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(17) A 20% yield of the sulfonic acid **1r** was also obtained.
(18) Demethylation of **3b** was a concurrent process.

Derivatives of Tetrahydro-1,4-benzodiazepines as Potential Antihypertensive Agents

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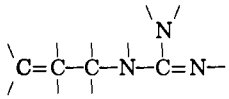
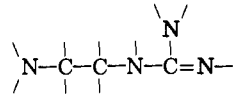
4-Amidino derivatives and quaternary salts of tetrahydro-1,4-benzodiazepines were synthesized and evaluated for antihypertensive activity in conscious rats by the oral route. Included in this study were derivatives of 1,2,4,5,-6,7-hexahydropyrrolo[3,2,1-*jk*][1,4]benzodiazepine and 1,2,3,4,8,9,10,11-octahydro[1,4]diazepino[6,5,4-*jk*]carbazole in which the 1 and 9 positions of tetrahydro-1,4-benzodiazepine are linked by an ethylene and a cyclohexenyl chain, respectively. Four compounds exhibited marked blood pressure lowering activity (>50 mmHg) at doses of 75 mg/kg. Further study indicated that these compounds are effective by impairing transmission in the sympathetic nervous system.

Many currently available clinically effective antihypertensive agents are derivatives of nitrogen heterocycles obtained as a result of careful selection among series of active compounds which were synthesized with systematic structural modification. In each active series, there seems to exist a common structural feature, the presence of which is essential for the activity.

Recently, Schier and Marxer postulated that for the aralkylguanidine type of antihypertensive agents, the partial structure I or II is a requisite feature for lowering blood pressure.[1,2]



I



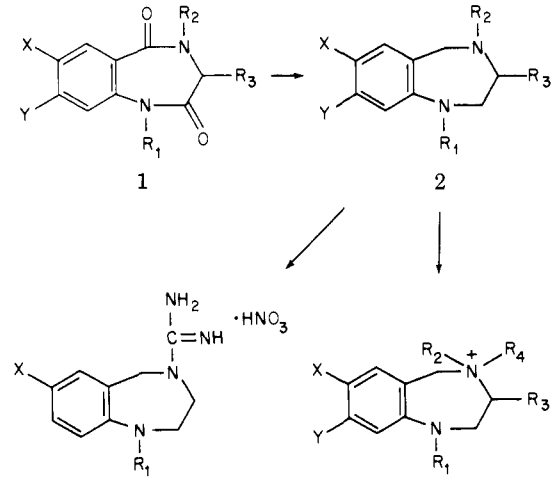
II

This proposition led us to examine amidino derivatives of tetrahydro-1,4-benzodiazepine as potential antihypertensive agents, for those compounds would satisfy simultaneously the above two essential structural requirements. Furthermore, since 1,4-benzodiazepines have a number of pharmacological actions on the central nervous system,[3] agents obtained from these basic structures are of interest as potential "centrally" acting antihypertensive agents.[4]

Also examined in this study were quaternary salts of the tetrahydro-1,4-benzodiazepines and related compounds. Many quaternary ammonium salts are known to possess hypotensive properties. Although most of them are known to be effective by blockade of ganglia, recently it has been shown that some quaternary salts lower blood pressure by other mechanisms, as demonstrated by bretylium.[5]

1,2,3,5-Tetrahydro-4*H*-1,4-benzodiazepine-4-acetamidoxime (**12**) was synthesized and evaluated for antihypertensive effects, since some amidoximes were reported to have blood pressure lowering activity when tested in hypertensive dogs.[6]

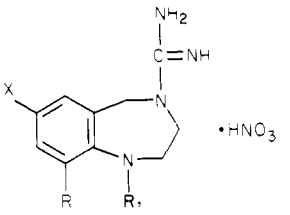
Scheme I



1,2a, R_1 = H; R_2 = H; R_3 = H; X = H; Y = H
1,2b, R_1 = Me; R_2 = H; R_3 = H; X = H; Y = H
1,2c, R_1 = H; R_2 = H; R_3 = Me; X = H; Y = H
1,2d, R_1 = H; R_2 = H; R_3 = H; X = Cl; Y = H
1,2e, R_1 = H; R_2 = H; R_3 = H; X = OMe; Y = OMe

Chemistry. Tetrahydro-1,4-benzodiazepines **2a–e** were obtained by a standard lithium aluminum hydride reduction of 1,4-benzodiazepine-2,4-diones **1a–d** which were prepared from appropriately substituted isatoic anhydrides and α-amino acids by a literature method.[7] In the case of **1e**, a synthetic route which is different from the above one was used and is shown in Scheme II. *N*-Amidino derivatives (see Table I) were prepared by fusion of the tetrahydro-1,4-benzodiazepines with 3,5-dimethylpyrazole-1-carboxamidine nitrate and were isolated as nitrate salts (Scheme I). Quaternary salts shown in Table II were obtained by the treatment of the cyclic amines **2a–e** with

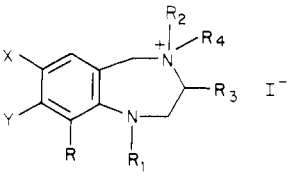
Table I. Amidino Derivatives of Tetrahydro-1,4-benzodiazepines and Related Compounds



Compd	R	R_1	X	Mp, °C	Recrystn solvent	Yield, %	Formula	Analyses[a]
3a	H	H	H	235-237	Water	57	$C_{10}H_{15}N_5O_3$	C, H, N
3b	H	H	Cl	172 dec	Water	52	$C_{10}H_{14}ClN_5O_3$	C, H, N
3c	H	Me	H	210-213	Water	49	$C_{11}H_{17}N_5O_3$	C, H, N
3d	CH_2—CH_2		H	270 dec	Water	13	$C_{12}H_{17}N_5O_3$	C, H, N
3e			H	240-246	Water	14	$C_{16}H_{21}N_5O_3$	C, H, N

[a] Analytical results for these elements were within ±0.4% of the theoretical values.

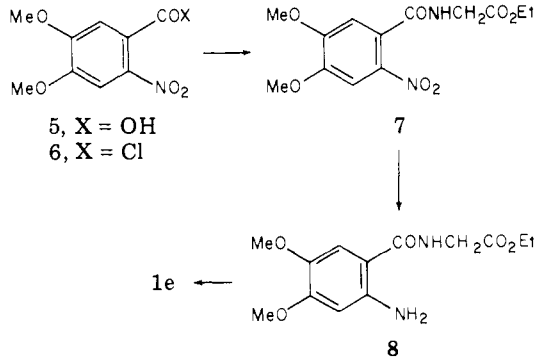
Table II. Quaternary Salts of Tetrahydro-1,4-benzodiazepines and Related Compounds



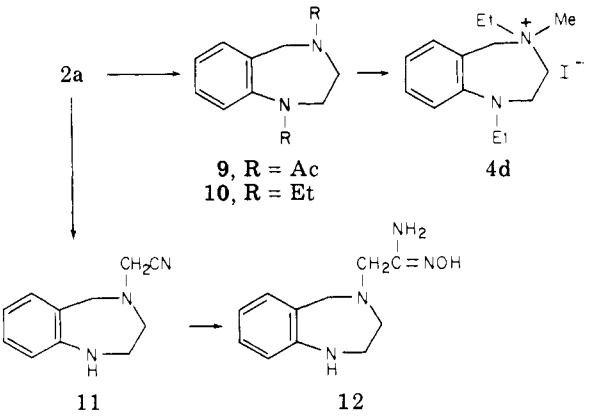
Compd	R	R_1	R_2	R_3	R_4	X	Y	Mp, °C	Recrystn solvent	Yield, %	Formula	Analyses[a]
4a	H	H	Me	H	Me	H	H	196-198	EtOH	32	$C_{11}H_{17}IN_2$	C, H, N
4b	H	Me	Me	H	Me	H	H	155-156	EtOH	43[b]	$C_{12}H_{19}IN_2$	C, H, N
4c	H	H	Me	Me	Me	H	H	226-227	Water	22	$C_{12}H_{19}IN_2$	C, H, N
4d	H	Et	Me	H	Et	H	H	174-176	EtOH	81	$C_{14}H_{23}IN_2$	C, H, N
4e	H	H	Me	H	Me	Cl	H	245-248	EtOH	25	$C_{11}H_{16}ClIN_2$	C, H, N
4f	H	H	Me	H	Me	OMe	OMe	228-230	EtOH	80[b]	$C_{13}H_{21}IN_2O_2 \cdot 0.5H_2O$	C, H, N
4g	CH_2—CH_2		Me	H	Me	H	H	206-208	EtOH	30	$C_{13}H_{19}IN_2$	C, H, N
4h			Me	H	Me	H	H	249-251	EtOH	94	$C_{17}H_{23}IN_3$	C, H, N

[a] Analytical results for these elements were within ±0.4% of the theoretical values. [b] A yield of crude product.

Scheme II



Scheme III



methyl iodide in ether at room temperature (Scheme I). Acetylation of **2a** with acetic anhydride and subsequent reduction with lithium aluminum hydride afforded **10** which on treatment with methyl iodide gave **4d** (Scheme III). Acetamidoxime **12** was obtained by allowing hydroxylamine to react with **11** which was prepared by the reaction of **2a** with chloroacetonitrile (Scheme III). Finally, the work was extended to include derivatives (**3d** and **4g**) of 1,2,4,5,6,7-hexahydropyrrolo[3,2,1-*jk*][1,4]benzodiazepine (**13**) and derivatives (**3e** and **4h**) of 1,2,3,4,8,9,10,11-octahydro[1,4]diazepino[6,5,4-*jk*]carbazole (**14**) in which the 1 and 9 positions of **2a** are linked by an ethylene and a cyclohexenyl chain, respectively. The amine **13** was prepared by the method described by Hester et al.[8] and the synthesis of compound **14** will be reported elsewhere.[9]

Pharmacology and Discussion. The compounds were screened for oral hypotensive activity in conscious female Sprague–Dawley rats which were made hypertensive by the application of a "figure-8" ligature to the left kidney and contralateral nephrectomy 1 week later.[10] Systolic

Table III. Results of Antihypertensive Tests

Compd	Dose, mg/kg	No. of rats	Blood pressure,[a] mmHg	
			Control	Change
3a	75	8	159	-55
3b	75	4	184	-34
3c	75	4	166	-60
3d	75	8	183	-12
3e	75	4	167	-8
4a	75	4	176	-46
4b	75	8	178	-51
4c	75	8	183	-4
4d	75	4	169	-21
4e	75	8	183	-4
4f	75	4	165	+2
4g	75	8	180	-48
4h	75	4	153	+3
12	75	4	171	-12

[a] Data represent group mean.

blood pressure was recorded from the animal's tail by means of an occluding and a sensing cuff and an appropriate pressure transducer.[10] Compounds were administered via a stomach tube and pressures were recorded prior to and 1.5 and 4 h after dosing.

In the amidino series, compounds **3a** and **3c** were shown to be the most potent, eliciting marked reduction (>50 mmHg) in blood pressure at doses of 75 mg/kg. The activity was diminished by the introduction of a chloro group at the 7 position and abolished by linking the diazepine ring to the benzene nucleus as shown by **3d** and **3e** (see Table III).

In the series of quaternary salts, **4a** caused a moderate decrease in blood pressure. The activity was enhanced when a methyl group was introduced at the 1 position of **4a**, i.e., **4b** which was found to be the most potent compound in the series. However, introduction of a substituent at a site other than the 1 position caused loss of the activity. Interestingly, **4g** was active, but **4h** was devoid of activity.

Further studies were performed with the four most potent compounds, **3a**, **3c**, **4a**, and **4g**, in order to obtain an indication of possible mechanisms of the hypotensive effect. Interactions with the sympathetic system were evaluated in pithed rats. Blood pressure responses to sympathetic outflow activitation at three frequencies and to iv injection of phenylephrine were obtained before and after administration.

Activation of sympathetic outflow and the iv injection of phenylephrine resulted in frequency or dose-related increases in blood pressure. The four compounds tested inhibited pressor responses to sympathetic stimulation and had little effect on enhanced responses to phenylephrine (Table IV). The adrenergic neuron blocking agent, guanethidine, exerted similar actions. These agents, therefore, appear to lower blood pressure by impairing transmission in the sympathetic nervous system.

Experimental Section

Melting points were obtained on a Thomas-Hoover melting point apparatus and are uncorrected. The IR spectra were obtained with a Perkin-Elmer Model 21 spectrophotomer. The NMR spectra were determined with a Varian A-60 spectrometer using Me_4Si as the internal reference. The elemental analyses were obtained with a Perkin-Elmer Model 240 elemental analyzer. The analytical results when indicated by C, H, and N agree with the theoretical values within ±0.4%.

6-Nitroveratryl Chloride (6). 6-Nitroveratric acid (**5**)[11] (45 g) was added in small portions to thionyl chloride (120 ml) with stirring, and the resulting mixture was heated under reflux for 3 h. Chilling of the reaction mixture in ice caused separation of a precipitate which was collected on a filter and washed with anhydrous ether. From the mother liquor was obtained another crop of the product. The product amounted to 43.6 g (81%) and melted at 122–124 °C. Anal. ($C_9H_8ClNO_5$) C, H, N.

4,5-Dimethoxy-2-nitrohippuric Acid Ethyl Ester (7). A mixture of **6** (5.0 g, 0.021 mol), $H_2NCH_2CO_2Et$·HCl (3.0 g, 0.0215 mol), and benzene (100 ml) was heated under reflux with introduction of slow stream of nitrogen gas for 5 h. The reaction mixture was then allowed to set at room temperature overnight. A precipitate was collected on a filter and washed with ether several times and then recrystallized from EtOH, giving 5.2 g (82%) of product: mp 148–150 °C; IR (KBr) 3.07 and 5.68 μ. Anal. ($C_{13}H_{16}N_2O_7$) C, H, N.

2-Amino-4,5-dimethoxyhippuric Acid Ethyl Ester (8). Compound **7** (3.1 g, 0.01 mol) was reduced by catalytic hydrogenation in the presence of Pd/C (5%) (1 g) in 250 ml of EtOH. After removal of the catalyst by filtration, ethanol was evaporated under reduced pressure on a rotary evaporator to give an oil which solidified on standing. The solid product was then recrystallized from EtOH, giving 1.3 g (46%): mp 109–110 °C; IR (KBr) 2.87, 2.95, 5.74 μ. Anal. ($C_{13}H_{18}N_2O_5$) C, H, N.

3,4-Dihydro-7,8-dimethoxy-1*H*-1,4-benzodiazepine-2,5-dione (1e). A mixture of **8** (5.6 g, 0.0198 mol), pyridine hydrochloride (2.5 g, 0.0216 mol), and pyridine (50 ml) was heated under reflux for 6 h. A precipitate started to separate in ~4 h. The reaction mixture was chilled in ice, and the precipitate was collected on a filter and washed with EtOH and then with water several times. The product amounted to 4.6 g (98.5%) and melted at 286–288 °C: IR (KBr) 6.00 μ (broad). Anal. ($C_{11}H_{12}N_2O_4$) C, H, N.

2,3,4,5-Tetrahydro-7,8-dimethoxy-1*H*-1,4-benzodiazepine (2e). Compound **1e** (40 g, 0.17 mol) was added in small portions to a slurry of $LiAlH_4$ (20 g, 0.54 mol) in THF (250 ml) at a rate which causes mild reflux. The resulting mixture was refluxed with stirring for 7 h and then allowed to set at room temperature overnight. The mixture was then treated with 20 ml of water,

Table IV. Effects on Systemic Pressor Responses[a] to Activation of Sympathetic Outflow and to Iv Phenylephrine in Pithed Rats

Compd	Dose, mg/kg	Percent change in response				
		Sympathetic activation			Phenylephrine (mg/kg)	
		2.5 Hz	5 Hz	10 Hz	2	4
3a	20	-39	-50	-58	+8	+5
	40	-82	-100	-86	+3	+10
3c	10	-73	-67	-57	-14	-5
	20	-83	-83	-82	-26	-20
4a	10	+10	+9	+1	+23	+39
	30	-38	-34	-35	+44	+74
4g	5	-42	-40	-47	+68	+18
	15	-93	-87	-91	+87	+12
Guanethidine	1	-85	-72	-65	+64	+57
	5	-100	-100	-94	+160	+105
Range of control response		24-69	35-90	52-116	16-45	29-67

[a] Data represent the mean of changes observed in two rats.

20 ml of 15% aqueous NaOH solution, and then with 60 ml of water. The white slurry thus formed was filtered, and the filter cake was washed with hot THF several times. The filtrate and washings were combined and chilled in a freezer overnight. The crystalline product (29.8 g, 84%) thus deposited was collected on a filter and recrystallized from ether: mp 101–104 °C; IR (KBr) 3.06 μ and no absorption in the carbonyl region; NMR ($CDCl_3$) δ 3.06 (s, 4 H, CH_2CH_2), 3.84 (s, 8 H, 2-OMe and $PhCH_2$). Anal. ($C_{11}H_{16}N_2O_2$) C, H, N.

2,3,4,5-Tetrahydro-1*H*-1,4-benzodiazepine (2a) was obtained in a 97% yield (15.5 g) from 3*H*-1,4-benzodiazepine-2,5-dione (19.1 g, 0.108 mol) by reduction with $LiAlH_4$ (11.3 g, 0.25 mol) following the above procedure. The analytical sample which was recrystallized from ether melted at 95–97 °C. Anal. ($C_9H_{12}N_2$) C, H, N.

1-Methyl-2,3,4,5-tetrahydro-1*H*-1,4-benzodiazepine (2b) was similarly prepared by reduction of **1b** (10.5 g, 0.055 mol) with $LiAlH_4$ (6.3 g, 0.17 mol) in THF. The product (9.0 g, 100%) thus obtained as an oil was used directly for the preparation of **4b**: IR (film) 3.06, 3.50 μ.

Similarly, **3-methyl-2,3,4,5-tetrahydro-1*H*-1,4-benzodiazepine (2c)** was prepared from **1c** (23 g, 0.121 mol) and $LiAlH_4$ (14 g, 0.369 mol) and obtained as an oil which was used without purification in the subsequent reaction: IR (film) 3.10 μ.

7-Chloro-2,3,4,5-tetrahydro-1*H*-1,4-benzodiazepine (2d). 3,4-Dihydro-7-chloro-1*H*-1,4-benzodiazepine-2,5-dione (**1d**) (31.4 g, 0.149 mol) was reduced with $LiAlH_4$ (17.1 g, 0.45 mol) in similar fashion, giving 25.5 g (94%) of the product. The analytical sample was obtained by recrystallization from ether: mp 103–105 °C. Anal. ($C_9H_{11}ClN_2$) C, H, N.

1,4-Diacetyl-2,3,4,5-tetrahydro-1*H*-1,4-benzodiazepine (9). To a stirring mixture of **2a** (18 g, 0.122 mol) and pyridine (70 ml) was added dropwise acetic anhydride (22 g, 0.216 mol) at room temperature. The resulting mixture was heated on a steam bath for 1 h and then evaporated on a rotary evaporator under reduced pressure to give an oil. The oil solidified on standing. Recrystallization from ether with addition of a small amount of EtOH gave 28 g (99%) of the product: mp 119–120 °C. Anal. ($C_{13}H_{16}N_2O_2$) C, H, N.

1,4-Diethyl-2,3,4,5-tetrahydro-1*H*-1,4-benzodiazepine (10). Compound **9** (12 g, 0.0517 mol) was reduced with $LiAlH_4$ (5.7 g, 0.15 mol) in THF (200 ml) heating under reflux for 5 h and then worked up by a standard procedure, giving 9.2 g (87%) of **10** as an oil. The oil was used directly in the next reaction: IR (film) 3.43 and 3.57 μ.

1,2,3,5-Tetrahydro-4*H*-1,4-benzodiazepine-4-carboxamidine Nitrate (3a). The following procedure exemplifies the preparations of **3a–e**. A well-blended mixture of **2a** (4.4 g, 0.0298 mol) and 1-amidino-3,5-dimethylpyrazole nitrate (6.0 g, 0.0298 mol) in a 50-ml round-bottom flask was immersed in an oil bath maintaining the temperature of 190–200 °C for 20 min. Sublimation of 3,5-dimethylpyrazole was observed as the reaction was taking place. The 3,5-dimethylpyrazole which condensed on the neck of the reaction flask was removed mechanically. The solid residue was triturated with ether and then recrystallized from a small amount of water with charcoal treatment, giving 3.65 g (57%) product: mp 232–235 °C. Another recrystallization from water afforded an analytical sample: mp 235–237 °C; IR (KBr) 3.02, 3.15, 6.00 μ; NMR (Me_2SO-d_6) δ 3.27 and 3.65 (crude s, NCH_2CH_2N), 4.55 (s, $PhCH_2$).

2,3,4,5-Tetrahydro-4,4-dimethyl-1*H*-1,4-benzodiazepinium Iodide (4a). The following procedure used for the preparation of **4a** represents preparations of **4a–h**. To an ether solution of **2a** (4.5 g, 0.026 mol) was added dropwise methyl iodide (4.5 g, 0.031 mol) dissolved in 10 ml of ether. The resulting mixture was stirred at room temperature for 5 h. A precipitate was collected on a filter, washed with ether and then ethanol, and recrystallized from ethanol, giving 2.5 g (32%) of **4a**: mp 196–198 °C; IR (KBr) 3.02, 3.15 μ; NMR (Me_2SO-d_6) δ 3.24 (s, 6 H, 2-Me), 3.50 (crude s, 4 H, NCH_2CH_2N) 4.65 (s, 2 H, $PhCH_2$).

1,2,3,5-Tetrahydro-4*H*-1,4-benzodiazepine-4-acetonitrile (11). To the stirring mixture of **2a** (14.8 g, 0.10 mol), sodium bicarbonate (8.4 g, 0.10 mol), and ethanol (150 ml) was added chloroacetonitrile (7.6 g, 0.10 mol) dropwise at room temperature. The reaction mixture was heated under reflux for 5 h. After the reaction mixture was cooled, it was filtered to remove an inorganic salt. The filtrate was evaporated on a rotary evaporator under reduced pressure to give an oil which solidified on standing. The solid residue was recrystallized from ether with charcoal treatment to give 18 g (94%) of 11: mp 87–89 °C; IR (KBr) 4.49 μ (weak). Anal. ($C_{11}H_{13}N_3$) C, H, N.

1,2,3,5-Tetrahydro-4*H*-1,4-benzodiazepine-4-acetamidoxime (12). To a stirring mixture of 11 (3.74 g, 0.0195 mol), hydroxylamine hydrochloride (1.4 g, 0.020 mol), and absolute EtOH (50 ml) was added a freshly prepared sodium ethoxide solution which was prepared by dissolving 0.4 g (0.0174 mol) of sodium in 25 ml of absolute EtOH. The resulting mixture was heated under reflux for 3 h and then set at room temperature overnight. The inorganic salt which deposited was removed by filtration. Nitric acid (70%, 1.8 g) was added dropwise to the filtrate. Addition of ether to the neutralized filtrate caused separation of a resinous material which solidified on chilling and scratching. The product was collected on a filter and washed with ether, giving 4.8 g (87%) of 12: mp 154–156 °C dec. Recrystallization from water with charcoal treatment improved the melting point to 159–161 °C dec: IR (KBr) 3.00, 3.35, 3.70, 3.73, 5.98 μ. Anal. ($C_{11}H_{17}N_5O_4$) C, H, N.

Pharmacology. Antihypertensive screening was performed by the method described by Baum et al.[10]

Interactions with the Sympathetic Nervous System. Rats were anesthetized with ether and placed on artifical respiration. The blood pressure was recorded from the carotid artery and drugs were injected into the jugular vein. A stainless steel pithing rod was introduced into the spinal cord through the orbit. The total sympathetic outflow was activated by using the pithing rod as a cathode and a hypodermic needle in the femoral region as the anode. Maximal stimuli (approximately 35 V) of 1-ms duration and frequency of 2.5, 5, and 10 Hz were applied for 5 s causing an elevation of systemic blood pressure. Phenylephrine was also injected iv. Animals were pretreated with 1 mg/kg of atropine and 1 mg/kg of *d*-tubocurarine chloride. Blood pressure responses to sympathetic activation and to injected amines were obtained prior to and after one or more iv doses of test compounds.

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