

Developmental Changes in Photosynthetic Gas Exchange in the Polyol-Synthesizing Species, *Apium graveolens* L. (Celery)¹

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ABSTRACT

Developmental changes in photosynthetic gas exchange were investigated in the mannitol synthesizing plant celery (*Apium graveolens* L. 'Giant Pascal'). Greenhouse-grown plants had unusually high photosynthetic rates for a C₃ plant, but consistent with field productivity data reported elsewhere for this plant. In most respects, celery exhibited typical C₃ photosynthetic characteristics; light saturation occurred at 600 micromoles photons per square meter per second, with a broad temperature optimum, peaking at 26°C. At 2% O₂, photosynthesis was enhanced 15 to 25% compared to rates at 21% O₂. However, celery had low CO₂ compensation points, averaging 7 to 20 microliters per liter throughout the canopy. Conventional mechanisms for concentrating CO₂ were not detectable.

MATERIALS AND METHODS

Plant Material. Celery plants (*Apium graveolens* L. 'Giant Pascal') were grown in a greenhouse with supplemental lighting as previously described (17). For measurements, leaves were sequentially numbered beginning with the youngest visible leaf (approximately 3 cm long = leaf No. 1) and categorized as young (leaf No. 1-4), first fully expanded (leaf No. 5-7), and mature (leaf No. 8 and greater). In general, young plants had 11 to 13 leaves, while mature plants had 18 to 21 leaves.

Gas Exchange Measurements. The three terminal leaflets of each leaf were sealed into a Plexiglas chamber and photosynthesis and transpiration measurements were made using an open IR gas analysis system described previously (18). Unless otherwise indicated, measurements were carried out in an atmosphere of 300 to 340 $\mu\text{l CO}_2 \text{ L}^{-1}$ flowing over the leaflets at approximately 50 to 75 L h^{-1} , with a leaf temperature of 26°C. Photon flux densities of 1200 to 2400 $\mu\text{mol photons m}^{-2} \text{ s}^{-1}$ at leaf blade level were provided by 1000 W Lucalux lamps. Light intensity was varied by shading the chamber with layers of Mira cloth. Leaves not being measured were shaded to prevent desiccation. Leaf areas were determined using a LiCor 3100 area meter. CE,³

Acyclic sugar alcohols (polyols) are primary photosynthetic products and the major form of translocated carbon in many higher plants (3, 20). Although physiological roles for polyols have been well established from studies in fungi, algae, and animals (7, 13, 19), little research has been done in higher plants to determine their roles in plant metabolism.

With the exception of apple, no detailed studies of photosynthesis in higher plants that synthesize polyols have been conducted. And even in apple, rarely has photosynthesis been studied developmentally (11, 21). Photosynthetic rates in apple, a sorbitol synthesizer, are generally very high (2, 11) and substantially higher than other trees (12) or many C₃ plants. In a survey of photosynthesis in trees (12), among the highest values reported were for *Pyrus*, a sorbitol synthesizer, and *Fraxinus*, a mannitol synthesizer. Whether these observations are related to polyols being the principal product of photosynthesis in these plants remains to be determined.

In the study reported here, our objective was to study photosynthesis in celery (*Apium graveolens*), which is both a mannitol synthesizer (17) and capable of tremendous biomass production (5). In a preliminary report (14), we demonstrated that celery is capable of high photosynthetic rates. Here, we present a more complete analysis of the gas exchange characteristics of celery in relation to leaf development.

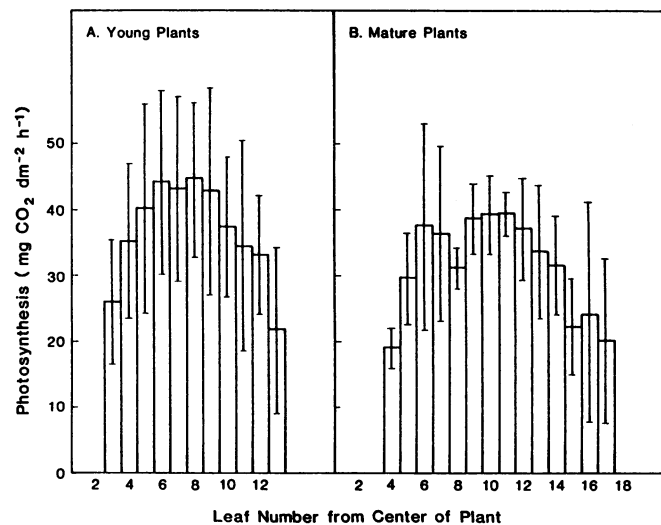


FIG. 1. Photosynthetic rates in leaves of (A) young and (B) mature celery plants as a function of leaf age within the canopy. Values are means \pm SD for 14 to 19 replications in young plants except for leaf No. 12 and 13 (6 and 2 replications, respectively) and in mature plants (4 replications).

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³ Abbreviations: CE, carboxylation efficiency; P, net photosynthesis; C_i, internal CO₂ concentration; R_m, mesophyll resistance to CO₂ transfer; Γ , CO₂ compensation point; PEP, phosphoenolpyruvate.

defined as $\Delta P/\Delta C_i$, and R_m were calculated according to Ehleringer and Björkman (4).

CO₂ Compensation Point. The three terminal leaflets of a celery leaf were excised and sealed in a 125-ml glass tube with the petiole immersed in 10 ml deionized H₂O. The tube was submerged in a water bath at 25°C and irradiated with approximately 1200 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$. Γ was determined with an IR gas analyzer in a closed system by circulating air over the leaflets

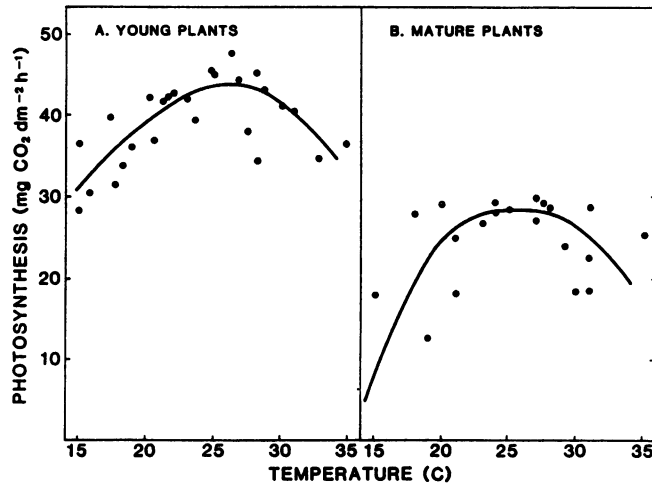


FIG. 2. Photosynthetic rates in leaves of (A) young and (B) mature celery plants as a function of temperature. Measurements were made on leaves No. 4 to 7. Each curve is a composite of data collected from three plants.

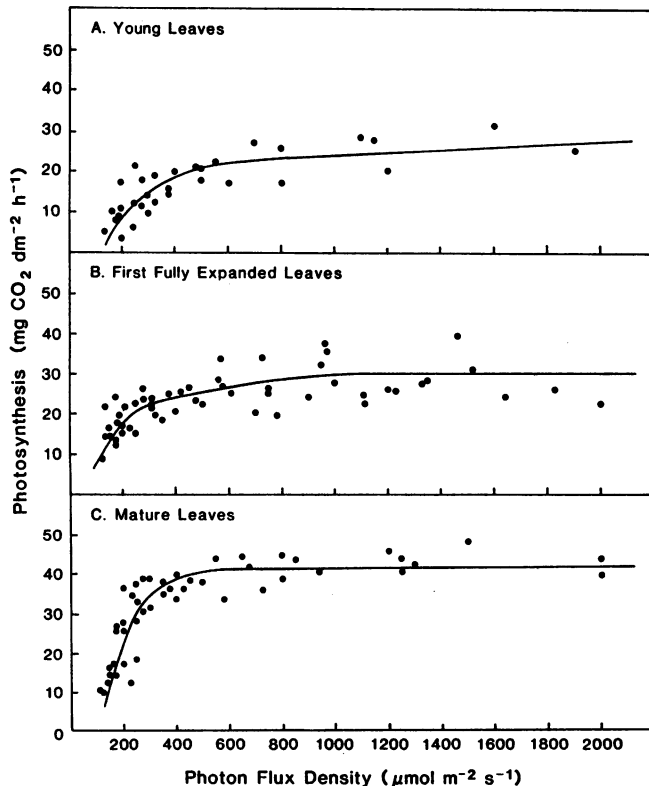


FIG. 3. Photosynthetic rates in celery leaves as a function of photon flux density in relation to leaf age: (A) young leaves, leaves No. 1 to 4; (B) first fully expanded leaves, leaves No. 5 to 7; and (C) mature leaves, leaves No. 8 and greater. Curves are composites of data collected from three young leaves, six first fully expanded leaves, and five mature leaves.

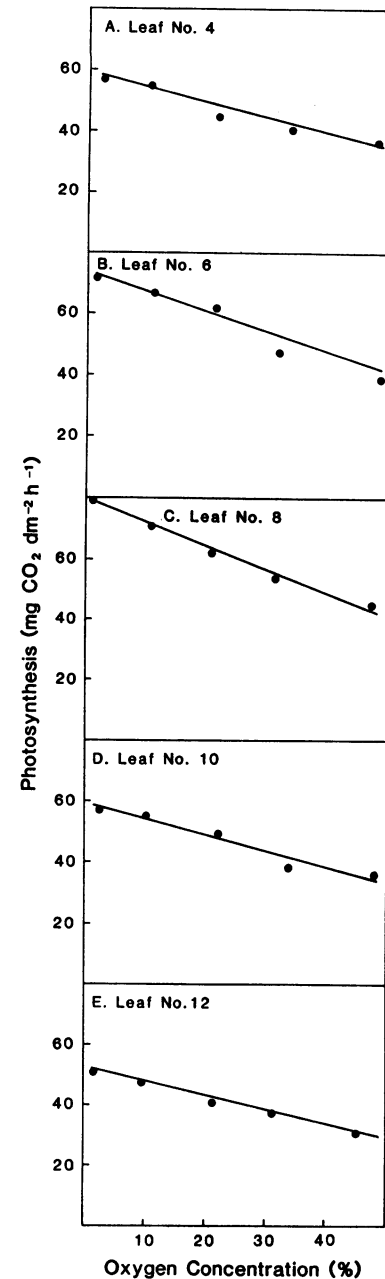


FIG. 4. Photosynthetic response to changes in ambient O₂ concentration at several stages of leaf development in young plants. Linear regression analysis yielded the following equations: (A) Leaf No. 4, $y = 57.71 - 0.46x$, $r = -0.97$; (B) Leaf No. 6, $y = 73.78 - 0.73x$, $r = -0.98$; (C) Leaf No. 8, $y = 79.59 - 0.77x$, $r = -0.97$; (D) Leaf No. 10, $y = 58.30 - 0.48x$, $r = -0.97$; and (E) Leaf No. 12, $y = 51.51 - 0.44x$, $r = -0.98$.

until the CO₂ concentration in the system was reduced to a steady state level. Soybean (*Glycine max*) and corn (*Zea mays*) were used for C₃ and C₄ comparison values of Γ .

RESULTS

Attempts to relate photosynthesis and crop productivity require photosynthetic measurements at several stages of leaf development (21). In the present study with celery, the highest photosynthetic rates were seen in young vigorously growing plants. In such plants, the highest rates were found in leaf No. 6 to 9 (Fig. 1A). Mature plants, on the other hand, exhibited maximum photosynthetic rates in leaf No. 6 to 14 (Fig. 1B). Not

until the leaves were quite old (leaf No. 15 and greater) did photosynthetic capacity decline.

Temperature response was typical of C_3 plants: the temperature optimum occurred over a broad range, peaking at 26°C (Fig. 2). With respect to temperature, young and mature plants responded similarly except that young plants (Fig. 2A) exhibited higher photosynthetic rates over the temperature range studied than did mature plants (Fig. 2B). Light saturation occurred at relatively low levels, 600 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$, for all leaf ages studied but older leaves had higher photosynthetic rates than younger leaves (Fig. 3).

In addition to changes in photosynthesis, photorespiration also changed with leaf development, as seen by the photosynthetic response at various O_2 concentrations (Fig. 4). In general, net CO_2 fixation decreased linearly between 1 and 50% O_2 . Compared to photosynthetic rates at 21% O_2 , photosynthesis was enhanced by 12 to 31% at 2% O_2 and inhibited by 17 to 37% O_2 at 47% O_2 . Sensitivity to O_2 varied with leaf age; leaf numbers 6 to 8 were most responsive to changes in O_2 concentration (see slopes of regression lines in legend of Fig. 4).

Photosynthetic response to changes in CO_2 concentration at 2 and 21% O_2 was characterized at various stages of leaf development. In young plants (Table I) the degree of photosynthetic enhancement under 2% O_2 increased with leaf age from leaf Nos. 6 to 10. This increase was accompanied by a decrease in CE and an increase in R_m , as indicated by the decrease in slope of the CO_2 response curves. Conversely, a decrease in both photosynthetic enhancement and R_m and an increase in CE occurred from leaf Nos. 4 to 6. Mature plants exhibited similar trends, although the patterns were shifted; leaf No. 10 exhibited the lowest photosynthetic enhancement and R_m and the largest CE (Table II).

Leaf development also affected CO_2 compensation point in celery (Fig. 5). The largest difference we observed was in young leaves. Γ decreased from 55 $\mu\text{L L}^{-1}$ in leaf No. 3 to 11 $\mu\text{L L}^{-1}$ by the time the leaf had reached leaf age 4. Within the remainder

of the canopy, Γ remained exceptionally low; leaf No. 8 in young plants and leaf No. 7 in mature plants exhibited the lowest values (7.7 and 9.6 $\mu\text{L L}^{-1}$, respectively). Older leaves exhibited a slight trend toward increased Γ , although even in the oldest leaves, Γ increased to only 15 $\mu\text{L L}^{-1}$ (young plants) and 20 $\mu\text{L L}^{-1}$ (mature plants). These values are remarkably low for a C_3 plant. For comparative purposes, we determined Γ for soybean and corn using the same methodology that we employed for celery. The values for both soybean (42.1 $\mu\text{L L}^{-1}$) and corn (1.6 $\mu\text{L L}^{-1}$) are typical for those C_3 and C_4 species, respectively.

DISCUSSION

Celery leaves are capable of exceptionally high rates of photosynthesis compared to commonly reported values for C_3 plants. When studied in the context of biomass production, however, these rates are also consistent with field productivity data (Table III). Celery produces more biomass per unit land area than other crop plants, with the possible exception of sugar cane. Indeed, the photosynthetic rates we obtained for celery (35–65 $\text{mg CO}_2 \text{ dm}^{-2} \text{ h}^{-1}$) compare favorably with those reported for sugar cane (42–49 $\text{mg CO}_2 \text{ dm}^{-2} \text{ h}^{-1}$) (8, 9). Such high photosynthetic rates for celery must contribute to this plant's high productivity. The large biomass production in celery may also result from an ability of the leaves to maintain maximum photosynthetic rates longer, a trait especially important in older plants where a large fraction of the canopy consists of mature leaves.

In addition to high photosynthetic rates, celery exhibits another interesting photosynthetic characteristic: its CO_2 compensation points are unusually low for a C_3 plant. Conventional explanations for low Γ revolve around structural or enzymological adaptations (15). Preliminary anatomical studies revealed that young celery leaves had a compact mesophyll, but did not have a well developed bundle sheath (ME Rumpho, personal communication). These observations confirm earlier anatomical investigations done at the light microscopy level (6). At the

Table I. Photosynthetic Enhancement under 2% O_2 , Carboxylation Efficiency, and Mesophyll Resistance in Young Plants

The plants had 11 to 13 leaves.

Leaf No.	Photosynthesis at 300 $\mu\text{L L}^{-1}$		Enhancement	Carboxylation Efficiency	Mesophyll Resistance
	21% O_2	2% O_2			
	$\text{mg CO}_2 \text{ dm}^{-2} \text{ h}^{-1}$		%	cm s^{-1}	
4	37.08	43.89	18.4	0.14	6.91
6	32.96	34.65	5.1	0.11	9.26
8	49.89	58.26	16.8	0.18	5.49
10	33.03	43.46	24.0	0.14	7.14
12	28.58	34.28	16.6	0.11	9.09

Table II. Photosynthetic Enhancement under 2% O_2 , Carboxylation Efficiency, and Mesophyll Resistance in Mature Plants

The plants had 18 to 21 leaves.

Leaf No.	Photosynthesis at 300 $\mu\text{L L}^{-1}$		Enhancement	Carboxylation Efficiency	Mesophyll Resistance
	21% O_2	2% O_2			
	$\text{mg CO}_2 \text{ dm}^{-2} \text{ h}^{-1}$		%	cm s^{-1}	
4	8.58	15.58	81.6	0.03	38.05
6	34.96	41.98	20.1	0.13	7.44
8	47.89	49.89	6.0	0.16	6.21
10	58.79	56.16	-4.5	0.23	4.27
12	57.49	62.77	9.2	0.23	4.27
14	38.02	44.29	16.5	0.14	7.30
16	18.41	25.55	38.8	0.07	15.02
18	24.36	35.30	24.4	0.09	10.75

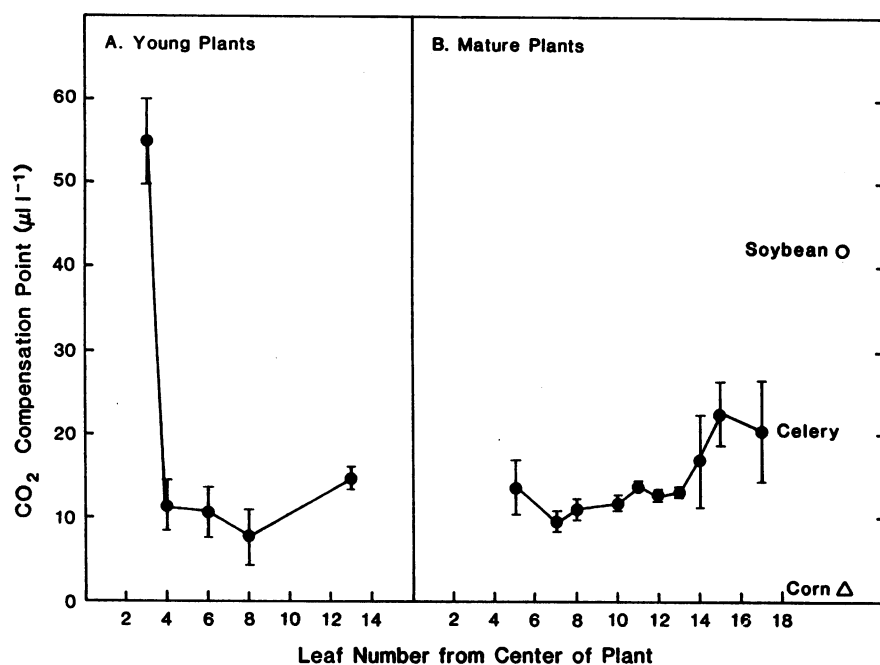


FIG. 5. CO₂ compensation points (Γ) as a function of leaf age in (A) young and (B) mature celery plants. Γ was determined by depletion of CO₂ in a closed system from ambient CO₂ level to a steady state level. Values are means \pm SD for four replications in young plants and two replications in mature plants. Values for soybean and corn are presented for comparative purposes.

Table III. Harvestable Yields Relative to Celery Productivity for Selected Crops on a Fresh Weight Basis
Data are 1982 production values compiled from Agricultural Statistics 1983, USDA (1). No attempt was made to correct to dry matter productivity.

Crop	Harvestable Yield metric tons/ha	Harvestable Yield Relative to Celery %	Crop	Harvestable Yield metric tons/ha	Harvestable Yield Relative to Celery %
Celery	54.92	100.0	Sunflower	1.30	2.4
Sugar beet	46.17	84.1	Soybean	2.16	3.9
Lettuce	31.37	57.1	Wheat	2.39	4.4
Carrots	32.95	60.0	Barley	3.68	6.7
Cauliflower	11.88	21.6	Rice	5.31	9.7
Potatoes	30.71	55.9	Rye	1.83	3.3
Onions	37.30	67.9	Cotton	0.66	1.2
Corn	9.01	16.4	Sweet corn	4.97	9.0
Sugar cane	71.00	129.3	Tobacco	2.45	4.5

ultrastructural level, Holaday *et al.* (10) have described an arrangement of organelles that may contribute to the C₃-C₄ nature of *Moricandia arvensis*. In *Moricandia*, bundle sheath cells contain a higher proportion of mitochondria, peroxisomes, and chloroplasts than the surrounding mesophyll cells. Furthermore, the organelles are arranged centripetally, with an abundance of mitochondria located along the cell membrane adjacent to the vascular bundle. This arrangement presumably allows for more efficient recycling of photorespiratory CO₂ before it can exit the leaf. The occurrence of similar mechanisms in celery remains to be determined.

In still other plants, CO₂ concentrating mechanisms require high activity of PEP carboxylase or carbonic anhydrase (15). Preliminary studies of the enzymology and ¹⁴C-fixation products in celery, however, indicate typical C₃ values for enzymes and metabolites (14). Currently, we have no evidence that an alternative to the reductive pentose phosphate pathway operates in celery tissues. The only unusual aspect of its carbon metabolism appears to be mannitol synthesis.

The relationship between polyol metabolism and photosynthesis may be analogous to the relationship between photorespiration and photosynthesis. One role postulated for photorespiration is dissipation of excess energy produced photochemically, thereby preventing photoinhibition of CO₂ fixation (16). The

stoichiometry of mannitol synthesis requires up to one third of the triose-P exported from the chloroplast for regeneration of reductant (17). Consequently, mannitol synthesis could present an alternative mechanism for dissipating reductant, possibly reducing the need for photorespiration. Our data, however, provide only circumstantial evidence to support this argument.

Despite the exceptionally low Γ values in celery, present evidence indicates an otherwise normal C₃ pattern of photosynthesis. Undoubtedly, the reduction of apparent photorespiration in this species, as evidenced by the low values for Γ , contribute to the high photosynthetic rates reported here and the large biomass production noted elsewhere.

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