pl*^{i}+, *I-Pl*$_6$ +	Total	Goodness of fit		
					χ^2	d.f.	P
[*ms-bo*] Shiokari × (H-478×H-406) F_1	Obs.	83	71	154	0.935	1	0.3-0.5
	C. R.	1	1				
	Cal.	77	77				
[*ms-bo*] Norin 9 go × (H-478×H-406) F_1	Obs.	17	21	38	0.421	1	0.5-0.7
	C. R.	1	1				
	Cal.	19	19				

Genotype: H-478: *Pl*^{i} *Pl*^{i} *I-Pl I-Pl* + +
H-406: *Pl*^{w} *Pl*^{w} + + + +
[*ms-bo*] Shiokari or Norin 9 go: + + *I-Pl I-Pl I-Pl*$_6$ *I-Pl*$_6$

TABLE 12. Occurrence of green plants in the F_2 populations

Cross combi.	Genotype of F_1	Theo. Ratio	Color type			Total
			Suppressed *Pl* or *Pl*^{w}	Fully purple (*Pl*, *Pl*^{w} or *Pl*^{i})	Green	
H-100× I -102	$\frac{Pl\ +}{Pl^i\ I\text{-}Pl}$	9:7	237	138	9	384
H-126×H-478	$\frac{Pl\ +}{Pl^i\ I\text{-}Pl}$	9:7	66	25	2	93
I - 92× I -102	$\frac{Pl^w\ I\text{-}Pl}{Pl^i\ I\text{-}Pl}$	3:1	283	53	1	337

As *I-Pl-4* and *I-Pl-5* inhibited only the coloration of pericarp by *Pl*^{w}, the new inhibitor for *Pl*^{i} must be designated as *I-Pl-6*. In B_1 generation of the two crosses, the selective suppressions by *I-Pl* and *I-Pl-6* were demonstrated as expected (Table 11).

Since the allelic relation between *Pl*^{i} and *Pl*^{w} were established, *Pl*^{i} joins in the multiple allelic series together with *Pl*^{w}, *Pl* and *Pl*$^+$ in which the dominancy, *Pl*^{w}>*Pl*^{i} and *Pl*>*Pl*^{i} were determined in this experiments. As to the inhibitory gene for *Pl*^{i}, the specific inhibitor, *I-Pl-6* corresponds to *Pl*^{i} and the action of the three inhibitors, *I-Pl-1*, *I-Pl-2* and *I-Pl-3* was not expressed for *Pl*^{i}.

In the crossing experiments between I-102 or H-478 and Japonica testers, F_1 plants showed various degrees of hybrid sterility. In F_2 populations of the crosses between purple leaf types, several plants showing green leaf

blade and sheath occurred as shown in Table 12. The causations of the green plants are considered as follows; (1) outcrossings with plants having *I-Pl-6* and (2) occurrence of the rare recombination within the *Pl* locus. In the progeny tests of the green plants, most of the F_3 families segregated into green and purple leaves. According to the off-type feature of F_2 green plants, it is highly plausible that the outcrossings with plants having *I-Pl-6* caused the green leaf plants in F_2. When F_1 and F_2 plants were bagged, no green plants appeared in their progenies. However, the authors also found one case where the green plant in F_2 was bred true to the F_3 family. In such a case, the second hypothesis of the recombination within the *Pl* locus could not be rejected completely.

3. Interrelation with *Pn* (Purple node)

It is already known that the distributing gene *Pn* inserts its effect to the coloration of pulvinus with pleiotropic effects to leaf tip, margin and collar through co-existence with the basic genes, *C* and *A*. F_1 of the crosses indicated the coloration of pulvinus and leaf collar by C^BAPn or $C^{Bp}APn$. Because the F_2 segregation of leaf color was 3 purple : 13 green as mentioned in Table 4 and purple : green nodes segregated into 3 : 1, the theoretical ratio was calculated into 9 : 3 : 39 : 13 depending on the independent relation between both genes.

As shown in Table 13, the independent relation was almost satisfied in the two crosses. It is noted that the purple leaf rice with colored leaf

TABLE 13. Independent relations between Pl^i and *Pn* (Purple node)

Phenotype	Leaf blade and sheath	Fully purple	Fully purple	Partially purple	Green	Total	Goodness of fit		
	Collar and pulvinus	Colored	Non-colored	Colored	Non-colored		χ^2	d. f.	P
Genotype		$Pl^i + Pn$	$Pl^i + +$	Pl^i *I-Pl₆, Pn* / $+$ *I-Pl₆, Pn* or $+ + Pn$	Pl^i *I-Pl₆* $+$, / $+$ *I-Pl₆* $+$, or $+ + +$				
A-58 ($+$, *I-Pl₆, Pn*) × I-102 (Pl^i, $+$, $+$)	Obs.	59	13	256	62	390	6.999	2	0.02-0.05
	C. R.	9	3	39	13				
	Cal.	54.8	18.3	237.7	79.2				
L-34 ($+$, *I-Pl₆, Pn*) × I-102 (Pl^i, $+$, $+$)	Obs.	63	18	226	77	384	1.786	2	0.3 -0.5
	C. R.	9	3	39	13				
	Cal.	54	18	234	78				

collar due to *Pl* were identical with the coloration produced by the combination of Pl^i and *Pn*.

4. Independent relation with some marker genes

In the three cross combinations, three marker genes, *er* (errect growth habit), *Ur-1* (Undulate rachis) and *d-10* (tillering dwarf of Toyohikari bunwai) were combined with the genes, Pl^i and *I-Pl-6*. As shown in Table 14, the expected ratio due to the independent relation between the genes, 9 : 3 : 39 : 13 were sustained in most of the crosses. Although the locus of Pl^i in the second linkage grous is already known, the locus of *I-Pl-6* must be explored.

TABLE 14. Independent relations between the genes for leaf color and marker genes in the three crosses

Cross combi	Gene pair		F_2 segregation				Total	Goodness of fit (9 : 3 : 39 : 13)		
	A[1]	B	AB	Ab	aB	ab		χ^2	d.f.	P
I-102×A- 32	Pl^i *I-Pl₆*	*er*	23	7	142	33	205	6.015	3	0.1 -0.2
H- 21× I-102	Pl^i *I-Pl₆*	*er*	29	12	162	58	261	2.145	3	0.5 -0.7
I-102×A- 32	Pl^i *I-Pl₆*	*Ur-1*	22	2	114	27	165	7.381	3	0.05-0.1
N- 70×H-478	Pl^i *I-Pl₆*	*d-10*	30	5	130	27	192	7.248	3	0.05-0.1

1) Segregation due to the inhibitory genes (3 : 13).

Discussion

In Japanese rice, NAGAO and TAKAHASHI[10,11] carried out extensive gene analysis on the anthocyanin coloration and established a scheme in which *C* produces chromogen and *A* activates *C* and turns the chromogen into anthocyanin. Later, the third gene, *P* was found out to spread the coloration over the apiculus. The intensity and hues of coloration depend on the various combinations of alleles from *C*, *A* and *P* loci which consist of multiple allelic series, respectively.[12,20]

As for the coloration of various organs, the pleiotropic actions of various distributing genes are responsible together with several inhibitors. Thus, *Pl* and Pl^w are assigned for the coloration of leaf blade and sheath and the three inhibitors, interacting with them, suppress the leaf blade color.[14,20]

In this paper, it was disclosed that a new allele, Pl^i joins in the *Pl*-locus and a specific inhibitor, *I-Pl-6* suppresses the action of Pl^i causing the ratio, 3 purple : 13 green leaves in F_2. As for the inhibitory ratios of F_2s,

various ratios such as 13 : 3, 55 : 9 and 229 : 27 of green : purple are mentioned in previous literatures.[1,2,3,6,18,19,22,23] On the basis of the assumption mentioned above, di-, tri-and tetra-genic ratios are given by the interaction of the genes, *C*, *A*, Pl^i and *I-Pl-6*.

From the complete inhibition by *I-Pl-6* for Pl^i, the three genotypes, *Pl I-Pl-6*, + *I-Pl-6* and + + show the green leaf plants. Therefore, it was possible that the F_2 segregation ratio of 3 green : 1 purple occurs from the cross between purple and green types. Inhibitors for *Pl* and Pl^w seldom exist among Japonica strains, while *I-Pl-6* is found to be common throughout Japonica and Indica strains.

Recently it was found that the *C-A-P* gene scheme for anthocyanin coloration in Japonica was also applicable to Indica except for the nature and number of localized genes. For example, any two of the genes, P_a, P_b, and P_c together acted instead of the alleles at *P* locus. Thus, it is needed to establish an integrated genic scheme which is common through Japonica and Indica rice owing to the actual crossings even if the same gene symbol is assigned to the coloration of a respective organ. Pl^i was adovocated as the new distributing gene of anthocyanin coloration together with *Pin-1* for purple internode,[8] by the identification of color characters between Japonica and Indica.

Summary

1. Mode of inheritance were investigated using the purple leaf type of Indica rice which was introduced from the International Rice Research Institute.

2. Under the co-existence of the high rank alleles of the basic genes, *C* and *A*, a distributing gene, Pl^i was responsible for the character and the specific inhibitor *I-Pl-6* for Pl^i was found through Japonica and Indica strains. Thus the F_2 segregation resulted in the inhibitory ratio of 3 purple : 13 green in the category of purple apiculus owing to the interaction between Pl^i and *I-Pl-6*.

3. From the genic identification, it was disclosed that Pl^i is one of the multiple alleles at *Pl* locus showing the dominancy of $Pl > Pl^w > Pl^i > Pl^+$. A suppressive effect of *I-Pl-6* is restricted in Pl^i and inhibits completely the coloration of leaf blade and sheath. There was a specific relationship between purple leaf genes and inhibitors showing the correspondence between Pl^i and *I-Pl-6* and between *Pl* or Pl^w and the three inhibitors, *I-Pl-1*, *I-Pl-2* and *I-Pl-3*.

4. In the crossing between purple and green types, it was demonstrated

that the segregation of *I-Pl-6* under the co-existence of Pl^i causes the ratio of 3 green : 1 purple in F_2. As a consequence, there are three genotypes, Pl^i *I-Pl-6*, + *I-Pl-6* and + + showing the green leaf plants.

5. An independent relationship was confirmed between the set of color genes and *Pn* (Purple node). The combined genotype, Pl^i *Pn* showed the same phenotype due to *Pl*, showing purple leaf blade, collar and sheath.

6. Further research is needed to carry out genic identification by actual crossings even if the same gene symbol is assigned to the coloration of a respective organ.

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Plate I

Anthocyanin coloration due to the distributing genes under the coloration genotype, C^B *A P*.

1. Purple node (pulvinus) due to *Pn*.
2. Purple leaf due to *Pl*.
3. Purple wash leaf and deeply colored internode due to Pl^w.
4. Purple leaf and non-colored collar and pulvinus due to Pl^i.