

Stomatal Limitation to Carbon Gain in *Paphiopedilum* sp. (Orchidaceae) and Its Reversal by Blue Light¹

Received for publication June 12, 1984

EDUARDO ZEIGER,* C. GRIVET, SARAH M. ASSMANN, GERALD F. DEITZER, AND M. W. HANNEGAN
Department of Biological Sciences, Stanford University, Stanford, California 94305 (E.Z., C.G., S.M.A.);
and Smithsonian Institution, Environmental Research Center, 12441 Parklawn Drive, Rockville, Maryland
20852-1773 (G.F.D., M.W.H.)

ABSTRACT

Leaves from *Paphiopedilum* sp. (Orchidaceae) having achlorophyllous stomata, show reduced levels of stomatal conductance when irradiated with red light, as compared with either the related, chlorophyllous genus *Phragmipedium* or with their response to blue light. These reduced levels of stomatal conductance, and the failure of isolated *Paphiopedilum* stomata to open under red irradiation indicates that the small stomatal response measured in the intact leaf under red light is indirect.

The overall low levels of stomatal conductance observed in *Paphiopedilum* leaves under most growing conditions and their capacity to increase stomatal conductance in response to blue light suggested that growth and carbon gain in *Paphiopedilum* could be enhanced in a blue light-enriched environment. To test that hypothesis, plants of *Paphiopedilum acmodontum* were grown in controlled growth chambers under daylight fluorescent light, with or without blue light supplementation. Total photosynthetic photon flux density was kept constant in both conditions. Blue light enrichment resulted in significantly higher growth rates—of up to 77%—over a 3 to 4 week growing period, with all evidence indicating that the blue light effect was a stomatal response. Manipulations of stomatal properties aimed at long-term carbon gains could have agronomic applications.

matal apparatus devoid of guard cell chloroplasts, provide information on the properties of stomatal movements in the absence of these organelles (12). *Paphiopedilum* stomata are functional, but fail to respond to red light in isolation (19). Very low, steady-state conductance levels in these orchids may also be correlated with their lack of guard cell chloroplasts (15).

In the work reported in this paper, we studied the properties of *Paphiopedilum* stomata in the intact leaf using gas exchange to measure net photosynthesis and stomatal conductance under blue and red light irradiation. These responses were compared with those of a related genus, *Phragmipedium*, of broadly similar morphology and habitat, but having chlorophyllous stomata. *Paphiopedilum* leaves irradiated with red light showed markedly reduced conductance levels and concomitant, low levels of intercellular CO₂ concentrations. These observations suggested that an enrichment of blue light in the environment of *Paphiopedilum* plants could increase the unusually low levels of conductance seen in these leaves under most environmental conditions, and result in a higher intercellular CO₂ concentration, which would allow increased rates of CO₂ fixation and growth. A test of that hypothesis confirmed that *Paphiopedilum* plants growing in a blue light-enriched environment have a higher growth rate, indicating that the manipulation of stomatal responses can increase leaf productivity.

MATERIALS AND METHODS

Plant Material. Plants of *Paphiopedilum insigne*, *Pa. harrisianum*, *Pa. barbatum*, *Pa. villosum*, *Pa. philippinense*, *Phragmipedium longifolium*, and *Ph. albopurpureum* were purchased from a local nursery (Rod McLellan, South San Francisco, CA). *Ph. longifolium* had been reported previously as *Ph. vittatum* (5, 8). The plants were originally labeled as *vittatum* when imported by the supplier and reproduced vegetatively under that classification. Specimens kept in our greenhouse have since flowered, making it possible to determine that they were, in fact, *Ph. longifolium*. We thank the Orchid Identification Center, Sarasota, FL for the taxonomic identification and Dr. R. Griesbach, USDA, Beltsville, MD, for pointing out the rarity of the *vittatum* specimens and the likelihood of a misidentification.

All plants except *Pa. acmodontum* were grown in a greenhouse at Stanford under natural daylight. Temperatures ranged between 15 and 25°C. As supplied by the grower, plants arrived in pots filled with redwood bark chips and were maintained under the same conditions throughout. To generate a microenvironment with high relative humidity, the plants were kept in a polyethylene plastic tent built on a greenhouse bench. The tent also decreased prevailing light intensities to a maximum of 750 $\mu\text{mol m}^{-2} \text{s}^{-1}$. The plants were watered daily and fertilized weekly with Spoonit Orchid food (30% nitrogen, 10% phosphoric acid, 10%

Stomata provide plants with a primary means for adjusting to changes in the environment and usually constitute the main resistance in the pathway of CO₂ uptake in leaves (4, 17). Because of their impact on CO₂ uptake, stomata impose limitations on photosynthesis in the mesophyll (7).

Prevailing stomatal conductances are a function of the physical dimensions of the stomatal pores, which are controlled by guard cell turgor. Turgor pressures in guard cells, in turn, are modulated by metabolic events transduced from guard cell responses to external and internal stimuli. Because guard cells respond simultaneously to different stimuli, any given conductance value represents an integrated output of the guard cell to prevailing environmental signals (17).

Chloroplasts are one of the functional components of guard cells. Their precise role in stomatal movements is not fully understood, but available data indicate that they participate in signal transduction in guard cells via wavelength sensitivity, and provide metabolic energy via photophosphorylation (17). Studies on the achlorophyllous stomata from the genus *Paphiopedilum* (Orchidaceae), which have the characteristic responses of a sto-

¹ Supported by the Department of Energy (81ER10924) and the National Science Foundation (PCM 8214378).

potassium salts, 0.1% Ferric DTPA, PlantSmith, Mountain View, CA) at about 1.5 g/l. Plants of *Pa. acmodontum* were obtained from the orchid collection at the Smithsonian Institution, Office of Horticulture in Washington, D.C., and were divided into 40 plants of relatively uniform size and then weighed to obtain total fresh weight before being repotted in 10 cm plastic pots with a fir bark-black sand mix (Sequoia Orchid Bark-Paph Mix) purchased from a local nursery (Kensington Orchid, Kensington, MD). After repotting, they were used in the growth experiments at Rockville.

Gas Exchange. Gas exchange experiments with *Pa. insignis* were conducted at Stanford in an open, differential system (16) at ambient CO₂ concentration of 300 to 320 μl l⁻¹, a VPD² of 0.35 to 0.40 kPa and a leaf temperature of 20°C. The light source was a M1000 Metalarc lamp (Sylvania GTE, Danvers, MA). Red or blue light was provided by a Kodak SAFE No. 1A (50% cutoff at 645 nm) or a Rohm and Haas Deep Blue Plexiglass No. 2424 (maximal transmittance at 470 nm; half band width, 100 nm) filter. Photon fluxes were measured with a silicon cell located in the leaf cuvette and calibrated against a quantum sensor (Li-Cor).

Porometry. At Rockville, measurements of stomatal conductance were obtained with a Li-Cor (Model LI-1600) steady-state porometer. Several plants, selected from among those used in the growth experiments, were transferred to a growth chamber equipped with a high-voltage DC drive fluorescent lamp canopy and allowed to equilibrate overnight in the dark. Conductances were measured on randomly selected individual leaves from separate plants. Photon fluence response curves were obtained using both daylight fluorescent (Sylvania F48T12/D/VHO) and daylight fluorescent supplemented with a 1:1 ratio of blue phosphor fluorescent lamps (Sylvania F48R12/246/VHO) with a spectral distribution shown in Figure 1. The emission spectra were measured with a spectral-radiometer (Gamma Industries, Model C-3, San Diego, CA) which was used to calibrate the Li-Cor quantum sensor attached to the porometer. Stomatal conductances were measured continuously at each irradiance until no change was observed over a 15 min period.

Growth Experiments. For the growth experiments, conducted at Rockville, freshly repotted *Pa. acmodontum* plants were randomized and placed in growth chambers with either daylight

fluorescent or daylight fluorescent supplemented with blue phosphor fluorescent lamps. The total PPFD was equalized in both conditions (Table I) by using a Gamma C-3 spectral-radiometer to measure the emission spectrum from 300 to 800 nm and multiplying the obtained values by the action spectrum for photosynthetic quantum yield (10). The PPFD was remeasured at weekly intervals and maintained constant to within 5% of the initial values. Both chambers were on 16 h photoperiods; temperatures were 25 ± 0.5°C during the day and 15 ± 0.5°C at night. RH was 70 ± 10%; CO₂ concentrations ranged from 380 to 450 μl l⁻¹. Plants were watered every 2 to 3 d with one-fourth strength Hoagland solution (9).

Leaf area measurements were made weekly from tagged leaves on each plant. Since only a limited number of plants was available, and since the plants grow very slowly, fresh weights were determined only at the end of each 33 to 42 d experimental period. Plants were then repotted, randomized again, and used for the next experiment. The results in Table III are, therefore, based on the initial 40 cloned individuals.

Statistics. In the growth experiments, the statistical significance of the differences between means (Table III) was assessed with a *t* test. For the grouped data of all three experiments, a *t* test was also used with $t = (\sum \Delta_i N_i) / (\sum N_i (S_{xi}^2 + S_{yi}^2))$, where Δ_i was the difference between means in experiment *i*, N_i was the corresponding sample size (taken as $(N_{xi} + N_{yi})/2$ in the case of the leaf areas), and S_{xi}^2 and S_{yi}^2 the sample variances for daylight and blue light-enriched daylight, respectively.

Preliminary analysis of the data showed that there was no proportionality between increments and initial values, allowing the use of the data themselves rather than their logarithms, and that there was no significant skewness or other features in the data which would have invalidated a *t* test.

RESULTS

Guard Cell Chloroplasts in *Paphiopedilum* and *Phragmipedium*. Both *Paphiopedilum* and *Phragmipedium* belong to the same subfamily of the Orchidaceae, Cyripedioideae (6), and have similar morphologies and growth characteristics. The two genera, however, occur in geographically separated habitats, with *Paphiopedilum* growing in tropical Asia and *Phragmipedium* in tropical North, Central, and South America. A survey of six *Paphiopedilum* species (*insigne*, *harrisianum*, *acmodontum*, *barbatum*, *ballosum*, and *philipinense*), undertaken with fluorescence microscopy showed that all were devoid of guard cell chloroplasts, as reported previously for *insigne* and *harrisianum* and three other species (*venustum*, *leanum*, and *aureum* *hycanum* (12–14). In contrast, the *Phragmipedium* species investigated in the present study, *longifolium* and *albopurpureum*, showed normally fluorescing guard cell chloroplasts. The structural difference, therefore, appears to be at a generic level and presumably arose as a relatively early evolutionary event.

Net Photosynthesis and Stomatal Conductance in Intact

Table I. Irradiances Calculated from Spectral Emission Curves as Reported in Figure 1 for Daylight Fluorescent or Daylight Fluorescent Supplemented with Blue Phosphor Fluorescent Lamps

Measurements made at six weekly intervals.

	Daylight	Daylight + Blue
	μmol m ⁻² s ⁻¹	
Total irradiance, 300–800 nm	125 ± 5*	125 ± 4
PAR, 400–700 nm	118 ± 6	114 ± 6
PPFD, 400–700 nm	102 ± 5	93 ± 4
Blue irradiance, 400–500 nm	30 ± 3	65 ± 5

* Mean ± SD of six measurements.

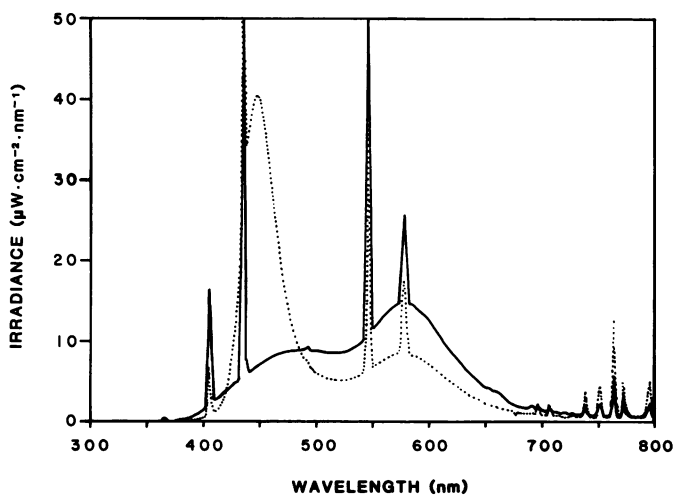


FIG. 1. Spectral emission curves for the daylight fluorescent (—) and the daylight fluorescent supplemented with blue phosphor fluorescent light (····) measured with a Gamma Scientific Model C-3 spectral radiometer calibrated against a NBS standard source (EV-22).

² Abbreviations: VPD, vapor pressure deficit; PPFD, photosynthetic photon flux density.

Leaves of *Paphiopedilum* and *Phragmipedium* under Blue and Red Irradiation. The physiological implications of the lack of guard cell chloroplasts on stomatal movements in the intact leaf were evaluated in gas exchange experiments with *Pa. insignis*. For comparison, identical experiments were performed with *Ph. longifolium*.

Under white light, net photosynthesis and stomatal conductance saturated in both species at around $200 \mu\text{mol m}^{-2} \text{s}^{-1}$ (for light curves of *Pa. insignis*, see Williams *et al.* [15]; data for *Ph. longifolium*, not shown), indicating that the two species have comparable photosynthetic rates.

Under colored light, net CO_2 fixation rates ranged between 2 and $3.5 \mu\text{mol m}^{-2} \text{s}^{-1}$ in *Paphiopedilum* and between 2 and $4.5 \mu\text{mol m}^{-2} \text{s}^{-1}$ in *Phragmipedium*. Curves typical of the relationships between photosynthetic rates, levels of stomatal conductance, and intercellular CO_2 concentrations in the two genera, under blue and red irradiation are shown in Figures 2 and 3. Photosynthetic rates in the light-limited portion of the curves in both *Phragmipedium* and *Paphiopedilum* were higher under red light than under blue, as reported for other species (18). In contrast, stomatal conductances were much lower in red than in blue light, particularly at moderate to high irradiances in *Paphiopedilum*.

Table II shows the blue/red ratios of stomatal conductances in both species, obtained from steady-state values at the highest ($150\text{--}180 \mu\text{mol m}^{-2} \text{s}^{-1}$) irradiance used in three replicate experiments. In *Phragmipedium*, the average blue/red ratio for stomatal conductances was slightly higher than unity, as observed with other species (18). In contrast, the average blue/red ratio for stomatal conductance in *Paphiopedilum* was 2.51. A significant number of additional experiments with *Pa. insignis* treated with red light confirmed the conclusion that *Paphiopedilum* leaves have very low levels of stomatal conductance under red irradiation.

Enhancement of Growth in *Paphiopedilum*, under Blue Light-Enriched Illumination. The slow growth rate and low steady state conductances observed in *Paphiopedilum*, coupled with observations showing that blue light can increase stomatal apertures

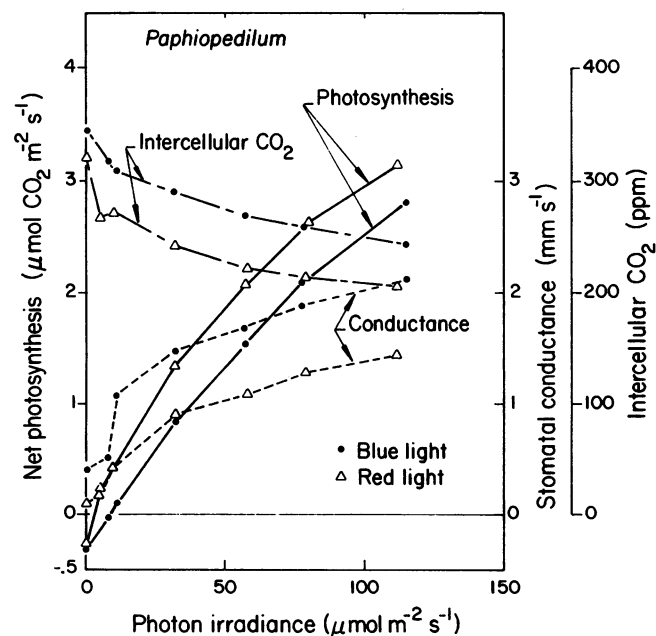


FIG. 2. Net photosynthesis, stomatal conductance, and intercellular CO_2 concentrations in attached leaves of *Pa. insignis* irradiated with blue or red light. Leaf temperature, 20°C ; VPD, $0.35\text{--}0.40 \text{ kPa}$; ambient CO_2 concentration, $300\text{--}320 \mu\text{l l}^{-1}$.

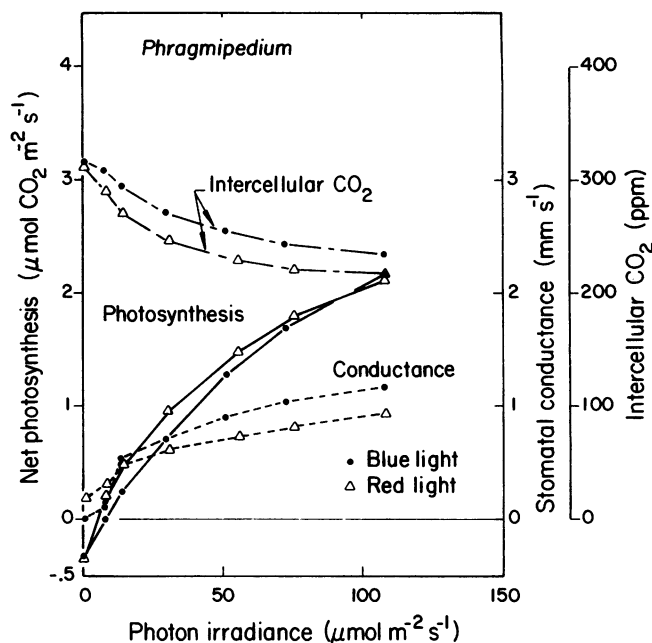


FIG. 3. Net photosynthesis, stomatal conductance, and intercellular CO_2 concentrations in attached leaves of *Ph. longifolium* irradiated with blue or red light. Leaf temperature, VPD, and ambient CO_2 concentrations as in Figure 2.

Table II. Stomatal Conductance in *Pa. insignis* and *Ph. longifolium*

The measurements were made under 150 to $180 \mu\text{mol m}^{-2} \text{s}^{-1}$ of red or blue light and the blue/red ratios, obtained in three different experiments. The same leaf was used for the blue and red light treatment in each experiment.

Stomatal Conductance					
<i>P. insignis</i>			<i>P. longifolium</i>		
Blue	Red	B/R	Blue	Red	B/R
mm s^{-1}		ratio	mm s^{-1}		ratio
0.60	0.50	1.20	1.63	1.45	1.12
2.30	0.47	4.90	1.20	0.92	1.30
2.10	1.45	1.44	1.24	1.34	0.92
Mean = 2.51			Mean = 1.11		

in this species (19), led us to test the long-term effect of blue light supplementation on the growth of this genus. The hypothesis was tested with *Pa. acmodontum* in controlled environment growth chambers available at Rockville. The use of an all-blue environment was avoided to eliminate morphogenetic effects (11). Instead light regimes were designed to simulate daylight (with about 25% of blue quanta in the $400\text{--}500 \text{ nm}$ spectral region) and a blue light-enriched environment with about 50% blue quanta (Table I).

In the first experiment, which lasted 33 d, plants lost fresh weight in both treatments (Table III), due to an unfavorable response to cloning and handling. Enhanced growth under the blue light-enriched environment was, however, evident in this first experiment because of the higher weight loss measured in the daylight treatment. Presumably, the 58.3% increase represents more rapid recovery of the plants in the blue light-enriched environment. In two subsequent experiments, of 42 d each, plants in both treatments gained fresh weight, with those growing in the blue light-enriched environment gaining an average of 76.9% (Exp. 2) and 61.1% (Exp. 3) more weight than the plants growing under the daylight regime. It should be mentioned that, as an additional safeguard against long-term, morphogenetic

Table III. Average Initial and Final Fresh Weights of Plants from *Pa. acmodontum* Growing in Growth Chambers

Plants were weighed prior to repotting before each experiment. After repotting, all plants were randomized and placed in a growth chamber either with daylight fluorescent alone or daylight fluorescent + blue light with the irradiances indicated in Table I.

	Exp. 1 (33 d; n = 19)*		Exp. 2 (42 d; n = 19)		Exp. 3 (42 d; n = 7)		Exp 1-3 Combined
	Daylight	Daylight + blue	Daylight	Daylight + blue	Daylight	Daylight + blue	
Average, initial fresh wt ± SD (g)	18.6 ± 5.9	17.6 ± 7.6	15.7 ± 6.0	14.8 ± 4.7	13.2 ± 2.6	12.2 ± 3.1	
Average, final fresh wt ± SD (g)	15.0 ± 4.5	16.1 ± 6.2	16.9 ± 6.3	17.1 ± 4.9	15.0 ± 3.1	15.1 ± 3.7	
Δ increase in fresh wt ± SD (g)	-3.6 ± 3.2	-1.5 ± 2.9	1.3 ± 2.2	2.3 ± 1.8	1.8 ± 1.2	2.9 ± 0.8	
% increase in fresh wt	-19.3	-8.6	8.0	15.7	13.3	23.9	
% increase in the blue light-enriched envi- ronment		58.3		76.9		61.1	
<i>t</i>		2.43		1.79		1.93	2.98
Significance level (%)		97.5		95.0		95.0	99.5

* *n*, number of plants/treatment.

effects of blue light, each population of plants was randomized at the beginning of each experiment. Leaf areas were also measured in randomly selected leaves in the three experiments (data not shown). The increases in leaf area were consistent with those in fresh weight although variability in the leaf area was higher. Because of that variability, the differences between treatments were only statistically significant ($t = 95\%$) upon pooling of the measurements from all three experiments.

Stomatal densities at the end of experiment 3 were obtained in randomly selected leaves from both experimental treatments. There were no significant differences between treatments (20.6 ± 2.8 and 21.6 ± 2.2 stomata/mm² for blue light-enriched and daylight treatment, respectively; $n = 3$). Light curves of stomatal conductance, measured with a steady-state porometer, were obtained with leaves from plants growing under the two light

regimes. Higher stomatal conductances were measured in the leaves growing under blue light-enriched daylight (Fig. 4).

DISCUSSION

Despite their apparent normalcy (12), *Paphiopedilum* stomata have been shown to be functionally distinct from their chlorophyllous counterparts because of their inability to open, in isolation, in response to red light (19). In this paper, we demonstrate that the lack of guard cell chloroplasts also correlates with unusual stomatal responses to light in the intact leaf. These anomalies, which are evident in comparisons of the stomatal responses to blue and red light in *Paphiopedilum* and on the differential responses of *Paphiopedilum* and *Phragmipedium*, indicate that under red light, a lack of guard cell chloroplasts results in a substantial decrease in stomatal conductance. This conclusion confirms an earlier interpretation of the reported stomatal opening under red light with *Pa. leeanum*, obtained with a transient porometer (12), as an indirect effect (19).

The responses of achlorophyllous stomata can be explained with the hypothesis that normal stomatal movements depend on the function of two photoreceptor systems in the guard cells, one relying on photosynthesis which requires guard cell chloroplasts and a second, nonphotosynthetic, blue light-dependent photosystem (17). Chloroplasts appear essential for direct stomatal responses to red light; in their absence, the red light response seems physiologically equivalent to a dark response. Under blue light, on the other hand, *Paphiopedilum* stomata can rely on the blue light photosystem for their light sensitivity, resulting in a light response despite their lack of chloroplasts.

The ecophysiological implications of the lack of guard cell chloroplasts in *Paphiopedilum* are intriguing. The invariance of this feature in all species of the genus analyzed to date indicate that the evolutionary change resulting in loss of guard cell chloroplasts is functionally stable. Because of physiological data indicating that, in natural conditions, guard cell chloroplast activity is required to support high conductance levels at moderate to high irradiances (17, 18), we have speculated that the loss of guard cell chloroplasts has been evolutionarily stable because of the unusually low levels of stomatal conductance

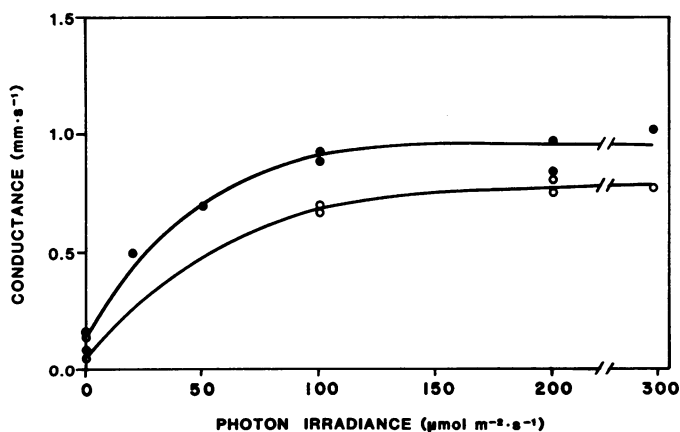


FIG. 4. Stomatal conductance in attached leaves of *Pa. acmodontum* irradiated either with daylight fluorescent (○) or daylight fluorescent supplemented with blue phosphor fluorescent light (●) as in Figure 1. Measurements made in a growth chamber equipped with a DC drive fluorescent lamp canopy using a Li-Cor Model 1600 steady state porometer. Each point is the average of two separate measurements made on different plants.

prevailing in *Paphiopedilum* (15). A conservative growth strategy that includes very slow growth rates and low levels of conductance, appears widespread in the Orchidaceae and is correlated with a wide structural divergency in their guard cell chloroplasts (5; D'Amelio and Zeiger, unpublished). *Paphiopedilum* appears as an extreme case in which the development of guard cell chloroplasts has been completely eliminated.

The enhancement of growth rates of *Paphiopedilum* by a blue light-enriched environment indicates that the stimulation of stomatal conductance by blue irradiation, and the increases in intercellular CO₂ concentrations observed in gas exchange experiments (Fig. 2), can result in long-term increases in carbon gain. Several lines of evidence point to a stomatal effect as the major, if not the only, consequence of the blue light-enrichment. Direct blue light effects on growth are usually inhibitory (3). Also, net photosynthesis is consistently higher in red light than in blue (18). A direct stimulation of growth or photosynthetic rate by blue light in the long-term growth experiments is, therefore, unlikely. Also unlikely is a morphogenetic effect of blue light enhancing chloroplast development, in view of the evidence indicating that a red light regime is more favorable for chloroplast development than blue (1). Further evidence for a stomatal-mediated stimulation of growth by blue light is provided by the higher levels of stomatal conductance measured in the plants grown in the blue light-enriched environment (Fig. 4) and the lack of significant differences in stomatal densities of leaves growing in the two treatments.

The interpretation of the observed higher growth rates as an enhanced carbon gain ensuing from the blue light-induced, higher stomatal conductances, implies that net photosynthesis rates were higher in the plants growing in the blue light-enriched environment. This conclusion is seemingly in conflict with the results obtained in the short-term gas exchange experiments, showing that photosynthesis rates measured under blue light were not higher than those seen under red light. However, pure blue and red light conditions cannot be directly compared with the light regimes used in the growth experiments, in which the plants were exposed to white light with different proportions of blue quanta. Pure blue light enhances stomatal conductance but also depresses net photosynthesis (Figs. 2 and 3), hence a net gain in photosynthetic rates would be observed only under conditions in which the photosynthetic stimulation ensuing from the higher intercellular CO₂ concentrations exceeds the effect of the lower photosynthetic quantum yield of blue light. Although unavailability of the required gas exchange equipment at Rockville did not allow us to measure directly net photosynthetic rates in the growth experiments, the demonstrated stimulation of stomatal conductances by blue light and the measured higher growth rates point to an enhanced net photosynthesis in the blue light-enriched environment.

The sustained stimulation of stomatal opening by blue light over long growth periods is unexpected when considered in the context of the integrated nature of stomatal responses. Because stomata respond to many environmental signals, overall strategies of stomatal regulation would be expected to be conserved despite relative variations in any single environmental input. The case could be more extreme with *Paphiopedilum* which, like

many Orchidaceae, appears to strongly conserve low conductance levels. The observation that a single environmental factor, such as blue light, can override that strategy indicates that stomatal manipulation could be utilized in management or breeding programs aimed at higher plant productivity. Boyer (2) has recently emphasized the importance that the manipulation of environmental responses could play in future breeding programs. The results with *Paphiopedilum* indicate that the establishment of conditions which specifically change stomatal responses for agronomic purposes, is conceptually feasible. In addition, higher growth rates of *Paphiopedilum* under blue light-enriched environments might be useful in commercial nurseries if the technique proves cost-effective.

Acknowledgments—We thank Professor H. Mooney for generous support with equipment and the Rod McLellan Co. for information on orchid taxonomy and supply of plant material. We also thank Mr. August Dietz, Foreman of the Smithsonian Greenhouse Nursery, for generous donation of the *Paphiopedilum acmodontum* plants.

LITERATURE CITED

- AKOYUNOGLU G, H ANNI 1984 Blue light effect on chloroplast development in higher plants. In H Senger, ed. *Blue Light Effects in Biological Systems*. Springer-Verlag, New York, pp 397-406
- BOYER JS 1982 Plant productivity and environment. *Science* 218: 443-448
- COSGROVE DJ 1981 Rapid suppression of growth by blue light. Occurrence, time course, and general characteristics. *Plant Physiol* 67: 584-590
- COWAN IR 1982 Regulation of water use in relation to carbon gain in higher plants. In OL Lange, PS Nobel, CB Osmond, H Ziegler, eds, *Physiological Plant Ecology*. Encyclopedia of Plant Physiology, New Series, Vol 12B. Springer-Verlag, Heidelberg, pp 589-613
- D'AMELIO E, E ZEIGER 1983 Structural and functional divergence of guard cell plastids in the Orchidaceae. *Plant Physiol* 72: S-101
- DRESSLER RL 1981 *The Orchids*. Harvard University Press, Cambridge
- FARQUHAR GD, TD SHARKEY 1982 Stomatal conductance and photosynthesis. *Annu Rev Plant Physiol* 33: 317-345
- GRIVET C, E ZEIGER 1983 Blue and red light responses of net photosynthesis (Ps) and stomatal conductance (g) in intact leaves of *Paphiopedilum*. *Plant Physiol* 72: S-101
- HOAGLAND DR, DI ARNON 1939 The water culture method for growing without soil. *Calif Agric Exp Stn Circ* 327: 1-39
- MCCREE KJ 1971 The action spectrum, absorption and quantum yield of photosynthesis in crop plants. *Agric Meteorol* 9: 191-216
- MOHR H 1980 Interaction between blue light and phytochrome in photomorphogenesis. In H Senger, ed, *The Blue Light Syndrome*. Springer-Verlag, New York, pp 97-109
- NELSON SD, JM MAYO 1975 The occurrence of functional non-chlorophyllous guard cells in *Paphiopedilum* spp. *Can J Bot* 53: 1-7
- RUTTER JC, CM WILLMER 1979 A light and electron microscopy study of the epidermis of *Paphiopedilum* spp., with emphasis on stomatal ultrastructure. *Plant Cell Environ* 2: 211-219
- THORPE N 1980 Accumulation of carbon compounds in the epidermis of five species with either different photosynthetic systems or stomatal structure. *Plant Cell Environ* 3: 451-460
- WILLIAMS WE, C GRIVET, E ZEIGER 1983 Gas exchange in *Paphiopedilum*. Lack of chloroplasts in guard cells correlates with low stomatal conductance. *Plant Physiol* 72: 906-908
- WINNER WE, HA MOONEY 1980 Ecology of SO₂ resistance. I. Effects of fumigations on gas exchange of deciduous and evergreen shrubs. *Oecologia* 44: 290-295
- ZEIGER E 1983 The biology of stomatal guard cells. *Annu Rev Plant Physiol* 34: 441-475
- ZEIGER E, C FIELD 1982 Photocontrol of the functional coupling between photosynthesis and stomatal conductance in the intact leaf. *Plant Physiol* 70: 370-375
- ZEIGER E, SM ASSMANN, H MEIDNER 1983 The photobiology of *Paphiopedilum* stomata: opening under blue but not red light. *Photochem Photobiol* 38: 627-630