Renal Vasodilators. The Role of the 4-Substituent in Isoquinolin-3-ol Cardiovascular Agents: 4-Ureido Derivatives of Isoquinolin-3-ol with Selective Renal Vasodilator Properties

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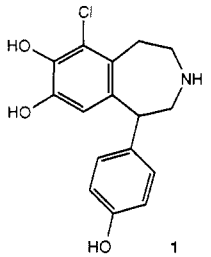
The synthesis and cardiovascular evaluation of a series of isoquinolin-3-ol derivatives bearing a variety of nitrogen substituents (amino, acylamino, carbamate, and ureido) at C-4 are described. Certain of these compounds have a selective renal vasodilating profile and have minimal effects on arterial blood pressure or heart rate when administered intravenously in the instrumented anesthetized dog. The most potent renal vasodilator in the series is 4-(allylureido)-6,7-dimethoxyisoquinolin-3-ol (**38**), which at a dose of 1.2 mg/kg iv produces a 97% maximal increase in renal blood flow without significant hypotensive or chronotropic effects. Structure–activity observations on the nature of the 4-substituent and the alkoxy substitution pattern in the aromatic ring of the isoquinolinol nucleus are discussed.

Current therapies for the treatment of hypertension include the use of peripheral vasodilators, diuretics, sympatholytic agents, calcium channel blockers, and angiotensin converting enzyme (ACE) inhibitors.[1] In clinical practice, these drugs are used singly or in various combinations appropriate for the individual patient. Each of these therapies has shortcomings including lack of broad efficacy for the total patient population and side effect liability. Selective renal vasodilation has been proposed as a new mechanism to gradually reduce high blood pressure in treatment of chronic human essential hypertension and may also have clinical utility in the treatment of a variety of renal disorders characterized by poor renal perfusion such as acute renal failure.[2,3] Among the compounds reported to have preferential renal vasodilating properties are the dopamine agonist fenoldopam (SKF 82526; **1**)[3-5] and the *N*-acetyl-γ-glutamyl derivative **2a**, which appears to act as a prodrug for selective release of the vasodilator **2b** in the kidney.[6,7]



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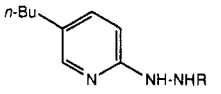
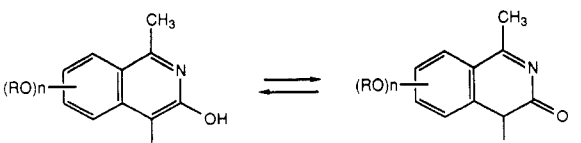


2a R = $COCH_2CH_2CH(NHAc)COOH$
2b R = H

We recently described[8] the synthesis and cardiovascular activity of a series of 4-substituted isoquinolin-3-ol derivatives **3**, which, depending upon conditions, might exist in several tautomeric forms, including lactam **3a**. These

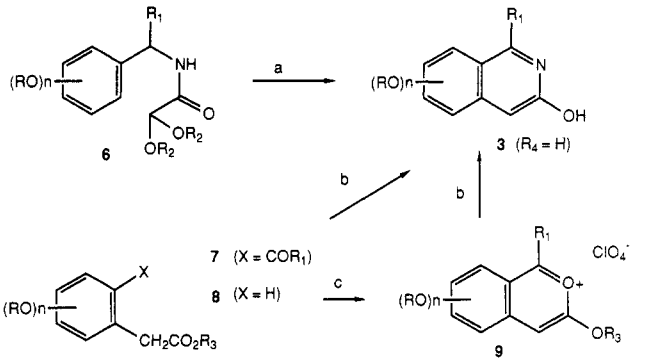


3 **3a**

compounds were of interest as isosteres of our quinazolinone cardiotonic agent bemarinone (ORF 16600; **4**).[9-11] Within our initial series, compounds **3** with hydrogen, alkyl, carboxyalkyl, halogen, and carboxyl-derived substituents at C-4 were identified as potent positive inotropic agents in the dog; vasodilating activity of weak to moderate potency was also observed for certain of these derivatives. The cardiovascular effects of our initial series were of rather short duration, and we noted that although variations in the 4-substituent and in the alkoxy substitution pattern affected potency, the overall cardiovascular profile within that series remained predominately inotropic with secondary vasodilating effects.

In continuing to explore substituent effects in the isoquinolinol series, we have found that further changes in

Scheme I



(a) PPA; (b) NH_4OAc or NH_4OH; (c) Ac_2O, $HClO_4$

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(1) Rudd, P.; Blaschke, T. F. *The Pharmacological Basis for Therapeutics*, 7th ed.; Gilman, A. G., Goodman, L. S., Rall, T. W., Murad, F., Eds.; Macmillan: New York, 1985; Chapter 32.
(2) Struyker-Boudier, H. A. J. *Drug Design*, Vol. 10; Ariens, E. J., Ed.; Academic Press: New York, 1980; pp 146–191.
(3) Ackerman, D. M.; Weinstock, J.; Wiebelhaus, V. D.; Berkowitz, B. *Drug. Devel. Res.* **1982**, *2*, 283.
(4) Ackerman, D. M.; Blumberg, A. L.; McCafferty, J. P.; Sherman, S. S.; Weinstock, J.; Kaiser, C.; Berkowitz, B. *Fed. Proc., Fed. Am. Soc. Exp. Biol.* **1983**, *42*, 186.
(5) Weinstock, J.; Hieble, J. P.; Wilson, J. W., III *Drugs Future* **1985**, *10*, 645.
(6) Hofbauer, K. G.; Sonnenburg, C.; Stalder, R.; Criscione, L.; Kraetz, J.; Fuhrer, W.; Habicht, E. *J. Pharmacol. Exp. Ther.* **1985**, *232*, 838.
(7) Smits, J. F. M.; Struyker-Boudier, H. A. J. *J. Pharmacol. Exp. Ther.* **1985**, *232*, 845.
(8) Kanojia, R. M.; Press, J. B.; Lever, O. W., Jr.; Williams, L.; Tobia, A. J.; Falotico, R.; Moore, J. B., Jr. *J. Med. Chem.* **1988**, *31*, 1363.
(9) Bandurco, V. T.; Schwender, C. F.; Bell, S. C.; Combs, D. W.; Kanojia, R. M.; Levine, S. D.; Mulvey, D. M.; Appollina, M. A.; Reed, M. S.; Malloy, E. A.; Falotico, R.; Moore, J. B.; Tobia, A. J. *J. Med. Chem.* **1987**, *30*, 1421.
(10) Falotico, R.; Moore, J. B., Jr.; Tolman, E. L.; Bandurco, V. T.; Bell, S. C.; Tobia, A. J. *Pharmacologist* **1985**, *27*, abstr. 679.
(11) Falotico, R.; Moore, J. B.; Bandurco, V. T.; Bell, S. C.; Levine, S. D.; Tobia, A. J. *Drug Devel. Res.*, in press.

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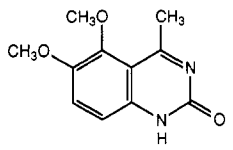

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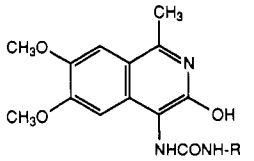

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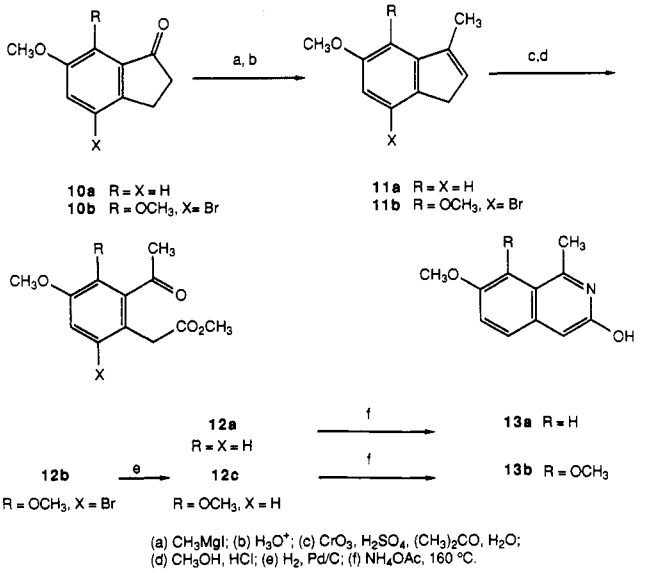
the nature of the 4-substituent of **3** can dramatically effect the overall cardiovascular profile. In the present report, we describe the synthesis and cardiovascular evaluation of isoquinolin-3-ol derivatives bearing amino and amino-derived substituents at C-4. We have found that, in a number of these compounds, particularly certain 4-ureido derivatives **5** in the 6,7-dimethoxy series, the profile shifts from the short-acting inotropic/generalized vasodilating activity of our previous series to a remarkably selective renal vasodilating profile of longer duration. The dramatic increases in renal blood flow provided by the most potent compounds in the ureido series are accompanied by minimal effects on arterial blood pressure and heart rate, indicating a selective vasodilating effect on the renal vasculature.

Chemistry

In our previous report, we described several synthetic approaches to isoquinolin-3-ol systems.[8] As summarized in Scheme I, these compounds may be prepared from amides **6** through an intramolecular cyclization related to the Pomeranz–Fritsch reaction, or through aminative cyclization of either a 2-acylphenylacetic acid derivative **7** or a benzopyrilium salt **9**, derived from **8**. As we noted previously, the 2-acylphenylacetic acids **7** are available in many cases by direct electrophilic acylation of **8**. In the present work, the acetophenone intermediates required for the 7-methoxy- and the 7,8-dimethoxyisoquinolin-3-ol systems were prepared through an alternative general synthesis involving oxidative cleavage of indene derivatives with chromic acid–sulfuric acid in aqueous acetone (Scheme II).[12] This method gave moderate yields of the keto acids which were converted to the esters **12** by Fischer esterification. The indenes were available from 1-indanones by Grignard addition followed by dehydration of the resulting carbinol as shown in Scheme II. The synthesis of the 7,8-dimethoxy derivative **13b** was carried out with the known[13] bromoindanone **10b** as the starting material and the halogen was removed by hydrogenolysis at the keto ester (**12b**) stage. Cyclization to the isoquinolinols **13** was carried out in molten ammonium acetate as we described earlier.[8]

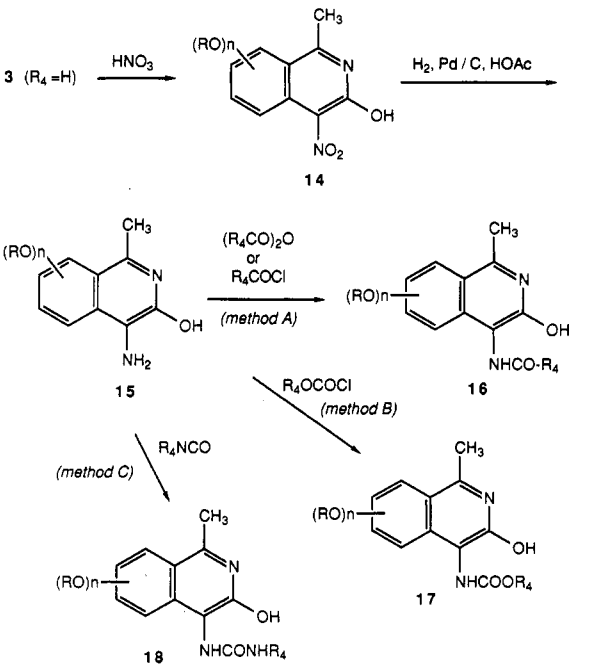
The isoquinolin-3-ol system undergoes preferential electrophilic substitution at C-4,[8,14] and nitration of the 4-unsubstituted isoquinolin-3-ols gave the 4-nitro derivatives **14** (Scheme III; Table I). In the highly activated 6,7,8-trimethoxy system, the 4,5-dinitro compound (**20**) was isolated along with the desired 4-nitro derivative (**21**). Catalytic reduction of the nitro derivatives readily provided the corresponding 4-amino compounds **15**. These amines were moderately unstable, particularly on exposure to air, and typically were used for further synthesis directly as obtained from workup of the reduction. However, in the 6,7-dimethoxy system the 4-amino compound (**28**, Table III) was completely characterized. As outlined in Scheme III, subsequent functionalization of the 4-amino group by standard methodology (methods A–C) provided 4-acylamino (**16**), 4-carbamate (**17**), and 4-ureido (**18**) derivatives. Carboxylic acid **53** was prepared by hydrolysis of the corresponding ester (**52**). Catechol **45** was prepared by boron tribromide demethylation of the dimethoxy analogue **39**. Compounds prepared in this study are listed with their physical properties in Table I and in Tables III–VI.

Scheme II



(a) CH_3MgI; (b) H_3O^+; (c) CrO_3, H_2SO_4, $(CH_3)_2CO$, H_2O; (d) CH_3OH, HCl; (e) H_2, Pd/C; (f) NH_4OAc, 160 °C.

Scheme III



Pharmacology

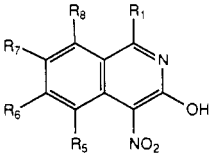
The cardiovascular properties of the isoquinolin-3-ol derivatives were evaluated after intravenous infusion in anesthetized instrumented dogs by using procedures previously described.[8,9,13,14] Renal blood flow (RBF), mean arterial blood pressure (MAP), and heart rate (HR) were measured directly, and renal vascular resistance (RVR) was calculated as the ratio of mean arterial blood pressure to renal blood flow. Effects on contractile force (CF) were also determined for certain compounds. These measurements were compared to pretreatment control values and

(12) For related syntheses of isoquinolines from indenes, see: (a) Miller, R. B.; Frincke, G. M. *J. Org. Chem.* **1980**, *45*, 5312. (b) Dime, D. S.; McLean, S. *J. Org. Chem.* **1981**, *46*, 4999.

(13) de Silva, S. O.; Ahmed, I.; Snieckus, V. *Can. J. Chem.* **1979**, *57*, 598.

(14) Castedo, L.; Estevez, R. J.; Saa, J. M.; Suau, R. *J. Heterocycl. Chem.* **1982**, *19*, 1469.

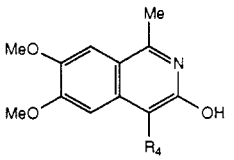
Table I. Physical Properties of 4-Nitroisoquinolin-3-ols



compd	R_1	R_5	R_6	R_7	R_8	mp, °C	formula[a]
19	Me	H	OMe	OMe	H	>310	$C_{12}H_{12}N_2O_5$
20	Me	NO_2	OMe	OMe	OMe	225 dec	$C_{13}H_{13}N_3O_8$
21	Me	H	OMe	OMe	OMe	250–252	$C_{13}H_{14}N_2O_6$
22	Me	H	OMe	OEt	H	257–260 dec	$C_{13}H_{14}N_2O_5$
23	Me	H	H	OMe	H	>250	$C_{11}H_{10}N_2O_4$
24	Me	H	H	OMe	OMe	214–216	$C_{12}H_{12}N_2O_5$[b]
25	H	H	OMe	OMe	H	>300 dec	$C_{11}H_{10}N_2O_5$

[a] Combustion analyses for C, H, and N were within ±0.4% of theory unless otherwise noted. [b] N: calcd, 10.60; found, 10.17.

Table II. Cardiotonic Activity of Selected 4-Substituted Isoquinolin-3-ol Derivatives



compd	R_4	dose[a]	CF, % Δ[b]
26	H	1.75	100[c]
27	Et	0.025	143[c]
28[d]	NH_2	1.88	114 ± 11[e]
29[d]	NHCOMe	8.75	47 ± 2[e]

[a] Dose in mg/kg administered intravenously to anesthetized dogs. [b] Maximal percent increase relative to pretreatment control values in myocardial contractile force (CF) at the total dose reported. [c] Cardiotonic data for this compound taken from ref 8. [d] Physical properties for this compound are reported in Table III. [e] $n = 2$.

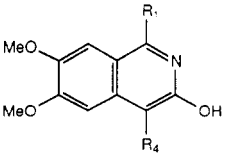
are reported as a percent change for the specified parameter. Activities for the compounds prepared in this study are presented in Tables II–VI.

Our initial studies on 4-amino and functionalized amino substituents centered on the 6,7-dimethoxy system since this alkoxy substitution pattern had provided the most potent analogues in our previous 4-alkyl cardiotonic series. Cardiotonic data were determined for several of the compounds and are presented in Table II. The 4-amino compound **28** has moderate inotropic activity, approximately equipotent with the 4-unsubstituted compound **26**[8] but considerably less potent than the best 4-alkyl derivatives from our earlier isoquinolinol series, such as **27**.[8] As shown in Table III, additional cardiovascular evaluation of amino compound **28** showed that this compound also has renal vasodilating properties as evidenced by a moderate increase in renal blood flow and a corresponding drop in renal vascular resistance without significant effects on blood pressure or heart rate. Acylation of the amino function dramatically reduces the inotropic potency of **29** (Table II) relative to that of **28** although the acylamino derivative **29** retains much of the renal vasodilator potency and has a renal profile similar to that of **28** (Table III). With these data in hand and in view of the potential utility of selective renal vasodilators (vide supra), our interests shifted from further developing the inotropic activity of the 4-amino derivatives and were redirected toward enhancing the potency and specificity of the renal vasodilating profile of the series (Tables III–VI).

Replacement of the 1-methyl group of **29** with hydrogen (**30**) does not significantly alter the renal effects, in contrast with our earlier studies of cardiotonic effects where the equivalent change reduced potency,[8] although increased bulk in the acyl substituent, such as in compounds **31–33**, is detrimental to renal vasodilating activity. We then focused our interest on 4-ureido derivatives as isosteres of the acylamino compounds (Table IV).

The parent 4-ureido derivative **34** in the 6,7-dimethoxy system exhibits an interesting cardiovascular profile. Cardiotonic activity is totally absent in this ureido compound (<10% increase in CF at 1.875 mg/kg iv), demonstrating that positive inotropic activity diminishes dramatically as the 4-substituent is changed from amino (**28**) to acylamino (**29**) to urea (**34**). However, urea **34** has

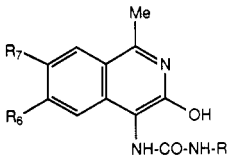
Table III. Cardiovascular Activity and Physical Properties of 4-Amino- and 4-(Acylamino)isoquinolin-3-ols



compd	R_1	R_4	dose[a]	(n)	RBF[b]	RVR[c]	MAP[d]	HR[e]	mp, °C	formula[f,g]
28	Me	NH_2	1.2	(2)	+23 ± 3	−13 ± 4	+6 ± 2	+4 ± 1	250–253 dec	$C_{16}H_{20}N_2O_5$
29	Me	NHCOMe	6.2	(2)	+39 ± 1	−24 ± 2	+6 ± 3	+14 ± 10	312–313 dec	$C_{14}H_{16}N_2O_4 \cdot {}^1/_4H_2O$
30	H	NHCOMe	6.2	(2)	+30 ± 14	−12 ± 16	+14 ± 8	−8 ± 2	284–286	$C_{13}H_{14}N_2O_4 \cdot {}^1/_4H_2O$[h]
31	Me	NHCO-*i*-Pr	6.2	(2)	−5 ± 0	−4 ± 4	−2 ± 2	−1 ± 5	292–294 dec	$C_{16}H_{20}N_2O_4 \cdot {}^1/_2H_2O$
32	Me	NHCO-*n*-Bu	6.2	(2)	−9 ± 5	+13 ± 3	+3 ± 2	+6 ± 4	283–285 dec	$C_{17}H_{22}N_2O_4 \cdot {}^1/_2H_2O$
33	Me	NHCO-*n*-pentyl	6.2	(2)	−27 ± 12	+40 ± 21	0 ± 0	−9 ± 6	276–278 dec	$C_{18}H_{24}N_2O_4$

[a] Cumulative dose in mg/kg administered intravenously to anesthetized dogs. [b] Renal blood flow as maximal % change relative to pretreatment control values. [c] Renal vascular resistance as % change relative to pretreatment control values. [d] Mean arterial blood pressure as % change relative to pretreatment control values. [e] Heart rate as % change from pretreatment control values. [f] Combustion analyses for C, H, and N were within ±0.4% of theory unless otherwise noted. [g] Compound **28** was obtained by reduction of **19** (see the Experimental Section) and compounds **29–33** were prepared by method A. [h] N: calcd, 10.49; found, 9.99.

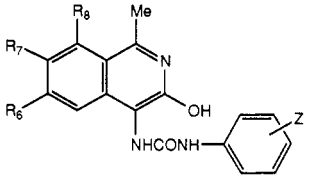
Table IV. Cardiovascular Activity and Physical Properties of 4-(Alkylureido)isoquinolin-3-ols



compd	R	R_6	R_7	dose[a]	(n)	RBF[b]	RVR[c]	MAP[d]	HR[e]	mp, °C	formula[f,g]
34	H	OMe	OMe	6.2	(3)	+59 ± 19	−23 ± 8	+18 ± 4	−10 ± 8	250–255	$C_{13}H_{15}N_3O_4$
35	Me	OMe	OMe	1.2	(2)	+4 ± 17	−5 ± 17	+4 ± 2	+3 ± 7	265–267 dec	$C_{14}H_{17}N_3O_4 \cdot {}^1/_4H_2O$
36	Et	OMe	OMe	1.2	(2)	+28 ± 10	−22 ± 3	0 ± 3	0 ± 1	236–238 dec	$C_{15}H_{19}N_3O_4$
37	*n*-Pr	OMe	OMe	1.2	(2)	+5 ± 16	0 ± 21	+1 ± 5	−7 ± 2	234–236 dec	$C_{16}H_{21}N_3O_4$[h]
38	allyl	OMe	OMe	1.2	(2)	+97 ± 1	−44 ± 4	+8 ± 6	−3 ± 17	224–226 dec	$C_{16}H_{19}N_3O_4 \cdot {}^1/_2H_2O$
39	*n*-Bu	OMe	OMe	6.2	(2)	+117 ± 11	−52 ± 5	+4 ± 5	−2 ± 7	230–232 dec	$C_{17}H_{23}N_3O_4 \cdot {}^1/_2H_2O$
39				1.2	(2)	+36 ± 19	−27 ± 12	−3 ± 3	+6 ± 4		
40	*i*-Pr	OMe	OMe	1.2	(2)	+11 ± 5	−10 ± 3	−9 ± 2	+3 ± 13	237–239 dec	$C_{16}H_{21}N_3O_4$
41	*t*-Bu	OMe	OMe	6.2	(2)	+10 ± 3	−8 ± 6	+1 ± 4	−4 ± 8	276–278 dec	$C_{17}H_{23}N_3O_4 \cdot {}^1/_4H_2O$
42	c-C_6H_{11}	OMe	OMe	1.2	(2)	+7 ± 17	−4 ± 12	0 ± 3	0 ± 2	248–250 dec	$C_{19}H_{25}N_3O_4 \cdot {}^1/_2H_2O$
43	allyl	OMe	OEt	1.2	(3)	+28 ± 11	−22 ± 9	−2 ± 4	+29 ± 14	208–210 dec	$C_{17}H_{21}N_3O_4 \cdot {}^1/_4H_2O$
44	*n*-Bu	H	OMe	1.2	(2)	−12 ± 8	+16 ± 10	+3 ± 0	−6 ± 2	217–220 dec	$C_{16}H_{21}N_3O_3$[i]
45	*n*-Bu	OH	OH	1.2	(2)	−2 ± 9	+2 ± 10	−1 ± 1	+2 ± 4	202–204 dec	$C_{15}H_{19}N_3O_4 \cdot HBr$[j]

[a] Cumulative dose in mg/kg administered intravenously to anesthetized dogs. [b] Renal blood flow as maximal % change relative to pretreatment control values. [c] Renal vascular resistance as % change relative to pretreatment control values. [d] Mean arterial blood pressure as % change relative to pretreatment control values. [e] Heart rate as % change from pretreatment control values. [f] Combustion analyses for C, H, and N were within ± 0.4% of theory unless otherwise noted. [g] Compounds prepared according to method C unless otherwise noted. [h] N: calcd, 13.16; found, 12.67. [i] N: calcd, 13.67; found, 13.16. [j] Obtained by BBr_3 demethylation of dimethoxy compound **39** (see the Experimental Section).

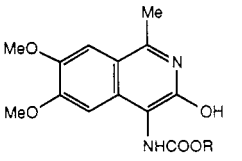
Table V. Cardiovascular Activity and Physical Properties of 4-(Arylureido)isoquinolin-3-ols



compd	Z	R_6	R_7	R_8	dose[a]	(n)	RBF[b]	RVR[c]	MAP[d]	HR[e]	mp, °C	formula[f,g]
46	H	OMe	OMe	H	1.2	(2)	−23 ± 15	+35 ± 30	0 ± 2	−13 ± 3	236–238 dec	$C_{19}H_{19}N_3O_4$
47	3-OMe	OMe	OMe	H	6.2	(2)	+59 ± 15	−36 ± 3	−1 ± 5	−2 ± 0	239–241 dec	$C_{20}H_{21}N_3O_5 \cdot {}^1/_2H_2O$
48	4-OMe	OMe	OMe	H	6.2	(2)	+68 ± 18	−32 ± 15	+12 ± 12	+2 ± 6	235–237 dec	$C_{20}H_{21}N_3O_5 \cdot {}^1/_2H_2O$
49	3-CF_3	OMe	OMe	H	6.2	(2)	+23 ± 7	−16 ± 2	+2 ± 2	−10 ± 8	257–259 dec	$C_{20}H_{18}F_3N_3O_4 \cdot {}^1/_4H_2O$
50	4-CF_3	OMe	OMe	H	6.2	(3)	+49 ± 7	−29 ± 0	+4 ± 4	+10 ± 8	242–244 dec	$C_{20}H_{18}F_3N_3O_4$
51	4-NO_2	OMe	OMe	H	1.2	(2)	+3 ± 7	−2 ± 7	+1 ± 1	−1 ± 1	270–272 dec	$C_{19}H_{18}N_4O_6 \cdot {}^1/_4H_2O$
52	4-CO_2Et	OMe	OMe	H	1.2	(2)	+2 ± 4	−4 ± 2	−2 ± 2	−1 ± 3	222–224 dec	$C_{22}H_{23}N_3O_6 \cdot {}^1/_4H_2O$
53	4-CO_2H	OMe	OMe	H	1.2	(2)	+40 ± 8	−28 ± 2	−1 ± 3	−12 ± 2	244–246 dec	$C_{20}H_{19}N_3O_6 \cdot {}^5/_2H_2O$[h]
54	4-OMe	OMe	OEt	H	6.2	(2)	−8 ± 2	+15 ± 4	+6 ± 2	+10 ± 8	237–240 dec	$C_{21}H_{23}N_3O_5$
55	4-OMe	OMe	OMe	OMe	6.2	(2)	+14 ± 4	−15 ± 3	−4 ± 5	+7 ± 3	211–214 dec	$C_{21}H_{23}N_3O_6$
56	3-OMe	H	OMe	OMe	1.2	(2)	+17 ± 11	−16 ± 9	−3 ± 2	−2 ± 12	185–187 dec	$C_{20}H_{21}N_3O_5 \cdot {}^7/_{10}H_2O$
57	3-OMe	H	OMe	H	1.2	(4)	+17 ± 20	−4 ± 14	+2 ± 2	+5 ± 3	197–200 dec	$C_{19}H_{19}N_3O_4$

[a] Cumulative dose in mg/kg administered intravenously to anesthetized dogs. [b] Renal blood flow as maximal % change relative to pretreatment control values. [c] Renal vascular resistance as % change relative to pretreatment control values. [d] Mean arterial blood pressure as % change relative to pretreatment control values. [e] Heart rate as % change from pretreatment control values. [f] Combustion analyses for C, H, and N were within ±0.4% of theory. [g] Compounds prepared by method C unless otherwise noted. [h] Obtained by basic hydrolysis of ester **52** (see the Experimental Section).

Table VI. Cardiovascular Activity and Physical Properties of Urethane Isosteres of 4-(Alkylureido)isoquinolin-3-ols



compd	R	dose[a]	(n)	RBF[b]	RVR[c]	MAP[d]	HR[e]	mp, °C	formula[f]
58	allyl	6.2	(2)	+2 ± 4	−11 ± 3	−9 ± 6	−2 ± 2	252–254 dec	$C_{16}H_{18}N_2O_5$
59	*n*-Bu	6.2	(3)	−23 ± 6	+29 ± 15	−3 ± 4	−6 ± 2	282–284 dec	$C_{17}H_{22}N_2O_5$

[a] Cumulative dose in mg/kg administered intravenously to anesthetized dogs. [b] Renal blood flow as maximal % change relative to pretreatment control values. [c] Renal vascular resistance as % change relative to pretreatment control values. [d] Mean arterial blood pressure as % change relative to pretreatment control values. [e] Heart rate as % change from pretreatment control values. [f] Combustion analysis for C, H, and N within ±0.4% of theory; prepared by method B.

significant renal vasodilating properties at the 6.2 mg/kg dose without untoward effects on other cardiovascular parameters (Table IV), similar to acetamide **29**. We were encouraged by the observation that the *n*-butylureido (**39**) analogue at the 6.2 mg/kg dose has potent and selective renal vasodilating activity and maintains the selectivity and substantial potency at the 1.2 mg/kg dose level. At the lower dose, although the methyl (**35**), ethyl (**36**), and *n*-propyl (**37**) analogues along with the more bulky isopropyl (**40**) and cyclohexyl (**42**) congeners all were significantly less potent than the *n*-butyl compound **39**, allyl derivative **38** emerged as the most potent compound in the series. At 1.2 mg/kg, this compound produces dramatic maximal increases (97%) in renal blood flow with no significant reduction in arterial blood pressure and no significant chronotropic effects.

The interesting levels of selective renal vasodilation shown by **38** and **39** led us to explore selected changes in the alkoxy substitution pattern. The allyl derivative **43** in the 6-methoxy-7-ethoxy system is less potent than the corresponding 6,7-dimethoxy compound **38**. At equivalent dose levels, the renal vasodilating potency shown by the *n*-butyl derivative (**44**) in the 7-methoxy series is diminished relative to that of the 6,7-dimethoxy compound **39**. These results indicate that the presence of a 6-methoxy group is advantageous, and a particular preference for alkoxy substitution at the 6- and 7-positions is indicated by the lack of renal vasodilating activity of *n*-butylureido catechol **45** compared with the excellent potency shown by the corresponding dimethoxy compound **39** at the same dose. These results suggest that 6,7-dimethoxy is the preferred alkoxy substitution pattern of those examined in the alkyl urea series.

To further explore the ureido substituent, we also examined a series of ureas with aryl substitution (Table V). On the basis of results with the alkylureas as discussed above, the initial aryl derivatives were prepared in the 6,7-dimethoxy series. Although phenylurea **46** and the 4-nitro (**51**) and 4-carbethoxy (**52**) analogues are not renal vasodilators at the 1.2 mg/kg dose, several compounds with substituted aromatic rings were identified as having renal vasodilating effects at the 6.2 mg/kg dose level. Thus, compounds with methoxy (**47**, **48**) or trifluoromethyl (**49**, **50**) substitution in either the 3- or 4-positions have moderate levels of activity. Interestingly, the 4-carboxylic acid derivative **53** is the most active of the arylureas and has renal vasodilating activity at the 1.2 mg/kg dose comparable to that of the *n*-butyl analogue **39** in the alkyl series.

We also examined the effect of selected changes in the alkoxy substitution pattern of the isoquinoline moiety of the arylureas. The data in Table V for the 6-methoxy-7-ethoxy (**54**), 6,7,8-trimethoxy (**55**), 7,8-dimethoxy (**56**), and 7-methoxy (**57**) systems relative to their 6,7-dimethoxy counterparts (cf. **47**, **48**) indicate that, as in the alkylurea series, 6,7-dimethoxy substitution is a preferred pattern. It was also of interest to determine the effect of isosteric replacements in the side chain of the more active alkylureas. Comparison of the data for amide **33** (Table III) and *n*-butylurea **39** (Table IV) shows that replacement of the N′ urea nitrogen with methylene diminishes renal vasodilating activity, and replacement of this nitrogen with oxygen also causes loss of renal potency as shown by comparison of the levels of renal vasodilation of ureas **38** and **39** and urethanes **58** and **59** (Table VI).

Several structure–activity conclusions may be drawn from the data discussed above for the 4-ureidoisoquinolinols. The 6,7-dimethoxy system is a preferred alkoxy substitution for the ureas, and for good renal vasodilator activity, the N′ nitrogen of the urea moiety is important. In the alkyl urea series, allyl and *n*-butyl side chains are optimal. Aromatic ring substituents such as 4-carboxyl and methoxy or trifluoromethyl in the 3- or 4-positions confer the greatest potency in the arylurea series; there is no definitive correlation of potency with electronic or lipophilic parameters in the arylureas. Of the ureido derivatives, the *n*-butyl (**39**), allyl (**38**), and 4-carboxyphenyl (**53**) compounds clearly are the most interesting selective renal vasodilating agents.

The selective cardiovascular profiles of the allyl (**38**) and *n*-butyl (**39**) analogues in the anesthetized dog are shown in Figures 1 and 2. This graphical representation dramatically illustrates the remarkably selective profile of these compounds. These derivatives provide a profound increase in renal blood flow and concomitant reduction in renal vascular resistance unaccompanied by significant changes in heart rate or arterial blood pressure. This profile persists for several hours, and the absence of chronotropic and hypotensive effects demonstrates that **38** and **39** do not have general vasodilating properties but instead cause a specific dilation of the renal vasculature.

In summary, we have shown that the substituent at C-4 has a significant role in determining the overall cardiovascular profile of the isoquinolinols. Our earlier studies showed that 4-unsubstituted, 4-halo and especially 4-alkyl derivatives are inotropic agents with a short duration of cardiotonic action accompanied by modest generalized vasodilating properties.[8] In the present study, we have shown that a 4-amino substituent confers both cardiotonic and renal vasodilating activities and that acylation of the 4-amino function diminishes cardiotonic activity but maintains renal activity. Moreover, we have shown that although cardiotonic activity is completely eliminated when the 4-substituent is a 4-ureido moiety, certain 4-ureido derivatives, especially **38** and **39**, are potent renal vasodilators with an overall cardiovascular profile that indicates remarkable selectivity for dilation of renal vasculature in the dog.

Experimental Section

Chemistry. Melting point determinations were performed on a Thomas-Hoover capillary melting point apparatus and are uncorrected. All compounds exhibited spectral data consistent with their assigned structures and were homogeneous by thin-layer chromatography (TLC). Proton nuclear magnetic resonance (^{1}H NMR) spectra were obtained on Varian XL-400 or FT-80A or IBM WP-100 spectrometers and are reported in δ downfield from tetramethylsilane (TMS) as internal standard. Mass spectra were obtained on a Finnigan 1015D quadrupole mass spectrometer coupled to a Finnigan 9500 gas chromatograph or on a Finnigan MAT 8230 double-focusing high-resolution mass spectrometer. Combustion analyses for C, H, and N were within 0.4% of theory unless otherwise noted. Compounds in the study were prepared according to the general procedures described below and physical properties of the compounds are summarized in Table I and in Tables III–VI. With the exception of those prepared by the indanone route described below, all isoquinolin-3-ol derivatives used as starting materials in this work were synthesized as described in our earlier report.[8]

Nitration of Isoquinolin-3-ols: General Procedures. (a) 3-Hydroxy-6,7-dimethoxy-1-methyl-4-nitroisoquinoline (19). 3-Hydroxy-6,7-dimethoxy-1-methylisoquinoline[8] (0.927 g, 4.23 mmol) was dissolved in glacial AcOH (60 mL) by warming and when the solution had cooled to 15 °C a crystalline solid separated. To this mechanically stirred slurry at 15 °C was added during a 15-min period 1.5 mL of a nitrating mixture composed of 0.6 mL of glacial AcOH and 0.9 mL of 90% nitric acid. The mixture, which contained a heavy yellow solid, was stirred for an additional 15 min and then poured into water (300 mL). The precipitate was collected by filtration, washed thoroughly with water, and dried in vacuo to provide **19** in 63% yield (0.70 g): mp >310 °C;

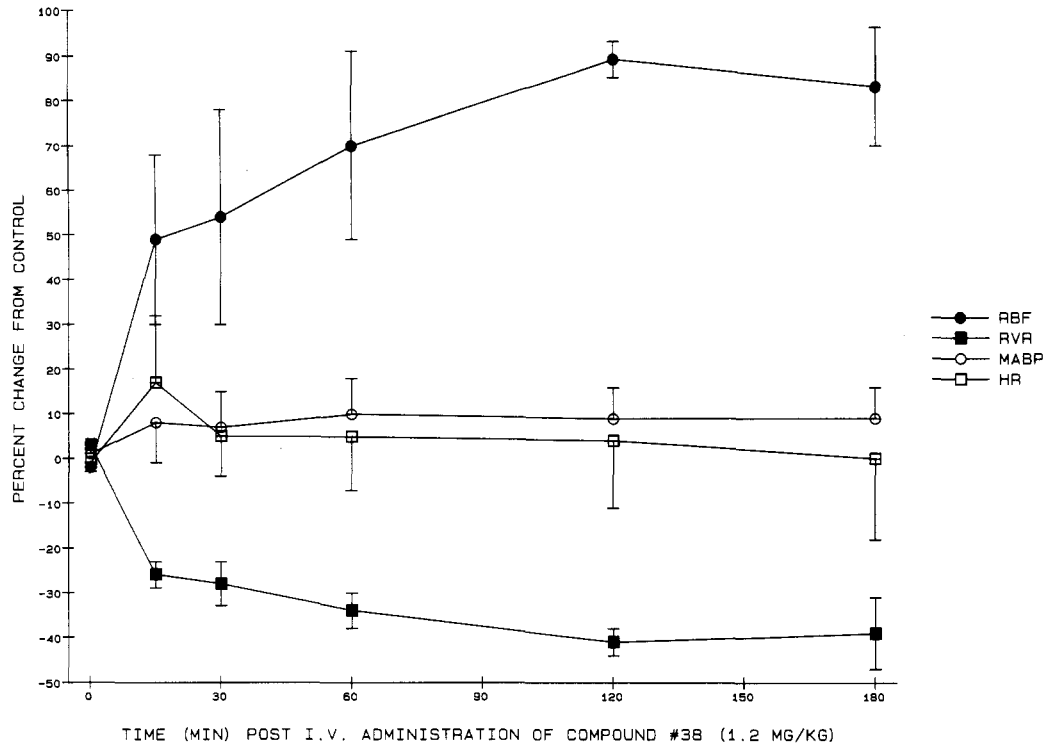


Figure 1. Renal vasodilator activity and specificity of allylureido analogue **38** after intravenous administration (1.2 mg/kg; $n = 2$) in the anesthetized dog.

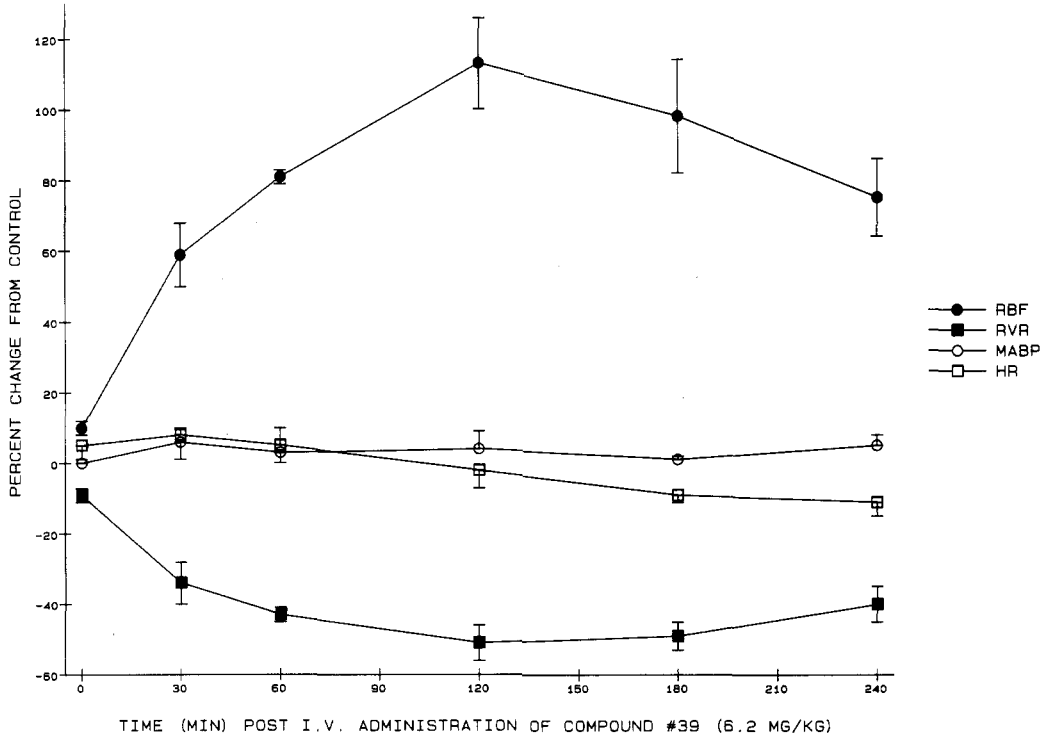


Figure 2. Renal vasodilator activity and specificity of *n*-butylureido analogue **39** after intravenous administration (6.2 mg/kg; $n = 2$) in the anesthetized dog.

NMR (TFA-*d*) δ 3.23 (s, 3 H), 4.20 (s, 3 H), 4.32 (s, 3 H), 7.65 (s, 1 H), 8.70 (s, 1 H); MS *m/z* 264.

(b) 3-Hydroxy-7,8-dimethoxy-1-methyl-4-nitroisoquinoline (24). Nitric acid (90%, 1 mL) was cautiously added dropwise (**caution**: exothermic) to cold (0 °C) Et_2O (12 mL). Then 3-hydroxy-7,8-dimethoxy-1-methylisoquinoline (0.510 g, 2.3 mmol) was added to the cold mixture in a single portion. The cooling bath was removed and the mixture was stirred for 10 min. The red solid was collected, washed successively with Et_2O and MeOH, and then dried in vacuo to provide **24** (0.250 g, 41%): mp 214–216 °C; NMR (DMSO-d_6) δ 2.95 (s, 3 H), 3.90 (two overlapping 3 Hs, 6 H total), 7.70 and 7.88 (each 1 H, AB pattern, J_{AB} = 9 Hz).

General Procedure for Reduction of the 4-Nitro Compounds: 4-Amino-3-hydroxy-6,7-dimethoxy-1-methylisoquinoline (28). Nitro compound **19** (16.0 g, 60.5 mmol) was slurried in glacial AcOH (600 mL) and hydrogenated over 10% Pd/C (1.5 g) at 17 psi for 1.5 h at room temperature to provide a yellow solution. The catalyst was removed by filtration through Celite and the filtrate was concentrated to dryness under reduced pressure to provide **28** as the diacetate solvate (20.48 g, 96%): mp 111–115 °C; NMR ($CDCl_3$) δ 2.07 (s, 6 H), 2.63 (s, 3 H), 3.90 (s, 3 H), 3.95 (s, 3 H), 6.57 (s, 1 H), 6.67 (s, 1 H), 9.18 (br, 5 H).

Treatment of the diacetate solvate with triethylamine in methanol gave the free base of **28** as a yellow solid, mp 250–254 °C.

Method A. Synthesis of 4-Acylamino Compounds. (a) 4-Acetamido-3-hydroxy-6,7-dimethoxy-1-methylisoquinoline (29). A slurry of nitro compound **19** (2.028 g, 7.67 mmol) in acetic anhydride (28 mL) and AcOH (180 mL) was hydrogenated over 10% Pd/C (0.8 g) at 25 psi for 3 h. The mixture was filtered through a pad of Celite and the collected solid was washed with acetic acid. The filtrate and washings were combined and evaporated to dryness under reduced pressure. The residue was recrystallized from MeOH to provide **29** (0.70 g, 33%) as the hemihydrate: mp 300–301 °C dec; NMR (TFA-*d*) δ 2.63 (s, 3 H), 3.12 (s, 3 H), 4.17 (s, 3 H), 4.20 (s, 3 H), 7.30 (s, 1 H), 7.48 (s, 1 H), 5.28 (br s, 1 H, NH); MS *m/z* 276.

(b) 3-Hydroxy-4-isobutyramido-6,7-dimethoxy-1-methylisoquinoline (31). A solution of amine **28** (1.77 g, 5 mmol) and triethylamine (2.5 mL, 16.5 mmol) in methylene chloride (50 mL) was treated with isobutyryl chloride (0.63 mL, 6 mmol) and then stirred under nitrogen for 1 h. The slurry was evaporated to dryness and boiled in MeOH (50 mL) and water (5 mL) for 15 min. The solid was collected by filtration and washed successively with MeOH and acetone and then dried under vacuum at 50 °C to provide amide **31** (0.60 g, 39%): mp 292–294 °C; NMR (TFA-*d*) δ 1.52 (d, *J* = 7 Hz, 6 H), 3.12 (s, 3 H), 4.15 (s, 3 H), 4.18 (s, 3 H), 7.23 (s, 1 H), 7.50 (s, 1 H), 9.20 (br s, 1 H, NH); MS *m/z* 304.

Method B. Urethane Formation. 4-[*N*-(*n*-Butoxycarbonyl)amino]-3-hydroxy-6,7-dimethoxy-1-methylisoquinoline (59). *n*-Butyl chloroformate (0.578 g, 4.24 mmol) was added to a slurry of amine **28** (0.900 g, 3.85 mmol) and triethylamine (0.409 g, 4.04 mmol) in $CHCl_3$ (50 mL). The mixture was stirred overnight and the solid was then collected by filtration and was washed with MeOH followed by Et_2O. Trituration of the solid with MeOH gave urethane **59** (0.520 g, 40%) of mp 282–284 °C: NMR (TFA-*d*) δ 3.28 (s, 3 H), 4.18 (s, 3 H), 4.21 (s, 3 H), 7.32–8.05 (m, 5 H), 8.15–8.38 (m, 3 H, includes NH), 10.5 (br s, 1 H, NH); MS (FAB) *m/z* 398 (M + 1).

Method C. Urea Formation. (a) 6,7-Dimethoxy-3-hydroxy-1-methyl-4-ureidoisoquinoline (34). Sodium cyanate (0.39 g, 6 mmol) was added in one portion to a solution of amine **28** (1.772 g, 5 mmol) in AcOH (50 mL) under nitrogen. A clear solution formed which soon became a thick slurry. The mixture was stirred overnight at room temperature and the solid was then collected by filtration, washed successively with AcOH, EtOAc, acetone, and Et_2O, and then dried to provide the monoacetate solvate of **34** as a yellow solid. This solid was slurried in MeOH (50 mL) and triethylamine (3 mL) was added and the mixture was heated under reflux under a nitrogen atmosphere for 1 h. The slurry was filtered and the yellow solid was washed with MeOH and dried under vacuum at 50 °C to give 0.80 g (58%) of **34**: mp 250–255 °C; NMR (TFA-*d*) δ 3.13 (s, 3 H), 4.18 (s, 3 H), 4.22 (s, 3 H), 7.42 (s, 1 H), 7.53 (s, 1 H), 8.08 (br s, 1 H); MS *m/z* 277.

(b) 4-(*N'*-*n*-Butylureido)-3-hydroxy-6,7-dimethoxy-1-methylisoquinoline (39). To a stirred solution of amine **28** (1.722 g, 5 mmol, diacetate solvate) in AcOH (9 mL) was added dropwise *n*-butyl isocyanate (0.68 mL, 6 mmol) during a 3-min period. Within the first 0.5 h the reaction mixture became a thick gel and was diluted with additional AcOH (2 mL). The viscous slurry was stirred overnight, and the solid was then isolated by filtration, washed successively with AcOH (small volume), acetone, and Et_2O, and then dried under vacuum to afford **39** in 41% yield (0.688 g) as the hemihydrate: mp 230–232 °C; NMR (TFA-*d*) δ 1.02 (t, *J* = 7 Hz, 3 H), 1.58 (br m, 4 H), 3.10 (s, 3 H), 3.50 (br t, *J* = 7 Hz, 2 H), 4.17 (s, 3 H), 4.20 (s, 3 H), 7.38 (s, 1 H), 7.50 (s, 1 H), 8.00 (br s, exchangeable).

4-[*N'*-(4-Carboxyphenyl)ureido]-3-hydroxy-6,7-dimethoxy-1-methylisoquinoline (53). A slurry of ester **52** (1.00 g, 2.35 mmol) in 2 N aqueous NaOH was stirred overnight at room temperature. The mixture was acidified with 3 N HCl and the yellow solid was collected by filtration and washed successively with water, acetone, and Et_2O and dried under vacuum to afford acid **53** (0.720 g, 77%): mp 224–246 °C dec; NMR (TFA-*d*) δ 3.28 (s, 3 H), 4.18 (s, 3 H), 4.21 (s, 3 H), 7.32–8.05 (m, 5 H), 8.15–8.38 (m, 3 H), 10.5 (br s, 1 H).

Isoquinolinol Synthesis from Indanones: 7,8-Dimethoxy-1-methylisoquinolin-3-ol (13b). To a solution of 4-bromo-6,7-dimethoxy-1-indanone[13] (**10b**; 45 g, 0.16 mol) in benzene (450 mL) was added an ethereal solution of methylmagnesium bromide (95 mL, 2.85 M, 0.27 mol), and the mixture was refluxed under nitrogen for 3 h and then allowed to cool. Concentrated HCl (30 mL) was cautiously added and the mixture was stirred for 0.5 h. Water (50 mL) and Et_2O (100 mL) were added, and the organic layer was washed with water, dried ($MgSO_4$), and concentrated to give 4-bromo-6,7-dimethoxy-1-methylindene (**11b**; 40 g, 93%) as an oil which slowly crystallized: NMR ($CDCl_3$) δ 2.30 (m, 3 H), 3.16 (m, 2 H), 3.83 (s, 6 H), 6.10 (m, 1 H), 6.83 (s, 1 H). Anal. ($C_{12}H_{13}BrO_2$) C, H.

To a solution of **11b** (40 g, 0.15 mol) in acetone (400 mL) at 5 °C was added a solution of chromium trioxide (32 g, 0.32 mol) and concentrated H_2SO_4 (32 mL, 0.6 mol) in water (200 mL). The reaction mixture was stirred 3 h at room temperature, and then Et_2O (300 mL) and water (300 mL) were added. The aqueous layer was extracted with several portions of Et_2O, and the combined Et_2O layers were extracted with 10% aqueous $NaHCO_3$ (4 × 150 mL). The basic extracts were then combined and acidified with 6 N HCl, and after 20 min the mixture was filtered to provide 2-acetyl-6-bromo-3,4-dimethoxyphenylacetic acid (22.5 g, 47%) as pale yellow granular crystals; mp 108–110 °C; NMR ($CDCl_3$) δ 2.53 (s, 3 H), 3.61 (s, 2 H), 3.83 (s, 3 H), 3.88 (s, 3 H), 7.16 (s, 1 H), 9.58 (br, 1 H, exchangeable). Anal. ($C_{12}H_{13}BrO_5$) C, H.

The keto acid obtained above (26 g, 82 mmol) was refluxed for 3 h in MeOH (150 mL) containing concentrated HCl (10 drops). The mixture was diluted with water and upon cooling deposited crystals which were collected by filtration and then dissolved in methylene chloride. This solution was filtered and evaporated under reduced pressure to afford methyl 2-acetyl-6-bromo-3,4-dimethoxyphenylacetate (**12b**; 23 g, 85%): mp 65–67 °C; NMR ($CDCl_3$) δ 2.55 (s, 3 H), 3.60 (s, 2 H), 3.63 (s, 3 H), 3.81 (s, 3 H), 3.85 (s, 3 H), 6.88 (s, 1 H). Anal. ($C_{13}H_{15}BrO_5$) C, H.

Bromo ester **12b** (15 g, 45 mmol) was then dehalogenated by hydrogenation over 10% Pd/C in AcOH (150 mL) containing NaOAc (8 g, 98 mmol). After hydrogenolysis was complete, the reaction mixture was filtered and the filtrate was evaporated to dryness under reduced pressure. The solid residue was thoroughly washed with water and then dried to give methyl 2-acetyl-3,4-dimethoxyphenylacetate (**12c**; 11.3 g, 97%): mp 72–74 °C; NMR ($CDCl_3$) δ 2.55 (s, 3 H), 3.65 (s, 2 H), 3.83 (s, 3 H), 3.85 (s, 3 H), 6.88 (s, 2 H). Anal. ($C_{13}H_{16}O_5 \cdot 0.1H_2O$) C, H.

Ester **12c** (11.2 g, 45 mmol) was added to molten (160 °C) ammonium acetate (400 g) and the mixture was stirred at 160 °C for 1 h. After cooling, water (400 mL) was added and the mixture was stirred at room temperature for 1 h and then filtered to provide 7,8-dimethoxy-1-methylisoquinolin-3-ol (**13b**; 8.5 g, 87%): mp 141–144 °C; NMR (DMSO-d_6) δ 2.93 (s, 3 H), 3.88 (s, 3 H), 3.90 (s, 3 H), 6.65 (s, 1 H), 7.43 (m, 2 H). Anal. ($C_{12}H_{13}NO_3 \cdot {}^3/_4H_2O$) C, H, N.

4-(*N'*-*n*-Butylureido)-3,6,7-trihydroxy-1-methylisoquinoline Hydrobromide (45). To a stirred cold (−78 °C) slurry of dimethoxy compound **39** (2.50 g, 50 mmol) in CH_2Cl_2 (100 mL) was added boron tribromide (8.46 g, 33.8 mmol). The mixture was allowed to slowly warm to ambient temperature and was then recooled to 0 °C and quenched with MeOH. Solvents were removed under reduced pressure, and the residue was triturated with Et_2O to give **45** (2.71 g, 94%) of mp 202–204 °C: NMR (DMSO-d_6) δ 0.85–1.75 (m, 7 H), 2.82 (s, 3 H), 3.15 (br s, 2 H), 7.07 (br s, 1 H, NH), 7.27 (s, 1 H), 7.40 (s, 1 H), 8.50 (br s, 1 H, NH).

Cardiotonic Activity.[8,15] Adult mongrel dogs were anesthetized with sodium pentobarbital (45 mg/kg, ip) and were artificially respired. A right thoracotomy was performed and myocardial contractile force (CF) was measured with a Walton Brodie strain gauge sutured to the right ventricle and adjusted to produce a baseline of 100 g tension. A standard dose (10–15 μg/kg per min × 3 min) of dopamine was administered to assess myocardial responsiveness to inotropic stimulation. Only animals producing over 100% increase in CF were used subsequently. Compounds were solubilized in dimethylformamide (10–15%) and administered at the total dose reported in Table II by intravenous infusion into a cannulated femoral vein. Dose-related effects of

(15) Alousi, A. A.; Farah, A. I.; Lesher, G. Y.; Opalka, C. J. *Circ. Res.* **1979**, *45*, 666.

the test compound on contractile force (CF) were compared to pretreatment control values and expressed as a percent change.

Renal Vasodilating Activity.[16] Adult mongrel dogs were anesthetized (sodium pentobarbital; 45 mg/kg ip) and surgically prepared for electromagnetic measurement of renal artery blood flow. A femoral artery was cannulated for measuring arterial blood pressure, and drugs were administered intravenously via the femoral vein. Heart rate was monitored with a cardiotachometer. Renal vascular resistance was calculated as the ratio of mean arterial blood pressure to renal artery blood flow. Dopamine was infused intravenously at 3 μg/kg per min for 10 min (at an infusion rate of about 1 mL/min) to determine responsiveness of each dog to a known renal vasodilator. At this dose, dopamine increases renal blood flow approximately 30% without appreciable α or β adrenergic receptor mediated vasoconstriction and cardiac stimulation. Cumulative dose–response data were obtained in groups of two or more dogs by administering the test compound at progressively increasing infusion rates (normally 5-fold increments: 0.2 and 1.0 mg/kg for the cumulative 1.2 mg/kg dose, and 0.2, 1.0, and 5.0 mg/kg for the cumulative 6.2 mg/kg dose), each dose being infused for 5 min. Compounds were solubilized in 0.9% physiological saline with some requiring aqueous acid or base (0.1–1.0 mequiv) for solubilization. These vehicles alone were shown not to alter any measured parameter by more than ±5% of baseline values. All animals were monitored for at least 30 min postdrug treatment and the maximum percent change from predrug control was quantitated for renal artery blood flow (RBF), renal vascular resistance (RVR), mean arterial blood pressure (MAP), and heart rate (HR). At the infusion dose of 3 μg/kg per min under the conditions described above, dopamine provided the following percent changes in the measured parameters (n = 25): RBF = +32 ± 3; RVR = −27 ± 2; MAP = −6 ± 1; HR = +5 ± 2.

(16) Goldberg, L. I.; Sonneville, P. F.; McNay, J. L. *J. Pharmacol. Exp. Ther.* **1968**, *163*, 188.

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Registry No. **10b**, 18028-29-0; **11b**, 118575-95-4; **12b**, 118575-96-5; **12c**, 118575-92-1; **13b**, 114130-58-4; **15** (R = 7-OMe; n = 1), 118575-97-6; **15** (R = 6-OMe, 7-OMe, 8-OMe; n = 3), 118575-99-8; **15** (R = 7-OMe, 8-OMe; n = 2), 118576-00-4; **16** (R = 6-OH, 7-OH; n = 2; R_4 = Bu), 118575-98-7; **19**, 113982-73-3; **20**, 113982-75-5; **21**, 113982-76-6; **22**, 113982-79-9; **23**, 113982-88-0; **24**, 113983-47-4; **25**, 118576-01-5; **26**, 16535-98-1; **27**, 114130-71-1; **28**, 113983-32-7; **28** diacetate, 118575-90-9; **29**, 113982-78-8; **30**, 113982-77-7; **31**, 113982-86-8; **32**, 113982-85-7; **33**, 118576-02-6; **34**, 113982-84-6; **34** acetate, 118575-91-0; **35**, 113983-40-7; **36**, 113983-45-2; **37**, 118576-03-7; **38**, 113983-08-7; **39**, 113982-89-1; **40**, 113983-46-3; **41**, 113983-13-4; **42**, 113983-23-6; **43**, 118576-04-8; **44**, 113983-29-2; **45**, 118576-05-9; **46**, 113983-35-0; **47**, 114052-45-8; **48**, 113982-87-9; **49**, 113983-02-1; **50**, 113983-00-9; **51**, 113983-11-2; **52**, 113983-21-4; **53**, 113983-42-9; **54**, 113982-91-5; **55**, 118597-18-5; **56**, 118576-06-0; **57**, 118576-07-1; **58**, 113983-44-1; **59**, 113983-31-6; 3-hydroxy-6,7-dimethoxy-1-methylisoquinoline, 16535-98-1; 3-hydroxy-7,8-dimethoxy-1-methylisoquinoline, 114130-58-4; n-butyl chloroformate, 592-34-7; 2-acetyl-6-bromo-3,4-dimethoxyphenylacetic acid, 118575-93-2; 4-amino-7-ethoxy-3-hydroxy-6-methoxy-1-methylisoquinoline, 118575-94-3; allyl chloroformate, 2937-50-0.

Synthesis and Antiproliferative Effects of Novel 5′-Fluorinated Analogues of 5′-Deoxy-5′-(methylthio)adenosine

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5′-Deoxy-5′-[(monofluoromethyl)thio]adenosine (**9**) and 5′-deoxy-5′-fluoro-5′-(methylthio)adenosine (**10**), two novel analogues of 5′-deoxy-5′-(methylthio)adenosine (MTA), have been synthesized and evaluated for their substrate and inhibitory activities toward MTA phosphorylase and for their biological effects in L1210 (MTA phosphorylase deficient) and L5178Y (MTA phosphorylase containing) murine leukemia cell lines. Compound **9** was a potent competitive inhibitor of MTA phosphorylase with a K_i value of 3.3 μM and was also a substrate, with activity approximately 53% that of MTA. Compound **10** was significantly less inhibitory toward the phosphorylase with a K_i value of 141 μM; its lack of substrate activity was attributed to rapid nonenzymatic degradation. The 50% growth inhibitory concentrations (48 h) of **9** were 300 and 200 μM in L1210 and L5178Y cells, respectively; for **10**, these respective values were 2 and 0.7 μM. The initial characterization of **9** in these systems reveals that it differs from MTA by not acting as a product regulator of the polyamine biosynthetic pathway.

5′-Deoxy-5′-(methylthio)adenosine (MTA) (**3**) is a metabolite of *S*-adenosylmethionine (AdoMet), which is formed as a byproduct during synthesis of the polyamines, spermidine (Spd), and spermine (Spm). The maintenance of low intracellular levels of MTA via further metabolism is of critical importance, since the nucleoside is a relatively potent inhibitor of cell growth. The enzyme MTA phosphorylase is responsible for the degradation of MTA to adenine and 5-(methylthio)ribose 1-phosphate (MTRP). Adenine is subsequently salvaged to the nucleotide pools, and MTRP is recycled to methionine by a multistep pathway which has not been fully elucidated.[1] Interest in chemotherapeutic strategies involving MTA and its analogues has been stimulated by Toohey's discovery in 1977 that certain malignant cell lines are devoid of MTA phosphorylase activity[2] and by the more recent documentation of MTA phosphorylase deficiency in a significant portion of clinically obtained human leukemias and solid tumors.[3,4]

Numerous analogues of MTA have been synthesized[5,6] and characterized in tumor cell lines containing or lacking

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(1) (a) Backlund, P. S., Jr.; Smith, R. A. *Biochem. Biophys. Res. Commun.* **1982**, *108*, 687. (b) Furfine, E. S.; Abeles, R. H. *J. Biol. Chem.* **1988**, *263*, 9598.
(2) Toohey, J. I. *Biochem. Biophys. Res. Commun.* **1977**, *78*, 1273.
(3) Fitchen, J. H.; Riscoe, M. K.; Dana, B. W.; Lawrence, H. J.; Ferro, A. J. *Cancer Res.* **1986**, *46*, 5409.
(4) Kamatani, N.; Yu, A. L.; Carson, D. A. *Blood* **1982**, *60*, 1387.
(5) Montgomery, J. A.; Shortnacy, A. J.; Thomas, A. T. *J. Med. Chem.* **1974**, *17*, 1197.
(6) Savarese, T. M.; Cannistra, A. J.; Parks, R. E., Jr.; Secrist, J. A., III; Shortnacy, A. T.; Montgomery, J. A. *Biochem. Pharmacol.* **1987**, *36*, 1881.

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