Synthesis of Monofluoro Analogs of Tri- and Dicarboxylic Acid Esters and of Fluorolactic Acid[1a]

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Procedures are described for the synthesis of triethylfluorooxalosuccinate, triethyl fluoroisocitrate (1-hydroxy-2-fluoro-1,2,3-propanetricarboxylate), diethyl α-oxo-β-fluoroglutarate, diethyl-α-fluoro-γ-hydroxyglutarate, diethyl-α-hydroxy-β-fluoroglutarate, and monofluorolactic acid.

As part of a systematic effort to obtain specific inhibitors of certain key enzymes strategically located at branching points of metabolic pathways, we have previously succeeded in preparing certain mono- and difluorocarboxylic acids.[2-7] These substances proved to be valuable tools in studying both enzymatic reaction mechanisms, as well as serving as enzyme reagents in complex biochemical systems.[8-11] For *in vitro* enzyme kinetic experiments it was necessary to obtain the crystalline free fluoro acids, but this goal was not always achieved. Although esters are more difficult to handle in aqueous *in vitro* enzyme systems, they may prove to be of interest in the course of *in vivo* experiments. With the exception of monofluorolactic acid, the enzyme substrate homologs described in this paper were obtained in pure form as their ethyl esters. Syntheses of other fluorocarboxylic acid esters were previously reviewed by Bergmann, *et al.*[12]

Triethyl fluorooxalosuccinate (I) was prepared from the potassium enolate of triethyl oxalosuccinate using perchloryl fluoride as the fluorinating reagent. When this reaction was carried out in ethanol at temperatures below 0°, good yields of the fluorinated ester were obtained. However, above 0° subsequent acyl cleavage of the fluoro ester was observed, leading to the formation of diethyl fluorosuccinate and diethyl oxalate as by-products. In the course of synthesis of monofluorosuccinate, Dean and Pattison[13] obtained triethyl fluorooxalosuccinate as an intermediate but did not isolate it.

Attempts to prepare the triamide of fluorooxalosuccinic acid by treating the ester with ammonia were unsuccessful and only fumaric diamide and oxalic diamide were obtained. Similarly, when diethyl fluorosuccinate was treated with aqueous ammonia, only fumaric diamide could be isolated. Similar results with diethyl fluorosuccinate have been reported.[14]

Attempts to hydrolyze triethyl fluorooxalosuccinate under basic conditions resulted in cleavage of the molecule, even when mildest conditions (*i.e.*, sodium bicarbonate–water) were employed. In every instance fumaric and oxalic acids were the products. The instability of esters of these types was also observed by Bergmann.[15] Treatment of triethyl fluorooxalosuccinate with 37% hydrochloric acid resulted in simultaneous hydrolysis and decarboxylation of one carboxyl group to yield α-oxo-β-fluoroglutaric acid as a sirup. Upon esterification the diethyl ester of this acid (III) was obtained in good yields. This fluoroketo acid ester reacted normally with 2,4-dinitrophenylhydrazine reagent to give the hydrazone. Crystalline α-oxo-β-fluoroglutaric acid could not be obtained by acid or alkaline hydrolysis of the diethyl ester.

Triethyl fluoroisocitrate (II) (1-hydroxy-2-fluoro-1,2,3-propanecarboxylate) was prepared from triethyl fluorooxalosuccinate by selective reduction of the carbonyl group according to the method of Barnett and Kent.[16] In order to prevent extensive decomposition of the starting material due to basic conditions prevailing during reduction by borohydride, the reaction had to be carried out below 0°. When the experiment was carried out at room temperature, only diethyl esters of oxalic and fluorosuccinic acids were obtained. Triethyl fluoroisocitrate forms a crystalline 3,5-dinitrobenzoate when treated with 3,5-dinitrobenzoyl chloride dissolved in pyridine.

Diethyl α-fluoro-γ-hydroxyglutarate (V) was synthesized by a condensation of diethyl fluoromalonate with ethyl pyruvate enol acetate. This reaction was predicted from the fact that diethyl fluoromalonate undergoes normal Michael-type condensations,[7,12,17] originally described by Ostaszynski.[12] The resulting oil showed a strong infrared absorption band at 3450 cm.$^{-1}$ (OH stretching), indicating that the substance is triethyl α-fluoro-α carboxy-γ-hydroxyglutarate. Ethyl acetate could be detected in this reaction mixture, formed by transesterification between the initial condensation product and the solvent, ethanol. Acidic (20% HCl) hydrolysis resulted in a viscous sirup, which upon esterification with ethanol yielded diethyl α-fluoro-γ-hydroxyglutarate (V). Treatment of this ester with 3,5-dinitrobenzoyl chloride gave the 3,5-

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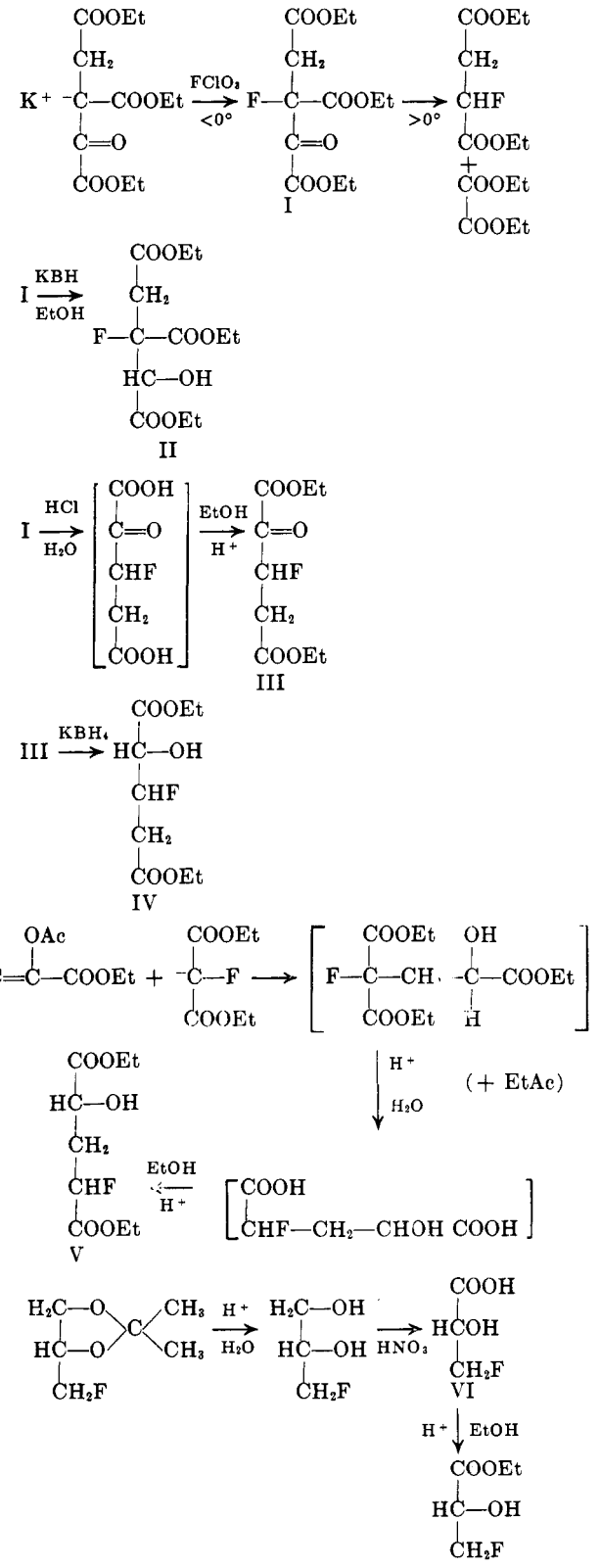
SCHEME I



dinitrobenzoate as an oil. The monoethyl lactone of α-fluoro-γ-hydroxyglutaric acid and its diamide were prepared earlier.[17]

Diethyl α-hydroxy-β-fluoroglutarate (IV) was obtained in good yields by reduction of diethyl α-oxo-β-fluoroglutarate (III) with potassium borohydride.

Monofluorolactic acid (VI) was prepared from 4-fluoromethyl-2,2′-dimethyl-2,3-dioxalane, which upon acidic hydrolysis yields 1-fluoropropane-2,3-diol. This intermediate was oxidized to fluorolactic acid with nitric acid. A similar reaction route was previously described by Bergmann[18] except conditions reported by this author had to be modified in order to obtain the desired products in reasonable yields. Pattison[19] previously prepared 1-fluoropropane-2,3-diol from epifluorohydrin, and by oxidation of the diol with nitric acid obtained ethyl fluorolactate. The modified procedure adopted in our work yielded fluorolactic acid which was purified by vacuum distillation as the ethyl ester. Treatment of the ester with aqueous hydrochloric acid gave pure fluorolactic acid, which was isolated by vacuum distillation. As expected, fluorolactic acid is a stronger acid than lactic acid itself (pK_a of lactic acid = 3.9; pK_a of fluorolactic acid = 3.1). Biochemical properties of fluorolactic acid will be reported elsewhere. The summary of these reactions is given in Scheme I.

Experimental

Triethyl α-Fluorooxalosuccinate.—An efficiently stirred suspension of potassium triethyl oxalosuccinate (37.4 g.) in absolute ethanol (90 ml.) was cooled with an ice–salt bath. Perchloryl fluoride was passed into the mixture by means of a subsurface tube while the reaction temperature was maintained at −5 to 0°. After 0.5 hr. the mixture showed pH 7 on pHydrion paper. Addition of perchloryl fluoride was discontinued and the reaction mixture was flushed with nitrogen to remove the excess perchloryl fluoride. The mixture was then poured into 300 ml. of water while stirring was continued in order to dissolve salts. The aqueous mixture was then extracted with three portions of ethyl ether (100 ml.) and the ether layer was dried with anhydrous Na_2SO_4. Distillation of the ether extract afforded triethyl α-fluoro-α-oxalosuccinate (27 g., 78%): b.p. 115° (0.2 mm.) or 123–124° (0.5 mm.), η^{25}_0 1.4300.

Anal. Calcd. for $C_{12}H_{17}FO_7$: C, 49.5; H, 5.9; F, 6.5. Found: C, 49.3; H, 5.8; F, 6.3.

If the reaction temperature is allowed to rise above 0°, diethyl fluorosuccinate as a lower boiling fraction is obtained in 10 to 20% yields: b.p. 79–80 (1 mm.), η^{25}_0 1.4120.

Anal. Calcd. for $C_8H_{13}FO_4$: C, 50.0; H, 6.7. Found: C, 50.1; H, 6.8.

Treatment of triethyl fluorooxalosuccinate with 2,4-dinitrophenylhydrazine reagent gave diethyl fluorooxalosuccinate 2,4-dinitrophenylhydrazone which was crystallized from ethanol: m.p. 117°.

Anal. Calcd. for $C_{18}H_{21}FN_4O_{10}$: C, 45.8; H, 4.5; N, 11.9. Found: C, 46.0; H, 4.4; N, 11.9.

Treatment of triethyl α-fluoro-α-oxalosuccinate with aqueous ammonia at 50° gave a mixture of fumaric diamide and oxalic diamide. Diethyl fluorosuccinate gave, under identical conditions, fumaric diamide.

Diethyl α-Oxo-β-fluoroglutarate.—Triethyl α-fluoro-α-oxalosuccinate (20 g.) and aqueous 37% hydrogen chloride (35 ml.) were mixed and left at room temperature for 36 hr. The solution was then heated at 65–70° until CO_2 evolution ceased (2 hr.). The solvent was removed under vacuum (bath temperature 70°). Water was then added and the process was repeated twice. The residue, a viscous yellow oil, was dissolved in absolute ethanol (100 ml.) containing *p*-toluenesulfonic acid (0.5 g.) and the mixture was heated under reflux for 24 hr. After standing an additional 36 hr., the solution was treated with excess solid sodium bicarbonate and then filtered. The solvent was removed by vacuum distillation and the residue was extracted into ether. After filtering off insoluble salts from the ether solution and distillation, diethyl α-oxo-β-fluoroglutarate was obtained (10 g., 66%, b.p. 110–112° at 1.0–1.5 mm.). A sample was distilled for analysis: b.p. 107–108° (2 mm.), η^{25}_0 1.4280.

Anal. Calcd. for $C_9H_{13}FO_5$: C, 49.09; H, 5.96; F, 8.5. Found: C, 49.27; H, 6.04; F, 8.4.

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The ester gave, with 2,4-dinitrophenylhydrazine reagent, the 2,4-dinitrophenylhydrazone which was recrystallized from ethanol; m.p. 101°.

Anal. Calcd. for $C_{15}H_{17}FN_4O_8$: C, 45.0; H, 4.3; N, 14.0. Found: C, 44.9; H, 4.4; N, 13.8.

Triethyl Fluoroisocitrate.—Triethyl fluorooxalosuccinate (5.84 g.), dissolved in anhydrous ethanol (25 ml.), was added dropwise to a cooled (−10 to 0°) suspension of potassium borohydride (1.08 g. in anhydrous ethanol, 50 ml.). The addition was completed in 1.5 hr. while the temperature was maintained between −10 and 0°. The reaction mixture was then put into the deep freeze (−5°) for 10 hr. Glacial acetic acid was then added to decompose excess borohydride, and the solvent was distilled under vacuum (bath 30°). Water (20 ml.) was added to the residue and the mixture was extracted twice with ether (50 ml.). The ether was washed with dilute sodium bicarbonate and dried with Na_2SO_4. Distillation gave triethyl fluoroisocitrate: 2.2 g., b.p. 143° (1 mm.). Redistillation for analysis gave 2 g. (34%), b.p. 135–140° (0.75 mm.).

Anal. Calcd. for $C_{12}H_{19}FO_7$: C, 49.0; H, 6.5; F, 6.5. Found: C, 49.2; H, 6.3; F, 6.4.

The infrared spectrum showed strong OH stretching absorption at 3450 cm.$^{-1}$. A 3,5-dinitrobenzoate of the alcohol was prepared with 3,5-dinitrobenzoyl chloride in pyridine. When recrystallized from ethanol, it melted at 118°.

Anal. Calcd. for $C_{10}H_{21}FN_2O_2$: C, 46.72; H, 4.34; N, 5.74. Found: C, 46.94; H, 4.53; N, 5.56.

Diethyl α-Fluoro-γ-hydroxyglutarate.—Diethyl fluoromalonate (9.3 g.) was added to a solution of sodium (0.2 g.) in absolute alcohol (100 ml.). After stirring this solution for 5 min., ethyl α-acetoxyacrylate (7.9 g.) was added dropwise while the stirring continued. After addition, the reaction mixture was left at room temperature for 12 hr. and then neutralized with glacial acetic acid. The solvents were removed under vacuum, and the residue was extracted into ether. Filtration and vacuum distillation of the ether left a yellow oil (12.8 g.). This oil was mixed with 20% HCl (100 ml.) and heated under reflux for 4 hr. The solvent was removed under vacuum and the remaining sirup was taken up in anhydrous ethanol (100 ml.) containing *p*-toluenesulfonic acid (0.5 g.). This ethanolic solution was heated under reflux for 24 hr. Ethanol was then removed under vacuum and the residue was extracted into ether. The ether solution was washed with dilute sodium bicarbonate, separated, and dried with anhydrous Na_2SO_4. After removal of ether, distillation gave diethyl α-fluoro-γ-hydroxyglutarate: 1.8 g., 16%, b.p. 130–135° (4 mm.).

Anal. Calcd. for $C_9H_{15}FO_5$: C, 48.8; H, 6.8; F, 8.5. Found: C, 48.7; H, 6.8; F, 8.4.

The infrared spectra showed OH and C=O stretching absorption at 3450 and 1750 cm.$^{-1}$, respectively. When treated with 3,5-nitrobenzoyl chloride, a dinitrobenzoate was obtained as an oil.[9] Its infrared spectra showed the disappearance of OH absorption at 3450 cm.$^{-1}$ and the appearance of aromatic C–H absorption at 3100, 725, and 730 cm.$^{-1}$, and C=O absorption at 1625 cm.$^{-1}$.

1-Fluoropropane-2,3-diol.—A mixture of 4-fluoromethyl-2,2-dimethyl-1,3-dioxalane[17] (29.8 g.) in water (36 ml.) and 37% hydrochloric acid (11 ml.) was heated under reflux for 0.5 hr. The solvents were removed under vacuum and water was added to the residue. This water was again removed under vacuum and the remaining sirup was taken up in ethanol (50 ml.). The ethanol solution was treated with solid sodium bicarbonate and filtered. Distillation gave pure 1-fluoropropane-2,3-diol: 14.3 g., 68%, b.p. 100–102° (10 mm.) or 117° (30 mm.), η^{25}_0 1.4225; lit.[18] b.p. 117° (30 mm.), η^{25}_0 1.4230.

Ethyl Fluorolactate.—A mixture of 1-fluoropropane-2,3-diol (11.0 g.), water (36 ml.), and 70% nitric acid (50 ml.) was heated on a steam bath. When the temperature of the mixture reached 80–90°. a spontaneous reaction set in and the reaction mixture was removed from the steam bath. (Caution: the reaction can become violent.) When the reaction subsided, the mixture was maintained at 55° for 5 hr. and then left at room temperature for 12 hr., heated an additional hour at 65°, and concentrated at 65° (20–30 mm.). After addition of water (25 ml.), the distillation was repeated and continued to dryness, leaving as a sirup crude fluorolactic acid.

The sirup was heated under reflux with anhydrous ethanol (20 ml.), benzene (60 ml.), and toluenesulfonic acid (0.5 g.) while water was continuously removed by distillation. When no more water was collected, the solvents were removed and the residue was dissolved in ether (50 ml.). The ether solution was treated with solid sodium bicarbonate and filtered, and the product was isolated by distillation, yielding ethyl fluorolactate: 8.8 g., 54%, b.p. 95–98° (30 mm.); lit.[17] b.p. 96–98° (30 mm.).

Fluorolactic Acid.—Ethyl fluorolactate (8.8 g.) was dissolved in 10% hydrochloric acid (50 ml.), and the mixture was heated on a steam bath at 95° for 40 min. After standing for 12 hr. at room temperature, the solvents were removed under vacuum (60° at 20–30 mm.). Water (30 ml.) was added and the distillation was repeated and continued to dryness. The remaining sirup was purified by distillation, yielding fluorolactic acid: 4.3 g., 61%, b.p. 124° (5 mm.). A sample was distilled for analysis: b.p. 122–123° (3 mm.).

Anal. Calcd. for $C_3H_5FO_3$: C, 33.4; H, 4.6; F, 17.6; neut. equiv., 108.1. Found: C, 33.4; H, 4.6; F, 17.4; neut. equiv., 108.9.

Configurations of Substituted 5-Cyanosorbic Acids. An Intramolecular Ritter Reaction

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The ultraviolet, infrared, and n.m.r. spectra of geometric isomers of 3-alkyl-5-cyanosorbic acids (III and V) and 4-carbethoxy-3-methyl-5-cyanosorbic acids (VI and VII) and the configuration of the isomers is found to be in agreement with chemical data. Hydrolysis of 5-cyano-3-methylsorbic acids affords an acid for which structure X is proposed; its formation involves an intramolecular Ritter reaction.

The reaction of substituted pyrylium salts (I) with aqueous alkali cyanides[1] affords 5-cyanopentadienones (II) which are oxidized by hypobromite to 5-cyanosorbic acids (III). Configurations about the C-2–C-3 double bond are most likely to be those indicated in formulas II and III (*cis* position of the carbonyl and the CH=CMeCN groups) if ring cleavage of the cyclic starting compound takes place without isomerization.

On treatment with concentrated mineral acids the 2-*cis*-5-cyanopentadienones (II) undergo two reactions. They eliminate hydrogen cyanide to re-form the initial pyrylium salt (I) and they isomerize into 2-*trans*-5-cyanopentadienones (IV). These latter isomers, and their arylhydrazones, have higher melting points than the 2-*cis* isomers and, unlike them, can no longer be cyclized to pyrylium and pyridinium salts. Hypobromite oxidation of the 2-*trans*-5-cyanopentadienones (IV) leads to 2-*trans*-5-cyanosorbic acids (V). No

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