Total Synthesis of 8-*epi*-$PGF_{2\alpha}$. A Novel Strategy for the Synthesis of Isoprostanes

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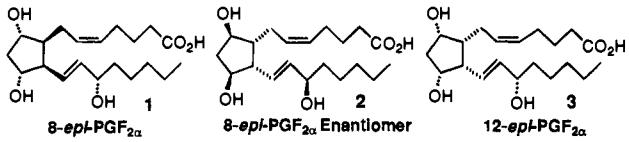
Recently a new biochemical pathway of arachidonic acid metabolism has been uncovered.[2] What is unusual about this pathway is that it is nonenzymatically mediated and initiated by free radicals. In addition, 8-*epi*-prostaglandin $F_{2\alpha}$ (8-*epi*-$PGF_{2\alpha}$, **1**) has been shown to be a product of such biotransformation and to be the most potent renal vasoconstrictor known, 10 times more potent than LTC_4. It is an important causative factor in renal diseases such as hepatorenal syndrome.[3] The thromboxane receptor appears responsible for this pharmacological action.[4] As the first step of our involvement in this program, we needed a general method of synthesis of isoprostanes and, in particular, 8-*epi*-$PGF_{2\alpha}$. We report here on the total synthesis of this natural mediator. Because of the expected

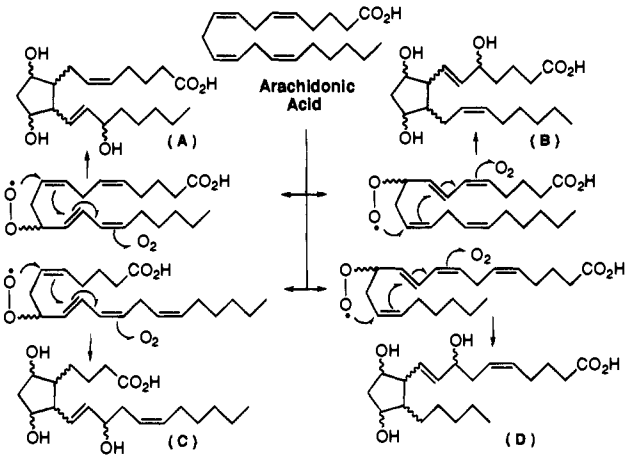


racemic nature of *in vivo* generated 8-*epi*-$PGF_{2\alpha}$ (Scheme 1), we elected to perform the total synthesis of its enantiomer to study the biomechanism of the free-radical oxidation and its biological activity.

Excessive production of free radicals has been claimed, for some time, to have a harmful effect on our systems.[5] Radicals such as HO•, HOO•, and ROO• have been implicated in the initiation and propagation steps in free-radical lipid peroxidation.[6] An increasing body of evidence points to the fact that the so-called antioxidant vitamins such as vitamin E, which are in fact radical inhibitors, protect our systems against cardiovascular and other types of diseases.[7]

Cyclization of secondary radicals to form *gem*-disubstituted cyclopentane derivatives yields mostly *cis*-disubstituted products.[8] In addition, three biomimetic studies using C_{18} linoleic acid[9,10] and C_{20} arachidonate ester[11] -derived peroxy radical intermediates have shown the formation of a preponderance of

Scheme 1



cis-cyclized products of the 8-*epi*-$PGF_{2\alpha}$ type and/or the 12-*epi*-$PGF_{2\alpha}$ type.

As a key component of our strategy we have chosen to use a radical cyclization step at the ring-forming junction of the five-membered ring (Scheme 2). This decision was predicated on our desire to obtain, by controlling the cyclization process, a general method of synthesis which will provide access not only to the *cis-anti-cis* configuration of 8-*epi*-$PGF_{2\alpha}$ (**1**) but also to the *all-syn* arrangement found in 12-*epi*-$PGF_{2\alpha}$ **3**.[12] The synthesis of synthons **10** and **13** was from the outset the central focus of our strategy. The availability of these synthons will also provide, in addition to class A compounds (Scheme 1), access to isoprostanes of types B, C, and D, to labeled isoprostanes necessary for the development of the analytical methodology necessary to accurately assess the involvement of isoprostanes and free-radical damage in disease states.

The key intermediate **8**, which was targeted for the cyclization studies, was prepared in seven efficient steps from L-glucose as shown in Scheme 2. We have selected the thionocarbonate group as a radical precursor for all cyclization reactions.[13,14] Cyclization of diol **8** under radical-generating conditions afforded the *all-syn* five-membered-ring lactone **10** as the major product in 40% isolated yield. The chair conformation (**9**) of the radical intermediate best explains the product formed. Hydrogen bonding of the type shown probably contributes in favoring this 1,3-diaxial arrangement of the two OH groups. In order to force the radical species to assume the desired chair conformation with the OHs in the equatorial positions for the generation of the *cis-anti-cis* structure of the lactone **13**, we opted to substitute the two hydroxy groups, with the result of eliminating the hydrogen bonding and at the same time providing bulk so as to ensure that the intermediate radical species assumes the conformation shown in **12**. When we carried out the cyclization on the bis-acetyl compound **11a**, and then on the more bulky bis-silyl derivative **11b**, $[\alpha]_D$ −26.1° (*c* 1.0, MeOH), we were gratified to see that the major product of this cyclization was indeed the desired **13b**, isolated in 41% yield.

Spectroscopic evidence confirms the structural and stereochemical assignment shown in Scheme 2.[15] In addition,

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(14) We have found the thionocarbonate group to be an extremely convenient secondary radical precursor for cyclization reactions. Rondot, B.; Durand, T.; Girard, J. P.; Rossi, J. C.; Schio, L.; Khanapure, S. P.; Rokach, J. *Tetrahedron Lett.* **1993**, *34*, 8245. The use of this group saves a protection and deprotection step of the primary alcohol. In addition, the thionocarbonate group is more stable than a thioxanthate group under the various conditions shown in Scheme 2.

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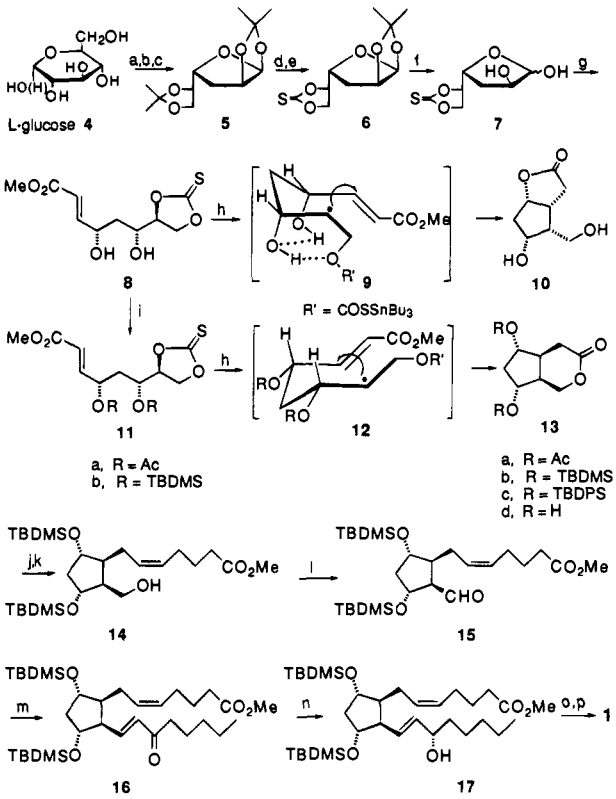
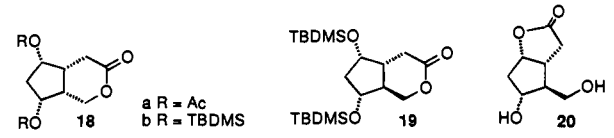
Scheme 2[a]



[a] Reaction conditions: (a) acetone, 85% H_3PO_4, $ZnCl_2$, room temperature, 18 h, 91%; (b) 1.5 equiv of NaH, THF, 0 °C, 30 min, CS_2, room temperature, 30 min, MeI, room temperature, 30 min, 98%; (c) 1.6 equiv of n-Bu_3SnH, toluene, reflux, 8 h, 87%; (d) 30% aqueous acetic acid, room temperature, 24 h, 80%; (e) 1.2 equiv of 1,1'-thiocarbonyldiimidazole, CH_2ClCH_2Cl, reflux, 3 h, 89%; (f) 4% aqueous H_2SO_4, THF, reflux, 3 h, 85%; (g) 2 equiv of methyl (triphenylphosphoranylidene)acetate, TDA, THF, 18 h, 60%; (h) 2 equiv of n-Bu_3SnH, 0.6 equiv of AIBN, C_6H_6, reflux, 4 h; (i) t-$BuMe_2SiCl$, imidazole, DMF, 60 °C, 18 h, 71% from **7**; (j) 3 equiv of DIBAL-H, CH_2Cl_2, −78 °C, 2 h, H_2O, 0 °C, 1 h, 87%; (k) 4 equiv of $[Ph_3P(CH_2)_4CO_2H]Br$, t-BuOK, THF, room temperature, 1.5 h, CH_2N_2, CH_2Cl_2, room temperature, 5 min, 75%; (l) 2 equiv of $(COCl)_2$, 3 equiv of DMSO, CH_2Cl_2, −78 °C, 15 min, then **14**, −78 °C, 30 min, 6 equiv of Et_3N, −78 °C, 93%; (m) 1.7 equiv of dimethyl 2-oxoheptanephosphonate, $NaN(SiMe_3)_2$, THF −78 °C, 10 min, then to 0 °C, 3 h, 93%; (n) 3 equiv of (*S*)-BINAL-H, THF, −100 °C, 2 h, −100 °C to −78 °C, 3 h, 94%; (o) n-Bu_4NF, THF, room temperature, 18 h, 95%; (p) KOH, H_2O, room temperature, 10 min, 10% aqueous oxalic acid, 93%.

compound **13a**, after basic hydrolysis followed by acidification, afforded lactone **13d**, indicating a *trans* relationship of the hydroxy group and the side chains (lactone ring). By contrast, the *all-syn* six-membered-ring lactone **18**, obtained in small amounts (6−8%) in the cyclization reaction, on fluoride deprotection of the silyl groups followed by hydrolysis, afforded the five-membered-ring lactone **10**, indicating the *all-cis* stereochemistry. In both cyclization reactions, minor amounts of a *trans*-fused lactone **19** are obtained. Fluoride desilylation afforded the five-membered-ring lactone **20**, identical with a commercial sample.



The reduction of **13b** with DIBAL-H yielded a mixture of lactol epimers which was used as such in the next step. Wittig reaction with commercial (4-carboxybutyl)triphenylphosphonium bromide afforded **14**. The oxidation of the alcohol in **14** afforded aldehyde **15**. Introduction of the lower side chain proceeded smoothly to yield **16**. The enantioselective reduction of the C_{15} keto function in **16** proceeded smoothly with (*S*)-BINAL-H with >95% enantiomeric excess under conditions already reported[12,16] and afforded the 15*S* derivative **17**, which, after fluoride removal of the silyl groups and basic hydrolysis, yielded the desired 8-*epi*-$PGF_{2\alpha}$ (**1**).

The synthesis of the enantiomer **2** of 8-*epi*-$PGF_{2\alpha}$ was performed by first preparing the enantiomer of synthon **8** from D-glucose.[14] The bis-TBDMS derivative was prepared, and this cyclization precursor, enantiomeric with **11b**, $[\alpha]_D$ +25.7° (*c* 1.0, MeOH), was used and the synthesis completed as shown in Scheme 2 for the L-series. The reduction of the keto function at C_{15} was carried out with (*R*)-BINAL-H instead of the (*S*)-BINAL-H used in the synthesis of 8-*epi*-$PGF_{2\alpha}$.

The control of the cyclization step we have achieved allows ready access to the two key synthons **10** and **13** from one starting material and one synthetic strategy.

The importance of the isoprostanes, in our view, is related not only to their bioactivity but to the biomechanism of their formation. Arachidonic acid is mostly stored in the esterified form in phospholipids. Phospholipids are the main constituents of the cell membranes. The hydrophobic nature of these membranes is what keeps the cell integrity. The free-radical-initiated oxygenation of these phospholipids will cause the formation of isoprostanes and other oxygenated polar molecules in the midst of a hydrophobic environment. This might result in changes in membrane fluidity, generation of leaks, and eventual cell death. In our view, it is almost certain that some of the protective action of antioxidant vitamins, such as vitamin E, is at the phospholipid level and plays a role in the preservation of cell integrity. This, however, remains to be shown. The present contribution is a first step in a program designed to measure 8-*epi*-$PGF_{2\alpha}$ and eventually other isoprostanes in biological fluids in the presence of enzymatic products as an *in vivo* index of degenerative diseases in which cell destruction is documented, such as myocardial infarct, atherosclerosis, arthritis, and Alzheimer's.

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Supplementary Material Available: IR, ^1H NMR, and ^{13}C NMR spectral data for all compounds described herein (35 pages). This material is contained in many libraries on microfiche, immediately follows this article in the microfilm version of the journal, and can be ordered from the ACS; see any current masthead page for ordering information.

(15) Extensive ^1H, ^{13}C, $^1H-^1H$ COSY, NOE, and DEPT NMR studies confirm the stereochemical assignment for **10** and **13** as well as all reported compounds in this manuscript. The TBDPS derivative **13c** was prepared for NOE experiments.

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