Reaction Products of Tocopherols[1]

W.A. SKINNER and **R.M. PARKHURST**, Life Sciences Research, Stanford Research Institute, Menlo Park, California 94025

ABSTRACT

Tocopherols readily undergo oxidation with a variety of oxidizing agents. Considerable effort has gone into isolation and identification of these various oxidation products. In many cases they can undergo further transformations upon treatment with various chemical reagents. This review will focus on the oxidation of α-tocopherol and transformation of its oxidation products to new derivatives. Dimeric and trimeric oxidation products will not be covered. Early work on the oxidation of α-tocopherol led to identification of α-tocopherol quinone as an oxidation product formed by $FeCl_3$ oxidation. Stronger oxidizing conditions with $FeCl_3$ or oxidation with $AgNO_3$ or HNO_3 led to the orthoquinone and the hydroxy-*p*-quinone due to loss of one or two methyl groups from the aromatic ring. These early studies pointed out the unusual reactivity of the 5-methyl group of α-tocopherol. Oxidation of α-tocopherol with benzoyl peroxide led to substitution of a benzoate on the 5-methyl group. A similar reaction occurs when diasobisisobutyronitrile is used as the oxidizing agent. The oxidation of α-tocopherol by tetrachloro-*o*-quinone in aqueous acetonitrile resulted in the formation of 9-hydroxy-α-tocopherone. When $FeCl_3$ was used as the oxidizing agent in the presence of α,α'-bipyridyl in ethanol, 9-ethoxy-α-tocopherone was formed. α-Tocopherolquinone can be reduced with Zn-HOAc or by catalytic hydrogenation to the hydroquinone or reductively cyclized to α-tocopherol with Zn-HBr. Reaction of α-tocopherolquinone with acetyl chloride resulted in the 5-chloromethyl-6-acetoxy derivative which has been converted to a variety of 5-methylsubstituted derivatives. Reaction of α-tocopherol with Br_2 led to the 5-bromomethyl derivative. When α-tocopherolquinone was treated with hydrochloric, phosphoric, citric or tartaric acid, in the absence of oxygen, a disproportionation took place forming α-tocopherol, α-tocored and other oxidation products. An interesting isomerization of α-tocored, the orthoquinone, occurs in the presence of aqueous HCl to yield the yellow *p*-quinone with the chroman ring closed. The 5-benzoyloxymethyl derivative upon treatment with HCl generates *o*-quinone methide which can be trapped by reaction with tetracyanoethylene or dihydropyran. Treatment of the 5-benzoyloxmethyl derivative with HCl in ethanol followed by sublimation yielded the 5-aldehyde of α-tocopherol. Recently, a series of phosphate derivatives of α-tocopherol or its model, 2,2,5,7,8-pentamethyl-6-chromanol, were synthesized. Tris(6-acetoxy-5-methyleneoxy-7,8-dimethyltocol)phosphate, tris(2,2,5,7,8-pentamethyl-6-chromanol)phosphate, 5-hydroxymethyl-2,2,7,8-tetramethyl-6-chromanol phosphate and the cyclic 5-methylenoxy-2,2,7,8-tetramethyl-6-chromanol phosphate, were prepared. These phosphates are of interest in view of a possible role of α-tocopherol in oxidative phosphorylation.

INTRODUCTION

Investigation into the chemistry of vitamin E started in 1927 with Evans and Burr (1) when they found that its biological activity was destroyed by bromination and not by hydrogenation. Evans et al. (2) later isolated α-, β- and γ-tocopherols from wheat germ oil in the form of solid allophonates. The chemical identification of α-tocopherol resulted from studies on its decomposition and oxidation products by Fernholz (3,4). Synthesis of the biologically active principle was performed by Karrer et al. (5) from trimethylhydroquinone and phytyl bromide. John, Smith et al. (6,7) confirmed the chroman nucleus of vitamin E. In due time, the structures of β and γ were ascertained and they were synthesized. Other tocopherols (5,7-dimethyltocol; 8-methyltocol; 7-methyltocol, 5-methyltocol and tocol) have been isolated from natural sources or synthesized since then. These all possess the saturated phytyl side chain. In addition, several tocopherols with unsaturation in the side chain have been isolated and synthesized.

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OXIDATION REACTIONS OF THE TOCOPHEROLS

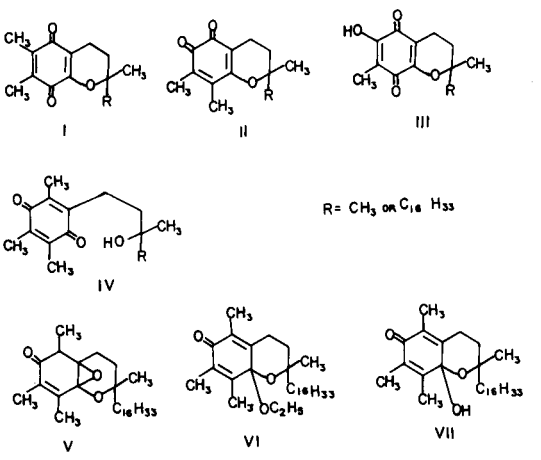
The most common reaction which the tocopherols undergo is one of oxidation. Interest in studying the oxidation reactions of the tocopherols are related to their use as anitoxidants and to interest in their biological role. Dimeric and trimeric oxidation products are excluded from this review.

In 1939, John and Emte (8) found that oxidation of α- or β-tocopherol with nitric acid in alcoholic solution gave an intense red coloration to the solution. Also silver nitrate oxidation in boiling alcohol yielded this red product. This reaction formed the basis of the Furter-Meyer (9) colorimetric method for analysis of tocopherols. Emmerie and Engel (10) have developed a colorimetric method for tocopherols based on oxidation with ferric chloride. John used the model compound 2,2,5,7,8-pentamethyl-6-chromanol, to study the oxidation reaction with silver nitrate. The red product obtained from the model was named chromanred 109 and that from α-tocopherol, α-tocored. Structure I was proposed for the red oxidation product by John.

This structure was later shown to be in error by Smith et al. (11) who showed that the red oxidation product, α-tocored, possessed Structure II. Compounds of Structure I could be formed from the red, orthoquinones, by treatment with hydrochloric acid. This latter isomerization was first noted by John and Emte (12).

In addition to this isomeric *p*-quinone, John and Emte (12) also isolated a hydroxy-*p*-quinone upon oxidation of α-tocopherol with silver nitrate in boiling ethanol. Structure III was assigned by John on the basis of elementary analysis of the product and its indicator properties (purple in base and yellow in acid). It was shown by Frampton et al. (13) that this hydroxy quinone (tocopurple) could also be produced by ferric chloride oxidation or by treatment of tocored with hydrochloric acid. These workers also showed that tocored could be produced by oxidation of tocopherol with ferric chloride in methanol at 50 C. Structure III for tocopurple was proved unequivocally in 1960 by Frampton et al. (14).

These oxidations under relatively mild conditions resulting in elimination of aromatic methyl groups are interesting and unusual in the field of organic chemistry. The extreme reactivity of the 5-methyl group of α-tocopherol and its model compounds becomes obvious as one studies their various reactions.

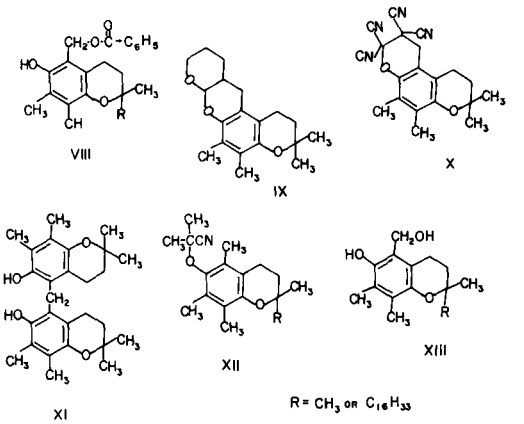


Mild oxidation of α-tocopherol with ferric chloride or silver nitrate yields α-tocopherolquinone (IV) (15). Boyer (16) isolated a product formed by oxidation with ferric chloride which was converted by acid to α-tocopherolquinone. Structure V was postulated for this product called α-tocopheroxide. Martius and Eilingsfeld (17) showed this structure to be in error with the correct structure as VI. More recently, Durckheimer and Cohn (18) studied the oxidation of α-tocopherol with tetrachloro-*o*-quinone or N-bromosuccinimide and prepared 9-hydroxy-α-tocopherone (VII). In the presence of alcohols, compounds of type VI were obtained. Deviation from a pH of 5.4 in either direction resulted in decomposition of VII to form α-tocopherol quinone.

Free radical initiated oxidation of α-tocopherol and its model compounds has led to interesting information about the mechanism of these oxidations and to new products. Inglett and Mattill (19) studied the oxidation of α- and γ-tocopherol and 2,2,5,7,8-pentamethyl-6-chromanol with benzoyl peroxide at 30 C. A surprisingly rapid oxidation of α-tocopherol occurs at this temperature yielding α-tocopherolquinone and a compound identified as VIII. γ-Tocopherol yielded the red, *ortho*-quinone. Goodhue and Risley (20) studied the reaction of *d*-α-tocopherol in hydrocarbon solvents with benzoyl peroxide. They obtained compound VIII which upon treatment with aqueous KOH yielded spirodienone dimer. These same workers (21) found that benzoyl peroxide oxidation of *d*-α-tocopherol in the presence of alcohols led to formation of 8a-alkoxy-α-tocopherones. Skinner and Parkhurst (22) confirmed the structure of the benzoyl peroxide oxidation product as VIII. Decomposition of the 5-benzoate with either hydrochloric acid in benzene or KOH in ethanol

yielded dimer and trimer. The ease of conversion of VIII to the intermediate quinone methide was used to prepare various tricyclic derivatives from dienophiles (IX and X). Treatment of the benzoate (VIII) with hydrochloric acid in ethanol followed by sublimation yielded the 5-aldehyde. Reduction of VIII with zinc-acetic acid gave the starting chroman. Refluxing VIII in aqueous hydrochloric acid in ethanol for several days under nitrogen gave a new product identified as dimer (XI).

Oxidation of 2,2,5,7,8-pentamethyl-6-chromanol with azobisisobutyronitrile, another free radical initiator, yielded a dihydroxy dimeric product (23) and a coupling product from the initiator and the phenoxy radical (24) (XII).



REDUCTION PRODUCTS

Treatment of α-tocopherolquinone with zinc and acetic acid causes reduction to the hydroquinone, which is very sensitive to air oxidation, and some cyclization to α-tocopherol (15). This reductive cyclization can also be obtained using zinc-hydrochloric acid. Catalytic hydrogenation of α-tocopherolquinone yields the hydroquinone.

HALOGENATION REACTIONS

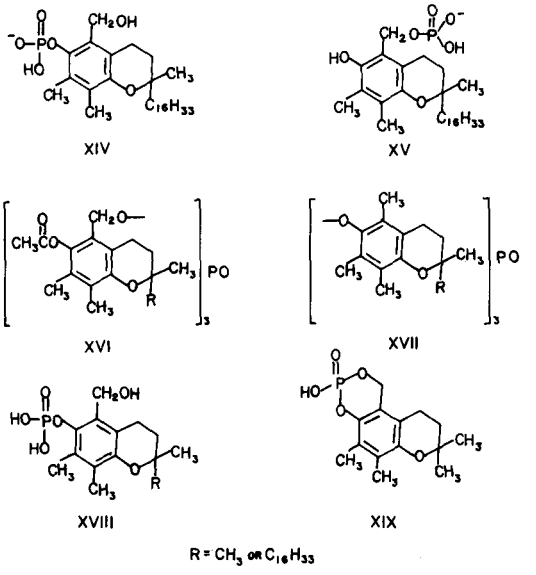
Bromination (21) of α-tocopherol led to the 5-bromomethyl derivative which was converted to the spirodienone dimer when treated with 1 N KOH. Treatment of α-tocopherol with acetyl chloride in benzene yielded the 5-chloromethyl-6-acetoxy derivative which has been converted to a variety of 5-methyl substituted derivatives of α-tocopherol (25).

ALKYLATION OF TOCOPHEROLS

Interest in conversion of γ-tocopherol to α-tocopherol has led to the study of alkylation of γ-tocopherol. Weisler (26) has hydroxymethylated γ-tocopherol using formalin and converted the crude 5-hydroxymethyl derivative to α-tocopherol by zinc-hydrochloric acid reduction. Haloalkylation of γ-tocopherol was described by Weisler and Chechak (27). Formylation of γ-tocopherol is covered by Weisler in other patents (28). Aminoalkylation (29) is covered by another patent (29). Recently, we prepared 2,2,7,8-tetramethyl-5-hydroxymethyl-6-chromanol (XIII) in analytically pure form as well as the corresponding α-tocopherol derivative.

ACID CATALYZED DISPROPORTIONATION REACTIONS

An interesting reaction of α-tocopherolquinone was discovered by Issidorides (30). She found that a disproportionation occurred when the quinone was treated with certain acids (phosphoric, citric or tartaric). Both α-tocopherol and α-tocored were formed from α-tocopherolquinone in this disproportionation which occurred in the absence of oxygen. These results again point to the unusual reactivity of the 5-methyl group of α-tocopherol.



PHOSPHATE DERIVATIVES

Phosphate derivatives of hydroquinones and chromans have been proposed as important intermediates in biological oxidative phosphorylation processes (31). Intermediates such as XIV and XV were proposed by Vilkas and Lederer (32).

In our laboratory we have synthesized a number of phosphate derivatives of α-tocopherol and its model; 2,2,5,7,8-pentamethyl-6-

chromanol. These are compounds XVI-XIX. The latter two compounds were synthesized from the 5-benzoyloxymethyl-6-hydroxychroman by treatment with phosphorus oxychloride followed by hydrolysis and by cyclization with dicyclohexylcarbodiimide.

EXPERIMENTAL PROCEDURES

2,2,7,8-Tetramethyl-5-hydroxymethyl-6-chromanol

2,2,7,8-Tetramethyl-6-chromanol (164.0 mg) was placed in a glass vial with 1 ml of water and 0.2 ml of 37% formalin. Nitrogen gas was bubbled through the mixture for 15 min and 25 mg of calcium hydroxide was added while the bubbling was continued and the glass vial was sealed with a flame. The vial was placed in a shaker and left over the weekend. The vial which now contained a thick slurry was acidified with acetic acid, extracted with ether and the ether washed with sodium bicarbonate solution and dried with sodium sulfate. Chromatography of the crude product on a thick layer silica gel plate with chloroform gave a band near the origin which was removed and eluted with ether. The product crystallized from ether-light petroleum ether mixture to give 60 mg of a white solid; mp 110-112 C.

Analysis calculated for $C_{14}H_{20}O_3$: C, 71.16; H, 8.53; Found: C, 71.35; H, 8.60.

5-Hydroxymethyl-γ-tocopherol

The procedure reported for the model compound was repeated exactly on 333 mg of γ-tocopherol. A colorless gum (52 mg) was recovered from chromatography.

Analysis calculated for $C_{29}H_{50}O_3$: C, 78.15; H, 11.08; Found: C, 78.25; H, 11.34.

2,2,7,8-Tetramethyl-5-bromomethyl-6-chromanol

Ten grams of model chromanol was dissolved in light petroleum ether and 9.2 g of bromine was added in light petroleum ether. After standing at room temperature for 0.5 hr, the solvent was removed in vacuo and the solid material remaining was recrystallized from light petroleum ether which gave a nearly quantitative yield of cream colored needles, mp 73-75 C.

Analysis calculated for $C_{14}H_{19}O_2Br$: C, 56.20; H, 6.40; Br, 26.71; Found: C, 56.16; H, 6.45; Br, 26.62.

Sodium methoxide converts this compound to the 5-methoxymethyl derivative (33).

Analysis calculated for $C_{15}H_{22}O_3$: C, 71.96; H, 8.86. Found: C, 71.77; H, 8.91.

Tris(2,2,5,7,8-pentamethyl-6-chromanol)phosphate

2,2,5,7,8-Pentamethyl-6-chromanol (2.2 g) was dissolved in hexane and 0.24 g of sodium hydride was added while stirring under nitrogen. After 1/2 hr a slight excess of $POCl_3$ was slowly added to the stirring slurry. The sodium chloride was filtered and the filtrate evaporated to a small volume, washed with sodium bicarbonate, dried with anhydrous sodium carbonate and ether added. Evaporation of the ether gave a white precipitate which was recrystallized from hexane-ether mixture to give 2 g of a white solid, mp, 174-175 C.

Analysis calculated for $C_{42}H_{57}O_7P$: C, 71.57; H, 8.15; Found: C, 71.57; H, 7.94.

Tris(6-acetoxy-5-methylenoxy-7,8-dimethyltocol)phosphate

5-Chloromethyl-γ-tocopherol acetate (6.08 g) and 1.67 g of yellow, silver phosphate were stirred in refluxing diglyme for 6 hr under a nitrogen atmosphere. During this time the yellow color of the silver phosphate changed to a grey-black. The solution was decanted into ice water and extracted with ether. The ether was removed and the brown product chromatographed on a silica gel column using light petroleum ether-ether mixtures. A center cut was rechromatographed on florisil and finally on Silica gel GF-254 (according to E. Stahl) thick layer plates with chloroform, giving a very viscous and almost colorless oil (in about 10% yield) which showed only one spot on thin layer chromatography; Silica gel GF-254 (according to Stahl)-$CHCl_3$, R_f = 10-23. $\gamma^{(\mu)}_{max}$ = 5.65 (CO) 6.30, 7.50, 7.95, 8.34, 8.96, 9.20, 10.15, 13.55 and 14.45.

Analysis calculated for $C_{93}H_{153}O_{13}P$: C, 73.97; H, 10.21; Found: C, 74.14; H, 10.00.

5-Hydroxymethyl-2,2,7,8-tetramethyl-6-chromanol phosphate

2,2,7,8-Tetramethyl-5-benzoyloxy-6-chromanol (2 g) was dissolved in dry pyridine (20 ml) and 2.4 ml of $POCl_3$ in 20 ml of dry pyridine was added to the ice cold, stirring mixture. The ice bath was removed and the mixture was allowed to stir under N_2 for 36 hr. The solution was poured into water, acidified and extracted with ether. The ether was washed with 6 N HCl, water and dried over sodium sulfate. Upon evaporation in vacuo a gum was isolated. This gum was dissolved in alcohol and NaOH (aqueous) added. The precipitate was filtered and dissolved in water and HCl added, extracted with ether and evaporated in vacuo. The white solid obtained was recrystallized from ethyl acetate-ether-petroleum ether to yield a solid; mp 145-151 C. NMR in pyridine and D_2O was consistent with the above structure as was the IR spectrum. The yield was 705 mg (38%, theory). A titration of this material

gave an equivalent weight of 164 or a molecular weight of 328 (calculated, 316.3).

The analysis was calculated for $C_{14}H_{21}O_6P$: C, 53.16; H, 6.69; P, 10.03; Found: C, 53.25; H, 6.60; P, 9.70.

Cyclic 5-methylenoxy-2,2,7,8-tetramethyl-6-chromanol phosphate

The 5-hydroxymethyl-2,2,7,8-tetramethyl-6-chromanol phosphate (250 mg) was added to 5 ml of pyridine and stirred until dissolved. One milliliter of water and 1 g of dicyclohexylcarbodiimide was added and the mixture stirred over the weekend. Water was added and the dicyclohexylurea filtered off. The filtrate was extracted with ether three times and then made acidic with HCl. Washing with water, drying with sodium sulfate and evaporation in vacuo gave a glassy gum which crystallized on treatment with light petroleum ether after refrigerating for one week. Recrystallization from ethanol-ethyl acetate gave 78.6 mg of a white solid; mp 230-233 C (dec.).

The analysis was calculated for $C_{14}H_{19}O_5P$; C, 56.45; H, 6.44; P, 10.38; Found: C, 55.90; H, 6.29; P, 10.22.

The molecular weight (in EtOH) by osmometer was 284 (calculated, 298).

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