4-Alkyl-5-aryl-4*H*-1,2,4-triazole-3-thiols as Hypoglycemic Agents

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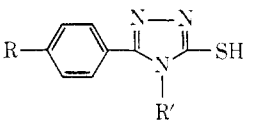
Twenty-one 4-alkyl-5-aryl-4*H*-1,2,4-triazole-3-thiols have been synthesized and screened for hypoglycemic activity in rats. Five compounds showed significant activity of which 5-*p*-chlorophenyl-4-ethyl-4*H*-1,2,4-triazole-3-thiol and 5-*p*-sulfamoylphenyl-4-ethyl-4*H*-1,2,4-triazole-3-thiol were the most active, having a long duration of action and producing blood sugar lowering in normal as well as alloxan-treated rats. They had a very low order of toxicity and showed no antibacterial action.

We described earlier[2,3] the synthesis and diuretic activity of various substituted 3-mercapto-4*H*-1,2,4-triazoles. During the screening of these compounds for possible hypoglycemic effect, some showed significant activity. The most active of these was 5-*p*-chlorophenyl-4-ethyl-4*H*-1,2,4-triazole-3-thiol (I, R = Cl; R′ = C_2H_5). The shifting of the Cl atom from the *para* to the *meta* position or its elimination lowered the hypoglycemic activity, while shifting the Cl atom to the *ortho* position abolished the activity. The work



I

R = CH_3, C_2H_5, Cl, F, NO_2, NH_2, and SO_2NH_2
R′ = CH_3, C_2H_5, C_3H_7, *n*(or *i*)-C_3H_7, *n*(or *i*)-C_4H_9, C_6H_{11}, and $CH_2CH_2OCH_3$

has now been extended further and a number of related 4,5-disubstituted-4*H*-1,2,4-triazole-3-thiols have been synthesized to exploit the lead.

Chemistry.—1-Aroyl-4-alkylthiosemicarbazides were prepared from the requisite benzhydrazides and isothiocyanates and then cyclized with aq NaOH to get the corresponding triazoles. 5-*p*-Aminophenyl-4-ethyl-4*H*-1,2,4-triazole-3-thiol was obtained by the reduction of the corresponding *p*-nitrophenyl analog. *p*-Ethylbenzhydrazide was obtained from ethyl *p*-ethylbenzoate.

Hypoglycemic Activity.—Of the 21 compounds synthesized and screened in normal rats at a dose level of 25 mg/kg, 5 compounds (**21, 22, 29, 30, 31**) showed blood sugar reduction of 30% or more and the blood sugar value returned to normal only after 48 hr. A study of the effect of substitutions in the 4 position revealed that Et and methoxyethyl gave the most active compounds, while *n*-Bu and cyclohexyl were the next best. Further, whereas the 5-*p*-nitrophenyl derivative **38** possessed substantial activity, the corresponding 5-*p*-aminophenyl derivative **39** was practically inactive. Two of the most active compounds in this series, *viz.*, 5-*p*-chlorophenyl-4-ethyl-4*H*-1,2,4-triazole-3-thiol (**21**) and 5-*p*-sulfamoylphenyl-4-ethyl-4*H*-1,2,4-triazole-3-thiol (**30**), were further tested at 10 mg/kg as well as at 50 mg/kg dose levels and produced significant blood sugar lowering. The crossover tests of these compounds with tolbutamide in rats at 25 mg/kg indicated that while the maximum reduction in blood sugar with tolbutamide was 25–27% at the end of the 5th hr, **21** and **30** produced a reduction of up to 45–47% at the 9th hr and even after 24 hr a good hypoglycemic effect was in evidence (Tables I and II). In alloxan-diabetic rats **21** and **30** also produced hypoglycemic responses of 34 and 28%, respectively, at the end of 9 hr and even after 24 hr significant reduction of blood sugar was present (Table III). When tested in rabbits, however, the activity of **21** and **30** was found to be of lower order (13.2 and 15.5%, respectively, at 25 mg/kg dose). These two compounds did not show any

TABLE I

COMPARATIVE HYPOGLYCEMIC ACTIVITY OF **21** AND TOLBUTAMIDE IN RATS[a]

	Percentage reduction in blood sugar		
	Compound **21**	Tolbutamide	Probability
Hr	Mean (±SE)	Mean (±SE)	
1.5	8.06 (±0.80)	12.60 (±0.67)	<0.001
3	16.08 (±1.23)	18.55 (±0.68)	NS
5	26.26 (±1.54)	26.76 (±0.79)	NS
7	34.75 (±2.00)	20.99 (±1.23)	<0.001
9	46.73 (±2.66)	15.37 (±0.94)	<0.001
24	39.01 (±1.75)	3.61 (±0.58)	<0.001

[a] Crossover tests at 25 mg/kg (oral) using 14 animals (7 per group).

TABLE II

COMPARATIVE HYPOGLYCEMIC ACTIVITY OF **30** AND TOLBUTAMIDE IN RATS[a]

	Percentage reduction in blood sugar		
	Compound **30**	Tolbutamide	Probability
Hr	Mean (±SE)	Mean (±SE)	
1.5	10.50 (±1.39)	9.29 (±0.62)	NS
3	16.90 (±1.99)	16.90 (±0.78)	NS
5	28.05 (±3.00)	25.08 (±0.90)	NS
7	36.15 (±2.98)	20.54 (±1.21)	<0.001
9	45.27 (±2.27)	15.16 (±1.14)	<0.001
24	36.21 (±1.88)	3.03 (±0.66)	<0.001

[a] Crossover tests at 25 mg/kg (oral) using 14 animals (7 per group).

TABLE III

BLOOD SUGAR REDUCTION IN ALLOXAN-DIABETIC RATS BY COMPOUNDS **21** AND **30** AT 25 MG/KG (ORAL)

	Percentage blood sugar reduction[a] (±SE)	
Hr	**21**	**30**
1.5	1.6 ± 0.70	6.3 ± 1.20
3	8.1 ± 2.02	17.8 ± 1.17
5	19.0 ± 0.89	20.9 ± 1.39
7	29.9 ± 2.10	24.8 ± 1.06
9	34.0 ± 1.40	28.1 ± 0.39
24	38.3 ± 0.95	23.6 ± 0.73

[a] Figures indicate mean values of 6 rats. Average fasting levels for **21** and **30** were 338 and 259.6 mg %, respectively.

(1) To whom the inquiries should be addressed.

(2) M. H. Shah, V. M. Patki, and M. Y. Mhasalkar, *J. Sci. Ind. Res., Sect. C*, **21**, 76 (1962).

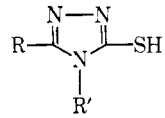
(3) M. H. Shah, M. Y. Mhasalkar, V. M. Patki, and C. V. Deliwala, *J. Pharm. Sci.*, **58**, 1398 (1969).

TABLE IV
1-AROYL-4-ALKYLTHIOSEMICARBAZIDES
RCONHNHCSNHR′

No.	R	R′	Yield, %	Mp, °C	Formula[a]
1	4-ClC_6H_4	CH_3	73	196–197	$C_9H_{10}ClN_3OS$
2	4-ClC_6H_4	$CH_2CH_2OCH_3$	85	186–187	$C_{11}H_{14}ClN_3O_2S$[b]
3	2,4-$Cl_2C_6H_3$	C_2H_5	75	178–179	$C_{10}H_{11}Cl_2N_3OS$[c]
4	4-FC_6H_4	C_2H_5	94	190–191	$C_{10}H_{12}FN_3OS$
5	4-$CH_3C_6H_4$	C_2H_5	91	194–195	$C_{11}H_{15}N_3OS$
6	4-$CH_3C_6H_4$	n-C_3H_7	89	165–166	$C_{12}H_{17}N_3OS$
7	4-$CH_3C_6H_4$	n-C_4H_9	85	158–159	$C_{13}H_{19}N_3OS$
8	4-$CH_3C_6H_4$	C_6H_{11}	87	210–211	$C_{15}H_{21}N_3OS$
9	4-$C_2H_5C_6H_4$	C_2H_5	78	174–175	$C_{12}H_{17}N_3OS$
10	4-$H_2NSO_2C_6H_4$	C_2H_5	92	225–226	$C_{10}H_{14}N_4O_3S_2$
11	4-$H_2NSO_2C_6H_4$	$CH_2CH_2OCH_3$	90	186–187	$C_{11}H_{16}N_4O_4S_2$
12	4-$H_2NSO_2C_6H_4$	$CH_2CH{=}CH_2$	90	211–213	$C_{11}H_{14}N_4O_3S_2$
13	4-$H_2NSO_2C_6H_4$	n-C_3H_7	84	208–209	$C_{11}H_{16}N_4O_3S_2$
14	4-$H_2NSO_2C_6H_4$	i-C_3H_7	89	209–210	$C_{11}H_{16}N_4O_3S_2$
15	4-$H_2NSO_2C_6H_4$	n-C_4H_9	93	200–201	$C_{12}H_{18}N_4O_3S_2$
16	4-$H_2NSO_2C_6H_4$	i-C_4H_9	83	201–203	$C_{12}H_{18}N_4O_3S_2$
17	4-$H_2NSO_2C_6H_4$	C_6H_{11}	88	214–216	$C_{14}H_{20}N_4O_3S$
18	4-$O_2NC_6H_4$	C_2H_5	93	200–201	$C_{10}H_{12}N_4O_3S$[d]

[a] All compounds were analyzed for N, S. [b] Calcd: N, 14.60. Found: 14.14. [c] Calcd: N, 14.38. Found: 14.83. [d] Calcd: N, 20.36. Found: 20.79.

TABLE V
4-ALKYL-5-ARYL-4*H*-1,2,4-TRIAZOLE-3-THIOLS



No.	R	R′	Yield, %	Mp, °C	Formula	Analyses	Maximum blood sugar reduction at a dose of 25 mg/kg % (hr)[a]
19	4-ClC_6H_4	H		286[b]			24.9 (7)
20	4-ClC_6H_4	CH_3	70	212–213	$C_9H_8ClN_3S$	C, H, N, S	Inactive
21	4-ClC_6H_4	C_2H_5		203–204[b]			53.7 (9)[c]
22	4-ClC_6H_4	$CH_2CH_2OCH_3$	78	172–173	$C_{11}H_{12}ClN_3OS$	C, H, N	42.9 (9)[d]
23	2,4-$Cl_2C_6H_3$	C_2H_5	80	191–193	$C_{10}H_9Cl_2N_3S$	C, H, N	19.2 (5)
24	4-FC_6H_4	C_2H_5	84	173–174	$C_{10}H_{10}FN_3S$	C, H, N, S	29.1 (7)
25	4-$CH_3C_6H_4$	C_2H_5	76	182–184	$C_{11}H_{13}N_3S$	C, H, S	21.9 (7)
26	4-$CH_3C_6H_4$	n-C_3H_7	81	155–156	$C_{12}H_{15}N_3S$	C,[e] H, S	Inactive
27	4-$CH_3C_6H_4$	n-C_4H_9	76	172–174	$C_{13}H_{17}N_3S$	C, H, S	19.6 (7)
28	4-$CH_3C_6H_4$	C_6H_{11}	69	189–190	$C_{15}H_{19}N_3S$	C, H, S	28.5 (7)
29	4-$C_2H_5C_6H_4$	C_2H_5	84	161–162	$C_{12}H_{15}N_3S$	C, H, N, S	34.4 (9)[f]
30	4-$H_2NSO_2C_6H_4$	C_2H_5	92	276–278	$C_{10}H_{12}N_4O_2S_2$	C, H, N, S	45.3 (9)[g]
31	4-$H_2NSO_2C_6H_4$	$CH_2CH_2OCH_3$	84	214–216	$C_{11}H_{14}N_4O_3S_2$	C, H, N	39.4 (9)[h]
32	4-$H_2NSO_2C_6H_4$	$CH_2CH{=}CH_2$	92	205–206	$C_{11}H_{12}N_4O_2S_2$	C, H, N, S	Inactive
33	4-$H_2NSO_2C_6H_4$	n-C_3H_7	89	191–192	$C_{11}H_{14}N_4O_2S_2$	C, H, S	Inactive
34	4-$H_2NSO_2C_6H_4$	i-C_3H_7	91	275 dec	$C_{11}H_{14}N_4O_2S_2$	C, H, N, S	Inactive
35	4-$H_2NSO_2C_6H_4$	n-C_4H_9	84	167–168	$C_{12}H_{16}N_4O_2S_2$	C, H, N, S	Inactive
36	4-$H_2NSO_2C_6H_4$	i-C_4H_9	92	220–222	$C_{12}H_{16}N_4O_2S_2$	C, H, S	23.7 (7)
37	4-$H_2NSO_2C_6H_4$	C_6H_{11}	81	233–235	$C_{14}H_{18}N_4O_2S_2$	C, H, N	17.7 (7)
38	4-$O_2NC_6H_4$	C_2H_5	88	228–229	$C_{10}H_{10}N_4O_2S$	C, H, N, S	27.8 (7)
39	4-$H_2NC_6H_4$	C_2H_5	58	246–248	$C_{10}H_{12}N_4S$	C, H, N,[i] S	Inactive

[a] Figures in parenthesis indicate the time in hours for this reduction. [b] *J. Pharm. Sci.*, **58**, 1398 (1969). [c] See Table I. [d] Reduction (26.9%) at 24 hr. [e] *Anal.* Calcd for $C_{12}H_{15}N_3S$: C, 61.80. Found: C, 61.15. [f] Reduction (23.5%) at 24 hr. [g] See Table II. [h] Reduction (27.3%) at 24 hr. [i] *Anal.* Calcd for $C_{10}H_{12}N_4S$: N, 25.45. Found: N, 24.85.

antibacterial action against *Salmonella typhi, Staphylococcus aureus, Vibro comma,* and *Escherichia coli.*

Acute toxicity studies showed that **21** and **30** were much less toxic than tolbutamide. The maximum tolerated single doses (oral) in albino mice for **21** and **30** were 4.0 and 5.0 g/kg, respectively, while LD_{50} of tolbutamide (oral) was 2.6 g/kg. Chronic toxicity experiments were carried out in rats fed orally at a dose level of 100 mg/kg for more than 8 weeks. The animals receiving **21** and **30** maintained good physical condition, showed no significant differences in weight gain as compared with controls, and no macroscopic or microscopic abnormalities were seen in the animals.

Experimental Section

Screening Method.—The hypoglycemic activity was tested in normal fasting albino rats weighing 180–200 g. The drug was

administered orally as a suspension in 2% gum acacia, and blood sugar was determined at 1.5, 3, 5, 7, 9, and 24 hr by Somogyi's method[4] using Nelson's reagent.[5] The crossover tests were carried out with tolbutamide at 25 mg/kg dose level using 7 animals in each groups. One group was given the test drug while the other received tolbutamide. Following a rest period of 1 week, the drugs were crossed over and the test was repeated.

For alloxan-diabetic rats, healthy male albino rats weighing about 200 g were fasted overnight and injected with an aq solution of alloxan monohydrate at 200 mg/kg dose level and food given immediately. Only diabetic animals were used in the test.

Chemical Method.[6] ***p*-Ethylbenzhydrazide.**—Ethyl *p*-ethylbenzoate (44.5 g, 0.25 mol), $N_2H_4 \cdot H_2O$ (25 ml of 98%) and EtOH (100 ml) were mixed and heated under reflux for 2 hr. The residue after removal of EtOH was triturated with Et_2O, filtered, and washed (Et_2O). It was crystd (H_2O), yield 39.1 g (95%), mp 89–90°. *Anal.* ($C_9H_{12}N_2O$) C, H.

(4) M. Somogyi, *J. Biol. Chem.*, **160**, 69 (1945).

(5) N. Nelson, *ibid.*, **153**, 375 (1944).

(6) The melting points were taken in capillary tubes with a partial immersion thermometer and are uncorrected. Where analyses are indicated only by symbols of the elements, the analytical results obtained for these elements were within 0.4% of the theoretical values.

1-*p*-Sulfamoylbenzoyl-4-ethylthiosemicarbazide. *p*-Sulfamoylbenzhydrazide (21.5 g, 0.1 mol) was dissolved in dioxane (150 ml) and heated under reflux with ethyl isothiocyanate (9.5 ml, 0.11 mol) for 4 hr. The solid that sepd on cooling was collected by filtration, washed (H_2O), and crystd (EtOH).

All the thiosemicarbazides required for the present work were similarly prepared and crystd from EtOH. See Table IV for the new thiosemicarbazides.

5-*p*-Sulfamoylphenyl-4-ethyl-4*H*-1,2,4-triazole-3-thiol. 1-*p*-Sulfamoylbenzoyl-4-ethylthiosemicarbazide (6.0 g, 0.02 mol) was dissolved in 2 *N* NaOH (60 ml) and refluxed for 2 hr. The reaction mixture was cooled and acidified with HCl (pH 4). The white precipitate of the desired compound was filtered, washed (H_2O), and crystd (85% EtOH).

All the new triazoles which were prepared similarly and crystd from EtOH are listed in Table V with their melting points, analytical data, etc.

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Saligenin Analogs of Sympathomimetic Catecholamines[1a,b]

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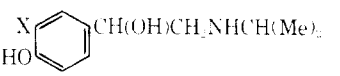
Analogs of isoproterenol have been prepared in which the catechol group has been replaced by salicylic acid (IX) or saligenin (VII) functions. Many of the latter are potent long-lasting β-adrenoreceptor stimulants that are effective orally and show a highly selective action on bronchial smooth muscle. Structure–activity relationships are discussed and related to current theories of molecular processes at β-adrenoreceptors.

Sympathomimetic amines that relax bronchial smooth muscle by stimulation of β-adrenoreceptors have been widely used as bronchodilators in reversible airways obstruction and extensive investigations of structure–activity relations have been made.[2] Maximum potency has always been associated with the presence of a catechol function as in isoproterenol (Ia). The catecholamines have only a short duration of parenteral action probably mainly owing to uptake into tissues[3,4] but also because of metabolism by catechol-*O*-methyl transferase (COMT) to a methyl ether, *e.g.*, Ib.[5] The latter is a β-adrenoreceptor blocker.[6] An additional metabolic barrier may underlie the ineffectiveness of the catecholamines when administered orally since it has been shown in the dog[3] and in man[4] that they are inactivated by conversion into an *O*-sulfate ester in the gut.

We hoped to circumvent these metabolic pathways and hence overcome some of the clinical deficiencies of isoproterenol by the preparation of compounds of formula I where X was a group which retained some of the attributes of the catechol but which would not be subject to attack by the enzymes that inactivate the latter.



Ia, X = HO Ic, X = $MeSO_2NH$ Ie, X = $HOCH_2$
Ib, X = MeO Id, X = HOOC

Previous replacement of one or both phenolic groups[7] by other substituents has drastically reduced sympathomimetic action except in the case of compounds such as Ic where the bioisosteric methanesulfonamide group produces a pseudocatechol.[8] The enhanced acidity of the *m*-phenolic group in the catecholamines, which is simulated by the methanesulfonamide group, is considered important for high biological activity.[8] An additional feature of possible importance is the ability of the catechol moiety to chelate with metals.[9] Both of these properties are displayed, with the minimum of steric disturbance, by the salicylic acid Id which was our first objective. A general synthetic route to Id, outlined in Scheme I, also leads to the saligenin derivative Ie which should still be capable of chelating

(1) (a) For preliminary communication of this work see D. Hartley, D. Jack, L. H. C. Lunts, and A. C. Ritchie, *Nature (London)*, **219**, 861 (1968). (b) Presented in part at the Fourth Rencontres Internationales de Chimie Thérapeutique, Clermont-Ferrand, 1968. (c) To whom inquiries should be addressed.

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