Short Communication

Photosynthetic $^{14}CO₂$ Fixation by Green Cymbidium (Orchidaceae) Flowers

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Orchid flowers occur in a great variety of colors and color combinations (2). Several genera, among them $Cymbidium$, contain species which bear green flowers. The pigment in these flowers was assumed to be chlorophyll for many years despite the absence of analytical evidence (1). Recent investigations (1, 10) have confirmed this assumption and have shown that green Cymbidium flowers owe their color to chlorophyll which, as might be expected, is localized in chloroplasts.

These chloroplasts have a normal appearance when examined under oil immersion, but lose their integrity with flower senescence. (T. R. Pray, University of Southern California, personal communication). Since chlorophyll containing stems and roots of several orchid species are capable of photosynthesis $(6, 7, 8)$ it appeared reasonable to assume that green $Cymbidium$ flowers might have the same capability. However, the occurrence andl contributions of photosynthesis in orchid flowers have not been investigated to date.

Flowers of Cymbidium XChelsea and C . XIndependence Day 'Yorktown' were used for the experiment. Since these flowers are produced on indeterminate racemes which have a slow flower-opening sequence, it was possible to obtain buds (arbitrarily designated as day 0), newly opened flowers (3 days later) and futlly opened flowers (7 days from bud) from each raceme. All experiments were repeated twice with flowers from 2 racemes of each Cym bidium, cross.

The experiments were carried out in sealed vessels half of which were painted black and covered with aluminum foil to serve as dark controls. Each vessel was fitted with a glass tuibe covered with an ampule cap and reaching into a shallow glass container within the experimental vessels. Fourteen μ c of Na₂¹⁴CO₃ in an aqueous carrier solution, $(377470 \text{ cpm/ml}, \text{pH } 8)$, adjusted to contain a total of 1.6 mm $CO₂$, were injected through the ampule cap followed by 7.5 ml of ¹ N $H₂SO₄$. After 15 minutes in the dark, to allow for gas equilibration, the unpainted vessels were placed in a well ventilated hood and subjected to illumination for 45 minutes. The light source consisted of one 300 watt, two 200 watt Westinghouse incandescent bulbs and one 12 inch cool-white Westinghouse fluorescent bulb producing a total of 0.13 w/cm^2 at a distance of 30 cm. (J. R. Jones, Westinghouse Electric Corporation, Los Angeles, personal communication). This somewhat long exposure period was chosen because metabolic processes and growth are relatively slow in orchids (2, 4, 5). Temperature was maintained at 22° by using 20 cm wide water-filled museum jars as heat filters between the lights and the reaction vessels.

The reaction was terminated by turning the lights off and freeing the vessels of CO₂ by

FIG. 1. $^{14}CO_2$ fixation by Cymbidium buds, newly and fully opened flowers and leaves. S-sepals; Ppetals; 0-ovary.

evacuation through 2 barium hydroxide traps. The flowers were then dissected, their parts weighed separately, and extracted in boiling 80% (v/v) ethanol (9). Chlorophyll content of flowers at the same stages of development from the same racemes was determined by the method of Arnon $(3, 9)$. Radioactivity of duplicate samples of the extract, was determined in a liquid scintillation counter.

Since the amount of chlorophyll in each sample varied, quenching was determined with an internal standard. The results (figs 1, 2) indicate that green Cymbidium flowers are capable of CO, fixation in the light, and that fixation rates vary between flower parts and varieties. In both crosses the amount of $CO₂$ fixation is highest in the sepals, lower in the petals and lowest in the ovary. This implies certain metabolic differences between the sepals and petals which are very similar in appearance and therefore often referred to by the common term tepals. It is also interesting to note that the flowers can fix $CO₂$ in the dark.

In Cymbidium \bar{X} Chelsea the efficiency of ¹⁴C fixation decreases with age, whereas in C . XIndependence Day 'Yorktown' it increases in the 3-day old flower and may or may not drop in the 7-day

FIG. 2. Net $14CO₂$ fixation by flowers in the light as a function of age in $Cymbdium$ XChelsea and C. XIndependence Day 'Yorktown'. The bar, representing $^{14}CO_{2}$ fixation by a mature leaf, is included for comparison purposes.

Table I. Net Carbon Fixation in the Light by Orchid Plant Organs Expressed as Percent of the Amount Fixed by Leaves

	Cymbidium		Epidendrum	Cattleya	Phalaenopsis	<i>Vanda</i>	
Organ	X Chelsea	XIndep. Day 'Yorktown'	xanthinum (6)	gigas (6)	hybrids (6)	suavis (6)	Cattleya (7, 8)
Bud							
Sepals	13.6	1.0					
Petals	5.5	0.1					
Ovary	1.2	0.4					
Newly opened							
Flowers							
Sepals	10.7	11.9					
Petals	0.5	1.0					
Ovary	0.1						
Fully opened							
Flowers							
Sepals	1.4	4.9					
Petals	0.6	1.1					
Ovary	0.2	0.4					
Lower leaves			100			100	
Upper leaves			111			129	
Stems and leaves			80				
Stem				113			
Leaf and stem					100		
Root			1814	645	3689		
Leaf	100	100		100			
1 yr old							100
2 yrs old							103
3 vrs old							103
4 yrs old							91.6
5 yrs old							70
6 yrs old							50
7 yrs old							38

old one (fig 2). This seems to indicate different rates of maturation and/or senescence of the photosynthetic apparatus in the flowers of each cross.

Orchids like Microcoelia smithii, Polyrrhiza *funalis* and *P*. *lindenii* which are leafless and almost stemless, but have well developed aerial roots and flower spikes, probably depend entirely upon the photosynthetic products produced by their roots $(6, 7)$. Roots of other orchids like Cattleya labiata, Cattleya hybrids, Epidendrum xanthinum, Phalaenopsis schilleriana, and Vanda suavis have a well developed photosynthetic capability and are apparently sufficiently autotrophic to either maintain themselves or contribute substantially to their needs $(6, 7)$. It is interesting therefore to compare the amount of photosynthesis in various organs to that in Cymbidium flowers. This can be done easily if in each case net photosynthesis in leaves is arbitrarily assigned a value of 100% and net photosynthesis in other organs expressed as fractions of that (table I). Such comparison indicates that although green Cymbidium flowers can fix CO₂ in the light their contribution of photosynthetic products is small.

The dark fixation of CO., is also of considerable interest. It is reminiscent of the behavior of thick (i.e. fleshy and succulent) orchid leaves. Such leaves exhibit increased acidity (12) and CO₂ uptake (11) in the dark, under certain conditions, in a fashion similar to that of the Crassulaceae, cacti and other succulents with known Crassulacean acid metabolism and dark fixation of CO.. However, these leaves can also fix CO₂ in the light (E. L. Nuerenbergk, Staatsinstitut fur Allegemeine Botanik und Botanischer Garten, Hamburg, Germany, personal communication). The Cymbidium flower parts used in this experiment were fleshy and therefore perhaps capable of dark CO. fixation in a manner not unlike that of fleshy orchid leaves.

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