1,3-Dialkyl-8-(p-sulfophenyl)xanthines: Potent Water-Soluble Antagonists for A_1- and A_2-Adenosine Receptors

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A series of 8-(substituted phenyl) derivatives of theophylline and other 1,3-dialkylxanthines were evaluated for potency and selectivity as antagonists at A_1- and A_2-adenosine receptors in brain tissue. Theophylline has a similar potency (K_i = 14 μM) at both A_1 and A_2 receptors. 8-Phenyltheophylline is 25–35-fold more potent as an adenosine receptor antagonist than theophylline, while 8-phenylcaffeine is only 2–3-fold more potent than caffeine. A *p*-hydroxyaryl substituent enhances the potency of 8-phenyltheophylline as an adenosine antagonist. *p*-Carboxy- and *p*-sulfoaryl substituents reduce potency of 8-phenyltheophylline, yielding water-soluble adenosine antagonists, which are some 2–5-fold more potent than theophylline at adenosine receptors. None of the 8-(substituted phenyl)theophyllines are particularly selective as antagonists toward A_1- and A_2-adenosine receptors. 1,3-Dipropyl-8-phenylxanthine represents a potent and somewhat selective A_1-receptor antagonist about 23-fold more potent at A_1 receptors than at A_2 receptors. A *p*-hydroxyaryl substituent further enhances potency of the 1,3-dipropyl-8-phenylxanthine at both A_1 and A_2 receptors. The 8-(2-amino-4-chlorophenyl)-1,3-dipropylxanthine is a very potent and selective antagonist for A_1 receptors, being nearly 400-fold more potent at A_1 than at A_2 receptors. The water-soluble 8-(*p*-sulfophenyl)- and 8-(*p*-carboxyphenyl)-1,3-propylxanthines no longer exhibit marked selectivity. Both compounds are much more potent as adenosine antagonists than theophylline. The striking selectivity of 1-isoamyl-3-isobutylxanthine as an A_1 antagonist is retained in the 8-phenyl derivative but is virtually lost in the 8-*p*-sulfophenyl derivative.

Adenosine has important roles in the control of the cardiovascular, nervous, and endocrine systems.[1] Two classes of receptors[2,3] mediate the actions of adenosine. One class consists of relatively high affinity A_1-adenosine receptors which have been extensively studied in adipocytes where adenosine agonists cause an inhibition of adenylate cyclase.[3] The other class consists of lower affinity A_2-adenosine receptors which have been studied extensively in various tissues, cultured cells, and platelets where adenosine agonists cause a stimulation of adenylate cyclase.[4,5] Adenosine agonists with some selectivity for A_1 receptors have been developed,[1] but no selective adenosine antagonists are as yet available. Theophylline and caffeine, prototypes for adenosine-receptor antagonists of the xanthine class, are virtually nonselective for A_1 and A_2 receptors.[1,6,7] It is, therefore, unclear to what extent the antiasthmatic,[8] diuretic,[9] respiratory stimulant,[10] central stimulant,[11] cardiac stimulant,[12] and analgesic adjuvant[13] activities of xanthines reflect interactions at A_1 or A_2 receptors or some other site. An 8-phenyl group markedly enhances the potency of theophylline at adenosine receptors[14–18] but does not confer selectivity.[7] The present paper describes the effect of polar aryl substituents on both potency and selectivity of 8-phenylxanthines as adenosine antagonists. The 8-(*p*-sulfophenyl)xanthines because of their solubility, potencies as A_1- and A_2-adenosine antagonists, and probable lack of effect on phosphodiesterases may prove useful in further delineation of physiological roles of A_1- and A_2-adenosine receptors.

Results and Discussion

The potencies of xanthines as antagonists of adenosine receptors were determined with two systems.

(1) Interaction of A_1-adenosine receptors was investigated by using a binding assay with N^6-[^{3}H]cyclohexyladenosine and rat cerebral cortical membranes. Binding of this ligand has been shown to occur only at A_1 receptors in brain membranes[16] with an affinity constant for rat of 1 nM or slightly higher.[6,17,19–21] A variety of xanthines have been investigated as antagonists of binding of N^6-[^{3}H]-cyclohexyladenosine.[6,7,11,16,18] The binding data for xanthines has been consonant with literature on xanthines as antagonists of A_1-adenosine receptors (see ref 1 and references therein).

(2) Interaction of xanthines at A_2-adenosine receptors was investigated by using an assay based on antagonism of 2-chloroadenosine-elicited accumulation of cyclic AMP in guinea pig cerebral cortical slices. 2-Chloroadenosine stimulates cyclic AMP generation in cerebral cortical slices through interaction with a relatively low affinity A_2-adenosine receptor.[4,6] Half-maximal stimulations of cyclic AMP generating systems in brain occur at at a concentration of 2-chloroadenosine of about 10–20 μM. Under the present assay conditions, the EC_{50} for 2-chloroadenosine was 8 μM (data not shown). Adenosine deaminase was present to eliminate any contributions from

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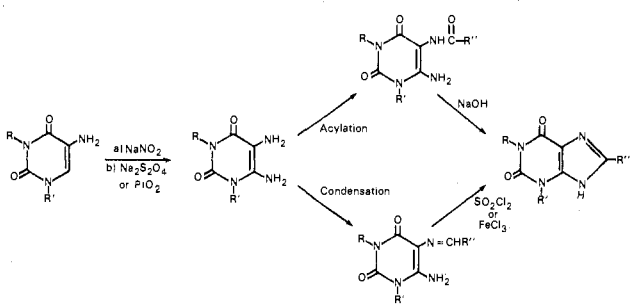
(1) Daly, J. W. *J. Med. Chem.* **1982**, *25*, 197.
(2) Van Calker, D.; Muller, M.; Hamprecht, B. *J. Neurochem.* **1979**, *33*, 99.
(3) Londos, C.; Cooper, D. M. F.; Wolff, J. *Proc. Natl. Acad. Sci. U.S.A.* **1980**, *77*, 2551.
(4) Daly, J. W.; Bruns, R. F.; Snyder, S. H. *Life Sci.* **1981**, *28*, 2083.
(5) Bruns, R. F. *Can. J. Physiol. Pharmacol.* **1980**, *58*, 673.
(6) Daly, J. W.; Butts-Lamb, P.; Padgett, W. *Cell. Mol. Neurobiol.* **1983**, *3*, 6.
(7) Fredholm, B. B.; Persson, C. G. A. *Eur. J. Pharmacol.* **1982**, *8*, 673.
(8) Fredholm, B. B.; Brodin, K.; Strandberg, K. *Acta Pharmacol. Toxicol.* **1979**, *45*, 336.
(9) Osswald, H. *Naunyn-Schmiedeberg's Arch. Pharmacol.* **1979**, *288*, 79.
(10) Stroud, M. W.; Lambertsen, C. J.; Ewing, J. H.; Kough, R. H.; Gould, R. A.; Schmidt, C. F. *J. Pharmacol. Exp. Ther.* **1955**, *114*, 461.
(11) Snyder, S. H.; Katims, J. J.; Annau, Z.; Bruns, R. F.; Daly, J. W. *Proc. Natl. Acad. Sci. U.S.A.* **1981**, *78*, 3260.
(12) Ogilvie, R. I.; Fernandez, P. G.; Winsberg, F. *Eur. J. Clin. Pharmacol.* **1977**, *17*, 409.
(13) Laska, E. M.; Sunshine, A.; Zighelboim, I.; Roure, C.; Marrero, I.; Wanderling, J.; Olson, N. *Clin. Pharmacol. Ther.* **1983**, *33*, 498.
(14) Bruns, R. F. *Biochem. Pharmacol.* **1981**, *30*, 325.
(15) Smellie, F. W.; Davis, C. W.; Daly, J. W.; Wells, J. N. *Life Sci.* **1979**, *24*, 2475.
(16) Bruns, R. F.; Daly, J. W.; Snyder, S. H. *Proc. Natl. Acad. Sci. U.S.A.* **1980**, *77*, 5547.
(17) Wu, P. H.; Phillis, J. W.; Nye, M. J. *Life Sci.* **1982**, *31*, 3857.
(18) Bruns, R. F.; Daly, J. W.; Snyder, S. H. *Proc. Natl. Acad. Sci. U.S.A.* **1983**, *80*, 2077.
(19) Murphy, K. M.; Snyder, S. H. *Mol. Pharmacol.* **1982**, *22*, 250.
(20) Patel, J.; Marangos, P. J.; Stivers, J.; Goodwin, F. K. *Brain Res.* **1982**, *237*, 203.
(21) Murray, T. F.; Cheney, D. L. *Neuropharmacol.* **1982**, *21*, 575.

Table I. Effect of 8-Substituents on Potency of Alkylxanthines at A_1- and A_2-Adenosine Receptors of the Central Nervous System

xanthine	8-substituent	K_i,[a] μM		ratio A_2/A_1
		A_1 receptor	A_2 receptor	
caffeine	H	55 ± 11	50 ± 5	0.9
	phenyl	17 ± 5	27 ± 7	1.6
theophylline	H	14 ± 3	14 ± 2	1.0
	methyl	6.5 ± 1.4	14 ± 0.3	2.2
	phenyl	0.40 ± 0.15	0.55 ± 0.04	1.4
	p-hydroxyphenyl	0.05 ± 0.02	0.13 ± 0.03	2.6
	m-hydroxyphenyl	0.34 ± 0.07	1.0 ± 0.2	3.0
	o-hydroxyphenyl	0.75 ± 0.2	1.7 ± 0.7	2.3
	p-aminophenyl	0.23 ± 0.1	0.13 ± 0.01	0.6
	m-aminophenyl	1.1 ± 0.15	0.9 ± 0.13	0.8
	o-aminophenyl	0.85 ± 0.3	1.7 ± 0.1	2.0
	p-carboxyphenyl	3.0 ± 1.2	2.5 ± 0.2	0.8
	m-carboxyphenyl	32 ± 11	15 ± 8	0.5
	o-carboxyphenyl	>50	>30	
	p-sulfophenyl	4.5 ± 0.5	6.3 ± 1.3	1.6
1,3-dipropylxanthine	H	0.7 ± 0.3	2.7 ± 0.8	3.8
	phenyl	0.013 ± 0.003	0.3 ± 0.08	23
	2-amino-4-chlorophenyl	0.0025 ± 0.001	1.0 ± 0.03	400
	p-hydroxyphenyl	0.0029 ± 0.0008	0.05 ± 0.01	17
	p-carboxyphenyl	0.17 ± 0.03	0.11 ± 0.02	0.6
	p-sulfophenyl	0.21 ± 0.05	0.71 ± 0.23	3.4
1-isoamyl-3-isobutylxanthine	H	13 ± 5	>150	>11
	phenyl	1.8 ± 0.9	>10	>6
	p-sulfophenyl	3.5 ± 0.5	7 ± 2	2.0

[a] IC_{50} values for A_1 receptors were obtained from antagonism of binding of 1 nM [^{3}H]cyclohexyladenosine to rat cerebral cortical membranes. IC_{50} values for A_2 receptors were obtained from antagonism of cyclic [^{3}H]AMP accumulation elicited by 15 μM 2-chloroadenosine in [^{3}H]adenine-labeled guinea pig cerebral cortical slices. K_i values were calculated from the equation $K_i = IC_{50}/(1 +$ concentration adenosine analogue$/K_a$ for adenosine analogue). Values are means ± SEM for two to four determinations.

endogenous adenosine and a potent phosphodiesterase inhibitor (rolipram)[22] was present to minimize any effects due to inhibition of this enzyme by the xanthines being tested. The blockade by xanthines of the effects of 2-chloroadenosine at the A_2-adenosine receptor of brain slices then provided a direct measure of antagonist activity.



The presence of an 8-phenyl substituent increased the potency of caffeine by only about 2–3-fold at A_1 and A_2 receptors (Table I). An 8-phenyl substituent had a much greater effect on the potency of theophylline as previously reported:[7,15,16] A 25–35-fold increase in potency occurred. An 8-methyl substituent had no effect on the potency of theophylline at A_2 receptors while increasing potency at A_1 receptors by less than 2-fold. Neither caffeine, theophylline, nor their 8-phenyl derivatives were selective for A_1 or A_2 receptors. 8-Phenyltheophylline is very insoluble in water, a maximal aqueous concentration being only 10 μM (unpublished results). Investigation of the effect of aryl substituents on selectivity was therefore directed toward polar substituents (OH, NH_2, $N(CH_3)_3^+$, COO^-, SO_3^-), which should also increase solubility. Synthesis of the various compounds followed standard routes as shown in the general scheme. A *p*-hydroxy substituent increased potency of 8-phenyltheophylline at both A_1 and A_2 receptors. The resulting 8-(*p*-hydroxyphenyl)theophylline was still relatively unselective and insoluble. The solubility was 5 μM in water. A *m*- or *o*-hydroxy substituent had little effect on potency at A_1 receptors while causing a significant reduction in potency at A_2 receptors. Amino substituents also did not yield any truly selective antagonists. A *p*-carboxy substituent reduced potency at both receptors by 5–7-fold. A *m*-carboxy or *o*-carboxy substituent was poorly tolerated by both classes of adenosine receptors. A *p*-sulfo substituent reduced potency at both receptors by about 10-fold. Since the *p*-carboxy and *p*-sulfo derivatives of 8-phenyltheophylline had reasonable potencies and solubilities, further investigation of such compounds appeared warranted. The solubility of 8-(*p*-carboxyphenyl)theophylline in water was 90 μM, while the solubility of 8-(*p*-sulfophenyl)theophylline was greater than 20 mM.

Replacement of the 1- and 3-methyl substituents of theophylline with *n*-propyl groups greatly enhances potency at both A_1- and A_2-adenosine receptors.[14,15,18] This was confirmed with the present assay systems (Table I). The 1,3-dipropylxanthine was found to be a somewhat selective A_1 antagonist with a potency at this receptor about 4-fold greater than at the A_2 receptor. Incorporation of an 8-phenyl substituent greatly enhanced potency at the A_1 receptor while enhancing potency at the A_2 receptor to a lesser extent. The resulting 1,3-dipropyl-8-phenylxanthine is a rather selective A_1 antagonist with a potency at A_1 receptors some 23-fold greater than at A_2 receptors. The presence of a *p*-hydroxyaryl substituent in the 1,3-dipropylxanthine series enhanced potency at both A_1 and A_2 receptors as was the case in the theophylline series. The 1,3-dipropyl-8-(*p*-hydroxyphenyl)xanthine had a K_i of about 3 nM at A_1 receptors and a K_i of about 50 nM at A_2 receptors and thus represents a remarkably potent and somewhat selective antagonist for A_1-receptor (17-fold) antagonist. Another very potent A_1 antagonist, 8-(2-amino-4-chlorophenyl)-1,3-dipropylxanthine,[18] proved to be a very selective A_1 antagonist with a potency at A_1 receptors some 400-fold greater than at A_2 receptors (Table I). The results with 1,3-dipropyl-8-(*p*-carboxyphenyl)-

(22) Schwabe, U.; Miyake, M.; Ohga, Y.; Daly, J. W. *Mol. Pharmacol.* **1976**, *25*, 900.

xanthine and 1,3-dipropyl-8-(p-sulfophenyl)xanthine were quite unexpected. Introduction of the polar p-carboxy or p-sulfo substituent decreased potency at the A_1 receptor by 60–70-fold while reducing potency at the A_2 receptor by only about 2-fold and 14-fold, respectively. Both compounds are, thus, only slightly selective: the p-carboxy analogue for A_2 receptors, the p-sulfo analogue for A_1 receptors.

The potent phosphodiesterase inhibitor 1-isoamyl-3-isobutylxanthine has virtually no antagonist activity at A_2-adenosine receptors.[15,23] The present study confirmed the lack of activity of 1-isoamyl-3-isobutylxanthine at A_2-adenosine receptors but revealed relatively potent activity, comparable to that of theophylline at A_1-adenosine receptors (Table I). Thus, this xanthine, in addition to being a potent phosphodiesterase inhibitor,[15,24,25] is also a relatively potent and selective antagonist for A_1-adenosine receptors. It would appear likely that the A_1 receptor has greater bulk tolerance at the 1-position of 1,3-dialkylxanthines than does the A_2 receptor. It was hoped that water-soluble 8-phenyl derivatives of 1-isoamyl-3-isobutylxanthine could be obtained that would retain selectivity and potency as antagonists for A_1 receptors. 1-Isoamyl-3-isobutyl-8-phenylxanthine was relatively selective for A_1 receptors but its low water solubility (<10 μM) would appear to limit its usefulness. Introduction of a p-sulfo group yielded 1-isoamyl-3-isobutyl-8-(p-sulfophenyl)xanthine, a compound that is nearly nonselective for A_1 and A_2 receptors.

The results indicate that the interactions of the 1, 3-, and 7-substituents of a xanthine with adenosine receptors are strongly influenced by the presence and nature of 8-aryl substituents and vice versa. This interdependence between different domains of the xanthine recognition site on adenosine receptors should provide further insights into its nature but has at present only frustrated efforts to design selective water-soluble antagonists for A_1- and A_2-adenosine receptors. As yet no clear indications of the structural or physical features which will ultimately lead to selective antagonists have been obtained.

It will be of interest to determine whether or not the modest selectivity of xanthines, such as 1,3-dipropyl-8-phenylxanthine, will allow for blockade of A_1 adenosine receptor mediated events such as cardiac depression and inhibition of lipolysis in fat cells while still allowing A_2 adenosine receptor mediated events such as smooth muscle relaxation (vasodilation, spasmolytic action) and inhibition of platelet aggregation. Initial studies have indicated that parenteral 8-(p-sulfophenyl)theophylline does not penetrate well in brain since it does not reverse the behavioral depressant effects of N^6-cyclohexyladenosine.[11] Such 8-p-sulfophenyl derivatives, therefore, may prove useful as peripheral adenosine antagonists void of the central stimulant properties of theophylline and caffeine. 8-(p-Sulfophenyl)theophylline effectively blocks adenosine-elicited potentiation of glucagon release from pancreas[26] and when applied iontophoretically blocks adenosine-elicited inhibition of firing of central neurons.[27] Further studies on inhibition of phosphodiesterase by 8-phenylxanthines are necessary. However, 8-phenyltheophylline had little activity as a phosphodiesterase inhibitor[15] and 8-phenyltheophylline and various 8-aryl-substituted analogues were reported to be weak inhibitors of a membrane phosphodiesterase.[17] In addition, the 8-(p-sulfophenyl)-xanthines should penetrate poorly into cells, thus making it even less likely that inhibition of phosphodiesterase by such polar analogues will contribute significantly to their pharmacological actions.

Experimental Section

Melting points were taken on a Kofler block hot stage and are corrected. Mass spectra were determined with a Finnegan 1015 quadrapole (chemical ionization with CH_4 or NH_3) and with VG 70/70 and Hitachi Perkin-Elmer RMU-6E (70 eV, electron impact) mass spectrometers. The spectra were consistent with structures of the compounds. In the case of the 8-(p-sulfophenyl)xanthines, mass spectral analyses were carried out by Dr. A. Yergey (NIH) using a modified Finnegan mass spectrometer with a thermospray inlet. Thin-layer chromatographic analysis on silica gel with $HCCl_3$/MeOH mixtures indicated the presence of a single compound in the final xanthine products. R_f values for $CHCl_3$/MeOH (10:1) were as follows: 8-phenylcaffeine, 0.7; 8-(p-hydroxyphenyl)theophylline, 0.6; 8-(p-nitrophenyl)theophylline, 0.67; 8-(p-aminophenyl)theophylline, 0.49; 1,3-dipropyl-8-phenylxanthine, 0.73; 1,3-dipropyl-8-(p-hydroxyphenyl)xanthine, 0.64; 1-isoamyl-3-isobutyl-8-phenylxanthine, 0.74. R_f values for $CHCl_3$/MeOH (2:1) were as follows: 8-(p-carboxyphenyl)theophylline, 0.35; 8-(p-sulfophenyl)theophylline, 0.32; 1,3-dipropyl-8-(p-carboxyphenyl)xanthine, 0.41; 1,3-dipropyl-8-(p-sulfophenyl)xanthine, 0.38; 1-isoamyl-3-isobutyl-8-(p-sulfophenyl)xanthine, 0.43. Analytical results are given or were within ±0.4% of theoretical values. Satisfactory analyses for the 8-(p-sulfophenyl)xanthines were difficult to obtain because of trace amounts of sodium salts in otherwise pure material. 8-(p-Sulfophenyl)theophylline is now available from Research Biochemicals Inc. (Wayland, MA) and a sample from that source exhibited identical adenosine antagonist activity to our own material. 8-Phenyltheophylline was from Calbiochem-Behring (San Diego, CA). 8-Methyltheophylline was synthesized as described.[28] 1,3-Dipropylxanthine was provided by G. D. Searle (Chicago, IL). The following compounds have been reported in the literature[18] and were provided by Dr. R. F. Bruns (Warner-Lambert Co., Ann Arbor, MI): 8-(m-hydroxyphenyl)theophylline, 8-(o-hydroxyphenyl)theophylline, 8-(m-aminophenyl)theophylline, 8-(m-carboxyphenyl)theophylline, 8-(o-carboxyphenyl)theophylline, and 1,3-dipropyl-8-(2-amino-4-chlorophenyl)xanthine. A detailed procedure for the synthesis of 1-isoamyl-3-isobutylxanthine has not been published, and since difficulties were encountered using the described route,[24] our final procedure to this xanthine is reported.

8-Phenylcaffeine. To a stirred mixture of 8-phenyltheophylline (123 mg, 0.48 mmol) and dry K_2CO_3 (66 mg, 0.48 mmol) in 2 mL of dry dimethylformamide was added dropwise iodomethane (680 mg, 4.8 mmol). The mixture was refluxed for 2 h and then poured into 20 mL of H_2O. The product slowly precipitated and was then filtered and recrystallized from DMF/H_2O to afford 81 mg (62%) of 8-phenylcaffeine: mp 186–188 °C. Anal. ($C_{14}H_{14}N_4O_2$) C, H, N.

8-(p-Hydroxyphenyl)theophylline. The synthesis followed the general procedure of Bariana.[29] 5,6-Diamino-1,3-dimethyluracil[30] (1.7 g, 10 mmol) was dissolved in 100 mL of MeOH/HOAc (5:1) and p-hydroxybenzaldehyde (1.24 g, 10 mmol) in 100 mL of MeOH was added. The mixture was stirred at room temperature for 15 h. The precipitate was filtered, washed with MeOH, and recrystallized from DMF/MeOH, affording 1.27 g (46%) of 6-amino-1,3-dimethyl-5-[(p-hydroxybenzylidene)-amino]uracil. The uracil (1.27 g, 4.6 mmol) was suspended in 125 mL of EtOH and $FeCl_3$ (0.75 g, 4.6 mmol) in 25 mL of EtOH was added with stirring. The mixture was refluxed for 6 h and then stirred at room temperature for 4 days. The precipitate was

(23) Smellie, F. W.; Daly, J. W.; Wells, J. N. *Life Sci.* **1979**, *25*, 1917.
(24) Kramer, G. L.; Garst, J. E.; Mitchel, S. S.; Wells, J. N. *Biochemistry* **1977**, *16*, 3316.
(25) Kramer, G. L.; Wells, J. N. *Mol. Pharmacol.* **1979**, *16*, 813.
(26) Petrack, B.; Czernik, A. J.; Ansell, J.; Cassidy, J. *Life Sci.* **1981**, *28*, 2611.
(27) Phillis, J. W.; Wu, P. H. "Regulatory Function of Adenosine"; Berne, R. M., Rall, T. W., Rubio, R., Eds.; Martinus Nijhoff: The Hague, 1983; p 419.
(28) Speer, J. H.; Raymond, A. L. *J. Am. Chem. Soc.* **1953**, *75*, 114.
(29) Bariana, D. S. *Can. J. Chem.* **1968**, *46*, 3413.
(30) Blicke, F. F.; Godt, H. C. *J. Am. Chem. Soc.* **1954**, *76*, 2798.

filtered, washed with MeOH, and recrystallized from DMF/MeOH, affording 0.77 g (62%) of 8-(*p*-hydroxyphenyl)theophylline: mp >300 °C. Anal. ($C_{13}H_{12}N_4O_3$) C, H, N.

8-(*p*-Aminophenyl)theophylline. 5,6-Diamino-1,3-dimethyluracil (1.7 g, 10 mmol) was dissolved in 50 mL of MeOH/HOAc (5:1) and *p*-nitrobenzaldehyde (1.5 g, 10 mmol) in 35 mL of MeOH was added with stirring. After the mixture was stirred at room temperature overnight, the precipitate was filtered, washed with MeOH, and recrystallized from DMF/MeOH to yield 1.58 g (52%) of 6-amino-1,3-dimethyl-5-[(*p*-nitrobenzylidene)amino]uracil. The following cyclization procedure follows the procedure of Senga et al.[31] The $FeCl_3$ procedure (vide supra) afforded only starting material. The uracil (0.5 g, 1 mmol) was refluxed for 5 min in $SOCl_2$, cooled to room temperature, and stirred overnight. After evaporation in vacuo of solvent, the residue was triturated with 6 N NH_4OH and filtered. The product (0.26 g) was recrystallized from DMF/MeOH to yield 0.11 g (36%) of 8-(*p*-nitrophenyl)theophylline: mp >300 °C. Anal. ($C_{13}H_{11}N_5O_4$) C, H, N.

The (nitrophenyl)theophylline (0.1 g) was hydrogenated at 40 psi of H_2 for 3 h in 10 mL of EtOH with 10 mg of 10% palladium on charcoal catalyst. After filtration, the solvent was evaporated in vacuo. Recrystallization from DMF/MeOH afforded 10 mg (10%) of 8-(*p*-aminophenyl)theophylline: mp >300 °C. Anal. ($C_{13}H_{13}N_5O_2 \cdot H_2O$) C, H, N.

8-(*p*-Carboxyphenyl)theophylline. 5,6-Diamino-1,3-dimethyluracil (1.7 g, 10 mmol) was dissolved in 100 mL of MeOH/HOAc (5:1), and *p*-carboxybenzaldehyde (1.5 g, 10 mmol) in 100 mL of MeOH was added with stirring. The mixture was stirred overnight and the precipitate was filtered, washed with MeOH, and recrystallized from DMF/MeOH to yield 1.5 g (50%) of 6-amino-1,3-dimethyl-5-[(*p*-carboxybenzylidene)amino]uracil. The uracil (1.5 g, 5 mmol) was suspended in 100 mL of EtOH, and $FeCl_3$ (0.81 g, 5 mmol) in 25 mL of EtOH was added with stirring. The mixture was refluxed for 6 h and then stirred at room temperature for 4 days. The precipitate was filtered, washed with MeOH, and recrystallized from DMF/MeOH, affording 690 mg (46%) of 8-(*p*-carboxyphenyl)theophylline: mp >300 °C. Anal. ($C_{14}H_{12}N_4O_4$) C, H, N.

8-(*p*-Sulfophenyl)theophylline. *p*-Sulfobenzoic acid potassium salt (2.4 g, 10 mmol) was dissolved in 50 mL of H_2O, and first 5,6-diamino-1,3-dimethyluracil (1.7 g, 10 mmol) and then 1-ethyl-3-[3-(dimethylamino)propyl]carbodiimide hydrochloride (1.91 g, 10 mmol) were added with stirring. The solution was stirred at room temperature for 45 min, and 1,3-dimethyl-6-amino-5-(*p*-sulfobenzamido)uracil was precipitated from solution with MeOH, removed by filtration, and washed with MeOH. The (*p*-sulfobenzamido)uracil was dissolved in 50 mL of 2.5 N NaOH and heated at 70 °C for 5 min. After cooling to 0 °C, the solution was adjusted to pH 6 with concentrated HCl. After filtration, washing with cold 1 N HCl and recrystallization by base/acid treatment afforded 1.68 (50%) of 8-(*p*-sulfophenyl)theophylline: mp >300 °C. Anal. ($C_{13}H_{12}N_4O_5S \cdot 1^1/_2H_2O$) C, H, N.

1,3-Dipropyl-8-phenylxanthine. The following modification of the standard nitrosation-reduction procedure for preparation of 1,3-dialkyl-5,6-diaminouracils was used. A solution of $NaNO_2$ (3.09 g, 45 mmol) in 10 mL of H_2O was added dropwise to a solution of 1,3-dipropyl-6-aminouracil[32] (6.33 g, 30 mmol) in 225 mL of 42% HOAc. The pH was kept at 3 by addition of 6 N HCl. A reddish purple precipitate formed. The mixture was stirred vigorously for another 15 min. The precipitate was collected by filtration and air-dried overnight to give 5.7 g (75%) of 1,3-dipropyl-6-amino-5-nitrosouracil. A small portion (300 mg) of the nitrosouracil was suspended in 30 mL of H_2O and the suspension was made basic (pH 10–11) with 50% NH_4OH. An aqueous solution of $Na_2S_2O_4$ was added until the purple color faded. An additional portion of 1,3-dipropyl-6-amino-5-nitrosouracil was added, the suspension was made alkaline and the $Na_2S_2O_4$ solution was again added until the color faded. This process was repeated until all the nitroso compound (3.1 g, 12.5 mmol) was reduced. The bluish white crystals were filtered, washed with water, and air-dried overnight to give 2.35 g (83%) of 5,6-diamino-1,3-dipropyluracil. Benzoic acid (1.22 g, 10 mmol) was dissolved in 20 mL of MeOH, and first the crude 5,6-diamino-1,3-dipropyluracil (2.26 g, 10 mmol) and then diisopropylcarbodiimide (1.26 g, 10 mmol) were added with stirring. The mixture was stirred at room temperature for 30 min and the precipitate removed by filtration and washed with MeOH. The crude benzamidouracil was heated to 100 °C for 10 min in 50 mL of 2.5 N NaOH and filtered while hot. After cooling, the filtrate was adjusted to pH 6 with concentrated HCl and the precipitate removed by filtration, washed with cold 1 N HCl, and recrystallized by base–acid precipitation to afford 1.15 g (37%) of 1,3-dipropyl-8-phenylxanthine: mp 260–261 °C. Anal. ($C_{17}H_{20}N_4O_2$) H, N; C: calcd, 65.38; found, 64.91.

1,3-Dipropyl-8-(*p*-hydroxyphenyl)xanthine. To a solution of 1,3-dipropyl-5,6-diaminouracil (0.76 g, 3.4 mmol) in 60 mL MeOH/HOAc (5:1) was added dropwise and with stirring 0.43 g of *p*-hydroxybenzaldehyde (3.5 mmol) in 20 mL of MeOH. The solution was stirred for another 10 min and the solution concentrated in vacuo. The yellow precipitate was filtered, washed with MeOH, and air-dried to afford 0.49 g (45%) of 1,3-dipropyl-8-(*p*-hydroxyphenyl)xanthine. It would appear that the oxidative cyclization occurred spontaneously in this case. Presumably, O_2 served as the oxidant. An analytical sample was obtained by chromatography on Sephadex LH-20 with MeOH and recrystallization from DMF/MeOH: mp >300 °C. Anal. ($C_{17}H_{20}N_4O_3 \cdot {}^1/_2H_2O$) C, H, N.

1,3-Dipropyl-8-(*p*-carboxyphenyl)xanthine. To a solution of 1,3-dipropyl-5,6-diaminouracil (1.85 g, 8 mmol) in 100 mL of MeOH/HOAc (5:1) was added dropwise with continuous stirring *p*-carboxybenzaldehyde (1.23 g, 8 mmol) in 20 mL of MeOH. After stirring for an additional 15 min, the suspension was cooled in an ice bath, filtered, and air-dried to give 2.3 g (80% yield) of 6-amino-5-[(*p*-carboxybenzylidene)amino]-1,3-dipropyluracil, which was used without further purification. To a solution of the 1,3-dipropyl-6-amino-5-[(*p*-carboxybenzylidene)amino]uracil (0.55 g, 1.5 mmol) in 25 mL of EtOH was added slowly and with constant stirring a solution of $FeCl_3$ (0.25 g, 1.5 mmol) in 25 mL of EtOH. The reaction mixture developed a deep blue color. It was refluxed for 4 h and allowed to stand overnight. The precipitate was filtered, washed with EtOH, and air-dried to give 0.12 g (23%) of 1,3-dipropyl-8-(*p*-carboxyphenyl)xanthine. An analytical sample was obtained by purification on LH-20 Sephadex with MeOH and recrystallization from DMF: mp >300 °C; NMR δ 0.88 (m, 6, CH_3), 1.57 (m, 2, 1-βCH_2), 1.75 (m, 2, 3-βCH_2), 3.85 (t, 2, 1-αCH_2), 4.02 (t, 2, 3-αCH_2), 8.02 (d, 2, C_6H_4), 8.22 (d, 2, C_6H_4). Anal. ($C_{18}H_{20}N_4O_4 \cdot {}^1/_2H_2O$) C, H; N: calcd, 15.34; found, 16.03.

1,3-Dipropyl-8-(*p*-sulfophenyl)xanthine. 1,3-Dipropyl-5,6-diaminoxanthine (0.78 g, 3.4 mmol) was suspended in 20 mL of H_2O, and first *p*-sulfobenzoic acid potassium salt (0.82 g, 3.4 mmol) and then 1-ethyl-3-[3-(dimethylamino)propyl]carbodiimide hydrochloride (0.65 g, 3.4 mol) were added. The mixture was stirred at room temperature for 1 h, followed by concentration in vacuo to a small volume. Addition of Et_2O caused precipitation of 1,3-dipropyl-6-amino-5-(*p*-sulfobenzamido)uracil, which was filtered and washed with ether. The crude (sulfobenzamido)uracil was dissolved in 20 mL of 2.5 N NaOH and heated at 70 °C for 5 min. After cooling to 0 °C, the product was precipitated by adjusting the pH to 5.0 with concentrated HCl. After filtration and washing with cold 1 N HCl, recrystallization by base/acid treatment yielded 647 mg (48%) of 1,3-dipropyl-8-(*p*-sulfophenyl)xanthine: mp >300 °C. Anal. ($C_{17}H_{20}N_4O_5S$) C, N; H: calcd, 4.89; found, 5.87.

1-Isoamyl-3-isobutylxanthine.[24] 1-Isobutyl-6-aminouracil was prepared by a modification of the method of Papesch and Schroeder[32] as follows: A mixture containing 23.2 g (200 mmol) of monoisobutylurea, 18.8 g (220 mmol) of cyanoacetic acid, and 40 mL of Ac_2O was heated to 75–80 °C for 2 h. After the mixture cooled, 100 mL of Et_2O was added and the reaction mixture was allowed to stand in an ice bath for several hours. The crystalline product was filtered, washed with Et_2O, air-dried, and suspended in 180 mL of H_2O–EtOH (2:1) for ring closure. To the suspension was rapidly added 30 mL of 10% NaOH and the reaction mixture was warmed to 85 °C for 1 h during which time more NaOH was added to keep the reaction mixture strongly alkaline. After several

(31) Senga, K.; Shimizu, K.; Nishiyaki, S. *Chem. Pharm. Bull.* **1977**, *25*, 495.
(32) Papesch, V.; Schroeder, E. F. *J. Org. Chem.* **1951**, *16*, 1879.

hours at room temperature, the reaction mixture was cooled in an ice bath, and the crystals were filtered and air-dried to yield 7.32 g (20%) of 1-isobutyl-6-aminouracil. Two alkylation procedures were used to prepare 3-isoamyl-1-isobutyl-6-aminouracil.

A. A mixture of 4.4 g (24 mmol) of 1-isobutyl-6-aminouracil, 7 mL of 15% NaOH, and 15 mL of 95% EtOH was refluxed for 15 min. To the refluxing solution was then added dropwise over a period of 10 min 7.25 g (48 mmol) of 1-bromo-3-methylbutane. The mixture was refluxed for 4 h and the solvent removed in vacuo. The residue was extracted with a mixture of 100 mL of $CHCl_3$ and 50 mL of H_2O. The $CHCl_3$ layer was washed with additional H_2O and evaporated in vacuo to afford 4.1 g (68%) of 1-isobutyl-3-isoamyl-6-aminouracil. The aqueous layer yielded unreacted starting material which could be recycled through the alkylation procedure. An analytical sample of product was obtained by three recrystallizations from EtOH–H_2O: mp 119–120 °C. Anal. ($C_{13}H_{23}N_3O_2$) C, H, N.

B. An alternate alkylation with isoamyl tosylate gave a lower yield of 1-isobutyl-3-isoamyl-6-aminouracil as follows. To a stirred solution of 3 g (17 mmol) of 1-isobutyl-6-aminouracil in 4.5 mL of EtOH was added 3.5 mL of 20% NaOH. After the mixture was warmed to 50 °C, 8.0 g (33 mmol) of freshly prepared isoamyl tosylate was added dropwise over 12 min. After an additional 30 min at 50 °C, the solution was refluxed for 2 h during which time an oil precipitated. The mixture was concentrated in vacuo to one-half volume and 6 mL of H_2O was added. The mixture was extracted six times with 25-mL portions of $CHCl_3$. The $CHCl_3$ was dried with Na_2SO_4/Drierite and evaporated in vacuo to yield 6.23 g of yellow oil. Column chromatography (100 g of silica gel) with 5–7% MeOH in CH_2Cl_2 afforded 1.66 g (38%) of nearly pure 1-isobutyl-3-isoamyl-6-aminouracil.

To a stirred solution of 4 g (16 mmol) of 1-isobutyl-3-isoamyl-6-aminouracil in 80 mL of 42% AcOH at 60 °C was added 3.3 g (48 mmol) $NaNO_2$ in 18 mL of H_2O over a period of 10 min. After stirring for an additional 10 min, the reaction mixture was cooled in an ice bath. The precipitate was filtered, washed several times with cold H_2O, and air-dried overnight to give 3.5 g (79%) of 1-isobutyl-3-isoamyl-5-nitroso-6-aminouracil. An analytical sample was obtained by two recrystallizations from MeOH: mp 226–228 °C. Anal. ($C_{13}H_{22}N_4O_3$) C, H, N.

Two reduction procedures were used to prepare 1-isobutyl-3-isoamyl-5,6-diaminouracil. The PtO_2 method (B) afforded more consistent results and is the preferred method.

A. A stirred suspension of 320 mg (1.13 mmol) of 1-isobutyl-3-isoamyl-5-nitroso-6-aminouracil in 3.5 mL of 6 N NH_4OH was warmed to 75 °C. $Na_2S_2O_4$ (1.22 g, 7.0 mmol) in 5 mL of H_2O was added dropwise to the suspension over 6–7 min. During the addition, the purple solid disappeared, with concomitant formation of a brown gummy solid on the stirring bar. Heating and stirring were continued for 40 min. After cooling, the stirring bar was removed from the clear solution. The adhering solid was dissolved in 20 mL of $CHCl_3$. The $CHCl_3$ extract was washed with H_2O and then dried over anhydrous $MgSO_4$. Evaporation of the solvent in vacuo gave 280 mg (92%) of 1-isobutyl-3-isoamyl-5,6-diaminouracil.

B. A suspension of 1.6 g (5.6 mmol) of 1-isobutyl-3-isoamyl-5-nitroso-6-aminouracil in 75 mL of MeOH and 0.03 g of PtO_2 was hydrogenated at 40 psi for 30 min. The catalyst was removed by filtration and the solvent removed in vacuo to yield 0.9 g (60%) of a yellow glass which was pure by thin-layer chromatographic analysis. Crystallization from MeOH/Et_2O afforded 1-isobutyl-3-isoamyl-5,6-diaminouracil. This compound is labile and should be stored in cold under N_2.

A solution of 977 mg (3.6 mmol) of *freshly* prepared 1-isobutyl-3-isoamyl-5,6-diaminouracil in 12 mL of 97% HCOOH was refluxed for 1 h. The HCOOH was removed in vacuo to give a light tan solid. Recrystallization from 30 mL of acetone gave 681 mg (64%) of 1-isobutyl-3-isoamyl-5-formamido-6-aminouracil. An analytical sample was obtained by a second crystallization from acetone: mp 186–187 °C (trans crystallization to needles at 182 °C). Anal. ($C_{14}H_{24}N_4O_3$) C, H, N.

A mixture of 550 mg (1.86 mmol) of 1-isobutyl-3-isoamyl-5-formamido-6-aminouracil and 25 mL of 10% NaOH was refluxed for 20 min. The colorless solution was cooled to room temperature and filtered through cotton to remove a small amount of insoluble material. The clear aqueous solution was then cooled and acidified with cold, concentrated HCl to Congo Red. The colorless solid was removed by filtration and washed repeatedly with cold H_2O. The solid was recrystallized from C_6H_6 to give 450 mg (87%) of 1-isoamyl-3-isobutylxanthine: mp 195.5–196 °C (lit.[24] mp 189–191 °C). Anal. ($C_{14}H_{22}N_4O_2$) C, H, N.

1-Isoamyl-3-isobutyl-8-phenylxanthine. To a solution of 0.75 g (2.8 mmol) of 1-isobutyl-3-isoamyl-5,6-diaminouracil in 50 mL of MeOH/AcOH (5:1) was slowly added with constant stirring a solution of 0.30 g (2.8 mmol) of benzaldehyde in 20 mL of MeOH. The reaction mixture was allowed to stand at room temperature for 24 h. The precipitate was filtered and air-dried to give 0.58 g (59%) of 1-isoamyl-3-isobutyl-8-phenylxanthine. In this case, oxidative cyclization occurred spontaneously; presumably O_2 served as the oxidant. Recrystallization from a DMF/MeOH mixture afforded an analytical sample: mp 259–260 °C. Anal. ($C_{20}H_{26}N_4O_2 \cdot H_2O$) C, H, N.

1-Isoamyl-3-isobutyl-8-(*p*-sulfophenyl)xanthine. To a solution of 0.54 g (2 mmol) of 1-isobutyl-3-isoamyl-5,6-diaminouracil in 5 mL of H_2O was added dropwise with constant stirring a solution of 0.51 g (2.1 mmol) of *p*-sulfobenzoic acid potassium salt in 20 mL of H_2O, followed by 0.41 g (2.1 mmol) 1-ethyl-3-[3-(dimethylamino)propyl]carbodiimide hydrochloride. The solution was stirred at room temperature for 1 h, followed by addition of 20 mL of MeOH and 10 mL of H_2O and stirring overnight. The reaction mixture was filtered and the volume of the filtrate was reduced in vacuo. After several hours at room temperature the concentrated reaction was cooled in an ice bath and the precipitate collected by filtration. The crude product was refluxed in 20 mL of 2.5 N NaOH for 25 min. The yellow oil which formed during this period dissolved on neutralization with HCl. The solution was adjusted to pH 5 and cooled to yield a white precipitate, which was filtered and air-dried. Recrystallization in DMF/MeOH yielded 0.8 g (92%) of 1-isobutyl-3-isoamyl-8-(*p*-sulfophenyl)xanthine: mp >300 °C; NMR δ 0.92 (m, 12, CH_3), 1.45 (m, 2, 1-βCH_2), 1.59 (m, 1, 1-γCH), 2.28 (m, 1, 3-βCH), 3.9 (m, 4, 1- and 3-αCH_2), 7.72 (d, 2, C_6H_4), 8.1 (d, 2, C_6H_4). Anal. ($C_{20}H_{26}N_4O_5S \cdot H_2O$) C; H: calcd 6.19; found, 6.71. N: calcd, 12.39; found, 13.37.

Biochemical Assay. Inhibition of binding of 1 nM N^6-[^{3}H]cyclohexyladenosine to A_1 receptors in rat cerebral cortical membranes was assayed as described.[6] Inhibition of binding by a range of concentrations of each xanthine was assessed in triplicate for at least two separate experiments. Inhibition of 2-chloroadenosine-stimulated cyclic AMP accumulation in [^{3}H]-adenine-labeled guinea pig cerebral cortical slices was assayed essentially as described.[6] In the present experiments 10 μg/mL of adenosine deaminase was present in incubations with slices to prevent effects of endogenous adenosine and 30 μM 4-[3-(cyclopentyloxy)-4-methoxyphenyl]-2-pyrrolidone (rolipram, ZK 62711)[22] was present to inhibit phosphodiesterases. Under these conditions 2-chloroadenosine elicited a maximal 10–20-fold increase in levels of radioactive cyclic AMP in guinea pig cerebral cortical slices with an EC_{50} of about 8 μM (data not shown). Inhibition of the response to 15 μM 2-chloroadenosine by a range of concentrations of each xanthine was assessed in triplicate in at least two separate experiments. The results (Table I) are in toto quite comparable to published results[6,7,11,16,18] on xanthines as adenosine antagonists. Differences could reflect the use of different assays, tissues, or species by various investigators. One report on the lack of activity of 8-(*p*-sulfophenyl)theophylline as an A_1-adenosine antagonist[34] is incorrect, being due to an error in citation of earlier literature.

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Registry No. *p*-HOC_6H_4CHO, 123-08-0; *p*-$O_2NC_6H_4CHO$, 555-16-8; *p*-$HO_2CC_6H_4CHO$, 619-66-9; *p*-$HO_2CC_6H_4SO_3K$, 5399-63-3; $C_6H_5CO_2H$, 65-85-0; *i*-$C_4H_9NHCONH_2$, 592-17-6; $NCCH_2$-

(33) Fieser, L. F.; Fieser, M. "Reagents for Organic Synthesis"; Wiley: New York, 1977; Vol. 1, p 1180.
(34) Katims, J. J.; Annau, Z.; Snyder, S. H. *J. Pharmacol. Exp. Ther.* 1983, *227*, 167.

CO_2H, 372-09-8; $(CH_3)_2CH(CH_2)_2Br$, 107-82-4; caffeine, 58-08-2; 8-phenylcaffeine, 6439-88-9; theophylline, 58-55-9; 8-methyltheophylline, 830-65-9; 8-phenyltheophylline, 961-45-5; 8-(*p*-hydroxyphenyl)theophylline, 85872-69-1; 8-(*m*-hydroxyphenyl)-theophylline, 85872-68-0; 8-(o-hydroxyphenyl)theophylline, 85872-57-7; 8-(*p*-aminophenyl)theophylline, 85872-66-8; 8-(*m*-aminophenyl)theophylline, 85872-65-7; 8-(o-aminophenyl)theophylline, 18830-58-5; 8-(*p*-carboxyphenyl)theophylline, 85872-58-8; 8-(*m*-carboxyphenyl)theophylline, 85872-52-2; 8-(o-carboxyphenyl)theophylline, 78164-01-9; 8-(*p*-sulfophenyl)theophylline, 80206-91-3; 1,3-dipropylxanthine, 31542-62-8; 1,3-dipropyl-8-phenylxanthine, 85872-53-3; 1,3-dipropyl-8-(2-amino-4-chlorophenyl)xanthine, 85872-51-1; 1,3-dipropyl-8-(*p*-hydroxyphenyl)-xanthine, 94781-76-7; 1,3-dipropyl-8-(*p*-carboxyphenyl)xanthine, 94781-78-9; 1,3-dipropyl-8-(*p*-sulfophenyl)xanthine, 89073-57-4; 1-isoamyl-3-isobutyl xanthine, 63908-26-9; 1-isoamyl-3-isobutyl-8-phenylxanthine, 94781-84-7; 1-isoamyl-3-isobutyl-8-(*p*-sulfophenyl)xanthine, 94781-85-8; 5,6-diamino-1,3-dimethyluracil, 5440-00-6; 6-amino-1,3-dimethyl-5-[(*p*-hydroxybenzylidene)-amino]uracil, 94781-72-3; 6-amino-1,3-dimethyl-5-[(*p*-nitrobenzylidene)amino]uracil, 76473-17-1; 8-(*p*-nitrophenyl)theophylline, 1094-63-9; 6-amino-1,3-dimethyl-5-[(*p*-carboxybenzylidene)amino]uracil, 94781-73-4; 1,3-dimethyl-6-amino-5-(*p*-sulfobenzamido)uracil, 94781-74-5; 1,3-dipropyl-6-aminouracil, 41862-14-0; 1,3-dipropyl-6-amino-5-nitrosouracil, 81250-33-1; 5,6-diamino-1,3-dipropyluracil, 81250-34-2; 6-amino-5-benzamido-1,3-dipropyluracil, 94781-75-6; 6-amino-5-[(*p*-carboxybenzylidene)amino]-1,3-dipropyluracil, 94781-77-8; 1,3-dipropyl-6-amino-5-(*p*-sulfobenzamido)uracil, 94781-79-0; 1-isobutyl-6-aminouracil, 56075-75-3; 1-isobutyl-3-isoamyl-6-aminouracil, 94781-80-3; isoamyl tosylate, 2431-75-6; 1-isobutyl-3-isoamyl-5-nitroso-6-aminouracil, 94781-81-4; 1-isobutyl-3-isoamyl-5,6-diaminouracil, 94781-82-5; 1-isobutyl-3-isoamyl-5-formamido-6-aminouracil, 94781-83-6; adenosine, 58-61-7.

Structure–Activity Relationship of Estrogens: Receptor Affinity and Estrogen Antagonist Activity of Certain (*E*)- and (*Z*)-1,2,3-Triaryl-2-propen-1-ones[1,2]

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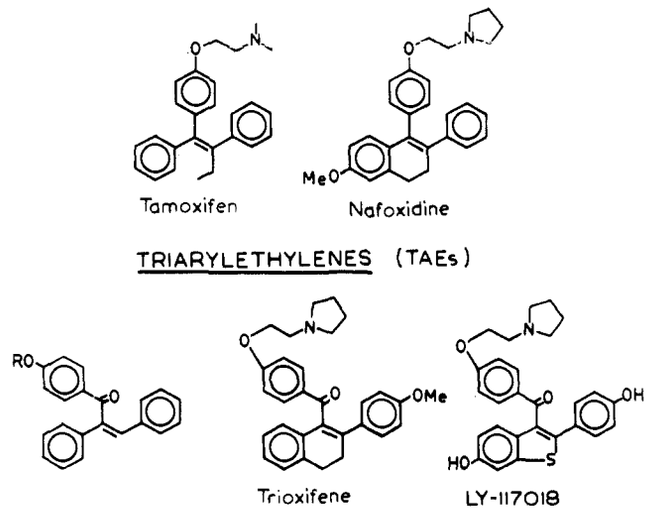
(*E*)- and (*Z*)-1,2,3-triphenyl-2-propen-1-ones and some of their phenolic and alkoxy analogues, substituted at the para position in one or more of the aromatic rings, were synthesized and assigned geometry on the basis of their spectroscopic data. The structure–activity relationship of the triarylpropenones was studied from the point of view of their estrogen receptor affinity and estrogen agonist and antagonist activities. (*E*)- as well as (*Z*)-propenones were found to compete with estradiol for binding with the mouse uterine cytosol receptors, with phenolic analogues usually more potent than the unsubstituted as well as alkoxypropenones. The (*E*)-propenones, which have now emerged as a new group of estrogen receptor ligands, were found to differ from *Z* isomers quite markedly in their binding specificities. The uterotrophic and antiuterotrophic assays in immature mice revealed that while some of the compounds were marginally estrogenic, nearly all the isomeric propenones were antiestrogenic to a varying degree.

The success of antiestrogen therapy in the treatment of certain hormone-responsive breast cancers[3,4] has led to a resurgence of interest in the molecules which can prevent estrogens from exercising their full biochemical effects. These interests relate both to the design of more effective antiestrogens and to elucidation of the mechanism of action of those already known. The majority of the present-day antiestrogens, such as, tamoxifen [(*Z*)-2-[4-(1,2-diphenyl-1-butenyl)phenoxy]-*N*,*N*-dimethylethanamine] and nafoxidine [1-[2-[4-(3,4-dihydro-6-methoxy-2-phenyl-1-naphthalenyl)phenoxy]ethyl]pyrrolidine], belong to a common chemical category, the triarylethylenes (TAEs) (see Chart I). The biological profile of the TAEs is characterized by mixed agonist–antagonist action, and their antagonist activity depends rather crucially on certain favorable circumstances of molecular geometry, substitution pattern, and even on the animal species employed for investigation.[5–11] While some of these features of TAE action are attracting considerable attention, the need for more effective antiestrogens continues to be an important one.

Chart I



Certain (*Z*)-1,2,3-triaryl-2-propen-1-ones (*Z*-TAPs) (see Chart I) have recently emerged as a new group of anti-

(1) CDRI Communication No. 3326.
(2) Dedicated to Dr. Nitya Anand on his 60th birthday.
(3) Horwitz, K. B.; McGuire, W. L. In "Breast Cancer: Advances in Research and Treatment"; McGuire, W. L., Ed.; Plenum Press: New York, 1978; Vol. 2, pp 155–204.
(4) Legha, S. S.; Carter, S. K. *Cancer Treat. Rep.* **1976**, *3*, 205.
(5) Harper, M. J. K.; Walpole, A. L. *J. Reprod. Fertil.* **1967**, *13*, 101.
(6) Lednicer, D.; Lyster, S. C.; Duncan, C. W. *J. Med. Chem.* **1967**, *10*, 78.
(7) Terenius, L. *Acta Endocrinol. (Copenhagen)* **1971**, *66*, 431.
(8) Katzenellenbogen, B. S.; Bhakoo, H. S.; Ferguson, E. R.; Lan, N. C.; Tatee, T.; Tsai, T. L.; Katzenellenbogen, J. A. *Rec. Prog. Horm. Res.* **1979**, *35*, 259.
(9) Jordan, V. C.; Haldeman, B.; Allen, K. E. *Endocrinology* **1981**, *108*, 1353.
(10) Robertson, D. W.; Katzenellenbogen, J. A.; Long, D. J.; Rorke, E. A.; Katzenellenbogen, B. S. *J. Steroid Biochem.* **1982**, *16*, 1.
(11) Robertson, D. W.; Katzenellenbogen, J. A.; Hayes, J. R.; Katzenellenbogen, B. S. *J. Med. Chem.* **1982**, *25*, 167.

0022-2623/85/1828-0492$01.50/0