New Phytol. (1976) **76**, 301–305.

DISTRIBUTION OF THE C_4 DICARBOXYLIC ACID PATHWAY OF PHOTOSYNTHESIS IN LOCAL MONOCOTYLEDONOUS PLANTS AND ITS TAXONOMIC SIGNIFICANCE

BY A. S. RAGHAVENDRA* AND V. S. R. DAS

Department of Botany, Sri Venkateswara University,
Tirupati 517502, *India*

(*Received* 21 *July* 1975)

SUMMARY

The photosynthetic characteristics of fifty-six monocotyledonous plants belonging to seventeen families were examined. As many as twenty-three plants, hitherto unreported, showed C_4 pathway characteristics. C_4 species were recorded in ten new genera. The C_4 plants were confined to the families, Cyperaceae and Gramineae. The findings confirm the highly evolved nature of the tribes Andropogoneae and Paniceae in contrast to the primitive features of Agrosteae and Festuceae. On the basis of photosynthetic characteristics, it is suggested that *Aristida* should be included in a separate tribe, Aristideae, whereas *Eragrostis* resembles more panicoid grasses rather than the members of Festuceae. The importance of the C_4 syndrome in relation to the evolution of grasses is stressed.

INTRODUCTION

Considerable attention has been given in recent years to the C_4 dicarboxylic acid pathway of photosynthesis (Hatch, Osmond and Slatyer, 1971), since its discovery (Kortschak, Hartt and Burr, 1965) and detailed characterization (Hatch and Slack, 1970). The C_4 plants are mostly tropical and subtropical species. Although the pathway had been found in many plants (Downton, 1971), subsequent studies revealed that the C_4 pathway was functional in a large number of then unreported species (Das and Raghavendra, 1973; Wynn *et al.*, 1973; McWilliam and Mison, 1974).

During the course of investigations on the C_4 dicarboxylic acid pathway of photosynthesis in our laboratory (Das, 1974; Raghavendra, 1975), we examined the local flora for the presence of the C_4 pathway. The existence of the C_4 pathway could be demonstrated by the occurrence of one or more characteristics associated with the syndrome, such as Kranz type leaf anatomy, high photosynthetic rates, insensitivity of carbon fixation to oxygen in ambient atmosphere and low carbon dioxide compensation point (Tregunna *et al.*, 1970). The above features have already been used to detect the functioning of the C_4 pathway in a local dicotyledonous weed flora (Das and Raghavendra, 1973). Here we report the presence of characteristics of the C_4 pathway in a large number of monocotyledonous plants.

---

* Present address: Central Plantation Crops Research Institute, Regional Station, Vittal 574243, Karnataka, India.

MATERIALS AND METHODS

Plants were collected from their natural habitat either in the University campus or from Tirumalai Hills. Some of the ornamental plants were being grown in the University Botanic Garden.

The methods used to screen for the C_4 syndrome were as already described (Das and Raghavendra, 1973). The anatomy of leaves was studied under a research light microscope after preparing free-hand sections. Carbon dioxide compensation points were determined by estimating CO_2 concentration in an enclosed chamber after allowing the plant to photosynthesize for sufficient time (Das and Raghavendra, 1973). Their photosynthetic rates were estimated both in air and in an aerobic atmosphere by a modified continuous gas flow technique (Raghavendra, 1975).

RESULTS

In all, fifty-six plants belonging to seventeen families were examined for their phosynthetic characters. Of them, twenty-three plants were found to possess the C_4 syndrome (Table 1). However, C_4 species were confined to two families only, namely, Cyperaceae and Gramineae. There were as many as ten genera with C_4 plants, which were hitherto unreported (Table 2). Of particular interest was the observation that, in the Gramineae, C_4 plants were represented by the genera of the following tribes only: Maydeae, Andropogoneae, Paniceae, Eragrosteae, Zoyseae, Chlorideae and Aristideae.

Table 1. *Photosynthetic characters of various monocotyledonous plants**

Family and plant species	Leaf anatomy Kranz (K) or normal (N)	Bundle sheath chloroplasts†	Compensation point (CO_2 ppm)	Photosynthetic rate‡ ($mg\ CO_2/dm^2/h$)	
				Air	O_2 free air
HYDROCHARITACEAE					
Hydrilla verticillata Royle.	N	–	–	–	–
COMMELINACEAE					
Commelina benghalensis L.	N	–	42	32	41 (28)
Cyanotis axillaris R & S.	N	–	61	18	26 (44)
Tradescantia virginiana L.	N	–	37	29	39 (34)
Zebrina pendula Schnizl.	N	–	58	16	22 (38)
ERIOCAULACEAE					
Eriocaulon truncatum Ham.	N	–	–	–	–
MUSACEAE					
Musa paradisiaca L.	N	–	59	22	32 (45)
Ravenala madagascarensis Sour.	N	–	48	26	38 (46)
ZINZEBERACEAE					
Zinziber officinale Rosc.	N	–	47	30	38 (27)
Elettaria cardamom Maton.	N	–	–	–	–
Curcuma longa L.	N	–	54	28	37 (32)
CANNACEAE					
Canna indica L.	N	–	62	28	39 (39)
LILIACEAE					
Allium cepa L.	N	–	62	35	45 (26)
Asparagus racemosus Willd.	N	–	65	28	34 (21)
Gloriosa superba L.	N	–	48	37	46 (24)
Ornithogalum spp.	N	–	52	32	44 (38)
Scilla indica Baker.	N	–	49	28	34 (21)
SMILACACEAE					
Smilax zeylanica W.	N	–	48	33	44 (33)
ARACEAE					
Colocasia antiquorum Scholt.	N	–	55	38	49 (29)
Pistia stratiotes L.	N	–	58	32	45 (41)
Pothos scandens L.	N	–	72	15	21 (40)

Table 1 (*contd.*)

LEMNACEAE					
Lemna gibba L.	N	–	–	–	–
AMARYLLIDACEAE					
Zephyranthus grandiflora Lindl.	N	–	68	22	30 (36)
DIOSCOREACEAE					
Dioscorea alata L.	N	–	45	39	51 (31)
D. oppositifolia L.	N	–	–	–	–
D. pentaphylla L.	N	–	–	–	–
AGAVACEAE					
Dracaena terniflora Roxb.	N	–	63	18	25 (39)
PALMAE					
Borassus flabellifer L.	N	–	42	29	37 (28)
Cocos nucifera L.	N	–	38	42	51 (21)
Phoenix sylvestris Roxb.	N	–	53	35	46 (31)
ORCHIDACEAE					
Habenaria platyphylla Spreng.	N	–	–	–	–
Vanda tesselata Hook.	N	–	63	18	24 (33)
CYPERACEAE					
Scirpae					
Fimbristylis monostachya Hassk.	K	+	0	55	55 (0)
F. ulginosa Strend.	K	+	2	52	53 (2)
Cyperae					
Kyllinga triceps Rottb.	K	+	3	62	59 (−5)
Sclerae					
Scleria lithosperma Sw.	K	+	–	–	–
GRAMINEAE					
Maydeae					
Coix aquatica R.	K	+	4	58	60 (3)
Andropogoneae					
Apluda aristata L.	K	+	2	62	62 (0)
Cymbopogon coloratus Stapf.	K	+	0	68	68 (0)
C. martini Wats.	K	+	–	–	–
C. nardus Rendle.	K	+	0	71	68 (−4)
Iseilema laxum Hack.	K	+	0	51	50 (−2)
Paniceae					
Brachiaria semiundulata Stapf.	K	+	2	63	60 (−5)
Digitaria adscendens H.B. & K.	K	+	5	48	50 (4)
Isachne dispar Trin.	K	+	0	67	69 (3)
Melanocenchris Royleana Nees.	K	+	4	52	53 (2)
Panicum repens L.	K	+	2	71	71 (0)
Eragrosteae					
Eragrostis ciliaris Link.	K	+	3	58	59 (2)
Leptochloa polystachya Benth.	K	+	0	68	68 (0)
Zoysieae					
Perotis indica O. Ktz.	K	+	2	58	60 (3)
Tragus biflorus Schult.	K	+	4	65	66 (2)
Chlorideae					
Chloris barbata Sw.	K	+	4	60	63 (5)
Enteropogon monostachyos Schum.	K	+	2	68	70 (3)
Aristideae					
Aristida funiculata Trin & Rupr.	K	+	0	72	73 (1)
A. setacea Retz.	K	+	2	63	63 (0)
Bambuseae					
Dendrocalamus strictus Nees.	N	–	42	38	50 (32)

* The plants were listed according to the sequence followed by Darlington and Wylie (1955). The arrangement of tribes in Cyperaceae was of Hutchinson (1959).

† Indicates the specialization of bundle sheath chloroplasts for starch accumulation.

‡ Rates were determined both in normal air with 21% oxygen and oxygen free air. Figures in parentheses represent the percentage enhancement in photosynthetic rate after removal of oxygen.

Table 2. *Monocotyledonous genera, hitherto unreported, found in the present investigation to have C_4 species*

Family	Genera	
Cyperaceae	*Fimbristylis*	*Scleria*
Gramineae	*Apluda*	*Brachiaria*
	Enteropogon	*Cymbopogon*
	Isachne	*Iseilema*
	Melanocenchris	*Perotis*

DISCUSSION

The C_4 plants reported in the present investigation form an addition to the list of plants already known to have C_4 characteristics (Downton, 1971; Das and Raghavendra, 1973; Wynn *et al.*, 1973; McWilliam and Mison, 1974). Radiate mesophyll tissue in the leaf lamina, which was considered to be one of the distinguishing characters of C_4 plants (Black, 1973), was observed in many of the C_4 genera we examined (Metcalfe, 1960, 1971).

The distribution of all the C_4 genera listed in Table 1 was invariably confined mostly to tropical regions of the world (Darlington and Wylie, 1955). The Andropogoneae and Paniceae which possess a number of C_4 plants are predominantly tropical in distribution (Hartley, 1950, 1958a, b). On the other hand the Agrosteae and Festuceae, with a very few C_4 plants reported among them, occur predominantly in temperate regions. A proviso should be made because the present geographical distribution of pasture grasses has been greatly influenced by man (Cooper, 1965). Nevertheless, all the tribes of Gramineae having C_4 species are known to be tropical in their distribution.

The C_4 pathway has been suggested to be a successful adaptation to ecological conditions of high temperature, high irradiance and limited availability of water (Hatch and Slack, 1970; Black, 1971, 1973). Such conditions normally occur in tropical and subtropical regions of the world. It is reasonable to assume that tropical and subtropical plants have developed successfully the C_4 dicarboxylic acid pathway of photosynthesis during evolution.

The occurrence of the C_4 pathway, which may be considered as an advanced character, can also be used to assist some of the controversies over the systematic position of genera such as *Aristida* and *Eragrostis*. Parodi and Frier (1945) placed *Aristida* in the Stipeae along with *Piptochaetium*, *Nassella*, *Oryzopsis* and *Stipa*, but Brown (1958) felt strongly that *Aristida* is distinct in its leaf anatomy from them because of its two parenchymatous bundle sheaths. The demonstration of the characteristics of the C_4 pathway in species of *Aristida* in the present investigation and also in previous reports (Downton, 1971) strongly supports the inclusion of *Aristida* in a separate tribe, Aristideae. The occurrence of the C_4 syndrome in *Eragrostis* spp. (present report; Downton, 1971) suggests its close affinity with panicoid grasses rather than with members of the Festuceae. The systematic position of *Eragrostis* was previously discussed by Metcalfe (1960).

The occurrence of the C_4 pathway also correlates with our ideas of evolution among the tribes of the Gramineae. The Bambuseae, with no C_4 species, appear to be primitive whereas the subfamily Panicoideae, with all the three tribes of Andropogoneae, Maydeae and Paniceae possessing a number of C_4 plants, is considered to be highly evolved (Hutchinson, 1959). The importance of the C_4 syndrome in relation to grass evolution, with special reference to *Panicum* has been stressed (Brown, 1958; Brown and Smith, 1972). The striking feature of confinement of the C_4 dicotyledonous plants to the Centrospermae (Das and Raghavendra, 1973) and that of C_4 monocotyledons to the Cyperaceae and Gramineae (present work; Downton, 1971) indicates the importance of knowledge of photosynthetic pathways for botanical classification.

ACKNOWLEDGMENTS

One of us (A.S.R.) was in receipt of an U.G.C. Junior Research Fellowship. Thanks are due to Dr K. N. Rao, Department of Botany, Sri Venkateswara University, Tirupati for identifying some of the plants.

REFERENCES

BLACK, C. C. (1971). Ecological implications of dividing plants into groups with distinct photosynthetic production capacities. *Adv. Ecol. Res.*, **7**, 87.

BLACK, C. C. (1973). Photosynthetic carbon fixation in relation to net CO_2 uptake. *Ann. Rev. Plant Physiol.*, **24**, 253.

BROWN, W. V. (1958). Leaf anatomy in grass systematics. *Bot. Gaz.*, **119**, 170.

BROWN, W. V. & SMITH, B. N. (1972). Grass evolution, the Kranz syndrome, $^{13}C/^{12}C$ ratios, and continental drift. *Nature, Lond.*, **239**, 345.

COOPER, J. P. (1965). The evolution of forage grasses and legumes. In: *Essays on Crop Plant Evolution* (Ed. by J. Hutchinson), p. 142. Cambridge University Press, London.

DARLINGTON, C. D. & WYLIE, A. P. (1955). *Chromosome Atlas of Flowering Plants.* George Allen and Unwin Ltd., London.

DAS, V. S. R. (1974). Stomatal dynamics, carbon economy and C4 photosynthesis in plant life. *Ind. J. Plant Physiol.*, **17**, 1.

DAS, V. S. R. & RAGHAVENDRA, A. S. (1973). A screening of the dicotyledonous weed flora for the occurrence of C_4 dicarboxylic acid pathway of photosynthesis. *Proc. Ind. Acad. Sci. Part B*, **77**, 93.

DOWNTON, W. J. S. (1971). Check list of C_4 species. In: *Photosynthesis and Photorespiration* (Ed. by M. D. Hatch, C. B. Osmond and R. O. Slatyer), p. 455. Wiley Interscience, New York.

HARTLEY, W. (1950). The global distribution of tribes of the Gramineae in relation to historical and environmental factors. *Aust. J. agric. Res.*, **1**, 355.

HARTLEY, W. (1958a). Studies on the origin, evolution and distribution of Gramineae. I. The tribe Andropogoneae. *Aust. J. Bot.*, **6**, 116.

HARTLEY, W. (1958b). Studies on the origin, evolution and distribution of Gramineae. II. The Paniceae. *Aust. J. Bot.*, **6**, 343.

HATCH, M. D., OSMOND, C. B. & SLATYER, R. O. (1971). (Ed.) *Photosynthesis and Photorespiration.* Wiley Interscience, New York.

HATCH, M. D. & SLACK, C. R. (1970). The C4-dicarboxylic acid pathway of photosynthesis. In: *Progress in Phytochemistry*, Vol. 2 (Ed. by L. Reinhold and Y. Lipschitz), p. 35. Interscience, London.

HUTCHINSON, J. (1959). *The Families of Flowering Plants.* Vol. II. *Monocotyledons.* Oxford University Press, London.

KORTSCHAK, H. P., HARTT, C. E. & BURR, G. O. (1965). Carbon dioxide fixation in sugarcane leaves. *Plant Physiol., Lancaster*, **40**, 209.

MCWILLIAM, J. R. & MISON, K. (1974). Significance of the C_4 pathway in *Triodia irritans* (Spinifex), a grass adapted to arid environments. *Aust. J. Plant Physiol.*, **1**, 171.

METCALFE, C. R. (1960). *Anatomy of the Monocotyledons.* I. *Gramineae.* Oxford University Press, London.

METCALFE, C. R. (1971). *Anatomy of the Monocotyledons.* II. *Cyperaceae.* Oxford University Press, London.

PARODI, L. R. & FRIER, F. (1945). Observaciones taxonomicas sobre las gramines estipeas. *Ciencia & Investigation*, **1**, 144.

RAGHAVENDRA, A. S. (1975). *A comparative study of C_4 and C_3 photosynthetic systems in some mono- and dicotyledonous plants.* Ph.D. Thesis. Sri Venkateswara University, Tirupati.

TREGUNNA, E. B., SMITH, B. N., BERRY, J. A. & DOWNTON, W. J. S. (1970). Some methods for studying the photosynthetic taxonomy of the Angiosperms. *Can. J. Bot.*, **48**, 1209.

WYNN, T., BROWN, H., CAMPBELL, W. H. & BLACK, C. C. (1973). Dark release of $^{14}CO_2$ from higher plant leaves. *Plant Physiol., Lancaster*, **52**, 288.