B-RING AROMATIZATION OF ESTROGEN DERIVATIVES

M.D. Hylarides[1], D.S. Wilbur[2], F.A. Mettler, Jr.[3]

[1, 3]Department of Radiology, University of New Mexico
Albuquerque, New Mexico 87131

[2]Medical Radioisotope Research (CNC-3); MS-E514
Los Alamos National Laboratory, Los Alamos, New Mexico 87545

Received 5-9-83

ABSTRACT

A novel method of B-ring aromatization of steroid derivatives is reported. Addition of BrCl in methanol to a non-aromatic B-ring double bond results in a rapid double elimination generating the aromatic B-ring. This procedure represents an effective method for conversion of an equilin to an equilenin nucleus.

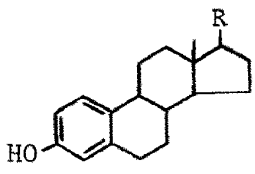
INTRODUCTION

The preparation of equilenin via B-ring aromatization has been previously reported. Some of the described procedures utilized a dehydrogenation step to generate the aromatic B-ring. For example, dehydrogenation of estrone 1a with Pd/C at elevated temperatures produced isoequilenin 2 (1). Unfortunately, epimerization at the C/D ring juncture accompanied the dehydrogenation, probably as a result of the elevated reaction temperature. Equilin 3a, however, was readily converted to equilenin 4a by dehydrogenation with Pd/C at 80°C (2). The milder reaction conditions yielded no epimerization at the C/D ring juncture (3). Other reactions that result in aromatization of the B-ring include the dehydrogenation of 6-dehydroestrone-3-acetate 5 with SeO_2 which produces equilenin-3-acetate 4c in moderate yields (4), and the incubation of equilin with rat liver homogenate which results in the formation of equilenin, probably through a similar dehydrogenation process (5).

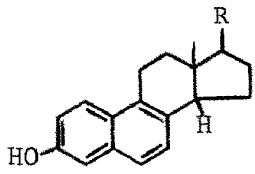
Other methods of B-ring aromatization include epoxidation of the non-aromatic B-ring double bond of equilin followed by acidic

rearrangement and dehydration to generate equilenin (6). In an analogous manner, equilenin was produced from 8-dehydroestrone 6 (7).

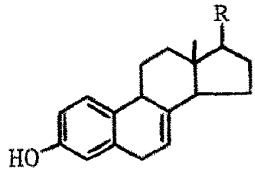
We now report a novel and mild conversion of an equilin nucleus to an equilenin nucleus that was observed during bromination studies involving the B-ring of estradiol 1b. This room temperature procedure, which employs the use of the electrophilic brominating agent BrCl, represents an additional method to effect the B-ring aromatization. In addition to high yields, simplicity and mildness of the reaction conditions, there is no evidence of epimerization at the C/D ring juncture.



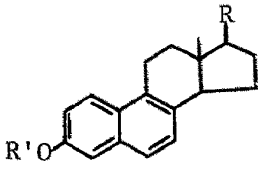
1a R = =O
1b R = OH



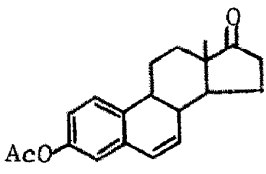
2



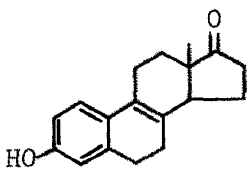
3a R = =O
3b R = OH



4a R = =O, R' = H
4b R = OH, R' = H
4c R = =O, R' = Ac



5



6

Figure 1

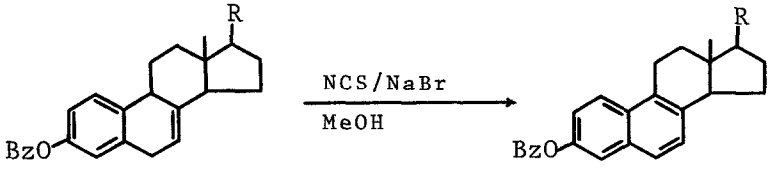
RESULTS AND DISCUSSION

Bromine monochloride is conveniently prepared in situ from the reaction of N-chlorosuccinimide (NCS) with NaBr in protic or aprotic media (8). It has been demonstrated that reaction of the brominating

agent with an olefin or a protic solvent, such as methanol, results primarily in the addition of one bromine atom and one methoxyl group (8).

To prevent A-ring bromination (9), the free phenols of equilin 3a and 17β-dihydroequilin 3b were protected as the corresponding benzoate esters 7a and 7b (10). Treatment of 7a or 7b with BrCl in methanol did not give the expected brominated adducts; rather, the addition was followed by a rapid double elimination (-HBr, $-CH_3OH$) to generate equilenin-3-benzoate 8a from 7a and 17β-dihydroequilenin-3-benzoate 8b from 7b. The reaction is complete within a few minutes at room temperature and 84-92% of aromatized product was obtained.

After subsequent removal of the benzoate groups from 8a and 8b, the resultant equilenin and 17β-dihydroequilenin were compared (mp, ^{1}H NMR, ^{13}C NMR) to known equilenin and 17β-dihydroequilenin standards. In each case, the physical characteristics of the synthesized material were identical to those of the standards.



7a R = =O
7b R = OH

8a R = =O
8b R = OH

Figure 2

EXPERIMENTAL SECTION

d,l-Equilin and d,l-equilenin (Sigma) were used as obtained. N-chlorosuccinimide was obtained from Aldrich and used as obtained. Sodium bromide was obtained from Mallinckrodt and was finely pulverized prior to use. Analytical grade methanol was dried over molecular sieves prior to use. THF (tetrahydrofuran) was freshly distilled from Na-benzophenone prior to use. All melting points were

obtained on a Thomas Hoover melting point apparatus and are corrected. Proton NMR spectra were obtained on a Varian EM-360 or a Jeol FX-90 instrument and are referenced to tetramethylsilane at 0.0 ppm. Carbon NMR data were obtained on either a Varian FT-80 or a Jeol FX-90 spectrometer and chemical shifts obtained are referenced to the center peak of the deuterated solvent used.

Reduction of Equilin: 1,3,5(10),7-estratetraene-3,17β-diol 3b

Commercially available equilin (100 mg, 0.37 mmol) was predried and dissolved in 10 mL of freshly distilled THF. The resultant solution was cooled to 0°C and 190 mg (0.75 mmol) of $LiAl(OtBu)_3H$ was added. The reaction mixture was allowed to stir for 16 h under N_2 at room temperature. After hydrolysis with 5% HCl and recrystallization of the crude product from toluene, 80 mg (80% yield) of 3b was obtained, mp = 174.5-175°C (reported (11) mp = 174-175°C). ^{13}C NMR (acetone-d_6) of the aromatic and olefinic carbons: δ138.0, 134.5, 129.4, 128.9, 114.4, 114.1, 114.0; ^1H NMR (acetone-d_6) of aromatic and olefinic protons: δ7.1(d, 1H), 6.8(m, 2H), 5.4(t, 1H).

Reduction of Equilenin: 1,3,5(10),6,8-estrapentaene-3,17β-diol 4b

In like manner as described for the preparation of 3b, equilenin 4a (39 mg, 0.15 mmol) was reduced with 150 mg (0.60 mmol) of $LiAl(OtBu)_3H$. After recrystallization of the crude product from ethyl acetate - petroleum ether, 22 mg (56% yield) of 4b was obtained, mp = 240-242°C (reported (4) mp = 248°C). ^{13}C NMR (DMSO-d_6) of the aromatic carbons: δ154.0, 133.1, 129.9, 126.1, 124.6, 124.4, 124.1, 118.2, 109.3. ^1H NMR (DMSO-d_6) of the aromatic hydrogens: δ7.2(d, 1H), 6.8(d, 1H), 6.3-6.6(m, 3H).

3-Hydroxyl-1,3,5(10),7-estratetraen -17-one 3-benzoate 7a

Commercially available equilin (500 mg, 1.86 mmol) was dissolved in 4 mL of dry pyridine. Benzoic anhydride (1.0 g, 4.42 mmol) was added and the resultant mixture was allowed to stir under N_2 for two days. Following hydrolysis with 5% HCl, the organic material was extracted with ethyl acetate - methylene chloride (1:1). After drying over anhydrous $MgSO_4$, removal of the solvents and recrystallization from ethyl acetate, 548 mg (80% yield) of 7a was obtained, mp = 195-196°C (reported (12) mp = 197-198°C). ^{13}C NMR ($CDCl_3$) for aromatic and olefinic carbons: δ148.9, 135.5, 134.9, 134.4, 133.4, 130.0, 129.5, 128.7, 128.4, 120.9, 119.5, 115.6; ^1H NMR ($CDCl_3$) for aromatic and olefinic hydrogens: δ6.9-7.6(m, 6H), 5.5(t, 1H).

1,3,5(10),7-Estratetraene-3,17β-diol 3-benzoate 7b

In like manner as described for the preparation of 7a, 17β-dihydroequilin 3b (200 mg, 0.74 mmol) was esterified with benzoic anhydride (0.44 g, 1.95 mmol). After recrystallization from ethyl acetate - petroleum ether, 100 mg (37% yield) of 7b was obtained, mp = 169-170°C. ^{13}C NMR ($CDCl_3$): δ189.4, 148.8, 136.8, 135.5, 134.7, 133.2, 130.0, 129.8, 128.6, 128.5, 128.4, 120.7, 119.3, 113.9, 81.9,

50.3, 45.1, 40.8, 37.5, ,32.4, 30.5, 29.7, 11.0; ^{1}H NMR ($CDCl_3$): δ8.1-8.3(m, 2H), 6.9-7.7(m, 6H), 5.4(t, 1H), 3.5(m, 2H), 3.1(t, 1H), 1.3-2.4(m, 10H), 0.8(s, 3H). IR (KBr): 3400-3600, 2800-3000, 1770, 1600, 1500, 1450, 1270, 1250, 1210 cm^{-1}. UV (MeOH): λ_{max} = 225 nm (ε = 21,300)

Anal. Cald. for $C_{25}H_{26}O_3$: C, 80.18; H, 7.00. Found: 79.98; H, 6.97.

3-Hydroxy-1,3,5(10),6,8-estrapentaen -17-one 3-benzoate 8a

Equilin-3-benzoate 7a (100 mg, 0.27 mmol) was added to a solution of a 30.5 mg (0.30 mmol) of NaBr in 15 mL of methanol. After the addition of 39.9 mg (0.30 mmol) of NCS in 20 mL of methanol, the resultant solution was stirred at room temperature for 1 h. After removal of the methanol under reduced pressure, 30 mL of 1:1 ethyl acetate-ethyl ether was added. The insoluble salts were filtered off and the organic filtrate was washed twice with 10 mL portions of water and dried over anhydrous $MgSO_4$. The resultant crude oil was chromatographed on a short alumina column (25% ethyl acetate-toluene) and 92 mg (92% yield) of 8a was obtained, mp = 217-219°C (reported (13) mp = 222-223°C). ^{13}C NMR ($CDCl_3$) for the aromatic carbons: δ165.1, 148.1, 134.4, 133.4, 132.9, 131.0, 130.0, 129.6, 128.4, 126.3, 124.7, 124.5, 121.2, 119.1; ^{1}H NMR ($CDCl_3$) for the aromatic hydrogens: δ8.2(d, 2H), 8.0(d, 1H), 7.1-7.8(m, 7H).

1,3,5(10),6,8-Estrapentaene-3,17β-diol 3-benzoate 8b

In like manner as described for the preparation of 8a, 17β-dihydroequilin-3-benzoate 7b (96 mg, 0.25 mmol) was treated with NaBr (29 mg, 0.28 mmol) and NCS (37.2 mg, 0.28 mmol) in a total of 33 mL of methanol. After chromatography on alumina with the same solvent system, 80 mg (84% yield) of 8b was obtained, mp = 213.5-215.5°C (reported (4) mp = 215°C). ^{13}C NMR ($CDCl_3$) for the aromatic carbons: δ182.4, 148.8, 136.1, 133.8, 132.5, 130.5, 130.1, 129.9, 129.8, 128.4, 128.3, 125.3, 124.7, 120.8, 119.0; ^{1}H NMR ($CDCl_3$) for the aromatic hydrogens: δ8.2(d, 2H), 8.0(d, 1H), 7.1-7.7(m, 7H).

3-Hydroxy-1,3,5(10),6,8-estrapentaen -17-one 4a

Equilenin-3-benzoate 8a (0.7 g, 0.46 mmol) was suspended in 10 mL of 10% ethanol KOH solution. After stirring for 12 h, removal of the ethanol under reduced pressure and acidification, 107 mg (88% yield of crude) equilenin was obtained. Recrystallization from methanol-water gave the purified material, mp = 252.5-254°C (reported (13) mp = 258-259°C). ^{13}C NMR (DMSO-d_6) for the aromatic carbons and carbonyl carbons: δ218.4, 154.4, 133.5, 131.2, 130.3, 126.7, 124.6, 124.4, 123.9, 119.4, 109.4; ^{1}H NMR (DMSO-d_6) for the aromatic hydrogens: δ7.9(d, 1H), 7.7(d, 1H), 619-7.3(m, 3H).

1,3,5(10),6,8-Estrapentaene-3,17β-diol 4b

In like manner as described for the preparation of 4a, 17β-dihydroequilenin-3-benzoate 8b (80 mg, 0.22 mmol) was saponified in 5 mL of 10% KOH-ethanol to obtain 50 mg (76% yield) of 4b mp =

241-243°C (reported (4) mp = 248°C). ^{13}C NMR (DMSO-d_6) for the aromatic carbons: δ154.0, 133.1, 130.0, 126.0, 124.7, 124.6, 124.2, 118.2, 109.3; ^{1}H NMR (DMSO-d_6) for the aromatic hydrogens: δ8.0(d, 1H), 7.7(d, 1H), 7.1-7.4(m, 3H).

ACKNOWLEDGEMENT

We are very grateful for the financial support of the Department of Energy (Contract #DE-AC04-81EV10596).

REFERENCES

1. Butenandt, A., Wolff, A., and Karlson, P. Chem. Ber. 74, 1308 (1941).
2. Dirscherl, W., and Hanusch, F. Z. Physiol. Chem. 233, 13 (1935).
3. Bachmann, W.E., and Dreiding, A.S. J. Amer. Chem. Soc. 72, 1323 (1950).
4. Kaufman, S., Pataki, J., Rosendranz, G., Roma, J., and Djerassi, C. J. Amer. Chem. Soc. 72, 4531 (1950).
5. Breuer, H., and Mittermayaer, C. Biochem. J. 86, 12 (1963).
6. Smith, H., Smith, R., and Reinhard, P. (American Home Products Corp.), S. African Patent 67 04,741, January 17, 1968.
7. Ginsig, G. (Syntex Corp.), U.S. Patent 3,647,831, March 7, 1972.
8. Wilbur, D.S., and Anderson, K.W. J. Org. Chem. 47, 358 (1982).
9. Wilbur, D.S., and O'Brien, H.A., Jr. J. Org. Chem. 47, 359 (1982).
10. The corresponding acetates were also investigated and found to undergo the B-ring aromatization.
11. Stein, R.P., Buzby, G.C., Jr., and Smith, H. Tetrahedron 26, 1917 (1970).
12. Girard, A., Sandulesco, G., Fridenson, A., and Rutgers, J. Comptes Rendus 194, 1020 (1932).
13. Girard, A., Sandulesco, G., and Rutgers, J. Comptes Rendus 195, 981 (1932).

GLOSSARY

1.	8-Dehydroestrone 6:	3-hydroxy-1,3,5(10),8(9)-estratetraen -17-one.
2.	6-Dehydroestrone-3-acetate 5:	3-hydroxy-1,3,5(10),6-estratetraen -17-one 3-acetate.
3.	Equilenin-3-acetate 4c:	3-hydroxy-1,3,5(10),6,8-estrapentaen -17-one 3-acetate.
4.	Estradiol 1b:	1,3,5(10)-estratriene-3,17β-diol.
5.	Isoequilenin 2:	3-hydroxy-14β-estra-1,3,5(10),6,8-pentaen -17-one.