

Table 1 Diagnostic characters of species of *Halophila*

<i>H. ovalis</i>	<i>H. ovata</i>	<i>H. decipiens</i>	<i>H. stipulacea</i>	<i>H. beccarii</i>
Dioecious	Dioecious	Monoecious	Dioecious	Dioecious
Leaves 2, oblong-elliptic, surface glabrous, 1-7 cm long: 1/2-2 cm wide: margin entire: intramarginal cross veins 10-12 pairs	Leaves 2, oblong-elliptic, glabrous; 7-14 mm long: 3-5 mm wide, margin entire, intramarginal cross veins 3-11 pairs	Leaves 2, oblong-elliptic, surface rough with stiff unicellular hairs: margin finely serrulate, cross veins 4-9 pairs: 20-30 mm long and 6-10 mm wide	Leaves 2, linear to oblong, glabrous, papillose or slightly hairy 3-6 cm long: 2 1/2-8 mm wide, margin serrulate	Leaves 6-10 on erect shoot: lanceolate, glabrous margin entire, 6-13 mm long: 1-2 mm wide, cross veins absent
Flowers unisexual	Flowers unisexual	Spathe encloses one male and one female flower	Flowers unisexual	Flowers unisexual

25 May 1987; Revised 14 October 1987

1. Hartog, C. den, *The seagrasses of the world*, North-Holland, Amsterdam, 1970, p. 254.

ALTERATION OF RIBULOSE BISPHOSPHATE CARBOXYLASE/OXYGENASE RATIO BY UREA

M. C. GHILDIYAL

Department of Agronomy, University of Illinois, Urbana, Illinois 61801, USA.

Present address: Division of Plant Physiology, Indian Agricultural Research Institute, New Delhi 110 012, India.

RIBULOSE biphosphate carboxylase/oxygenase (Rubisco) catalyses carboxylation and oxygenation of RuBP to initiate photosynthesis and photorepiration respectively. Carbon dioxide and oxygen are mutually competitive substrates in these reactions¹. Modification of enzyme reactivity towards its alternate substrate might enhance plant productivity^{2,3}. The question arises whether the activity of Rubisco can be modified. Efforts have been made to seek chemical agents which modify its activity. However, most of the apparent effects were the result of artifacts in the assay procedures used^{4,5}. The present study by utilizing a sensitive assay procedure indicates that urea decreases the carboxylase/oxygenase ratio.

RuBP carboxylase/oxygenase was purified from spinach following the method of Jordan and Ogren⁴. Purified enzyme was preincubated with 10 mM NaHCO₃, 10 mM MgCl₂ with 25 mM tris (pH 8) and 0.25 mM EDTA for 1 h at 20°C. Assays were

initiated by adding 70 µg of activated protein in 25 µl to reaction mixtures at 25°C containing varying amounts (0.5, 1, 2, 5, 10 mM) of NaH¹⁴CO₃, 0.0236 mM H³RuBP, 10 mM MgCl₂, 50 mM Bicine at pH 7.85, with and without 2 M urea in a total volume of 0.5 ml. Reactions were performed in 6 ml scintillation vials which were sealed with serum stoppers. The reaction mixtures were flushed with pure oxygen for 15 min before adding NaH¹⁴CO₃ and enzyme. Reactions were terminated after 30 min by adding 0.1 ml of a solution containing 0.05 N HCl and 50 mM ZnSO₄ and stored at -15°C.

Carboxylation and oxygenation rates were determined according to Jordan and Ogren⁴. The labelled products of carboxylase reaction [(³H, ¹⁴C) glycerate-P] and the oxygenase reaction (³H-glycolate-P) were separated. Glycolate-P phosphatase was used to convert (³H) glycolate-P to (³H) glycolate which was readily separated from labelled organic phosphates by ion exchange chromatography. Radioactivity in these compounds was quantified by scintillation spectroscopy. The ratio of RuBP carboxylase/oxygenase was determined by measuring the two activities simultaneously in the same reaction mixture. This procedure eliminates the possible discrepancy when two activities were measured separately under different conditions particularly at low CO₂ concentration^{4,5}.

The ratio of carboxylase to oxygenase activity (vc/v_o) plotted against the ratio of CO₂ and O₂ concentrations present during assay is shown in figure 1. The ratio increases with increase in the ratio of the CO₂ and O₂ concentrations. However, this ratio was lower when urea was included in the assay medium.

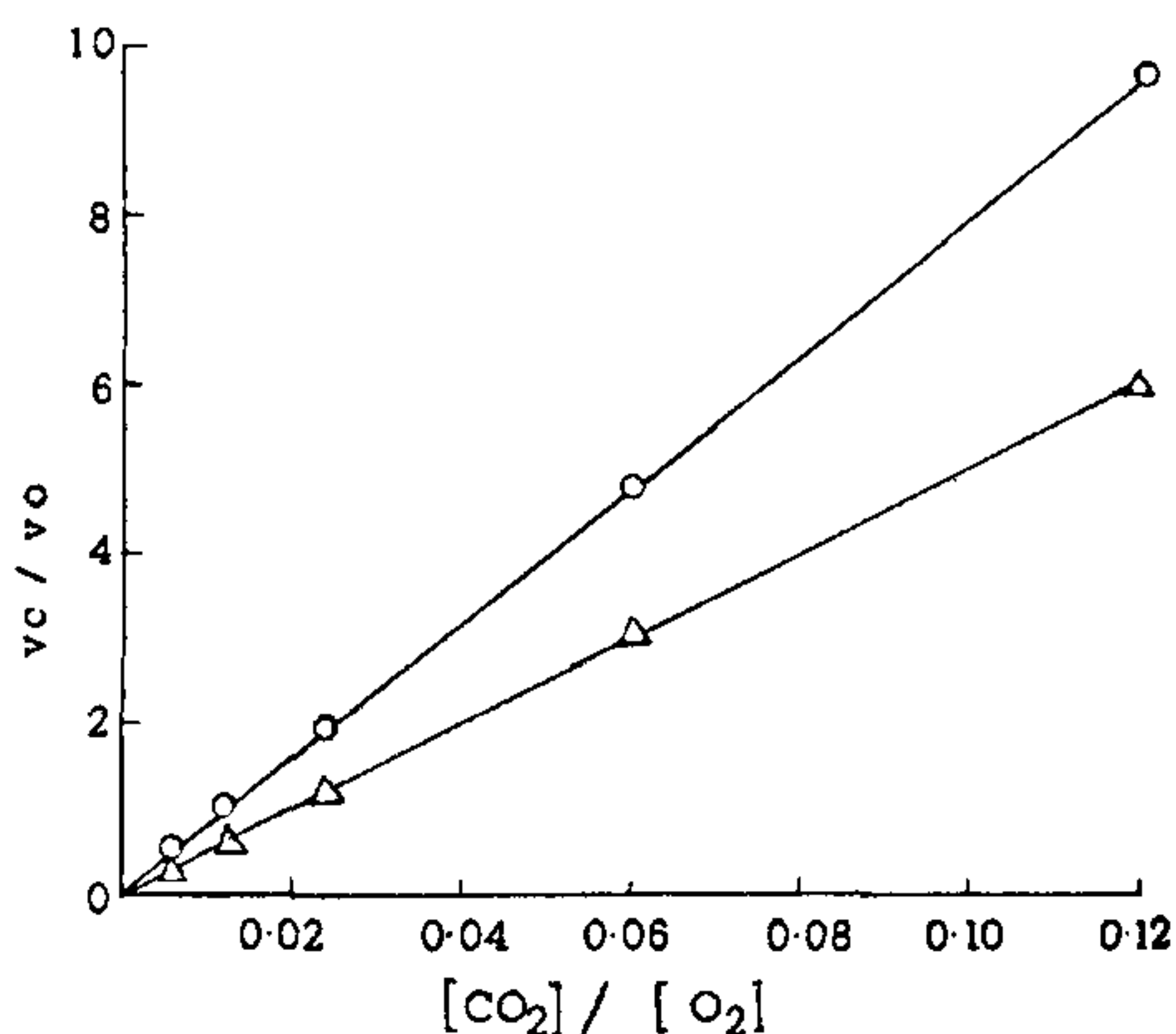


Figure 1. Effect of urea on the ratio of RuBP carboxylase to oxygenase (v_c/v_o) plotted against the ratio of $(CO_2)/(O_2)$ concentration present during assay. Control ○, with urea △.

The substrate specificity factor, $V_c K_o/V_o K_c$, determines the relative rates of two reactions at any given CO_2 and O_2 concentrations¹. A high specificity value indicates a greater specificity for CO_2 . Both the enzyme activities assayed simultaneously under several $(CO_2)/(O_2)$ values permit direct determination of the specificity factor. The specificity factor calculated from the slope of this plot was found to be 80. Similar value has been reported for other C_3 plants⁵. Urea treatment decreased the specificity factor to 50.

It has been argued that RuBP carboxylase/oxygenase cannot completely discriminate between CO_2 and O_2 , so that photorespiration is unavoidable^{6,7}. However, Rubisco enzyme from diverse species showed substantial differences in CO_2/O_2 specificity and that carboxylase/oxygenase ratio increased during the natural evolution of photosynthesis⁸. Furthermore, Mn^{2+} ⁹, and temperature¹ have been shown to alter the ratio of two activities. The present study indicates that urea also alters the carboxylase/oxygenase ratio.

The author is thankful to Prof. W. L. Ogren and Prof. G. S. Sirohi for facilities and encouragement. The fellowship received from FAO of the United Nations is gratefully acknowledged.

12 June 1987; Revised 24 August 1987

1. Liang, W. A., Ogren, W. L. and Hageman, R. H., *Plant Physiol.*, 1974, **54**, 678.

- Ogren, W. L., *CO₂ metabolism and plant productivity*, (eds) R. H. Burris and C. C. Black, University Park Press, Baltimore, 1976, p. 19.
- Ogren, W. L., *Proc. 4th Int. Congr. on Photosynthesis*, (eds) D. O. Hall, J. Coombs and T. W. Goodwin, Biochem. Soc., London, 1978, p. 721.
- Jordan, D. B. and Ogren, W. L., *Plant Physiol.*, 1981, **67**, 237.
- Ogren, W. L., *Annu. Rev. Plant Physiol.*, 1984, **35**, 415.
- Lorimer, G. H. and Andrews, T. J., *Nature (London)*, 1973, **243**, 359.
- Andrews, T. J. and Lorimer, G. H., *FEBS Lett.*, 1978, **90**, 1.
- Jordan, D. B. and Ogren, W. L., *Nature (London)*, 1981, **291**, 513.
- Wildner, G. F. and Henkel, J., *FEBS Lett.*, 1978, **91**, 99.

INHERITANCE OF PERICARP COLOUR IN RICE, *ORYZA SATIVA* LINN.

P. T. ANNIE and K. PAVITHRAN

Department of Botany, University of Calicut, Calicut 673 635, India.

INHERITANCE of pericarp colour in rice has been studied earlier and genetical ratios $3r:1w^{1-6}$, $12p:3r:1w^1$, $9p:6b:1w^7$, $9p:3b:4w^{8,9}$ and $15w:1r^{10,11}$ have been reported. A genic scheme for pericarp colouration has also been proposed¹². The present study reports the inheritance of purple pericarp.

Inheritance of purple pericarp was studied in Jaya × 7019 and Jaya × 7010 up to F_3 generation. Jaya has white pericarp, 7019 and 7010 and the markers supplied by Dr Nelson E. Jodon of the USDA Louisiana, have purple pericarp.

F_1 showed purple pericarp (dominant) in both crosses and the F_2 population of 274 plants of Jaya × 7019 segregated into 176 purple:98 white, giving a good fit to the ratio 162:94 with $\chi^2 = 0.11$; the F_2 population of Jaya × 7010 segregated into 438 purple:148 white, giving a good fit to 3:1 with $\chi^2 = 0.02$ (table 1). The ratios were confirmed by the breeding behaviour of families in F_3 generation.

Out of 50 F_3 families of Jaya × 7019 studied, 1 bred true for purple, 11 segregated for 3:1, 11 for 9:7, 2 for 15:1, 1 for 27:37, 2 for 45:19, 2 for 54:10, 4 for 162:94 and 16 bred true for white pericarp, giving a good fit to the expected ratio with $\chi^2 = 11.48$ for 8 d.f., and out of 89 F_3 families of