mice without involving either a cell mediated or humoral immune response.[24] It was found that in order to inhibit MLV in mice, small doses of polymer had to be administered daily; a cumulative high dose injected once was ineffective.[24] Thus, circulating polymer was required for antiviral activity to be expressed. This observation coupled with those reported here provides some information concerning the active form/site of these compounds. We have shown that poly(vA) accumulates in the spleen of mice, an organ where murine leukemia virus replicates. Furthermore, by the parameters examined (electric charge and molecular weight distribution), this accumulated polymer is very similar to the administered control even after 3 weeks. Thus, the polymer contained in mouse organs should sustain its antiviral activity. However, as mentioned above, daily administration of polymer was required for virus inhibition. These results show that the accumulation of polymer in tissues is accompanied by its gradual conversion to an inactive form. This conversion may be explained by the hypothesis that the polymer is segregated into certain cells within the tissue or into compartments within the cell, away from primary sites of viral replication.

Conclusion

Judged by the rate of uptake of polymers into cells in culture and their relatively rapid distribution to different tissues in mice, it appears that the macromolecular nature of drugs does not slow down their action drastically. However, to obtain optimal pharmacological activity with electroneutral polymers, compounds with a short half-life (greater than 1 h but not over 1 day) should be synthetized. Nondegradable polymers, even when nontoxic, are without any notable advantage as they lose their biological potency in animals probably by being segregated into different cells or into subcellular components where they cannot influence virus replication.

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Disteroidyl Ethers. 1. Synthesis and Oral Long-Lasting Uterotrophic Activity of 1,3,5(10)-Estratrien-17-yl Enol Ethers of 3-Keto Steroids

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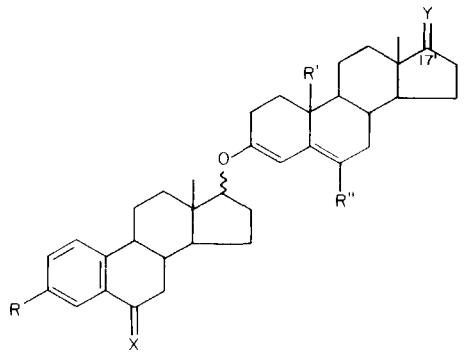
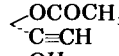
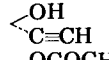
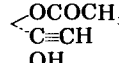
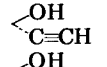
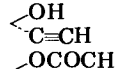
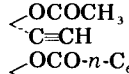
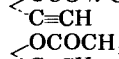
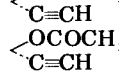
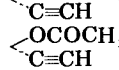
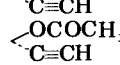
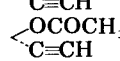
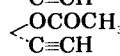
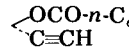
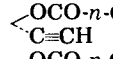
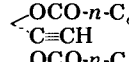
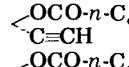
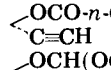
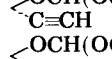
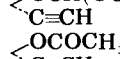
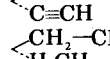
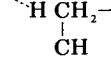
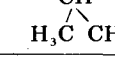
A series of disteroidyl enol ethers derived from a 17-hydroxyestratriene and a 3-keto steroid has been synthetized and tested for prolonged uterotrophic activity after oral treatment. Most of the compounds derived from 17β-estradiol displayed a high, prolonged activity, many of them being more active than quinestrol.

In the last 15 years many steroid ethers have been investigated in our laboratories and several of them proved to be outstanding for their oral activity. In particular, compounds like 3-cyclopentyl ethers of ethinylestradiol and other estrogens,[1] as well as estradiol 17-cycloalkenyl ethers,[2] displayed a quite unusual oral long-lasting activity.

Table I

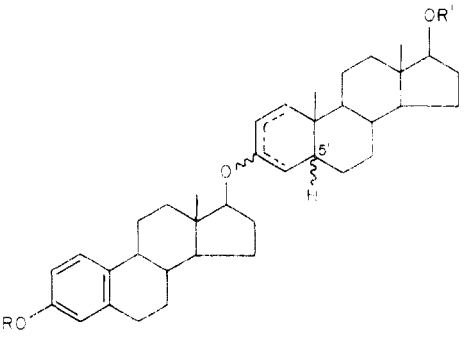


No.	C(17)	R	X	R'	R''	Y	Method[a]	Yield,[b] %	Mp, °C	[α]D	Formula	Analyses
1	β	HO	H_2	CH_3	H	O	C	86	230-232	-22.5	$C_{37}H_{48}O_3$	C, H
2	β	C_6H_5COO	H_2	CH_3	H	O	A	73	276-279	25	$C_{44}H_{52}O_4$	C, H
3	β	HO	H_2	CH_3	H		D	65	270-274	-54	$C_{37}H_{50}O_3$	C, H
4	β	C_6H_5COO	H_2	CH_3	H		B	82	221-224	-58	$C_{47}H_{58}O_5$	C, H
5	β	HO	H_2	CH_3	H		C	82	236-239	-67	$C_{38}H_{52}O_3$	C, H
6	β	C_6H_5COO	H_2	CH_3	H		A	54	241-243	-50	$C_{47}H_{58}O_5$	C, H
7	β	HO	H_2	H	H	O	C	80	290-293	-32.5	$C_{36}H_{46}O_3$	C, H
8	β	C_6H_5COO	H_2	H	H	O	A_2	88	283-286	33	$C_{43}H_{50}O_4$	C, H
9	β	$HOCOCH_2Cl_2COO$	H_2	H	H	O	E	59	170-173[c]	-19.5	$C_{40}H_{52}O_6$	C, H
10	β	C_5H_9O[d]	H_2	H	H	O	B	55	206-209	-31.7	$C_{41}H_{54}O_3$	C, H
11	β	HO	H_2	H	H		C	87	238-241	-67.2	$C_{36}H_{48}O_3$	C, H
12	β	C_6H_5COO	H_2	H	H		A_1	72	208-211	-65.4	$C_{45}H_{54}O_5$	C, H
13	β	C_6H_5COO	H_2	H	H		A	60	218-223	-14.5	$C_{50}H_{50}O_5$	C, H
14	β	C_5H_9O[d]	H_2	H	H		D	82	134-137	-60	$C_{41}H_{56}O_3$	H[e]
15	β	C_5H_9O[d]	H_2	H	H		E_1	63	158-160[f]	-60	$C_{45}H_{60}O_6$	C, H
16	β	C_6H_5COO	H_2	CH_3	H		A	40	250-253	-10.7	$C_{46}H_{56}O_4$	C, H
17	β	C_6H_5COO	H_2	CH_3	H		A_1	82	274-278	-72	$C_{48}H_{58}O_6$	C, H
18	β	C_6H_5COO	H_2	CH_3	CH_3		A_1	68	268-271	-88.5	$C_{49}H_{60}O_6$	C, H
19	α	C_6H_5COO	H_2	H	H		A_1	38	244-247	-37.2	$C_{45}H_{54}O_4$	C, H
20	β	C_6H_5COO	H_2	H	H		A_1	72	237-241	-11[g]	$C_{45}H_{54}O_4$	C, H

21	β	H	H_2	H	H		A_1	30	230-234	−95.6	$C_{40}H_{50}O_3$	C, H
22	α	HO	H_2	H	H		C	71	173-175	−162.5	$C_{38}H_{48}O_3 \cdot 0.5H_2O$	C[h]
23	α	C_6H_5COO	H_2	H	H		A	70	262-265[i]	−132	$C_{47}H_{54}O_5$	C, H
24	β	HO	H_2	H	H		C	75	260-263[j]	−119	$C_{38}H_{48}O_3 \cdot H_2O$	C, H
25	β	CH_3COO	H_2	H	H		E	95	281-283	−113.5	$C_{40}H_{50}O_4 \cdot H_2O$	C, H
26	β	HO	H_2	H	H		C_1	88	281-284[k]	−111	$C_{40}H_{50}O_4$	C, H
27	β	HO	H_2	H	H		C_1	89	233-236	−98.5	$C_{45}H_{60}O_4$	C, H
28	β	CH_3COO	H_2	H	H		A_1	71	200-203	−110	$C_{42}H_{52}O_5$	C, H
29	β	n-C_3H_7COO	H_2	H	H		A_1	65	176-178	−109	$C_{44}H_{56}O_5$	C, H
30	β	n-$C_6H_{13}COO$	H_2	H	H		A_1	53	156-159	−101	$C_{47}H_{62}O_5$	H[l]
31	β	C_6H_5COO	H_2	H	H		A_1	74	231-234	−101	$C_{47}H_{54}O_5$	C, H
32	β	C_5H_9O[d]	H_2	H	H		A_1	59	227-230	−108	$C_{45}H_{58}O_4$	C, H
33	β	$CH_3CH(OC_2H_5)O$	H_2	H	H		F	25	185-188	−104.5	$C_{44}H_{58}O_5$	C, H
34	β	CH_3COO	H_2	H	H		A	63	140-143[m]	−92.5	$C_{47}H_{62}O_5$	C, H
35	β	i-C_3H_7COO	H_2	H	H		E	81	154-157[n]	−92	$C_{49}H_{66}O_5$	C, H
36	β	C_6H_5COO	H_2	H	H		A	62	137-141[o]	−80.5	$C_{52}H_{64}O_5$	C, H
37	β	CH_3O	H_2	H	H		A	45	127-130	−94	$C_{46}H_{62}O_4$	C, H
38	β	C_5H_9O[d]	H_2	H	H		A	42	170-173[p]	−91	$C_{50}H_{68}O_4$	C, H
39	β	CH_3COO	H_2	H	H		F	70	126-131	−98	$C_{40}H_{58}O_5$	C, H
40	β	$CH_3CH(OC_2H_5)O$	H_2	H	H		F	45	128-131[q]	−10.5	$C_{46}H_{64}O_5$	C, H
41	β	C_6H_5COO	O	H	H		A_1	25	238-241	−102.3	$C_{47}H_{52}O_6$	C, H
42	β	C_6H_5COO	H_2	H	H		A	75	193-196[r]	−41	$C_{52}H_{70}O_3$	C, H

[a] See the Experimental Section for the letter that corresponds to each synthetic method. [b] Yield is of analytically pure material and based, for methods A–B, on the enol ether or acetal used. No attempts were made to maximize yields. [c] With softening at 115 °C. [d] C_5H_9O denotes cyclopentyloxy. [e] C: calcd, 82.50; found, 82.98. [f] With softening at 123 °C. [g] Determined in Py. [h] H: calcd, 8.79; found, 9.25. [i] With softening at 255 °C. [j] With softening at 225 °C. [k] With softening at 270 °C. [l] C: calcd, 79.84; found, 79.35. [m] With softening at 120 °C. [n] With softening at 150 °C. [o] With softening at 120 °C. [p] With softening at 164 °C. [q] With softening at 120 °C. [r] With softening at 188 °C.

Table II



No.	R	R'	C(5')	Δ^a	Method[b]	Yield,[c] %	Mp, °C	$[\alpha]_D$	Formula	Analyses
43	C_6H_5CO	CH_3CO	α	1', 3'	A_1	58	270-275	+41.6	$C_{46}H_{56}O_5$	C, H
44	C_6H_5CO	C_2H_5CO	α	2'	B	80	195-199	+52	$C_{47}H_{60}O_5$	C, H
45	C_6H_5CO	C_2H_5CO	β	3'	A	30	138-142	+83	$C_{47}H_{60}O_5$	C, H
46	H	H	α		C	97	275-278	+26.1	$C_{37}H_{54}O_3$	C, H
47	C_6H_5CO	C_2H_5CO	α			75	232-234	+18	$C_{47}H_{62}O_5$	C, H
48	H	H	β		C	98	292-294	+56.2	$C_{37}H_{54}O_3$	C, H
49	C_6H_5CO	C_2H_5CO	β			68	216-218	+52.3	$C_{47}H_{62}O_5$	C, H

[a] Ring A' unsaturation. [b,c] See footnotes *a* and *b*, respectively, in Table I.

The ability of cycloalkyloxy groups to strongly affect the pharmacokinetics of the molecules prompted us to extend the investigation to new classes of modified steroids, obtained by linking two or more steroid units by reversible ether linkages. Our working hypothesis was to consider the steroid units as both polycyclic lipophilic structures and biologically active entities, thus capable to give rise to molecules with a potential wide range of pharmacological properties.

Oligomeric steroids of different natures have been known for a long time. Steroids joined directly by a covalent bond have displayed no interesting pharmacological properties. The polysteroidal molecules which showed an actual or potential therapeutic utility, with the exception of some azines,[3] were compounds containing ester linkages.[4,5] Like monomeric esters, dimeric and polymeric esters were mainly remarkable for their parenteral, long-lasting activity. Taking into account our previous experience on steroid ethers, the objective of our program on di- and polysteroidyl ethers was to obtain new molecules not only parenterally but also orally long active, possibly endowed with selective biological profiles.

In this paper we report a first class of disteroidyl enol ethers derived from a 17-hydroxyestratriene and a 3-keto steroid.

Chemistry. Disteroidyl enol ethers were prepared by suitable modifications of procedures already described for the cycloalkenyl ethers of 17β-hydroxyestratrienes and androstanes.[6] Acid-catalyzed exchange etherification of the parent 17α- or 17β-hydroxyestratrienes with the ethyl enol ether of the appropriate Δ^4-3-keto steroids afforded the $\Delta^{3,5}$-diene structures (Table I). The same reaction with dimethyl acetals of 3-keto-5α- and 3-keto-5β-steroids gave rise to Δ^2-ene and Δ^3-ene structures, respectively, while a 1α,3,3-trimethoxy intermediate was used to obtain the $\Delta^{1,3}$-diene structure[7] (Table II). Reactions were carried out in benzene using pyridine *p*-toluenesulfonate (method A) or *p*-toluenesulfonic acid (method A_1) as catalyst, in toluene (method A_2), or in DMF (method B). Some compounds were converted into related derivatives by alkaline deacylation (methods C and C_1), ketone reduction (method D), acylation (methods E and E_1), etherification (method F), and catalytic hydrogenation (method G).

Biology and Discussion. The compounds have been tested for their oral prolonged uterotrophic activity in spayed rats; autopsies were performed 1 and 2 weeks after a single treatment. The results are given in Table III. The compounds not listed in Table III exhibited no significant activity even at a ten times higher dose.

Almost all the compounds derived from 17β-estradiol displayed a very prolonged activity, giving rise to uterus weights higher than those obtained after ethinylestradiol. Compounds **7–9**, **11**, **1**, and **24** exhibited a greater activity than quinestrol, a well-known oral long-lasting agent developed in our laboratories,[1,8,9] after 1 and 2 weeks. Other compounds, like **2**, **4**, **5**, **12**, **25**, **26**, **28**, **35**, and **36**, displayed an activity of the same order as quinestrol.

All the most active compounds are 3,5-dien-3-ol ethers and are derived from 3-ketoandrost-4-enes or 19-norandrost-4-enes. The few derivatives of 4,5-saturated 3-ketones prepared and investigated show little activity. Also compounds derived from 3-ketopregnanes or 19-norpregnanes, **16–18** and **20**, or from cholestenone, **42**, are not endowed with any appreciable oral long-lasting uterotrophic activity. The leading compounds are 19-norandrostenedione derivatives **7–9**, followed by derivatives of androstenedione, **1** and **2**, 19-nortestosterone, **11** and **12**, testosterone, **3** and **4**, methyltestosterone, **5**, and norethindrone, **24–26**, **28**, **35**, and **36**.

Dimeric steroids **19**, **22**, **23**, **21**, and **41**, derived from poorly active estrogens such as 17α-estradiol, 3-deoxyestradiol, and 6-ketoestradiol, displayed, as expected, a low uterotrophic activity.

Very likely the disteroidyl enol ethers act not as such but as prodrugs, after releasing the estrogenic moiety in a proper site and with a suitable timing. This is supported by the negligible activity displayed by the hardly reversible, saturated ethers **46–49**.

However, the activity order cannot be reasonably attributed to the intrinsic biological properties of the 3-keto steroid moiety. Androstanes and 19-norandrostanes are uterotrophic but norethindrone is also antiuterotrophic, like progesterone and related compounds. Moreover, the steroids employed as parent 3-ketones display their hormonal and antihormonal activity at dose levels not comparable with those of estradiol. Accordingly, in a separate trial, the uterotrophic activity of estradiol was not significantly modified by the concomitant administration

Table III. Oral Long-Lasting Uterotrophic Activity in Spayed Rats

Compd[a]	Uterus wt[c] After 1 week	Uterus wt[c] After 2 weeks
Controls	15.1 ± 0.51	15.9 ± 0.50
Ethinylestradiol	22.5 ± 0.55	20.5 ± 0.75
Quinestrol	39.0 ± 1.41	29.0 ± 0.93
1	56.9 ± 2.13	38.9 ± 1.89
2	53.0 ± 3.86	31.6 ± 0.92
3	50.2 ± 2.10	36.8 ± 1.15
4	49.4 ± 2.37	31.0 ± 0.91
5	48.1 ± 2.32	33.5 ± 1.14
6	26.4 ± 1.15	21.5 ± 0.60
7	71.7 ± 5.33	43.6 ± 2.41
8	59.8 ± 3.49	43.7 ± 2.58
9[b]	60.7 ± 5.37	36.8 ± 1.43
10	29.1 ± 0.95	26.2 ± 1.50
11	59.9 ± 2.48	41.6 ± 1.74
12	48.0 ± 3.18	31.6 ± 1.09
13	31.3 ± 1.78	18.1 ± 0.55
14	35.8 ± 3.89	27.1 ± 1.50
15[b]	28.9 ± 1.54	31.8 ± 3.85
16	26.3 ± 1.12	20.6 ± 0.45
17	27.5 ± 1.53	23.3 ± 1.06
18	26.6 ± 1.90	21.9 ± 0.52
20	29.1 ± 1.48	28.5 ± 2.29
24	47.8 ± 3.22	36.3 ± 1.22
24[b]	40.5 ± 1.23	23.3 ± 1.13
25	39.2 ± 2.62	32.7 ± 2.16
26	38.1 ± 2.26	26.7 ± 1.67
27	33.8 ± 1.09	22.1 ± 1.50
28	36.6 ± 1.97	25.4 ± 0.52
29	26.5 ± 1.20	18.3 ± 0.73
30	29.5 ± 1.18	18.9 ± 0.88
31	25.6 ± 1.28	21.4 ± 0.89
32	17.9 ± 0.46	
33	30.3 ± 2.04	19.3 ± 1.10
34	31.7 ± 2.68	28.7 ± 1.68
35	37.2 ± 2.48	30.4 ± 2.56
36	35.6 ± 2.17	24.2 ± 0.96
37	18.2 ± 0.57	15.4 ± 0.80
38	19.8 ± 0.84	15.2 ± 0.70
39	26.8 ± 1.88	21.6 ± 1.22
40	23.6 ± 0.80	18.0 ± 0.75
42	20.7 ± 1.30	20.8 ± 0.50
43	21.3 ± 0.87	20.1 ± 0.62
44	23.3 ± 0.84	21.3 ± 0.71
45	21.2 ± 0.53	19.9 ± 0.58

[a] Single administration of 0.1 μmol in sesame oil. [b] Single administration in suspending vehicle. [c] In milligrams, average ± SE from ten rats per group.

of an equimolecular amount of the same 3-keto steroids.

The nature of the functions at C-3 and C-17′ seems to play an important role in determining the activity of the disteroidyl ethers. Thus, among 19-norandrostenedione derivatives, free alcohol **7** exhibits a higher activity than benzoate **8**, while cyclopentyl ether **10** is by far less active. Also in the other series of disteroids derived from a same 3-ketone, free alcohols like **11**, **1**, **3**, **5**, and **24** are more active than the relevant esters and the still more lipophilic cyclopentyl ethers **32** and **38** show little activity.

The oral long-lasting activity of quinestrol[10] and of other 3- and 17-cycloalkenyl ethers of estrogenic steroids[2,11] was attributed to their lipophilic properties and their storage in body fat. Although also in the case of disteroidyl ethers the lipophilic properties may play a key role in determining the long-lasting activity, we may infer that the presence of a hydrophilic function improves their bioavailability.

The compounds were administered in oil solution, most of them being poorly active when given in fat-free vehicle, as already proved for other lipophilic compounds.[2] The presence of a hydrophilic group modifies this pattern, like in hemisuccinate **9**, highly active after administration in aqueous vehicle, and in alcohol **24**, considerably active in the same vehicle, although less than in oil.

Conclusions

The discovery of a great number of oral long-lasting estrogens may already justify some interest for this new class of disteroidyl ethers. However, the possibility of obtaining tailor made molecules by joining two different hormonal moieties was not proved. Only a deeper evaluation may define the possible manifold biological aspects of these molecules and emphasize the role of the nonestrogenic moiety. On the basis of this first screening for the uterotrophic activity and of the structure–activity relationships discussed above, we may infer that the ketonic moiety acts simply as a lipophilic structure able to strongly affect the pharmacokinetics of the active moiety. Its role is not significantly different from that of cycloalkanones previously investigated.[2] Accordingly, the chemical and physical properties of the disteroid, like ether lability and partition coefficient, seem to be more important than the intrinsic biological properties of the nonestrogenic component.

Experimental Section

All temperatures are uncorrected. Optical rotations were determined in dioxane, unless otherwise indicated, at 24 °C (*c* ~1). UV spectra were determined in dioxane with an Optica CF_4 spectrometer; IR spectra were measured in Nujol mull on a Perkin-Elmer 457 instrument. Absorption bands of these spectra were in agreement with the assigned structure. All analytical samples moved as a single spot on TLC using Fluorosil G plates. Where analyses are indicated only by symbols of the elements, analytical results obtained for those elements were within ±0.4% of the theoretical values.

General Methods of Preparation. A. 17β-(17′-Oxoandrosta-3′,5′-dien-3′-yloxy)estra-1,3,5(10)-trien-3-ol Benzoate (2). To a boiling solution of estradiol 3-benzoate (6.6 g) in C_6H_6 (3 l.) under anhydrous conditions, androst-4-ene-3,17-dione 3-ethyl enol ether (5 g) was added, followed by Py TsOH (50 mg). Heating was continued for 90 min, about two-thirds of the solvent being removed by distillation. After addition of a few drops of Py and concentration under reduced pressure to about 500 ml, the mixture was percolated through an Al_2O_3 (120 g) column, followed by fresh C_6H_6 (500 ml). After complete removal of the solvent in vacuo, the residue was taken up in MeOH, filtered, and crystallized from CH_2Cl_2–MeOH.

A_1. TsOH was used as the catalyst in method A.

A_2. $PhCH_3$ was used as the solvent in method A.

B. 17β-(17′β-Propionoxy-5′α-androst-2′-en-3′-yloxy)estra-1,3,5(10)-trien-3-ol Benzoate (44). Estradiol 3-benzoate (1.5 g), **50** (1.5 g), and TsOH (15 mg) in DMF (20 ml) were heated at 120–150 °C for 40 min and then at 180–200 °C for 20 min under a gentle stream of N_2. After addition of a few drops of Py and complete removal of the solvent in vacuo, the residue was worked up in the usual way.

C. 17β-(17′β-Hydroxy-19′-nor-17′α-pregna-3′,5′-dien-20′-yn-3′-yloxy)estra-1,3,5(10)-trien-3-ol (24). A suspension of **31** (11.9 g) in MeOH (250 ml) was treated with 5% KOH–MeOH (500 ml) and heated under reflux for 7 h. After concentrating under reduced pressure, the residue was diluted with H_2O, 10% HCl (160 ml) was added, and CO_2 was bubbled for 30 min through the mixture. The precipitate was filtered and crystallized from MeOH.

C_1. 17β-(17′β-Hydroxy-19′-nor-17′α-pregna-3′,5′-dien-20′-yn-3′-yloxy)estra-1,3,5(10)-trien-3-ol (26). A suspension of **31** (2 g) in THF (20 ml) and MeOH (80 ml) was treated with 10% K_2CO_3 aqueous solution (10 ml). After keeping under stirring at room temperature for 4 h, the mixture was poured into H_2O (300 ml). The precipitate was filtered and recrystallized from CH_2Cl_2–MeOH.

D. $NaBH_4$ (THF) reduction of the corresponding 17′-ketone.

E. The corresponding 3-hydroxy derivative was conventionally acylated in Py at room temperature by treatment with the proper anhydride.

E_1. Acylation as in method E of the corresponding 17′-hydroxy derivative.

F. **17β-[17′β-(1″-Ethoxyethoxy)-19′-nor-17′α-pregna-3′,5′-dien-20′-yn-3′-yloxy]estra-1,3,5(10)-trien-3-ol Acetate (39).** A solution of **25** (1 g) in anhydrous THF (10 ml) was treated with Py TsOH (10 mg) and ethyl vinyl ether (2 ml) and kept at room temperature for 15 h. After addition of a few drops of Py, the mixture was percolated through an Al_2O_3 (10 g) column. The solvent was then removed under reduced pressure and the residue crystallized from EtOH.

G. **17β-(17′β-Propionoxy-5′α-androstan-3′ξ-yloxy)estra-1,3,5(10)-trien-3-ol Benzoate (47).** To a solution of 44 (5 g) in anhydrous THF (300 ml), 10% Pd/C (5 g) was added and the resulting suspension was charged in an autoclave with 50 atm of H_2 and shaken for 24 h at room temperature. After removal of the catalyst by filtration, the mixture was percolated through an Al_2O_3 (20 g) column. The solvent was removed under reduced pressure and the residue crystallized from C_6H_6.

Intermediates. All enol ethers and acetals of 3-keto steroids, 3-esters, and 3-ethers of 17α- and 17β-estradiol required for preparation of compounds in Table I and II were obtained according to known procedures.[12] Among these, the following compounds appeared not yet described: 3,3-dimethoxy-5α-androstan-17β-ol propionate (**50**) [mp 135–138 °C; $[\alpha]_D$ +9.5°. Anal. ($C_{24}H_{40}O_4$) C, H]; 3,3-dimethoxy-5β-androstan-17-ol propionate (**51**) [mp 100–102 °C; $[\alpha]_D$ +17.3°. Anal. ($C_{24}H_{40}O_4$) C, H]; 3-ethoxyestra-3,5-dien-17β-ol benzoate (**52**) [mp 170–172 °C; $[\alpha]_D$ −74°. Anal. ($C_{27}H_{34}O_3$) C, H]; and estra-1,3,5(10)-trien-3,17α-diol 3-benzoate (**53**) [mp 157–159 °C; $[\alpha]_D$ +40°. Anal. ($C_{25}H_{28}O_3$) C, H].

The ethyl enol ether of norethindrone enanthate was not fully isolated and characterized.

Pharmacological Testing. Immature Wistar female rats, 23–25 days old, weighing 40–50 g, were spayed. On the day following surgery, the animals were gavaged by a single treatment. The autopsies were scheduled 1 and 2 weeks later. The uteri were separated from the vagina by cutting through the cervix and weighed fresh, on a torsion balance, after pressing out the intrauterine fluid.

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Bis-Basic-Substituted Polycyclic Aromatic Compounds. A New Class of Antiviral Agents.[1-3] 8. Bis-Basic Derivatives of Carbazole, Dibenzofuran, and Dibenzothiophene

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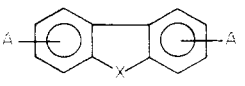
A series of bisalkamine esters, bis-basic ethers, and bis-basic ketones of carbazole, *N*-ethylcarbazole, dibenzofuran, and dibenzothiophene was synthesized and evaluated for antiviral activity. The series also included two bis-basic alkanes of *N*-ethylcarbazole and one bis-basic carboxamide of dibenzofuran. Structure–activity relationships indicated that within the carbazole and *N*-ethylcarbazole series the bisalkamine esters gave the most active derivatives while the bis-basic ketone derivatives of dibenzofuran and dibenzothiophene afforded the more potent compounds within the respective series. The [6,5,6] heterocyclic nuclei were compared with the [6,5,6] aromatic nuclei (fluorene and fluoren-9-one) including tilorone with respect to antiviral activity against encephalomyocarditis (EMC) virus. Maximum activity was associated with the bis-basic ketone side chain and fluoren-9-one nucleus.

Reports describing the antiviral activity of bisalkamine esters Ia,[4] bis-basic ethers Ib,[5] and bis-basic ketones Ic[6] of fluorene and fluorenone (IIa–c) led to the investigation of a variety of other bis-basic-substituted derivatives of various aromatic nuclei including anthraquinone,[7] fluoranthene,[8] and xanthene.[3] We now wish to report the



I, X = CH_2
II, X = C=O
III, X = NH
IV, X = NEt
V, X = O
VI, X = S

a, A = $CO_2(CH_2)_nNR_2$
b, A = $O(CH_2)_nNR_2$
c, A = $CO(CH_2)_nNR_2$
d, A = $(CH_2)_nNR_2$

synthesis and antiviral properties of a series of bis-basic derivatives of carbazole (III), *N*-ethylcarbazole (IV), dibenzofuran (V), and dibenzothiophene (VI).

Chemistry. The synthesis of bis-basic-substituted carbazoles is outlined in Scheme I. Carbazole-3,6-dicarboxylic acid prepared by the method of Preston et al.[9] and *N*-ethylcarbazole-3,6-dicarboxylic acid prepared by the method of Gilman and Spatz[10] were treated with 3-diethylaminopropyl chloride in the presence of a catalytic amount of benzyltrimethylammonium chloride to give the bisalkamine esters (Table I) **1** and **2**, respectively. The bis-basic ethers **3–6** (Table I) were prepared by the direct alkylation of *N*-ethylcarbazole-3,6-diol diacetate derived from 3,6-diacetyl-*N*-ethylcarbazole[11] via the Baeyer–Villiger oxidation reaction with *m*-chloroperbenzoic acid. This procedure, previously described for the synthesis of bis-basic ethers of fluoranthene,[8] was the method of choice since all attempts to isolate *N*-ethylcarbazole-3,6-diol