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PHOTOSYNTHETIC RESPONSES OF PLANTS FROM HABITATS WITH CONTRASTING THERMAL ENVIRONMENTS

Comparison of Photosynthetic Characteristics of Intact Plants

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Last year we reported on the responses of growth and development of a number of species native to habitats with contrasting thermal regimes in our experimental stations on the hot desert floor of Death Valley and the cool oceanic bluff of Bodega Head (*Year Book 73*, p. 748). On the basis of their growth response under adequate water supply in the Death Valley garden, the 17 species tested could be separated into three main categories: (1) those which are unable to survive the hottest period of the year, (2) those which are able to survive this period but do not grow, and (3) those whose growth mainly occurs during the hottest part of the year. All of the species native to cool oceanic habitats and several of those native to warm-temperate environments, including both C_3 and C_4 species, fall in the first category. All species native to the floor of Death Valley, except *Tidestromia oblongifolia*, fall in category 2. This C_4 species which is summer active and winter deciduous in its native Death Valley habitat, is the only one that falls into category 3. This plant exhibited a very fast growth rate under the extreme summer heat in the Death Valley garden.

In contrast, all of the species tested, except *T. oblongifolia*, were able to grow in the cool Bodega Head garden. Here the species native to cool coastal habitats such as *Atriplex glabriuscula* (C_3) and *A. sabulosa* (C_4) had the highest productivities.

On the basis of these transplant experiments *Atriplex glabriuscula* (cool coastal C_3), *A. sabulosa* (cool coastal C_4), *A. hymenelytra* (Death Valley evergreen C_4), and *Tidestromia oblongifolia* (Death Valley summer active C_4) were selected for further detailed comparative studies of productivity under contrasting controlled thermal regimes and for investigation of the photosynthetic and other physiological mechanisms that underlie the striking differences in the ability of plants to grow in contrasting natural thermal environments. The comparative growth experiments in which temperature was the only variable and in which other stress factors, including atmospheric drought, were completely eliminated clearly show that the widely different abilities of these species to grow in the contrasting habitats of Bodega Head and the Death Valley floor are primarily due to intrinsic differences in their response to *temperature* (*Year Book 73*, p. 757). For example, under a 16°C day/10°C night regime, simulating a cool coastal environment, the daily growth rates of coastal *A. glabriuscula* and *A. sabulosa* were as high as 0.16 $g^{-1}day^{-1}$ or 65% to 75% of the maximum growth rate which occurred at about 25°C day/17°C night. *T. oblongifolia* proved to be incapable of sustained growth under the 16°/10°C regime. In contrast, under a very hot 45°C day/31°C night regime, simulating a typical summer day in Death Valley, *T. oblongifolia* is very near its optimum temperature for growth and under these conditions its growth rate was a record high of 0.26 g^{-1} day^{-1}, i.e., this extraordinary plant doubled its dry matter content in less than 3 days. This high temperature regime is lethal to both of the coastal species. *A. hymenelytra* grew under both of these extremes of temperature, but the growth rates were lower than those of the coastal species under the cool regime and much lower than that of *T. oblongifolia* under the hot regime.

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Our growth analyses further show that although temperature has a marked influence on the growth habits and other morphological characteristics, the strikingly different temperature dependences of dry matter production among the four species analyzed in detail cannot even in part be due to differences in the allocation of carbon to the leaves in relation to nonphotosynthetic organs. These results indicate that the contrasting interspecific responses of dry matter production to temperature must be the result of intrinsic differences in the temperature dependence of primary growth processes. During the year detailed studies of the photosynthetic and respiratory characteristics of the four species have been conducted both in the field and on plants grown under controlled temperature regimes in the laboratory. Since processing of the field data was not yet completed when this report was prepared (May 1975) the results presented here will concentrate on the characteristics of material grown under controlled conditions. It is evident, however, that these results are consistent with the field data.

The photosynthetic performances of the different species discussed in the following section, are compared under similar conditions of irradiance, CO_2 and O_2 concentrations, water vapor pressure, stomatal conductance, and plant water status. These results, therefore, reflect intrinsic characteristics of the photosynthetic apparatus. A detailed account of these studies will be published elsewhere.

The striking differences that can exist in the photosynthetic temperature dependence of higher plants native to habitats with contrasting thermal regimes are illustrated in Fig. 1. The curve for *T. oblongifolia* was obtained at the peak of summer heat in Death Valley, while that for coastal *A. glabriuscula* was measured on a plant grown under a thermal regime simulating a cool oceanic environment.

The coastal C_3 species, *A. glabriuscula*, and the coastal C_4 species, *A. sabulosa* (not shown), are capable of high photosynthetic rates at low and moderate temperatures but the rates do not increase with increased temperature beyond 25° to 30°C; they decline above 35°C and rapidly fall to zero at 45°C. The photosynthetic performance of *T. oblongifolia* is in sharp contrast with the coastal plants. When grown in a hot temperature regime either in Death Valley or under controlled conditions in the laboratory, the photosynthetic rate is poor at low and moderate temperatures but increases steeply with increased temperature to reach a very high maximum rate at about 45°C, and even at 50°C the rate exceeds that of *A. glabriuscula* at its optimum temperature. Total inhibition of photosynthesis in *T. oblongifolia* does not occur until 52° to 54°C.

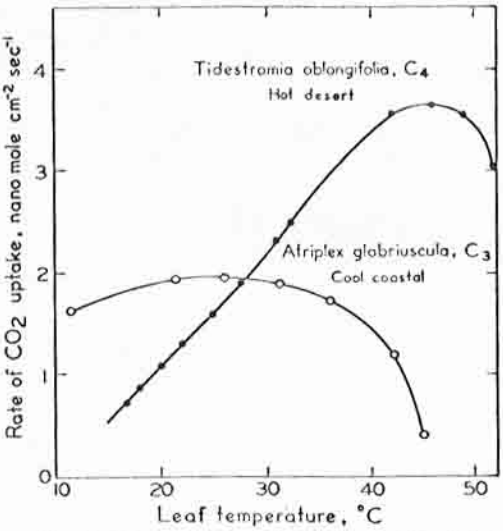

Fig. 1. Temperature dependence of photosynthesis in *Tidestromia oblongifolia* and *Atriplex glabriuscula* at a high light intensity of 160 nanoeinstein $cm^{-2}sec^{-1}$, a CO_2 partial pressure of approximately 320 μbar, and an O_2 concentration of 21%. Stomatal conductance values were almost identical in the two species.

Both curves shown in Fig. 1 were determined at a high light intensity of 160 nanoeinstein $cm^{-2}sec^{-1}$, which is approximately equal to 80% of maximum noon sunlight. This light intensity is rate-saturating for *A. glabriuscula* but

is insufficient to provide maximum photosynthetic rate at the temperature optimum for *T. oblongifolia*. The latter species is thus capable of even higher photosynthetic rates. Rates in excess of 5 nanomole CO_2 $cm^{-2}sec^{-1}$ (80 mg CO_2 $dm^{-2}hr^{-1}$) are not uncommon for *T. oblongifolia*.

The temperature regime under which the plants are grown markedly affects the temperature dependence of photosynthesis in all of the species examined. Comparisons of the different species grown at a temperature regime simulating the summer in Death Valley cannot be made, since this is lethal to the coastal plants. The comparisons were therefore carried out under 40°C day/30°C night and 16°C day/11°C night, the highest and lowest temperature regime at which all four species are able to survive. As shown in Table 1, growth at 40°C results in strikingly reduced photosynthetic rates both at high and low temperatures in the two coastal species. In *T. oblongifolia*, on the other hand, growth at the low temperature regime causes a drastic reduction in photosynthetic capacity. This low temperature inhibition of photosynthesis is present at low, intermediate, and high temperatures, irradiances, and CO_2 concentrations.

Atriplex hymenelytra (C_4) is capable of a wider range of photosynthetic acclimation to temperature than either the two coastal *Atriplex* species or *T. oblongifolia*. Growth under the cool temperature regime results in a somewhat higher photosynthetic capacity at low temperature than does growth under the hot regime and vice versa. However, the photosynthetic capacity of this species is lower than the capacity of the coastal plants when grown under a low temperature regime and greatly inferior to that of *T. oblongifolia* when grown under a hot regime. Although *A. hymenelytra* can tolerate higher temperatures than the coastal *Atriplexes* without permanent damage, it is less extreme in this respect than *T. oblongifolia* and it is incapable of sustained net photosynthesis above 47°C.

These and other results show that the responses of photosynthesis to different temperatures are quite similar to those of overall growth rates and thus provide further evidence that the temperature dependence of dry matter production is closely linked to the temperature dependence of photosynthetic capacity. Also, the genetically determined range of environmentally induced photosynthetic acclimation to temperature is strikingly different among the study species. This undoubtedly reflects profound adaptive differentiations of the photosynthetic machinery to the contrasting thermal regimes of the respective native environments and indicates that these adaptations are major determinants in the different abilities of the plants to grow and succeed in their widely diverse habitats. The results further suggest that excellence in photosyn-

TABLE 1. Effect of Temperature for Growth on Photosynthetic Rates*

	Photosynthesis at 16°C			Photosynthesis at 40°C		
Species	A: Grown at 16°C	B. Grown at 40°C	Ratio A/B	C. Grown at 40°C	D. Grown at 16°C	Ratio C/D
A. glabriuscula	2.20	0.73	3.01	1.01	1.80	0.56
A. sabulosa	2.80	0.70	4.00	1.54	2.80	0.55
A. hymenelytra	1.62	1.10	1.47	2.41	2.18	1.11
T. oblongifolia	0.42	1.70	0.25	5.00	0.42	11.90

* Rates measured at a high light intensity of 150 nanoeinstein $cm^{-2}sec^{-1}$, 320 microbar CO_2, and a water vapor pressure deficit $\lesssim$ 15 millibar. Rates are given in nanomole CO_2 $cm^{-2}sec^{-1}$.

thetic performance in one extreme may preclude excellence in the opposite extreme.

The superior photosynthetic performance of *T. oblongifolia* at high temperature is not caused by differences in stomatal conductance to CO_2 diffusion. Figure 2 shows the relationship between the photosynthetic and stomatal conductance at 40°C leaf temperature of plants grown at this temperature. The data are derived from experimental determinations of the rate of CO_2 uptake as a function of the CO_2 concentration in the intercellular spaces in the leaf. It is clear that *T. oblongifolia* is superior to *A. hymenelytra* at any stomatal conductance at which either plant is capable of a substantial rate of CO_2 uptake. Recent field measurements on these two species in Death Valley show that the responses are similar to those of the laboratory grown material but the differences between the two species are somewhat greater with even higher rates for *T. oblongifolia* and lower rates for *A. hymenelytra*. The rates of the coastal C_3 plant *A. glabriuscula* are lower than those of the other species at any stomatal conductance. Clearly, the interspecific differences in photosynthetic performance are unr lated to differences in stomatal characteristics.

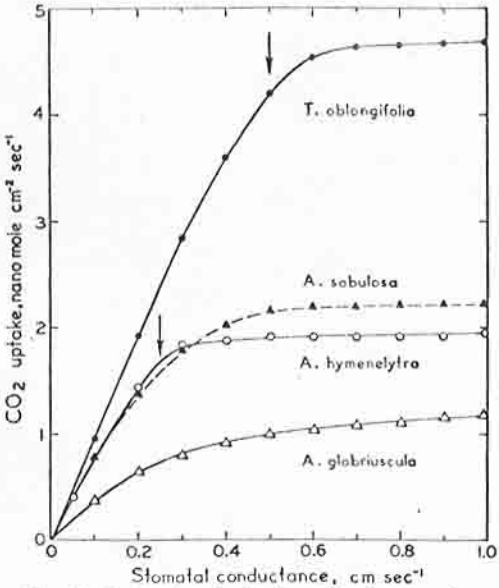

Fig. 2. Rate of net photosynthesis as a function of stomatal conductance to water-vapor diffusion in *T. oblongifolia*, *A. hymenelytra*, *A. sabulosa*, and *A. glabriuscula*. Light intensity was 160 nanoeinstein cm^{-2}sec^{-1}, the CO_2 partial pressure of the external air was 325 μbar, and leaf temperature was 40°C. The plants were grown at a 40°C day/30°C night regime. The relationship was determined from measurements of CO_2 uptake as a function of CO_2 concentration at known stomatal conductances. Arrows indicate the stomatal conductance values usually found in *T. oblongifolia* and *A. hymenelytra* under optimum conditions both in the field and in the laboratory.

From the results presented in Fig. 2 it also follows that for any given photosynthetic rate and any given wate. vapor pressure difference between the leaves and the air, the photosynthetic water use efficiency (CO_2 fixed/water transpired) is considerably greater in *Tidestromia* than in *A. hymenelytra* and much greater than in the coastal C_3 species. It is interesting to note that the stomatal conductance values usually found under optimal conditions in the two Death Valley species are so adjusted that they permit a near maximum photosynthetic water use efficiency together with a high photosynthetic rate. A further increase in stomatal conductance would lead to a proportional increase in water loss but a much smaller increase in photosynthesis. A decrease in conductance would, on the other hand, result in a decreased photosynthetic rate with little improvement in water use efficiency.

There is now little doubt that C_4 photosynthesis is an important mechanism underlying the capacity for a high photosynthetic water use efficiency. Photosynthetic gas exchange studies on a great number of C_3 and C_4 plants including the heat-adapted *T. oblongifolia* and the cool-adapted *A. sabulosa* show without exception that C_4 photosynthesis is markedly superior in utilizing low CO_2 concentrations in the leaf intercellular spaces. This is illustrated in Fig. 3 which

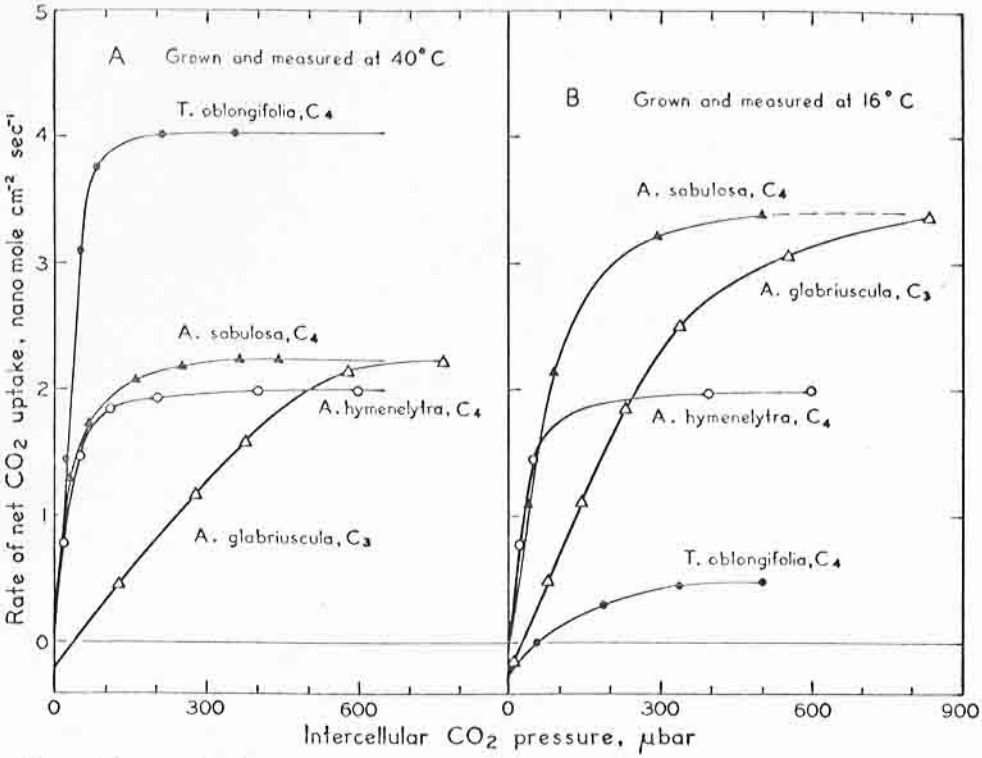

Fig. 3. Photosynthesis as a function of the CO_2 partial pressure in the intercellular spaces for different C_3 and C_4 species. Measurements were made at a high light intensity of 160 nanoeinstein $cm^{-2}sec^{-1}$, an O_2 concentration of 21%, and two different leaf temperatures, 40°C and 16°C.

shows the rate of photosynthesis as a function of intercellular space CO_2 concentration in the four species grown and measured at 40°C and at 16°C. At the high temperature the C_4 species are greatly superior to the C_3 plant. With the exception of *T. oblongifolia*, which does not tolerate extended periods at low temperatures, this difference between C_3 and C_4 plants is also present at 16°C although it is not as pronounced as at the higher temperature.

One would thus predict that the C_4 pathway would confer an adaptive advantage under any condition where photosynthesis in the absence of this pathway would be markedly limited by the intercellular space CO_2 concentration. Where this concentration exerts no effect or only a small effect, as is the case at low light intensities or low temperatures, the advantage would of course be at best marginal, but there is no reason to assume that C_4 photosynthesis *per se* would be disadvantageous in any terrestrial environment. All of our experimental results to date are in full agreement with these predictions. Conversely, one would also predict that the advantages of the C_4 pathway would be maximal under conditions where photosynthesis is limited largely by the intercellular CO_2 concentration. Such conditions are present when photosynthesis is operating at high light intensities and temperatures and in particular when stomatal conductance to the exchange of CO_2 and water vapor is low. Thus, for a given high light intensity and leaf temperature and a given rate of water loss, a C_4 plant should have a higher photosynthetic rate and, consequently, a

higher water use efficiency. However, this can only be realized if other essential adaptations are also present. For example, the high thermal stability of the photosynthetic apparatus of *T. oblongifolia* permits a full expression of the adaptive advantage of C_4 photosynthesis at very high temperatures. In *Atriplex sabulosa*, which lacks this thermal stability, the photosynthetic rate at high temperatures is limited by factors other than intercellular CO_2 concentration, and the potential advantage of C_4 photosynthesis cannot be fully realized.

Thermal Stability of the Photosynthetic Apparatus in Intact Leaves

Olle Björkman

In the preceding pages we reported on the dramatic differences that exist in the temperature dependence of photosynthesis among plants from habitats with contrasting thermal regimes. It seems evident that the truly remarkable performance at high temperatures and normal air of *Tidestromia oblongifolia* is dependent on the presence of the C_4 pathway of photosynthesis but that other independent adaptations, such as an unusually high thermal stability of the photosynthetic apparatus, must also be present. This year several studies have been initiated in an effort to uncover the biochemical and biophysical mechanisms that underlie the differences among our study species in their genetically determined abilities to photosynthesize and grow at extreme temperatures, as well as the changes induced by the temperature under which a given genotype is grown. Brief accounts of some of the early results are given here and in the following reports.

Measurements of CO_2 and water vapor exchange rates clearly show that in none of the four species analyzed is the decline in photosynthetic rate at high temperatures due to a decreased conductance of the stomata to CO_2 diffusion. On the contrary, conductances reached their maximum values at the highest measurement temperatures.

Temperature dependence curves of photosynthesis determined at rate-saturating CO_2 concentrations in the leaf intercellular spaces rather than in normal air show that high CO_2 does not prevent high-temperature inhibition in any of the four species. Although, as expected, high CO_2 greatly increases the absolute photosynthetic rate at high but noninhibiting temperatures in the C_3 species, *Atriplex glabriuscula*, and little or not at all in the three C_4 species, there is scarcely any effect of CO_2 concentration on the leaf temperature at which a rapid high-temperature–induced decline of photosynthesis sets in. For plants grown at 40°C day/30°C night regime these critical temperatures are: *A. glabriuscula*, 36°–37°C; *A. sabulosa*, 37°–38°C; *A. hymenelytra*, 41°–42°C; and *T. oblongifolia*, 46°–47°C. For plants grown under 16°C day/10°C night regime these temperatures are 2° to 3°C lower.

These results show that the high-temperature–induced decline in photosynthesis is unrelated to changes in the *diffusive* transport of CO_2 fixation sites inside the parenchyma cells. Further experiments show that a reduction of the oxygen concentration from the usual 21% to 2% at normal atmospheric CO_2 concentration did not significantly change the temperature at which thermal inhibition occurred in the C_3 or in the C_4 species. This indicates that factors associated with the O_2 inhibition of CO_2 fixation, catalyzed by RuDP carboxylase, and the O_2-stimulated photorespiratory CO_2 release via the glycolate pathway are not responsible for high-temperature inhibition of photosynthesis. (It should be noted, however, that in *A. glabriuscula* and other C_3 plants, these factors are almost certainly partly responsible for the relatively low net photosynthetic rates in normal air at