Genetic Control for Leaf Photosynthesis in Rice, *Oryza sativa* L.

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Inheritance of leaf photosynthesis in rice and relationships between leaf photosynthesis and various characters were studied with infrared analysis of net CO₂ exchange method in a cross experiment between Japanese variety Nakate Shinsenbon(P₁) and USA variety CP-SLO(P₂).

F₁ mean for leaf photosynthesis in terms of apparent photosynthesis (Pa) was very close to P₁ that was the low parent in Pa. F₂ segregation in Pa distributed in a wide range from P₂ to even lower than P₁. Moreover, a bimodal distribution, one mode corresponding to P₁ with a predominant number of plants and the other mode corresponding to P₂ with much less plants, was clearly established. Hence, we concluded that the expression of leaf photosynthesis would be controlled by single major gene and that low leaf photosynthesis is dominant.

Pa significantly correlated negatively with specific leaf area and positively with leaf areal nitrogen content in F₂ progeny. Significant partial correlation existed only between Pa and leaf areal nitrogen content, indicating that areal nitrogen content is the major factor associating with Pa. Still, other undetected factors are indicated to be equally involved in the expression of Pa.

In F₂ progeny, Pa did not correlate with days to heading, culm length, panicle length and length/width ratio of grain. It neither correlated with some characters controlled by single major gene such as esterase isozyme, apiculus color, awn presence, and blade pubescence, indicating that gene(s) controlling Pa may not link with any of them. Besides, a possible effective introduction of glabrous habit into Japanese rice was demonstrated by the absence of any significant difference in seven agronomic characters between glabrous and pubescent F₂ plants.

In conclusion, with its rather simple manner of inheritance and its independency from various agronomic characters, leaf photosynthesis can possibly be increased by intentional selection in future rice breeding.

Introduction

In a series of experiments on efficiencies of solar energy conversion Japanese high yielding rice varieties were high in the efficiencies of utilizing the absorbed energy, but they were not necessarily high in the net assimilation rate measured with plants grown singly (HAYASHI 1968, 1969, 1972). The importance to increase leaf photosynthetic rate and its feasibility in rice breeding were demonstrated in the extensive studies with infrared analysis of net CO₂ exchange on various strains of *Oryza sativa* L. (TSUNODA and TAKANO 1967, TAKANO and TSUNODA 1971).

Despite its importance for growth and yield of crop plants, detailed study on inheritance of leaf photosynthesis has so far been reported only for dry beans, *Phaseolus vulgaris* L. (IZHAR and WALLACE 1967) and for soybean, *Glycine max*(L.) Merrill (OJIMA, KAWASHIMA and MIKOSHIBA 1969). Consequently, we made a cross experiment between a Japanese and

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a USA rice variety chosen from our experience in the previous studies on the efficiencies of solar energy conversion. The objectives of the present study were 1) to study the inheritance of leaf photosynthesis, and 2) to examine relationships between leaf photosynthesis and various characters with a particular concern to their implication for practical rice breeding.

Materials and Methods

Two rice varieties with contrasting characteristics as shown in Table 1 were chosen as the parents. Female parent Nakate Shinsenbon (P_1) was crossed with male parent CP-SLO(P_2) in August 1974. A part of the F_1 seeds were sown in December 1974 in the greenhouse to advance the generation, and F_2 seeds were harvested in April 1975. The seeds of P_1 , P_2 , F_1 and F_2 thus prepared were sown in the outdoor nursery on May 8, 1975. Careful managements including frequent top dressing of nitrogenous fertilizer allowed the plants to grow healthy and vigorously. During the period of most vigorous vegetative growth, sets of five plants each included four F_2 and one P_1 or P_2 or F_1 plants were investigated one after another for their leaf photosynthesis. Sample leaves were two uppermost fully expanded attached leaves per an intact plant. They were in a growth stage one to two leaves below the flag leaf.

Leaf photosynthesis was estimated by infrared analysis of net CO_2 exchange and was expressed in terms of apparent photosynthesis per unit leaf area (hereafter, Pa). The system used to study Pa included channels of air intake, a unit of chambers and an infrared gas analyzer. The air that gave the initial CO_2 content was taken from a large empty glassroom annexed to the laboratory. This air was blown into a storage tank and constantly mixed with a fan to eliminate instantaneous fluctuation of CO_2 content. The air was supplied to the chambers at a rate of 1.5 litre per minute. Six chambers, each had a size of $29.5 \times 9.0 \times 1.8$ cm, composed a unit to accommodate two leaves each of five intact plants. One chamber out of six was used as blank. The whole unit of chambers was

Table 1. Characteristics of parental rice varieties

Character	Parents	
	Nakate Shinsenbon(P_1)	CP-SLO(P_2)
Maturity	medium	late
Culm length	slightly short	short
Tillering	heavy	medium
Flag leaf angle	errect	errect
Blade pubescence	pubescent (gl^+)	glabrous (gl)
Grain type	short (type a) ¹⁾	long (type c) ¹⁾
Apiculus color	white (C^+)	red (C)
Awn presence	slight (An)	absent (An^+)
Esterase isozyme	band 6 A absent (Est_2^0)	band 6 A present (Est_2^s)

1) : After Matsuo (1952)

suspended in a water bath regulated in a temperature range of 27~30°C. The light source was six iodine lamps that gave 80,000 lux at the upper surface of the chambers after passing through the water filter of 10 cm. Sampling air from each chamber to the analyzer was rotated in a period of exactly 50 seconds by electro-magnetic regulators, and CO₂ content at the end of each period was automatically recorded. Differences in CO₂ content between the air from the blank and from the sample chambers were read and used to calculate Pa. The expression of Pa was adjusted at 330 ppm of CO₂, taking the mean of inlet and outlet CO₂ content as the average CO₂ content inside the chamber. The daily measurement of Pa involved a total of 35 to 40 plants in seven to eight series.

The sample leaves were detached after the measurement of Pa and their area was measured by the automatic area meter (MURATA and HAYASHI 1967). Then the leaves were dried at 95°C for 16 hours and weighed. Specific leaf area was expressed in cm² per one gram dry weight of leaf blade. Leaf areal nitrogen content was determined by the CN coder and was expressed in mg nitrogen per dm² of leaf. Electrophoresis of esterase isozyme of leaves was conducted following to the methods described elsewhere (NAKAGAHARA, AKIHAMA and HAYASHI 1975). Other characters were measured by the conventional methods.

Results

1. Inheritance of leaf photosynthesis

Fig. 1 shows the frequency distribution for Pa of P₁, P₂, F₁ and F₂ plants. The expected difference in Pa between the parents was realized, giving a high value of 35.0 for P₂ and a low value of 27.5 for P₁. The mean value for F₁ was 29.4 and was very close to that for P₁ which was the low parent in Pa. For F₂ plants, Pa distributed in a very wide range from as high values as for P₂ to even lower than for P₁. Moreover, a bimodal distribution was clearly established; one mode involving a predominant number of plants corresponded to the value for the low parent (P₁), and the other mode involving much less plants corresponded to the value for the high parent (P₂). Above results in F₁ and F₂ plants would indicate that the expression of leaf photosynthesis is controlled by single

Fig. 1. Frequency distribution (number of plants) for apparent photosynthesis of rice variety Nakate Shinsenbon (P₁), and CP-SLO (P₂), their F₁, and F₂ progenies.

major gene and that low leaf photosynthesis is dominant.

2. Segregation of other agronomic characters

Table 2 gives the mean values and variances of six agronomic characters in F_2 plants, together with the mean values of P_1 , P_2 and F_1 plants. The means of F_1 plants for number of days from seeding to heading and culm length far exceeded that of the parent with higher value, indicating a considerable overdominance. F_1 values for other characters were somewhere between P_1 and P_2 . The F_2 plants distributed in a very wide range far exceeding the both parents in all of the six characters and their frequency distribution was in a manner of quantitative inheritance.

Table 2. Means for P_1 , P_2 , F_1 and F_2 , and variances for F_2 , of six agronomic characters

Generation	Days to heading		Culm length		Panicle length		Grain length/width		Specific leaf area		Areal N content	
	$\bar{x}$	s^2	$\bar{x}$	s^2	$\bar{x}$	s^2	$\bar{x}$	s^2	$\bar{x}$	s^2	$\bar{x}$	s^2
	(days)		(cm)		(cm)		(ratio)		(cm ² ·g ⁻¹)		(mgN·dm ⁻²)	
P_1	120		88		25.8		2.34		248		13.2	
P_2	128		79		23.0		3.18		226		16.1	
F_1	135		111		24.4		2.58		238		13.8	
F_2	127	105.5	93	143.0	22.5	7.67	2.65	0.040	236	526.4	15.2	3.60

3. Correlation between some agronomic characters in F_2 progeny

Table 3 gives correlation coefficients between seven agronomic characters in F_2 progeny. No significant correlation existed between P_a and any of the following four characters, *viz.* days to heading, culm length, panicle length, and length/width ratio of grain. P_a , however, correlated negatively with specific leaf area ($r = -0.658$) and positively with leaf areal nitrogen content ($r = +0.733$), both significant at one percent level. Other highly significant correlations included those between culm length and panicle length and between specific leaf area and leaf areal nitrogen content. The former was rather a common case, but the latter indicated a close association between the leaf characters.

Table 3. Correlations between each of the seven agronomic characters including apparent photosynthesis in F_2 progeny

Character	Apparent photo.	Days to heading	Culm length	Panicle length	Grain length/width	Specific leaf area
Days to heading	-.097					
Culm length	+.011	+.130				
Panicle length	+.042	-.193*	+.455**			
Grain length/width	+.004	+.066	-.106	+.062		
Specific leaf area	-.658**	+.128	+.145	-.044	+.007	
Areal N content	+.733**	+.068	-.171	+.024	+.034	-.790**

*, **, Significant at the 5 and 1% levels of probability, respectively

Partial correlation coefficient, eliminating the effect of specific leaf area, between Pa and leaf areal nitrogen content was +0.461, significant at one percent level. On the other hand, partial correlation coefficient, eliminating the effect of leaf areal nitrogen content, between Pa and specific leaf area was only -0.189 and was not significant. This implies that leaf areal nitrogen content which accounts for about 50 percent of the variation in Pa would be the major factor correlating with Pa.

4. Comparison of Pa between phenotypes of some characters in F₂ progeny

Based on the above findings that Pa would be under major genic control, possible relationship between Pa and some characters each controlled by a major gene was examined for F₂ progeny. However, as shown in Table 4, none of the presence or absence of leaf blade hair (pubescent or glabrous), of esterase isozyme, of red color in apiculus, and of awn, made any significant difference in Pa. This indicates that gene(s) controlling Pa may not link with any gene controlling these four characters.

Table 4. Comparison of apparent photosynthesis between phenotypes for four characters in F₂ progeny

Phenotype	Character			
	Blade pubescence (<i>gl</i>)	Esterase isozyme (<i>Est</i> ₂)	Apiculus color (<i>C</i>)	Awn presence (<i>An</i>)
Dominant	29.4	29.7	29.3	29.2
Recessive	30.2	29.0	29.8	30.3
t-value	1.34 ^{ns}	0.95 ^{ns}	0.44 ^{ns}	1.91 ^{ns}

(mgCO₂·dm⁻²·hr⁻¹)

Of these characters, "glabrous" habit receives an increasing attention particularly as a desirable character for machine harvesting of rice. Additional comparison thus made in Table 5 shows no significant difference in the seven agronomic characters including Pa between the pubescent and glabrous F₂ plants.

Table 5. Means of seven agronomic characters for pubescent and glabrous F₂ plants

Blade pubescence	Agronomic character						
	Apparent photo.	Days to heading	Culm length	Panicle length	Grain length/width	Specific leaf area	Areal N content
	(mgCO ₂ ·dm ⁻² ·hr ⁻¹)	(days)	(cm)	(cm)	(ratio)	(cm ² ·g ⁻¹)	(mgN·dm ⁻²)
Pubescent plants	29.4	127	93	22.5	2.64	236	15.2
Glabrous plants	30.2	126	92	22.3	2.70	236	15.2
t-value	1.34 ^{ns}	0.220 ^{ns}	0.501 ^{ns}	0.605 ^{ns}	1.589 ^{ns}	ns	ns

Discussion

As reported earlier, USA rice varieties usually exceeded Japanese varieties in the net assimilation rate for singly grown plants (HAYASHI 1968). This was supported also in the present study estimating leaf photosynthesis under strong artificial light. The choice of the parents, therefore, was relevant with their big difference not only in leaf photosynthesis but also in various other characters.

The key point of the present paper exists in the study on inheritance of leaf photosynthesis. Not many paper demonstrated positive heterosis of leaf photosynthesis such as for maize (HEICHEL and MUSGRAVE 1969). On the other hand, two papers, one for dry beans (IZHAR and WALLACE 1967) and the other for soybean (OJIMA, KAWASHIMA and MIKOSHIBA 1969), demonstrated quite different results. The F_1 value was significantly lower than the midparent value for the former, or was not significantly different from the low parent for the latter case. In both cases low leaf photosynthesis was considered to be dominant. The present results for F_1 of rice agreed very well to the above results. Moreover, F_2 segregants of rice reported here clearly demonstrated a bimodal distribution for leaf photosynthesis. Consequently, we concluded for the results of the present study leaf photosynthesis would be under a major genic control.

In both cases of dry beans (IZHAR and WALLACE 1967) and soybean (OJIMA, KAWASHIMA and MIKOSHIBA 1969), the observed F_2 distribution was thought to fit normal curve, and hence quantitative inheritance was suggested. This was concluded on the basis of relatively small number of F_2 plants. In the soybean crosses, some parents differed only slightly in leaf photosynthesis. In the present study the number of F_2 plants was increased upto 174, and also the parents differed widely in leaf photosynthesis. These would certainly account for a more clear trend of F_2 distribution demonstrated here. Actually, the both cases of the dry beans and soybean did admit that only a few genes were involved in the inheritance of leaf photosynthesis. More detailed analysis on our results can be made by future backcross experiment.

A very close association between P_a and leaf areal nitrogen content that has been revealed in their highly significant simple and partial correlation coefficients agrees with other results reported earlier (MURATA and OSADA 1959, TAKEDA and KUMURA 1959, TSUNODA and TAKANO 1967, TAKANO and TSUNODA 1971). Similar relationships have been reported also for soybean (OJIMA, KAWASHIMA and MIKOSHIBA 1969) and for wheat and its wild relatives (KHAN and TSUNODA 1970). On the other hand, specific leaf area which was understood as the reverse expression of leaf thickness (FRIEND, HELSON and FISHER 1962, HAYASHI 1968) related with P_a not directly but through its close association with leaf areal nitrogen content. Accordingly, generally accepted advantage of thicker leaves in having higher P_a may exist on the basis that thicker leaves generally have higher content of areal nitrogen. Still, even its pronounced effect on P_a leaf areal nitrogen content accounts for about 50 percent of variation in P_a , implying that some undetected factors other than leaf areal

nitrogen content are equally involved in the expression of Pa.

Independency of Pa from some agronomic characters such as days to heading, culm length, panicle length, and length/width ratio of grain, may have a practical implication for rice breeding. The same applies to another independency of Pa from major genic characters such as esterase isozyme, apiculus color, awn presence, and blade pubescence. In conclusion, with its rather simple manner of inheritance and its independency from various agronomic characters that appeared in the present study, leaf photosynthesis can possibly be increased by intentional selection in future rice breeding.

Besides, no significant difference in seven agronomic characters including Pa between pubescent and glabrous F_2 plants may receive attention for effective introduction of glabrous habit into future rice breeding of Japan. Similar possibility was also reported for B_1F_2 lines of rice derived from a cross between a pubescent Japanese and a glabrous USA variety (FUJIMAKI 1975).

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Literature Cited

- FRIEND, D. J. C., V. A. HELSON and J. E. FISHER 1962. Leaf growth in Marquis wheat, as regulated by temperature, light intensity, and daylength. *Can. J. Bot.* **40** : 1299~1311.
- FUJIMAKI, H. 1975. Genetic variability of some quantitative traits associating with a gene for smoothness in backcross lines of rice. *Japan. J. Breed. Suppl.* **1** : 136~137 (in Japanese with English tables).
- HAYASHI, K. 1968. Response of net assimilation rate to differing intensity of sunlight in rice varieties. *Proc. Crop Sci. Soc. Japan* **37** : 528~533.
- 1969. Efficiencies of solar energy conversion and relating characteristics in rice varieties. *Ibid.* **38** : 495~500.
- 1972. Efficiencies of solar energy conversion in rice varieties. *Bull. Nat. Inst. Agr. Sci.* **D 23** : 1~67 (in Japanese with English tables, figures and summary).
- HEICHEL, G. H. and R. B. MUSGRAVE 1969. Varietal differences in net photosynthesis of *Zea mays* L. *Crop Sci.* **9** : 483~486.
- IZHAR, S. and D. H. WALLACE 1967. Studies of the physiological basis for yield differences. III. Genetic variation in photosynthetic efficiency of *Phaseolus vulgaris* L. *Crop Sci.* **7** : 457~460.
- KHAN, M. A. and S. TSUNODA 1970. Evolutionary trends in leaf photosynthesis and related leaf characters among cultivated wheat species and its wild relatives. *Japan. J. Breed.* **20** : 133~140.
- MURATA, Y. and A. OSADA 1959. Studies on photosynthesis in rice plant. XII. The effect of three major nutrient element supply upon the photosynthetic activity of rice leaves. *Proc. Crop Sci. Soc. Japan* **28** : 184~187 (in Japanese with English tables, figures and summary).
- and K. HAYASHI 1967. On a new, automatic device for leaf-area-measurement. *Ibid.* **36** : 463~467.

- NAKAGAHRA, M., T. AKIHAMA and K. HAYASHI 1975. Genetic variation and geographic cline of esterase isozymes in native rice varieties. Japan. J. Genetics **50** : 373~382.
- OJIMA, M., R. KAWASHIMA and K. MIKOSHIBA 1969. Studies on the seed production of soybean. VII. The ability of photosynthesis in F_1 and F_2 generations. Proc. Crop Sci. Soc. Japan **38** : 693~699 (in Japanese with English table, figures and summary).
- TAKANO, Y. and S. TSUNODA 1971. Curvilinear regression of the leaf photosynthetic rate on leaf nitrogen content among strains of *Oryza sativa* species. Japan. J. Breed. **21** : 69~76.
- TAKEDA, T. and A. KUMURA 1959. Analysis of grain production in rice plant. III. Studies of the effects of various nitrogenous nutrient levels and light conditions on the factors constituting field photosynthetic ability and dry matter productivity in paddy rice. Proc. Crop Sci. Soc. Japan **28** : 175~178 (in Japanese with English tables, figures and summary).
- TSUNODA, S. and Y. TAKANO 1967. The regression between leaf areal nitrogen content and leaf photosynthetic rate in strains of *Oryza sativa* L. and its significance in rice breeding. Japan. J. Breed. **17** Suppl. 2 : 125~126 (in Japanese with English figures).

イネ葉身光合成能力の遺伝様式

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イネの葉身光合成能力の遺伝ならびに光合成能力と他形質との関係を調べる目的で、日本イネ品種中生新千本 (P_1) とアメリカ品種 CP-SLO (P_2) との雑種集団について、炭酸ガス赤外線分析法により、葉身光合成能力を測定した。

光合成能力の F_1 平均値は、低光合成能力親である P_1 に非常に近かった。 F_2 では、高光合成能力親である P_2 から P_1 までの幅広い変異を示し、 P_2 ならびに P_1 付近にモードがあった。後者のモードにふくまれる低光合成能力個体の頻度は、前者の頻度よりもはるかに高かった。以上の F_1 および F_2 における結果から、光合成能力の発現には単一の主働遺伝子が関与している可能性が大であり、また低光合成能力が優性と考えられる。

F_2 集団において、光合成能力は葉面積／葉乾量比 (specific leaf area) と有意な負の、また葉面当たり窒素含量と有意な正の相関を示した。後者の関係のみが偏相関係数においても有意であり、光合成能力と関連する主な要因は葉面当たり窒素含量であることがわかった。しかし、光合成能力の発現には、本試験では明らかにできなかった他の要因も関与することが示唆された。

到穂日数、稈長、穂長、粒の長／幅比のそれぞれと光合成能力との間には、 F_2 集団ではいずれも有意な相関がなかった。また、光合成能力とエステラーゼ同位酵素、稈先色、芒性、葉毛性などの単遺伝形質との間にも有意な相関がなかった。一方、光合成能力をふくめた七つの農業形質においては、有毛性 F_2 個体群と無毛性 F_2 個体群との間に有意差がなく、日本イネに無毛性を効果的に導入することが可能であると思われた。

以上、光合成能力の遺伝様式は比較的単純であり、また種々の農業形質とは独立的であることから、将来のイネ育種における意識的選抜により、光合成能力を増大しうる可能性がある結論される。