Synthesis of Compounds Related to Coenzyme Q

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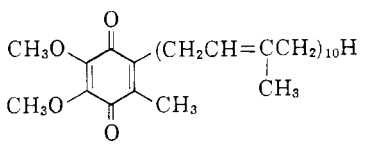
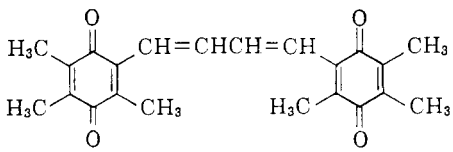
Several compounds structurally related to coenzyme Q have been prepared. Unfortunately, it was not possible to place quinoid groups at the end of the chain in these compounds because the related hydroquinone ethers could not be cleaved without complete loss of the unsaturated side chain. In connection with this work, there was developed a new and convenient method for syntheses of highly substituted diphenylbutadienes, and of highly substituted terphenyls.

Since the discovery of coenzyme Q in animal tissue by Crane[2] in 1957 and by Bouman[3] and Morton[4,5] in 1958, many publications have appeared concerning the isolation, structure, and synthesis of various members of the coenzyme Q family. The structure was established as I by Folkers and co-workers[6] in 1958–1959 through NMR spectra and degradation studies.

The various co Q compounds isolated to date differ in the number of isoprenoid units in the side chain and by the presence of methyl rather than methoxy groups in the nucleus.[6,7]



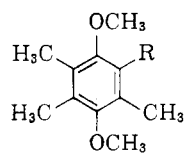
I



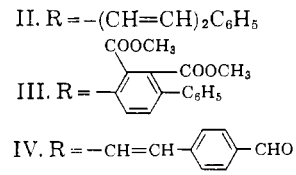
Ia

In order to explore somewhat the structural limits of compounds with Q activity, the synthesis of some related compounds was undertaken. In particular, it was desired to replace the isoprenoid chain by one containing conjugated double bonds and to place at each end of the chain a quinoid nucleus. The research was therefore directed toward the synthesis of the simpler model compound Ia. The complete synthesis of Ia was not successful because it was not possible to cleave the hydroquinone ethers (which were necessary intermediates) without complete loss of the unsaturated side chain. Nevertheless, the synthesis of some closely related compounds of type II was successful, and incidental to this, a new and convenient method for synthesis of certain highly substituted terphenyls was developed.

The route to compounds of type II began with chloromethylation of the dimethyl ether of trimethylhydroquinone[8] (IIa, R = H) to give the chloromethyl compound VI. The Wittig reaction was then employed as the method for introduction of the unsaturated side chain. Reaction of VI with triphenylphosphine in dimethylformamide gave the phosphonium salt VII in nearly quantitative yield. Action of lithium ethoxide in absolute ethanol upon VII produced the intermediate phosphine methylene, and action of the appropriate aldehyde upon the ylide led to the final product.



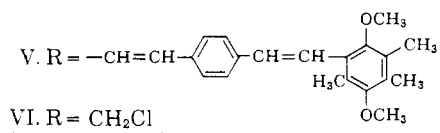
IIa



II. R = $-(CH{=}CH)_2C_6H_5$

III. R =

IV. R =



V. R =

VI. R = CH_2Cl

VII. R = $-CH_2\overset{+}{P}(C_6H_5)_3\bar{C}l$

Reaction of yield with cinnamaldehyde proceeded smoothly, yielding only one isomeric form of the butadiene II, assumed to be the thermodynamically more stable *trans-trans* form. Presence of the 1,3-diene system in II was confirmed through reaction of II with typical dienophiles and isolation of 1:1 adducts.

The reaction between II and tetracyanoethylene was characterized by formation of the dark colored π complex. After twenty-four hours a nearly quantitative yield of the adduct VIII was obtained. Treatment of VIII with pyridine yielded an isomer, probably resulting from migration of

(1) Abstracted from a thesis by John J. Baldwin, presented to the Graduate Faculty of the University of Minnesota in partial fulfillment of the requirements for the Ph.D. degree, August, 1960.

(2) Crane, Hatefi, Lester, and Widmer, *Biochem. et Biophys. Acta*, **25**, 220 (1957).

(3) Bouman, Slater, Rudney, and Links, *Biochem. et Biophys. Acta*, **29**, 456 (1958).

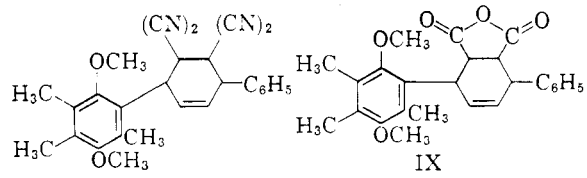
(4) Fahmy, Hemming, Morton, Paterson, and Pennock, *Biochem. J.*, **70**, 1 P (1958).

(5) Morton, Gloor, Schendler, Wilson, Chopar-dit-Jean, Hemming, Isler, Leat, Pennock, Ruegg, Schwieter, and Wiss, *Helv. Chim. Acta*, **41**, 2343 (1958).

(6) Wolf, Hoffman, Trenner, Arison, Shunk, Lenn, McPherson, and Folkers, *J. Am. Chem. Soc.*, **80**, 4752 (1958).

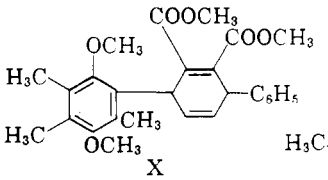
(7) Shunk, Erickson, Wong, and Folkers, *J. Am. Chem. Soc.*, **81**, 5000 (1959).

(8) Smith and Opie, *J. Am. Chem. Soc.*, **63**, 937 (1941).

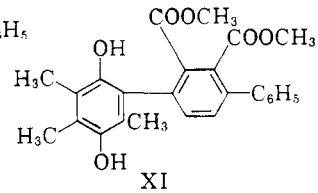


VIII

IX



X



XI

the double bond into conjugation with the aromatic ring.

The maleic anhydride adduct IX formed in 62.8% yield using dry refluxing xylene as the solvent.

The reaction of II with dimethyl acetylenedicarboxylate did not proceed in benzene; only tar was produced using excess dienophile at the reflux temperature. But when *o*-dichlorobenzene was used as the solvent, X was isolated as the major reaction product along with some of the terphenyl III. Action of chloranil upon X in refluxing xylene converted the compound smoothly into III.

The 1,4-distyryl benzene V resulted from the reaction of two equivalents of the ylide with one of terephthaldehyde. The reaction proceeded readily at room temperature and was complete in three hours. The *p*-styrylbenzaldehyde IV could be isolated only when an equivalent amount of lithium ethoxide (based on the weight of lithium) was used. The formation of IV under these conditions was attributed to a partial decomposition of the base, thereby decreasing the amount of ylide available for reaction with the terephthaldehyde. IV could be converted to V by reaction with a second equivalent of the ylide.

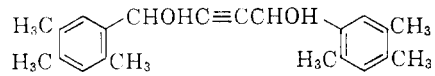
Many attempts were made to cleave the other linkages of II and IV in an effort to generate the corresponding hydroquinone. Action of hydrobromic acid in acetic acid, of pyridine hydrochloride, and of potassium hydroxide in ethylene glycol at 225° were unsuccessful. Some success in cleaving the ether linkage was achieved, but the reaction proceeded beyond a simple ether cleavage: Thus, when either II or IV was subjected to action of aluminum chloride in refluxing *o*-dichlorobenzene, the unsaturated side chain was eliminated, and only trimethylhydroquinone was isolated. A somewhat similar reaction has been reported by Buu-Hoï.[9]

Action of aluminum chloride in *o*-dichlorobenzene or of hydrobromic acid in acetic acid upon III produced a new compound. Infrared evidence suggested this to be the hydroquinone XI. However, XI could not be obtained in an analytically pure state and its structure, therefore, has not been established with certainty.

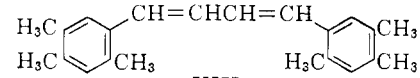
In preparation of II, IV, and V it was noteworthy that the bulky ylide generated from VII reacted under the normal mild conditions with the carbonyl group, emphasizing the generality of the Wittig reaction.

Since the derivatives of the methyl ether of trimethylhydroquinone could not be cleaved, the dibenzyl ether was tried, but this compound could not be chloromethylated. The diacetate of the hydroquinone was investigated. This could be chloromethylated, although with some difficulty, and the chloromethyl compound could be converted into the triphenylphosphonium salt. However, this salt either failed to form the ylide, or else the ylide failed to react with aldehydes, for no product was obtained when the phosphonium salt was subjected to the sequence of these two reactions. In the hope that a phenol ether might be more tractable, and that later the second hydroxyl group could be introduced, the benzyl ether of 2,3,5-trimethylphenol was chloromethylated. Here, however, the chloromethyl group entered the *para* position, rather than the *ortho* position, and the resulting compound was useless for the purpose at hand.

In a model experiment for another approach to compounds of type II, trimethylbenzaldehyde was subjected to the action of acetylene. The acetylenic glycol XII was obtained in fair yield, and this, when reduced by action of lithium aluminum hydride, was smoothly converted into the butadiene XIII. However, the necessary aldehyde for the purpose in hand—trimethyldimethoxybenzaldehyde, itself a difficult compound to prepare—failed to react with acetylene.



XII



XIII

The experiments reported here open the way to convenient syntheses of many highly substituted 1,4-diphenylbutadiene and distyrylbenzenes, and from these, to the preparation of a wide variety of highly substituted terphenyls. The reactions proceed smoothly, and the yields in all the steps are quite acceptable for preparation purposes.

Experimental[10]

Triphenyl(2,5-dimethoxy-3,4,6-trimethylbenzyl)phosphonium Chloride (VII).—A solution of triphenylphosphine

(9) Buu-Hoï and Lavit, *Proc. Chem. Soc.*, 120 (1960).

(10) Microanalysis by O. Hamerston, J. Keenan, and J. Johnson.

(15 g., 0.057 mole) and 2,5-dimethoxy-3,4,6-trimethylbenzyl chloride VI[8] (11.0 g., 0.049 mole) in *N,N*-dimethylformamide was heated at 65° for 16 hr. The solvent was removed under reduced pressure until the product began to precipitate, and dry ether (250 ml.) was then added. If the phosphonium salt separated as an oil, it could be induced to crystallize by vigorous agitation with a glass rod. The white solid (22.75 g., 0.046 mole, 94.6%, m.p. 233–235.5°) was filtered and washed with dry ether. The product was pure enough for further reaction but it could be recrystallized from absolute ethanol–ether.

Anal. Calcd. for $C_{30}H_{32}O_2PCl$: C, 73.38; H, 6.56. Found: C, 73.65; H, 6.78.

1-Phenyl-4-(2,5-dimethoxy-3,4,6-trimethylphenyl)-1,3-butadiene (II).—To a solution of the phosphonium salt VII (22 g., 0.05 mole) and redistilled cinnamaldehyde (7.0 g., 0.053 mole) in absolute ethanol at 50° was added an alcoholic solution of lithium ethoxide (0.7 g., 0.1 g.-atom of lithium in 100 ml. of absolute ethanol). An orange color developed after the addition of base. The reaction mixture was stirred and maintained at 50° until the product had precipitated from solution (4 hr.). Water (100 ml.) was added and the flocculent yellow precipitate was removed by filtration and recrystallized from ethanol and cyclohexene. It weighed 8.8 g. to 9.4 g., 0.0387 mole to 0.03 mole, 57.4% to 61% yield, and had m.p. 113–113.5°.

Anal. Calcd. for $C_{22}H_{24}O_2$: C, 81.78; H, 7.84. Found: C, 81.50; H, 7.93.

There was no effect on the yield when the reaction was allowed to proceed for an additional 24 hr. or when a nitrogen atmosphere was maintained. When the reaction time at room temperature was reduced to 45 min., the yield fell to 45%.

1,4-Di(2,5-dimethoxy-3,4,6-trimethylstyryl)benzene (V).A.— To a solution of terephthaldehyde (1.6 g., 0.014 mole) and the phosphonium salt VII (12 g., 0.0287 mole) in absolute ethanol (90 ml.) was added ethanolic lithium ethoxide (0.36 g., 0.05 g.-atom of lithium in 100 ml. of absolute ethanol). A yellow-green solid began to precipitate from the solution after 10 min. and the reaction was complete after 3 hr. The solid was removed by filtration, washed with ethanol, and recrystallized from benzene. It then weighed 3.75 g., 65% yield, and had m.p. 209–210.5°.

Anal. Calcd. for $C_{32}H_{38}O_4$: C, 78.98; H, 7.87. Found: C, 78.56; H, 7.79.

When only an equivalent amount of lithium ethoxide (based on weight of lithium) was used, 4-(2,5-dimethoxy-3,4,6-trimethylstyryl)benzaldehyde (IV) could be isolated from the mother liquor in 35% yield (m.p. 99.5–101°).

Anal. Calcd. for $C_{20}H_{22}O_3$: C, 77.39; H, 7.14. Found: C, 77.40; H, 7.28. The 2,4-dinitrophenylhydrazone melted at 236–237°.

Anal. Calcd. for $C_{26}H_{26}O_6N_2$: C, 63.66; H, 5.34; N, 11.42. Found: C, 64.11; H, 5.21; N, 11.41.

B. **From 4-(2,5-Dimethoxy-3,4,6-trimethylstyryl)benzaldehyde (IV).**—The phosphonium salt VII (0.5 g., 0.0011 mole) and IV (0.31 g., 0.001 mole) were dissolved in absolute ethanol (20 ml.). To this was added with stirring an ethanolic solution of lithium ethoxide (0.002 g.-atom of lithium in 25 ml. of absolute ethanol). The reaction mixture was heated to 50° and after 5 min. the distyrylbenzene VIII began to appear in the reaction mixture as a greenish yellow solid. The mixture was allowed to stand overnight, and the product was then removed by filtration and washed with ethanol. It weighed 0.4 g., 0.00082 mole, 82.3% yield, and had m.p. 209–210.5°.

Action of Aluminum Chloride on II.—Aluminum chloride (3.74 g., 0.028 mole) was added to a stirred solution of II (2.1 g., 0.007 mole) in *o*-dichlorobenzene (50 ml.). The red reaction mixture was heated slowly to refluxing under a nitrogen atmosphere, and then allowed to cool to room temperature. The mixture was acidified with 10% aqueous sulfuric acid (75 ml.) and extracted with ether. The extracts were dried over sodium sulfate, the solvent was removed under reduced pressure, and the residue was triturated with petroleum ether (b.p. 60–68°). The resulting brown solid was removed by filtration and sublimed to 140° and 1 mm. It weighed 0.4 g., and had m.p. 179–180°, and was shown to be trimethylhydroquinone on comparison with an authentic sample.

Action of Aluminum Chloride on V.—Aluminum chloride (11.1 g., 0.086 mole) was added to a stirred solution of V (5.5 g., 0.011 mole) in *o*-dichlorobenzene (60 ml.). The red reaction mixture was slowly heated to refluxing while a nitrogen atmosphere was maintained. Heating was discontinued and the mixture was allowed to cool to room temperature. Dilute sulfuric acid (150 ml., 10%) was added and the mixture was extracted with ether. The combined ether extracts were dried over sodium sulfate; the solvent was removed under reduced pressure and the residue was triturated with petroleum ether (150 ml., b.p. 60–68°). The greenish brown solid was removed by filtration and sublimed at 140° and 1 mm. It weighed 1.73 g., and had m.p. 179.5–180°. It was identified as trimethylhydroquinone.

1-Phenyl-4-(2,5-dimethoxy-3,4,6-trimethylphenyl)-5,5,-6,6-tetracyanocyclohexene-2 (VIII).—To a solution of tetracyanoethylene (1.6 g., 0.0125 mole) in dry benzene (50 ml.) was added dropwise over 15 min. a benzene (35 ml.) solution of II (3.8 g., 0.0123 mole).

The reaction mixture immediately became very dark green, indicating the formation of a π complex between the reactants. The mixture was refluxed for 23 hr. (now dark yellow). The benzene was removed and the residue was triturated with petroleum ether (b.p. 60–68°). The resulting gray-white solid was removed and recrystallized from ethanol. It weighed 4.7 g., 87.6%, and had m.p. 166–168°.

Anal. Calcd. for $C_{27}H_{24}O_2N_4$: C, 74.24; H, 5.54; N, 12.83. Found: C, 74.24; H, 5.65; N, 12.57.

Action of Pyridine on VIII.—The tetracyanoethylene adduct VIII (4 g., 0.0091 mole) in dry pyridine (50 ml.) was refluxed for 1 hr. Pyridine was removed by distillation from the black solution and the dark residue was dissolved in ethanol. On cooling the product crystallized as white needles. It weighed 1.55 g., 38.75% yield, and had m.p. 226–228°. This was an isomer of VIII.

Anal. Calcd. for $C_{27}H_{24}O_2N_4$: C, 74.24; H, 5.54; N, 12.83. Found: C, 74.22; H, 5.41; N, 12.60.

3-Phenyl-6-(2,5-dimethoxy-3,4,6-trimethylphenyl)-Δ_4-tetrahydrophthalic Anhydride (IX).—A solution of II (2.15 g., 0.007 mole) and maleic anhydride (0.68 g., 0.007 mole) in dry xylene (40 ml.) was refluxed for 24 hr. The solvent was then removed by heating the solution on a steam bath under a stream of nitrogen, and the residue was triturated with petroleum ether (b.p. 60–68°). The viscous oil solidified; the solid was removed by filtration and recrystallized from ethanol. It weighed 1.8 g., 0.0044 mole, 62.8% yield, and had m.p. 209–210°.

Anal. Calcd. for $C_{25}H_{26}O_5$: C, 73.86; H, 6.45. Found: C, 73.35; H, 6.52.

1-Phenyl-4-(2,5-dimethoxy-3,4,6-trimethylphenyl)-2,3-dicarbomethoxycyclohexa-2,5-diene (X).—A solution of II (4 g., 0.013 mole) and dimethyl acetylenedicarboxylate (2.3 g., 0.016 mole) in *o*-dichlorobenzene (10 ml.) was heated with stirring at 140° for 14 hr. The resulting dark red solution was heated on a steam bath under a stream of nitrogen until the solvent had been removed. The dark brown, gummy residue slowly crystallized when it was triturated with petroleum ether (b.p. 60–68°). The solid was removed by filtration and recrystallized from ethanol (200 ml.). It weighed 2.8 g., 0.0062 mole, 47.7% yield, and had m.p. 178°.

Anal. Calcd. for $C_{27}H_{30}O_6$: C, 71.98; H, 6.71. Found: C, 72.25; H, 6.55.

Concentration of the mother liquor yielded a white solid (0.5 g., 0.0011 mole, 8.4% yield, m.p. 143–144°). This

compound was the terphenyl III resulting from the dehydrogenation of the Diels-Alder adduct X. Its infrared spectrum was identical with that of the compound obtained through the reaction of X with chloranil.

2,5-Dimethoxy-3,4,6-trimethyl-2′,3′-dicarbomethoxy-*p*-terphenyl (III).—A solution of X (0.9 g., 0.002 mole) and chloranil (0.49 g., 0.002 mole) in dry xylene (20 ml.) was refluxed for 24 hr. The red solution was cooled to room temperature and diluted with ether (50 ml.). The solution was extracted with aqueous 10% sodium hydroxide until the washings were no longer colored. The organic layer was then washed with water (100 ml.). The solvent was removed on a steam bath under a stream of nitrogen and the dark residue was triturated with petroleum ether (b.p. 60–68°). The oil solidified to a yellow solid which was recrystallized from ethanol–water. It weighed 0.7 g., 0.0016 mole, 78% yield, and had m.p. 143–144°.

Anal. Calcd. for $C_{27}H_{28}O_6$: C, 72.30; H, 6.29. Found: C, 72.30; H, 6.32.

Action of Aluminum Chloride upon III.—Aluminum chloride (1 g., 0.008 mole) was added to a solution of III (0.45 g., 0.001 mole) in *o*-dichlorobenzene (12 moles). The dark solution was refluxed for 5 min. under a nitrogen atmosphere and then cooled to room temperature. Dilute sulfuric acid was added (100 ml., 10%) and the mixture was extracted with ether (100 ml.). A yellow, insoluble material was removed by filtration. The ether solution was evaporated on a steam bath under a stream of nitrogen. After trituration with petroleum ether (b.p. 60–68°) the residue solidified and was filtered. The white solid XI was recrystallized from benzene–ethanol. It weighed 1.5 g., ethanol. It weighed 1.5 g., 0.00357 mole, 35.7% yield, and had m.p. 292–295°.

Action of Hydrobromic Acid on III.—To a solution of acetic acid (20 ml.) and aqueous hydrobromic acid (10 ml., 48%) was added III (0.55 g., 0.0012 mole). The stirred solution was refluxed; after 45 min. a precipitate formed in the red reaction mixture. Following an additional 15 min. of refluxing, the mixture was cooled to room temperature and the solid was removed by filtration and recrystallized from glacial acetic acid. It weighed 0.3 g., 0.0007 mole, 58.3% yield, and had m.p. 292–295°. The infrared spectrum of this material (XI) was identical with that of the compound produced by the action of aluminum chloride on III. The compound sublimed at 240°/0.5 mm., but this did not give a pure product, for the impurities sublimed also. No satisfactory method of purification was found for this substance.

1,4-Di(2,4,5-trimethylphenyl)but-2-yne-1,4-diol (XII).—A Grignard solution was prepared from magnesium (12 g., 0.5 g.-atom), ethyl bromide (80 g., 0.75 mole) in ether (100 cc.). Acetylene was bubbled into this solution for 12 hr., and then durylicaldehyde[11] (75 g., 0.5 mole) in ether (100 cc.) was added over the course of 1 hr. The solution was refluxed for 1.5 hr., then poured over iced sulfuric acid (500 cc., 5%), and the gray solid was removed and washed with ether. The solid was crystallized from ethanol; it then weighed 20.0 g. (0.08 mole, 32%) and melted at 214–216°.

Anal. Calcd. for $C_{22}H_{26}O_2$: C, 81.95; H, 8.13. Found: C, 81.70; H, 8.35.

1,4-Di(2,4,5-trimethylphenyl)-1,3-butadiene (XIII).—Lithium aluminum hydride (1 g., 0.044 mole) in ether (100 cc.) was added to stirred suspension of XII (2.6 g., 0.008 mole) in ether (75 cc.) at 0° and under nitrogen. The mixture was warmed slowly to room temperature; at 15° the reaction began. The mixture was then refluxed for 2 hr., cooled to 0°, and ethyl acetate (25 cc.), water (25 cc.), and sulfuric acid (20 cc., 10%) were added consecutively. More ethyl acetate (50 cc.) was added, and the organic layer was removed and dried (sodium sulfate). The solution was concentrated and cooled; the solid was removed and recrystallized from ethyl acetate. It weighed 1.0 g., 0.0034 mole, 42.5%, and melted at 199–200°,

Anal. Calcd. for $C_{22}H_{26}$: C, 91.03; H, 8.97. Found: C, 90.46; H, 9.24.

(11) R. R. Holmes, Doctoral dissertation, University of Minnesota, 1950, pp. 77, 78.

Synthesis of Specifically Iodine-131- and Carbon-14-Labeled Thyroxine[1]

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A simple synthesis of various forms of radioactive L-thyroxine, carrying the label (I^{131} or C^{14}) either in the phenolic ring or in the nonphenolic ring and in the side chain, is described. It is based on the coupling of 4-hydroxy-3,5-diiodophenylpyruvic acid and 3,5-diiodotyrosine. Labeled 4-hydroxy-3,5-diiodophenylpyruvic acid was prepared by iodination of *p*-hydroxybenzaldehyde followed by condensation with acetylglycine and hydrolysis of the azlactone formed, or by enzymic oxidative deamination of diiodotyrosine.

The only commercially available radioactive thyroxine is the one in which the iodine atoms of the phenolic ring are labeled. Syntheses of other forms of radioactive thyroxine in which various other atoms carry the label, have been reported.[3–9] Most of these syntheses are either tedious or give low yields. This seems to be the only reason for the nonavailability of such other radioactive forms, which would be extremely valuable for metabolic or other studies. The major pathway

(1) A preliminary report of this work was given at the 140th National Meeting of the American Chemical Society, Chicago, Ill., September, 1961.

(2) Visiting Scientist from the Department of Chemistry, Faculty of Science, Osaka University, Osaka, Japan.

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(8) R. Michel, R. Truchot, H. Tron-Loisel, and B. Poillot, *Compt. rend.*, **250**, 2632 (1960).

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