dition, understanding the mechanism by which LDV decreases ANA and antibody to DNA may offer possible leads for the suppression of ANA responses in man.

MICHAEL B. A. OLDSTONE
FRANK J. DIXON

Department of Experimental Pathology, Scripps Clinic and Research Foundation, La Jolla, California 92037

References and Notes

1. G. Tonietti, M. B. A. Oldstone, F. J. Dixon, *J. Exp. Med.* **132**, 89 (1970).
2. M. Bielschowsky, B. J. Heyler, J. B. Howie, *Proc. Univ. Otago Med. School* **37**, 9 (1959); M. C. Holmes and F. M. Burnet, *Ann. Int. Med.* **59**, 265 (1963); J. B. Howie and B. J. Heyler, *Advan. Immunol.* **9**, 215 (1968); B. H. Hahn and L. E. Shulman, *Arthritis Rheum.* **12**, 355 (1969); B. J. Heyler and J. B. Howie, *Nature* **197**, 197 (1963); F. M. Burnet and M. C. Holmes, *Aust. Ann. Med.* **14**, 185 (1965); R. C. Mellors, *J. Exp. Med.* **122**, 25 (1965).
3. P. H. Lambert and F. J. Dixon, *J. Exp. Med.* **127**, 507 (1968).
4. M. B. A. Oldstone and F. J. Dixon, *ibid.* **129**, 483 (1969).
5. P. H. Lambert and F. J. Dixon, *Clin. Exp. Immunol.* **6**, 829 (1970).
6. A. D. Steinberg, S. Baron, N. Talal, *Proc. Nat. Acad. Sci. U.S.* **63**, 1102 (1969).
7. A. Notkins, *Bacteriol. Rev.* **29**, 143 (1965).
8. Seed LDV was provided by Dr. Abner Notkins of National Institute of Dental Research, Bethesda, Maryland. Animals were assayed for persistent LDV infection by plasma infectivity testing.
9. M. B. A. Oldstone and F. J. Dixon, *Amer. J. Pathol.* **52**, 251 (1968).
10. R. Farr, *J. Infec. Dis.* **103**, 239 (1958); R. Wold, F. Young, E. Tan, R. Farr, *Science* **161**, 806 (1968); F. J. Dixon, G. Tonietti, M. Oldstone, T. Aoki, P. McConahey, in *Proc. Int. Immunopathol. Symp. 6th*, P. Miescher, Ed. (Schwabe, New York, 1971), p. 377.
11. A. Notkins and C. Scheele, *J. Nat. Cancer Inst.* **33**, 741 (1964); J. Bailey, J. Clough, M. Stearman, *Proc. Soc. Exp. Biol. Med.* **117**, 350 (1964); V. Riley, H. Campbell, C. Stock, *ibid.* **133**, 38 (1970).
12. A. Notkins, S. Mergenhagen, A. Rizzo, C. Scheele, T. Waldmann, *J. Exp. Med.* **123**, 347 (1966).
13. G. De-Thé and A. Notkins, *Virology* **26**, 512 (1965); R. Evans and M. Salaman, *J. Exp. Med.* **122**, 993 (1965); D. Porter and A. Notkins, personal communications.
14. R. Peterson, R. Hendrickson, R. Good, *Proc. Soc. Exp. Biol. Med.* **114**, 517 (1963); M. Salaman and N. Wedderburn, *Immunology* **10**, 445 (1966); R. Siegel and J. Morton, *ibid.*, p. 559; N. Cremer, D. Taylor, S. Hagens, *J. Immunol.* **96**, 495 (1966).
15. S. Mergenhagen, A. Notkins, S. Dougherty, *J. Immunol.* **99**, 576 (1967).
16. R. Howard, A. Notkins, S. Mergenhagen, *Nature* **221**, 873 (1969).
17. B. Greenwood and A. Voller, *Clin. Exp. Immunol.* **7**, 793 (1970).
18. We thank P. McConahey for assistance in the DNA antibody assay, and T. Tishon and M. Magner for technical help. This is publication No. 535 from the Department of Experimental Pathology, Scripps Clinic and Research Foundation, La Jolla, California. Supported by PHS grants AI-09484 and AI-07007, and the Violet June Kertell Memorial grant for research on multiple sclerosis. M.B.A.O. is recipient of PHS career development award KO4 AI-42580.

5 August 1971; revised 6 October 1971

Photosynthetic Adaptation to High Temperatures: A Field Study in Death Valley, California

Abstract. *The photosynthesis of* Tidestromia oblongifolia *(Amranthaceae) is remarkably well adapted to operate at the very high summer temperatures of the native habitat on the floor of Death Valley. The photosynthetic rate was very high and reached its daily maximum when the light intensity reached its noon maximum at the high leaf temperatures of 46° to 50°C which occurred at this time. At the intensity of noon sunlight the rate decreased markedly when the leaf temperature was experimentally reduced to below 44°C. The optimum rate occurred at 47°C. At this temperature the photosynthetic rate was essentially directly proportional to light intensity up to full sunlight.*

The intense heat and the very limited supply of water that prevail during the summer in many of the world's desert regions impose special demands on plants occupying such habitats. Most species escape the requirement of efficient photosynthesis and other metabolic activity under such severe conditions by carrying out most or all of their photosynthesis and growth during milder periods of the year. However, some higher plant species grow primarily during the hot summer. Obviously these species are uniquely suited for investigations of photosynthetic adaptation to high temperature. We report here the results of a study of the photosynthetic performance of one such species, *Tidestromia oblongifolia* Wats (Standl.) in its native habitat on the floor of Death Valley, California, during the first week of July 1970. The results show that the photosynthetic apparatus of this species is remarkably well adapted to function at the extremely high temperatures prevailing in its native habitat; even at leaf temperatures as high as 50°C the plant was capable of photosynthetic rates as high as those reported for any species under much more favorable conditiions and far greater than for other species growing in hot, arid environments.

Tidestromia oblongifolia is a low-growing herbaceous perennial which occupies dry sandy places, such as washes, below an altitude of 700 m throughout the Colorado and eastern Mojave deserts of the southwestern United States. It is very common on the floor of Death Valley, particularly on the foot of alluvial gravel fans. Earlier observations in this habitat indicated that this plant carries out most of its growth during May through August and dies back to at most a few basal leaves during the mild winter.

The floor of Death Valley is the hottest and driest part of the Western Hemisphere and one of the most severe habitats in the world. Long-time weather records show that it receives an average of 42 mm of rain per year, of which 75 percent falls during November through April (*1*). Air temperatures are consistently very high during the months of May through September, and a temperature of 50°C or higher is not uncommon during these months. The long-time average daily maximum temperature for the hottest month, July, measured at the U.S. Weather Bureau Station at Furnace Creek (54 m below sea level) is 47°C, and the average daily mean temperature is 39°C. Temperatures measured at Badwater, 85 m below sea level and 26 km south of Furnace Creek, are as high as or even higher than those at Furnace Creek. Ground surface temperatures may be more than 25°C higher than those measured at standard height (*1*).

Our experimental site was located on a gravel fan, 60 m below sea level and 2.5 km south of Furnace Creek, just off the road to Badwater. The meteorological data measured at Furnace Creek should therefore be approximately valid also for the experimental site. Our temperature measurements at this site during the period of the investigation closely agreed with those recorded by the U.S. Weather Bureau Station at Furnace Creek.

The vegetation of the gravel fan on which the site was located is characterized by three species: *T. oblongifolia, Atriplex hymenelytra,* and *Euphorbia* sp. These species occur predominantly in small washes crossing the gravel fan. Except in phreatophytic zones, plants are sparsely distributed on the gravel fans in this area at densities of about 125 per hectare (*2*). Anatomical examination of the leaves showed that all three species at this site had the anatomical characteristics of plants possessing the C_4 dicarboxylic acid pathway (*3*) of photosynthesis. In situ labeling experiments with ^{14}C-labeled carbon dioxide, in which the method described by Berry *et al.* (*4*) was used, showed that, after 10 seconds of exposure to labeled carbon dioxide, 57 to 92 percent of the total radioactivity incorporated was in the C_4 dicarboxylic acids malate and aspartate. Thus, there is no doubt that the species possess the C_4 pathway of photosynthesis.

Simultaneous measurements of carbon dioxide and water vapor exchange were made with an open system incorporating a ventilated controlled-environment plant chamber, an infrared carbon dioxide analyzer modified for differential measurements (Beckman model 315), and a dew-point hygrometer (Cambridge model 880). These and other measuring and controlling devices, with the exception of the plant chamber and the temperature and light sensors, were contained in an air-conditioned mobile laboratory unit. Details of this unit, the plant chamber, and the measuring and control system are described elsewhere (*5*).

In order to determine the daily course of the carbon dioxide and water vapor exchange of *T. oblongifolia*, the above-ground parts of an intact plant in situ were enclosed in the plant chamber and the chamber was sealed around the basal part of the stem. Great care was taken to disturb the soil as little as possible. The temperatures of leaves enclosed in the plant chamber as well as those of an adjacent plant outside the chamber were measured with small thermocouples attached to the lower leaf surfaces. The leaf temperatures of the outside plant were recorded, and the temperature of the plant chamber was automatically adjusted so that the temperatures of the leaves inside the chamber were the same as those of the leaves outside. The system reproduced the leaf temperatures measured outside to within 1°C, but it did not follow rapid fluctuations. Since the dew points of the ambient air were very low and transpiration rates were very high, no attempt was made to control the humidity of the air inside the chamber at ambient levels. However, the humidity of the chamber was kept sufficiently low so that the water vapor pressure gradient from the leaf to the air was always greater than 80 percent of that outside. The ventilation rate inside the leaf chamber was very high, and consequently the resistances of the leaf boundary layer to the diffusion of carbon dioxide and water vapor were probably small.

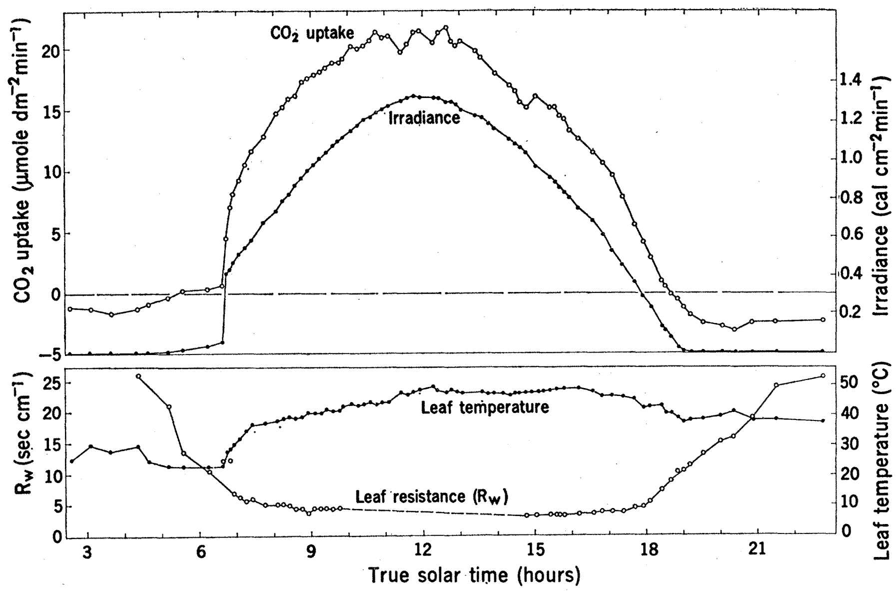

Fig. 1. Diurnal course of irradiance and of leaf temperature, rate of carbon dioxide uptake, and resistance to water vapor transfer of *T. oblongifolia* on 1 July 1970, in Death Valley, California. Leaf resistances were calculated from transpiration rates and leaf temperatures (*16*).

Figure 1 shows the daily course of photosynthesis on 1 July 1970, a bright clear day with irradiances and temperatures representative of that time of the year. The rate of uptake of carbon dioxide closely followed the irradiance throughout the day, with the maximum rate occurring in the period when irradiance was at a maximum. Leaf temperatures exceeded 40°C for nearly the entire light period and exceeded 45°C between 11 and 17 hours when rates of carbon dioxide uptake were highest. The maximum leaf temperature during the day was 49.5°C. Outside air temperatures were 2° to 3°C lower than the leaf temperatures. The maximum rate of photosynthesis during the day was 22 µmole per square decimeter of leaf area per minute, or 58 mg of carbon dioxide per square decimeter per hour. This is an unusually high rate of photosynthesis, particularly since this value represents the average value of all leaves on the plant; maximum rates of individual leaves can be expected to be appreciably higher. The sum of the resistances of the leaf boundary layer and the stomata to water vapor transfer between the leaf and the air (R_w) started to decrease at dawn, an indication that the stomata were opening, and reached its minimum value during the period of maximum photosynthesis. It was not possible to determine R_w during the middle of the day in this experiment because of equipment failure; however, minimum resistances measured on subsequent days under similar conditions were as low as 2 sec cm^{-1}. At an ambient dew point of 3°C and a leaf temperature of 48°C this resistance value corresponds to a transpiration rate of 0.21 g of water per square decimeter per minute. It seems unlikely that such high transpiration rates could be sustained for an extended period without loss of turgor if the plant were under severe water stress.

In order to obtain an estimate of the water status of the *Tidestromia* plants, we determined the xylem sap pressures of twigs of several individuals by using the Scholander pressure bomb

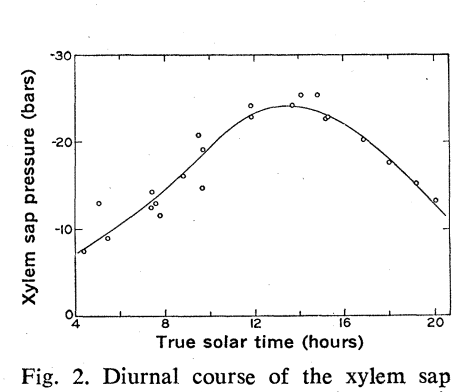

Fig. 2. Diurnal course of the xylem sap pressure of *T. oblongifolia*. The determinations were made from 1 to 5 July 1970. Each point represents the mean of three to five measurements.

technique (6). As shown in Fig. 2, sap pressures ranged from a minimum negative pressure of −10 bars in the morning to a maximum of −25 bars in the midafternoon when transpiration rates reached their maximum values. These sap pressures are similar to those reported previously for several other desert species; they are far less negative than the highest negative pressures reported for *Atriplex* sp. (≈ −55 bars), *Juniperus californica* (≈ −55 bars), and *Larrea divaricata* (≈ −80 bars) (6). Determinations of soil moisture also revealed that, although the top 20 cm of soil was almost completely dry, the water content at depths of 25 to 40 cm was 8 to 10 percent (weight/weight) in the < 2-mm fraction. Excavation of a plant showed that finely divided roots occurred in the zone from 25 to 40 cm. Evidently, the root system was capable of taking up water at rates sufficiently high to sustain a rapid transpiration at moderate sap pressures. These results clearly indicate that the T. *oblongifolia* plants were not under a severe water stress.

The daily course of photosynthesis in *T. oblongifolia* differs strikingly from previously published results for plants from hot and arid habitats. No evidence was found of the pronounced midday depression present in several species of the Mojave Desert in California (7), the Negev Desert in Israel (8), or in California chaparral (9), in spite of the considerably higher leaf temperatures measured in the study presented here. Moreover, the maximum photosynthetic rates obtained with *T. oblongifolia* are among the highest recorded in any natural habitat and are comparable to those of the highly productive crop species corn and sugar cane, when these plants are grown under highly favorable conditions. The rates are far greater than rates previously measured in the field for species native to arid or semiarid habitats (7–9).

Further experiments were done to distinguish between the influences of light intensity and temperature upon the photosynthetic rate of *T. oblongifolia*. Measurements of temperature dependence were made when irradiance was high and essentially constant. In these experiments the temperature-tracking system was disconnected and the temperature was decreased in steps. Measurements of light dependence were made by covering the plant chamber with large neutral optical-density screens. The leaf temperature was held constant at the different light intensities by adjusting the temperature of the plant chember.

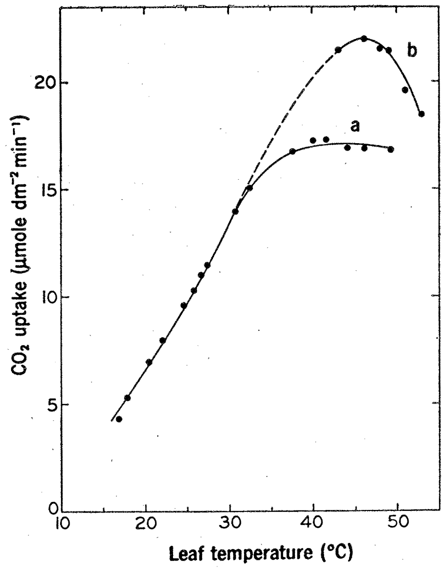

Fig. 3. Dependence of photosynthetic carbon dioxide uptake on leaf temperature in *T. oblongifolia* at two levels of irradiance: (a) 1.07 cal cm^{-2} min^{-1} and (b) 1.33 cal cm^{-2} min.$^{-1}$

The curve for photosynthesis as a function of temperature (Fig. 3) shows that at the highest level of irradiance the maximum photosynthetic rate is reached at about 47°C, and the rate is sharply reduced when the temperature is lowered to below 44°C. At 20°C, a temperature sufficiently high for maximum photosynthetic rates in many species from temperate environments, the rate in *T. oblongifolia* is only about one-third of its maximum value. As can be expected, the temperature required for the maximum photosynthetic rate decreases when the light intensity is reduced to a lower level. We have found no reports of an optimum temperature for photosynthesis of a terrestrial plant exceeding that of *T. oblongifolia*. Optimum temperatures exceeding 47°C have been reported only for thermophilic blue-green algae occurring in hot springs (*10*).

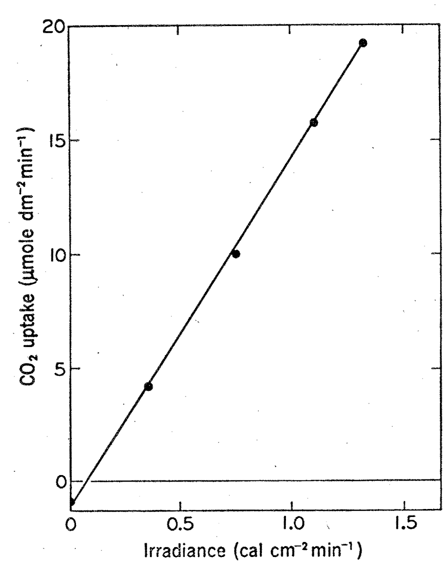

Fig. 4. Dependence of photosynthetic carbon dioxide uptake on light intensity in *T. oblongifolia* at a leaf temperature of 47.5°C. Irradiance values are for solar radiation in the wavelength range from 0.3 to 3.0 μm, incident on a horizontal plane inside the plant chamber.

As shown in Fig. 4, the photosynthetic rate was a linear function of light intensity up to the highest intensity received by the plant enclosed in the plant chamber. This was equal to 85 percent of the light energy flux incident upon the plant chamber at noon. An essentially linear relationship between photosynthesis and irradiance up to the highest level of irradiance received by the plant inside the leaf chamber (1.3 cal cm^{-2} min^{-1}) was also obtained on another day when light intensity was variable because of intermittent cloudiness. The decrease in the photosynthetic rate that resulted when light intensity decreased was not attributable to an increased stomatal resistance to carbon dioxide diffusion. Lack of light saturation of photosynthesis at irradiance values as high as 1.4 cal cm^{-2} min^{-1} has been reported previously for several species which, like *T. oblongifolia*, possess the C_4 dicarboxylic acid pathway of photosynthesis (*11*), but the light dependence obtained with *T. oblongifolia* is extreme (*12*). It is likely that the lack of even a partial light saturation may be attributed to the relatively high reflectance of the tomentose leaf surfaces and also, to some extent, to mutual shading among the leaves. The high temperature used in these experiments may also have resulted in a decreased steepness of the slope at low light intensities; although close to optimum at high light intensities (Fig. 3), this temperature may well be above optimum at low light intensities. There is, nevertheless, little doubt that, in spite of the high irradiances prevailing in Death Valley, light was the primary external factor that limited photosynthesis in *T. oblongifolia*.

It seems likely that the extraordinary photosynthetic performance of *T. oblongifolia* at high temperatures is largely attributable to the presence of the C_4 pathway of photosynthesis coupled with an unusually high thermal stability of its photosynthetic apparatus. There is strong evidence that the C_4 pathway serves in effect as a mechanism for concentrating carbon dioxide for the carboxylation step in the Calvin-Benson cycle (*13, 14*), resulting in high rates of carbon dioxide uptake under conditions of high irradiance, high temperature, and limited water supply (*13*). The inhibitory effect of atmospheric oxygen concentrations on net photosynthesis, present in species lacking the C_4 pathway, is absent in plants possessing this pathway. Since this inhibitory effect increases with increasing temperature, one might predict that its presence would be particularly detrimental to net photosynthesis in the Death Valley habitat of *T. oblongifolia*. Moreover, since C_4 photosynthesis evidently enables carbon dioxide of low concentrations in the leaf intercellular spaces to be utilized with a higher efficiency (lower "mesophyll resistance"), the efficiency of water use, that is, the amount of carbon dioxide fixed divided by the amount of water lost through transpiration, is considerably improved (*13, 15*). Whether higher plant species exist which do not possess the C_4 pathway and yet are capable of a photosynthetic performance at high temperatures equaling that of *T. oblongifolia* remains an interesting and important question.

OLLE BJÖRKMAN*, ROBERT W. PEARCY
Department of Plant Biology, Carnegie Institution of Washington, Stanford, California 94305
A. TYRONE HARRISON
HAROLD MOONEY
Department of Biological Sciences, Stanford University, Stanford 94305

References and Notes

1. C. B. Hunt, T. W. Robinson, W. A. Bowles, A. L. Washburn, *U.S. Geol. Surv. Prof. Pap. 494-B* (1966), pp. B 5–B 11; *U.S. Weather Bureau 1964 Climatic Summary of the United States, Supplement for 1951–1960, California* (Government Printing Office, Washington, D.C., 1964).
2. C. B. Hunt, *U.S. Geol. Surv. Prof. Pap. 509* (1966).
3. M. D. Hatch and C. R. Slack, *Annu. Rev. Plant Physiol.* **21**, 141 (1970).
4. J. A. Berry, W. J. S. Downton, E. B. Tregunna, *Can. J. Bot.* **48**, 777 (1970).
5. H. A. Mooney, E. L. Dunn, A. T. Harrison, P. A. Morrow, B. Bartholomew, R. L. Hays, *Photosynthetica*, in press. Certain modification of the laboratory unit proved necessary for the present studies. The unit was covered with aluminized Mylar ("space blanket"), an extra air-conditioner was installed, and both the air-conditioners and the electric generator were cooled with evaporative coolers; an ice bath was used in place of an electric refrigeration unit as the source of coolant for the temperature control system of the plant chamber.
6. P. F. Scholander, H. T. Hammel, E. D. Bradstreet, E. A. Hemmingsen, *Science* **148**, 339 (1965).
7. B. R. Strain, *Ecology* **50**, 511 (1969); *Photosynthetica* **4**, 118 (1970).
8. O. L. Lange, W. Koch, E. D. Schulze, *Ber. Deut. Bot. Ges.* **82**, 39 (1969).
9. H. A. Mooney and E. L. Dunn, *Evolution* **24**, 292 (1970).
10. T. D. Brock, *Nature* **214**, 882 (1967).
11. J. Hesketh and D. Baker, *Crop Sci.* **7**, 285 (1967); *Planta* **78**, 79 (1968); O. Björkman, E. Gauhl, M. A. Nobs, *Carnegie Inst. Wash. Year B.* **68**, 620 (1970); O. Björkman, R. W. Pearcy, M. A. Nobs, *ibid.* **69**, 640 (1971).
12. J. Hesketh and M. El-Sharkaway (personal communication) have observed a light dependence of photosynthesis for *Amaranthus edulis* in Tucson, Arizona, approaching that reported here for *T. oblongifolia*.
13. O. Björkman, in *Photosynthesis and Photorespiration*, M. D. Hatch, C. B. Osmond, R. O. Slayter, Eds. (Interscience, New York, 1971), p. 18.
14. M. D. Hatch, *ibid.*, p. 139.
15. R. W. Downes, *Planta* **88**, 261 (1969); R. O. Slayter, *ibid.* **93**, 175 (1970).
16. R. O. Slatyer and J. F. Bierhuizen, *Aust. J. Biol. Sci.* **17**, 115 (1964).
17. Operation of the mobile laboratory was supported by NSF research grants GB 21441 and GB 14168 to H.M. We acknowledge the assistance of Dr. E. Medina, Instituto Venezolano de Investigaciones Cientificas, Caracas, Venezuela, and the cooperation of P. G. Sanchez, chief naturalist, National Park Service, Death Valley National Monument. Carnegie Institution of Washington Department of Plant Biology publication No. 481.
* Present address: Research School of Biological Sciences, Australian National University, Canberra City, A.C.T.

24 May 1971

Rapid Increase of Phenylethanolamine N-Methyltransferase by Environmental Stress in an Inbred Mouse Strain

Abstract. *The activity of phenylethanolamine* N-*methyltransferase in mice of the C57Bl/Ka strain was determined after a 4°C stress. The enzyme activity increased 1.2-fold at the end of 3 hours and by 1.4-fold by the end of 6 hours of the stress. The results are in contrast to those from other species with intact animals in which the enzyme changes only after several days of chronic stress. Cycloheximide prevents the rise in enzyme activity, suggesting the increase may be due to protein synthesis. The increase may provide a model system for studying regulation of catecholamine biosynthetic enzymes.*

Phenylethanolamine *N*-methyltransferase (PNMT) is the adrenal medullary enzyme involved in epinephrine formation (*1*). In the rat, this enzyme has been shown to respond to exogenous glucocorticoid administration along an extremely slow time course (*2, 3*). The half-life of the enzyme has been estimated to be 6 days in the hypophysectomized rat (*3*) and 20 to 50 days in the intact rat (*4*). Because of the extremely slow turnover of the enzyme in the rat, studies which have attempted to probe the relation between this enzyme and acute environmental stress situations have not been possible.

Studies from this laboratory have suggested that PNMT in an inbred mouse strain may be under both neuronal and glucocorticoid control (*5*). In one inbred mouse strain, a maximal PNMT response to exogenous glucocorticoid administration was seen 3 hours after injection of the drug (*5*). Because of the rapid response of the enzyme in these animals, it would appear that inbred mouse strains could serve as excellent models in which to study the relation between acute stress and PNMT activity.

We now report our studies in an inbred mouse strain of the C57Bl/Ka line. The mice (20 to 25 g, male) were obtained from the department of radiobiology, Stanford Medical Center. Animals were housed in a soundproof animal room maintained at constant temperature and humidity and were allowed to acclimatize to these facilities and the light-dark cycle (LD, 13 : 11; lights on 7:00 a.m.) for 5 to 7 days before the study. Cold-stress exposure was carried out as follows: Animals were housed in groups of three in a litter-free stainless steel cage in a cold room maintained at 4°C. Animals were allowed free access to water and Purina Lab Chow during the period of cold exposure. Immediately after the cold exposure period was ended, animals were killed by cervical dislocation and their adrenals were removed. The glands were cleaned and homogenized as a pair in ice-cold 0.32*M* sucrose. Homogenates were centrifuged at 30,000*g* for 20 minutes. The PNMT activity was estimated by the modification of the method of Deguchi and Barchas (*6*) of the phenylethanolamine procedure originally described by Axelrod (*1*). Protein determinations were made according to the method of Lowry (*7*).

Table 1. Effects of cold exposure on adrenal PNMT activity in C57Bl/Ka mice. Each group contained nine animals; the values shown are the means and standard errors of those groups. One unit of PNMT activity is the formation of 1 nmole of *N*-methylphenylethanolamine per hour.

Treatment	PNMT activity (unit/mg protein)
Unstressed control	0.300 ± 0.005
3-hour cold stress	0.370 ± 0.024*
6-hour cold stress	0.417 ± 0.018†

* $P < .01$ greater than unstressed control.
† $P < .001$ greater than unstressed control.