Attempted Latentiation of 2-Acetamidoethanethiol with α,β-Unsaturated Acids[1,2,†]

Pramod K. Srivastava and Lamar Field*

Department of Chemistry, Vanderbilt University, Nashville, Tennessee 37235. *Received July 6, 1972*

For several years we have been interested in synthesis of compounds which would modify radioprotective thiols in such a way as to reduce their toxicity and/or improve their activity.[1,2,†] Essentially, this approach has been one of "latentiation."[3,4] Latentiation of a drug may provide a means of favorably influencing absorption, transport, distribution, localization, metabolism, and toxicity,[3,4] as well as stability. Hopefully, a radioprotective thiol affords a valid model for developing principles of latentiation applicable to *any* medicinally significant thiol.

This paper describes the use of maleic and cinnamic acids for latentiating 2-acetamidoethanethiol (**1**) by reaction with the thiol function. These two acids were chosen as models on the basis of reactivity in undergoing conjugate addition of thiols. Also, if the aromatic adduct were promising, its properties could be varied markedly by substitution. As a model thiol, **1** was selected because of ready availability and adaptability to addition, combined with reasonably favorable properties as an antiradiation drug.[5] Our hope was that the adducts would show improved properties as antiradiation drugs and that at a biologically appropriate site reverse conjugate addition would reform **1**. Latentiation of 2-mercaptoethylamine hydrochloride with fluoral and chloral led to active compounds,[6] which presumably function by release of the aminothiol.

Addition of **1** to disodium maleate was first tried, the thought being that reaction of the salt would lead to enough thiolate ion for successful addition. This method proved unsatisfactory (only a large amount of fumaric acid could be isolated). Use of the acid alone with **1** (eq 1) proved successful, without a base catalyst, although again some fumaric acid resulted. Thus 2-(2-acetamidoethylthio)succinic acid (**2**) was obtained in about 51% yield by condensation of maleic acid and **1** (eq 1). The cinnamic acid adduct **3** was obtained using piperidine as a catalyst in about 82% yield (eq 2). However, this **3** could not be obtained pure and therefore was converted into the *p*-chloroanilide **4** (eq 2).

$$\underset{1}{AcNH(CH_2)_2SH} + HO_2CCH{=}CHCO_2H \longrightarrow \underset{2}{AcNH(CH_2)_2SCH(CO_2H)CH_2CO_2H} \quad (1)$$

$$\underset{1}{} + PhCH{=}CHCO_2H \xrightarrow{C_5H_{11}N} \underset{3}{AcNH(CH_2)_2SCH(Ph)CH_2CO_2H} \xrightarrow[2.\ H_2NC_6H_4\text{-}p\text{-}Cl]{1.\ SOCl_2} \underset{4}{AcNH(CH_2)_2SCH(Ph)CH_2CONHC_6H_4\text{-}p\text{-}Cl} \quad (2)$$

Biological tests were carried out essentially as recently described [*e.g.*, 30-day survival of mice against lethal radiation, usually of 975 rads (1023**R**) (230 rads/min) from ^{60}Co].[1,‡] Activities of **1** and 2-mercaptoethylamine also have been given previously.[1,5] Compound **2** was inactive (ALD_{50}, mg/kg, 880 ip; the 30-day survival of mice was 0% at dose levels of 250–667 mg/kg ip in H_2O at pH ~3.2). Compound **4** showed activity, although slight (ALD_{50}, 880 mg/kg, ip; the 30-day survival of mice with **4**, given ip in aqueous 0.2% methylcellulose plus 0.4% Tween 80, was 7% at a dose of 500 mg/kg and 20% at a dose of 250 mg/kg; the higher activity at the lower dose perhaps reflects a reduced toxic effect of the **4**).

Although the activity of **4** conceivably could stem from the intact molecule, rather than from released **1**, this activity suggests that latentiation of medicinally significant thiols deserves further investigation with α,β-unsaturated acceptors of such structure that adducts will release the thiol more readily *in vivo*.

Experimental Section[§]

2-(2-Acetamidoethylthio)succinic Acid(2). A mixture of maleic acid (11.6 g, 100 mmol) and **1** (11.9 g, 100 mmol)[7] in 200 ml of EtOAc was heated under reflux with good stirring for 4 hr. A white granular solid (2.3 g, 20%) separated with cooling and was identified as fumaric acid. The filtrate on removal of solvent afforded a viscous oil (12 g, 51%) which was washed several times with C_6H_6 and Et_2O and which then solidified after several days at ~5°, mp 91–98°. This **2** was recrystallized from a mixture of Me_2CO and EtOAc to give **2** having a constant melting point of 119–120°. Tlc showed one spot (1:2 MeOH–Me_2CO); ir 3380, 3300, 3200–2350, 1725, 1700, 1630, 1600, 1590, and 1550 cm^{-1}. *Anal.* ($C_8H_{13}NO_5S$) C, H, N, S. In other experiments, yields of 45–50% were obtained.

3-Phenyl-3-(2-acetamidoethylthio)propanoic Acid(3). A mixture of cinnamic acid (14.8 g, 100 mmol) and **1** (11.9 g, 100 mmol) was heated under reflux for 6 hr in 80 ml of piperidine. The resulting yellow solution was cooled, diluted with H_2O (150 ml), and acidified (pH 1) with concentrated HCl. Semisolid material that separated was extracted with $CHCl_3$ (200 ml) in 50-ml portions. The $CHCl_3$ solution was washed first with H_2O (100 ml) and then with 10% aqueous $NaHCO_3$. The basic solution was acidified and was extracted again with $CHCl_3$. After drying ($MgSO_4$) and removal of solvent, a viscous oil (22 g, 82%) was left, and this was rubbed several times with small amounts of C_6H_6 to remove unchanged cinnamic acid. Tlc of this oil showed two spots and ir absorption at 3300, 1700, 1650, and 1550 cm^{-1}. This oil could not be recrystallized or purified further and therefore was converted into **4**.

***p*-Chloroanilide Derivative 4 of 3.** Crude **3** (20 g, 74.9 mmol) was mixed with $SOCl_2$ (10.6 g, 89 mmol), and the reaction mixture was stirred at ~25° for 24 hr. After the reaction was over, excess $SOCl_2$ was removed at ~20 mm on a steam bath. The light brown material (16 g) that remained, rubbed with two portions of $CHCl_3$ (50 ml) to remove acid chloride, left an unidentified residue. The $CHCl_3$ solution was treated with *p*-chloroaniline (14.3 g, 112.1 mmol), and the reaction mixture was stirred for 1 hr. Solvent then was removed, and the residue was rubbed two times with dilute HCl (20 ml) to remove excess amine and then was washed with H_2O. The resulting semisolid **4** (8.5 g, 30%) was kept at 5° for 24 hr. Three recrystallizations from C_6H_6 gave **4** as white amorphous powder of constant mp 126–127°. Tlc showed one spot (1:2 MeOH–C_6H_6); ir 3340, 1630, and 1590 cm^{-1}. *Anal.* ($C_{19}H_{21}ClN_2O_2S$) C, H, Cl, N, S. In similar experiments yields of 25–30% were obtained.

References

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‡We are indebted for these results to M. M. Grenan and for helpful discussion to D. L. Klayman and T. R. Sweeney, Walter Reed Army Institute of Research, Washington, D. C.

§Melting points, determined in capillary tubes using a Mel-Temp apparatus, are corrected. Ir spectra were obtained using KBr pellets and a Beckman Model IR10 spectrophotometer; bands reported were at least of medium intensity. Tlc spots were obtained using Brinkmann F-254 precoated sheets of silica gel (0.25 mm) on aluminum and were developed by exposure to I_2 vapor in a sealed container. Elemental analyses were by Galbraith Microanalytical Laboratories, Knoxville, Tenn. Where analyses are indicated only by symbols of the elements, analytical results for those elements were within ±0.4% of the theoretical values.

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(3) N. J. Harper, *J. Med. Pharm. Chem.*, 1, 467 (1959).
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Communications to the Editor

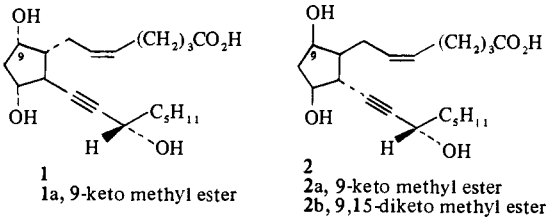
Synthesis and Biological Effects of 13-Dehydro Derivatives of Natural Prostaglandin $F_{2\alpha}$ and E_2 and Their 15-Epi Enantiomers¶

Sir:

In recent communications[1,2] we have described synthetic routes leading to all the primary prostaglandins *via* acetylenic intermediates of type **3** and **11**. The convertibility of these same intermediates into acetylenic prostaglandins, *e.g.*, **1** and **2**, appeared an attractive synthetic goal, in view of the possibility that such substances containing a propargyl in place of the allylic alcohol moiety might turn out to be nonsubstrates, or even antagonists, for the highly specific prostaglandin 15-dehydrogenase[3,4] and thus possess longevity of action not observed with the natural prostaglandins. Such blockage of dehydrogenation had previously been achieved by substituting a methyl group for the 15-hydrogen.[5] We wish to report here the synthesis and biological properties in five test systems of *nat*-13-dehydroprostaglandin $F_{2\alpha}$† (**1**) and *ent*-13-dehydro-15-epiprostaglandin $F_{2\alpha}$† (**2**) and their corresponding E_2 methyl esters **1a** and **2a**. The data presented show that substitution of an acetylenic for the trans-olefinic function does indeed render these substances antagonists of the 15-dehydrogenase and, moreover, gives rise to remarkable changes in their activity profiles.

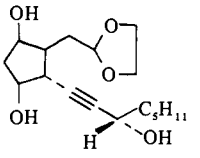
In planning the synthesis of **1** and **2** it was evident that one of the features of our synthetic route, namely, the resolution of the racemic cyclopentane moiety, *e.g.*, **9**, by introduction of the acetylenic 8-carbon side chain in optically active form[1,2] as in **10** or **11** appeared to be precluded since chromatographic separation of the acetylenic diastereomers of type **3** and **11** could not be achieved. Fortunately, one of the diastereomers, **3**,‡ could be obtained in crystalline form,§ mp 102°, $[\alpha]_D$ −2.0° (*c* 2.15),# and the synthesis of **2** completed by hydrolysis of the former with 0.1 *N* HCl in acetonitrile-water (2:1) at 25° for 24 hr to the hemiacetal **4**, $[\alpha]_D$ +1.4° (*c* 2.60), followed by a Wittig reaction as previously described.[1] The resulting *ent*-13-dehydro-15-epiprostaglandin $F_{2\alpha}$ (**2**) obtained in 67% yield from **3**, after purification by high-pressure chromatography, had mp 22–23° and $[\alpha]^{EtOH}_D$ −39.5° (*c* 0.85).** Conversion of the methyl ester of **2** into the corresponding PGE_2 derivative by selective 11,15-silylation with *N*-trimethylsilyldiethylamine in acetone at −40° followed by Collins oxidation[6] furnished **2a**,** $[\alpha]^{EtOH}_D$ −5.6° (*c* 0.45), in 40% yield, together with the recovered methyl ester of **2** (18%) and the 9,15-diketo methyl ester **2b** (20%), λ^{alc}_{max} 222 nm (ε 9600).

Since the other diastereomer corresponding to **3** could not be obtained in pure crystalline form, it was necessary to carry out the synthesis of **1** with resolved (+)-**5**, and, of course, (3*S*)-3*t*-butyloxy-1-octynyldimethylalane,[7] the resolution and determination of absolute configuration of



1
1a, 9-keto methyl ester

2
2a, 9-keto methyl ester
2b, 9,15-diketo methyl ester



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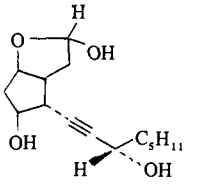

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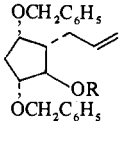
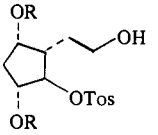
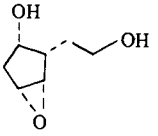


5, R = H
6, R = Tos



7, R = $CH_2C_6H_5$
8, R = H



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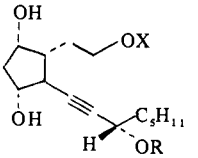
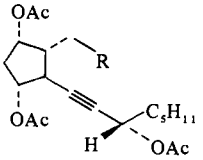
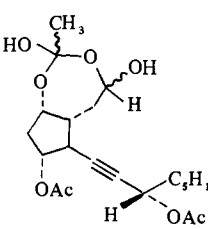


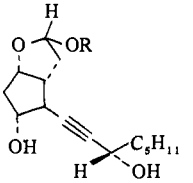
10, X = H; R = *tert*-Bu
11, X = H; R = H
12, X = Tr; R = H



13, R = CH_2OTr
14, R = CH_2OH
15, R = CHO



15a



16, R = CH_3
17, R = H

¶Presented in part at the International Conference of Prostaglandins, Vienna, Sept 25–28, 1972.

†The prefixes nat and ent are employed here to indicate that all the chiral centers present in these prostaglandin analogs correspond to those in the natural prostaglandins (nat) or in their enantiomers (ent).

‡All structural formulas represent the absolute configurations shown.

§*Cf.* footnote 13 in ref 1. The purity of this substance was verified by $LiAlH_4$ reduction to the allylic alcohol, which proved to be a single diastereomer by tlc, a technique which readily distinguishes between such allylic 15-epimers.

#Rotations in chloroform at 28–30° unless indicated otherwise.

**The ir and nmr spectra of this substance as well as the low-resolution mass spectrum of its trimethylsilyl ether methyl ester which showed the M^+ peak were in accord with the assigned structure. Mass spectra were taken on a Finnigan 1015 quadrupole instrument equipped with gc inlet and interfaced with a Systems Industries computer system.