6-Methyl-2,4-dioxytetrahydro-*s*-triazine (X) was prepared according to Ostrogovich[5] and its absorption spectrum in the ultraviolet compared to that of allantoxaidin at different *p*H values. A great similarity of the absorption curves of the two compounds was noted. The wave lengths of the maximal absorption and the extinction coefficients are given in the Experimental part. Parabanic acid and hydantoin, whose structures would be similar to the old structure for allantoxaidin, do not have an ultraviolet absorption of the above-mentioned type, but instead exhibit only end absorption above 215 mμ.

The 6-methyltriazine also shows many chemical similarities to allantoxaidin such as ease of oxidation to cyanuric acid, hydrolysis in concentrated ammonia to acetic acid and biuret and reduction by sodium amalgam with uptake of two hydrogens.[6]

TABLE I

OXIDATION OF C^{14}-LABELED URIC ACIDS

Compound	Specific activity, c.p.m./μM. Uric acid-4-C^{14}	Specific activity, c.p.m./μM. Uric acid-5-C^{14}
Uric acid	84	22.5
First CO_2	0.2	0.4
Second CO_2	0.1	18.5
Allantoxaidin	89	0.0
Formic acid	87	..
Biuret	0.1	..

Experimental

4-C^{14}-Uric Acid.—4-C^{14}-Hypoxanthine was synthesized by Dr. Walter Brooks by the method of Shaw and Woolley.[7,8] This was converted to 4-C^{14}-uric acid by the action of xanthine oxidase.

5-C^{14}-Uric Acid.—5-C^{14}-Hypoxanthine was prepared from 2-C^{14}-glycine in an *in vitro* pigeon liver system, synthesizing inosinic acid.[9] The hypoxanthine, obtained by hydrolysis of the inosinic acid, was converted to uric acid with xanthine oxidase. Uric acid was isolated by a standard procedure after addition of carrier.[10]

Potassium Oxonate and First CO_2 Fraction.—Five hundred milligrams of uric acid was dissolved in a solution of 1.7 g. of KOH in 15 ml. of water and 7.5 ml. of 30% hydrogen peroxide. After standing for 24 hours, a small amount of manganese dioxide was added to destroy the excess peroxide, and the MnO_2 then removed by centrifugation. Four ml. of glacial acetic acid was added to bring the solution to about *p*H 5, and the carbon dioxide evolved was collected in saturated $Ba(OH)_2$ solution. The reaction mixture was cooled in ice while the potassium oxonate crystallized out; yield 360 mg., 63%.

Anal. Calcd. for $C_4H_2O_4N_3K$: K, 20.04. Found: K, 20.66.[11]

Allantoxaidin and Second CO_2 Fraction.—The potassium oxonate was dissolved in the minimum amount of water and 3 ml. of 10% silver nitrate solution added. The precipitate of silver oxonate was centrifuged off and washed with water. This was suspended in 3 ml. of water and 2 ml. of concentrated hydrochloric acid was added. The solution was warmed to 60° to hasten the decarboxylation reaction and the carbon dioxide evolved was collected as $BaCO_3$. When carbon dioxide evolution ceased, the silver chloride was centrifuged off, the supernatant taken to dryness *in vacuo* and washed with water to remove the HCl. The product was recrystallized from methanol; yield 109 mg., 52%. This product was dried at 100° over P_2O_5 for 24 hours for analysis.

Anal. Calcd. for $C_3H_3N_3O_2$: C, 31.87; H, 2.67; N, 37.17. Found: C, 32.03; H, 2.69; N, 37.36.[11]

Hydrolysis of Allantoxaidin to Formic Acid and Biuret.—Seventy milligrams of allantoxaidin were heated with 5 ml. of concentrated ammonium hydroxide on the steam-bath until the volume was reduced to 2 ml. Then 3 ml. more of ammonium hydroxide was added and the volume taken down again to 2 ml. One ml. of 0.5 *M* lead nitrate solution and 10 ml. of ethanol were added. The lead formate precipitate was centrifuged off and washed with ethanol. These washings were added to the supernatant solution. The lead formate was treated with 2 ml. of 10% $HgSO_4$ and 1 ml. of concentrated sulfuric acid to oxidize the formic acid to carbon dioxide, which was collected as barium carbonate.

Oxidation of Allantoxaidin and 6-Methyl-2,4-dioxytetrahydrotriazine to Cyanuric Acid.—The acid permanganate method of Biltz[4] was used to oxidize both compounds. Oxidation of the 6-methyltriazine to cyanuric acid also could be accomplished by boiling with 10% nitric acid or by heating with bromine water.

Reduction of Allantoxaidin with Sodium Amalgam.—One hundred milligrams of allantoxaidin was dissolved in 10 ml. of water and heated to 100° on a steam-bath. Ten grams of 2% sodium amalgam was added slowly with stirring while the solution was maintained slightly acid at all times with HCl. When hydrogen evolution stopped, the solution was cooled in ice and the product precipitated. After recrystallization from hot water the product sublimed at 260° (uncorrected) and melted with decomposition at 287° when sealed in a capillary. It exhibited only end absorption in the ultraviolet region; yield 40 mg., 39%.

Anal. Calcd. for $C_3H_5N_3O_2$: C, 31.32; H, 4.38; N, 36.53. Found: C, 31.90; H, 4.38; N, 36.68.[11]

ULTRAVIOLET ABSORPTION OF ALLANTOXAIDIN AND 6-METHYL-2,4-DIOXYTETRAHYDRO-*S*-TRIAZINE

Compound	Maximum, mμ, at *p*H 2.2	Molecular extinction coeff. × 10^{-3} at *p*H 2.2	Maximum, mμ, at *p*H 11.6	Molecular extinction coeff. × 10^{-3} at *p*H 11.6
Allantoxaidin	235	5.75	252.5	7.75
6-Methyl-2,4-dioxytetrahydro-*s*-triazine	232.5	1.85	250	2.15

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[11] All microanalyses were carried out by the Microchemical Laboratory, M.I.T., Dr. S. M. Nagy.

The Synthesis of 9,10-Dimethyl-1,2-benzanthracene-9,10-C^{14} [1]

BY HERBERT I. HADLER

RECEIVED AUGUST 25, 1954

The potent carcinogen 9,10-dimethyl-1,2-benzanthracene has been labeled with C^{14} in the 9,10-positions as indicated below. By means of the Grignard reaction, *o*-bromotoluene and $C^{14}O_2$ gave carboxy-labeled *o*-toluic acid. Oxidation with alkaline potassium permanganate followed by sublimation[2] resulted in phthalic anhydride in 67% yield from $BaC^{14}O_3$. After condensing the anhydride with naphthalene by conducting the Friedel–Crafts reaction in ethylene chloride (a solvent recom-

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[2] W. Werth, *Ber.*, **7**, 1057 (1874).

mended for α-acylation[3]) the resultant keto acid was cyclized to 1,2-benz-9,10-anthraquionone by the procedure of Badger and Cook.[4] The desired hydrocarbon was obtained by the method of Sandin and Fieser,[5] who after condensing the quinone with the Grignard reagent of methyl iodide, conducted a two-step reduction with hydrogen iodide followed by stannous chloride. The over-all yield of 9,10-dimethyl-1,2-benzanthracene-9,10-C^{14} (2.77 millicuries per millimole) from barium carbonate was 18%.[6]

(3) G. Baddeley, *J. Chem. Soc.*, S 59 (1949).

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(6) The experimental details of this synthesis have been deposited as Document number 4392 with the ADI Auxiliary Publications Project, Photoduplication Service, Library of Congress, Washington 25, D. C. A copy may be secured by citing the Document number and by remitting in advance $1.25 for photoprints, or $1.25 for 35 mm. microfilm by check or money order payable to Chief, Photoduplication Service, Library of Congress.

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Synthesis of Centrally Labeled (15,15′)-β-Carotene

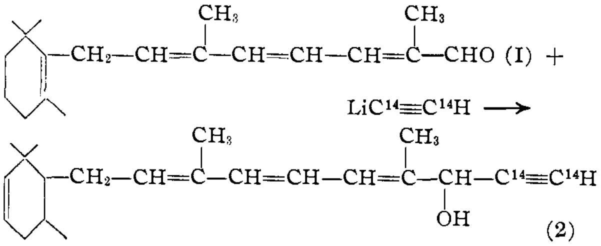
BY H. H. INHOFFEN,[1a] U. SCHWIETER,[1a] C. O. CHICHESTER[1b] AND G. MACKINNEY[1b]

RECEIVED OCTOBER 16, 1954

The synthesis of β-carotene with centrally-labeled carbons 15 and 15′ may be accomplished by use of radioactive acetylene in the reaction, expressed schematically[2]

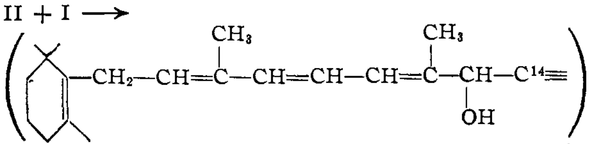
$$C_{19} + C^{14}_2 + C_{19} \longrightarrow C_{19}\text{–}C_2^{14}\text{–}C_{19} \qquad (1)$$

Grignard acetylene is normally prepared with a large excess of the gas, reacting for a considerable period. It is formed slowly, and the reaction is incomplete when the acetylene is limiting, and since dilution of C^{14}-acetylene with inert gas must be kept at a minimum, it has been necessary to modify the procedure,[3a] and the acetylene is first partially condensed with β-C_{19}-aldehyde[3b] and the β-C_{21}-acetylenecarbinol, (compound II) is obtained, then coupled, in a second reaction with additional C_{19}-compound to form the C_{40}-yn-diol. Lithium acetylide is formed readily, the reaction proceeds quantitatively in liquid ammonia, and no large excess of acetylene is required. The over-all scheme, equation 1, may be represented by steps 2–6



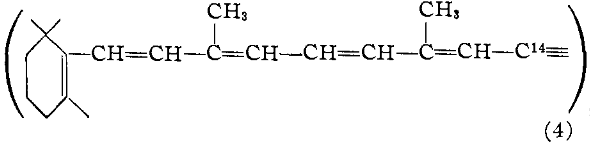
(2)

II + I ⟶



(3)

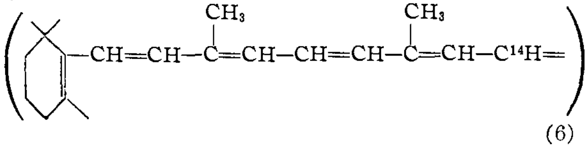
III − H_2O ⟶



(4)

IV + H_2 ⟶ central mono-*cis*-β-carotene (V) (5)

V = hν ⟶



(6)

The condensation of II with a second mole of I proceeds smoothly to form the 15,15′-yn-diol III. The removal of water to yield IV, a dehydrocarotene $C_{40}H_{54}$ and subsequent selective partial hydrogenation of the central triple bond gives rise experimentally only to central mono-*cis*-β-carotene.[4] This is then irradiated for a suitable length of time. The course of the reaction is followed spectrophotometrically, to determine maximum yield of β-carotene. This is then isolated chromatographically and crystallized.

Experimental[5,6]

Acetylene.—The yield of acetylene from the original $BaC^{14}O_3$ was 94 to 96% of theoretical, achieved by placing 100 mg. of carbonate between layers of finely ground barium metal. The subsequently evolved acetylene was condensed as a solid at the temperature of liquid nitrogen. Inactive acetylene completes the reaction with lithium ammonia.

β-C_{21}-Acetylenecarbinol.—The acetylene is in excess of the theoretical amount to couple with the C_{19}-aldehyde. As the reaction progresses, some acetylene is released from the mixture. It is trapped as silver acetylide and recycled to conserve activity. The removal of the lithium from the lithium acetylide addition compound with the C_{19}-aldehyde, to form the C_{21}-acetylenecarbinol is presumably a hydrolysis. The ammonium chloride used to effect the removal of the lithium was dried under vacuum at 80° as the presence of water is detrimental, and acetylene will be released.

Irradiation.—Formation of the all-*trans*-β-carotene from the central mono-*cis* isomer was followed spectrophotometrically, by noting the ratio of the maximum at 450 mμ to that of the *cis* peak at 338 mμ. The reaction is stopped when the ratio is *ca.* 10:1. The ratio will continue to rise for a while on prolonging the irradiation, but the absolute value at 450 mμ begins to fall. This must be avoided. The solution is then concentrated under vacuum and chromatographed. The all-*trans*-β-carotene is adsorbed above the residual mono-*cis* isomer and also above any dehydrocarotene that may be present. The all-*trans* isomer is crystallized and the other two components recycled.

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