PREPARATION OF B-RING BROMINATED DERIVATIVES OF ESTRADIOL

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ABSTRACT

The preparation of 6β-methoxy-7α-bromoestradiol (1) is reported. Addition of in situ-generated BrOMe to 6-dehydroestradiol 3-tetrahydropyranyl ether (4a) afforded the expected addition product. Removal of the tetrahydropyranyl protective group was carried out by mild acid hydrolysis to afford 1 in good yields. This base-sensitive compound is chemically stable over long periods of time.

INTRODUCTION

The synthesis of halogenated estradiol derivatives has been an area of interest in recent years, since their radiohalogenated analogs are potentially useful as imaging agents in nuclear medicine [1]. Several A-ring and D-ring radiohalogenated estradiol derivatives have been prepared and tested for potential clinical applications [2,3], but little work has been reported concerning the radiohalogenation of the B- or C-rings, possibly due to the suspected low in vivo stability of some of these compounds [4]. The preparation of 6-[^{125}I]iodo-6-dehydroestradiol and its reasonably high tumor/blood ratio as reported by Caspi and co-workers [5], prompted us to further investigate halogenation procedures that could be applied to the B-ring of estradiol.

RESULTS AND DISCUSSION

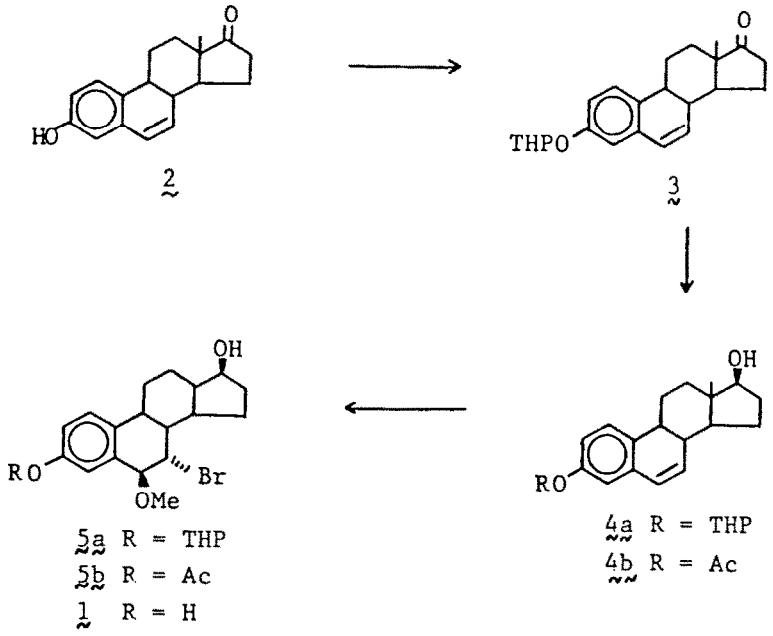
The addition of an electrophilic halogenated species to an unsaturated site has been used in the past to produce B- and C-ring halogenated steroids [6,7]. An adaptation of this

procedure to the radiohalogenation of estradiol derivatives has also been recently reported by our group [4].

Treatment of 6-dehydroestradiol with BrOMe, generated in situ from sodium bromide/N-chlorosuccinimide (NaBr/NCS) in methanol under conditions identical to those previously used [8], led to a complex mixture of products. In contrast, treatment of synthetically available 6-dehydroestradiol 3-acetate under the same conditions afforded the desired 6β-methoxy-7α-bromoestradiol 3-acetate (5b). Attempts to remove the 3-acetate group, under a variety of standard conditions, resulted in loss of the halogen as verified using the ^{77}Br-radiobrominated analog of 4b.

After investigation of several other phenolic protective groups the tetrahydropyranyl group (THP) was chosen. Reaction of dihydropyran (DHP) with commercially obtained 6-dehydroestrone (2) afforded the 3-OTHP derivative 3 in 74% yield. Reduction of 3 with lithium aluminum hydride triterbutoxide [$LiAl(OtBu)_3H$] afforded the estradiol analog 4a which, after isolation, was reacted with BrOMe (prepared in situ from NaBr and NCS in methanol), generating the desired addition product 5a. Deprotection was accomplished in situ by the addition of diluted HCl to the reaction mixture, affording 6β-methoxy-7α-bromoestradiol (1) in 64% overall yield from 2 (Scheme I).

Scheme I



Addition of BrOMe to an isolated double bond is known to proceed in a trans-fashion via a bromonium ion [9], making the benzylic 6-position the most likely to be attacked by the incoming methoxy group from the ß-face. We predicted an α-bromonium ion intermediate, based on the steric environment of the ß-face as viewed using molecular models. The trans-stereochemistry of 1 was also confirmed by the values of the coupling constants for protons 6α and 7ß (1.90 Hz and 2.10 Hz respectively), which are typical values for trans-equatorial-equatorial vicinal protons. The stereochemistry and regio-chemistry shown in Scheme I are in agreement with those

suggested by Caspi and co-workers in their synthesis of the halohydrin analogs [6], and the chemical shifts of protons 6 and 7 (4.40 ppm and 4.30 ppm respectively) are also in agreement with the assigned structure.

EXPERIMENTAL

6-Dehydroestrone was used as obtained from Sigma Co. All other reagents were purchased from Aldrich Co. and used as obtained. Analytical grade methanol was dried over molecular sieves prior to use. Tetrahydrofuran (THF) was distilled from Na-benzophenone before use. Melting points were obtained on an electrothermal melting point apparatus and are uncorrected. Elemental analyses were performed by Ruby Ju of the UNM chemistry department. Proton NMR spectra were recorded at 60 MHz on a Hitachi Perkin Elmer R-24B instrument and at 300 MHz on a Bruker WM-300 wide bore, and are referenced to tetramethylsilane as an internal standard using deuterated chloroform as the solvent.

Product purity and reaction progress were followed by analytical thin-layer chromatography (TLC) using GF-Baker plates. Medium pressure liquid chromatography (MPLC) was performed at ~80 psi using a fluid metering pump, 9x100 mm glass column, and Woelm 32-64 micron silica gel as the stationary phase.

3-Hydroxy-1,3,5(10),6-estratetraen-17-one 3-tetrahydropyranyl ether (3)

To a suspension of 0.30 g (1.12 mmol) of 6-dehydroestrone (2) in 12 mL of CH_2Cl_2, kept at room temperature and under a N_2 atmosphere, was added 0.20 mL (2.24 mmol) of DHP, followed by 5 mg (0.03 mmol) of paratoluenesulfonic acid (p-TsOH·H_2O). The resultant purple solution was stirred for 2h, and 15 mL of 5% aqueous $NaHCO_3$ solution and 15 mL of CH_2Cl_2 was added. The organic layer was then washed with water (1x10 mL), dried over anhydrous $MgSO_4$, filtered, and the solvent was removed under reduced pressure to afford 0.40 g of a pale yellow solid. Purification was performed by means of an MPLC system using 10% ethyl acetate in toluene as the eluant to give 0.29 g (75% yield) of colorless crystals, mp 150-152°C. TLC: Rf = 0.53 (15% ethyl acetate in toluene). ^{1}HNMR: δ 7.25-6.85 (m, 3H); 6.60 (d, 1H); 6.10 (d, 1H); 5.45 (t, 1H); 4.10-1.25 (m, 19H); 0.89 (s, 3H). IR (KBr): 1740 cm^{-1} (-C=O). Anal. calcd for $C_{23}H_{28}O_3$: C, 78.37; H, 8.01. Found: C, 78.46; H, 7.95.

1,3,5(10),6-Estratetraene-3,17β-diol 3-tetrahydropyranyl ether (4a)

To a solution of 0.19 g (0.54 mmol) of 3 in 20 mL of dry THF was added 0.57g (1.08 mmol) of $LiAl(OtBu)_3H$. The resultant mixture was stirred at room temperature under a N_2 atmosphere for 12 h, after which 5 mL of water, 15 mL of 10% aqueous NaOH, and 0.27 g of Rochelle Salt (potassium and sodium tartrate) was added. The milky suspension obtained was stirred for 5 min, and 20 mL of ethyl acetate was added. The resultant two-phase mixture was stirred for 10 min and then separated, and the aqueous phase was extracted with ethyl acetate (2x15 mL). The combined organic layers were then washed with H_2O (1x20 mL), dried over anhydrous $MgSO_4$, filtered, and the solvent was removed under reduced pressure to afford 0.19 g (99% yield) of 4a as a colorless oil. An analytical sample was obtained by MPLC purification using 15% ethyl acetate in toluene as the eluant. TLC: Rf = 0.36 (25% ethyl acetate in toluene). ^{1}HNMR: δ 7.27-6.81 (m, 3H); 6.45 (d, 1H); 5.92 (d, 1H); 5.40 (t, 1H); 3.95-1.24 (m, 21H); 0.75 (t, 3H). IR (KBr): 3600-3200 cm^{-1} (-OH). Anal. calcd for $C_{23}H_{30}O_3$: C,77.93; H, 8.53. Found: C, 77.49; H, 8.66.

7α-Bromo-1,3,5(10)-estratriene-3,6β,17β-triol 6-methyl ether (1)

To a solution of 0.07 g (0.53 mmol) of NCS in 20 mL of dry methanol was added 0.055 g (0.53 mmol) of NaBr. The resultant pale yellow solution was stirred at room temperature for 10 min and a solution of 0.17 g (0.48 mmol) of 4a in 5 mL of methanol was added. After stirring at room temperature for 30 min, 100 μL (1.2 mmol) of concentrated HCl solution was added and the colorless mixture was stirred at room temperature for an additional 30 min. The solvents were removed under reduced pressure and the obtained residue was partitioned between water (10 mL) and ethyl ether (30 mL). The ethereal layer was then washed with water (1x15 mL), dried over anhydrous $MgSO_4$, filtered, and the solvent removed under reduced pressure to afford 0.16 g (87% yield) of colorless solid, mp 95-107 °C. An analytical sample was obtained by MPLC purification using 20% ethyl acetate in toluene as the eluant. TLC: Rf = 0.52 (50% ethyl acetate in toluene). ^{1}HNMR: δ 7.19-6.69 (m, 3H); 4.67 (bs, 1H); 4.30 (bd, 1H); 3.72 (s, 3H); 2.48-1.19 (m, 15H): 0.76 (s, 3H). IR (KBr): 3620-3060 cm^{-1} (-OH). Anal. calcd for $C_{19}H_{25}O_3Br$: C, 59.84; H, 6.61. Found: C, 59.70, H, 6.83.

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NOTES AND REFERENCES

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NOMENCLATURE

6β-Methoxy-7α-bromoestradiol (1)	7α-Bromo-1,3,5(10)estratriene-3,6,17β-triol 6-methyl ether
6-Dehydroestradiol 3-acetate	1,3,5,(10),6-estratetraene-3,17β-diol 3-acetate
6-Dehydroestradiol (3)	1,3,5(10),6-estratetraene-3,17β-diol
6-Dehydroestradiol 3-tetrahydropyranyl ether (4a)	1,3,5(10),6-estratetraene-3,17β -diol 3-tetrahydropyranyl ether
17β-Dihydroequilenin 3-benzoate	1,3,5,(10),6,8-estrapentaene-3,17β-diol 3-benzoate
17β-Dihydroequilin 3-benzoate	1,3,5(10),7-estratetraene-3,17β-diol 3-benzoate
Estradiol	1,3,5,(10)-estratriene-3,17β-diol