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Variation in leaf photosynthetic characteristics in wild rice species

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Abstract

Variations in leaf gas-exchange characteristics, leaf pigment content, and other important leaf traits were investigated in seven wild *Oryza* species, five hybrids and improved varieties. The significant variations were observed in photosynthetic pigment contents amongst different species of *Oryza*. The mean chlorophyll (Chl) content was higher in *O. sativa* (varieties and hybrids), while *O. eichengeri* showed the lowest Chl content. The mean carotenoid (Car) content in *O. sativa* (varieties and hybrids) was higher than in other wild rice species. *O. eichengeri* and *O. barthii* had significantly lower Car contents than other rice species. Significant differences were noticed in the rate of photosynthesis (P_N), stomatal conductance (g_s), transpiration rate (E), internal CO_2 concentration (C_i), specific leaf mass (SLM), and leaf thickness amongst different *Oryza* species. The mean P_N was the highest in *O. nivara* followed by *O. eichengeri.* The mean P_N was the lowest in *O. glumaepatula,* which was lower than that of cultivated varieties and hybrids of *O. sativa*. High rates of photosynthesis were observed in *O. nivara* (ACC. No. CR 100097)*, O. rufipogon* (ACC.No. CR 100267*)*, and *O. nivara* (ACC.No. CR 100008). The *O. nivara* and *O. rufipogon* genotypes with high P_N might be used in rice improvement programmes for an increase of leaf photosynthesis in rice. Multiple correlations performed between different gas-exchange characteristics and other physiological traits revealed that the rate of photosynthesis was not dependent on the leaf pigment content or the leaf thickness. A strong positive correlation between P_N and the P_N/C_i ratio, which represents the carboxylation efficiency, indicated that the observed variation in P_N was not based on pigment content or other leaf traits.

Additional key words: gas exchange; photosynthesis; rice; specific leaf area; specific leaf mass; stomatal conductance; wild rice.

Introduction

Rice (*Oryza sativa.* L.) is one of cereal crops vital for providing food for most countries of the world. Contrary to maize and wheat, a major portion of 600 million t of rice produced per year is consumed directly by humans. Rice production has increased during the past three decades and China, India, Indonesia, Bangladesh, and Vietnam are the top rice-producing countries in the world. The increase in a rice yield was lesser in the recent years (2005–2010), when compared to the period of the green revolution in 1980 (FAOSTAT 2010). It shows clearly that the level of rice production needs to be increased to meet the demand for food due to the increase in population. Since most of the plant biomass is derived from carbon originated from photosynthesis, enhancing the photosynthesis at the level of a single leaf would eventually increase the grain yield (Makino 2011). The further increase in the grain yield of the major crops will depend on improving photosynthetic efficiency (Long *et al*. 2006, Zhu *et al*. 2010). Wild rices are known for their tolerance to biotic and abiotic stresses, high biomass formation, and low nutritional requirements, which were lost in the cultivated rice genotypes (Vaughan 1989, Sitch 1990). Assessment of the photosynthetic efficiency within the genus *Oryza* is essential for improving yield potential; it is necessary to identify donors with high photosynthetic rates and to use them in a wide crossing programme. Variation in photosynthesis and related traits have been studied in wild *Oryza* species (Yeo *et al*. 1994, Zhao *et al.* 2010). The light-saturated assimilation rate in wild germplasm of a number of species was higher than

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Abbreviations: Car – carotenoids; CE – carboxylation efficiency; Chl – chlorophyll; C_i – intercellular CO_2 concentration; E – transpiration rate; g_s – stomatal conductance; SLA – specific leaf area; SLM – specific leaf mass; WUE – water-use efficiency; WUE_i – intrinsic water-use efficiency.
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that of the cultivars and the elite breeding material of *O. sativa* (Yeo *et al*. 1994). These results indicated a significant diversity of photosynthetic traits in genus *Oryza*, which could be exploited for enhancing the yield and breaking the yield plateau in rice (Zhao *et al*. 2010).

Masumoto *et al.* (2004) investigated whether wild rice has genes to improve the photosynthesis of cultivated rice and they concluded that *O. rufipogon* can be used as a source of germplasm to enhance the photosynthetic capacity of *O. sativa*.

The objective of this investigation was to identify variations in leaf photosynthetic characteristics and other related traits in selected wild rice genotypes and popular hybrids and varieties.

Materials and methods

Plant material: Twenty five rice genotypes consisting of seven *Oryza* wild species, two hybrids, and three released varieties (Table 1) were obtained from Plant Breeding section of Directorate of Rice Research (DRR), India. They were grown in earthen pots, 25 cm in a diameter, filled with soil collected from DRR farm. To break dormancy, wild rice seeds were treated with 0.2 M nitric acid followed by repeated washing with distilled water and then they were germinated on a wet filter paper in Petri dishes at 27°C. One-week-old seedlings (2 seedlings/ pot) were transplanted into earthen pots (20 cm in diameter). The pots were kept in a net house under natural sunlight. Irrigation was provided according to the requirement. Each genotype was planted in three pots and each pot was treated as a separate replication; all measurements were performed three times.

Gas exchange: Leaf photosynthetic characteristics were measured between 10–13 h on fully matured leaves at tillering stage (40 d after transplanting) using *LI6400XT* portable photosynthesis measuring system (*LI-COR Environmental*, USA) connected to *Leaf Chamber Fluorometer* (*6400-40, LI-COR*, USA), which was used as a light source. Leaf temperature was maintained at 35°C, which was equal to the ambient temperature prevailing at the time of measurements and PAR was maintained at 1,000 μmol m^{-2} s^{-1}. Measurements were made at ambient CO_2 levels. The mean CO_2 concentration during measurements was 387 μmol mol^{-1}. Leaf photosynthetic pigments were extracted with a mortar and pestle in cold 80% acetone and a pinch of sea sand. The extract was centrifuged at 4°C for 5 min × *g*. Chl and Car contents were determined spectrophotometrically (*Spectrascan UV 2600*,

Table 1. Details of wild rice germplasm used in the study. Wild rice seeds were obtained from rice germplasm collection collected and maintained by Plant Breeding Section of Directorate of Rice Research.

S/N	Species	Acc. No.	Country
1	*Oryza glaberrima*	IR 100983	Nigeria
2	*O. glaberrima*	IR 101800	Liberia
3	*O. glaberrima*	IR 102445	Mali
4	*O. glaberrima*	IR 104020	Tanzania
5	*O. glaberrima*	IR 104033	Chad
6	*O. barthii*	IR 103580	Chad
7	*O. barthii*	IR 80433	India
8	*O. rufipogon*	IR 80774	Myanmar
9	*O. rufipogon*	IR 103404	Philippines
10	*O. rufipogon*	CR 100018	India
11	*O. rufipogon*	CR 100267	India
12	*O. rufipogon*	CR 100309	India
13	*O. nivara*	IR 104650	Thailand
14	*O. nivara*	CR 100008	India
15	*O. nivara*	CR 100097	India
16	*O. nivara*	CR 100100	India
17	*O. longistaminata*	IR 105262	Kenya
20	*O. longistaminata*	IR 104301	Gambia
18	*O. eichengeri*	IR 100881	Srilanka
19	*O. glumaepatula*	IR 104387	Brazil
21	*O. sativa (Moroberekan)*	Tropical upland Japonica variety	West Africa
22	*O. sativa (Akshayadhan)*	Variety	India
23	*O. sativa (Varadhan)*	Variety	India
24	*O. sativa (PA–6444)*	Hybrid	India
25	*O. sativa (DRR–Dhan–38)*	Hybrid	India

Toshniwal Instruments Pvt. Ltd., India) by measuring the absorbance at 663.2 nm (Chl *a*), 646.8 (Chl *b*), and 470 nm (Car). The pigment concentration was calculated according to Lichtenthaler and Wellburn (1983).

Specific leaf mass, specific leaf area, and leaf thickness: Leaf area was measured by using *LI-3100C* electronic leaf area meter (*LI-COR Environmental,* USA). Leaf thickness was measured with a digital calipers (*Model 06-664-16, Fisher Scientifics,* USA). Specific leaf area (SLA) and specific leaf mass (SLM) were calculated from the leaf area and leaf dry mass.

Statistical analysis: One-way analysis of variance was performed on leaf photosynthetic characters and other traits using *Statistix 8.1 (Analytical Software Inc.* USA) software and the statistical significance of the parameter means were determined by performing the *Fisher's* LSD test and standard deviations (SD) were calculated and reported. Multiple correlation between gas exchange traits and related parameters were performed using *Microsoft Excel 2007*.

Results and discussion

Variations in the photosynthetic characteristics of wild rice were studied by gas-exchange measurements and the results were compared with the photosynthetic characteristics of cultivated varieties and hybrids. The significant variation was observed in the photosynthetic pigment content among different species of *Oryza* (Table 2). Chl *a*, Chl *b,* and total Chl content were the highest in DRRH–3, followed by PA–6444 and *O. glaberrima*, ACC. No. IR 104033. The lowest Chl content was found in *O. glaberrima,* ACC. No. IR 101800. The mean Chl content was higher in *O. sativa* (varieties and hybrids), while *O. eichengeri* recorded the lowest Chl. The mean Chl content did not differ significantly in the remaining species (Fig. 1). The total Car content varied between 1.03 (DRRH–3) to 0.49 mg g^{-1}(FM) (*O. glaberrima*, ACC. No. IR 101800) with a mean of 0.79 in all genotypes. The mean Car content in *O. sativa* (varieties and hybrids) was higher than in other wild rice species. *O. eichengeri* and *O. barthii* had significantly lower Car content than other rice species. The differences in Chl *a/b* ratio was found to be insignificant among the tested genotypes (Table 2). The mean Chl *a/b* ratio did not differ significantly between wild rice species and cultivated varieties and hybrids. Significant differences were noticed in total Chl/Car ratio amongst different rice species (Table 2). However, the mean Chl/Car ratio was constant across different wild rice species, hybrids, and varieties (Fig. 1).

Genotypes maintaining higher leaf Chl *a* and Chl *b* during the growth period may be considered as potential donors for their ability to produce higher biomass and photosynthetic capacity (Hassan *et al*. 2009). Significant variations in SPAD values among different *Oryza* species have been reported (Zhao *et al*. 2010).

Significant differences were noticed in P_N, g_s, E, and C_i among different *Oryza* species. P_N varied between 24.2 (*O. nivara*, ACC. No. CR 100097) and 6.79 (*O. glumaepatula*, ACC. No. IR 104387) with a mean of

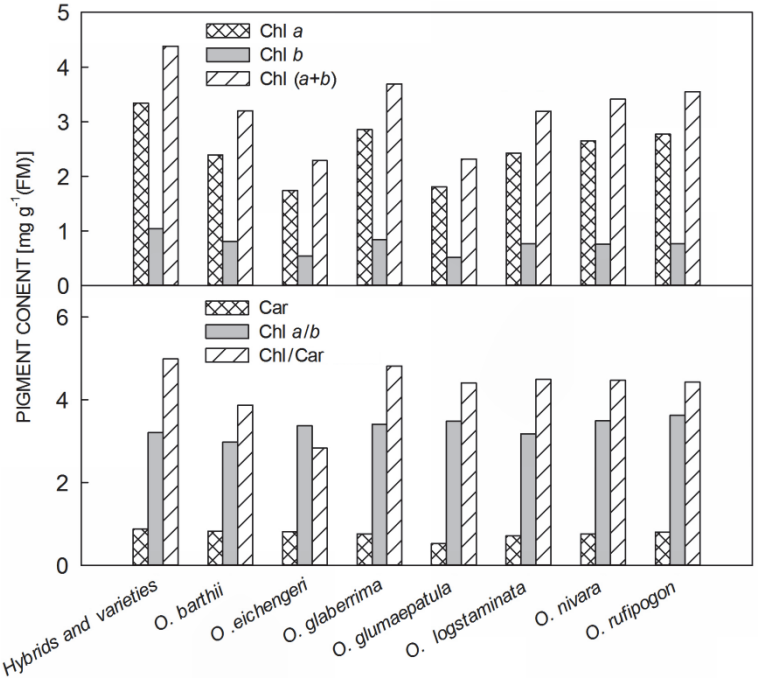

Fig. 1. Variation in leaf photosynthetic pigment content in different wild rice species and cultivated varieties. Each value represents the mean of the respective species. Chl – chlorophyll; Car – carotenoids; FM – fresh mass.

Table 2. Variation in leaf pigment content [mg g^{-1}(FM)] and other traits in rice genotypes. Each value represents the mean of three replications. Each value represents mean of three replications ± SD. Means followed by *different letters* in the same column are significantly different at $P<0.05$ according to *Fisher's* LSD test. The numbers in the parenthesis indicate accession numbers. Chl – chlorophyll; Car – carotenoids; ns – non-significant; CV – coefficient of variation.

Genotypes	Chl *a*	Chl *b*	Chl (*a*+*b*)	Car	Chl *a*/*b*	Chl/Car
Akshyadhan	2.7 ± 0.12^{a}	0.82 ± 0.01^{defghij}	3.52 ± 0.13^{cdefg}	0.680 ± 0.05^{defghi}	3.29 ± 0.09^{abcd}	5.18 ± 0.20^{ab}
DRR H3	4.0 ± 0.48^{bcde}	1.27 ± 0.15^{a}	5.27 ± 0.63^{a}	1.030 ± 0.14^{a}	3.15 ± 0.01^{cde}	5.12 ± 0.06^{ab}
Moroberekan	3.1 ± 0.60^{klm}	0.98 ± 0.16^{bcde}	4.08 ± 0.76^{bcd}	0.860 ± 0.13^{abcde}	3.16 ± 0.10^{bcd}	4.74 ± 0.16^{abcd}
O. barthii (IR 103580)	2.0 ± 0.05^{defgh}	0.77 ± 0.06^{efghijk}	2.77 ± 0.12^{ghij}	0.831 ± 0.03^{bcdef}	2.60 ± 0.15^{e}	3.33 ± 0.03^{h}
O. barthii (IR 80433)	2.8 ± 0.15^{lm}	0.84 ± 0.03^{defghi}	3.64 ± 0.19^{cdef}	0.824 ± 0.05^{bcdef}	3.33 ± 0.05^{abcd}	4.42 ± 0.02^{cdefg}
O. eichengeri (IR 100881)	1.7 ± 0.15^{lm}	0.54 ± 0.14^{lm}	2.24 ± 0.30^{ij}	0.806 ± 0.04^{bcdef}	3.15 ± 0.62^{abcd}	2.78 ± 0.22^{h}
O. glaberrima (IR 102445)	3.8 ± 0.28^{ab}	1.07 ± 0.12^{abc}	4.87 ± 0.40^{ab}	0.921 ± 0.05^{abc}	3.55 ± 0.12^{abcd}	5.29 ± 0.14^{a}
O. glaberrima (IR 100983)	2.0 ± 0.33^{ijklm}	0.65 ± 0.09^{ijkl}	2.65 ± 0.41^{ghij}	0.556 ± 0.10^{hi}	3.08 ± 0.09^{de}	4.77 ± 0.16^{abcd}
O. glaberrima (IR 101800)	1.6 ± 0.05^{m}	0.42 ± 0.02^{m}	2.02 ± 0.06^{j}	0.491 ± 0.01^{i}	3.81 ± 0.04^{ab}	4.11 ± 0.06^{g}
O. glaberrima (IR 104020)	3.2 ± 0.53^{bcd}	0.98 ± 0.06^{bcde}	4.18 ± 0.59^{bcd}	0.863 ± 0.19^{abcde}	3.27 ± 0.35^{bcd}	4.84 ± 0.42^{abc}
O. glaberrima (IR 104033)	3.7 ± 0.00^{ab}	1.08 ± 0.01^{abc}	4.78 ± 0.01^{ab}	0.948 ± 0.01^{ab}	3.43 ± 0.02^{abcd}	5.04 ± 0.05^{ab}
O. glumaepatula (IR 104387)	1.8 ± 0.02^{ab}	0.52 ± 0.01^{lm}	2.32 ± 0.03^{ij}	0.527 ± 0.02^{i}	3.46 ± 0.02^{abcd}	4.40 ± 0.21^{cdefg}
O. langstaminata (IR 104301)	2.5 ± 0.00^{efghijk}	0.81 ± 0.00^{defghij}	3.31 ± 0.00^{defgh}	0.673 ± 0.00^{efghi}	3.09 ± 0.00^{bcd}	4.92 ± 0.00^{abc}
O. langstaminata (IR 105262)	2.3 ± 0.00^{ghijkl}	0.72 ± 0.00^{fghijkl}	3.02 ± 0.00^{efghi}	0.758 ± 0.00^{bcdefg}	3.19 ± 0.00^{bcd}	3.98 ± 0.00^{g}
O. nivara (IR 104650)	3.2 ± 0.13^{bcde}	0.88 ± 0.04^{cdefgh}	4.08 ± 0.17^{bcd}	0.875 ± 0.03^{abc}	3.64 ± 0.02^{abcd}	4.66 ± 0.02^{bcde}
O. nivara (CR 100008)	2.4 ± 0.05^{fghijkl}	0.67 ± 0.01^{hijkl}	3.07 ± 0.03^{efghi}	0.732 ± 0.01^{cdefgh}	3.58 ± 0.15^{abcd}	4.19 ± 0.02^{efg}
O. nivara (CR 100097)	3.0 ± 0.04^{cdefg}	0.92 ± 0.02^{cdefg}	3.92 ± 0.02^{cde}	0.799 ± 0.01^{bcdefg}	3.26 ± 0.11^{bcd}	4.91 ± 0.01^{abc}
O. nivara (CR 100100)	2.0 ± 0.05^{jklm}	0.57 ± 0.02^{klm}	2.57 ± 0.07^{hij}	0.626 ± 0.01^{ghi}	3.51 ± 0.00^{abcd}	4.10 ± 0.05^{fg}
O. rufipogon (IR 80774)	2.7 ± 0.16^{defghij}	0.78 ± 0.09^{defghijk}	3.48 ± 0.26^{cdefg}	0.832 ± 0.04^{bcdef}	3.46 ± 0.19^{abcd}	4.18 ± 0.09^{efg}
O. rufipogon (IR 103404)	3.0 ± 0.25^{cedr}	0.83 ± 0.10^{defghi}	3.83 ± 0.36^{cde}	0.874 ± 0.00^{abcd}	3.61 ± 0.14^{abc}	4.38 ± 0.39^{cdefg}
O. rufipogon (CR 100018)	3.2 ± 0.10^{bcd}	0.94 ± 0.12^{bcdef}	4.14 ± 0.22^{bcd}	0.896 ± 0.04^{abc}	3.40 ± 0.34^{abcd}	4.62 ± 0.43^{bcdef}
O. rufipogon (CR 100267)	2.7 ± 0.06^{defghi}	0.70 ± 0.03^{bhijkl}	3.40 ± 0.03^{cdefgh}	0.749 ± 0.01^{cdefgh}	3.86 ± 0.26^{a}	4.54 ± 0.02^{cdefg}
O. rufipogon (CR 100309)	2.3 ± 0.16^{hjkl}	0.60 ± 0.01^{jklm}	2.90 ± 0.18^{fghi}	0.660 ± 0.07^{fghi}	3.83 ± 0.19^{ab}	4.39 ± 0.20^{defg}
PA 6444	3.7 ± 0.07^{abc}	1.16 ± 0.05^{ab}	4.86 ± 0.12^{ab}	0.919 ± 0.04^{abc}	3.19 ± 0.08^{cd}	5.29 ± 0.10^{a}
Varadhan	3.3 ± 0.08^{bcd}	0.99 ± 0.04^{bcd}	4.29 ± 0.12^{bc}	0.912 ± 0.03^{abc}	3.33 ± 0.05^{abcd}	4.70 ± 0.05^{bcde}
Mean	2.75	0.82	3.57	0.794	3.37	4.50
LSD	0.92	0.30	1.18	0.26	ns	0.72
CV [%]	11.94	13.13	11.84	12.00	8.03	5.70

Table 3. Variations in leaf photosynthetic characteristics in rice genotypes. P_N – net photosynthetic rate [μmol(CO_2) m^{-2} s^{-1}]; g_s – stomatal conductance [mol (H_2O) m^{-2} s^{-1}]; E – transpiration rate [mmol(H_2O) m^{-2} s^{-1}]; C_i – intercellular CO_2 concentration [μmol mol^{-1}]; WUE (P_N/E) – water-use efficiency; WUE_i (P_N/g_s) – intrinsic water-use efficiency; CE – carboxylation efficiency. The numbers in the parenthesis indicate local accession numbers. Each value represents mean of three replications ± SD. Means followed by *different letters* in the same column are significantly different at $P<0.05$ according to *Fisher's* LSD test.

Genotype	P_N	g_s	E	C_i	WUE	WUE_i	CE
Akshyadhan	8.20 ± 0.53^{ab}	0.146 ± 0.01^{abcdefg}	5.58 ± 0.47^{def}	214 ± 5.42^{abcd}	1.48 ± 0.06^{bcd}	61.9 ± 3.06^{bcde}	0.038 ± 0.00^{abc}
DRR H3	13.15 ± 5.66^{cdf}	0.167 ± 0.05^{bcdefgh}	8.54 ± 1.68^{ab}	213 ± 9.15^{abc}	1.51 ± 0.17^{bcd}	59.8 ± 4.95^{bcd}	0.063 ± 0.02^{cdefg}
Moroberekan	8.36 ± 1.06^{ab}	0.087 ± 0.03^{a}	5.37 ± 1.48^{abcde}	199 ± 20.15^{a}	1.65 ± 0.32^{cde}	73.9 ± 13.49^{bcdef}	0.042 ± 0.00^{abcd}
O. barthii (IR 103580)	11.62 ± 3.73^{bcde}	0.126 ± 0.03^{abcd}	7.01 ± 1.16^{bcde}	248 ± 8.61^{fg}	1.64 ± 0.10^{cde}	75.6 ± 4.21^{bcdef}	0.047 ± 0.01^{abcde}
O. barthii (IR 80433)	11.66 ± 1.00^{bcde}	0.153 ± 0.01^{abcdefg}	7.74 ± 0.66^{fg}	254 ± 2.34^{fgh}	1.51 ± 0.04^{bcd}	72.2 ± 0.44^{bcdef}	0.046 ± 0.00^{abcde}
O. eichengeri (IR 100881)	15.64 ± 1.57^{defgh}	0.227 ± 0.01^{hij}	10.52 ± 0.42^{fg}	268 ± 2.38^{gh}	1.49 ± 0.05^{bcd}	65.1 ± 1.02^{bcdef}	0.058 ± 0.00^{bcdefg}
O. glaberrima (IR 102445)	18.31 ± 1.77^{h}	0.249 ± 0.01^{ij}	10.60 ± 0.38^{cdefg}	244 ± 1.74^{cdefg}	1.73 ± 0.06^{cde}	77.3 ± 1.34^{cdefg}	0.075 ± 0.01^{fg}
O. glaberrima (IR 100983)	11.11 ± 0.16^{abcd}	0.123 ± 0.01^{abc}	6.40 ± 0.19^{cdefg}	241 ± 9.33^{cdefg}	1.74 ± 0.03^{cde}	84.4 ± 5.03^{fg}	0.046 ± 0.00^{abcde}
O. glaberrima (IR 101800)	11.61 ± 0.01^{bcde}	0.132 ± 0.01^{abcde}	7.02 ± 0.75^{defg}	244 ± 8.29^{defg}	1.67 ± 0.18^{cde}	82.1 ± 6.04^{fg}	0.048 ± 0.00^{abcde}
O. glaberrima (IR 104020)	13.32 ± 2.07^{cdefg}	0.199 ± 0.01^{efghi}	9.13 ± 0.05^{fgh}	257 ± 20.71^{fgh}	1.46 ± 0.17^{bcd}	71.2 ± 11.66^{bcdef}	0.053 ± 0.01^{bcdef}
O. glaberrima (IR 104033)	14.36 ± 1.95^{defgh}	0.185 ± 0.00^{cdefghi}	9.13 ± 0.26^{cdefg}	242 ± 10.21^{cdefg}	1.57 ± 0.11^{bcd}	78.6 ± 6.55^{defg}	0.060 ± 0.01^{cdefg}
O. glumaepatula (IR 104387)	6.79 ± 0.26^{a}	0.098 ± 0.01^{ab}	4.80 ± 0.50^{fgh}	254 ± 2.33^{fgh}	1.43 ± 0.11^{bcd}	74.7 ± 3.22^{defg}	0.027 ± 0.00^{a}
O. langstaminata (IR 104301)	11.61 ± 0.00^{bcde}	0.194 ± 0.00^{defghi}	8.63 ± 0.00^{def}	207 ± 0.00^{ab}	1.35 ± 0.00^{bc}	59.9 ± 0.00^{bcd}	0.056 ± 0.00^{bcdef}
O. langstaminata (IR 105262)	13.19 ± 0.00^{cdefg}	0.410 ± 0.00^{k}	16.01 ± 0.00^{fg}	249 ± 0.00^{fg}	0.82 ± 0.00^{a}	32.2 ± 0.00^{a}	0.053 ± 0.00^{bcdef}
O. nivara (IR 104650)	11.81 ± 0.33^{bcdef}	0.163 ± 0.00^{bcdefgh}	7.44 ± 0.07^{fgh}	254 ± 3.43^{fgh}	1.59 ± 0.02^{bcd}	72.7 ± 1.38^{bcdef}	0.047 ± 0.00^{abcde}
O. nivara (CR 100008)	16.38 ± 4.77^{fgh}	0.166 ± 0.03^{bcdefgh}	9.07 ± 1.14^{cdefg}	240 ± 5.54^{cdefg}	1.79 ± 0.15^{de}	81.2 ± 2.84^{efg}	0.069 ± 0.02^{efg}
O. nivara (CR 100097)	24.21 ± 1.26^{i}	0.273 ± 0.04^{j}	11.18 ± 1.81^{bcde}	198 ± 32.20^{a}	2.24 ± 0.44^{f}	109.0 ± 23.61^{h}	0.127 ± 0.03^{h}
O. nivara (CR 100100)	12.13 ± 0.13^{bcdef}	0.139 ± 0.00^{abcdef}	7.56 ± 0.29^{bcdef}	232 ± 2.24^{bcdef}	1.61 ± 0.05^{bcde}	84.7 ± 1.69^{fg}	0.052 ± 0.00^{abcdef}
O. rufipogon (IR 80774)	17.89 ± 3.27^{gh}	0.226 ± 0.04^{hij}	8.92 ± 1.12^{abcde}	215 ± 14.38^{abcde}	2.00 ± 0.01^{ef}	96.6 ± 7.36^{gh}	0.083 ± 0.01^{g}
O. rufipogon (IR 103404)	9.58 ± 2.72^{abc}	0.206 ± 0.04^{fghij}	7.82 ± 1.34^{h}	280 ± 3.45^{h}	1.22 ± 0.04^{ab}	58.3 ± 2.55^{bc}	0.034 ± 0.01^{ab}
O. rufipogon (CR 100018)	14.88 ± 2.18^{defgh}	0.208 ± 0.02^{fghij}	9.46 ± 0.93^{efg}	245 ± 2.34^{efg}	1.57 ± 0.01^{bcd}	77.4 ± 1.44^{cdefg}	0.061 ± 0.01^{cdefg}
O. rufipogon (CR 100267)	16.04 ± 0.62^{efgh}	0.189 ± 0.01^{cdefghi}	9.08 ± 0.57^{fg}	247 ± 4.86^{fg}	1.77 ± 0.06^{de}	80.4 ± 2.28^{efg}	0.065 ± 0.00^{defg}
O. rufipogon (CR 100309)	14.33 ± 4.70^{defgh}	0.145 ± 0.05^{abcdefg}	8.99 ± 1.53^{fgh}	256 ± 4.60^{fgh}	1.58 ± 0.10^{bcd}	74.0 ± 2.01^{bcdef}	0.056 ± 0.01^{bcdef}
PA 6444	12.10 ± 0.14^{bcdef}	0.169 ± 0.01^{cdefgh}	8.10 ± 0.35^{ab}	204 ± 8.47^{ab}	1.50 ± 0.08^{bcd}	67.2 ± 4.74^{bcdef}	0.059 ± 0.00^{bcdefg}
Varadhan	12.35 ± 1.41^{bcdef}	0.210 ± 0.01^{ghij}	9.07 ± 0.67^{abcde}	216 ± 4.15^{abcde}	1.36 ± 0.01^{bc}	56.7 ± 2.65^{b}	0.057 ± 0.01^{bcdef}
Mean	13.23	0.187	8.52	236	1.57	73.0	0.057
LSD ($P<0.05$)	4.71	7.01	2.62	30.9	0.55	20.1	2.5
CV (%)	17.3	18.2	14.9	6.3	12.6	13.3	19.1

Table 4. Relationship between leaf photosynthetic efficiency and other related traits in *Oryza* species. P_N – net photosynthetic rate [μmol(CO_2) m^{-2} s^{-1}]; g_s – stomatal conductance [mol(H_2O) m^{-2} s^{-1}]; E – transpiration rate [mmol(H_2O) m^{-2} s^{-1}]; C_i – intercellular CO_2 concentration [μmol mol^{-1}]; WUE (P_N/E) – water-use efficiency; WUE_i (P_N/g_s)– intrinsic water-use efficiency; CE – carboxylation efficiency; Chl – chlorophyll; Car – carotenoids; SLA – specific leaf area; SLM – specific leaf mass; $^{*}P<0.05$, $^{**}P<0.01$.

	P_N	g_s	E	C_i	WUE_i	CE	WUE	Chl *a*	Chl *b*	Chl (*a*+*b*)	Car	Chl *a*/*b*	Chl/Car	SLA	SLM	Leaf thickness
P_N	1.00															
g_s	0.49*	1.00														
E	0.62**	0.97**	1.00													
C_i	–0.15	0.05	0.10	1.00												
WUE_i	0.51**	–0.45*	–0.28	–0.20	1.00											
CE	0.95**	0.41*	0.51*	–0.43*	0.55**	1.00										
WUE	0.59**	–0.36*	–0.23	–0.34	0.95**	0.65**	1.00									
Chl *a*	0.14	0.20	0.18	–0.23	–0.20	0.15	–0.10	1.00								
Chl *b*	0.03	0.20	0.15	–0.23	–0.30	0.05	–0.19	0.96**	1.00							
Chl (*a*+*b*)	0.11	0.20	0.18	–0.23	–0.22	0.13	–0.13	1.00	0.97**	1.00						
Car	0.22	0.22	0.21	–0.05	–0.19	0.17	–0.07	0.85**	0.87**	0.86**	1.00					
Chl *a*/*b*	0.34	–0.03	0.08	0.00	0.34	0.30	0.29	–0.07	–0.34	–0.13	–0.23	1.00				
Chl/Car	–0.10	0.09	0.05	–0.34	–0.19	–0.02	–0.17	0.72	0.67**	0.72**	0.26	0.02	1.00			
SLA	0.05	0.06	0.07	0.13	–0.06	0.00	–0.06	0.11	0.10	0.11	0.26	0.01	–0.16	1.00		
SLM	0.12	0.16	0.20	–0.16	–0.10	0.15	–0.14	0.18	0.12	0.17	0.14	0.21	0.12	–0.02	1.00	
Leaf thickness	0.24	0.19	0.25	–0.15	0.01	0.26	–0.03	0.12	0.00	0.09	0.08	0.40	0.06	0.10	0.90**	1.00

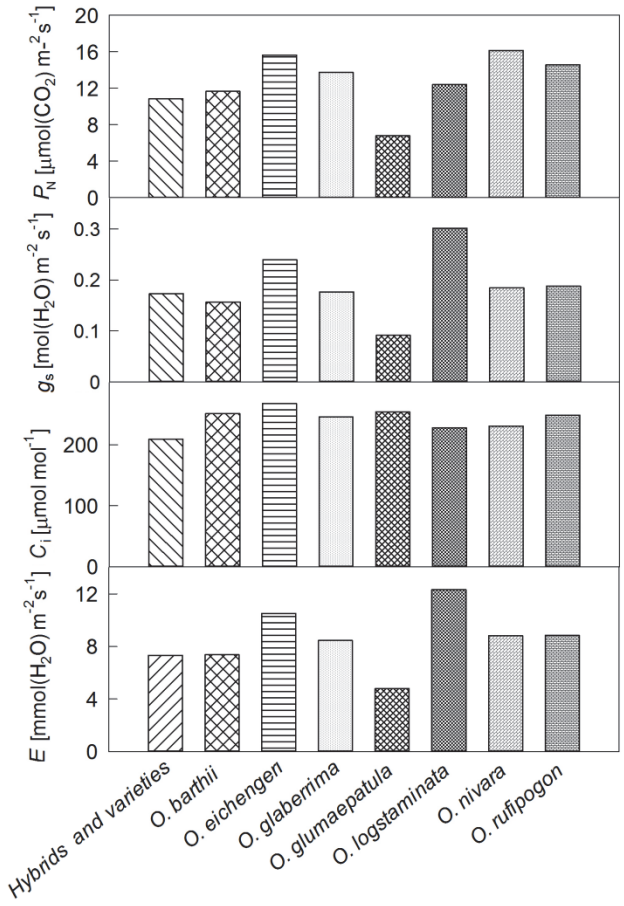

Fig. 2. Variation in leaf photosynthetic traits among different wild rice species and cultivated varieties. P_N – rate of photosynthesis; g_s – stomatal conductance, E – transpiration rate, C_i – internal CO_2 concentration. Each value represents the mean of the respective species.

13.2 μmol (CO_2) m^{-2} s^{-1} (Table 3). *O. rufipogon W–61* (ACC. No. IR 80774)*, O. rufipogon* (ACC. No. CR 100267), and *O. nivara* (ACC. No. CR 100008) were the other genotypes with higher P_N [>16 μmol(CO_2) m^{-2} s^{-1}]. The mean of P_N was the highest in *O. nivara* followed by O. *eichengeri*, whereas the lowest mean value of P_N was found in *O. glumaepatula;* it was lower than the mean of cultivated varieties and hybrids of *O. sativa*. Photosynthesis tended to be lower in the wild *Oryza* species compared with *O. sativa* (Cook and Evans 1983). However, higher photosynthetic rates were reported for wild rice species by Yeo *et. al*. (1994).

In the present study, the photosynthetic rate recorded in *Oryza* species was lower than the published values of P_N reported by other workers (Yeo *et al*.1994, Zhao *et al*. 2010). The lower values of P_N could be due to high temperatures prevailing during crop growth and during photosynthetic measurements. A wide variation in g_s was observed. g_s varied between a maximum of 0.410 mol(H_2O) m^{-2} s^{-1} (*O. langstaminata* ACC. No. IR 105262) and minimum of 0.087 (Moroberekan) with a mean of 0.187 mol(H_2O) m^{-2} s^{-1} (Table 3). The mean of g_s was the highest in *O. longstaminata* followed by *O. eichengeri.* With the exception of *O. glumaepatula*, the mean of g_s was higher than that of cultivated varieties and hybrids (Fig. 2). A wide variation in E was also observed. E varied between 16.0 (*O. langstaminata* ACC. No. IR 105262) and 4.8 (*O. glumaepatula* ACC No. IR 104387) with a mean of 8.52 mmol(H_2O) m^{-2} s^{-1}. The mean E of cultivated varieties and hybrids was 7.38 mmol(H_2O) m^{-2} s^{-1}, which was much lower than that of all other *Oryza* species. The mean of E (Fig. 2) was the highest in *O. langstaminata* (12.3 mmol m^{-2} s^{-1}), followed by *O. eichengeri* [10.5 mmol(H_2O) m^{-2} s^{-1}].

Water-use efficiency (WUE, P_N/E), intrinsic water-use efficiency (WUE_i, P_N/g_s), and carboxylation efficiency, CE (P_N/C_i), varied significantly among the tested rice genotypes. WUE varied between 2.24 (*O. nivara*, ACC. No.CR 100097) and 1.35 (*O. langstaminata*, ACC. No. IR 104301) with a mean of 1.57. The maximum WUE_i was reached by *O. nivara* (ACC. No. CR 100097), while *O. langstaminata* (ACC. No. IR 105262) had the lowest WUE_i. CE differed significantly amongst the *Oryza* species. The maximum CE was recorded in *O. nivara* (ACC. No. CR 100097), *O. glumaepatula* (ACC. No. IR 104387) had the lowest CE, which was much lower than the mean CE for all tested genotypes (Table 3).

Significant variation in leaf thickness was noticed among different genotypes of *Oryza*. The leaf thickness varied between 0.13 (*O. nivara*, ACC. No. CR 100008, ACC. No. IR 104650, and *O. langstaminata*, ACC. No. IR 105262) and 0.07 mm (*O. barthii*, ACC. No. IR 103580) with a mean thickness of 0.13 mm. Marginal differences were noticed among different species in SLA. However, SLM varied significantly among different *Oryza* species (Fig. 3). SLM varied between a maximum of 7.1 (*O. nivara*, ACC. No. IR 104650) and 3.7 (*O. rufipogon*, ACC. No. IR 103404).

Multiple correlation analysis was performed between different gas-exchange characteristics and other physiological traits (Table 4). The results revealed that the rate of photosynthesis was not significantly influenced by SLA, SLM, leaf thickness, and Chl or Car contents (Table 4). The correlation coefficients indicated that P_N was not dependent on the leaf pigment content or leaf thickness. A strong positive correlation between P_N and the P_N/C_i ratio, which represents CE, indicated that the observed variation in P_N was not based on pigment content or other leaf traits, but related to the leaf CE. A positive association of photosynthesis with light saturation and CE was reported by Yeo *et al*. (1994). Zhao *et al.* (2010) reported that for choosing rice genotypes with high P_N, selecting for high Chl content or leaf thickness may not be a useful strategy. The highest photosynthesis was noticed in *O. nivara* followed by *O. eichengeri* and *O. rufipogon*, which was higher than in the hybrids and cultivars of cultigen *O. sativa*.

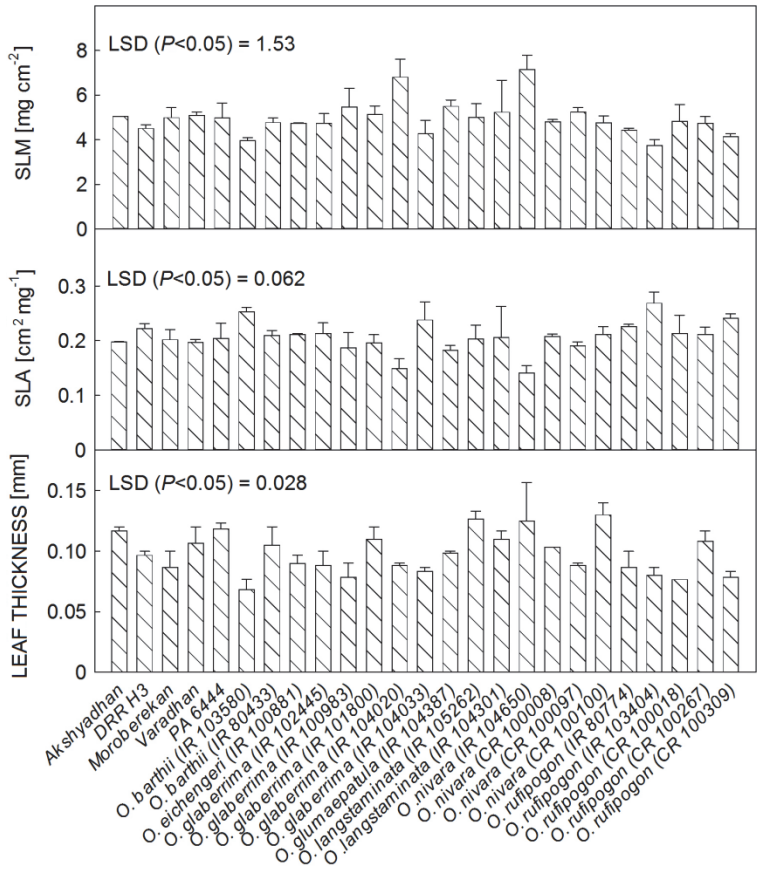

Fig. 3. Variation in specific leaf mass (SLM), specific leaf area (SLA), and leaf thickness in wild rice species and cultivated varieties. Each value represents the mean of three replications ± SD.

However, the difference amongst these species was not substantial. Yeo *et al.* (1994) studied photosynthetic characteristics of 22 *Oryza* species and reported that the species *O. australiensis* was superior to the *O. sativa* genotypes. However, Zhao *et. al.* (2010) reported that among 17 *Oryza* species, *O. rufipogon* and *O. australiensis* exhibited the highest photosynthesis, and they suggested that *O. rufipogon* was the best wild resource species for improving rice photosynthesis. Ming *et al*. (2004) estimated the variation in photosynthesis in 16 *Oryza s*pecies and suggested *O. sativa* and *O. rufipogon* as potential sources of the enhanced P_N. Substantial genotypic differences in leaf photosynthetic efficiency was observed throughout development in rice (Ohsumi *et al*. 2007) and the variation of P_N was correlated with that of g_s at a given stage and leaf N content. Our data on leaf photosynthetic traits showed significant, positive association between P_N and g_s (Table 4). Leaf nitrogen and g_s have been suggested to be major factors limiting leaf photosynthesis and simultaneous improvement in both these traits is essential for breeding varieties with higher P_N (Ohsumi *et al*. 2007). Our data indicated that *O. nivara* (ACC. No. CR 100097)*, O. rufipogon* (ACC. No. IR 80774), *O. rufipogon* (ACC.No. CR 100267), and *O. nivara* (ACC. No. CR 100008) recorded higher P_N and they could be useful as suitable donors for improving leaf photosynthesis.

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