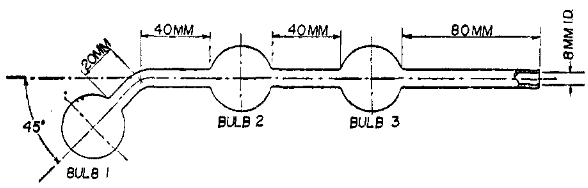

Fig. 1.—Inside diameter of all bulbs about 37 mm.

The residual aminoalcohol was distilled in high vacuum by first putting bulbs 1 and 2 of the distilling apparatus into a small metal box-shaped oven, fitted with a mica window on one side, a removable cover, a slit on one end and a thermometer, and heated with a bunsen burner. During the distillation, the slit in the box was covered with a piece of thick asbestos paper and wet cloth was wrapped around the bulbs remaining outside of the box.

A temperature of 142° was maintained inside of the oven for 15 minutes, allowing the forerun to distil at 0.15 mm. into bulb 3. After that period, the distilling apparatus was moved so that only bulb 1 remained in the box. Small amounts of the forerun condensed in bulb 2 were driven into bulb 3 by careful heating with a bunsen burner. The desired aminoalcohol was then distilled into bulb 2 as a colorless oil at 0.15 mm. and an oven temperature of 160–178°, the operation taking about 25 minutes.

Bulb 2 was then separated by cutting the connections to the other bulbs and the aminoalcohol (1.1205 g.) poured into a Pyrex tube of 100 mm. length and 22 mm. inside diameter. Three ml. of chloroform was used to wash the bottle.

(2) **C^{14}-Labeled N-Benzyl-N-(1-phenoxyisopropyl)-β-chloroethylamine Hydrochloride.**—The tube containing the chloroform solution of the aminoalcohol was cooled in an ice-bath and the procedure given in the literature[2] was followed in preparing the desired compound. Shiny white crystals (1.0414 g.) of the m.p. 137.5–140° (lit.[2] 137.5–140°) were obtained.

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A Convenient Synthesis of Uracil 2-C^{14} from Urea[1]

BY H. GEORGE MANDEL AND CURTIS L. BROWN

Uracil, a normal constituent of pentose nucleic acid, has been shown by many authors to act as a growth factor for various organisms.[2–6] It therefore became desirable to prepare this compound labeled with C^{14} in order to study its physiological disposition in several species. Since C^{14} urea is commercially available,[7] it was desirable to devise a synthesis with this substance as the limiting reagent. Non-radioactive uracil has been prepared in a 25% yield, based on urea, by Davidson and Baudisch.[8] The yield was improved slightly by temperature modifications introduced by Chi and Chen.[9] Hilbert[10] has observed that the amount of sulfur trioxide in the fuming sulfuric acid, the temperature of heating and the order of addition of the reagents influence the success of related condensation reactions.

The procedure outlined below permits the preparation of uracil in a better than 60% yield based on C^{14}-urea.

Experimental

Twenty ml. of fuming sulfuric acid (18% of SO_3) was placed in a three-necked 50-ml. flask equipped with a mercury seal stirrer, a thermometer and a funnel-shaped inlet tube. After cooling the solution in a Dry Ice–alcohol-bath to −5°, 4.4 g. (0.033 mole) of finely pulverized malic acid was added with stirring, keeping the temperature below 0°. When the material had been finely dispersed, 1.7 g. (0.028 mole) of urea, previously pulverized and desiccated, and containing 1 mc. of C^{14}-urea was introduced in small portions over a period of ten minutes, keeping the temperature below 5°, and stirring vigorously. The mixture was then slowly warmed to 80°, whereupon all solid material dissolved. The solution was stirred at 80–85° for one hour, cooled and poured over 60 g. of crushed ice. After 48 hours in the ice-box, the uracil had separated. It was centrifuged, resuspended repeatedly in ice water, filtered off and dried. Recrystallization from hot water, carried out with non-radioactive uracil, showed that this step was unnecessary.

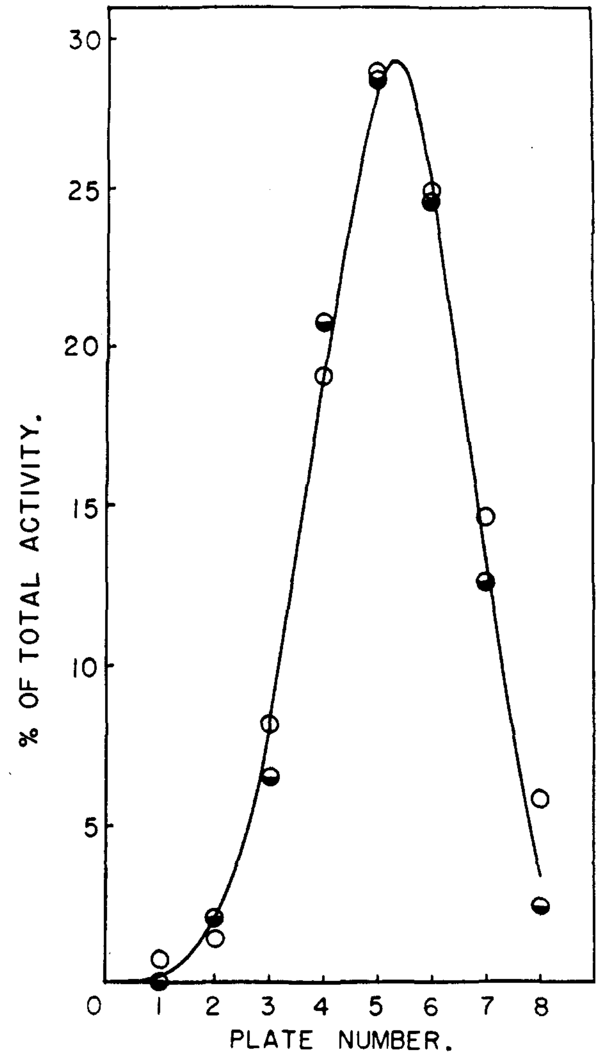

Fig. 1.—Eight-plate countercurrent distribution of uracil 2-C^{14}. Theoretical curve $K = 1.85$ system 1 *M* phosphate buffer *p*H 6.8, *n*-butanol and *t*-butanol; ○, optical density at 260 mμ; ⊖, radio assay.

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[1] Aided by grants from the National Cancer Institute, of the National Institutes of Health, Public Health Service, and the Damon Runyon Fund.

[2] R. D. Housewright and S. A. Koser, *J. Infectious Diseases*, **75**, 113 (1944).

[3] S. H. Hunter, *Arch. Biochem.*, **4**, 119 (1944).

[4] R. E. Feeney, J. H. Mueller and P. A. Miller, *J. Bact.*, **46**, 559 (1943).

[5] E. Diczfalusy and H. v. Euler, *Arkiv Kemi, Mineral. Geol.*, **24A**, No. 38 (1947).

[6] G. W. Kidder, *Ann. N. Y. Acad. Sci.*, **49**, 99 (1947).

[7] Purchased from U. S. Atomic Energy Commission, Los Alamos Scientific Laboratory.

[8] D. Davidson and O. Baudisch, THIS JOURNAL, **48**, 2382 (1926).

[9] Y. F. Chi and Y.-H. Chen, *Trans. Science Soc. China*, **8**, 83 (1934).

[10] G. Hilbert, THIS JOURNAL, **54**, 2081 (1932).

A yield of 1.95 g. of uracil (62% based on urea) was obtained with a specific activity of 0.33 μc./mg.

The substance exhibited an ultraviolet absorption spectrum and an extinction coefficient identical with those reported in the literature.[11,12] A descending chromatogram of the material in a mixture of *t*-butanol–glacial acetic acid–water (65:25:10 v./v.), using Whatman No. 1 paper, showed a single radioactive component having an R_f value of 0.60. The product was subjected to an 8-plate countercurrent distribution in a system of 1 *M* potassium phosphate buffer at *p*H 6.8 and a mixture of equal volumes of *n*-butanol and *t*-butanol. The optical density at 260 mμ for the aqueous and organic layer of each plate was measured, and the sum of the values for the two phases of the various plates is plotted in Fig. 1. The relative radioactivity of each plate was determined by the addition of a constant volume of methanol and water to each plate to make the two layers mutually soluble. Aliquots of these solutions were then plated in plastic cups, dried and assayed for radioactivity in a gas-flow proportional counter. It was found that in addition to the background a correction for naturally occurring K^{40} of the buffer was necessary. The resulting values are plotted in Fig. 1. The close agreement, within the accuracy of the technique, with the calculated curve for authentic uracil having a distribution of 1.85 in such a system, indicated that the substance was of high purity.

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(11) R. D. Hotchkiss, *J. Biol. Chem.*, **175**, 315 (1948).

(12) F. F. Heyroth and J. R. Loofbourow, THIS JOURNAL, **56**, 1728 (1934).

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Absence of Rapid Exchange of Sulfur Atoms between Sulfate and Persulfate Ions

BY P. C. RIESEBOS AND A. H. W. ATEN, JR.

In aqueous solutions a rapid exchange between Hg^{++} and Hg^{2++}-ions has been reported.[1] This observation suggests that an investigation of exchange reactions between ion pairs of the same type might be a matter of some interest. As the sulfate–persulfate combination is fairly easy to handle, we have performed some experiments with this system. It may be pointed out that in this exchange process an oxygen–oxygen bond is affected, whereas in the sulfur exchanges studied earlier, like the $SO_4^=-HS^-$, $SO_4-SO_3^=$, $S_2O_3^=-HS^-$ and $S_2O_3^=-SO_3^=$ reactions,[2] bonds between a sulfur and an oxygen atom or between two sulfur atoms were attached.

Solutions containing radioactive potassium sulfate, labeled with S^{35} (about 0.001 or 0.002 molar) and inactive potassium persulfate (about 0.0005 or 0.001 molar) were kept at room temperature for a week. The sulfate fraction was precipitated as barium sulfate after which the persulfate was decomposed by boiling with hydrochloric acid. Experiments were performed at *p*H values of about 1, about 7 and about 10. In all cases the average value of the specific activity of the sulfur in the persulfate amounted to less than 2% of the specific activity of the sulfate sulfur. (Large differences between figures obtained in duplicate experiments suggest, however, that most or all of the small activity found in the persulfate fraction may well be due to incomplete separation of the two fractions.) Under these circumstances the half-time of exchange amounts to about half a year at least.

Another series of exchange experiments was performed at *p*H about 10 in which the solutions were boiled for 5 minutes. This resulted in the decomposition of about $^1/_3$ of the persulfate, after which the average of the radioactivity in this fraction still did not amount to more than 1.5% of the total activity in the system. (Here again the wide variation of the results suggests that this limit may be far too high.)

We are pleased to express our gratitude to the "Nederlandse Organisatie voor Zuiver Wetenschappelijk Onderzoek" and to the "Stichting voor Fundamenteel Onderzoek der Materie" for their support.

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AMSTERDAM, NETHERLANDS

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(1) S. Ruben, G. T. Seaborg and J. W. Kennedy, *J. Appl. Phys.*, **12**, 308 (1941).

(2) H. Voge, THIS JOURNAL, **61**, 1032 (1939); D. Ames, in A. C. Wahl and N. A. Bonner, "Radioactivity Applied to Chemistry," John Wiley and Sons, Inc., New York, N. Y., 1951, p. 347.

Synthesis of Histamine-2-C^{14}-Imidazole[1]

BY RICHARD W. SCHAYER[2]

Certain bacteria possess an enzyme which converts L-histidine into histamine and carbon dioxide.[3] Rodwell[4] has isolated unspecified strains of *Lactobacilli* possessing a very high histidine decarboxylase activity. Using an acetone powder preparation of these bacteria,[5] radioactive L-histidine has been decarboxylated and the radioactive histamine isolated as the dipicrate.

Experimental

Thiol-L-histidine-2-C^{14}-imidazole.—Radioactive sodium cyanide (approximately 3 mc.) was prepared from C^{14}-barium carbonate without dilution of the isotope by the method of Belleau and Heard.[6] The sodium cyanide was converted to sodium thiocyanate by the method of Castiglioni[7] as adapted by Borsook, *et al.*[8] After dilution with carrier equal to 1.5 times the estimated weight of the isotopic material, the sodium thiocyanate was treated with α,δ-diamino-γ-ketovaleric acid[9] (γ-ketoörnithine) producing 155 mg. of crystalline thiol-L-histidine which failed to melt up to 300°, as reported by Ashley and Harington.[9] Additional radioactive thiolhistidine was crystallized from the mother liquor after addition of carrier.

L-Histidine-2-C^{14}-imidazole.—One hundred and fifty mg. of thiolhistidine was oxidized with ferric sulfate to histidine[8,]

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(1) Supported in part by research grants from the U. S. Public Health Service and the Chicago Heart Association. Radioactive barium carbonate allocated by the Isotopes Division, U. S. Atomic Energy Commission.

(2) The author is indebted to Rosa L. Smiley for assistance.

(3) E. F. Gale, "Advances in Enzymol." Vol. 6, Interscience Publishers, Inc., New York, N. Y., 1946.

(4) A. W. Rodwell, private communication to Dr. Hutton Slade of this Institute.

(5) The author is greatly indebted to Dr. A. W. Rodwell, Research Officer, Commonwealth Scientific and Industrial Research Organization, Melbourne, Australia, for his generosity in supplying the acetone powder of the *Lactobacilli*.

(6) B. Belleau and R. D. H. Heard, THIS JOURNAL, **72**, 4268 (1950).

(7) A. Castiglioni, *Gazz. chim. ital.*, **63**, 171 (1933).

(8) H. Borsook, C. L. Deasy, A. J. Haagen-Smit, G. Keighley and P. H. Lowy, *J. Biol. Chem.*, **187**, 839 (1950).

(9) J. N. Ashley and C. R. Harington, *J. Chem. Soc.*, 2586 (1930).