2-Phenylindenes: Development of a New Mammary Tumor Inhibiting Antiestrogen by Combination of Estrogenic Side Effect Lowering Structural Elements

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A new antiestrogenic, mammary tumor inhibiting 2-phenylindene was developed by the use of structural elements that we have shown to decrease estrogenic side effects but to increase antiestrogenic activity and retain the antitumor effect of certain stilbenes. The new 2-phenylindenes were synthesized from their corresponding methoxy-substituted 3,4-diphenylhexane-3,4-diols by cyclization with acetyl chloride and acetic anhydride and subsequent ether cleavage and acetylation. In this series, the 2-phenylindene derivative (compound **13**) with a 5,6,3′,4′-tetraacetoxy and a 1-methyl-3-ethyl substitution had the highest affinity for the estrogen receptor, the strongest antiestrogenic effect, and the lowest estrogenic effect. This compound was superior to the 2-phenylindenes with 5,3′-diacetoxy substitution or 1,1-dimethyl and 3-isopropyl moieties, respectively. Compound **13** exhibited a strong, significant inhibiting effect on the growth of the hormone-dependent MXT mouse mammary tumor without estrogenic side effects.

Tamoxifen, a nonsteroidal antiestrogen, is now routinely used for the treatment of the hormone-dependent mammary carcinoma.[1] However, this drug, like other antiestrogens, also displays weak estrogenic activity that may stimulate mammary tumor growth.[2] Therefore, the development of new antiestrogens with a strong mammary tumor inhibiting activity but with no or only very low estrogenic side effects is of great interest for the treatment of breast cancer.

During the last few years, we have obtained antiestrogens with a strong antitumor effect and diminished estrogenic properties by various modifications of synthetic estrogens. Transformation of diethylstilbestrol to 3,3′-dihydroxy-α,β-diethylstilbene or its acetoxy derivatives (**1**, Scheme I) led to antiestrogens with a strong tumor inhibiting activity but only low estrogenic side effects.[3-5] The introduction of additional acetoxy groups into the aromatic rings yielded the catechol compound **2** (Scheme I) with biological properties similar to those of **1**.[6,7] The transformation of diethylstilbestrol into its indene analogue indenestrol A (Scheme I) reduced the estrogenic activity.[9]

Therefore, we assumed that the 2-phenylindene analogue of **1** (**3**, Scheme I) also has a lower estrogenic potency.[8] However, the effects of **1** and **3** were comparable.[4,5,8]

Additionally, modifications of the alkyl substitution of the 2-phenylindene skeleton (especially the $C(CH_3)_2$ fragment) can be of interest since, in the class of 1,2-diphenylethanes, tetraalkylation in the 1- and 2-positions, e.g. 1,1,2,2-tetramethyl-1,2-bis(4-hydroxyphenyl)ethane, led to strong antiestrogens with almost no estrogenic effects.[10]

Scheme I

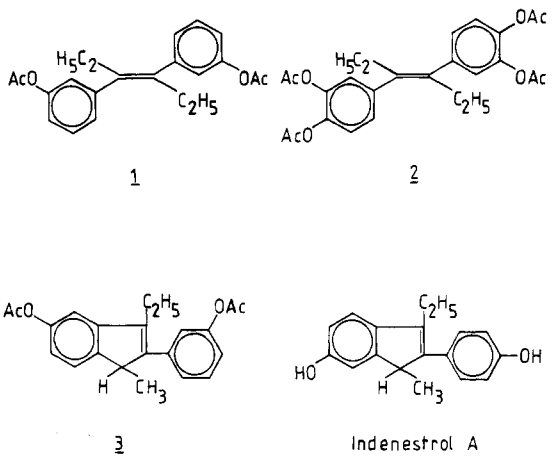
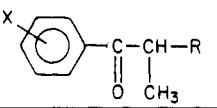


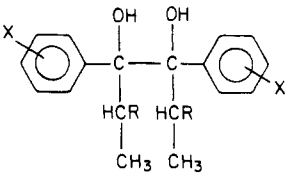
Table I. Substituted Phenylpropan-1-ones



no.	X	R	synth[a] method	yield, %	mp, °C	formula
4[b]	3,4-$(OCH_3)_2$	H	A	54	61	$C_{11}H_{14}O_3$
5[c]	3,4-$(OCH_3)_2$	CH_3	A	73	oil	$C_{12}H_{16}O_3$
6[d]	3-OCH_3	CH_3	B	60	oil	$C_{11}H_{14}O_2$

[a] Synthetic methods A and B under Experimental Section. [b] See ref 11. [c] See ref 12. [d] See ref 13.

Table II. Substituted 3,4-Diphenyl-3,4-hexanediols[a]



no.	X	R	yield, %	formula
7	3,4-$(OCH_3)_2$	H	85	$C_{22}H_{30}O_6$
8	3,4-$(OCH_3)_2$	CH_3	70	$C_{24}H_{34}O_6$
9	3-OCH_3	CH_3	75	$C_{22}H_{30}O_4$

[a] Synthetic method C under Experimental Section. The mixtures of the meso and racemic forms were not separated.

Consequently, the aim of this study was the combination of the above-listed structural elements in the class of 2-

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(7) Because of better handling and crystallization, we are using in some cases the acetoxy- instead of the hydroxy-substituted compounds.
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Scheme II

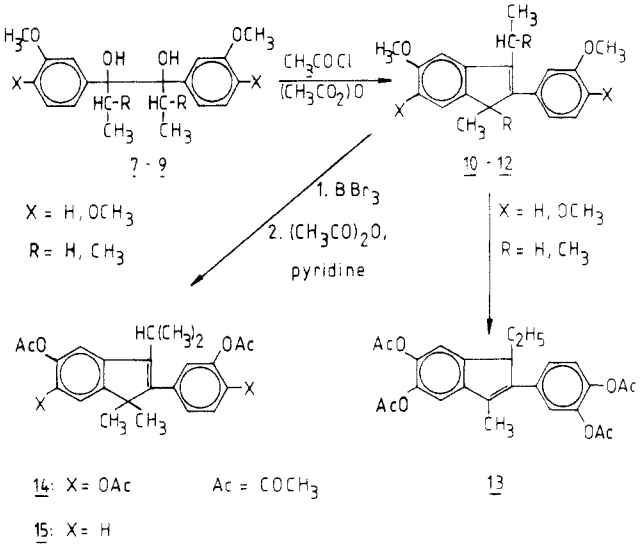
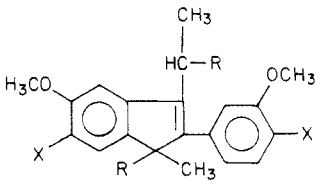


Table III. Methoxy Substituted 2-Phenylindenes[a]



no.	X	R	yield, %	mp,[b] °C	formula
10	OCH_3	H	60	108	$C_{22}H_{26}O_4$
11	OCH_3	CH_3	10	155	$C_{24}H_{30}O_4$
12	H	CH_3	35	92	$C_{22}H_{26}O_2$

[a] Synthetic method D under Experimental Section. [b] All compounds were crystallized from EtOH.

phenylindenes to get new mammary tumor inhibiting antiestrogens.

Chemistry. The acetoxy-substituted 2-phenylindenes **13–15** were synthesized as we previously described in detail.[8] Briefly, the phenylpropan-1-ones **4** and **5** were obtained by Friedel–Crafts acylation of veratrole with propionyl of 2-methylpropionyl chloride, respectively. Compound **6** was prepared by a Grignard reaction of 2-methylpropionyl chloride with (3-methoxyphenyl)magnesium chloride at −70 °C with use of THF as solvent.[14] These phenylpropan-1-ones (**4–6**) (Table I) were dimerized by reductive coupling with $TiCl_4/Zn$ to the corresponding 3,4-diphenyl-3,4-hexandiols **7–9** (Table II) as mixtures of the meso and racemic forms (Scheme II).[8,15]

The methoxy-substituted 2-phenylindenes **10–12** (Table III) were directly obtained from the corresponding diols **7–9** by a treatment with acetyl chloride and acetic anhydride.[8,16]

The acetoxy-substituted 2-phenylindenes were prepared from **10–12** (Table IV, Scheme II) by ether cleavage with BBr_3 and subsequent acetylation with acetic anhydride and pyridine.[7,8] In the case of compound **13**, but not of **12** and **15**, a displacement of the double bond to the 1,2-position took place during ether cleavage and acetylation. This was proved by ^1H NMR spectroscopy. As indenestrol A (Scheme I) and indenestrol B, its analogue with a 1,2-double bond, show nearly identical pharmacological properties,[9] we used compound **13** in this form (Scheme II) in all biological tests.

Table IV. Acetoxy-Substituted 2-Phenylindenes[a]

no.	yield, %	mp,[b] °C	formula[c]
13	71	129	$C_{26}H_{26}O_8$
14	70	124	$C_{28}H_{30}O_8$
15	65	99	$C_{24}H_{26}O_4$

[a] See Scheme II. Synthetic method E under Experimental Section. [b] All compounds were crystallized from EtOH. [c] All compounds were analyzed for C and H within 0.40% of the calculated values.

Table V. Relative Binding Affinities of Compounds **1–3** and **13–15** for Calf Uterine Estrogen Receptor[a]

compd	RBA value[a]	compd	RBA value[a]
1	2.2	13	3.8
2	1.0	14	1.7
3	1.8	15	2.0

[a] RBA = [E2]/[J] × 100; [E2] and [J] are the molar concentrations of nonradioactive E2 and inhibitor required to decrease the bound radioactivity by 50%. E2 = 17β-estradiol.

The structures of compounds **13–15** were proved by ^1H NMR, IR, and mass spectroscopy. A ring closure as shown in Scheme II in the para position of the 5-methoxy group was established by IR spectroscopy by using the γ-C–H bonds of the indene system.[27] The purity was proved by HPLC analysis.

Biological Properties. The 2-phenylindenes **3** and **13–15** as well as the stilbenes **1** and **2** showed an affinity to the estrogen receptor from calf uterine cytosol in vitro (Table V). All compounds exhibited a competitive inhibition of the interaction of [^3H]estradiol with its receptor, as the binding curves were parallel to that of estradiol.

In the series of the 2-phenylindenes, compound **13** with a 5,6,3′,4′-tetraacetoxy substitution (catechol type) had the highest receptor affinity with a RBA value of 3.8. This was rather surprising, as the affinity of **13** was higher than that of the stilbene **2** (RBA = 1.0).[6] This can be due to the shifted double bond in **13** (Scheme II). The 5,3′-diacetoxy-substituted 2-phenylindene **3** had nearly the same receptor affinity as its analogous stilbene **1**.[5,8] In the case of the 5,3′-diacetoxy-substituted compounds **3** and **15**, the transformation of the 1-methyl-3-ethyl to the 1,1-dimethyl-3-isopropyl substitution type did not alter the RBA value. Because of the shifted double bond, **13** cannot be compared exactly with **14** in this regard.

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Table VI. Antiestrogenic Activity of Compounds 2, 3, and 13–15 in the Mouse Uterine Weight Test

compd	dose,[a] μg	effect[b]	inhibn,[c,d] %
solvent		14.2 ± 2.5	
estrone	0.1	44.8 ± 6.8	
2	1.0	33.8 ± 3.7	31[e]
	5.0	27.4 ± 3.6	54[e]
	25.0	40.4 ± 5.2	8
	100.0	41.3 ± 5.0	5
solvent		14.2 ± 3.0	
estrone	0.1	42.5 ± 3.4	
3	1.0	30.1 ± 2.5	48[e]
	5.0	34.8 ± 2.9	30[e]
	25.0	40.2 ± 3.4	9
solvent		10.6 ± 1.3	
estrone	0.1	42.6 ± 3.7	
13	1.0	32.9 ± 4.3	30[e]
	5.0	33.3 ± 2.0	29[e]
	25.0	26.1 ± 2.2	52[e]
	100.0	25.3 ± 3.6	54[e]
solvent		13.8 ± 2.2	
estrone	0.1	34.8 ± 5.0	
14	1.0	29.0 ± 6.2	28[e]
	25.0	32.5 ± 2.8	11
15	1.0	30.3 ± 5.0	21
	25.0	44.7 ± 1.1	0

[a] Dose per animal and day. [b] [Uterus dry weight (mg)/body weight (g)] × 100. [c] Percent inhibition = $[(E_S - E_{S,T})/(E_S - E_V)] \times 100$; E_S = effect of estrone standard, $E_{S,T}$ = effect of standard under simultaneous application of test substance, E_V = effect of vehicle. [d] The U test according to Wilcoxon, Mann, and Whitney was used. [e] Significant ($p < 0.01$).

From the estradiol receptor affinity of a compound, no direct conclusion can be drawn if a drug acts as an estrogen agonist or antagonist. The antiestrogenic activity of the new 2-phenylindenes was determined by the inhibition of the estrone stimulated uterine growth of the immature mouse.

Among the new 2-phenylindenes, compound **13** showed the strongest antiestrogenic effect (Table VI). The estrone-stimulated uterine growth of the immature mouse was inhibited up to 54%. The antiuterotrophic activity could be shown from 1.0 to 100.0 μg/day per animal. Compound **3** was also antiuterotrophic. However, the effect was only apparent at low doses. Thus, the 5,6,3′,4′-tetraacetoxy-substituted 2-phenylindenes are stronger antiestrogens than the corresponding 5,3′-diacetoxy-substituted ones. This is also the case when the effects of **14** vs. **15** are compared.

Changing the alkyl substitution from 1-methyl-3-ethyl to 1,1-dimethyl-3-isopropyl led to a strong decrease of the antiestrogenic activity. This can be clearly seen by comparing the inhibiting effects of **13** vs. **14** and **3** vs. **15**.

Compound **2**, the stilbene with a catechol substitution like **13**, was also antiestrogenic. However, whereas **13** had antiuterotrophic activity over a wide dose range, the inhibiting effect of the stilbene **2** was only apparent at low doses and was nearly completely diminished at a dose of 100 μg of this drug.

These results are in very good agreement with the estrogenic activity of the compounds, which was determined in the immature mouse uterine weight test (Figure 1). Compound **13** exhibited only very low estrogenic effects over the dose range used. Thus, the strong antiestrogenic activity of **13** even at high doses can be explained (Table VI). Compound **3**, however, showed estrogenic properties at high doses in the same way as its antiuterotrophic activity decreased. The 5,6,3′,4′-tetraacetoxy-substituted 2-phenylindenes have lower estrogenic effects than the corresponding 5,3′-diacetoxy-substituted ones. The comparison of **13** vs. **3** and **14** vs. **15** clearly reveals that.

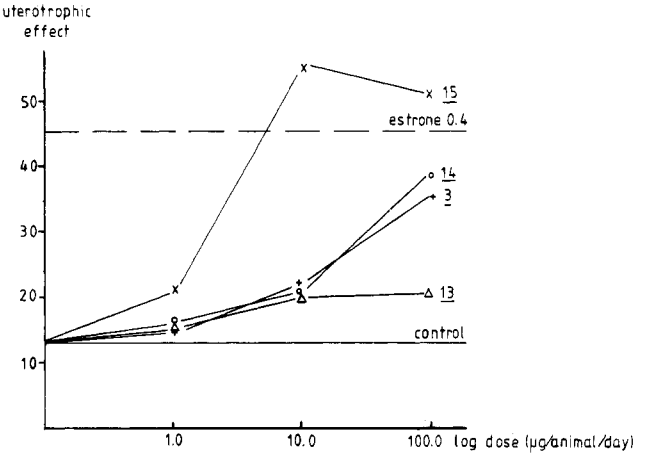

Figure 1. Estrogenic effect of **3** and **13–15** in the immature mouse uterine weight test. Compounds were administered at three consecutive days sc; the uteri were removed 24 h after the last injection. The uterotrophic effect is given as [uterine dry weight (mg)/body weight (g)] × 100.

A change in the alkyl substitution from 1-methyl-3-ethyl to 1,1-dimethyl-3-isopropyl strongly increased the uterotrophic effect as can be seen by comparison of **13** vs. **14** and **3** vs. **15**.

The stilbene **2** had a stronger estrogenic effect than **13** and a lower antiestrogenic activity at doses above 5 μg/mouse.[6]

As compound **13** had the highest affinity for the estrogen receptor and the strongest antiestrogenic and the lowest estrogenic activity of the new 2-phenylindenes, it was selected for evaluation of its mammary tumor inhibiting effects.

The transplantable hormone-dependent MXT mammary carcinoma is a good tumor model for determining the tumor-inhibiting effect of new compounds.[17,18] Its growth is inhibited by ovariectomy and also by the administration of antiestrogens like tamoxifen as well as estrogens like diethylstilbestrol.[18] In our experiments, ovariectomy caused a strong inhibition of tumor growth (% T/C = 2). Diethylstilbestrol was also inhibitory to nearly the same extent. However the estrogenic side effect, determined by the uterus dry weight at the end of therapy, was very high (estrogenic effect % T/C = 200) (unpublished results). Therefore we use this tumor model to determine the tumor-inhibiting effect by the weight of the tumors at the end of therapy as well as the estrogenic side effects by the uterus weight. Compounds that strongly reduce the tumor weight without affecting the uterus weight compared to the untreated control might be of great interest for the treatment of the hormone-dependent mammary carcinoma.

Compound **13** was administered three times a week at doses of 2.0, 4.0, and 16.0 mg/kg for 6 weeks. Whereas at a dose of 2 mg no antitumor effect was observed, **13** exhibited a strong, significant tumor-inhibiting activity at a dose of 4 mg. The tumor weight of the treated animals was only 26% of that of the untreated control. The increase of the dose to 16 mg lowered the antitumor effect (Table VII). At all doses used, **13** had no estrogenic side effect on the adult mouse during 6 weeks of therapy. Its uterotrophic effect was even lower, though not significantly, than that of the control.

Discussion

The results shown above demonstrate that it is possible to develop a mammary tumor inhibiting antiestrogen with only low estrogenic side effects like compound **13** by the

Table VII. Effect of 13 on the Growth of the Hormone-Dependent MXT Mouse Mammary Carcinoma and on Uterus Growth

compd	dose,[a] mg/kg	tumor wt,[b] mg	T/C, %	uterotrophic effect[b,c]	T/C, %
control		221		83.4 ± 12.6	
13	2.0	230	104	71.5 ± 18.1	86
	4.0	57[d]	26	76.2 ± 6.7	91
	16.0	135	61	65.0 ± 12.8	78

[a] Compound 13 was administered sc three times a week for 6 weeks. [b] Determined at the end of therapy. [c] [Uterus dry weight (mg)/body weight (g)] × 100. [d] Significant ($p < 0.05$).

use of certain structural elements. These include the substitution with four acetoxy groups in positions 5,6 and 3′,4′ instead of a 5,3′-diacetoxy substitution and an alkyl substitution with one methyl and an ethyl group in positions 1 and 3 instead of two methyl and an isopropyl group. The 1,1-dimethyl-3-isopropyl substitution type did not resemble the 1,1,2,2-tetramethyl type in the 1,2-diphenylethane series[10] and thus did not lead to strong antiestrogens, as we had hoped.

As stated above, the high receptor affinity of compound 13 was surprising. It is not clear if the displacement of the double bond of 13 compared to 14 or 15 is the reason for the high affinity, as indenestrol A and B, its analogue with a shifted double bond, have identical affinities for the estrogen receptor as well as identical uterotrophic activities.[9]

The antitumor effect of compound 13 on the hormone-dependent MXT mammary carcinoma was strong though only in the 4-mg dose; at the 16-mg dose the effect was lower. A decrease in the antitumor activity at higher doses was also observed with other catechol-type antiestrogens like 3,4-bis(3,4-diacetoxyphenyl)hexane.[19]

It is likely that compound 13 exerts its antitumor effect through its antiestrogenic properties, as the estrogenic effects are very low in the immature mouse test or even not apparent in the MXT assay. However, whereas the antitumor activity of 13 is dose dependent, it shows similar effects on the uterus in the MXT assay at all doses used (Table VII). This implicates that the mode of action of a compound on a hormone-dependent tumor and on an estrogen receptor target organ like the uterus is not unconditionally the same. Several other modes of action of catechol-type estrogens are discussed. The endogenous catechol estrogens, especially 2-OH-substituted estrogens, may play the antiestrogen role in endocrine regulation by an influence on hypothalamic releasing factors or on pituitary hormones and through their affinity for the estrogen receptor in several target organs.[20-23] A further mechanism of action can be cytotoxic effects. Compounds with a catechol structure can be oxidized to semiquinones or quinones, e.g. by the enzymes peroxidase, which is found in high concentration in hormone-dependent mammary tumors,[25] or tyrosinase.[23,24] Such compounds were shown to inhibit the growth of the lymphatic leukemia P-388 in vivo.[26] Catechol estrogens like 2-hydroxyestrone, are also metabolized to semiquinone-like electrophiles, which can bind covalently to the nucleophilic sites of biological macromolecules.[24] The irreversible action of 2-hydroxyestrone on the growth of the hormone-dependent MCF-7 breast cancer cell line can be the result of such covalent bonding to macromolecules including the estrogen receptor itself. If compound 13 acts in one or more of these modes is yet unresolved.

As a conclusion, it can be stated that among the 2-phenylindenes of this study, compound 13 has the most interesting biological properties as it exerted a strong tumor-inhibiting effect with only very low estrogenic properties on the immature as well as the mature mouse and as it had the best antiuterotrophic activity and the highest estrogen receptor affinity in this series.

Experimental Section

General Procedures. Melting points were determined on a Büchi 510 melting point apparatus and are uncorrected. Elemental analyses were performed by the Mikroanalytisches Laboratorium, University of Regensburg. ^1H NMR spectra were obtained with a Varian EM 390 A instrument, 90 MHz (internal standard Me_4Si; chemical shifts in δ (ppm)). IR spectra were recorded on a Beckman Acculab instrument. Mass spectra were obtained with a Varian MATCH 5 instrument. TLC of each compound was accomplished on Merck F 254 silica gel plates. HPLC was performed with an Altex 110 A pump and a Kontron Uvikon 720 LC spectrophotometer; column lichrosorb Si 60, 5 μM, Merck.

Syntheses. Synthetic methods A–F are representatives for compounds reported in Tables I–IV.

Method A. 1-(3,4-Dimethoxyphenyl)propan-1-one (4). Propionyl chloride (19.5 g, 0.21 mol) was added dropwise at 0 °C to a mixture of $AlCl_3$ (33.4 g, 0.25 mol) and veratrole (27.6 g, 0.2 mol) in 200 mL of 1,2-dichloroethane. The whole was stirred for 1 h at 35 °C. After hydrolysis with water, the 1,2-dichloroethane layer was separated, and the aqueous layer was extracted with 1,2-dichloroethane. The organic extracts were washed with water and dried over Na_2SO_4, and the solvent was removed. The crude product was crystallized from EtOH to give 21.9 g of 4.

Method B. 1-(3-Methoxyphenyl)-2-methylpropan-1-one (6). A solution of (3-methoxyphenyl)magnesium bromide (44.4 g, 0.21 mol) in 200 mL of THF was added dropwise at −70 °C to a solution of 2-methylpropionyl chloride (21.3 g, 0.2 mol) in 150 mL of THF. The reaction mixture was brought to room temperature. After 300 mL of water was added, the aqueous layer was extracted with ether. The extract was washed with 1 N NaOH and dried over Na_2SO_4, and the solvent was removed. The crude product was purified by fractional destillation to give 18.6 g of 6.

Method C. 3,4-Bis(3,4-dimethoxyphenyl)-3,4-hexanediol (7). A suspension of Zn (7.85 g, 0.12 mol) in 75 mL of dry THF was added in portions to a solution of 4 (7.8 g, 0.04 mol) and $TiCl_4$ (15.2 g, 0.08 mol) in 150 mL of dry THF at a temperature of −10 °C. The mixture was stirred for 2 h and then hydrolyzed with 10% Na_2CO_3 solution until alkaline reaction occurred. After extraction with ether and crystallization from ethanol, 6.6 g of 7 was obtained as a mixture of the meso and racemic forms.

Method D. 5,6-Dimethoxy-2-(3,4-dimethoxyphenyl)-3-ethyl-1-methyl-1*H*-indene (10). A mixture of 7 (3.9 g, 0.01 mol), acetyl chloride (10.2 g, 0.129 mol), and acetic anhydride (19.7 g, 0.19 mol) was heated under reflux for 5 h, cooled, poured into ice–water, and extracted with ether. After purification by chromatography [SiO_2; CH_2Cl_2–$CH_3COOC_2H_5$ (9:1) as the eluent], 2.19 g of 10 was obtained.

Method E. 5,6-Diacetoxy-2-(3,4-diacetoxyphenyl)-3-ethyl-1-methyl-3*H*-indene (13). A solution of 10 (0.77 g, 2 mmol) in 250 mL of dry CH_2Cl_2 was cooled to −60 °C under N_2, and BBr_3 (3.0 g, 12 mmol) was added with stirring. After the mixture was warmed to room temperature within 0.5 h and stirred for 3 h, 10 mL of dry methanol was added, and the solvents were evaporated. The crude product was acetylated with acetic anhydride and pyridine, and after recrystallization from ethanol, 0.66 g of 7 was obtained as colorless crystals.

Biological Methods. Estradiol Receptor Binding Assay. The method described in ref 4 was used with some modifications. The relative binding affinity (RBA) of the test compounds was determined by the displacement of [^3H]estradiol. Test compounds were incubated with cytosol from calf uteri and [^3H]estradiol at 4 °C for 16 h. Incubation was stopped by adding dextran-coated charcoal. After centrifugation, the radioactivity of a 100-μL supernatant aliquot was counted. The percentage bound radioligand was plotted vs. the concentration of unlabeled test compounds. Five or six concentrations of the competitors were tested. They were chosen to provide a linear portion on a semilog plot crossing the point of 50% competition. From this plot, the molar concentrations of unlabeled estradiol and of test compounds

reducing radioligand binding by 50% were determined.

Estrogen and Antiestrogen Assays. Estrogenic and antiestrogenic properties were determined by stimulation of the uterine growth or by inhibition of the uterine growth stimulated by estrone, respectively, using immature NMRI mice as described previously.[4,5] Female mice (body weight, 10–12 g; age, 20 days at test beginning, 10 mice per group) were injected sc daily for 3 days with solutions of the test compounds in olive oil (0.1 mL/mouse). The uteri were removed 24 h after the last injection, fixed with Bouin's solution, dried, and weighed.

Hormone-Dependent, Transplantable MXT Mammary Tumor of the BDF1 Mouse.[17,18] The MXT tumor used in these studies was the MXT line 3.2 provided by Dr. Bogden, Laboratory of Experimental Oncology, EG & G Bogden Laboratories, Worcester, MA, in a frozen state. The tumor was transplanted in pieces of about 2 mm^3 subcutaneously in female, 8-weeks-old BDF1-mice (body weight, 20 ± 1.6 g; Charles River Wiga, West Germany). After the tumor had reached a diameter of about 1 cm, it was transplanted to 20 mice to determine the hormone dependence. After transplantation the animals were randomly distributed in two groups of 10. The animals of one group were ovariectomized. The tumor grew well in control animals but only very slowly in the ovariectomized mice. Take rate of control animals was >95%. In an experiment to determine the tumor inhibiting activity of new compounds, transplantation is carried out as above (one tumor piece/animal). After transplantation, the animals are randomly distributed into groups of 10. Starting with the first day after transplantation, the test compounds were injected sc three times a week (Monday, Wednesday, Friday) as olive oil solutions (0.1 mL/mouse). The duration of treatment was 6 weeks. At the end of treatment, the animals were killed by cervical dislocation and weighed. The tumors were removed, washed in 0.9% sodium chloride solution, blotted dry, and weighed, and the average tumor weight was calculated. The uteri were also removed and prepared as described in ref 6 to serve as an indicator of the estrogenic side effects of the compounds.

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Registry No. 4, 1835-04-7; 5, 14046-55-0; 6, 6026-75-1; *meso*-7, 98876-44-9; (±)-7, 98876-45-0; *meso*-8, 98876-46-1; (±)-8, 98900-86-8; *meso*-9, 98876-47-2; (±)-9, 98876-48-3; 10, 98876-49-4; 10 (demethylated), 98876-50-7; 11, 98876-51-8; 11 (demethylated), 98876-52-9; 12, 98876-53-0; 12 (demethylated), 98876-54-1; 13, 98876-55-2; 14, 98876-56-3; 15, 98876-57-4; $(CH_3)_2CHCOCl$, 79-30-1; CH_3CH_2COCl, 79-03-8; *m*-BrC_6H_4OMe, 2398-37-0; veratrole, 91-16-7.

Supplementary Material Available: ^1H NMR data (Tables VIII and IX) of methoxy- and acetoxy-substituted 2-phenylindenes (2 pages). Ordering information is given on any current masthead page.

Synthesis and Antiviral Activity of the Carbocyclic Analogues of 5-Ethyl-2′-deoxyuridine and of 5-Ethynyl-2′-deoxyuridine

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The carbocyclic analogue of the antiviral agent 5-ethyl-2′-deoxyuridine (EDU) was synthesized by two routes. The pivotal step in the first route is the reaction of lithium dimethylcuprate with the carbocyclic analogue of 5-(bromomethyl)-2′-deoxyuridine dibenzoate (**6**). The second route is based on the synthesis of the carbocyclic analogue of 5-ethynyl-2′-deoxyuridine (**12**) by a coupling reaction catalyzed by bis(triphenylphosphine)palladium(II) chloride and copper(I) iodide, a method reported recently (Robins and Barr) for the synthesis of the true nucleoside 5-ethynyl-2′-deoxyuridine (**1b**). The carbocyclic analogue of EDU inhibits the replication of type 1 and type 2 herpes simplex viruses in Vero cells. The carbocyclic analogue of 5-ethynyl-2′-deoxyuridine has modest activity against herpes simplex virus, types 1 and 2.

5-Ethyl-2′-deoxyuridine (**1a**, EDU), synthesized originally from 5-ethyluracil,[1-3] was shown to have antiviral activity against herpes simplex and vaccinia viruses.[2-6] EDU inhibits the replication of both type 1 (HSV-1) and type 2 (HSV-2) herpes simplex viruses in cells in culture.[7-9] EDU, as well as related 5-substituted 2′-deoxyuridines, inhibits the replication of strains of HSV-1 that induce thymidine kinase, but it is ineffective against strains that lack the capacity to induce virus thymidine kinase in infected cells (TK^- strains).[7,8] Its affinities for thymidine kinases induced by HSV-1 and HSV-2 are about the same as the affinities of thymidine for these kinases, but the analogue binds much less firmly to cytoplasmic and mitochondrial thymidine kinases from human cells.[8] Furthermore, it has been reported that EDU is active in vivo against HSV-1 and HSV-2 encephalitis,[9,10] was somewhat more effective in vitro against clinical isolates of HSV-2 than against clinical isolates of HSV-1,[11] inhibits replication in cultured cells of several strains of Varicella-Zoster virus,[12] and is not immunosuppressive[13] or mutagenic.[3,14]

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