

renaturation. Thermal transition curves<sup>12</sup> of such DNA show a hyperchromicity of 19.5 per cent, whereas none is seen with DNA denatured in the presence of formaldehyde. Thermally denatured chick embryo DNA, on re-heating, resembles formaldehyde-denatured calf thymus DNA and shows only a slight hyperchromic effect. These observations, together with the fact that chick embryo DNA also has a relatively low guanine-cytosine content (41 per cent)<sup>10</sup>, support the suggestion made above, that the composition of DNA influences its reaction with antibody. Therefore, DNA's of varying base composition are being prepared in order to examine their precipitability with the antisera. If a correlation between the base specificity of an antibody and its reaction with DNA molecules of varying G-C content were found, such studies could provide a basis for using immunochemical methods in more subtle investigations of the structure of DNA.

This work was supported by grants from the U.S. Public Health Service and contracts between the Office of Naval Research and Columbia University.

S. M. BEISER  
S. W. TANENBAUM  
B. F. ERLANGER

Department of Microbiology,  
College of Physicians and Surgeons,  
Columbia University, New York.

- <sup>1</sup> Butler, V. P., jun., Beiser, S. M., Erlanger, B. F., Tanenbaum, S. W., Cohen, S., and Bendich, A., *Proc. U.S. Nat. Acad. Sci.*, **48**, 1597 (1962).  
<sup>2</sup> Tanenbaum, S. W., and Beiser, S. M., *Proc. U.S. Nat. Acad. Sci.*, **49**, 662 (1963).  
<sup>3</sup> Erlanger, B. F., and Beiser, S. M., *Proc. U.S. Nat. Acad. Sci.*, **52**, 68 (1964).  
<sup>4</sup> Marmur, J., *J. Mol. Biol.*, **3**, 208 (1961).  
<sup>5</sup> Kabat, E. A., and Mayer, M. M., *Experimental Immunochimistry*, second ed. (Thomas, C. C., Springfield, Ill., 1961).  
<sup>6</sup> Heidelberger, M., and MacPherson, C. F. C., *Science*, **97**, 405 (1943); **98**, 63 (1943).  
<sup>7</sup> Delcher, H. R. G., Holman, H. R., and Kunkel, H. G., *J. Exp. Med.*, **109**, 97 (1959).  
<sup>8</sup> Murukami, W. T., Van Vunakis, H., Grossman, L., and Levine, L., *Virology*, **14**, 190 (1961).  
<sup>9</sup> Stollar, D., and Grossman, L., *J. Mol. Biol.*, **4**, 31 (1962).  
<sup>10</sup> We thank Dr. H. S. Rosenkranz for determining the G-C contents of the calf thymus and chick embryo DNA's from the mid-point of the thermal transition curves and from the density in caesium chloride. Values for calf thymus DNA may also be found in Chargaff, E., in *The Nucleic Acids*, 1, edit. by Chargaff, E., and Davidson, J. N., 354 (Academic Press, New York, 1955).  
<sup>11</sup> Haselkorn, R., and Doty, P., *J. Biol. Chem.*, **236**, 2738 (1961).  
<sup>12</sup> Marmur, J., and Doty, P., *J. Mol. Biol.*, **5**, 109 (1962).

### A C<sub>15</sub>H<sub>22</sub>O<sub>4</sub> Compound produced by the Fungus *Aspergillus fischeri* var. *glaber*

In late August 1962, Dr. C. W. Hesseltine of our laboratory collected a dry soil sample from under a Douglas fir in House Rock Camp Grounds near Corvallis, Oregon. From this sample was isolated a strain of *Aspergillus fischeri* var. *glaber* Fennell and Raper, now maintained in the A.R.S. Culture Collection as NRRL 3088.

During our culture work with this strain, we observed abundant crystal formation after about 6 days when the fungus was grown on malt extract agar. In still culture, as much as 4.87 g of crystalline product per litre of culture liquor was produced in 45 days at 28° C. The medium had the following composition: malt extract, 20 g; peptone, 1 g; *d*-glucose, 20 g; and distilled water, 1,000 ml.

The new fermentation product was isolated by filtration of the culture liquor and by acetone extraction of the dried mixture of mycelium and crystals. The compound separated from the acetone-hexane as colourless needles which melted at 49°-50° C. On cooling, molten samples crystallized and then melted at 54°-55° C. The compound has  $[\alpha]_D^{25} - 40^\circ$ . Elementary analyses and a Signer molecular weight determination were in agreement with a molecular formula of C<sub>15</sub>H<sub>22</sub>O<sub>4</sub>. Preliminary chemical and spectroscopic investigations indicate that the substance is a dilactone.

When compared with the type strain of *A. fischeri* var. *glaber* (described by Fennell and Raper<sup>1</sup> and deposited

in our collection as NRRL 2163), strain NRRL 3088 differed only in being somewhat darker in colony colour, in lacking a faint pinkish reverse in the medium, and in having slightly larger cleistothecia. One sub-culture of the type strain also produced a few scattered crystals in malt extract agar after repeated transfer. Several other isolates of the variety produced no crystals.

*Note added in proof.* Since submission of this manuscript it has come to our attention that Brookes, Tidd and Turner<sup>2</sup> recently reported that *Aspergillus avenaceus* produces a C<sub>15</sub>H<sub>22</sub>O<sub>4</sub> product apparently identical with our lactone, judging from its double melting point, rotation and nuclear magnetic resonance spectrum.

J. J. ELLIS  
FRANK H. STODOLA  
RONALD F. VESONDER  
CURTIS A. GLASS

Northern Utilization Research and  
Development Division,  
Agricultural Research Service,  
U.S. Department of Agriculture,  
Peoria, Illinois.

- <sup>1</sup> Fennell, D. I., and Raper, K. B., *Mycologia*, **47**, 68 (1955).  
<sup>2</sup> Brookes, Tidd and Turner, *J. Chem. Soc.*, 5385 (1963).

### Pyridoxine Deficiency in the Rat produced by *D*-Penicillamine

*D*-PENICILLAMINE is used in the treatment of Wilson's disease<sup>1</sup> and in cases of lead poisoning<sup>2</sup>. Recently, Crawhall *et al.*<sup>3</sup> reported that the administration of *D*-penicillamine to two cystinuric patients resulted in a marked fall in the excretion of cystine in urine. This was believed to be due to disulphide exchange resulting in the formation of the mixed disulphide of *D*-penicillamine and cysteine. It has therefore been suggested that *D*-penicillamine might prove useful in the management of this condition.

In a series of investigations, Du Vigneaud and his collaborators<sup>4-6</sup> showed that the inclusion of *L*-penicillamine in the diet of white rats caused a depletion of vitamin B<sub>6</sub>, resulting in inhibition of growth, reduced transaminase activity and abnormal excretion of xanthurenic acid following the administration of a test dose of tryptophan. They also showed that these effects could be prevented by inclusion of large amounts of pyridoxine in the diet. It was suggested that vitamin B<sub>6</sub> deficiency was the result of a chemical combination between *L*-penicillamine and pyridoxal phosphate to form a thiazolidine structure.

Similar results on transaminases were observed by Hedde *et al.*<sup>7</sup>, who, however, investigated the effect of *DL*-penicillamine on white rats.

Ueda *et al.*<sup>8</sup> demonstrated that, like the *L*-isomer, *D*-penicillamine reacts non-enzymatically with pyridoxal phosphate to form a thiazolidine compound. It was therefore considered of interest to examine whether B<sub>6</sub> deficiency resulted in the rat following the administration of *D*-penicillamine.

Six male albino rats (300-340 g wt.) were maintained on Oxoid rat diet (Oxoid, Ltd.). Before starting the administration of *D*-penicillamine the rats were placed in metabolic cages and 24 h urine was collected before and after subcutaneous injections of a test dose (40 mg/rat) of *L*-tryptophan<sup>9</sup>. A solution of 50 mg *D*-penicillamine hydrochloride (Dista Products, Ltd.) was given, by stomach tube, twice daily to each of three rats (Group A). The other three rats received the same dose by intramuscular injection (Group B). Since solutions of penicillamine were strongly acidic, adequate amounts of sodium bicarbonate were always added to bring the pH to neutrality. After a period of four weeks 24 h urine was collected again before and after injections of 40 mg *L*-tryptophan.