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 COMMUNICATIONS TO THE EDITOR
 

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A NEW CYCLIZATION REACTION<sup>1</sup>

Sir:

We wish to report that  $\alpha$ -(2-biphenyl)-acetonitrile as well as certain  $\alpha$ -alkyl- and  $\alpha$ -aroyl derivatives in the presence of concentrated sulfuric acid will undergo isomerization to yield phenanthrylamines. After three hours at 0°, 2-biphenyl-acetonitrile afforded an 85% yield of 9-phenanthrylamine, m.p. 134–136° (lit.<sup>2</sup> 137.5–138°).

From  $\alpha$ -(2-biphenyl)-butyronitrile<sup>3</sup> and the homologous valeronitrile,<sup>4</sup> 10-ethyl-9-phenanthrylamine (m.p. 117.5–119.5°; benzamide, m.p. 241–243°) and 10-propyl-9-phenanthrylamine (m.p. 101–102.5°; benzamide, m.p. 220–221.5°) were obtained in 80–85% yield.

This new cyclization has been applied to  $\alpha$ -(*o*-methoxybenzoyl)- $\alpha$ -(2-biphenyl)-acetonitrile (obtained in a manner analogous to that used in preparing the isomer<sup>5</sup>) which in the presence of sulfuric acid yielded an amine (m.p. 175–176°) having the composition expected for 10-(*o*-methoxybenzoyl)-9-phenanthrylamine. This, as well as the other phenanthrylamines above, gave the characteristic diazonium coupling test with sodium  $\beta$ -naphtholate. On the basis of this same test, the compound obtained by the sulfuric acid cyclization of  $\alpha$ -(*p*-methoxybenzoyl)- $\alpha$ -(2-biphenyl)-acetonitrile, and originally described by Bradsher and Kittila<sup>6</sup> as 9-(*p*-methoxyphenyl)-10-phenanthramide is actually 10-(*p*-methoxybenzoyl)-9-phenanthrylamine.

(1) This investigation was supported by a research grant (C-1743) from the National Cancer Institute of the National Institutes of Health, Public Health Service.

(2) L. F. Fieser, R. P. Jacobsen and C. C. Price, *THIS JOURNAL*, **58**, 2163 (1936).

(3) C. K. Bradsher and W. J. Jackson, Jr., *ibid.*, **73**, 3235 (1951).

(4) C. K. Bradsher and W. J. Jackson, Jr., unpublished work.

(5) C. K. Bradsher and R. S. Kittila, *THIS JOURNAL*, **72**, 277 (1950).

CHEMISTRY DEPARTMENT  
DUKE UNIVERSITY  
DURHAM, N. C.

CHARLES K. BRADSHER  
DOROTHY J. BEAVERS  
EDWIN D. LITTLE

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2-METHYLADENINE, AN HYDROLYSIS PRODUCT OF PSEUDOVITAMIN B<sub>12d</sub>

Sir:

Holdsworth, *et al.*,<sup>1</sup> using paper ionophoresis with bioautographic techniques, demonstrated two microbiologically active components in crystalline pseudovitamin B<sub>12</sub>.<sup>2</sup> This material had satisfied the accepted criteria of chemical homogeneity, including paper chromatographic study in a variety of solvent systems. Quantitative electrophoretic analysis of the same preparation examined by Holdsworth, *et al.*,<sup>1</sup> yielded 90% pseudovitamin B<sub>12</sub>, 9% of a faster moving, microbiologically active, red pigment, and 1% of a very rapidly moving,

(1) E. S. Holdsworth, J. E. Ford, S. K. Kon and J. W. G. Porter, *Nature*, **171**, 148 (1953).

(2) J. J. Pfüfner, D. G. Calkins, *et al.*, Abst. of 120th Meeting of Am. Chem. Soc., New York, 1951, 23 C.

microbiologically inactive, red pigment. The separation was conducted on cellulose columns in 0.1 *M* acetic acid containing a trace of NaCN with a potential of 200 v. and 0.002 amp. The microbiologically active contaminant readily crystallized from aqueous acetone and is referred to as pseudovitamin B<sub>12d</sub>.<sup>3</sup> *Anal.*<sup>4</sup>: C, 52.68; H, 6.64; N, 16.90; Co, 4.36; P, 2.29. The composition is approximately that of pseudovitamin B<sub>12</sub>. *Anal.* (before electrophoresis): C, 52.06; H, 6.60; N, 16.37; Co, 4.40; P, 2.31. (After electrophoresis): C, 52.35; H, 6.68; N, 17.15; Co, 4.41; P, 2.32.

Pseudovitamins B<sub>12</sub> and B<sub>12d</sub> have identical absorption spectra<sup>5</sup> in water with maxima at 278, 308, 320, 361, 518 and 548–50 m $\mu$  with  $E_{1\%}^{1\text{cm}}$  of 130, 62, 60, 204, 54.5, 57.5, respectively.

Pseudovitamin B<sub>12</sub> is 0.7 and pseudovitamin B<sub>12d</sub> 0.3 as active as vitamin B<sub>12</sub> in growth assay with *L. leichmannii*.<sup>6</sup>

Pseudovitamin B<sub>12</sub> differs from vitamin B<sub>12</sub> in containing adenine<sup>7</sup> in nucleotide linkage in place of 5,6-dimethylbenzimidazole. The corresponding base in pseudovitamin B<sub>12d</sub> is 2-methyladenine.

Pseudovitamin B<sub>12d</sub> (4.47 mg.) in 4 ml. of 1 *M* HCl was heated in a sealed tube for 4 hours at 100°. The hydrolysate was percolated through a column (0.9 cm.  $\times$  5.5 cm.) of Amberlite 1R-120. The resin was washed with water (25 ml.), and then eluted with 2 *M* NH<sub>4</sub>OH (30 ml.). The eluate absorbed ultraviolet light (max. 270 m $\mu$ ). It was concentrated and examined for purines by paper chromatography, using successively the three systems: *n*-butanol-acetic acid-water ( $R_f$  0.35), isoamyl alcohol-5% disodium phosphate ( $R_f$  0.38), and 65% aqueous isopropyl alcohol-2 *M* HCl ( $R_f$  0.64).<sup>8</sup> The ultraviolet absorption properties of the resin eluate as well as the eluates from the three paper chromatograms were identical with those of 2-methyladenine<sup>9</sup> (maxima at 265 m $\mu$  in 0.1 *M* HCl and 270 m $\mu$  at pH 11). Comparative and mixed paper chromatograms with 2-methyladenine yielded identical  $R_f$  values in the three solvent systems.

The eluate from the 65% aqueous isopropyl alcohol-2 *M* HCl chromatogram, containing by spectrographic analysis 275 $\gamma$  of the purine base was concentrated and a crystalline picrate prepared

(3) This pigment was found by Holdsworth, *et al.*,<sup>1</sup> to have the same mobility and microbiological growth activity as the main ionophoretic component of their Factor A from calf faeces.

(4) By Mr. William Saschek, University of Chicago.

(5) By Dr. J. M. Vandenberg and associates.

(6) By Dr. O. D. Bird and associates.

(7) H. W. Dion, D. G. Calkins and J. J. Pfüfner, *THIS JOURNAL*, **74**, 1108 (1952); *Federation Proc.*, **11**, 269 (1952). The purine base has since been isolated as its crystalline picrate and identity with adenine picrate established by direct comparison of the X-ray powder diagrams.

(8) G. R. Wyatt, *Biochem. J.*, **48**, 584 (1951).

(9) J. Baddiley, B. Lythgoe, D. McNeil and A. R. Todd, *J. Chem. Soc.*, **382** (1943); J. Baddiley, B. Lythgoe and A. R. Todd, 318 (1944). A sample of 2-methyladenine was supplied by Dr. E. F. Godefroi and Dr. E. L. Wittle.