Synthesis and Evaluation of (17α,20E)-21-[^{125}I]Iodo-19-norpregna-1,3,5(10),20-tetraene-3,17-diol and (17α,20E)-21-[^{125}I]Iodo-11β-methoxy-19-norpregna-1,3,5(10),20-tetraene-3,17-diol (17α-(Iodovinyl)estradiol Derivatives) as High Specific Activity Potential Radiopharmaceuticals

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Two 17α-[^{125}I]iodovinyl estradiol derivatives **4b,d** possessing high specific activity have been prepared and tested as potential radiopharmaceuticals. The use of the 3-acetyl derivatives **2c,e** and the replacement of iodine monochloride with sodium iodide and Chloramine-T in THF/phosphate buffer (pH 7.0) permitted us to synthesize no-carrier-added (17α,20E)-21-[^{125}I]iodo-19-norpregna-1,3,5(10),20-tetraene-3,17-diol (**4b**) and (17α,20E)-21-[^{125}I]iodo-11β-methoxy-19-norpregna-1,3,5(10),20-tetraene-3,17-diol (**4d**) with 50% radiochemical yield and high purity. Although the specific activity represents only half of the theoretical value in some cases, this modified approach is a substantial improvement over the previously published method. Our preliminary distribution studies indicate that although both **4b** and **4d** localize in the tissues known to have a large concentration of estrogen receptors, **4d** accumulates in higher amounts in target tissues and provides a high target to nontarget ratio.

A number of radiohalogenated estrogen analogues have been prepared and tested as potential receptor imaging agents for human breast tumors.[2-5] Since the selective uptake of those imaging agents is mediated by low-capacity, high-affinity receptor systems, the requirements on specific activity are very stringent. The presence of radiolabeled low-affinity and unlabeled high-affinity impurities leads to a poor image and diagnostic errors due to high background and low effective specific activity.[6]

Synthesis of one of the potential diagnostic agents (17α,20E)-21-[^{125}I]iodo-19-norpregna-1,3,5(10),20-tetraene-3,17-diol (**4b**) was recently reported by Kabalka et al.[7] The use of [^{125}I]iodine monochloride as described in that procedure gave us a compound of very low specific activity, making the receptor studies extremely difficult. In addition, the reported method[7] failed to provide us with a pure compound even in its nonradioactive form.

For that reason we decided to alter the method to make it suitable for a no-carrier-added radioiodination. The synthetic route developed for **4b** was then applied to the synthesis of (17α,20E)-21-[^{125}I]iodo-11β-methoxy-19-norpregna-1,3,5(10),20-tetraene-3,17-diol (**4d**) from moxestrol (**1d**) as shown in Scheme I. Moxestrol (**1d**), which was synthesized from 17,17-(ethylenedioxy)estra-1,3,5(10)-triene-3,11β-diol,[8,9] exhibits high uterine biological activity due to decreased metabolism and virtually total absence of plasma binding.[10] The 17α-ethynyl group is responsible for decreased metabolism and reduced serum binding to sex-binding globulin (SBG) whereas the 11β-methoxy substituent is responsible for the decreased binding to serum SBG and albumin as compared to that of estradiol.[10] The iodinated derivative of moxestrol **4d** gave the highest uterine to blood ratio reported to date.[11]

Chemistry. Our approach (Scheme I) involved the acetylation of boronic acid derivates **2a,d** in order to protect the aromatic ring from iodination. An attempt to prepare the acetylated intermediate **2c** by reacting **1c** with catecholborane failed, due to removal of the acetyl group during the reaction. The acetylation of the intermediates **2a,d** with acetic anhydride/pyridine followed by purification provided the pure acetylated boronic acid derivaties **2c** and **2e** (85% and 75% yield, respectively).

Initial attempts to prepare nonradioactive **4a** in a pure form and reasonable yield by reacting **2c** with iodine monochloride followed by hydrolysis failed due to the presence of many impurities which were difficult to separate from the product. An alternate and successful method applied by us to produce both nonradioactive **3a–c** and hence **4a,c** used sodium iodide and Chloramine-T. When **2b,c,e** were allowed to react with sodium iodide and Chloramine-T in THF/phosphate buffer (pH 7.0) at room temperature, the yields after column chromatography were **3a** (50%), **3b** (25%), **3c** (40%). The ^{1}H NMR spectra of ^{127}I **3a,c** show doublets at δ 6.21 and 6.74 (**3a**) and at 6.32 and 6.74 (**3c**) corresponding to the protons of the vinyl group at C-17. The coupling constant (J = 14.4 and 15 Hz for **3a** and **3c**, respectively) confirmed Brown's[12] finding that the iodine was incorporated in E (trans) position. Deacetylation of **3a,c** to **4a** (90%), **4c** (77%) was carried out at room temperature using a mixture of sodium acetate, sodium carbonate, methanol, water, and ether. The

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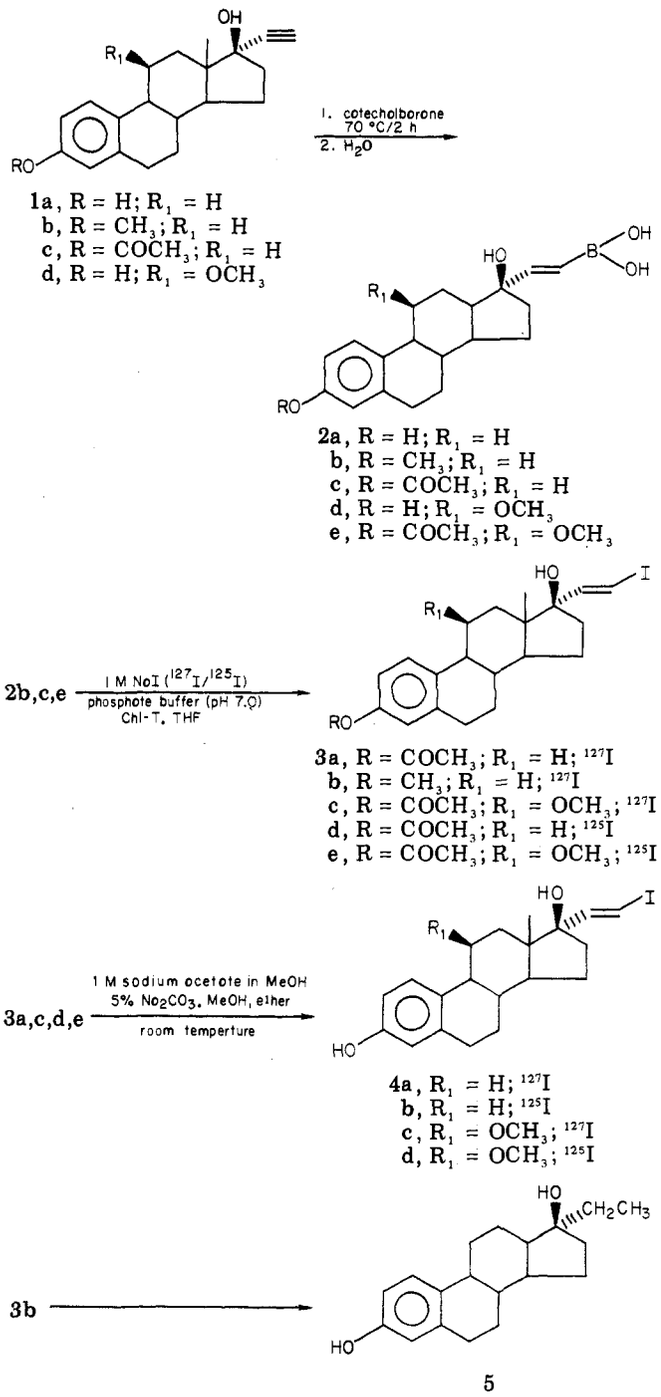
Table I. Distribution of **4b** and **4d** in Uterus and Plasma (Percent Dose/Gram of Tissue)

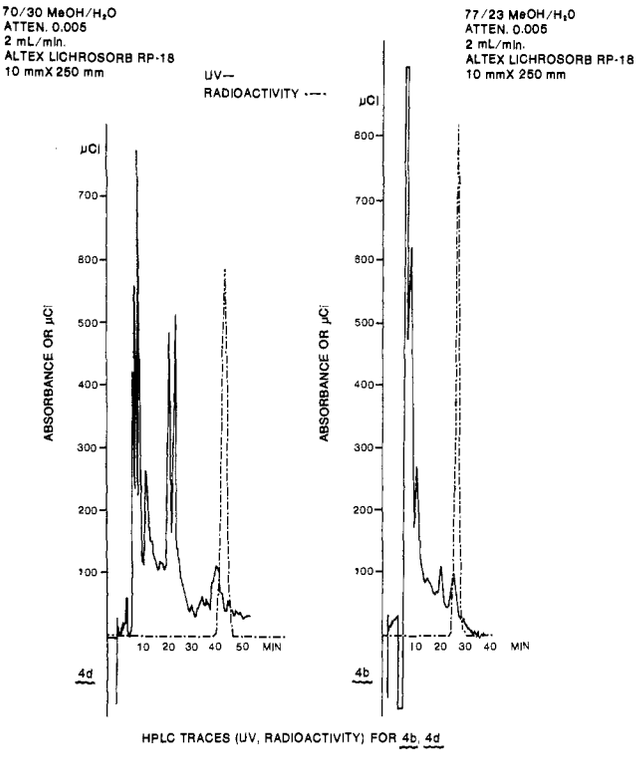
	4b				4d[b]			
time, h	uterus	plasma	uterus/ plasma	sp act., Ci/mmol	uterus	plasma	uterus/ plasma	sp act., Ci/mmol
1	5.89 ± 0.84[a]	1.09 ± 0.12	5.5	2200	8.18 ± 0.50	0.28 ± 0.02	29.0	1800
2	0.96 ± 0.11	0.192 ± 0.02	5.0	2152	17.2 ± 1.37	0.25 ± 0.03	68.3	572
3	1.10 ± 0.23	0.113 ± 0.02	9.7	2200	15.9 ± 1.31	0.24 ± 0.02	66.3	1888

[a] Mean of ≥5 rats ± SE. [b] Data from ref 19.

Scheme I



product **4a** crystallized with a solvent molecule of chloroform. To confirm the stereochemistry of **4a** and hence **4c** at C-17, the following set of reactions was carried out. (17α,20*E*)-21-Iodo-3-methoxy-19-norpregna-1,3,5(10),20-tetraen-17-ol (**3b**) was prepared from **2b** in a manner identical with the preparation of **3a**. Hydrogenation of **3b** in the presence of 10% Pd/C gave **5** (mp 84–86 °C, $[\alpha]^{25}_D$ +56° (*c* 0.01, $CHCl_3$)) identical with an authentic sample.[13]



HPLC TRACES (UV, RADIOACTIVITY) FOR 4b, 4d

Figure 1.

That finding confirms the absence of epimerization at C-17 during the hydroboration and iodination.

Radiochemistry. Initial attempts to prepare radioactive **4b** from **2a** using carrier-free iodine (^{125}I) monochloride generated in situ according to the procedure of Doran and Spar,[14] followed by hydrolysis, gave **4b** in very low yields. Similar results were obtained with use of elemental iodine prepared from NaI, KIO_3, and H_2SO_4.[15] The NaI and Chloramine-T procedure developed by us was therefore applied successfully to radioiodination. The major difference between the nonradioactive and the no-carrier-added synthesis was a large ratio of the boronic acid derivatives **2c** and **2e** to radioiodide. The radioactive derivatives **3d**, **3e** and **4b**, **4d** were cochromatographed with **3a**, **3c** and **4a**, **4c**, respectively, on TLC. We assume therefore they are identical with their respective nonradioactive counterparts. Figure 1 shows the UV and radioactivity traces for **4b** and **4d**. The yields were 50% for both **4b** and **4d**. Specific activities of **4b,d** were determined according to previously published methods[16] and ranged from 500 to 2200 Ci/mmol (theoretical value, 2200 Ci/mmol).

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Table II. Effect of Coinjection with Estradiol on the Distribution of 4d after 1 h[a]

tissue	% dose/g control[b]	% dose/g coinjected[c]
plasma	0.34 ± 0.03[d]	0.34 ± 0.02
uterus	8.87 ± 0.68	1.25 ± 0.10
liver	6.42 ± 1.05	4.52 ± 0.28
muscle	1.12 ± 0.11	1.29 ± 0.12

[a] Data from ref 19. [b] Controls were determined on the same day as the experimental coinjection study. [c] Fifty micrograms of "cold" estradiol and 5 μCi of 4d were administered simultaneously. [d] Mean of six rats ± SE.

Distribution Studies. The in vivo distribution of **4b** and **4d** in immature rats was determined at 1, 2, and 3 h following injection. The percent injected dose per gram was found for plasma and uterus (Table I).

Results and Discussion

The methodology developed for the synthesis of the 17α-iodovinyl estrogens in high purity and higher yields (overall yields for both **4a** and **4c** were about 24%) represents a significant improvement over existing methods. The generality of the procedure should make the synthesis of 17α-iodovinyl estrogens containing other substituents on the steroid nucleus an easier task. This method can also be applied to the synthesis of 17α-bromovinyl estrogens as is indicated by our preliminary studies. Further, this procedure meets the requirements for the preparation of clean, high specific activity products for the purposes of imaging with receptor-binding radiopharmaceuticals as is shown by our radiohalogenation studies. Finally, our preliminary in vivo distribution studies with rats indicate that **4d** would be superior to **4b** as a possible imaging agent. **4d** accumulates in high levels in target tissues and provides high target to nontarget ratios. Although in 1 h **4b** localizes to a similar extent in the uterus as **4d**, the activity washes out rapidly from the uterus (by 2 h only 16% of the activity observed at 1 h remains). In contrast, after 2 h the concentration of **4d** in the uterus increases by twofold and remains elevated at 3 h. These results are in agreement with in vitro kinetic studies with moxestrol.[17,18] Uterus to plasma ratios for **4d** are six- to tenfold higher than those observed for **4b**. This is due to the higher localization of **4d** in the uterus, except at 1 h in which case the injected dose/gram in the blood of **4b** is three- to fourfold higher than that observed for **4d**. The affinity constant of **4d** (6.8×10^9 M^{-1}) is about twofold less than that of [^{3}H]estradiol (1.65×10^{10} M^{-1}),[19] and its specificity is shown by in vivo competition studies with "cold" estradiol (Table II),[19] wherein it was found that estradiol blocked 95% of localization of **4d** in the uterus. The results suggest that **4d** has less nontarget binding, which would be an important asset for clinical use.

Experimental Section

Melting points were taken on a Fisher-Johns apparatus and are uncorrected. ^{1}H NMR spectra were recorded at 60 MHz or at 90 MHz on a Varian EM-360 and EM-390 or JEOL FX 60 spectrometer with Me_4Si as an internal standard. ^{13}C NMR spectra were determined at 15 MHz on a JEOL FX 60 spectrometer with Me_4Si as an internal standard. Mass (EI) spectra were obtained at 70 eV on a Du Pont Model 21-491 or Finnigan GC/MS 3300 spectrometer. Mass (CI) spectra were measured on a Finnigan 1015D spectrometer using NH_3 or NON_2. IR spectra were recorded on a Beckman IR-20A spectrophotometer. UV spectra were measured on a Beckman ACTA-CIII spectrophotometer. Specific rotations were obtained on a Perkin-Elmer 241 MC polarimeter using a 1-dm cell. HPLC analyses were carried out on a Altex 153 (labeled compounds) and Waters Associates ALC 202/6000 (unlabeled compounds) high-performance liquid chromatograph (HPLC). All preparative chromatographic separations were done by column chromatography using silica gel (Kieselgel 60, 0.040–0.060 mm, EM reagents, Germany) or using preparative TLC plates (silica gel GH, 250 μm, Analtech).

Moxestrol (**1d**) was synthesized according to the procedures outlined in ref 8 and 9 from 17,17-(ethylenedioxy)estra-1,3,5-(10)-triene-3,11β-diol, designated SC 16093, which was generously donated by G. D. Searle Labs., Chicago, IL. All other reagents were analytical grade and were used without further purification. Elemental analyses were performed by Galbraith Laboratories, Inc., Knoxville, TN. The results obtained are within ±0.4% of the theoretical values.

((17α,20*E*)-3,17-Dihydroxy-19-norpregna-1,3,5(10),20-tetraen-21-yl)boronic Acid (2a). A stirred mixture of 17α-ethynylestradiol **1a** (3 g, 10 mmol) and catecholborane (5 mL, 47 mmol) was heated at 70 °C for 2 h under nitrogen. Water (120 mL) was added dropwise and the mixture was stirred overnight. The crystalline product formed was collected by filtration, washed with water, and dried to give 3.2 g (94%) of **2a**: mp >300 °C; ^{1}H NMR (Me_2SO-d_6) δ 0.85 (s, 3 H, 18-CH_3), 1.00–3.00 (br m, steroid nucleus), 5.42 (d, J = 17 Hz, 1 H), 6.30–7.20 (m, 4 H, aromatic and CH=). Anal. ($C_{20}H_{27}O_4B$) C, H, B.

((17α,20*E*)-3-(Acetyloxy)-17-hydroxy-19-norpregna-1,3,5(10),20-tetraen-21-yl)boronic Acid (2c). A mixture of **2a** (1 g, 2.9 mmol), acetic anhydride (5 mL), and anhydrous pyridine (7 mL) was stirred at room temperature overnight. The mixture was poured into ice water. The precipitate formed was filtered, washed with water, and dried to give the crude acetate **2c** (quantitative yield), which was purified by column chromatography (silica gel, CH_2Cl_2–MeOH, 95:5) to afford pure **2c** (60%): mp 215–217 °C; IR (KBr) 3300, 2900, 1740, 1350, 1200, 1000 cm^{-1}; ^{1}H NMR (Me_2SO-d_6) δ 0.83 (s, 3 H, 18-CH_3), 1.00–3.00 (br m, steroid nucleus), 2.20 (s, 3 H, $COCH_3$), 5.38 (d, J = 17 Hz, 1 H, =$CHB(OH)_2$), 6.35–7.60 (m, 4 H, aromatic and CH=); MS (CI, NH_3), m/e 340 (MH^+ – 45). Anal. ($C_{22}H_{29}O_5B$) C, H, B.

(17α,20*E*)-21-Iodo-19-norpregna-1,3,5(10),20-tetraene-3,17-diol 3-Acetate (3a). To a mixture of **2c** (500 mg, 1.3 mmol), THF (5 mL), 0.066 M phosphate buffer (pH 7) (5 mL), and 1 M NaI (1.3 mL, 1.3 mmol) was added Chloramine-T (366 mg, 1.3 mmol), and the mixture was stirred for 2 h at room temperature. The reaction mixture was poured into water and extracted with ether. The ethereal extracts were washed with 5% sodium thiosulfate, dried over Na_2SO_4, and evaporated to give the crude product, which was purified by column chromatography (silica gel, petroleum ether–CH_2Cl_2, 7:3) to give **3a** (300 mg, 50%); mp 105–107 °C dec. Recrystallization from EtOH and water gave prisms: mp 107–109 °C dec; IR (KBr) 3500, 2920, 1745, 1370, 1200, 1010, 940 cm^{-1}, ^{1}H NMR ($CDCl_3$) δ 0.90 (s, 3 H, 18-CH_3), 1.00–3.00 (br, m, steroid nucleus), 2.26 (s, 3 H, $COCH_3$), 6.21 (d, J = 14.4 Hz, 1 H, =CHI), 6.74 (d, J = 14.4 Hz, 1 H, CH=), 6.50–7.40 (m, 3 H, aromatic); MS (CI, NH_3), m/e 467 (MH^+), 466 (M^+). Anal. ($C_{22}H_{27}O_3I$) C, H, I.

(17α,20*E*)-21-Iodo-19-norpregna-1,3,5(10),20-tetraene-3,17-diol (4a). A mixture of **3a** (200 mg, 0.43 mmol), 1 M methanolic solution of sodium acetate (1.3 mL), MeOH (0.7 mL), 5% Na_2CO_3 (0.7 mL), and ether (1.3 mL) was stirred for 3 h at room temperature. The reaction mixture was poured into water and extracted with ether. The ether extracts were washed with water, dried over Na_2SO_4, and evaporated to give a residue. Purification was effected by column chromatography (silica gel, petroleum ether–CH_2Cl_2, 6:4) to give **4a** (217 mg, 90%), which formed a crystalline adduct with $CHCl_3$: mp 120–125 °C dec; ^{1}H NMR (Me_2SO-d_6) δ 0.77 (s, 3 H, 18-CH_3) 1.00–3.00 (br m, steroid nucleus), 4.80 (s, 1 H, 17-OH), 6.18 (d, J = 14.4 Hz, 1 H, =CHI), 6.75 (d, J = 14.4 Hz, 1 H, CH=), 6.30–7.20 (m, 3 H, aromatic), 8.27 (s, 1 H, $CHCl_3$), 8.90 (s, phenolic OH). Anal. ($C_{21}H_{26}O_2Cl_3I \cdot H_2O$) C, H, Cl, I.

Recrystallization of the adduct from EtOH and water gave **4a**

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as needles: mp 113–115 °C dec; $[\alpha]^{25}_D$ +8.20° (c 0.009, EtOH); IR (KBr) 3500, 3280, 2900, 1610, 1450, 1230, 1000, 950 cm^{-1}; MS (EI), m/e 424 (M^+), 406, 391, 298, 254, 172, 159; ^1H NMR (Me_2SO-d_6) δ 0.77 (s, 3 H, 18-CH_3), 1.00–3.00 (br m, steroid nucleus), 6.10 (d, J = 14.4 Hz, 1 H, =CHI), 6.70 (d, J = 14.4 Hz, 1 H, CH=), 6.25–7.10 (m, 3 H, aromatic); UV (EtOH) λ_{max} 280 nm (ε 1780), 220 (14500), 207 (2520), ^{13}C NMR (Me_2SO-d_6) δ 14.0 (C-18), 22.9 (C-15), 26.0 (C-11), 27.1 (C-7), 29.2 (C-6), 32.3 (C-16), 35.4 (C-12), 39.3 (C-8), 43.2 (C-9), 46.7 (C-13), 48.6 (C-14), 74.3 (C-21), 85.8 (C-17), 112.7 (C-2), 114.9 (C-4), 125.9 (C-1), 130.3 (C-10), 137.1 (C-5), 152.0 (C-20), 154.9 (C-3). Anal. ($C_{20}H_{25}O_2I$·1.5H_2O) C, H, I.

((17α,20*E*)-17-Hydroxy-3-methoxy-19-norpregna-1,3,5(10),20-tetraen-21-yl)boronic Acid (2b). This compound was synthesized from mestranol (**1b**) according to the procedure of Kabalka et al.[7] **2b**: mp 150 °C; MS (CI, NH_3), m/e 311 (MH^+ − 45).

(17α,20*E*)-21-Iodo-3-methoxy-19-norpregna-1,3,5(10),20-tetraen-17-ol (3b). This compound was obtained from **2b** in a manner similar to that of **3a** from **2c**. **3b**: mp 102–105 °C dec; ^1H NMR ($CDCl_3$) δ 0.90 (s, 3 H, 18-CH_3), 1.00–3.00 (br m, steroid nucleus), 3.80 (s, 3 H, OCH_3), 6.25 (d, J = 14.4 Hz, 1 H, =CHI), 6.78 (d, J = 14.4 Hz, 1 H, CH=), 6.50–7.30 (m, 3 H, aromatic).

(17α)-3-Methoxy-19-norpregna-1,3,5(10)-trien-17-ol (5). A mixture of **3b** (150 mg, 0.34 mmol), 10% Pd–C (15 mg), 5% Na_2CO_3 (0.5 mL), and EtOH (20 mL) was shaken under H_2 (20 kg/cm^2) pressure for 4 h. Catalyst was removed by filtration and washed with EtOH. The filtrate and washings were combined and evaporated to give a residue, which was taken up in ether and washed with water. The ether extract was dried over Na_2SO_4 and evaporated to give a residue. The product was purified by column chromatography (silica gel, petroleum ether–CH_2Cl_2, 8:2) to afford a crystalline residue (86 mg, 81%), which was recrystallized from EtOH and water, giving needles: mp 84–86 °C (lit.[13] mp 86–87 °C); $[\alpha]^{25}_D$ +56.0° (c 0.01, $CHCl_3$) (lit.[13] $[\alpha]^{25}_D$ +55.6° (c 0.01, $CHCl_3$)); MS (EI), m/e 314 (M^+), 242, 240, 212, 200, 175, 159, 149, 143; ^1H NMR ($CDCl_3$) δ 0.90 (s, 3 H, 18-CH_3), 1.02 (t, 3 H, CH_2CH_3), 1.20–3.10 (several m, 17 H, steroid nucleus and CH_2CH_3), 3.73 (s, 3 H, OCH_3), 6.68 (m, 2 H, aromatic), 7.20 (d, 1 H, aromatic). Anal. ($C_{21}H_{30}O_2$·1.5H_2O) C, H.

((17α,20*E*)-3,17-Dihydroxy-11β-methoxy-19-norpregna-1,3,5(10),20-tetraen-21-yl)boronic Acid (2d). A stirred mixture of **1d** (2 g, 3 mM) and catecholborane (7 mL, 52.5 mM) was heated at 70 °C for 2 h under nitrogen. Water (100 mL) was added to this mixture after cooling. The resulting mixture was stirred overnight and the product isolated by filtration. This was washed with water and dried to give impure **2d**, which was purified by column chromatography (300 g, silica gel, hexane–EtOAc gradient) to give pure **2d** (0.70 g, 60%): mp 290 °C dec; ^1H NMR (Me_2SO-d_6) δ 1.00 (s, 3 H, 18-CH_3), 3.13 (s, 3 H, 11β-OCH_3), 4.10 (m, 1 H, 11α-H), 5.4 (d, J = 17 Hz, 1 H, =$CHB(OH)_2$) 6.40–7.00 (m, 4 H, aromatic and CH=); IR (KBr) 3380, 1640, 1510, 1460, 1270, 1250, 1080, 1030 cm^{-1}. Anal. ($C_{21}H_{29}O_5B$·1.5H_2O) C, H, B.

((17α,20*E*)-3-(Acetyloxy)-11β-methoxy-17-hydroxy-19-norpregna-1,3,5(10),20-tetraen-21-yl)boronic Acid (2e). A mixture of **2d** (1 g, 2.7 mmol), acetic anhydride (5 mL), and anhydrous pyridine was stirred overnight at room temperature and then poured into ice–water. Product isolation (ether, Na_2SO_4) yielded 1.0 g of impure **2e**, which upon purification by column chromatography (150 g, silica gel, hexane–EtOAc gradient) gave pure **2e** (800 mg, 72%): mp 248–250 °C; ^1H NMR (Me_2SO-d_6) δ 0.86 (s, 3 H, 18-CH_3), 2.28 (s, 3 H, $COCH_3$), 5.45 (d, J = 17 Hz, 1 H, =$CHB(OH)_2$), 6.75–7.40 (m, 4 H, aromatic and CH=); IR (KBr) 3400, 1735, 1625, 1500, 1370, 1220, 1080, 1020 cm^{-1}. Anal. ($C_{23}H_{31}O_6B$) C, H, B.

(17α,20*E*)-21-Iodo-11β-methoxy-19-norpregna-1,3,5(10),20-tetraene-3,17-diol 3-Acetate (3c). To a mixture of **2e** (200 mg, 0.48 mmol), THF (1.5 mL), phosphate buffer (0.066 M, pH 7, 1.5 mL), and 1 M sodium iodide (0.48 mL, 0.48 mmol) was added Chloramine-T (136 mg, 2.3 mM). This was stirred for 1.5 h at room temperature and then poured into water and extracted with ether. The combined extracts were washed with 5% sodium thiosulfate and dried over Na_2SO_4, and the ether was evaporated off to give the crude product, which was purified by preparative TLC (toluene–EtOAc, 8:2), yielding pure **3c** (100 mg, 42%): mp 162–164 °C dec; ^1H NMR ($CDCl_3$) δ 1.10 (s, 3 H, 18-CH_3), 2.26 (s, 3 H, $COCH_3$), 3.28 (s, 3 H, 11β-OCH_3), 4.15 (m, 1 H, 11α-H), 6.32 (d, J = 15 Hz, 1 H, =CHI), 6.74 (d, J = 15 Hz, 1 H, CH=), 6.80–7.15 (m, 3 H, aromatic); IR (KBr) 3490, 3050, 1740, 1600, 1495, 1450, 1375, 1235, 1210, 1090, 1030, 950 cm^{-1}. Anal. ($C_{23}H_{29}O_4I$) C, H, I.

(17α,20*E*)-21-Iodo-11β-methoxy-19-norpregna-1,3,5(10),20-tetraene-3,17-diol (4c). A mixture of **3c** (50 mg, 0.1 mmol), 1 M methanolic solution of NaOAc (0.4 mL), MeOH (0.2 mL), 5% Na_2CO_3 (0.2 mL), and ether (0.4 mL) was stirred for 1.5 h at room temperature. The reaction mixture was poured into water and extracted with ether. The combined ether extracts were washed with water and dried over Na_2SO_4. Evaporation of the solvent yielded the crude product, which was purified by preparative TLC (petroleum ether–MeOH–EtOAc, 30:2.5:2.5) to give pure **4c** (35 mg, 77%): mp 168–170 °C dec; ^1H NMR (Me_2SO-d_6) δ 0.98 (s, 3 H, 18-CH_3), 3.13 (s, 3 H, 11β-OCH_3), 4.11 (m, 1 H, 11α-H), 6.22 (d, J = 15 Hz, 1 H, =CHI), 6.75 (d, J = 15 Hz, 1 H, CH=), 6.45–6.98 (m, 3 H, aromatic), 8.90 (s, 1 H, PhOH); IR (KBr) 3360, 3030, 1625, 1595, 1510, 1455, 1360, 1300, 1255, 1080, 1030 cm^{-1}; MS (EI) m/e (relative intensity) 454 (M^+, 10). Anal. ($C_{21}H_{27}O_3I$) C, H, I.

(17α,20*E*)-21-[^{125}I]Iodo-19-norpregna-1,3,5(10),20-tetraene-3,17-diol 3-Acetate (3d) and (17α,20*E*)-21-[^{125}I]Iodo-11β-methoxy-19-norpregna-1,3,5(10),20-tetraene-3,17-diol 3-Acetate (3e). To a mixture of sodium [^{125}I]iodide (6 mCi, 0.0027 μmol), 0.06 M phosphate buffer (pH 7) (35 μL), THF (95 μL), and **2c** (5 mg, 0.013 mmol) or **2e** (5.4 mg, 0.013 mmol) was added Chloramine-T (1.13 mg, 0.004 mmol). The mixture was stirred at room temperature for 1 h (**2c**) or 34 °C for 30 min (**2e**) and then injected on an HPLC system (column, Lichrosorb RP-18, 10 mm × 250 mm; mobile phase, MeOH–water, 77:23, v/v (**2c**) or 70:30, v/v (**2e**); flow rate, 2 mL/min). Fractions were collected via a fraction collector (1 min/tube). The ones containing the product **3d** (50–54 min) or **3e** (33–36 min) were combined and evaporated, giving the following R_f values on TLC (silica gel): **3d**, 0.42 (MeOH–CH_2Cl_2, 1:99), 0.56 (petroleum ether–EtOAc–MeOH, 25:3.5:3.5), 0.58 (toluene–ethyl acetate, 8:2) and **3e**, 0.47 (MeOH–CH_2Cl_2, 2:98), 0.24 (petroleum ether–EtOAc–MeOH, 30:2.5:2.5), 0.35 (toluene–EtOAc, 8:2); yields, 50% (**3d**), 60% (**3e**).

(17α,20*E*)-21-[^{125}I]Iodo-19-norpregna-1,3,5(10),20-tetraene-3,17-diol (4b) and (17α,20*E*)-21-[^{125}I]Iodo-11β-methoxy-19-norpregna-1,3,5(10),20-tetraene-3,17-diol (4d). A mixture of **3d** (3.4 mCi) or **3e** (3.6 mCi), 1 M methanolic solution of NaOAc (0.4 mL), 5% Na_2CO_3 (0.2 mL), ether (0.4 mL), and MeOH (0.2 mL) was stirred for 1 h at room temperature, diluted with water (0.5 mL), and extracted with ether (4 mL). The ether extracts were washed with water (0.5 mL) and evaporated. The residue was taken up in 200 μL of the HPLC solvent to be used (MeOH–Water = 77:23, v/v for **3d** or 70:30, v/v for **3e**) and injected as before (column, Lichrosorb RP-18, 10 mm × 250 mm; flow rate, 2 mL/min). Fractions were collected (1 min/tube) and the ones containing **4b** (29–32 min) or **4d** (41–45 min) were combined and evaporated, giving the following R_f values on TLC (silica gel): **4b**, 0.20 (MeOH–CH_2Cl_2, 1:99), 0.42 (petroleum ether–EtOAc–MeOH, 25:3.5:3.5), 0.45 (toluene–ethyl acetate, 8:2) and **4d**, 0.20 (MeOH–CH_2Cl_2, 2:98), 0.14 (petroleum ether–EtOAc–MeOH, 30:2.5:2.5), 0.18 (toluene–ethyl acetate, 8:2). Radioactive products **4b** and **4d** (80% yield) were identical on TLC and HPLC with the nonradioactive products (**4a** and **4c**, respectively). The overall yields (both reactions) for the radioactive products **4b** and **4d** were on the order of 50% with specific activities from 500 to 2200 Ci/mmol, which were measured by radioreceptor technique.[16]

In Vivo Distribution Studies. Three to five microcurics of the radioligand **4d** or **4b** was injected via the femoral vein of female Sprague–Dawley rats (20–25 days old), and the animals were sacrificed by cervical dislocation at 1, 2, and 3 h. Samples of uterus and blood were removed. Tissues blotted free of excess blood and 10–30-mg samples were weighed. The heparinized blood samples were centrifuged to separate the plasma and 20-μL samples were removed for counting. Samples were counted on a Searle auto-gamma counter and dpm were determined by the absolute counting method.

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Registry No. **1a**, 57-63-6; **1b**, 72-33-3; **1d**, 34816-55-2; **2a**, 91176-86-2; **2b**, 81844-94-2; **2c**, 91085-37-9; **2d**, 91085-38-0; **2e**, 91085-39-1; **3a**, 91085-40-4; **3b**, 78479-31-9; **3c**, 91085-41-5; **3d**, 91085-42-6; **3e**, 91085-43-7; **4a**, 91085-44-8; **4b**, 82123-96-4; **4c**, 91085-45-9; **4d**, 90857-55-9; **5**, 2553-34-6; catecholborane, 274-07-7.

Synthesis and β-Adrenergic Blocking Activity of New Aliphatic and Alicyclic Oxime Ethers†

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We describe the synthesis and pharmacological properties of two new series of aliphatic and alicyclic β-adrenergic blockers, most of them containing a cyclopropyl ring. They belong either to 2-hydroxy-3-(*tert*-butylamino)propyl ether A or 2-hydroxy-3-*tert*-(butylamino)propyl ketoxime ether B derivatives. The *O*-[2-hydroxy-3-(*tert*-butylamino)propyl] dicyclopropyl ketoxime **5** exhibited a β-adrenergic antagonist activity comparable to that of propranolol. It was found that ketoxime ethers B generally showed higher potency than the corresponding ethers A. We confirm that the presence of an aromatic nucleus is not crucial for the β-adrenergic activity. Structure–activity relationships among these series are discussed.

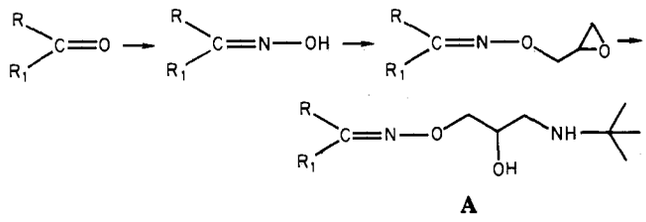
In a previous paper,[1] we have shown that some aliphatic oxime ethers could exhibit interesting β-adrenergic blocking activities. This result encouraged us to pursue our efforts to increase the activity of these molecules and to clarify structure–activity relationships within this series. In this paper we report the synthesis and the pharmacology of these new β-blockers, most of which contain a cyclopropyl ring.

Chemistry. Scheme I shows the classical synthetic route starting with the appropriate ketone oximes for the preparation of compounds **1–10** (Table I). Compounds **11–18** (Table II) were similarly prepared from the sodium salt of the corresponding alcohol (Scheme II).

Most of the ketones used in this work are commercially available or can be obtained by known procedures. Several attempts to prepare ketone **19** were unsuccessful. Thus, the action of MeMgI on crotonyl chloride led to a complex mixture of 1,1-dimethylbutadiene, methyl crotyl ketone, dimethylcrotylcarbinol, methyldicrotylcarbinol, and the desired allyl methyl ketone as evidenced by NMR, MS, and GLC. Similarly, the action of 3-butenyltrimethylsilane on MeCOCl, using $TiCl_4$ as a catalyst,[2] afforded a mixture of β-chloro ketone, 3-butenyl ketone, and cyclopropylacetone from which the last compound could not be isolated satisfactorily. Finally, the ketone **19** was obtained by using the procedure described in Scheme III. The reaction of crotonyl chloride with EtOH in the presence of $N(Et)_3$ gave the rearranged ethyl vinylacetate;[3] this ester was submitted to a Simmons–Smith reaction by using diiodomethane and Zn/Cu couple.[4] The direct action of MeLi (2 equiv) on cyclopropylacetic acid gave a poor yield of I (ca. 10%). Similar findings were mentioned in the literature[5] for lower aliphatic acids. As an alternative, the corresponding acid chloride was reacted with dimethylcadmium[6] to give the cyclopropylacetone with an overall yield of approximately 36%.

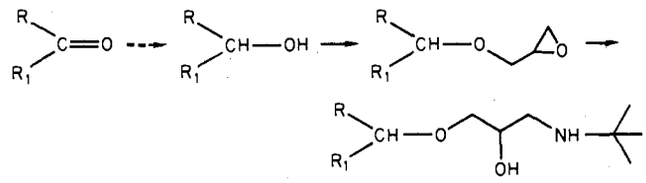
$NaBH_3CN$ reduction of the oxime ether **3** gave **8**.

Scheme I



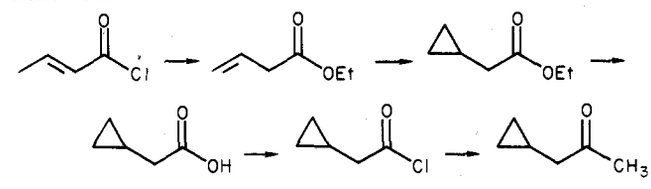
A

Scheme II



B

Scheme III



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