Chemically Reactive Estrogens: Synthesis and Estrogen Receptor Interactions of Hexestrol Ether Derivatives and 4-Substituted Deoxyhexestrol Derivatives Bearing Alkylating Functions

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A series of chemically reactive derivatives of the nonsteroidal estrogen hexestrol have been synthesized as potential affinity labels for the estrogen receptor or as cytotoxic agents with selective activity against receptor-containing cells. These compounds are hexestrol ethers with halo ketone, halohydrin, or epoxide functions or 4-substituted deoxyhexestrols with halo ketone, benzyl halide, nitro, azide, sulfonyl fluoride, or sulfonyl azide groups. The alkylating activity of the electrophilic derivatives was measured using the colorimetric reagent nitrobenzylpyridine, the bromo derivatives being considerably more reactive than the chloro ones. Their reversible binding to the lamb uterine estrogen receptor was measured by competitive binding assays, and their irreversible reaction with receptor was measured by exchange assays that determine the rate and extent of receptor inactivation. In general, monoetherification of hexestrol or substitution of deoxyhexestrol produces compounds with relatively low affinity for the estrogen receptor (0.3–10% that of estradiol). Most of the electrophilic derivatives are rapid and effective inactivators of receptor (24–70% inactivation within 0.5–5 h at 25 °C). Of the photosensitive derivatives, 4-azidodeoxyhexestrol appears to be the most efficient receptor inactivator (49%). The high reactivity of these compounds toward the estrogen receptor and the lack of interference by their reaction with other cellular nucleophiles suggest that these compounds may be useful as affinity-labeling agents or as selective cytotoxic agents in intact systems.

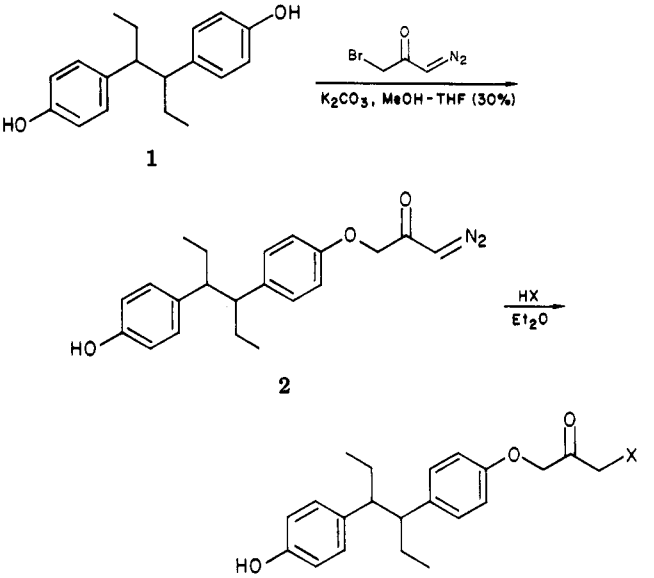
The action of estrogens in target tissues is believed to be mediated by specific cellular binding proteins called estrogen receptors.[1] The molecular interactions that take place between estrogens and these estrogen receptors are complicated, involving several complexes with different physical properties, interacting in different subcellular compartments.

We have been interested in developing chemically reactive estrogen derivatives and analogues that can be used as affinity labels to study the physical and chemical properties of the estrogen receptor and to investigate some of the dynamic features of estrogen receptor interactions.[2] In earlier work, we have described the synthesis of several simple derivatives of estradiol and hexestrol that are photochemically reactive,[3] and we have studied their interaction with the uterine estrogen receptor from immature rat and lamb.[4,5] These earlier contributions culminated in a report in which we described the photoaffinity labeling of the ovine uterine estrogen receptor using *o*-azidohexestrol.[6] Despite our successful use of *o*-azidohexestrol to label the receptor, the behavior of this reagent in terms of binding selectivity and reaction efficiency was not ideal; thus, we have continued our search for affinity-labeling reagents with improved characteristics for estrogen receptor labeling.

In this report, we describe the preparation of a series of hexestrol analogues that bear various alkylating functions on ether groups or on 4-substituted deoxyhexestrols. We have quantitated the alkylating activity of these compounds by chemical assay using the chromogenic agent nitrobenzylpyridine, and we have studied their reversible and irreversible interaction (in nonradiolabeled form) with the estrogen receptor by indirect competitive assays. Many of these compounds show sufficient reactivity toward the estrogen receptor to indicate that they may be useful in in vivo studies either as affinity-labeling agents or as selective cytotoxic agents.[7,8]

Results

Synthesis of Hexestrol Ethers. The conversion of hexestrol (1) to the 3-halo-2-oxopropyl ethers (**3b,c**) was



(1) For a recent review see: Katzenellenbogen, B. S., *Annu. Rev. Physiol.* **1980**, *42*, 17. Gorski, J.; Gannon, E., *Ibid.* **1976**, *38*, 425.
(2) For reviews of earlier work see: (a) Katzenellenbogen, J. A. *Biochem. Actions Horm.* **1972**, *4*, 1. (b) Katzenellenbogen, J. A. *Fed. Proc. Fed. Am. Soc. Exp. Biol.* **1978**, *37*, 174. (c) Katzenellenbogen, J. A.; Johnson, H. J., Jr.; Myers, H. N.; Carlson, K. E.; Kempton, R. J. In "Bioorganic Chemistry"; van Tamelen, E. E., Ed.; Academic Press: New York, 1978, Vol IV, p 207.
(3) Katzenellenbogen, J. A.; Myers, H. N.; Johnson, H. J., Jr. *J. Org. Chem.* **1973**, *38*, 3525.
(4) Katzenellenbogen, J. A.; Johnson, H. J., Jr.; and Myers, H. N. *Biochemistry* **1973**, *12*, 4085.
(5) Katzenellenbogen, J. A.; Johnson, H. J., Jr.; Carlson, K. E.; Myers, H. N. *Biochemistry* **1974**, *13*, 2986.
(6) (a) Katzenellenbogen, J. A.; Myers, H. N.; Johnson, H. J., Jr.; Kempton, R. J.; Carlson, K. E. *Biochemistry* **1977**, *16*, 1964. (b) Katzenellenbogen, J. A.; Carlson, K. E.; Johnson, H. J., Jr.; Myers, H. N. *Biochemistry* **1977**, *16*, 1970.
(7) Katzenellenbogen, J. A. *Cancer Treat. Rep.* **1979**, *62*, 1243.
(8) Raus, J.; Martens, H.; Leclerq, G. "Cytotoxic Estrogens in Hormone Responsive Tumors"; Academic Press: London, 1980.

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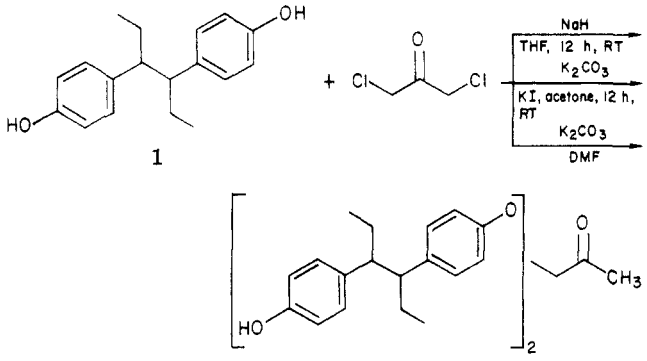
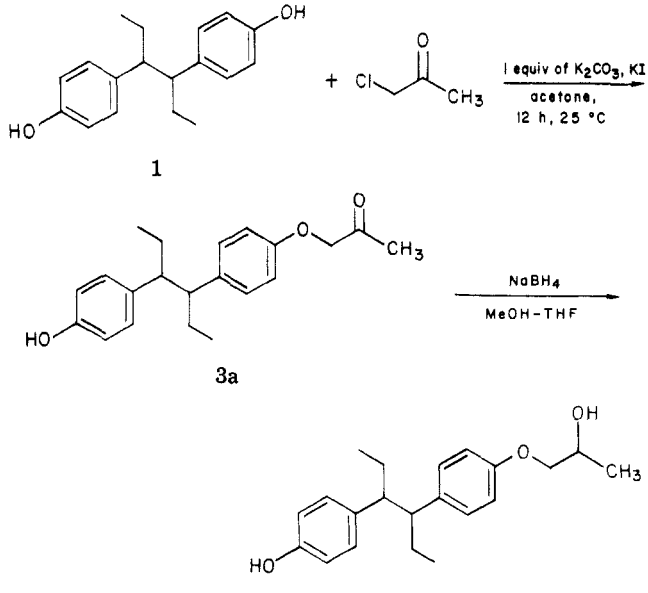
accomplished via acid decomposition of hexestrol 3-diazo-2-oxopropyl ether (**2**).[6a] Prior to our development of this sequence, we attempted to prepare the chloroacetonyl ether by direct treatment of hexestrol with 1,3-dichloroacetone. These trials afforded only starting material and the 1,1-bis(aryloxy) derivative **4**.[9] In contrast, alkylation



1

4

of hexestrol with chloroacetone did give the monosubstituted acetonylhexestrol (**3a**), which served as the precursor to the hydroxypropyl hexestrol ether (**5a**).

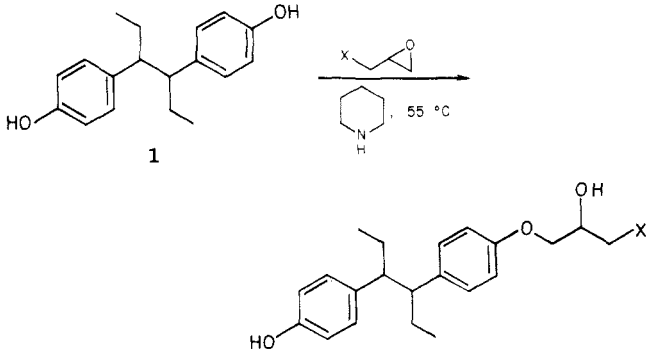


1

3a

5a

Synthesis of the halohydrin ethers **5b** and **5c** was conveniently performed in one step with an excess of epihalohydrin reagent according to the method of Stephenson.[10] Hexestrol epoxypropyl ether (**7a**) was obtained by

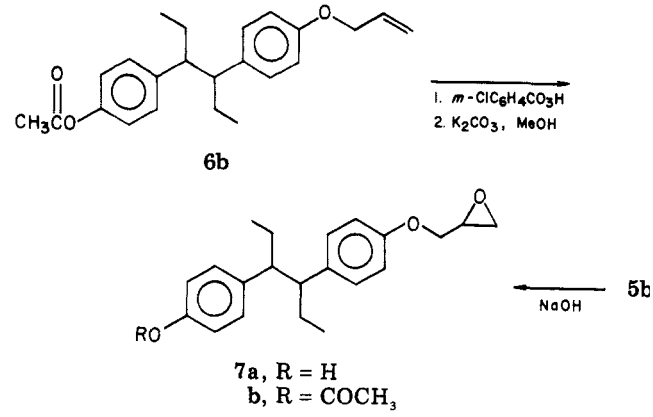


1

5b, X = Cl (40%)
c, X = Br (34%)

(9) Hill, J. *J. Chem. Soc. C* **1970**, 462.

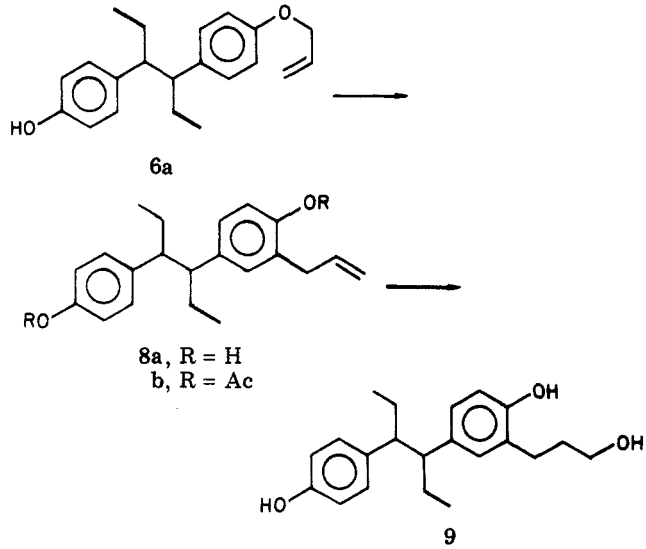
two routes: by epoxidation of allyl hexestrol acetate (**6b**) followed by deacetylation, or by base-induced epoxide formation from the chlorohydrin **5b**.



6b

7a, R = H
b, R = $COCH_3$

The Claisen rearrangement product **8a** resulted from



6a

8a, R = H
b, R = Ac

9

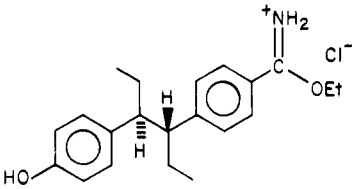
thermolysis of hexestrol allyl ether (**6a**) in diphenyl ether. This procedure represents a substantial improvement of similar rearrangements using *N,N*-diethylaniline as solvent.[11] *o*-Allylhexestrol (**8a**) was hydrated by hydroboration–oxidation to give the alcohol **9**.

Synthesis of 4-Substituted Deoxyhexestrols. Approach via 4-Bromodeoxyhexestrol (15) and 4-Cyanodeoxyhexestrol (17a). The approach utilized by Wilson[12] for the synthesis of 4-deoxyhexestrol was adapted to the synthesis of 4-bromo-4-deoxyhexestrol methyl ether (**15**, Scheme I). Acylation of anisole with *p*-bromophenylacetyl chloride and stannic chloride gave *p*-methoxy-*p'*-bromodeoxybenzoin (**10**), which was alkylated cleanly with sodium hydride and ethyl iodide. Addition of ethylmagnesium halide to **11** followed by dehydration of the benzylic alcohol **12** gave a mixture of the isomeric stilbenes (**13**) containing some of the 2-hexene isomers **14**. Catalytic hydrogenation of this olefin mixture proceeded very sluggishly and was accompanied by significant debromination. However, a mixture of only the *E* and *Z* 2-hexene isomers (**14**), obtained by the reaction of ketone **11** with ethylidene triphenylphosphorane, was reduced more easily to two diastereomeric 4-bromo-4-deoxy-

(10) (a) Stephenson, O. *J. Chem. Soc.* **1954**, 1571. (b) Bradley, W.; Forrest, J.; Stephenson, O. *J. Chem. Soc.* **1951**, 1589.
(11) Kaiser, E.; Svarz, J. J. *J. Am. Chem. Soc.* **1946**, *68*, 636.
(12) Wilson, J. W.; Burger, A.; Turnbull, L. B.; Peters, M.; Milnes, F. *J. Org. Chem.* **1953**, *18*, 96.

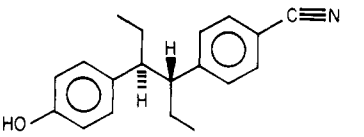
hexestrol methyl ethers (**15**). While these isomers were not readily separable by preparative column chromatography, the isomer composition (erythro/threo, 4:6) was evident by ^1H NMR and GLC.

Introduction of a carbon substituent on the hexestrol ring was achieved by treatment of the methyl ether diastereomers (**15**) with cuprous cyanide in refluxing *N*-methyl-2-pyrrolidinone. The corresponding phenols (**17**) were cleanly separated by chromatography, and their stereochemistry was established by hydrolysis to the corresponding acids, both of which are known.[12] Cyanodeoxyhexestrol (**17a**) gives the ethyl imidate hydrochloride



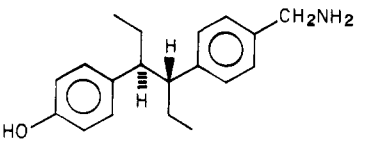
19

HCl | EtOH, 75%



17a

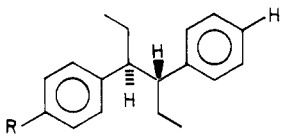
H_2, Raney Nickel 70%



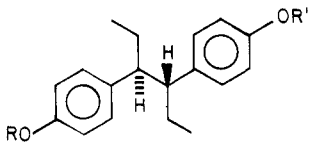
20

19 upon treatment with anhydrous hydrochloric acid in ethanol. The aminomethyl compound **20** can be prepared by reduction of **17a** with Raney nickel W-2.

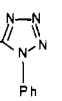
Approach via Deoxyhexestrol. Deoxyhexestrol (**21a**)



21a, R = OH
b, R = H



1, R′, R = H
22a, R = H; R′ = SO_2CH_3
b, R, R′ = SO_2CH_3

c, R = H; R′ = 

d, R, R′ = 

has been prepared in the past by two multistep routes that produce a relatively unfavorable ratio of diastereomers.[12,13] We have found it more convenient to prepare monodeoxyhexestrol by reduction of the hexestrol methanesulfonate (**22a**)[14] or 1-phenyl-1*H*-tetrazole derivatives (**22c**),[15] and, while this approach is not regioselective, its directness makes it quite practical.

Introduction of a substituent in the 4 position of 4-deoxyhexestrol through electrophilic aromatic substitution provides an interesting challenge in regioselectivity: This position is activated by the *p*-(3-hexyl) group and is not sterically hindered; however, the other ring is more electron rich, due to the phenolic hydroxyl substituent, but is 1,4 disubstituted, providing positions ortho to another substituent as the only activated sites for reaction. Thus, nitration of deoxyhexestrol methyl ether (**23a**) gives ortho substitution, but nitration of the deactivated toluenesulfonate (**23b**) or *p*-nitrobenzenesulfonate (**23c**) gives only the 4-substituted products (**24b** and **24c**, respectively, Scheme II). These sulfonate esters can then be hydrolyzed easily to 4-nitro-4-deoxyhexestrol (**24a**). This compound is reduced to the corresponding amino compound **25a**, which can be converted further into 4-azido-4-deoxyhexestrol (**25b**) by diazotization followed by quenching with azide ion.

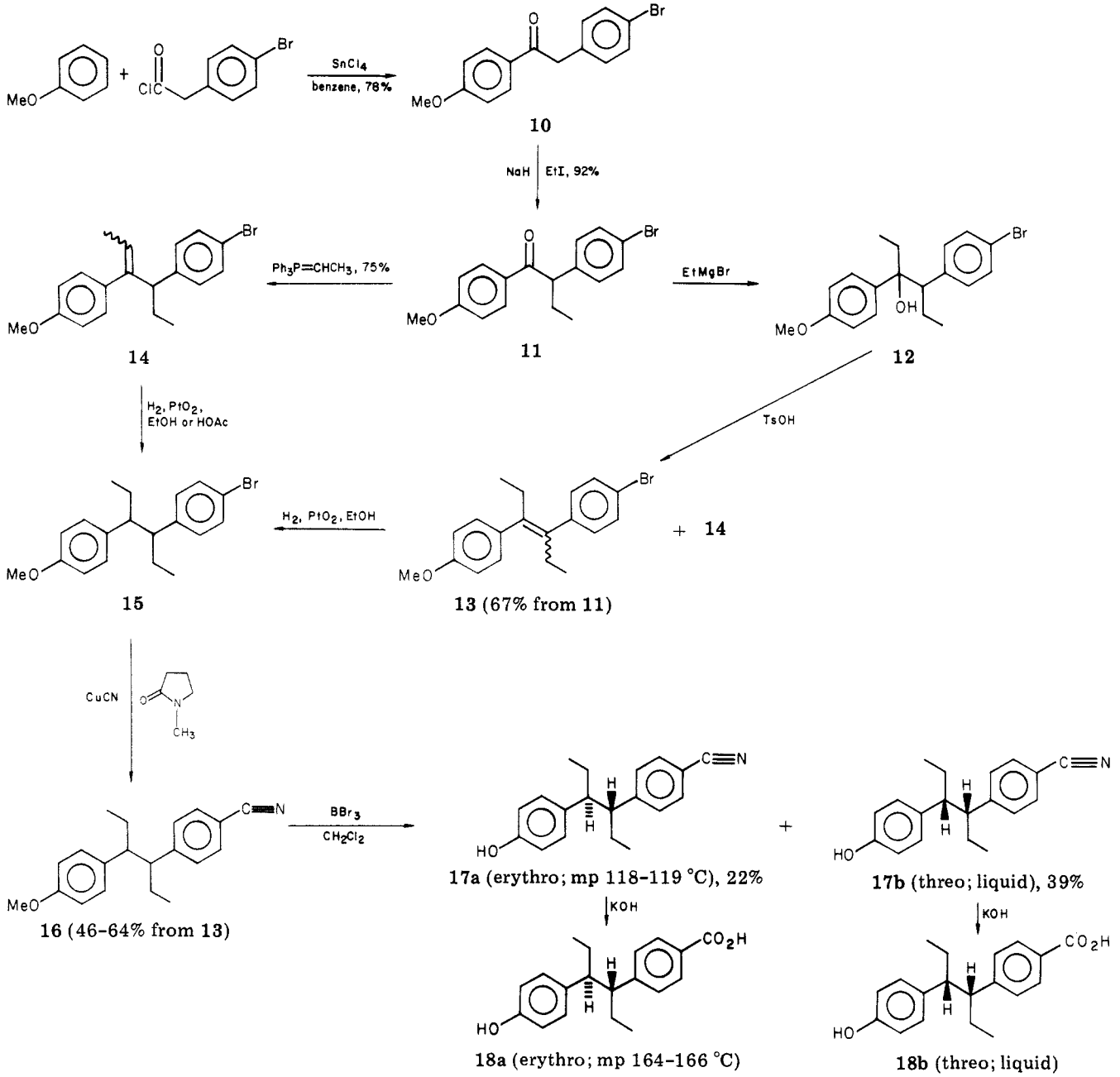
The fluorosulfonate **26a** was prepared by fluorosulfonation of deoxyhexestrol trifluoroacetate (**23d**), with sulfonation proceeding exclusively at the 4 position; mild hydrolysis furnishes the free phenol **26b**. The related chlorosulfonate **26d** was prepared in an analogous fashion and was converted into the sulfonyl azide **26e** by treatment with excess sodium azide.

Chloroacetylation of the methyl ether (**23a**) or toluenesulfonate (**23b**) is not regioselective (Scheme III); however, acylation of the corresponding benzoate (**23e**) proceeded cleanly to give only the para-substituted products **27b** and **27c**, which were hydrolyzed to the free phenols by base and acid, respectively. Bromoacetyldeoxyhexestrol (**27f**) was prepared from **27d** by heating with cupric bromide. Haloform cleavage gave the benzoate **29** which was hydrolyzed to the known acid **18a**,[12] confirming both the structure and stereochemistry of the compounds in this series. The benzoate **29** was reduced to the benzylic alcohol **31a**, a compound that was also produced by carboxylation[16] of **23e**, followed by hydride reduction of the catechol half ester **30**. The benzyl bromide **31b** was formed from **31a** by treatment with HBr, and the benzyl chloride **31c** was formed by treatment with HCl. Treatment of **31a** with hydrogen iodide in ethanol gave the ethyl ether derivative **31d**.

Alkylating Activity. Reaction of Electrophilic Hexestrol Derivatives with Nitrobenzylpyridine. The electrophilic (S_N2) reactivity of various alkylating agents can be measured conveniently by reaction with the chromogenic agent 4-(4-nitrobenzyl)pyridine.[17] The pyridinium salt[18] that results from the alkylation is converted

(13) Wawzonek, S. *J. Am. Chem. Soc.* **1946**, *68*, 1157.
(14) Clauss, K.; Jensen, H. *Angew, Chem., Int. Ed. Engl.* **1973**, *12*, 918.
(15) Musliner, W. J.; Gates, J. W. *Org. Synth.* **1971**, *51*, 82.
(16) (a) Gross, H.; Rieche, A.; Hoft, E. *Chem. Ber.* **1961**, *94*, 544. (b) Gross, H.; Rusche, J.; Mirsch, M. *Chem. Ber.* **1963**, *96*, 1382.
(17) (a) Preussmann, R.; Schneider, H.; Epple, F. *Arzneim.-Forsch* **1969**, *19*, 1059. (b) Swaisland, A. J.; Grover, P. L.; Sims, P. *Biochem. Pharmacol.* **1973**, *22*, 1547. (c) Hammock, L. G.; Hammock, B. D.; Casida, J. E. *Bull. Environ. Contam. Toxicol.* **1974**, *12*, 759.

Scheme I. Functionalization of Hexestrol via Cyanodeoxyhexestrol



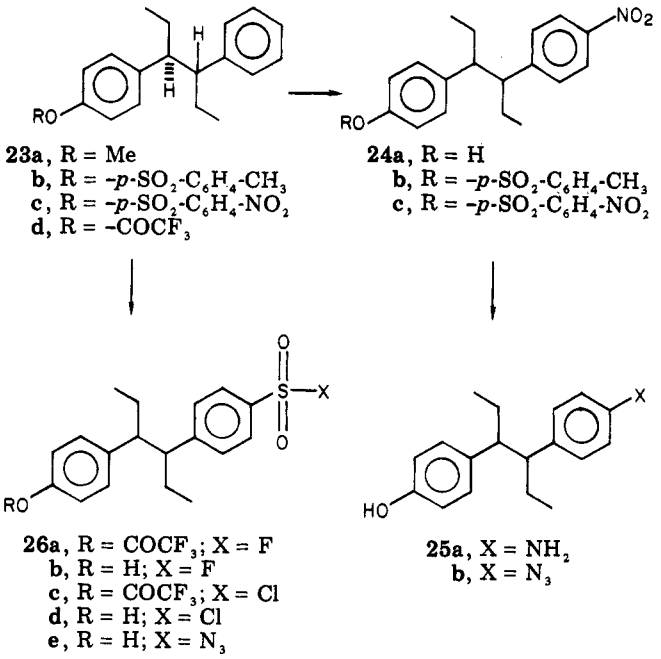
into a blue-violet dye (λ_{max} = 560 nm, ϵ = 37 000 in acetone–10% water) when treated with base. Figure 1 shows the time course of reaction of the reactive hexestrol ethers (panel A) and deoxyhexestrol derivatives (panel B) with nitrobenzylpyridine in aqueous ethylene glycol at 37 °C, pH 7.32. The second-order rate constants for this reaction are given in Table I, together with the half-times for the observed pseudo-first-order rate constant and the extent of reaction .

Of the hexestrol ethers (Figure 1A, Table I), the bromo ketone **3c** reacts most rapidly, but the reaction ceases at only a fraction of the expected maximum level of color generation. Since the pyridinium salt appears to be stable for 24 h (maximum color does not decrease), the lack of complete reaction suggests that solvolysis is competing with the alkylation of nitrobenzylpyridine. The chloro ketone **3b**, which is much less reactive toward the chromogenic agent, still appears to solvolyze extensively. The glycidyl ether **7a** is a moderately reactive compound, and it reacts to 55% completion. The bromohydrin **5c** is somewhat less reactive, and the chlorohydrin **5b** has a reactivity comparable to that of the chloro ketone (**3b**).

The greater reactivity of the bromo vs. chloro derivatives is also evident in the reaction of the deoxyhexestrol halo ketones (**27e** and **27f**) and benzyl halides (**31b** and **31c**) (Figure 1B, Table I). The extent of reaction with these compounds is greater than with the hexestrol ethers (**3b,c** and **5b,c**), indicating that solvolysis is not a major competitive process. The chloro compounds do appear to solvolyze more extensively than the bromo compounds, however, as was the case with the ethers. For reference, the stability of the pyridinium salt formed from phenacyl bromide is shown in Figure 1B (top curve); its rate of decomposition is relatively slow. The more reactive compounds (**27f** and **31b**) show a decay from maximum ab-

(18) Epstein, J.; Rosenthal, R. W.; Ess, R. J. *Anal. Chem.* **1955**, *27*, 1435.
(19) Wells, P. R. "Linear Free Energy Relationships"; Academic Press: New York, 1968; pp 12 and 13.

Scheme II. Deoxyhexestrol Derivatives with Nitrogen or Sulfonyl Groups



23a, R = Me
b, R = -*p*-SO_2-C_6H_4-CH_3
c, R = -*p*-SO_2-C_6H_4-NO_2
d, R = -$COCF_3$

24a, R = H
b, R = -*p*-SO_2-C_6H_4-CH_3
c, R = -*p*-SO_2-C_6H_4-NO_2

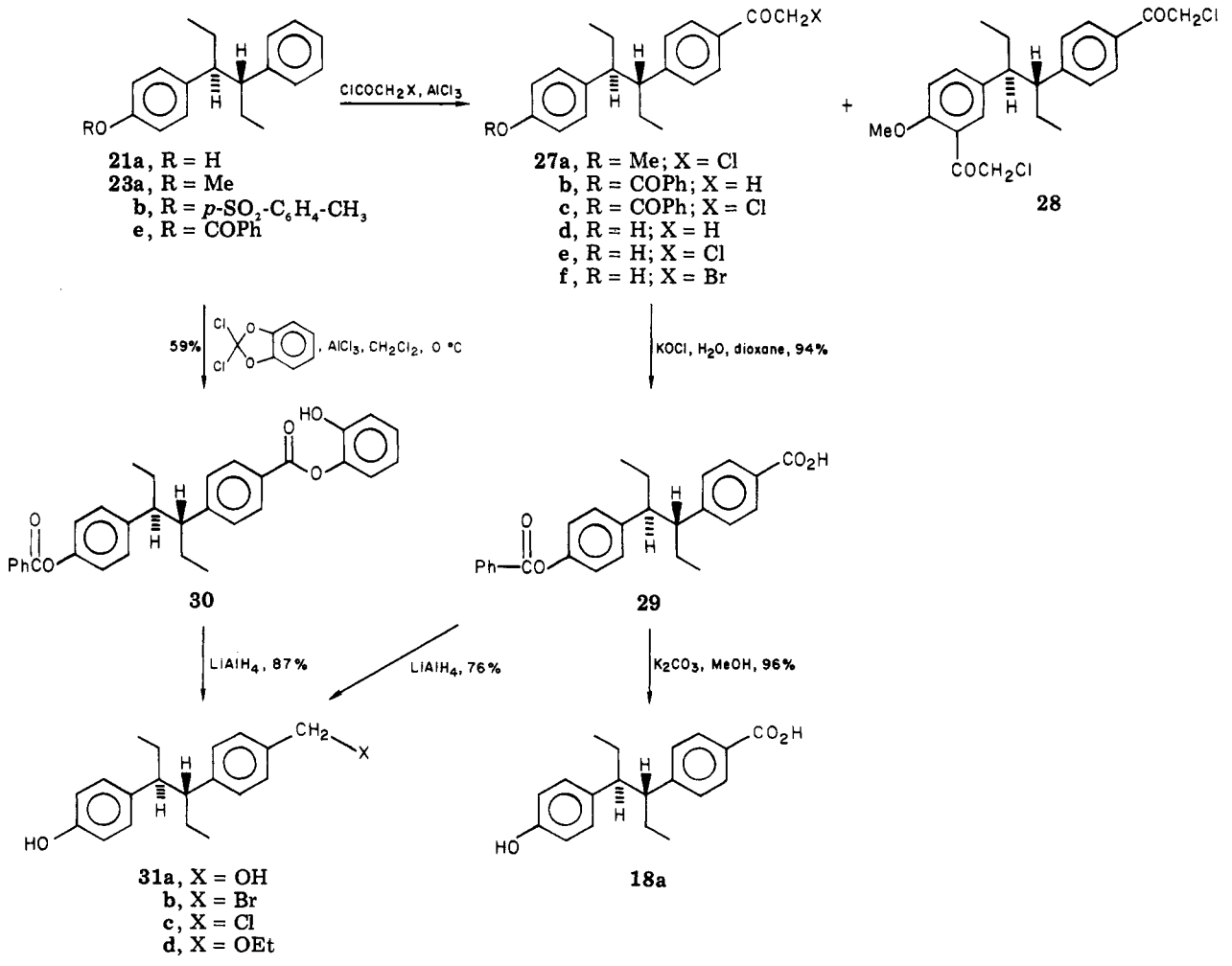
26a, R = $COCF_3$; X = F
b, R = H; X = F
c, R = $COCF_3$; X = Cl
d, R = H; X = Cl
e, R = H; X = N_3

25a, X = NH_2
b, X = N_3

sorbance that parallels the decay of the phenacyl bromide adduct. A number of interesting comparisons can be made: phenacyl bromide and bromoacetyldeoxyhexestrol (**27f**) react with nitrobenzylpyridine at virtually identical rates; this indicates that a *p*-alkyl substituent has little effect on the rate of S_N2 displacement in this system. On the other hand, bromomethyldeoxyhexestrol (**31b**) reacts twice as fast as benzyl bromide; this is consistent with the known rate enhancements caused by *p*-alkyl substituents in S_N2 displacements of benzylic halides.[19] The rates of alkylation of bromo- and chloroacetyldeoxyhexestrols (**27e** and **27f**) (Figure 1B) are quite similar to those of the hexestrol bromo- and chloroacetonyl ethers (**3b** and **3c**; Figure 1A; Table I). This indicated that the alkylating activity of the halo ketones is relatively insensitive to structural changes distal to the carbonyl group. On the other hand, solvolysis is a more important competing reaction with the hexestrol ethers.

Interaction of Hexestrol Derivatives with the Estrogen Receptor. Binding Affinity. The binding affinity of nonradiolabeled compounds to the estrogen receptor can be measured relative to [^{3}H]estradiol in a competitive assay.[4] It is convenient to express the binding affinities as a percent of the ratio of association constants [RAC × 100% or K_a (compd)/K_a (estradiol) × 100%]. In most cases we have used lamb uterine cytosol as a source of estrogen receptor; similar values are obtained using immature rat uterine cytosol. The receptor binding affinity of the compounds described in this paper, together with some related compounds prepared previously, are summarized in Table II. While it is possible that the irreversible reaction of some of these compounds with receptor or with other cytosol proteins (vide infra) may affect the estimate of reversible binding obtained by this assay, it

Scheme III. Derivatives Bearing Acyl and Alkyl Functions



21a, R = H
23a, R = Me
b, R = *p*-SO_2-C_6H_4-CH_3
e, R = COPh

27a, R = Me; X = Cl
b, R = COPh; X = H
c, R = COPh; X = Cl
d, R = H; X = H
e, R = H; X = Cl
f, R = H; X = Br

28

30

29

31a, X = OH
b, X = Br
c, X = Cl
d, X = OEt

18a

Table I. Reactive Hexestrol Derivatives. Reaction with Nitrobenzylpyridine and Reversible Binding and Inactivation of Estrogen Receptor

no.	derivative: hex-	nitrobenzylpyridine[a] k_2[b] (%)[c]	nitrobenzylpyridine[a] $t_{1/2}$, h[b]	receptor inactivation[d] $t_{1/2}$, h	receptor inactivation[d] %	receptor binding affinity,[e] RAC × 100%
3c	$OCH_2C(=O)CH_2Br$	23 (23)	0.57	<0.5	24	9.5
3b	$OCH_2C(=O)CH_2Cl$	1.21 (6)	10.8	0.25	31	10
5c	$OCH_2CH(OH)CH_2Br$	0.70 (41)	19	0.5	36	3.2
5b	$OCH_2CH(OH)CH_2Cl$	1.13 (7)	11.5	0.5	24	4.0
7a		2.3 (55)	5.8	4	44	7.4
27f	$COCH_2Br$	30 (83)	0.43	<0.5	44	1.1
27e	$COCH_2Cl$	1.6 (32)	8	0.5	29	0.44
31b	CH_2Br	68 (92)	0.19	2.5	70	9.3
31c	CH_2Cl	2.9 (75)	4.5	13	63	8.0
26b	SO_2F			1	24	0.33
	$PhC(=O)CH_2Br$	32	0.41			
	$PhCH_2Br$	34	0.39			

[a] The assay procedure was adapted from that of A. J. Swaisland.[17b] The following were incubated in a three-neck flask equipped with thermometer and stirring bar: 20 mL of 9.2×10^{-2} M 4-(4-nitrobenzyl)pyridine in ethylene glycol and 10 mL of 0.1 M Tris buffer, pH 7.32. After the temperature had stabilized at 37 °C, 5 mL of alkylating agent (2.42×10^{-4} M in methyl ethyl ketone) was added. The flask was then sealed with a rubber septum. The final concentrations were: nitrobenzylpyridine, 5.2×10^{-2} M; hexestrol derivative, 3.4×10^{-5} M; solvent (v/v) ethylene glycol–Tris buffer–2-butanone, 20:10:5 (v/v). At the indicated times, 3-mL aliquots were removed and treated with 2.5 mL of 1:1 triethylamine–2-butanone. The absorbance was read at 560 nm within 2 min against a solvent blank (the color was stable for 15 min at 25 °C and 60 min at 0 °C). [b] The second-order rate constants (k_2, M^{-1} h^{-1}) were obtained by dividing the pseudo-first-order rate constants [given as half times, $t_{1/2}$ (h)] by the concentration of nitrobenzylpyridine. The reaction was shown to be second order by running the assay at one-half and one-quarter the concentration of nitrobenzylpyridine; the pseudo-first-order rates under these conditions were very nearly 0.5 and 0.25, respectively, that of the standard assay. [c] The theoretical maximum extent of color generation was based on a molar absorptivity of 37 000 at 560 nm; this is the value determined for the dyes generated from the 4-(4-nitrobenzyl)pyridine salts from benzyl bromide and phenacyl bromide.[18] These salts were prepared by us and were characterized as analytically pure. The maximum extent of reaction was determined by a least-square fit of the data to an exponential function. [d] Receptor inactivation was determined by an adaption of our method for determining photoinactivation[5] and is described under Experimental Section. The half-times ($t_{1/2}$, h) of the inactivation process was determined by inspection of the time course of inactivation, and the extent of inactivation (%) was determined by comparing the maximum level of estrogen specific receptor inactivation with the remaining binding capacity determined with estradiol alone. [e] These data are taken from Table II and are included for comparison purposes.

should be noted that the binding assay is conducted at 0 °C, a temperature at which we have found no irreversible reaction (data not shown). Therefore, we feel that these values are reasonable estimates of receptor binding affinity.

Hexestrol binds to the estrogen receptor three times as well as estradiol; however, modification of or substitution of the phenolic hydroxyl group invariably decreases receptor binding. Etherification decreases receptor affinity, but the extent of the decrease depends upon the nature of the ether group in a manner that suggests that receptor binding is sensitive both to the bulk and the functionality pattern of the ether. Monomethylation decreases the binding affinity about 15-fold. Though larger, the hydroxyethyl ether has about the same affinity as the methyl ether, but the bulkier allyl and chloroethoxyethyl ethers bind with lower affinity. The 2-hydroxypropyl (**5a**), the 2,3-dihydroxypropyl, and the glycidyl (**7a**) ethers have similar but low affinities; the affinities of the bromo- and chlorohydrins (**5b** and **5c**) are still lower. In the acetonyl ether series, the parent compound (**3a**) and the bromo and chloro ketones (**3c** and **3b**) all have very similar affinities. The diazo ketone (**2**) and the carbethoxymethyl ether both have lower affinity; the acid, which would be ionized at the pH of the assay, binds particularly poorly.

The affinities of deoxyhexestrol (**21a**), hydroxymethyldeoxyhexestrol (**31a**), and aminodeoxyhexestrol (**25a**) are reasonably high and are comparable to data published on these compounds by Terenius.[20] The two photoreactive nitrogen derivatives nitro- (**24a**) and azidodeoxyhexestrols (**25b**) have lower affinities, as do the two deoxyhexestrol sulfonyl compounds (**26b,e**).

From the binding affinities of the 4-acyl- and 4-alkyldeoxyhexestrols listed in Table II, it is apparent that hydrogen bonding must play an important role in the binding of hexestrol to the estrogen receptor, since removal of one of the phenolic hydroxyl groups (**21a**) drops the affinity by a factor of 20, while some affinity is regained with a hydroxymethyl group in place of the hydrogens (**31a**). The other benzylic compounds, **31b** and **31c**, bind less well than the alcohol but better than the ethyl ether (**31d**). In the acetyl series, the unsubstituted analogue **27d** has the highest affinity; curiously, the bromoacetyl derivative **27f** binds with significantly higher affinity than the chloroacetyl compound **27e**. The latter two α-halo ketones bind less well than the related hexestrol bromoacetonyl and chloroacetonyl ethers (**3b** and **3c**). The nitrile (**17a**) and imino ester (**19**) show relatively modest affinities for the receptor. The Claisen rearrangement produced *o*-allylhexestrol (**8a**) and its hydroboration–oxidation product *o*-(3-hydroxypropyl)hexestrol (**9**), which have excellent and good binding affinities, respectively.

Inactivation Efficiency. We have previously described a photolysis-exchange assay[6] that can be used to determine the photointeraction between photosensitive estrogens and the estrogen receptor. In this assay, the light-mediated loss of binding activity of a sample of estrogen receptor is followed as a function of time, the biphasic decrease representing an initial chromophore-mediated inactivation, followed by a slower inactivation once the photosensitive agent has been fully consumed. This latter process has been shown to be due to the action of UV light directly on the receptor and parallels the loss of activity seen with sites filled only with (photoinert) es-

(20) Terenius, L. *Acta Pharm. Toxicol.* **1972**, *31*, 441.

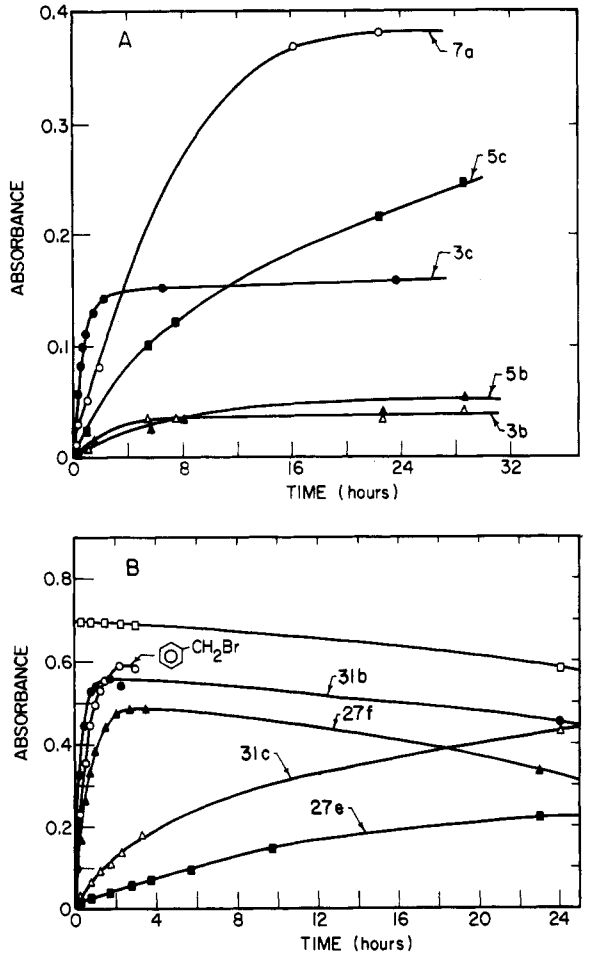


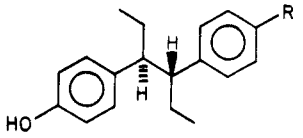
Figure 1. Colorimetric estimate of the rate of reaction of hexestrol derivatives with 4-(4-nitrobenzyl)pyridine: panel A, hexestrol ethers; panel B, 4-substituted deoxyhexestrols. The hexestrol derivatives and 4-(4-nitrobenzyl)pyridine were allowed to react under the conditions described in Table I, footnote *a*. The second-order rate constants, the half-times of the observed pseudo-first-order reaction, and the maximum extents of reaction are given in Table I. The topmost curve in panel B (□) represents the stability of the nitrobenzylpyridine salt formed with phenacyl bromide.

tradiol. The site specificity of the inactivation is established by a protection experiment in which an excess of estradiol displaces the photosensitive ligand from the binding site, producing a consequent reduction in the chromophore-dependent component of the inactivation.

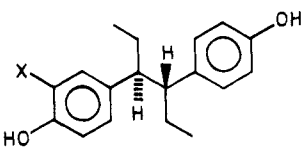
The results from assay of the three photosensitive compounds we have prepared (**24a**, **25b**, and **26e**) are shown in Figure 2. All three compounds are selective in their photoinactivation; the azide analogue (**25b**) also has the highest efficiency that we have observed for any nonsteroidal estrogen derivative. The sulfonyl azide (**26e**) is also reasonably efficient as an inactivator, but the nitro compound (**24a**) shows only modest inactivation activity.

We have adapted the photoinactivation assay to investigate the alkylating activity of the electrophilic hexestrol derivatives. While the photoinactivation version of this assay involved incubations for only several minutes at 0 °C during brief periods of UV irradiation, assay of receptor inactivation by the electrophilic estrogens requires incubations at higher temperatures and, in some cases, for longer periods of time. Under these conditions the receptor itself, in the absence of inactivating agents, loses some binding activity. Furthermore, during the longer-term 25 °C incubations it is more difficult to protect effectively the binding site with estradiol in order to demonstrate site specificity. Therefore, we have chosen to plot the data as a time course showing the site-specific inactivation as a percent of the sites that survive during incubation with estradiol alone. This method tends to make the inactivation process appear to cease, once receptor-specific in-

Table II. Estrogen Receptor Binding Affinity of Hexestrol Derivatives[a]



no.	R	binding affinity,[b] RAC × 100%
1	OH	300[c]
21a	H	17 (36)[d]
	OCH_3	19[e]
	OCH_2CH_2OH	21[f]
	$OCH_2CH_2OCH_2CH_2Cl$	0.90[f]
6a	$OCH_2CH{=}CH_2$	5.4
5a	$OCH_2CH(OH)CH_3$	4.3
	$OCH_2CH(OH)CH_2OH$	6.9[e]
5b	$OCH_2CH(OH)CH_2Cl$	4.0
5c	$OCH_2CH(OH)CH_2Br$	3.2
7a	$OCH_2CH(O)CH_2$	7.4
3a	$OCH_2C(=O)CH_3$	11
3b	$OCH_2C(=O)CH_2Cl$	10
3c	$OCH_2C(=O)CH_2Br$	9.5
2	$OCH_2C(=O)CH{=}N_2$	2[c]
	OCH_2CO_2Et	2.2[c]
	OCH_2CO_2H	0.12[c]
21a	H	17 (36)[d]
24a	NO_2	11
25a	NH_2	95 (225)[d]
25b	N_3	5.7
26b	SO_2F	0.33
26e	SO_2N_3	1.8
18a	CO_2H	1.8
31a	CH_2OH	40 (200)[d]
31c	CH_2Cl	8.0
31b	CH_2Br	9.3
31d	$CH_2OCH_2CH_3$	4.9
27d	$COCH_3$	4.1
27e	$COCH_2Cl$	0.44
27f	$COCH_2Br$	1.1
17a	C≡N	5.2
19	C(OEt)=NH·HCl	1.9



X	
8a	$CH_2CH{=}CH_2$ 54
9	$CH_2CH_2CH_2OH$ 12

[a] The binding affinities of the hexestrol derivatives to the estrogen receptor in lamb uterine cytosol were determined relative to that of [^{3}H]estradiol by a competitive binding assay. The details of this procedure have been described elsewhere.[4] [b] The binding affinity is expressed as the ratio of association constants [RAC = K_a (compd)/K_a (estradiol) × 100%]. The source of data on related compounds whose preparation is not described in this paper is given in the table footnotes. [c] This datum is from ref 4. [d] The affinities of compound **21a** (36%), **25a** (225%), and compound **31a** (200%) were reported by Terenius[20] for binding to mouse uterine receptor. These values are in reasonable agreement with our own, even though the assay conditions and the species are different. [e] This datum is from ref 29. [f] The datum is from ref 30.

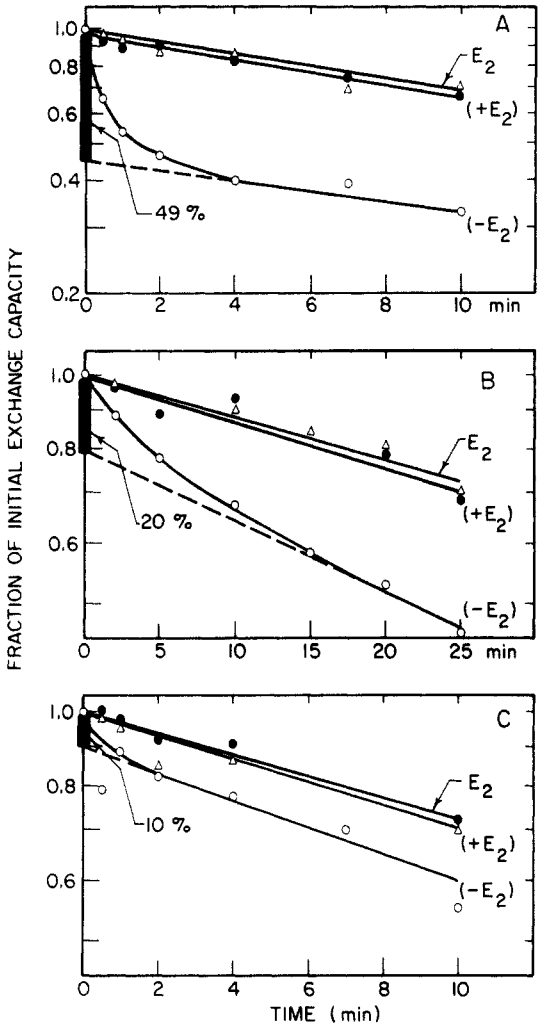


Figure 2. Time course of photoinactivation of 4-azidodeoxyhexestrol (**25b**, A), 4-(azidosulfonyl)deoxyhexestrol (**26e**, B), and 4-nitrodeoxyhexestrol (**24a**, C). The compounds were incubated with an estrogen receptor preparation from lamb uterus and irradiated at 254 nm for the time periods indicated. The quantity of binding activity that remains after photolysis is determined by subsequent exchange with [^{3}H]estradiol. In each case, the lowest curve ($-E_2$, open circles) represents the inactivation due to the photosensitive estrogen alone, the top curve (E_2, closed circles) is a control showing the inactivation that occurs when binding sites are filled with (photoinert) estradiol, and the middle curve ($+E_2$, open triangles) demonstrates site specificity by indicating that the addition of excess estradiol to displace the photosensitive estrogen from the binding sites also blocks the photoinactivation. Extrapolation of the $-E_2$ and $+E_2$ curves back to zero time gives the site specific, chromophore-mediated inactivation efficiency. Details of this assay have been described.[5]

activation has reached the point where further loss is proceeding at the same rate as thermal inactivation. The apparent cessation of inactivation may also indicate that there is a subclass of receptor that is insensitive to the alkylating agent. Nevertheless, this assay protocol and data presentation seem to us to provide the most reliable means for evaluating the rate and extent of receptor inactivation effected by the electrophilic hexestrol derivatives.

It is also important to note that 10% dimethylformamide has been included in these inactivation assays. In work we have described elsewhere,[8] we have found that inclusion of low concentrations of certain organic solvents in steroid binding assays can produce a dramatic decrease in nonreceptor binding, with little effect on binding to receptor. The effectiveness of the reactive hexestrol derivatives as receptor inactivators is markedly enhanced by inclusion of 10% dimethylformamide. The receptor inactivation data of the reactive hexestrol derivatives are in Figure 3 and in Table I.

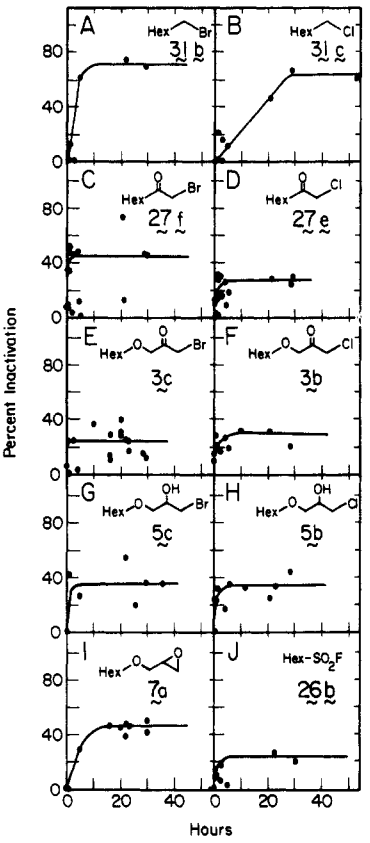


Figure 3. Time course of inactivation of the lamb uterine estrogen receptor exposed to reactive hexestrol derivatives. For procedural details, see Experimental Section: A, 4-(bromomethyl)deoxyhexestrol (**31b**); B, 4-(chloromethyl)deoxyhexestrol (**31c**); C, 4-(bromoacetyl)deoxyhexestrol (**27f**); D, 4-(chloroacetyl)deoxyhexestrol (**27e**); E, hexestrol 3-bromo-2-oxopropyl ether (**3c**); F, hexestrol 3-chloro-2-oxopropyl ether (**3b**); G, hexestrol 3-bromo-2-hydroxypropyl ether (**5c**); H, hexestrol 3-chloro-2-hydroxypropyl ether (**5b**); I, hexestrol 2,3-oxidopropyl ether (**7a**); J, 4-(fluorosulfonyl)deoxyhexestrol (**26b**).

The most complete inactivation (63–70%) is evident with the two deoxyhexestrol derivatives bearing benzylic halides **31b** and **31c**. The rate of inactivation by the chloro compound **31c** is considerably slower than with the bromo compound **31b**, which reflects its lower alkylating activity, as was evident by its slower reaction with nitrobenzylpyridine. Rather comparable levels of inactivation (29–44%) are seen with the α-halo ketones **3b**, **3c**, **27e**, and **27f**, and their inactivation rates are rapid. The greater reactivity of the bromo vs. the chloro ketones toward nitrobenzylpyridine does not appear to be reflected to any significant extent in the relative rates at which these compounds inactivate the receptor. This may indicate that the site of reaction on the receptor is more nucleophilic than nitrobenzylpyridine. Roughly equivalent inactivation was also observed with the two halohydrins **5b** and **5c** and the glycidyl ether **7a**. The halohydrins appear not to be inactivating via the epoxide, since that latter compound (**7a**) inactivates the receptor at a considerably slower rate. The inactivation rate of the halohydrins also exceeds the rate of their reaction with nitrobenzylpyridine. It is possible, though as yet unsubstantiated, that the halohydrins are being oxidized in the incubation to the corresponding α-halo ketones, which are effective inactivators. The sulfonyl fluoride (**26b**) shows a moderate rate and

extent of receptor inactivation.

The preparations of estrogen receptor that were employed in these inactivation experiments contain about 0.45 mM thiols (assayed by the Ellman method),[21] about 50% of which appears to be glutathione, and it is possible that the receptor inactivation produced by the alkylating hexestrol derivatives is limited through the competing reaction with these thiols. While our studies of the reaction of the electrophilic hexestrol derivatives with nitrobenzylpyridine established their relative reactivity, it was not clear from this work at what rate the individual compounds would react with these cytoplasmic thiols under the conditions of the receptor-inactivation assay. Therefore, we conducted a preliminary study of the reaction of selected compounds with glutathione. The reaction was followed by monitoring the consumption of glutathione (assayed by Ellman's method) upon exposure to the electrophilic compounds; alternatively, the nitrobenzylpyridine assay was used to follow the surviving concentration of alkylating agent after exposure to excess (5 mM) glutathione.

These assays (data not shown) indicated that there is a rapid reaction between the bromo ketone function and glutathione that is essentially complete by 5 min [pseudo-first-order conditions, using an excess of glutathione (5 mM) at pH 7.32, 25 °C]. Reaction of glutathione with the chloro ketone and benzyl bromide functions proceeds at a slower rate ($t_{1/2} \approx 1.5$ h) and that with the benzyl chloride function is yet slower ($t_{1/2} \approx 5$ h). Thus, it is only with the two hexestrol bromo ketones (**3c** and **27f**) that competitive reaction with glutathione might interfere. The effectiveness of these compounds as receptor inactivators and the rapidity of their receptor inactivation indicate that this competitive reaction is not a major problem. Furthermore, we have found that it is possible to reduce by >90% the concentration of thiols in estrogen receptor preparations by treatment with an equivalent of bromoacetone. (This treatment causes only a 20% reduction in the concentration of receptor sites.) Bromoacetone pretreatment did appear to cause a significant increase in the extent of receptor inactivation by the two bromo ketones (**3c** and **27f**) and the sulfonyl fluoride (**26b**) but did not affect the extent of inactivation by the other compounds. This again suggests that the endogenous thiols are not a significant factor in reducing the extent of inactivation, except with the three compounds mentioned.

Discussion

Our previous experience in developing photosensitive estrogen derivatives as affinity labels for the estrogen receptor has led us to conclude that the synthetic estrogen hexestrol is a more promising ligand for derivatization than estradiol, because the hexestrol derivatives generally have higher binding affinity than the corresponding estradiol derivatives.[4] We rationalized this on the basis of the symmetry and conformational flexibility of hexestrol that provides more modes for the binding of its derivatives in the receptor site.[4] In the present paper we describe the synthesis and the preliminary investigation of the estrogen receptor interaction of a series of hexestrol derivatives bearing electrophilic (alkylating) functions, as well as three new photosensitive hexestrol derivatives. These derivatives are either ethers of hexestrol or are 4-substituted deoxyhexestrols. While the preparation of the former type was achieved by relatively straightforward etherification reactions and functional-group manipulations, the most efficient route to the latter derivatives was by selective acylation of monodeoxyhexestrol in which the phenolic ring was deactivated by esterification with a suitable acid.

Reaction with the chromogenic agent nitrobenzylpyridine provided a convenient means for comparing the relative reactivity of the alkylating hexestrol derivatives. As expected, the bromo derivatives proved to be considerably more reactive than the corresponding chloro compounds.

The interaction of these compounds with the uterine estrogen receptor was investigated in two ways. The affinity for the receptor was assayed by a competitive binding assay, using tritium-labeled estradiol as a tracer. By this assay, most of the analogues showed lower affinity for the receptor than did estradiol. This indicated that both hydroxyl groups of hexestrol, one which has been replaced in these compounds, make an important contribution to receptor binding affinity.

The irreversible interaction of these compounds with the estrogen receptor was measured by a receptor-inactivation assay. The three photosensitive derivatives were studied by our previously published photolysis-exchange assay,[5] and azidodeoxyhexestrol (**25b**) proved to be an efficient receptor photoinactivator. It is interesting to compare this compound with the 3-azidohexestrol that we prepared previously: While the latter compound had a higher affinity for the receptor (70%),[4] its photoinactivation efficiency was much lower (15%).[5] This was presumed to be due to a rearrangement of the *o*-hydroxynitrene to a less reactive *o*-quinoneimine.[6] Such a rearrangement does not occur in the azidodeoxy compound, so its inactivation efficiency is higher. Its receptor affinity is considerably lower, however.

The deoxyhexestrol sulfonyl azide (**26e**) has particularly attractive photochemical properties as a photoattaching function, because the corresponding reactive intermediate, the sulfonylnitrene, should be highly reactive and not prone to deactivating Curtius-type rearrangements.[22] This compound was an effective inactivator of the estrogen receptor; however, the sulfonyl group is quite bulky, and so the affinity of this derivative for the receptor was low. While there is no obvious mechanism for photocovalent attachment of an aromatic nitro group to a protein, there are some reports of the use of aromatic nitro compounds in photoaffinity labeling.[5,23] We have found that 4-nitro-4-deoxyhexestrol (**24a**) has a low inactivation efficiency.

The inactivation assay was modified somewhat to study the alkylating hexestrol derivatives, and by this assay most of the compounds demonstrated substantial capacity to inactivate the estrogen receptor. Their rate of receptor inactivation in many cases exceeded the rate of their reaction with nitrobenzylpyridine and suggested that they might be alkylating a more nucleophilic site on the protein. Further evaluation of the potential of these compounds as affinity-labeling agents for the estrogen receptor would require their preparation in tritium-labeled form.

Another possible use for these alkylating agents is as receptor-carried cytotoxic agents. There is interest in compounds of this type because they may be selectively toxic toward cells that contain estrogen receptors, such as hormone-dependent breast cancer cells.[7,8] Despite the reactivity of some of the alkylating hexestrol derivatives

(21) Ellman, G. L. *Arch. Biochem. Biophys.* **1959**, *82*, 70.

(22) Breslow, D. S. In "Nitrenes"; Lwowski, W., Ed.; Wiley-Interscience: New York, 1970, Chapter 8.

(23) (a) Escher, E.; Schwyzer, R. *FEBS Lett.* **1974**, *46*, 347. (b) Cooperman, B. S. In "Aging, Carcinogenesis and Radiation Biology"; Smith, K. C., Ed.; Plenum: New York, 1976; pp 315–340.

(i.e., the bromo ketones) with thiols such as glutathione, the capacity of these reactive hexestrol derivatives to inactivate the receptor does not appear to be severely limited by their reaction with other cellular nucleophiles which are present in the uterine cytosol preparations. This suggests that these compounds may have activity in more intact systems, and studies are in progress to evaluate their selective toxicity toward estrogen-responsive cells in culture.

Experimental Section

The following compounds were obtained from the indicated sources: 17β-estradiol (Searle); 17β-[6,7-^{3}H]estradiol, 40-50 Ci/mmol (Amersham); *meso*-hexestrol, tris(hydroxymethyl)-aminomethane (Tris), charcoal Norit A, dextran grade C, 5,5′-dithiobis(2-nitrobenzoic acid) (Sigma); sodium azide (Eastman); (ethylenedinitrilo)tetraacetic acid (EDTA) (Baker); *N,N*-dimethylformamide (Mallinckrodt); Lipidex 5000 (Packard).

Tetrahydrofuran was distilled from sodium benzophenone ketyl, and diisopropylamine and triethylamine were refluxed over calcium hydride and then distilled. The organolithium reagents were titrated[24] periodically to determine the organic base present. Unless indicated otherwise, reaction products were isolated according to a standard protocol: The reaction mixture was quenched with water and extracted with several volumes of organic solvent; the extracts were sometimes washed with an aqueous solution before being dried over an anhydrous salt and filtered; solvent was removed by rotary evaporation. The extraction solvent and drying agent (and aqueous washing solution where used) are indicated in parentheses.

^{1}H NMR spectra were recorded on Varian Associates spectrometers, Models T-60, A-60, EM-390, and HR-220; chemical shifts are reported in parts per million downfield from a tetramethylsilane internal standard (δ scale). The ^{1}H NMR data are presented in the form: δ value of signal (peak multiplicity, coupling constant (if any), integrated number of protons). Infrared spectra were obtained on neat compounds held between sodium chloride plates. Data are presented in reciprocal centimeters and only the important diagnostic bands are reported. Mass spectra were obtained from a Varian MAT CH-5731 (high-resolution mass spectrum) or 311A (field desorption) spectometer and were at 70-eV ionization voltage unless otherwise indicated. Data are presented in the form: *m/e* (intensity relative to base peak). Elemental analyses were provided by the microanalytical service laboratory of the University of Illinois.

Medium pressure liquid chromatography (MPLC) was performed with a system described previously,[25] and dry column chromatography was performed according to the method of Loev and Goodman.[26] The physical properties and selected spectroscopic data on the isolated products are reported in Tables III and IV. All ^{1}H NMR spectra showed resonances characteristic for the hexestrol skeleton: δ ~0.50 (t, J = 7 Hz, 6 H, CH_3), ~1.30 (m, 4 H, CH_2), ~2.40 (m, 2 H, CH), ~6.6–7.0 (8 H, aromatic AA′BB′ patterns). All mass spectra showed prominent fragment ions from cleavage of the doubly benzylic bond. In each case, the assigned structures are consistent with the complete spectroscopic data.

Biochemical Methods. Cytosol was prepared from lamb uteri and stored in liquid nitrogen as described previously.[6b] The cytosol was prepared in TEA buffer (0.01 M Tris-HCl, 0.0015 M EDTA, 0.02% sodium azide, pH 7.4 at 25 °C); dimethylformamide was included in the binding assays at a final concentration of 7% and in the inactivation assays at a final concentration of 10%.[8,27] Free estrogens were removed by a charcoal–dextran slurry consisting of 5% Norit A and 0.5% dextran C in TEA buffer.[28]

The relative binding affinities of the compounds were determined as previously published[4] and expressed as the ratio of association constants [RAC = K_a (compd)/K_a (estradiol) × 100%].

The inactivation assay for electrophilic compounds was adapted from our inactivation assay for photoreactive compounds.[5] The cytosol was diluted to contain ~5 nM of estrogen receptor and incubated with a saturating concentration of compound: 30 nM of estradiol or 30/RAC nM of the derivatives. A parallel incubation was first "protected" with 30 nM of estradiol (1 h at 0 °C) to fill the receptor sites with nonreactive estradiol and then 30/RAC nM of the derivative was added. The electrophilic derivative, in this incubation, can only react with nonspecific sites, since the receptor site is already filled ("protected") with estradiol.

The inactivation assay was conducted at room temperature (24–25 °C). Samples were removed at various times up to 50 h and were treated with charcoal–dextran at 0 °C to remove free compound. They were then exchanged, at room temperature, with 30 nM [^{3}H]estradiol or 30 nM [^{3}H]estradiol plus 300 nM cold estradiol (hot and cold). After an exchange period of 16–20 h, the samples were cooled on ice for 1 h before treatment with charcoal–dextran. The radioactivity was measured as described previously.[5]

The difference in the exchangable sites in the "protected" sample and the sample with derivative alone represented those sites inactivated (covalently bound) by the derivative. These have been expressed as a percent of the sites remaining in the incubation with estradiol alone, since at the temperature and time scale of this experiment some sites are lost due to protein denaturation. Assays with estradiol alone and those "protected" with estradiol were corrected for the change in the specific activity of the [^{3}H]estradiol in the exchange as previously described.[28]

The protein was assayed for sulfhydryl groups by use of a modification of the Ellman assay[21] designed to conserve sample: 100 μL of cytosol was mixed with 2.90 mL of 0.05 M phosphate buffer, pH 8.0. To this was added 20 μL of 5,5′-dithiobis(2-nitrobenzoic acid). After 5 min at room temperature, the absorbance was read at 412 nm against a similar solution lacking the 5,5′-dithiobis(2-nitrobenzoic acid). The cytosol, diluted for the experiment, contained ~4×10^{-4} M SH groups. These could be lowered by 90% by addition of an equimolar quantity of bromoacetone.

4-*O*-(3-Diazo-2-oxopropyl)hexestrol (2). This procedure is a modification which improves the yield of our earlier method.[6a] A solution of hexestrol (1; 2.00 g, 7.41 mmol) and 3.06 g (22.2 mmol) of anhydrous potassium carbonate in 63 mL of methanol–tetrahydrofuran (4:1) was treated with 1.51 g (9.26 mmol) of 3-bromodiazoacetone[6a] in 63 mL of 4:1 methanol–tetrahydrofuran. The reaction mixture was flushed with nitrogen and then stirred at 25 °C. After 12 h, the reaction was quenched with water and acidified with 10% aqueous hydrochloric acid. Product isolation (EtOAc, $MgSO_4$) provided 3.44 g of crude product mixture. Preparative TLC (4% ethanol–chloroform, three developments) furnished 780 mg (30%) of a yellow crystalline product: mp 92 °C dec (lit.[3] 92 °C).

4-*O*-(2-Oxopropyl)hexestrol (3a). Potassium carbonate (330 mg, 2.39 mmol) was added to a solution of 2.00 g (7.40 mmol) of hexestrol (1) in 8 mL of anhydrous acetone, and the resultant suspension was refluxed for 15 min and then cooled to room temperature. One-fourth of a mixture which had been reacted overnight, comprising 410 mg (4.46 mmol) of chloroacetone, 500 μL of dry acetone, and 10 mg of potassium iodide, was added dropwise to the hexestrol solution with stirring. The remaining chloroacetone mixture and 990 mg (7.17 mmol) of potassium carbonate were added in three portions over 6 h, followed by continued stirring for an additional 12 h. The reaction was quenched with water, the solvent was evaporated, and the crystalline residue was acidified with 10% aqueous HCl. Product isolation (EtOAc, 10% aqueous $NaHCO_3$, $MgSO_4$) afforded 2.40 g (99%) of a crystalline residue consisting principally of monosubstituted acetonyl ether **3a** and hexestrol, in addition to the diacetonyl ether derivative. Chromatography of a 100-mg sample

(24) Watson, S. C.; Eastham, J. F. *J. Organomet. Chem.* **1967**, *9*, 165.
(25) Heiman, D. F.; Senderoff, S. G.; Katzenellenbogen, J. A.; Neeley, R. J. *J. Med. Chem.* **1980**, *23*, 994.
(26) Loev, B.; Goodman, M. M. *Chem. Ind. (London)* **1967**, 2026.
(27) Katzenellenbogen, J. A.; Carlson, K. E.; Heiman, D. F.; Goswami, R. *J. Nucl. Med.* **1980**, *21*, 550.
(28) Katzenellenbogen, J. A.; Johnson, H. J., Jr.; Carlson, K. E. *Biochemistry* **1973**, *12*, 4092.
(29) Tatee, T.; Carlson, K. E.; Katzenellenbogen, J. A.; Robertson, D. W.; Katzenellenbogen, B. S. *J. Med. Chem.* **1979**, *22*, 1509.
(30) Hsiung, H. M. Ph.D. Thesis, Department of Chemistry, University of Illinois, Urbana, IL, 1975.

on preparative TLC (SiO_2, 5:5:1 benzene–chloroform–ethyl acetate, four developments) afforded 12 mg (57%, based on consumed hexestrol) of **3a** as a colorless solid, which was recrystallized from benzene.

4-*O*-(3-Chloro-2-oxopropyl)hexestrol (3b). Gaseous hydrogen chloride was bubbled through a solution of 4-*O*-(3-diazo-2-oxopropyl)hexestrol (**2**; 100 mg, 0.284 mmol) in 35 mL of ether at 0 °C for 1 min. The reaction mixture was stirred at 0 °C for an additional 20 min, quenched while cold with 10% aqueous $NaHCO_3$ (dropwise). Product isolation ($MgSO_4$) gave 100 mg of crude crystalline product. Recrystallization from petroleum ether–ether afforded 90 mg (88%) of the colorless crystalline chloro ketone (**3b**).

4-*O*-(3-Bromo-2-oxopropyl)hexestrol (3c). Gaseous hydrogen bromide was bubbled through a solution of 4-*O*-(3-diazo-2-oxopropyl)hexestrol (**2**; 130 mg, 0.369 mmol) in 45 mL of ether at 0 °C for 1 min. The reaction mixture was stirred for an additional 15 min at 0 °C and quenched while cold with 10% aqueous $NaHCO_3$ (dropwise). Product isolation ($MgSO_4$) gave 90 mg (60%) of crude product. Recrystallization from petroleum ether–ether afforded 50 mg (33%) of the colorless crystalline bromo ketone **3c**.

4-*O*-(2-Hydroxypropyl)hexestrol (5a). Sodium borohydride (6 mg, 0.124 mmol) was added to a solution of 20 mg (0.062 mmol) of 4-ketopropylhexestrol (**3a**) in 750 μL of 2:1 methanol–THF at 0 °C. After 0.5 h, the reaction mixture was quenched with water and acidified with 10% aqueous hydrochloric acid. Product isolation (Et_2O, $MgSO_4$) afforded 20 mg of crude product mixture. Preparative TLC (5:1:5 benzene–ethyl acetate–chloroform, two developments) furnished 18 mg (88%) of a colorless crystalline product.

4-*O*-(3-Chloro-2-hydroxypropyl)hexestrol (5b). A mixture of 3.00 g (11.1 mmol) of hexestrol (**1**), 12.60 g (136 mmol) of epichlorohydrin, and 136 mg of piperidine was heated at 55 °C for 5 h. After the mixture cooled, 10 mL of water was added and excess epichlorohydrin was azeotropically removed at reduced pressure. The residual oil was acidified with 10% aqueous hydrochloric acid and isolated (EtOAc, 10% aqueous $NaHCO_3$, Na_2SO_4), giving a quantitative yield of mono- and disubstituted chlorohydrin ethers and unconsumed starting material. Preparative medium-pressure chromatography (SiO_2, 40% ether–hexane) afforded as the middle fraction 1.60 g (58%, based on consumed hexestrol) of a colorless crystalline product identified as the chlorohydrin **5b**. An analytical sample was obtained by recrystallization from benzene.

An analytical sample of the more mobile fraction was recrystallized from hexane and identified as 4,4′-*O*-bis(3-chloro-2-hydroxypropyl)hexestrol: mp 56–58 °C.

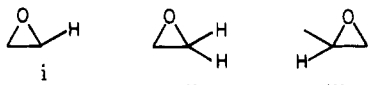
4-*O*-(3-Bromo-2-hydroxypropyl)hexestrol (5c). A mixture of 1.00 g (3.7 mmol) of hexestrol (**1**), 6.22 g (45.5 mmol) of epibromohydrin, and 45 mg of piperidine was heated at 55 °C for 5 h. After excess epibromohydrin was removed at reduced pressure, the residue was acidified with 10% aqueous hydrochloric acid; product isolation (EtOAc, 10% aqueous $NaHCO_3$, Na_2SO_4) gave a quantitative yield of mono- and disubstituted bromohydrin ethers and unconsumed hexestrol. Purification by preparative TLC (SiO_2, 1:1 ether–hexane, four developments) gave 510 mg (66%, based on consumed hexestrol) of **5c** as a colorless crystalline product. An analytical sample was obtained by recrystallization from benzene.

4-*O*-Allylhexestrol (6a). Hexestrol (**1**; 6.00 g, 22.2 mmol) was dissolved in 300 mL of *N,N*-dimethylformamide and treated with 9.20 g (66.7 mmol) of anhydrous potassium carbonate, and the resultant suspension was heated at 45 °C for 0.5 h. After the suspension cooled, 2.68 g (22.2 mmol) of allyl bromide was added dropwise, and the mixture was stirred at room temperature for 12 h. The reaction was quenched with 250 mL of water and acidified with 10% aqueous hydrochloric acid. Product isolation (EtOAc, 10% aqueous sodium bicarbonate, Na_2SO_4) produced 6.8 g of a crude mixture comprising mono- and diallylhexestrol, and unreacted hexestrol. Purification by dry column chromatography (SiO_2, 1:2 ether–hexane) furnished 2.2 g (51%, based on converted hexestrol) of **6a** as a colorless crystalline product, which was recrystallized from hexane.

An analytical sample of the most mobile fraction was recrystallized from petroleum ether and identified as 4,4′-*O*-diallylhexestrol: mp 84–85 °C (lit.[27] mp 81.5 °C).

4-*O*-(2,3-Oxidopropyl)hexestrol (7a). Method 1. By Epoxidation of Hexestrol Allyl Ether (6a). 4-*O*-Allylhexestrol (**6a**; 753 mg, 2.43 mmol) was acetylated with 2 mL of pyridine and 1.4 mL (14.6 mmol) of acetic anhydride. After 12 h at 40 °C, the reaction mixture was diluted with water. Product isolation (EtOAc, 10% aqueous HCl, $MgSO_4$) gave 855 mg (100%) of crude acetate. Recrystallization from ether–hexane afforded 727 mg (85%) of the acetate **6b** as a colorless crystalline solid: mp 89–90 °C; IR (KBr) 1775 (C=O), 930, 915 (C=C) cm^{-1}; ^1H NMR ($CDCl_3$) δ 2.27 [s, 3 H, C(=O)CH_3], 4.48 (dd, J = 1.5 Hz, 2 H, $ArOCH_2$), 5.13–5.53 (m, J = 1.5 Hz, 2 H, C=CH_2), 5.83–6.26 (m, 1 H, CH=C); mass spectrum, m/e 352 (1, M^+), 175 (100), 135 (9).

This acetate **6b** (400 mg, 1.14 mmol) was dissolved in 12 mL of chloroform and treated with a solution of 435 mg (1.26 mmol) of 50% *m*-chloroperoxybenzoic acid in 3 mL of chloroform over a 10-min interval. After the solution was stirred at room temperature for 12 h, product isolation (10% aqueous $NaHCO_3$, $CHCl_3$, $MgSO_4$) afforded 400 g of a 50:50 mixture (NMR) of starting olefin (**6b**) and epoxide (**7b**). Preparative TLC (2:1 petroleum ether–ether) and recrystallization from benzene gave 200 mg (96%, based on recovered olefin) of **7b** as colorless crystals: mp 108–110 °C; IR (KBr) 1760 (C=O), 1255 (i) cm^{-1}; ^1H NMR



i ii iii

(CCl_4) δ 2.23 [s, 3 H, C(=O)CH_3], 2.40–2.86 (br m, 4 H, CH, ii), 3.20 (m, 1 H, iii), 3.98 (d, 2 H, $ArOCH_2$), mass spectrum, m/e 368 (1, M^+), 191 (100), (2), 135 (21).

The epoxide acetate **7b** (100 mg, 0.272 mmol) was dissolved in 6 mL of methanol, and 75 mg (0.543 mmol) of potassium carbonate was added. After the mixture was stirred for 2.5 h at 25 °C, product isolation (EtOAc, $MgSO_4$) and recrystallization from ether–hexane provided 60 mg (68%) of **7a** as a colorless crystalline solid.

Method 2. From 4-*O*-(3-Chloro-2-hydroxypropyl)hexestrol (5b). A solution of 60 mg (0.166 mmol) of 4-*O*-(3-chloro-2-hydroxypropyl)hexestrol (**5b**) in 1.5 mL of Me_2SO was treated with 83 μL (0.415 mmol) of 20% aqueous NaOH and stirred for 2.5 h at 25 °C. The reaction was diluted with water and acidified with 10% aqueous hydrochloric acid. Product isolation (Et_2O, Na_2SO_4) and recrystallization from ether–hexane provided 43 mg (80%) of **7a**.

***o*-Allylhexestrol (8a).** A solution of 1.10 g (3.55 mmol) of hexestrol ally ether (**6a**) in 3 mL of diphenyl ether was blanketed under a nitrogen atmosphere and heated to 220 °C in a sand bath. After 2.5 h, the reaction mixture was cooled, diluted with ether, and filtered, providing 1.05 g of crude allylphenol. Purification by alumina column chromatography (1:1 petroleum ether–ether) afforded 870 mg (79%) of **8a** as colorless crystals.

***o*-(3-Hydroxypropyl)hexestrol (9).** To a solution of 100 mg (0.322 mmol) of *o*-allylhexestrol (**8a**) in 2 mL of THF at 0 °C was added dropwise 5.2 mL (3.86 mmol) of a 0.75 M solution of diborane in THF. After gradually warming to room temperature over 6 h, the reaction mixture was cautiously quenched with 2 mL water and 1 mL of a 40% aqueous NaOH solution. Following dropwise addition of 1.6 mL of 30% aqueous hydrogen peroxide, the solution was stirred for an additional 12 h at room temperature. Acidification with 10% aqueous HCl and product isolation (EtOAc, $MgSO_4$) yielded a quantitative recovery of crude product mixture. Column chromatography on silica gel (5:1:5 benzene–ethyl acetate–chloroform) provided 89 mg (84%) of **9** as colorless crystals.

α-(*p*-Bromophenyl)-*p*-methoxyacetophenone (10). This compound was prepared using a modification of the procedure of Wilson and co-workers.[12] Phosphorous pentachloride (25 g, 0.12 mol) was added in portions to a solution of 25 g (0.116 mol) of *p*-bromophenylacetic acid in 50 mL of benzene. The mixture was refluxed for 30 min, then cooled to 0 °C, and 55 mL (55 g, 0.51 mol) of anisole was added, followed by the dropwise addition of 40 mL (89.1 g, 0.34 mol) of anhydrous stannic chloride in 50 mL of benzene. Stirring was continued overnight at room temperature. The deep red solution was then poured into ice and concentrated HCl. Product isolation (ethyl acetate, 5% aqueous

Table III. Physical and Selected Spectroscopic Data[a]

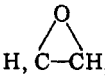
no.	mp, °C	IR (neat or KBr pellet), cm^{-1}	^1H NMR ($CDCl_3$),[b] δ	MS (70 eV)[c]
3a	151-152	3390, 1728	2.30 (s, 3 H, $COCH_3$), 4.52 (s, 2 H, $ArOCH_2$)	326 (0.7, M^+), 191 (100), 135 (35)
3b	157-158	3420, 1730	4.38 (s, 2 H, CH_2Cl), 4.70 (s, 2 H, $ArOCH_2$)	360, 362 (0.3, 0.1, M^+), 225, 227 (53, 17), 135 (100)
3c	146-148	3420, 1730	4.13 (s, 2 H, CH_2Br), 4.73 (s, 2 H, $ArOCH_2$)	404, 406 (0.2, 0.2, M^+), 269, 271 (8), 135 (100)
5a	89-91	3420	1.85 (m, 3 H, $CHCH_3$), 3.65-3.85 (m, 1 H, CHOH), 4.48 (m, 2 H, $ArOCH_2$)	328 (0.7, M^+), 193 (100), 135 (25)
5b	121-122	3440	3.72 (d, J = 4.5 Hz, 2 H, CH_2Cl), 3.96-4.3 (m, 3 H, $ArOCH_2$, CHOH)	362, 364 (1, 0.3, M^+), 227, 229 (100, 33), 135 (100)
5c	30-31	3440	3.60 (dd, J = 4.5 Hz, 2 H, CH_2Br), 3.90-4.33 (m, 3 H, $ArOCH_2$, CHOH)	406, 408 (0.3, 0.3, M^+), 271, 273 (73, 71), 135 (100)
6a	91-92	3440, 995, 950	CCl_4: 4.55 (m, 2 H, $ArOCH_2$), 5.13-5.52 (m, 3 H, $C=CH_2$, OH), 5.80-6.45 (m, 1 H, CH=C)	310 (1, M^+), 175 (100), 135 (15)
7a	108-110	3420, 1250	2.73-3.00 (br m, 2 H, ), 3.38 (m, 1 H, ), 3.87-4.33 (dd, J = 3.5 Hz, 2 H, $ArOCH_2$)	326 (0.8, M^+), 191 (100), 135 (49)
8a	65-67	3430, 918	3.40 (d, 2 H, $ArOCH_2$), 5.13 (dd, J = 12 and 15 Hz, $C=CH_2$), 5.76-6.24 (br m, 1 H, CH=C)	310 (3, M^+), 175 (100), 135 (27)
9	212-214	3430	acetone-d_6: 1.93-2.10 (m, 2 H, CH_2COH), 2.67 (t, J = 7.5 Hz, 2 H, CH_2CCOH), 3.56 (t, 2 H, CH_2OH)	328 (1, M^+), 193 (100), 135 (23)
17a	118-119	3422, 2245		279 (1, M^+), 135 (100)
20	170-173		3.96 (br s, 2 H, $ArCH_2$)	283 (16, M^+), 148 (13), 135 (100)
21a	148-149	3250	7.17 (m, 5 H)	254 (2, M^+), 135 (100)
23c	167-168	1540, 1385, 1358, 880	8.13 (AA'BB', Δν = 0.34 ppm, J = 9 Hz, 4 H)	320 (98), 292 (19), 119 (100), 91 (97)
24a	(liquid)	3440, 1525, 1355	6.86 (AA'BB', Δν = 0.19 ppm, J = 8.5 Hz, 4 H), 7.69 (AA'BB', Δν = 0.86 ppm, J = 8.5 Hz, 4 H)	299 (2, M^+), 135 (100), 107 (39)
25a	183-185		3.50 (s, 2 H, NH_2)	269 (4, M^+), 135 (17), 134 (100)
25b	98-100 dec	3470, 2135	CCl_4: 6.50-7.13 (m, 8 H, Ar)	295 (2, M^+), 160 (1), 135 (100), 107 (33)
26b	(liquid)	3470, 1410, 1220, 790	6.87 (AA'BB', Δν = 0.21 ppm, J = 8 Hz, 4 H), 7.64 (AA'BB', Δν = 0.57 ppm, J = 8 Hz, 4 H)	336 (0.35, M^+), 135 (100), 107 (54)
26d	71-73	3400, 1380, 1190, 375	6.86 (AA'BB', Δν = 0.21 ppm, J = 9 Hz, 4 H), 7.64 (AA'BB', Δν = 0.56 ppm, J = 8 Hz, 4 H)	
26e	45-47	3400, 2145, 1378	6.86 (AA'BB', Δν = 0.21 ppm, J = 8 Hz, 4 H), 7.63 (AA'BB', Δν = 0.54 ppm, J = 8 Hz, 4 H)	359 (0.6, M^+), 152 (4), 135 (45)
27b	122-124	1727, 1683	2.58 (s, 3 H, $COCH_3$)	10 eV: 400 (0.5, M^+), 240 (19), 239 (100)
27c	114-117	1732, 1703, 1688	4.66 (s, 2 H, $ClCH_2CO$)	239 (57), 105 (100)
27d	156-157	3345, 1670	2.60 (s, 3 H, CH_3CO), 6.90 (AA'BB', Δν = 0.19 ppm, J = 9 Hz, 4 H), 7.57 (AA'BB', Δν = 0.67 ppm, J = 9 Hz, 4 H)	296 (3, M^+), 162 (12), 135 (100)
27e	114-117	3390, 1697	acetone-d_6: 4.97 (s, 2 H, $ClCH_2CO$), 6.91 (AA'BB', Δν = 0.26 ppm, J = 9 Hz, 4 H), 7.70 (AA'BB', Δν = 0.56 ppm, J = 9 Hz, 4 H)	332, 330 (0.3, 1, M^+), 135 (100)
27f	83-85	3420, 1695, 1685	4.42 (s, 2 H, $BrCH_2CO$), 6.85 (AA'BB', Δν = 0.20 ppm, J = 9 Hz, 4 H), 7.57 (AA'BB', Δν = 0.67 ppm, J = 9 Hz, 4 H)	374, 376 (0.6, 0.6, M^+), 135 (100)
29	206-209	3700-2200, 1746, 1698	7.0-8.3 (13 H, Ar)	10 eV: 402 (0.7, M^+), 239 (100)
30	211-212	3445, 1740, 1718	6.7-8.6 (m, 17 H, Ar)	386 (24), 385 (85), 239 (69), 105 (100), 494 (M^+, by field desorption)
31a	165-167	3480	acetone-d_6: 4.04 (t, J = 6 Hz, 1 H, CH_2OH), 4.57 (d, J = 6 Hz, 2 H, CH_2OH), 6.90 (AA'BB', Δν = 0.26 ppm, J = 9 Hz, 4 H)	284 (1, M^+), 135 (100)
31b	84-88	3460	4.48 (s, 2 H, $ArCH_2$), 6.84 (AA'BB', Δν = 0.24 ppm, J = 9 Hz, 4 H), 7.17 (AA'BB', Δν = 0.22 ppm, J = 9 Hz, 4 H)	348, 346 (0.3, 0.3, M^+), 135 (100)

Table III (*Continued*)

no.	mp, °C	IR (neat or KBr pellet), cm^{-1}	^1H NMR ($CDCl_3$),[b] δ	MS (70 eV)[c]
31c	86–88	3480	4.56 (s, 2 H, $ClCH_2$), 6.85 (AA'BB', $\Delta\nu$ = 0.24 ppm, J = 9 Hz, 4 H), 7.18 (AA'BB', $\Delta\nu$ = 0.18 ppm, J = 9 Hz, 4 H)	304, 302 (0.5, 1.6, M^+), 135 (100)
31d	93–94	3360	1.25 (t, J = 7 Hz, 3 H, OCH_2CH_3), 3.55 (q, J = 7 Hz, 2 H, OCH_2CH_3), 4.47 (s, 2 H, $ArCH_2O$), 6.84 (AA'BB', $\Delta\nu$ = 0.23 ppm, J = 9 Hz, 4 H), 7.16 (AA'BB', $\Delta\nu$ = 0.19 ppm, J = 9 Hz, 4 H)	312 (3, M^+), 135 (100)

[a] Instrumentation and methods are described at the start of the Experimental Section. [b] Other solvents are noted where used. [c] Other ionizing voltages are noted where used.

Table IV. Microanalyses or Exact Mass of Hexestrol Derivatives

		calculated (M_r)			found (M_r)		
no.	formula	C	H	other	C	H	other
3a	$C_{21}H_{26}O_3$		(326.1902)			(326.1904)	
3b	$C_{21}H_{25}O_3Cl$		(360.1511)			(360.1513)	
3c	$C_{21}H_{25}O_3Br$		(404.1012)			(404.1014)	
5a	$C_{21}H_{28}O_3$		(328.2105)			(328.2107)	
5b	$C_{21}H_{27}O_3Cl$		(362.1648)			(362.1647)	
5c	$C_{21}H_{27}O_3Br$	61.92	6.68	Br: 19.62	61.70	6.85	Br: 19.78
6a	$C_{21}H_{26}O_2$	81.29	8.39		81.07	8.20	
7a	$C_{21}H_{26}O_3$		(326.1894)			(326.1895)	
8a	$C_{21}H_{26}O_2$		(310.1793)			(310.1779)	
9	$C_{21}H_{28}O_3$		(328.2071)			(328.2074)	
17a	$C_{19}H_{21}NO$	81.68	7.58	N: 5.01	81.41	7.58	N: 4.92
20	$C_{19}H_{25}ON$		(283.1930)			(283.1932)	
21a	$C_{18}H_{22}O$	84.99	8.72		84.98	8.70	
23c	$C_{24}H_{25}O_5NS$	65.60	5.69	N: 3.19	65.73	5.60	N: 3.15
23e	$C_{25}H_{26}O_2$	83.76	7.31		84.02	7.30	
24a	$C_{18}H_{21}NO_3$		(299.1516)			(299.1515)	
25a	$C_{18}H_{23}NO$		(269.1780)			(269.1780)	
25b	$C_{18}H_{21}N_3O$	73.19	7.17	N: 14.23	73.06	7.23	N: 14.30
26b	$C_{18}H_{21}O_3SF$		(336.1197)			(336.1197)	
26e	$C_{18}H_{21}SO_3N_3$		(359.1303)			(359.1300)	
27b	$C_{27}H_{28}O_3$	80.97	7.05		81.10	7.11	
27c	$C_{27}H_{27}ClO_3$	74.56	6.26	Cl: 8.15	74.32	6.24	Cl: 8.27
27d	$C_{20}H_{24}O_2$	81.04	8.16		80.81	8.19	
27e	$C_{20}H_{23}ClO_3$	72.61	7.01	Cl: 10.71	72.64	7.19	Cl: 10.60
27f	$C_{20}H_{23}BrO_3$	64.01	6.18	Br: 21.29	63.82	6.41	Br: 21.46
29	$C_{26}H_{26}O_4$	77.59	6.51		77.89	6.49	
30	$C_{32}H_{30}O_5$	77.71	6.11		77.43	6.14	
31a	$C_{19}H_{24}O_2$	80.14	8.51		80.11	8.38	
31b	$C_{19}H_{23}BrO$		(346.0932)			(346.0934)	
31c	$C_{19}H_{23}ClO$	75.36	7.65	Cl: 11.71	75.58	7.82	Cl: 11.52
31d	$C_{21}H_{28}O_2$		(312.2089)			(312.2089)	

$NaHCO_3$, $MgSO_4$) gave a red oil, which was taken up in hexane. Pale pink crystals precipitated immediately and were collected by filtration and washed with hexane five times. Concentration of the mother liquor gave a second crop of crystals, which were recrystallized from methanol and combined with the first crop: total yield 27.4 g (78%); mp 140–141.5 °C (lit.[12] 140–141 °C).

α-(*p*-Bromophenyl)-*p*-methoxybutyrophenone (11). Sodium hydride (1.48 g of a 56% dispersion in mineral oil, 34.5 mmol) was washed twice with dry THF and then added slowly as a slurry in THF (in a Pasteur pipette) to a stirred solution of 10.0 g (32.8 mmol) of α-(*p*-bromophenyl)-*p*-methoxyacetophenone (**10**) in 55 mL of THF. After the addition was completed, ethyl iodide (5.4 g, 34.5 mmol) was added dropwise, and the stirring and heating were continued overnight. The product was isolated (ether, $MgSO_4$) and redissolved in hexane. A small amount of starting material that crystallized from the hexane solution was removed by filtration. Concentration gave a pale yellow oil (10.1 g, 92%), showing on TLC a trace of a more mobile impurity, presumed to be the bisethylated product: ^1H NMR (CCl_4) δ 3.72 (s, 3 H, OCH_3), 4.26 (br t, $W_{1/2}$ = 1.5 Hz, J = 7.3 Hz, 1 H, CH), 6.67–7.48 (m, 6 H, aromatics), 7.70–7.98 (m, 2 H, aromatic protons ortho to carbonyl). This material was carried on to the next step without further purification.

3-(*p*-Bromophenyl)-4-(*p*-anisyl)-3-hexene (13, Mixture of *E* and *Z* Isomers). Ethylmagnesium iodide was prepared from 185 mg (7.6 mmol) of magnesium turnings and 935 mg (6 mmol) of ethyl iodide and added in portions to a solution of the ketone **11** (2.0 g, 6.0 mmol) in 15 mL of dry ether. After 18 h at 25 °C, the reaction mixture was poured into aqueous ammonium chloride. Product isolation (ether, $MgSO_4$) gave the alcohol (**12**), which was transferred to a 500-mL round-bottom flask with 600 mg of *p*-toluenesulfonic acid and heated over a steam bath under an aspirator vacuum for 2 h. The product olefin was separated from the acid and unconsumed starting material by dry column chromatography (silica gel, CCl_4). In this fashion, 910 mg (3.9 mmol) of the product as well as 620 mg of recovered ketone **11** were secured. The yield based on consumed starting material was 67%: ^1H NMR (CCl_4) δ 3.64, 3.70, and 3.76 (s, total of 3 H, OCH_3). The appearance of a third methoxyl resonance, in addition to the presence of a weak resonance (integrating to ca. 0.5 protons) at δ 5.6, suggests the presence of a small amount of the 2-hexene isomer (**14**) in the product mixture.

3-(*p*-Anisyl)-4-(*p*-bromophenyl)-2-hexene (14, Mixture of *E* and *Z* Isomers). A three-necked flask charged with 5.2 g (14 mmol) of ethyltriphenylphosphonium bromide and 15 mL of dry ether was chilled to 0 °C and with rapid stirring 10 mmol of

n-butyllithium in hexane was added dropwise. Stirring was continued at room temperature for 2 h. At this time, excess phosphonium salt was still present in the flask. The dark red contents of the flask were again chilled to 0 °C, and a solution of 2.92 g (8.75 mmol) of *p*-anisyl 1-(*p*-bromophenyl)propyl ketone (**11**) in 10 mL of ether was added over 10 min. After the solution was stirred overnight at room temperature, product isolation (ether, $MgSO_4$) gave material that was purified by dry column chromatography (silica gel, CCl_4) to yield 2.28 g (75%) of the isomeric 2-hexenes (**14**): ^1H NMR (CCl_4) δ 1.32–1.92 [m (containing a br d centered at δ 1.43, $W_{1/2}$ = 2 Hz, J = 6.5 Hz, and a d centered at δ 1.83, J = 7 Hz), 5 H, CH_2 plus allylic CH_3], 3.05–3.40 and 3.73–4.05 (m, total of 1 H, CH), 3.61 and 3.63 (s, total of 3 H, OCH_3), 5.30–5.68 (m, 1 H, C=CH); mass spectrum, *m/e* 344, 346 (M^+, 14.5, 13.9), 236 (29), 148 (11), 147 (100), 135 (24). The product was used without further purification.

3-(*p*-Bromophenyl)-4-(*p*-anisyl)hexane (15, Mixture of Erythro and Threo Isomers). A rapidly stirred mixture of 76 mg of 83% platinum oxide in 15 mL of absolute ethanol was equilibrated with hydrogen at atmospheric pressure and room temperature. After 20 min the uptake of hydrogen ceased (18 mL). The stirring was discontinued while 900 mg (2.6 mmol) of 3-(*p*-anisyl)-4-(*p*-bromophenyl)-2-hexene (**14**) in 2 mL of absolute ethanol was injected through an ethanol-washed septum. An additional 2 mL of ethanol was used to wash in any remaining traces of olefin. Stirring was resumed until approximately 1.05 equiv of hydrogen was consumed (70 min). The spent catalyst was removed, and the colorless ethanol solution was diluted with ethyl acetate and washed with 5% sodium bicarbonate and water. After the solvent was dried and removed under vacuum, the crude product (100% yield) was analyzed by gas chromatography (5 ft × 0.125 in. 3% OV-17, on Supelcoport 100–120 mesh, 230 °C), showing the presence of less than 10% of the debrominated product. The identity of this side product was confirmed by a comparison of retention times with an authentic sample of 3-phenyl-4-(*p*-anisyl)hexane prepared in an analogous sequence beginning with the acylation of anisole with phenylacetic acid. The desired product (**15**) displayed: mass spectrum, *m/e* 348 (0.9, M^+), 346 (1.5, M^+), 150 (11), 149 (100), 121 (20); ^1H NMR (C_6D_6) δ 0.37–0.90 (m, 6 H, CH_3), 0.96–2.06 (m, 4 H, CH_2), 2.25–2.67 (m, 2 H, CH), [3.31 (s, threo) and 2.41 (s, erythro), ratio 3:2, 3 H together, OCH_3], 6.43–7.46 (m, 8 H, aromatics).

Optimum yields were obtained under the above conditions and when the catalyst to substrate ratio was approximately 0.08. Less than 8% catalyst greatly increased the length of time of the reaction, and larger percentages of catalyst led to an increase in the rate of hydrogenolysis of the bromine–carbon bond.

3-(*p*-Cyanophenyl)-4-(*p*-anisyl)hexane (16, Mixture of Erythro and Threo Isomers). A mixture of 2.1 g (6 mmol) of 3-(*p*-bromophenyl)-4-(*p*-anisyl)hexane (**15**), 1.2 g (13.5 mmol) of copper(I) cyanide, and 15 mL of dry *N*-methyl-2-pyrrolidinone was refluxed under nitrogen for 3 h. The contents of the flask were then cooled to 65 °C and stirred for 20 min at this temperature with 25 mL of aqueous acidified ferric chloride. This mixture was cooled to room temperature, and products were isolated ($CHCl_3$, H_2O wash, $MgSO_4$), yielding 1.25 g of a light brown oil. The crude isomeric nitriles were purified by dry column chromatography (silica gel; 20% CH_2Cl_2 in hexane), yielding 1.07 g (61%) of the diastereomeric liquid nitriles: ^1H NMR (C_6D_6) δ 0.35–0.9 (m, 6 H, CH_3), 0.9–1.9 (m, 4 H, CH_2), 2.2–2.65 (m, 2 H, CH), 3.30 (threo) and 3.41 (erythro) (s, 3 H, ratio 3:2, OCH_3), 6.36–7.26 (m, 8 H, aromatics); mass spectrum, *m/e* 293 (0.34, M^+), 291 (4), 149 (100), 121 (27).

***erythro*-3-(*p*-Cyanophenyl)-4-(*p*-hydroxyphenyl)hexane (17a) and *threo*-3-(*p*-Cyanophenyl)-4-(*p*-hydroxyphenyl)-hexane (17b).** A solution of 612 mg (2.1 mmol) of 3-(*p*-cyanophenyl)-4-(*p*-anisyl)hexane (**16**) in 10 mL of methylene chloride was purged with nitrogen and chilled to −78 °C. Boron tribromide (1.25 g, 5 mmol) was injected into this solution via a rubber septum. The temperature was then allowed to gradually warm to room temperature overnight. Products were isolated from the dark orange solution (ether, 5% aqueous $NaHCO_3$, $MgSO_4$), giving a quantitative yield of the diastereomeric phenolic nitriles (**17a** and **17b**) as a pale yellow, viscous oil. The more mobile and crystalline isomer (erythro, **17a**) was isolated by dry column chromatography (silica gel; CH_2Cl_2). The first fractions were found to contain 175 mg (31%) of **17a**, which was recrystallized from hexane.

The remaining fractions from the column contained the less mobile isomer (threo, **17b**), as well as a small fraction of **17a**.

3-(*p*-Carboxyphenyl)-4-(*p*-hydroxyphenyl)hexane [Erythro (18a) and Threo (18b) Isomers]. The isomeric nitriles **17a** and **17b**, previously separated by dry column chromatography, were each taken up in 2 mL of 50% potassium hydroxide solution and 2 mL of ethanol. The solution was allowed to stand for 2 days and then refluxed for 5 h and acidified with HCl. Products were isolated (ether, $MgSO_4$). The acid **18a**, corresponding to the more mobile, crystalline nitrile **17a**, crystallized slowly from methanol–water, (mp 155–157 °C). Recrystallization from the same solvent system raised the melting point to 164–166 °C (lit.[12] 166–168 °C). The other isomer (**18b** from **17b**) did not crystallize. Both isomers showed essentially identical mass spectra: *m/e* 298 (3, M^+), 135 (100), 107 (39).

The erythro acid **18a** was also prepared by debenzoylation of **29** (K_2CO_3, methanol, 25 °C, 18 h) in 96% yield.

***erythro*-3-[*p*-(Ethylimidato)phenyl]-4-(*p*-hydroxyphenyl)hexane Hydrochloride (19).** A small test tube with side arm was charged with 55 mg (0.2 mmol) of 3-(*p*-cyanophenyl)-4-(*p*-hydroxyphenyl)hexane (**17a**) and 0.5 mL each of absolute ethanol and chloroform. The side arm was fitted with a $CaCl_2$ drying tube and the solution was saturated at 0 °C with dry HCl gas for 1 h. The resulting slightly yellow solution was allowed to stand at room temperature for 2 days. Removal of the solvent under vacuum left, after trituration with ether, 48 mg (75%) of white crystals melting at 166–168 °C with gas evolution. Recrystallization from ethanol–ether raised the melting point to 169.5–171 °C (gas evolution): mass spectrum (10 eV), *m/e* 325 (6.0, M^+), 324 (1), 323 (4), 192 (13), 191 (100), 135 (17).

***erythro*-3-[*p*-(Aminomethyl)phenyl]-4-(*p*-hydroxyphenyl)hexane (20).** A solution of 24 mg (0.086 mmol) of the crystalline nitrile **17a** in 5 mL of absolute ethanol saturated with ammonia was hydrogenated at atmospheric pressure with ca. 20 mg of Raney nickel W-2. After removal of the catalyst and solvent, the residue was taken up in hot $CHCl_3$ and filtered clear of some insoluble material. Crystallization of the product from $CHCl_3$–hexane gave 17 mg (70%) of colorless crystals, mp 168–170 °C.

***erythro*-3-Phenyl-4-(4-hydroxyphenyl)hexane (Deoxyhexestrol) (21a). Method 1. Via the 1-Phenyl-1*H*-tetrazole (22c).** Hexestrol (5 g, 18.5 mmol) was refluxed with 3.7 g (20.5 mmol) of 5-chloro-1-phenyl-1*H*-tetrazole and 5.15 g of potassium carbonate in 50 mL of acetone for 17 h. Solvent was removed and the residue was washed with water and dried (9.18 g). This material, shown to be a 1:2:1 mixture of hexestrol, monoalkylated (**22c**) and dialkylated material (**22d**) by TLC, was directly subjected to hydrogenolysis in a Parr shaker with 5% palladium on charcoal in ethanol at 37–37 psi at 35 °C. The product mixture was separated by column chromatography (silica gel, petroleum ether–ether gradient from 4:1 to 1:1). First to elute was *meso*-3,4-diphenylhexane (**21b**); mp 90–91 °C from hexane, lit.[13,20] 89 and 89–91 °C). The second component (1.79 g, 38% based on starting material and 47% based on consumed hexestrol) was the desired deoxyhexestrol (**21a**), which was purified by recrystallization from hexane–benzene, mp 148–149 °C (lit.[20] 143–144 °C).

Method 2. Via the Monomethanesulfonate (22a). A solution of 16.0 g (59.3 mmol) of hexestrol in 100 mL of pyridine was treated dropwise with 8.2 g (71.2 mmol) of methanesulfonyl chloride. After the solution was stirred for 6 h at ambient temperature, an additional portion of methanesulfonyl chloride (1.5 g, 13.1 mmol) was added, followed by heating at 70 °C for 7 h. The reaction mixture was quenched with water, and the solvent was removed. Product isolation (EtOAc–$CHCl_3$) furnished 24.2 g of a crude solid, comprising an approximate 1:2:1 ratio of the dimethanesulfonate **22b**, monomethanesulfonate **22a**, and starting hexestrol by TLC.

The crude mixture of hexestrol methanesulfonates was dissolved in 120 mL of ethanol, treated with 28.5 g (282 mmol) of freshly distilled triethylamine and 2.1 g of 5% palladium–charcoal catalyst, and subjected to hydrogenolysis in a Parr shaker at 45 °C, 45–50 psi hydrogen pressure. After 12 h, the catalyst was filtered and the solvent was evaporated to provide 7.60 g of a product mixture, consisting of **21b**, **21a**, and 1 by TLC. Dry column

chromatography (SiO_2, 2:1 petroleum ether–ether) afforded as the middle fraction 4.60 g (35%, based upon consumed hexestrol) of a colorless crystalline product identified as **21a**.

***erythro*-3-(4-Hydroxyphenyl)-4-(4-nitrophenyl)hexane (Nitrodeoxyhexestrol) (24a) via the Nitrobenzenesulfonate 23c.** A solution of 4.50 g (17.7 mmol) of deoxyhexestrol (**21a**) and 15.67 g (53.1 mmol) of 75% *p*-nitrobenzenesulfonyl chloride was dissolved in 100 mL of pyridine and heated to 70 °C. After 12 h, water was added, and the solvent was removed azeotropically at reduced pressure. Product isolation (EtOAc, 10% aqueous HCl, Na_2SO_4) provided 7.53 g (97%) of a colorless crystalline product identified as the *p*-nitrobenzenesulfonate **23c**: mp (from benzene–hexane) 167–168 °C.

Finely powdered cupric nitrate trihydrate (6.60 g, 27.3 mmol) was added quickly to a solution of 4.00 g (9.11 mmol) of the nitrobenzenesulfonate **23c** in 40 mL of acetic anhydride. The mixture was heated to 80 °C, stirred rapidly for 24 h, and then cooled. Product isolation (EtOAc, $MgSO_4$) gave 4.01 g of a solid that upon further purification by column chromatography (silica gel, 1:1 benzene–hexane) yielded 3.55 g (81%) of the nitrodeoxyhexestrol *p*-nitrobenzenesulfonate **24c**: mp (from ethanol–water) 201–202 °C; IR (KBr) 1540 (NO_2), 1382 (SO_2), 1355 (NO_2), 875 (SO), 860 cm^{-1}; ^1H NMR ($CDCl_3$) δ 6.82–7.33 (m, 8 H, aromatic), 7.88–8.40 (m, 4 H, aromatic); mass spectrum, *m/e* 322 (7), 321 (18), 320 (100), 292 (20), 186 (11), 165 (20), 164 (2), 135 (5), 134 (18), 133 (16), 122 (18), 91 (9).

A solution of 2.00 g (4.13 mmol) of the nitrodeoxyhexestrol nitrobenzenesulfonate (**24c**) in 200 mL of methanol was treated with 2.56 g (18.6 mmol) of potassium carbonate and stirred rapidly at room temperature. After 2 h, the reaction was quenched with water and acidified with 10% aqueous hydrochloric acid. Product isolation (EtOAc, $MgSO_4$) gave 1.00 g (81%) of a solid that was purified further by column chromatography (silica gel, 1:1 petroleum ether–ether). A mobile fraction (90 mg; 7%) was identified as *erythro*-3-(4-hydroxy-3-nitrophenyl)-4-phenylhexane: mp 117–119 °C; IR (neat) 3400 (OH), 1530 (NO_2), cm^{-1}. The major fraction gave 860 mg (70%) of a yellow gum which failed to crystallize despite persistent efforts; this material was identified as *p*-nitrodeoxyhexestrol (**24a**).

***erythro*-3-(4-Aminophenyl)-4-(4-hydroxyphenyl)hexane (25a).** A mixture of 100 g (0.334 mmol) of *p*-nitrodeoxyhexestrol (**24a**) and 35 mg of 83% platinum oxide catalyst in 4 mL of absolute ethanol was stirred under a hydrogen atmosphere at 45 °C and ambient pressure. After 12 h, 24 mL (1.00 mmol) of hydrogen was consumed and uptake was stopped. Following removal of the catalyst by filtration, the solvent was concentrated to dryness, affording a quantitative yield of crude crystalline product. Chromatography on a micro Florisil column (ether) and recrystallization from aqueous methanol furnished 77 mg (86%) of a colorless solid, which became tan upon standing.

***erythro*-3-(4-Azidophenyl)-4-(4-hydroxyphenyl)hexane (Azidodeoxyhexestrol; 25b).** A solution of aminodeoxyhexestrol (**25a**; 55 mg, 0.204 mmol) in 4.4 mL of acetone and 1.5 mL of 1 N hydrochloric acid was cooled to 0 °C and diazotized in the dark with 14 mg (0.204 mmol) of sodium nitrite in 60 μL of water. After 30 min, the cold solution of diazonium chloride was added to 133 mg (20.4 mmol) of sodium azide in 3 mL of water and the solution was overlayed with 30 mL of ether and stirred in the dark for 30 min. Product isolation (Et_2O, Na_2SO_4) gave a crude yellow oil which crystallized upon standing. Recrystallization from methanol–water afforded 42 mg (70%) of *p*-azidodeoxyhexestrol (**25b**).

***erythro*-3-(4-Hydroxyphenyl)-4-[4-(fluorosulfonyl)phenyl]hexestrol (26b).** Deoxyhexestrol (**21a**; 246 mg, 0.969 mmol) was suspended with efficient magnetic stirring in 2.5 mL of a solution of trimethylamine (2.0 g, 5.00 mmol) in dry benzene. The mixture was cooled to 0 °C and flushed with nitrogen, and 407 mg (1.94 mmol, 274 μL) of trifluoracetic anhydride was added dropwise. When the addition was complete, the reaction was stirred for 15 min at 0 °C, followed by gradual warming to room temperature over 50 min. The unreacted anhydride was removed by rotary vacuum evaporation at room temperature with three successive 10-mL portions of carbon tetrachloride, providing 332 mg (98%) of the trifluoroacetate (**23d**) as a white crystalline solid: mp 85–87 °C; IR (liquid film) 1670 (C=O), 1485, 1200, 1140 (CF) cm^{-1}. This material was used immediately in subsequent reactions without further purification.

Fluorosulfonic acid (500 μL, 8.72 mmol) was cooled to 0 °C and added dropwise to 182 mg (0.538 mmol) of deoxyhexestrol trifluoroacetate (prepared above) (**23d**), which was suspended in a dry ice–carbon tetrachloride bath (−23 °C). When the addition was complete, traces of undissolved fluoroacetate were reacted by momentary warming to 0 °C, resulting in a clear yellow solution. The mixture was allowed to warm gradually to 0 °C over 30 min and was quenched while cold by careful dropwise addition into ice–water. Product isolation (Et_2O, 10% aqueous $NaHCO_3$, $MgSO_4$) gave 145 mg of an oily residue consisting of a mixture of the acetylated and deacetylated sulfonyl fluoride (**26a, 26b**), along with a less mobile component consistent with the *o*-(fluorosulfonyl)phenol. Complete deprotection was effected by hydrolyzing the product mixture in 20 mL of moist ether for 12 h at 4 °C. Chromatography on preparative TLC (silica, 2:1 petroleum ether–ether, three developments) and elution of the more mobile fraction provided 65 mg (36%) of a clear oil identified as *p*-(fluorosulfonyl)deoxyhexestrol (**26b**).

***erythro*-3-[*p*-(Chlorosulfonyl)phenyl]-4-(*p*-hydroxyphenyl)hexane (26d).** Chlorosulfonic acid (3.0 mL, 45.1 mmol) was cooled to 0 °C and added dropwise to 540 mg (1.54 mmol) of deoxyhexestrol trifluoroacetate (**23d**), which was suspended in a dry ice–carbon tetrachloride bath (−23 °C). After addition was completed, occasional warming to 0 °C was necessary to prevent freezing and to solubilize traces of undissolved trifluoroacetate, resulting in an orange-yellow solution. The mixture was stirred with gradual warming to 0 °C over 30 min and quenched while cold by careful dropwise addition into ice–water. Product isolation (Et_2O, $MgSO_4$) gave 550 mg of an oily residue comprised of a mixture of the acetylated and deacetylated sulfonyl chlorides (**26c, 26d**) by silica TLC (2:1 hexane–ether), accompanied by a less mobile component. Complete deprotection was achieved by hydrolyzing the product mixture in 30 mL of moist ether for 12 h at 4 °C. Chromatography on a micro Florisil column (2:1 petroleum ether–ether) provided 218 mg (40%) of a gummy solid, which was identified as the sulfonyl chloride (**26d**). This material was immediately used in subsequent reactions without further purification.

***erythro*-3-[*p*-(Azidosulfonyl)phenyl]-4-(*p*-hydroxyphenyl)hexane (26e).** To a solution of 242 mg (0.69 mmol) of *p*-(chlorosulfonyl)deoxyhexestrol (**26d**) in 6 mL of acetone at 40 °C was added a solution of 102 mg (1.57 mmol) of sodium azide in 600 μL of 1:1 acetone–water. Following efficient mixing of the reagents, additional water was added as necessary to maintain one phase. After stirring for 2 h at room temperature, the mixture was poured into 2 mL of water and filtered. Product isolation (Et_2O, 10% aqueous $NaHCO_3$, Na_2SO_4) provided 180 mg (73%) of crude oily product. Chromatography on preparative TLC (SiO_2, 2:1 petroleum ether–ether) afforded a more mobile pale yellow gummy solid, which after two recrystallizations from aqueous methanol gave 100 mg (41%) of deoxyhexestrol-*p*-sulfonyl azide (**26e**).

***erythro*-3-[*p*-(Benzoyloxy)phenyl]-4-phenylhexane (23e).** Deoxyhexestrol (**21a**; 0.44 g, 1.73 mmol) was treated with 0.40 g (2.8 mmol) of benzoyl chloride and 2 mL of pyridine at room temperature overnight. Product isolation (ether, $MgSO_4$) gave 0.88 g of material which was purified by dry column chromatography (CH_2Cl_2) on 50 g of silica gel, furnishing 0.56 g of the benzoate **23e** (90%; mp 105–108 °C). Further purification was by recrystallization from hexane: mp 115–116 °C; IR (KBr) 1733 (CO_2R), 1275, 1210, 703 cm^{-1}; ^1H NMR ($CDCl_3$) δ 0.56 (t, J = 7 Hz, 6 H, CH_3), 1.4 (m, 4 H, CH_2), 2.6 (m, 2 H, CH), 7.0–8.3 (m, 14 H, aromatics); mass spectrum, *m/e* 358 (1, M^+), 240 (15), 239 (69), 105 (100), 91 (14), 77 (19).

***erythro*-3-[*p*-(Benzoyloxy)phenyl]-4-(*p*-acetylphenyl)hexane (27b).** To a suspension of 601 mg (4.51 mmol) of aluminum chloride in 5 mL of carbon disulfide were added 216 mg (2.75 mmol) of acetyl chloride in 7 mL of carbon disulfide and 781 mg (2.18 mmol) of the benzoate **23e** in 10 mL of carbon disulfide. The mixture was stirred under reflux for 7 h and then quenched with water. Product isolation (Et_2O, $MgSO_4$) gave 866 mg of crude product. Purification by dry column chromatography on 60 g of silica gel (CH_2Cl_2) gave 238 mg of unreacted benzoate **23e** (29%) and 572 mg of the acetyl benzoate **27b** (65% based on starting benzoate, 92% based on consumed benzoate). The product was recrystallized from ethanol.

***erythro*-3-[*p*-(Benzoyloxy)phenyl]-4-[*p*-(chloroacetyl)-phenyl]hexane (27c).** To a suspension of 0.75 g (5.60 mmol) of aluminum chloride in 15 mL of carbon disulfide were added 0.48 g (4.20 mmol) of chloroacetyl chloride in 5 mL of carbon disulfide and 1.00 g (2.80 mmol) of the benzoate **23e** in 25 mL of carbon disulfide. The reaction mixture was stirred under reflux for 6 h. Product isolation (ether, $MgSO_4$) gave 1.77 g of oil, which was separated by dry column chromatography in 130 g silica gel using CH_2Cl_2. The more mobile compound was unreacted benzoate **23e** (0.30 g, 30%) and the less mobile compound was the chloroacetylated benzoate **27c** (802 mg, 66% based on starting benzoate; 94% based on consumed benzoate).

***erythro*-3-(*p*-Hydroxyphenyl)-4-(*p*-acetylphenyl)hexane (27d).** The benzoate **27b** (478 mg, 1.19 mmol) was refluxed for 3 h in 2% ethanolic potassium hydroxide. Saturated salt solution was added, and product isolation (ethyl acetate, $MgSO_4$) gave 482 mg of a solid that was recrystallized from ethanol, furnishing 342 mg of **27d** (97%): mp 156–157 °C (lit.[12] 159–160 °C).

***erythro*-3-(*p*-Hydroxyphenyl)-4-[*p*-(chloroacetyl)-phenyl]hexane (27e).** The benzoate of chlorocarbonyl compound **27c** (300 mg, 0.69 mmol) was suspended in 10 mL of methanol and mixed with 5 mL of methanol saturated with hydrogen chloride gas. The resulting solution was refluxed for 9 h and the solvent was then evaporated under reduced pressure. The residue (0.47 g) was purified by dry column chromatography on 40 g of silica gel (light petroleum ether–ether, 10:1 increasing to 1:1) to give 226 mg (99%) of the chloro ketone **27e**. Recrystallization from 2-propanol gave an analytical sample.

***erythro*-3-(*p*-Hydroxyphenyl)-4-[*p*-(bromoacetyl)-phenyl]hexane (27f).** The phenolic carbonyl compound **27d** (249 mg, 0.84 mmol) in 20 mL of ethyl acetate was treated with 254 mg of finely ground cupric bromide under reflux for 9 h and then treated with water. Product isolation (ether, $MgSO_4$) gave 377 mg of residue, which was purified on a Lipidex 1000 chromatography column (1 × 47 cm, 7:3 hexane–chloroform) to give 211 mg (67%) of bromocarbonyl compound **27f**. Recrystallization from methanol–H_2O gave an analytical sample.

***erythro*-3-[*p*-(Benzoyloxy)phenyl]-4-(*p*-carboxy-phenyl)hexane (29).** The chlorocarbonyl compound **27c** (2.0 g, 4.60 mmol) in 60 mL of dioxane was mixed with 60 mL of 1.4 M potassium hypochlorite solution (made from calcium hypochlorite, potassium carbonate, and potassium hydroxide) and stirred at 60 °C for 30 min. Acidification with 6 mL of 4 N HCl and addition of 10 mL of 20% aqueous sodium hydrogen sulfite (negative to starch–iodide test), followed by product isolation (Et_2O, $MgSO_4$), gave 3.24 g of crude product that was purified by dry column chromatography on 160 g of silica gel (elution, light petroleum ether–ether, 1:1) to give 1.75 g (94%) of carboxylic acid **29**. An analytical sample was obtained by recrystallization from benzene.

***erythro*-3-[*p*-(Benzoyloxy)phenyl]-4-[*p*-[(*o*-hydroxy-phenoxy)carbonyl]phenyl]hexane (30).** The benzoate **23e** (2.0 g, 5.59 mmol) was dissolved in 20 mL of dichloromethane, and 1.40 g (7.33 mmol) of catechol dichloromethylene ether[16] (bp 83–88 °C at 12 mmHg) in 10 mL of dichloromethane and 1.50 g (11.3 mmol) of aluminum chloride were added at 0 °C. The reaction mixture was stirred at 0 °C for 3 h and then quenched with water. Product isolation (ether, $MgSO_4$) gave 3.16 g of a solid, which was purified by dry column chromatography on 130 g of silica gel using CH_2Cl_2, furnishing 1.62 g of the acylated compound **30**. This material was purified by recrystallization from hexane–ethyl acetate.

***erythro*-3-(*p*-Hydroxyphenyl)-4-[*p*-(hydroxymethyl)-phenyl]hexane (31a).** The carboxyl benzoate **30** (200 mg, 0.497 mmol) in 10 mL of dry tetrahydrofuran was reduced with 60 mg (1.58 mmol) of lithium aluminum hydride at 0 °C. The reaction was quenched with and acidified with concentrated HCl. Product isolation (ether, $MgSO_4$) gave 317 mg of crude product. Purification by dry column chromatography on 20 g of silica gel (petroleum–ether, 1:1) gave 123 mg (87%) of phenolic benzyl alcohol **31a**, which was recrystallized from benzene.

This material was also prepared by lithium aluminum hydride reduction of the hydroxyphenyl acid **29** in 76% yield. An analytical sample was obtained by recrystallization from benzene: mp 165–167 °C (lit.[20] 164–166 °C).

***erythro*-3-(*p*-Hydroxyphenyl)-4-[*p*-(bromomethyl)-phenyl]hexane (31b).** Gaseous hydrogen bromide was bubbled through a solution of 470 mg (1.65 mmol) of the benzylic alcohol **31a** in 15 mL of ethyl acetate at room temperature for 5 min. The reaction mixture was washed with water and dried over magnesium sulfate to give 848 mg of crude product. Purification by dry column chromatography on 50 g of silica gel (chloroform) gave 313 mg (89%) of the bromo compound **31b**, which was recrystallized from hexane.

***erythro*-3-(*p*-Hydroxyphenyl)-4-[*p*-(chloromethyl)-phenyl]hexane (31c).** Hydrogen chloride gas was bubbled into 10 g of 100% ethanol cooled in ice. Enough gas (3.66 g) was absorbed to make 15 mL of solution. The benzylic alcohol **31a** (200 mg, 0.703 mmol) was dissolved in 5 mL of ethanol, and 5 mL of the above mentioned ethanolic hydrogen chloride solution (containing 1.22 g of HCl gas) was added. The mixture was refluxed, and solvent was evaporated under reduced pressure. The residue (0.33 g) was dissolved in petroleum ether–ether (5:1) and purified by dry column chromatography on 40 g of silica gel (5:1 petroleum ether–ether). The benzyl chloride **31c** obtained (204 mg, 96%) was recrystallized from hexane.

***erythro*-3-(*p*-Hydroxyphenyl)-4-[*p*-(ethoxymethyl)-phenyl]hexane (31d).** A solution of 200 mg (0.703 mmol) of the benzylic alcohol **31a** in 10 mL of ethanol and 2 mL of aqueous hydriodic acid (48%) was refluxed for 9 h. Addition of aqueous sodium thiosulfate, followed by product isolation (ether, $MgSO_4$), gave 346 mg of crude product, which was purified by dry column chromatography on 40 g of silica gel (eluted with 5:1 hexane–ether) and recrystallized from hexane to afford 181 mg (82%) of **31d**.

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