[CONTRIBUTION FROM THE LOS ALAMOS SCIENTIFIC LABORATORY OF THE UNIVERSITY OF CALIFORNIA]

Micro-syntheses with Tracer Elements. VII. The Synthesis of 2-Methyl-C^{14}-1,4-naphthoquinone (Provitamin K)[1]

BY ARTHUR MURRAY III AND ANTHONY R. RONZIO

RECEIVED OCTOBER 11, 1951

2-Methyl-C^{14}-1,4-naphthoquinone, possessing a specific activity of 57.4 mg./mc., was synthesized in an over-all yield of 48.7% based on methyl-C^{14} iodide. The reaction sequence involved treatment of 2-naphthyllithium with methyl-C^{14} iodide, followed by oxidation of the hydrocarbon so obtained to the quinone. An improved synthesis of methyl-C^{14} iodide is reported. Conditions for the 85% interconversion of 2-iodonaphthalene and *n*-butyllithium are given.

A sample of 2-methyl-1,4-naphthoquinone labeled with C^{14} and possessing the highest possible specific radioactivity was desired for metabolic studies in conjunction with studies using the C^{14}-labeled drugs Pelentan and Dicumarol. The compound has been labeled in the ring with C^{14} by Collins.[2] Since the ring labeled compound was not necessary for our purpose, a much simpler synthesis yielding the compound labeled at the methyl group was devised and is here reported.

The introduction of the methyl-C^{14} group was thoroughly explored using methyl halides and the organometallic reagent obtained by: (1) the halogen–metal interconversion reaction; (2) the halogen replacement reaction; (3) the Grignard reaction. The latter two reactions were unsatisfactory because naphthalene was formed as a by-product.

The 2-naphthyllithium reacted with methyl-C^{14} iodide to give 2-methyl-C^{14}-naphthalene which was oxidized to the quinone.

A study of the halogen–metal interconversion reaction showed that the only by-products were 2,2′-dinaphthyl (m.p. 180–181°), *n*-butyl halide and a trace of 2-naphthyl ethyl ether, all easily removed.

The effect of lowering the temperature of reaction was to decrease the formation of the dinaphthyl. At −60° the reaction was too slow to be useful, while at −26° the yield of product was constant when the duration of interconversion was varied from ten minutes to one hour. The solutions of 2-naphthyllithium were stable for 24 hours when stored at 0°.

The coupling of 2-naphthyllithium with methyl bromide and methyl iodide gave 95–100% yields of 2-methylnaphthalene. Since the *n*-butyl halides interfered in the subsequent oxidation of the methylnaphthalene, they were removed by forming the tertiary amine salts with piperidine.[3]

Several methods for the oxidation of the methylnaphthalene to the quinone were tried. The method of Lucas and Pressman[4] showed the most promise and was modified to adapt it to a micro scale and for use with a radioactive compound. The modified procedure[5] gave 50% yields of purified product. Control experiments in which purified 2-methylnaphthoquinone was carried through this oxidation procedure gave a quantitative return of unchanged quinone.

Experimental

(1) **2-Naphthyllithium.**—This material was prepared essentially by the method of Gilman and Moore[6] using the halogen–metal interconversion reaction between 2-iodonaphthalene and *n*-butyllithium (at −30°, 10–30 min.) in yields of 85–87%.

(2) **Methyl-C^{14} Iodide.**—To one end of a thick-walled Pyrex glass tube (20 × 250 mm.) was sealed a thick-walled glass chamber of 10-ml. capacity and containing a glass enclosed magnetic stirrer and to the opposite end was sealed a conventional pressure stopcock (4 mm. bore) which was in turn sealed to a semi-ball joint. The tubular portion of the apparatus was enclosed with a water jacket for cooling. Through a fine-stemmed funnel passing through the bore of the stopcock was added 7 ml. of hydriodic acid (d. 1.7). The apparatus was then attached to the vacuum system. The acid was frozen with liquid nitrogen and the chamber evacuated to 0.01 mm. Weighed amounts of methanol (2.81–5.10 millimoles) were transferred in vapor phase into the reaction chamber which was then closed off from the system. Water was circulated through the cooling jacket and the contents were stirred while being heated for two hours at 80–85° with an oil-bath. Heating periods tried, varying up to six hours, indicated two hours to be satisfactory. The reaction chamber was cooled to room temperature and the methyl iodide was slowly distilled into an evacuated reservoir cooled with Dry Ice–acetone, through a purifying train of dried soda lime and phosphorus pentoxide. The transfer was completed by warming the reaction chamber with hot water and cooling the reservoir with liquid nitrogen. The reservoir was then freed of non-condensible residual gases, while still cooled with liquid nitrogen, and warmed to room temperature and weighed. The yields of methyl iodide from three trial runs were 99.3, 100.3 and 96.7%. One sample of the methyl iodide was distilled into a solution of 1.5 g. of silver nitrate in 100 ml. of alcohol and sealed off. After warming at 40° for several days, the precipitated silver iodide was filtered and weighed. The yield of iodide was 100.3% based on methyl iodide and 97.0% based on methanol.

The yield of colorless, homogeneous methyl-C^{14} iodide from 0.1295 g. (4.04 millimoles, 12 mc.) of methanol-C^{14} was 0.5735 g. (100%).

(3) **2-Methyl-C^{14}-naphthalene.**—Into a 100-ml. flask equipped with a magnetic stirrer, a connection to the vacuum system, and a closed addition tube, and which had been flushed out with pure, dry nitrogen, was introduced a solution of 3.08 g. (12.13 millimoles) of 2-iodonaphthalene in 50 ml. of ether. The flask was cooled to −30° and a solution of *n*-butyllithium (8.08 millimoles) in ether was added during 2.5 minutes. The mixture was allowed to react 30 minutes, then was immersed in liquid nitrogen, evacuated to a pressure of 0.01 mm. and the methyl-C^{14} iodide distilled into it. The vacuum was replaced by slightly less than one atmosphere of nitrogen. The flask was then allowed to warm to not over 0° and the mixture was stirred for 3 hours. The flask was then again cooled to −30°, an additional 6.05 millimoles of *n*-butyllithium was added (2 minutes) and the mixture was stirred for 30 minutes. The flask was then cooled to −60° and the reaction mixture

(1) Work done under the auspices of the A.E.C.

(2) C. J. Collins, THIS JOURNAL, **73**, 1038 (1951).

(3) J. Semb and S. M. McElvain, *ibid.*, **53**, 690 (1931).

(4) H. J. Lucas and D. Pressman, "Principles and Practice in Organic Chemistry," John Wiley and Sons, Inc., New York, N. Y., 1949, p. 442.

(5) For complete experimental details of this oxidation order Document 3565 from American Documentation Institute, 1719 N Street, N. W., Washington 6, D. C., remitting $1.00 for microfilm (images ½ inch high on standard 35 mm. motion picture film) or $1.00 for photocopies (6 × 8 inches) readable without optical aid.

(6) H. Gilman and F. W. Moore, THIS JOURNAL, **62**, 1843 (1940).

carbonated with dry carbon dioxide gas for 20 minutes at 1 atm. The product was hydrolyzed with 50 ml. of water then transferred into a 3-liter flask with ether and 1.5 liters of water. The 2-methyl-C^{14}-naphthalene was steam distilled directly into a 1-liter liquid–liquid extractor. The distillate was extracted for 28 hours using as small a volume of ether as was practicable. The raffinate showed no radioactivity when checked with a counter. The extract (20 ml.) was treated with 15 ml. of redistilled piperidine in a 200 ml. flask equipped with a reflux condenser and a guard tube filled with KOH pellets. The system was heated under gentle refluxing for 16 hours. The solution was then cautiously acidified with 1:4 hydrochloric acid and extracted with ether in a liquid–liquid extractor for 5.5 hours. The ether extract was dried, treated with "Norit," and filtered directly into the oxidation flask. The ether was removed by distillation through a column.

LOS ALAMOS, N. MEXICO

[CONTRIBUTION FROM THE LOS ALAMOS SCIENTIFIC LABORATORY OF THE UNIVERSITY OF CALIFORNIA (UNDER THE AUSPICES OF THE A.E.C.)]

Micro Syntheses with Tracer Elements. VIII. The Synthesis of Thiamin Labeled with C^{14}

BY D. LLOYD WILLIAMS AND ANTHONY R. RONZIO

RECEIVED AUGUST 22, 1951

2-Amino-4-methyl-5-(β-hydroxyethyl)-thiazole-2-C^{14} has been prepared and converted to 4-methyl-5-(β-hydroxyethyl)-thiazole-2-C^{14}, in high yield, by the reduction of the diazonium compound. The deaminated thiazole has been used to prepare C^{14} labeled thiamin with a specific activity of 170 mg./mc. It has been shown that 2-methyl-4-amino-5-bromomethylpyrimidine hydrobromide reacts with butanol to form an ether.

The availability of thiourea[1] labeled with either S^{35} or C^{14} makes possible the labeling of thiamin with these isotopes.

The condensation of thiourea with γ-chloro-γ-acetopropanol in boiling aqueous medium gave 83 to 90% yields of 2-amino-4-methyl-5-(β-hydroxyethyl)-thiazole. In the only published preparation of this compound, from the above intermediates, giving yield data either no solvent[2] or alcohol solvent[3] was used. The reported yields were 72 and 29%, respectively. Hatcher[4] reported 80% yields of this aminothiazole from thiourea and γ-chloro-γ-acetopropyl ether.

A modification of the procedure of Hatcher,[4] who prepared the desired thiazole in 31% yield, for the deamination of this aminothiazole gave reproduceably high yields (70%) of 4-methyl-5-(β-hydroxyethyl)-thiazole.

Published procedures for the condensation of the pyrimidine and thiazole moieties utilize a one to two molar excess of the thiazole. Williams, *et al.*,[5] obtained a 45% yield of thiamin from two moles of thiazole to one of pyrimidine with butanol as solvent. Gravin,[6] using 2.4 moles of thiazole to one of pyrimidine in bromoform solvent, obtained a 54% yield of thiamin, while a 70% yield of a thiamin isomer was obtained by Baumgarten[7] using a 3 to 1 molar ratio of 4-methyl-5-(α-hydroxyethyl)-thiazole to pyrimidine in nitromethane solvent.

Obviously it is not feasible to use an excess of thiazole when this is the isotope labeled moiety. In an effort to determine why an excess of the thiazole is conducive to good yields, an experiment was made to determine if the pyrimidine reacts with itself to form a quaternary base or with butanol to form an ether. A practically quantitative yield of material melting at 135–136° was obtained which was indicated by analysis to be the hydrobromide of 2-methyl-4-amino-5-butoxymethylpyrimidine. The free base, m.p. 84°, was also prepared. These compounds appear not to have been previously described in the literature.

Use of the conditions described by Baumgarten[7] with nitromethane as solvent but with an excess of pyrimidine (3 to 1 molar ratio) gave a lower yield of thiamin than the experiments with butanol as solvent and a 1 to 1 ratio of reactants. All attempts to isolate unreacted thiazole from the mother liquors from the latter experiments were without success. This result was further confirmed when C^{14} labeled thiazole was used in the condensation.

In view of the above results, the major side reactions appear to involve thiazole and possibly some thiamin which has already formed. Both the unreacted thiazole and the desired product, thiamin, have the β-hydroxyethyl group which is capable of reacting with the 5-bromomethyl group of the pyrimidine moiety to form an ether.

Experimental

2-Amino-4-methyl-5-(β-hydroxyethyl)-thiazole-2-C^{14}.—C^{14} Thiourea, 0.1697 g. (0.0022 mole) reacted with γ-chloro-γ-acetopropanol, 0.394 g. (0.0029 mole), in boiling aqueous medium for 2.5 hours. An 86.7% yield (0.3746 g.) of the aminothiazole was isolated as the hydrochloride from dry ether solution. In preliminary experiments with unlabeled thiourea, in which reaction time and ratio of reactants were varied, 83 to 90% yields of the aminothiazole were isolated as the hydrochloride in the same manner, m.p. 153–154°. Inasmuch as Hatcher[4] claimed to have obtained a di-hydrochloride there was some question as to the composition of our compound. Therefore a sample of the unlabeled hydrochloride, m.p. 153°, was analyzed for chlorine.

Anal. Calcd. for $C_6H_{10}N_2OS.HCl$: Cl, 18.2. Found: Cl, 18.3, 18.6.

The free base was prepared and crystallized from benzene in colorless needles, m.p. 93°. This material formed a

(1) C. W. Bills and A. R. Ronzio, THIS JOURNAL, **75**, 5510 (1950).

(2) A. R. Todd, F. Bergel, H. L. Fraenkel-Conrat and A. Jacob, *J. Chem. Soc.*, 1601 (1936).

(3) U. P. Basu and S. J. Das-Gupta, *J. Indian Chem. Soc.*, **15**, 160, 164 (1938) (*C. A.*, **32**, 7040[4] (1938)).

(4) J. B. Hatcher, THIS JOURNAL, **69**, 465 (1947).

(5) Joseph K. Cline, Robert R. Williams and Jacob Finkelstein, *ibid.*, **59** 1052 (1937).

(6) A. I. Gravin, *J. Applied Chem. (USSR)*, **16**, 105 (1943); *C. A.*, **38**, 1239 (1944).

(7) P. Baumgarten, A. Dornow, K. Gutschmidt and H. Krebl, *Ber.*, **75B**, 444 (1942).