di - acetyl - mono - triphenylmethyl - β - methyl-*d*-xylosides. These results show that triphenylmethyl chloride reacts with the secondary hydroxyl groups in the β-methyl-*d*-xyloside molecule.

WASHINGTON, D. C. RECEIVED DECEMBER 28, 1933

[CONTRIBUTION FROM THE LABORATORY FOR PURE RESEARCH OF MERCK & CO., INC.]

An Investigation of Certain Derivatives of Alkyl γ-Oxalylcrotonates

BY ALBERT B. BOESE, JR., AND RANDOLPH T. MAJOR

In connection with certain work which was being done in this Laboratory, an attempt was made to synthesize ethyl α-oxalyl-vinylacetate, $CH_2{=}CHCH(COCOOC_2H_5)COOC_2H_5$, by the condensation of ethyl vinylacetate and ethyl oxalate in the presence of sodium. The substances reacted readily, hydrogen was evolved and a yellow sodium derivative was formed. However, the free ester which was obtained when an aqueous solution of the sodium derivative was acidified, was not ethyl vinyl-α-oxalylacetate, but was, instead, ethyl γ-oxalylcrotonate, $C_2H_5OOCC(OH){=}CHCH{=}CHCOOC_2H_5$, which previously had been prepared by Prager,[1] by the condensation of ethyl crotonate and ethyl oxalate. Apparently, under the conditions of the reaction, ethyl vinylacetate rearranged to ethyl crotonate which then reacted normally to give ethyl γ-oxalylcrotonate.[2]

$$CH_2{=}CHCH_2COOC_2H_5 \longrightarrow CH_3CH{=}CHCOOC_2H_5$$

$$CH_3CH{=}CHCOOC_2H_5 + (COOC_2H_5)_2 + Na \longrightarrow C_2H_5OOCC(ONa){=}CHCH{=}COOC_2H_5 + {}^1/_2H_2 + C_2H_5OH$$

As comparatively little work has been done on ethyl γ-oxalylcrotonate, it was thought desirable to make a further study of this interesting compound, particularly of its O-acyl derivatives. Lapworth[3] prepared γ-oxalylcrotonic acid, $HOOCCH{=}CHCH_2COCOOH$ by careful alkaline hydrolysis of the sodium derivative of ethyl γ-oxalylcrotonate. This compound was described as a bright yellow microcrystalline substance which melted with decomposition at about 190°. It was regarded by Lapworth as probably a mixture of tautomeric forms. The ethyl γ-oxalylcrotonates which we obtained by the condensation between either ethyl vinylacetate or ethyl crotonate, with ethyl oxalate were hydrolyzed by a slight modification of the process used by Lapworth. γ-Oxalylcrotonic acid was obtained in the form of a colorless, crystalline compound which melted sharply with decomposition at 212°. When it was oxidized with 30% hydrogen peroxide according to the method of Holleman,[4] it reacted as a normal α-ketonic acid, and glutaconic acid was formed in good yield.

$$HOOCCH{=}CHCH_2COCOOH + H_2O_2 \longrightarrow HOOCCH{=}CHCH_2COOH + H_2O + CO_2$$

Although it has long been generally accepted that ethyl oxalylcrotonate and its sodium salt have the enol structure,[1,3] until recently no O-acyl derivatives of this substance have been prepared. Börsche and Manteuffel[5] prepared ethyl O - acetyl - γ - oxalylcrotonate, $C_2H_5OOCC(OCOCH_3){=}CHCH{=}CHCOOC_2H_5$, by the action of acetic anhydride on the potassium salt of ethyl γ-oxalylcrotonate. They showed that this compound could be reduced catalytically in the presence of colloidal palladium to diethyl α-acetoxyadipate, $C_2H_5OOC(OCOCH_3)CH_2CH_2CH_2 - COOC_2H_5$. We have prepared ethyl O-carbethoxy-γ-oxalylcrotonate, $C_2H_5OOCC(OCOOC_2H_5){=}CHCH{=}CHCOOC_2H_5$, by the action of ethyl chlorocarbonate on the sodium derivative of ethyl γ-oxalylcrotonate.

In a similar manner, methyl-O-carbomethoxy-γ-oxalylcrotonate, $CH_3OOCC(OCOCH_3){=}CHCH{=}CH{-}COOCH_3$, was prepared from methyl chlorocarbonate and methyl-γ-oxalylcrotonate, which in turn had been made in the normal manner by the condensation of methyl crotonate with methyl oxalate. The alkyl O-carbalkoxy-γ-oxalylcrotonates prepared were reduced catalytically at room temperature in a solution of methyl alcohol in the presence of a platinum catalyst.[6] Approximately two and a half moles of hydrogen were absorbed and the principal product obtained was dialkyl adipate. Possibly the formation of car-

(1) Prager, *Ann.*, **338**, 375 (1905).

(2) Another mechanism for this reaction is suggested by the recent work of Borsche and Manteuffel, *Ann.*, **505**, 179 (1933).

(3) Lapworth, *J. Chem. Soc.*, **79**, 1279 (1901).

(4) Holleman, *Rec. trav. chim.*, **23**, 169 (1904).

(5) Börsche and Manteuffel, *Ber.*, **65**, 871 (1932).

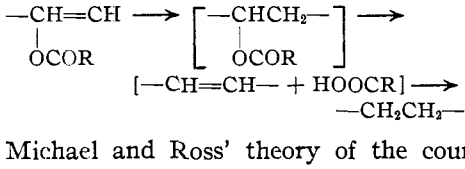
(6) Adams and Voorhees, "Organic Syntheses," John Wiley and Sons, Inc., New York, 1928, Vol. VIII, p. 10.

bon dioxide from the decomposition of the acid carbonic ester made the observed amount of hydrogen absorbed less than three molecules.

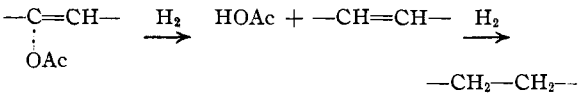
Ethyl O-carbethoxyoxalylacetate, $C_2H_5OOCC(OCOOC_2H_5)=CHCOOC_2H_5$, was prepared from the sodium salt of ethyl oxalylacetate and ethyl chlorocarbonate. When this compound was catalytically reduced, the reaction followed a course similar to that described above. One and one-half moles of hydrogen were absorbed; the product isolated was diethyl succinate.

In the light of these results, it appeared that with a platinum catalyst ethyl O-acetyl-γ-oxalylcrotonate should be reduced to diethyl adipate. This was found to be the case. Three moles of hydrogen were absorbed and the product isolated was diethyl adipate. Moreover, we were unable to isolate any intermediate product in this reduction. When the reaction was stopped after one and one-half moles of hydrogen had been absorbed, the reaction mixture was found to contain the starting material and diethyl adipate. Had the course of the reaction involved two separate steps; first, the hydrogenation of the ethylenic linkages followed by the hydrogenolysis of the O-acyl group, it should have been possible to isolate diethyl γ-acetoxyadipate as the intermediate product. We have also found that diethyl α-acetoxyadipate is not further reduced by hydrogen in the presence of a platinum catalyst.

Roll and Adams[7] and Michael and Ross[8] have recently shown that when certain O-acyl enolic compounds containing the grouping —C(OAc)=CH— are reduced, the acyloxy group is removed and compounds with the group —CH_2—CH_2— are obtained. Roll and Adams suggested that the mechanism of the reduction involved first the addition of hydrogen to the double linkage, followed by decomposition of the saturated O-acyl compound, forming another double bond which was in turn reduced.



Michael and Ross' theory of the course of the reaction was very similar. However, since we found that when the unsaturated linkages in the compounds which we have studied were fully reduced, no further reduction took place, it appears more likely that the addition of hydrogen takes place first at the O-acyl linkage or at least simultaneously with the reduction of the double bonds, as follows



Experimental Part

Alkyl γ-Oxalylcrotonate.—To 4 g. of sodium wire in 500-cc. of dry ether contained in a flask with a dropping funnel and reflux condenser, was added 0.19 mole of dialkyl oxalate. To this mixture was slowly added 20 g. of ethyl vinylacetate. It took from fifteen to thirty minutes for the reaction to commence after the first few cc. of ethyl vinylacetate had been added. The remainder was then introduced at such a rate as to maintain a gentle refluxing of the ether. During the reaction the sodium dissolved with evolution of hydrogen and the solution became deep red in color. When the reaction was complete, the solution was cooled and the yellow sodium derivative precipitated. This was removed by filtration, washed with petroleum ether and dried *in vacuo* over calcium chloride. Evaporation left a pale yellow microcrystalline compound. This was dissolved in 500 cc. of water and the solution made acid with dilute sulfuric acid; the compound which separated was collected on a filter, washed with cold water and dried *in vacuo* over sulfuric acid. It was crystallized from a mixture of ethyl acetate and petroleum ether. Small colorless prisms were obtained; yield 60%. The material obtained when diethyl oxalate was used melted at 78–80°. When mixed with an authentic sample of ethyl γ-oxalylcrotonate, the melting point remained unchanged.[9]

Anal. Calcd. for $C_{10}H_{14}O_5$: C, 56.05; H, 6.63. Found: C, 56.08; H, 6.54.

The material obtained when dimethyl oxalate was used melted at 124–126°.

Anal. Calcd. for $C_8H_{10}O_5$: C, 51.61; H, 5.37. Found: C, 51.52; H, 5.33.

γ-Oxalylcrotonic Acid.—Nineteen grams of the sodium derivative of ethyl γ-oxalylcrotonate, which had been made from ethyl vinylacetate and ethyl oxalate as previously described, was triturated in a mortar with 200 cc. of cold 30% solution of sodium hydroxide. The sodium hydroxide was added in small quantities and the mixture kept cool. The solution which resulted was allowed to stand for several hours at room temperature until a small sample, when diluted, gave no precipitate with acetic acid. The solution was then diluted with water to 700 cc., filtered, cooled and made acid to congo red with dilute sulfuric acid. After standing a short time, 11.7 g. of crude oxalylcrotonic acid separated. This was removed by filtration, washed with a small amount of water and dried When it was recrystallized from ethyl acetate, it occurred as almost colorless small needles which melted at 212°.

Anal. Calcd. for $C_6H_6O_5$: C, 45.57; H, 3.80. Found: C, 45.71; H, 4.00.

(7) Roll and Adams, THIS JOURNAL, **53**, 3470 (1931).

(8) Michael and Ross, *ibid.*, **54**, 392 (1932).

(9) Prager, *Ann.*, **338**, 375 (1905), gives 78–80° as the melting point of ethyl γ-oxalylcrotonate.

Oxidation of γ-Oxalylcrotonic Acid.—Ten grams of γ-oxalylcrotonic acid was suspended in 50 cc. of water and to the suspension was added 6.8 cc. of 30% hydrogen peroxide. The suspension was heated at 60° with occasional shaking. During the reaction, carbon dioxide was evolved and the suspension became clear. The solution was then treated with decolorizing carbon, filtered and evaporated to dryness *in vacuo* at 40°. A yellow crystalline mass remained which was repeatedly extracted with ether in a Soxhlet extractor. When petroleum ether was added to the ether extract, glutaconic acid was precipitated which after recrystallization from ethyl acetate melted at 137°; yield, 5.3 g. When this compound was mixed with an authentic sample of glutaconic acid, no depression of the melting point was observed.[10]

Anal. Calcd. for $C_5H_6O_4$: C, 46.15; H, 4.61. Found: C, 46.17; H, 4.73.

Alkyl O-Carbalkoxy-γ-oxalylcrotonate.—One-twentieth of a mole of the sodium derivative of alkyl γ-oxalylcrotonate was suspended in 250 cc. of dry ether contained in a flask fitted with a dropping funnel and reflux condenser. To the suspension was slowly added 5.5 g. of the corresponding alkyl chlorocarbonate. Gentle ebullition of the ether occurred and when the addition of the alkyl chlorocarbonate was complete, the mixture was refluxed for two hours. During the reaction sodium chloride was formed and the mixture became quite dark. The reaction mixture was poured into water and the ether layer was washed twice with water and twice with a dilute solution of sodium carbonate, then dried over anhydrous sodium sulfate. The ether was removed by distillation and the remaining oil was distilled under reduced pressure. The alkyl O-carbalkoxy-γ-oxalylcrotonates which were obtained were very viscous, sirupy straw-colored liquids, insoluble in water but soluble in all common organic solvents. They gave no color reaction with ferric chloride.

TABLE I

ANALYTICAL AND OTHER DATA OF ALKYL O-CARBALKOXY-γ-OXALYLCROTONATES

Alkyl groups	B. p., °C.	mm.	Yield, %	Carbon, % Calcd.	Carbon, % Found	Hydrogen, % Calcd.	Hydrogen, % Found
Methyl	160–163	6	71	49.16	49.08	4.96	5.05
Ethyl	158–160	3	68	54.52	54.32	6.32	6.22

Reduction of Alkyl O-Carbalkoxy-γ-oxalylcrotonates.—Ten grams of alkyl O-carbalkoxy-γ-oxalylcrotonate in 30 cc. of methyl alcohol was catalytically reduced by hydrogen under a pressure of 1–3 atmospheres in an Adams reduction apparatus, in the presence of 0.2 g. of a platinum catalyst.[11] Two and one-half moles of hydrogen were absorbed in two hours. When no more hydrogen was absorbed, the catalyst was removed by filtration and the filtrate was poured into 500 cc. of water. The aqueous mixture was extracted with ether, the ether extract washed with water and then dried over anhydrous sodium sulfate. The ether was distilled off and the remaining liquid distilled under reduced pressure.

Reduction of methyl O-carbomethoxy-γ-oxalylcrotonate gave dimethyl adipate, b. p. (10 mm.) 105°; yield, 70%.

Anal. Calcd. for $C_8H_{14}O_4$: C, 55.17; H, 8.04. Found: C, 55.12; H, 7.87.

Reduction of ethyl O-carbethoxy-γ-oxalylcrotonate gave diethyl adipate, b. p. (3 mm.) 108–110°.

Anal. Calcd. for $C_9H_{16}O_4$: C, 57.44; H, 8.51. Found: C, 57.25; H, 8.58.

Hydrolysis of this ethyl adipate by concentrated potassium hydroxide gave adipic acid, m. p. 153°.[12]

Anal. Calcd. for $C_6H_{10}O_4$: C, 49.31; H, 6.84. Found: C, 49.26; H, 6.60.

Ethyl O-Carbethoxy-oxalylacetate.—Ethyl O-carbethoxyoxalylacetate was obtained by the condensation of the sodium derivative of ethyl oxalylacetate[13] with ethyl chlorocarbonate by the same process that was used in the preparation of alkyl O-carbalkoxy-γ-oxalylcrotonates; yield, 65%. It was a colorless liquid of b. p. (4 mm.) 143–146°, insoluble in water or aqueous sodium hydroxide; it gave no color reaction with ferric chloride.

Anal. Calcd. for $C_{11}H_{16}O_7$: C, 50.75; H, 6.19. Found: C, 50.77; H, 6.11.

Reduction of Ethyl O-Carbethoxy-oxalylacetate.—A solution of 12 g. of ethyl O-carbethoxy-oxalylacetate in 20 cc. of methyl alcohol was reduced catalytically in the manner previously described. One and one-half moles of hydrogen were absorbed in one hour. The reduction product was treated as described above. Eight grams of diethyl succinate, which boiled at 216–218°,[14] was obtained. The product was identified as diethyl succinate by its boiling point and its hydrolysis product. When it was hydrolyzed with aqueous sodium hydroxide, succinic acid of m. p. 186° was obtained. When this acid was mixed with an authentic sample of succinic acid no depression of the melting point was observed.[15]

Reduction of Ethyl O-Acetyl-oxalylcrotonate.—Ten grams of ethyl O-acetyl-oxalylcrotonate (Ref. 5, p. 869) in 30 cc. of methyl alcohol was catalytically reduced as before. Three moles of hydrogen was absorbed in two hours. The reduction product was treated by the previously described methods. Diethyl adipate was isolated and identified by the methods above mentioned.

In another reduction under the same conditions, the reaction was stopped when approximately one and one-half moles of hydrogen had been absorbed. The reaction mixture was found to contain only two substances, diethyl adipate and the starting material, ethyl O-acetyl-oxalylcrotonate. No diethyl α-acetoxyadipate was obtained.

Attempted Reduction of Diethyl α-Acetoxyadipate.—Twelve grams of diethyl α-acetoxyadipate[16] was dissolved in 50 cc. of methyl alcohol and shaken with hydrogen in the presence of 0.3 g. of a platinum catalyst as previously described. No absorption of hydrogen was observed and the compound was recovered unchanged.

(10) Guthzeit and Bolam, *J. prakt. Chem.*, [2] **54**, 362 (1896), give 137–138° as the melting point of glutaconic acid.

(11) Adams and Shriner, THIS JOURNAL, **45**, 2171 (1923).

(12) Ciamician and Silber, *Ber.*, **29**, 485 (1896), give 153–153.5° as the melting point of adipic acid.

(13) Wislicenus, *Ann.*, **246**, 315 (1888).

(14) Perkin, *J. Chem. Soc.*, **45**, 515 (1884), gives 216.5° as the boiling point of diethyl succinate.

(15) Davidoff, *Ber.*, **19**, 407 (1886), gives 185° as the melting point of succinic acid.

(16) Staudinger and Ruzicka, *Helv. Chim. Acta*, **7**, 448 (1924).

Summary

1. Alkyl γ-oxalylcrotonates were prepared by the condensation of alkyl vinylacetate and alkyl oxalate in the presence of sodium.

2. γ-Oxalylcrotonic acid was prepared by the hydrolysis of the sodium derivative of ethyl γ-oxalylcrotonate. It was oxidized with 30% hydrogen peroxide to glutaconic acid.

3. Ethyl O-carbethoxy-γ-oxalylcrotonate, methyl O-carbomethoxy-γ-oxalylcrotonate and ethyl O-carbethoxy-γ-oxalylacetate were prepared by the action of ethyl chlorocarbonate on the corresponding sodium derivative. Diethyl adipate, dimethyl adipate and diethyl succinate, respectively, were formed by the reduction of these compounds.

4. Ethyl O-acetyl-γ-oxalylcrotonate was catalytically reduced to diethyl adipate.

5. The mechanism of these reductions has been discussed.

RAHWAY, N. J. RECEIVED DECEMBER 29, 1933

[CONTRIBUTION FROM THE DEPARTMENT OF AGRICULTURAL CHEMISTRY OF THE UNIVERSITY OF WISCONSIN]

The Chemistry of Mold Tissue. IV. The Lipids of Aspergillus Sydowi[1,2]

BY F. M. STRONG AND W. H. PETERSON

In comparison with the large number of papers on the production of organic acids and other compounds by the common molds, relatively few publications deal with the composition of the mycelium itself. In previous investigations[3] the lipid and sterol contents of a large number of molds have been determined, and one of the lipid constituents, ergosterol, has been isolated and identified. In the present study the lipids of one of these molds, *Aspergillus sydowi*, have been examined in detail. This mold was selected because it can be grown readily in large quantities,[3d] and also contains a large percentage of lipoidal material.

Previous investigations of the lipids of fungus tissue have been limited mainly to yeasts and to the higher fungi. Most of the work dealing with molds has been concerned with variations in the amount of fat produced under different conditions of growth.[4] With the exception of linoleic acid, which Takata[5] has shown to be present in *A. oryzae*, and of ergosterol, which has been found in various molds,[3b] apparently no rigorous characterization of the constituents of any mold fat has ever been made. Palmitic, oleic and other fatty acids have been reported by certain investigators,[6,7] but the evidence in most cases rests merely on melting points without supporting analytical data. Likewise in most cases the mold studied was not definitely identified. Various other authors have determined some of the fat constants on the lipids obtained from certain molds, without claiming to have identified any particular constituent of the fat.[8]

The method of extraction employed in this investigation was in general similar to that followed by Anderson in his extensive study of the lipids of tubercle and other acid fast bacilli.[9]

Experimental Part

Extraction of the Crude Lipids.—The mold used was grown in large sterilized incubators as previously described.[3d] When ready to harvest, it was killed by steaming, separated from the culture medium and dried for several days at 65°. The dry material was then finely ground.

Five kilograms of the ground mycelium was thoroughly extracted at room temperature with successive portions of alcohol–ether (1:1) during a period of two weeks.[10] The combined extracts were concentrated to one liter, dissolved in ether, washed with water, and the solvent removed under reduced pressure. The residual oil was a clear, deep red liquid weighing 607 g., or 12% of the mycelium extracted. It dissolved readily in alcohol, ether, chloroform and acetone at room temperature. Titration revealed the presence of about 22% of free acid, calculated as oleic. On standing, crystals of ergosterol

(1) This work was supported in part by a grant from the Wisconsin Alumni Research Foundation.

(2) Presented in part at the Chicago Meeting, American Chemical Society, September, 1933.

(3) Peterson and co-workers, (a) *J. Biol. Chem.*, **90**, 369 (1931); (b) *ibid.*, **97**, 483 (1932); (c) *Biochem. Z.*, **246**, 401 (1932); (d) *Ind. Eng. Chem.*, **25**, 213 (1933); (e) *Zentr. Bakt. Parasitenk.*, II Abt., **89**, 370 (1934).

(4) Terroine and co-workers, *Bull. soc. chim. biol.*, **9**, 12, 588, 604, 678 (1927); Behlin, *ibid.*, **8**, 1081, 1120 (1926); Bohn, *Compt. rend.*, **193**, 441 (1931); Pontillon, *ibid.*, **191**, 1148, 1367 (1930); Pearson and Raper, *Biochem. J.*, **21**, 875 (1927); Porges, *Botan. Gazz.*, **94**, 197 (1932).

(5) R. Takata, *J. Soc. Chem. Ind., Japan*, **32**, 171B (1929).

(6) H. H. Barber, *Biochem. J.*, **23**, 1158 (1929).

(7) M. X. Sullivan, *Science*, **38**, 678 (1913).

(8) Browne, THIS JOURNAL, **28**, 453 (1906); Sumi, *Biochem. Z.*, **195**, 161 (1928); Pontillon, *Compt. rend.*, **191**, 1148 (1930).

(9) Anderson, *Physiol. Rev.*, **12**, 166 (1932).

(10) The extraction and working up of all unsaturated lipids was carried out under atmospheres of carbon dioxide or nitrogen.