1600 (C=C), 1220 (P=O) cm^{-1}; NMR ($CDCl_3/Me_2SO$-d_6) δ 3.35-3.90 (m, 16 H, 8 CH_2), 7.42-8.10 (m, 6 H, arom); MS, m/z (relative intensity) 602 (M^+, 49). Anal. ($C_{18}H_{22}Cl_6N_2O_4P_2$) C, H, N.

1,5-Bis[*N*,*N*-bis(2-chloroethyl)phosphorodiamidic acid ester] of 1,5-Dihydroxynaphthalene (4b). Ammonia was bubbled into a benzene solution (100 mL) of **4a** (1.8 g, 3 mmol) for 30 min, the suspension was stirred for 30 min and filtered, and the filtrate was evaporated in vacuo. The residue was extracted with hot acetone and filtered. Upon cooling, the filtrate formed the product (0.5 g, 34%) as a white, crystalline material: mp 174-175 °C; IR 3120, 3220 and 3310 [P(O)NH_2], 1600 (C=C), 1200 (P=O), 970 (P=N); NMR (Me_2SO-d_6) δ 3.10-3.82 (m, 16 H, 8 CH_2), 4.90 (d, 4 H, 2 NH_2), 7.31-8.01 (m, 6 H, arom). Anal. ($C_{18}H_{26}Cl_4N_4O_4P_2$) C, H, N.

X-ray Diffraction. Crystals of (±)-**3a** formed as colorless blocks from ether. The cell constants [a = 26.366 (13), b = 8.826 (7), c = 19.224 (10) Å; β = 96.67 (8)°; vol = 4443.3 Å^3] were determined at 138 (2) K. The space group was found to be $C_{2/c}$ with Z = 8. From the 4556 unique data, 3969 were judged observed [$F > 4\sigma(F)$]. The structure was solved by MULTAN[21] and refined by SHELX.[22] The final conventional R and weighted R were 0.035 and 0.047 for the observed data. A final difference electron density map showed no peaks greater than 0.31 e$^-$/Å^3.

Crystals of (±)-**3b** grew as colorless rods from ether with $P2_{1/a}$ (Z = 4) symmetry. The cell constants [a = 6.939 (3), b = 26.747 (13), c = 11.828 (6) Å; β = 102.91 (4)°; vol = 2139.8 Å^3] were determined at 138 (2) K. A total of 3514 data were judged observed [$F > 4\sigma(F)$] from the 4392 unique data. The structure was solved by the SHELX direct methods package and refined using SHELX.[22] The final conventional R and weighted R were 0.040 and 0.047 for the observed data. A final difference electron density map had no peaks larger than 0.39 e$^-$/Å^3.

Data for both compounds were collected at 138 (2) K on an Enraf-Nonius CAD-4 diffractometer using Mo $K\alpha$ radiation (λ = 0.71069Å). For both compounds, intensity data were collected using the θ-2θ scan method with $2\theta \leq 53°$. Lattice constants were determined from a least-squares fit of the $\pm 2\theta$ values of 48 intensity maxima taken from all regions of reciprocal space. The function minimized in the structure refinements was $\sum\omega(|F_o|-|F_c|)^2$ with $\omega = 1/\sigma^2(F)$. Tables of fractional coordinates, thermal parameters, bond distances and bond angles for **3a** and **3b** are given in the supplementary material.

Acknowledgment. This research was supported in part by grants from the National Institutes of Health, National Cancer Institute (Grant CA 24970) and the Robert A. Welch Foundation [Grant E-920 (to L.A.C.) and Grant CA 17562 (to D.v.d.H.)]. The generous supply of propranolol hydrochloride from Mead Johnson Co. is greatly appreciated.

Registry No. **1a**, 88181-16-2; **1b**, 88181-17-3; **2a**, 88181-18-4; **2b**, 88181-19-5; (±)-**3a**, 88181-20-8; (±)-**3b**, 88181-21-9; **4a**, 88181-22-0; **4b**, 88181-23-1; DES, 56-53-1; 9-hydroxypsoralen, 2009-24-7; propanolol, 525-66-6; (±)-propanolol hydrochloride, 3506-09-0; 1,5-dihydroxynaphthalene, 83-56-7; phosphoramide mustard, 10159-53-2.

Supplementary Material Available: Tables of fractional coordinates, thermal parameters, bond distances and bond angles for **3a** and **3b** (8 pages). Ordering information is given on any current masthead page.

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Notes

(±)-4-Aryl-4,5-dihydro-3*H*-1,3-benzodiazepines. 3. 2-Phenyl and 2-Amino Analogues as Potential Antihypertensive Agents[1]

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A series of 2-phenyl- and 2-amino-4-aryl-4,5-dihydro-3*H*-1,3-benzodiazepines was prepared and submitted for broad biological screening, including evaluation for potential antihypertensive activity. Compound **4a** [(±)-4,5-dihydro-2,4-diphenyl-3-methyl-3*H*-1,3-benzodiazepine hydrochloride] was the most active member of the series in the spontaneously hypertensive rat (SHR) model, producing a 56 mmHg decrease in systolic blood pressure at an oral screening dose of 50 mg/kg. The synthesis of **4a** analogues containing nuclear substituents in the 4-phenyl moiety resulted in a marked decrease of antihypertensive activity. It was not possible to improve on the antihypertensive properties of **4a** through further synthetic modifications.

We previously reported the synthesis and biological evaluation of many 2-alkyl and several 2-aryl analogues of 4-aryl-4,5-dihydro-3*H*-1,3-benzodiazepines as potential psychotropic agents.[2,3] While optimum antidepressant-like properties were associated with a small 2-alkyl substituent, the 2-aryl series was virtually inactive in assays reflecting antidepressant-like activity. However, in broad

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(1) This paper has been presented in part; see: "Abstracts of Papers"; International Symposium on Medicinal Chemistry, 7th, Costa del Sol, Torremolinos (Malaga) Spain, Sept 2-5, 1980; Cotswold Press Ltd.: Oxford; Abstr P 178.
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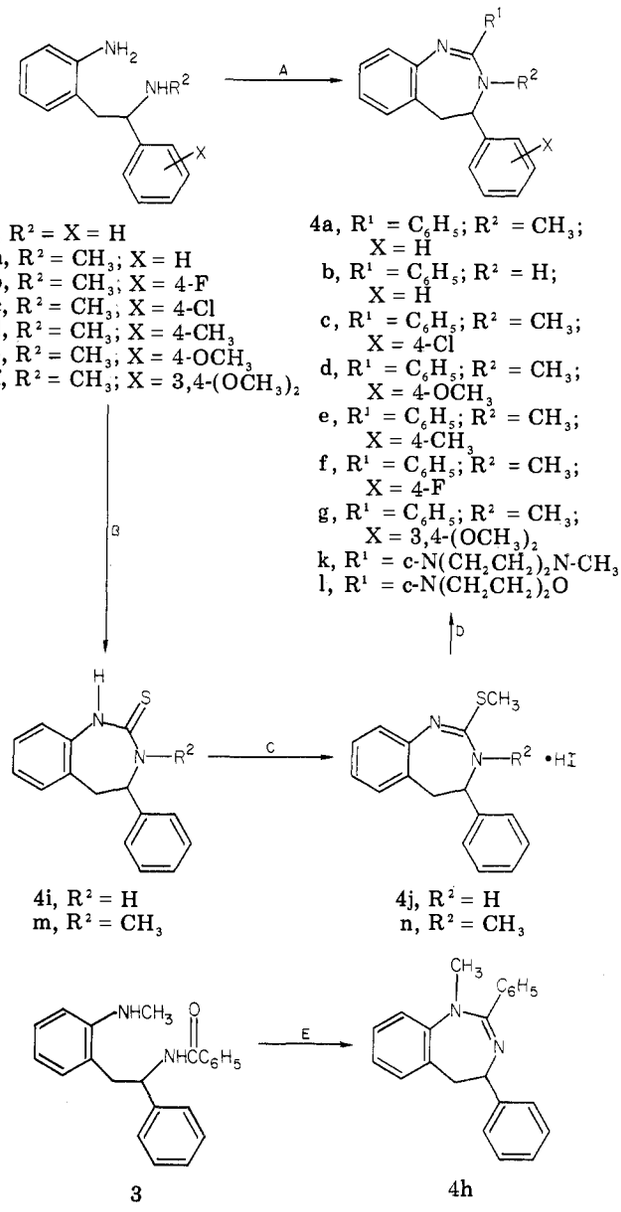
0022-2623/84/1827-0401$01.50/0

Scheme I[a]



[a] A = $R^1C(OR)_3$; B = CS_2; C = CH_3I; D = CH_3-c-$N(CH_2CH_2)_2NH$ (4k), c-$NH(CH_2CH_2)_2O$ (4l); E = $SOCl_2$.

screening, (±)-4,5-dihydro-2,4-diphenyl-3-methyl-3*H*-1,3-benzodiazepine hydrochloride (**4a**) was found active in the spontaneously hypertensive rat (SHR) model. We thus embarked on modification of this lead compound in an effort to maximize potential antihypertensive activity. Additionally, we thought that guanidine-type derivatives, that is, compounds having a 2-amino functionality, might display antihypertensive activity, since other guanidine derivatives, such as guanethidine and clonidine, are clinically useful antihypertensives. Thus, a limited series of 2-amino derivatives was also prepared.

Chemistry. The synthesis of 1,3-benzodiazepines **4a–n** is illustrated in Scheme I, and their properties are reported in Table I. R^1 phenyl analogues **4a–g** were prepared as previously described by cyclization of diamines **1** and **2** with the appropriate ortho esters under acid catalysis.[2,3] Compound **4h** was previously synthesized by cyclic dehydration of **3** with thionyl chloride.[3] The synthesis of R^1 amino derivatives involved the cyclization of **1** or **2a** with carbon disulfide to afford the thiones **4i** and **4m**, which were alkylated with iodomethane to give methylthio intermediates **4j** and **4n**, respectively. Displacement of methyl mercaptan from **4j** with 1-methylpiperazine and morpholine gave **4k** and **4l**, respectively. Attempts to prepare analogues of **4k**,l, where R^2 is methyl, by similar displacement of methyl mercaptan from **4n** were unsuccessful.

Results and Discussion

Potential antihypertensive activity was evaluated in the spontaneously hypertensive rat (SHR) model by the indirect tail-cuff method at an oral screening dose of 50 mg/kg. The results of this testing are summarized in Table I.

Maximum antihypertensive activity was observed with **4a** ($R^1 = C_6H_5$; $R^2 = CH_3$). When R^2 was hydrogen (**4b**), the antihypertensive activity was significantly diminished. The synthesis of nuclear-substituted analogues of **4a** was based initially upon the Topliss[4] operational scheme for analogue synthesis of a lead compound. These analogues, **4c–g**, also displayed significantly reduced activity in comparison with **4a**. As suggested by Topliss,[4] the reduced activity of the 4-Cl (**4c**) and 4-OCH_3 (**4d**) analogues in comparison with the unsubstituted derivative (**4a**) indicates unfavorable prospects for para substitution, and this suggestion is supported by the low activity of **4e–g**. Compound **4h** ($R^1 = C_6H_5$; $R^2 = CH_3$), which is isomeric with **4a**, did not display significant antihypertensive activity. Analogues **4k**,l, which contain R^1 amino substituents, also did not display significant activity.

In summary, the most interesting member of the group is **4a**, and oral doses of 5–50 mg/kg bring about a lowering of blood pressure in the SHR. It was not possible to improve on the properties of this compound through a limited program of molecular modification.

Experimental Section

The structures of all compounds are supported by their IR (Perkin-Elmer 457) and ^1H NMR (JEOL C60 HL; tetramethylsilane) spectra. Melting points were taken on a Thomas-Hoover capillary melting point apparatus and are uncorrected. Elemental analyses were performed by Micro Tech Laboratories, Skokie, IL. Results are within ±0.4% of theoretical values unless otherwise noted in the table. Reactions with moisture-sensitive reagents were maintained under a dry nitrogen atmosphere. Solvents dried over molecular sieves were employed for reactions requiring anhydrous solvents.

(±)-2-Phenyl-4,5-dihydro-4-aryl-1,3-benzodiazepines (4a–h). Compounds **4a–g** were prepared as shown in Scheme I from **1** and **2a–f** according to the procedures previously described for **4a**.[2,3] The synthesis of **4h** was previously reported by cyclic dehydration of **3** [(±)-*N*-benzoyl-2-(methylamino)-α-phenylbenzeneethanamine] with thionyl chloride.[2] Properties of **4a–h** are included in Table I.

(±)-1,3,4,5-Tetrahydro-4-phenyl-2*H*-1,3-benzodiazepine-2-thione (4i). A warm, stirred solution of (±)-2-amino-α-phenylbenzeneethanamine (**1**; 6.5 g, 0.031 mol) and 95% ethanol (15 mL) was treated dropwise with carbon disulfide (2.7 g, 0.035 mol). The mixture was diluted with 95% ethanol (55 mL) and warmed at 60 °C for 1 h. Concentrated hydrochloric acid (0.5 mL) was added, and the mixture was refluxed for 24 h. While the mixture was cooling, the crude product crystallized. Recrystallization from isobutyl alcohol gave **4i** (4.6 g, 58%), mp 210–212 °C. Properties of **4i**, and of **4m** prepared in a similar manner from **2a**, are included in Table I.

(±)-4,5-Dihydro-2-(methylthio)-4-phenyl-3*H*-1,3-benzodiazepine Hydriodide (4j). A warm stirred solution of **4i** (3.8 g, 0.015 mol) and 2-methoxyethanol (50 mL) was treated with a

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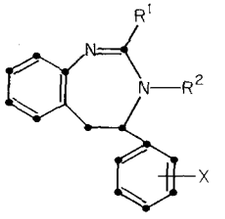
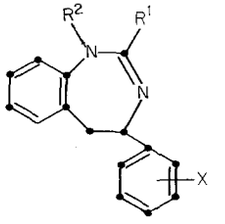
Table I. (±)-4-Aryl-4,5-dihydro-3*H*-1,3-benzodiazepines[a]



4a–g, i–n



4h

compd	R^1	R^2	X	mp,[b] °C	yield,[c] %	recrystn solvent[d]	formula	anal.[e]	base line systolic press., mmHg	SHR[f] BP, mmHg ± SEM, 50 mg/kg po
4a	C_6H_5	CH_3	H	258–259	29	A	$C_{22}H_{20}N_2 \cdot HCl$[g]	C, H, N	189	−56 ± 14
									199	−13 ± 5.1[h]
4b	C_6H_5	H	H	244–247	40	A	$C_{21}H_{18}N_2 \cdot HCl$	C, H, N	183	−15
4c	C_6H_5	CH_3	4-Cl	269–270	51	C-E	$C_{22}H_{20}ClN_2 \cdot HCl$	C, H, N	189	−10
4d	C_6H_5	CH_3	4-OCH_3	247–249	79	A	$C_{23}H_{22}N_2O \cdot HCl$	C, H, N	168	−26 ± 9.2
4e	C_6H_5	CH_3	4-CH_3	268–270	57	C-E	$C_{23}H_{22}N_2 \cdot HCl$	C, H, N	189	−6
4f	C_6H_5	CH_3	4-F	270–272	66	C-E	$C_{22}H_{19}N_2 \cdot HCl$	C, H, N	185	−8
4g	C_6H_5	CH_3	3,4-$(OCH_3)_2$	175–180	42	C-E	$C_{24}H_{24}N_2O_2 \cdot HCl$	C, H, N	207	−24 ± 20.8
4h	C_6H_5	CH_3	H	98–101	33	D	$C_{22}H_{20}N_2$	C, H, N	173	+4
4i	SH	H	H	210–212	58	G	$C_{15}H_{14}N_2S$	C, H, N	202	−40 ± 7.8
									178	−2[i]
4j	SCH_3	H	H	212–216	79	F-E	$C_{16}H_{16}N_2S \cdot HI$	C, H, N, S	185	−25 ± 4
4k	c-$N(CH_2CH_2)_2N$-CH_3	H	H	85–100[j]	63	B	$C_{20}H_{24}N_4$	C, H	200	+2
4l	c-$N(CH_2CH_2)_2O$	H	H	128–129	61	H	$C_{19}H_{21}N_3O$	C, H, N	223	−16
4m	SH	CH_3	H	174–178	52	C	$C_{16}H_{16}N_2S$	C, H, S	NT[k]	
4n	SCH_3	CH_3	H	175–183	92	A-E	$C_{17}H_{18}N_2S \cdot HI$	C, H, N	NT[k]	
guanethidine									174	−20 ± 4.9

[a] All compounds exhibited IR and ^1H NMR spectra consistent with assigned structures. [b] Melting points are uncorrected. [c] Yield of analytically pure material; yields were not optimized. [d] A = acetonitrile; B = cyclohexane; C = absolute ethanol; D = 95% ethanol; E = ether; F = methanol; G = isobutyl alcohol; H = toluene. [e] Analytical results within ±0.4% of theoretical values. [f] Spontaneously hypertensive rat; po = per os; generally, a drop in systolic pressure greater than 30 mmHg is considered significant. [g] Hemihydrate. [h] At 5 mg/kg. [i] At 25 mg/kg. [j] Isolated as crystalline solvate with cyclohexane, mp 93.5–98 °C, which was dried (Abderhalden pistol, benzene) to afford 4k as an amorphorous powder. [k] NT = not tested.

solution of iodomethane (2.4 g, 0.017 mol) and 2-methoxyethanol (10 mL). After the solution was stirred for 1 h with exclusion of light, additional iodomethane (0.5 g) in 2-methoxyethanol (5 mL) was added, and the solution was stirred at ambient temperature overnight. The solution was concentrated under reduced pressure, and the residual oil was triturated with ether to afford a solid. Recrystallization from methanol–ether gave **4j** as colorless crystals. Properties of **4j**, and **4n** prepared in a similar manner from **4m**, are included in Table I.

(±)-4,5-Dihydro-2-(4-methyl-1-piperazinyl)-4-phenyl-3*H*-1,3-benzodiazepine (4k). A mixture of **4j** (4.8 g, 0.012 mol) and 1-methylpiperazine (35 mL) was refluxed with stirring under a nitrogen atomosphere until the evolution of methyl mercaptan ceased. The 1-methylpiperazine hydriodide that crystallized was removed by filtration. The filtrate was diluted with chloroform and washed with 5% NaOH solution. The dried (Na_2SO_4) organic phase was filtered, the filtrate was concentrated, and the crude product was crystallized twice from cyclohexane to afford **4k** as a colorless solid. Properties of **4k** are included in Table I.

(±)-4,5-Dihydro-2-(4-morpholinyl)-4-phenyl-3*H*-1,3-benzodiazepine (4l). This compound was prepared from **4j** (3.0 g, 0.076 mol) and morpholine (30 mL) to afford **4l** in a manner analogous to that described for **4k**. Properties of **4l** are included in Table I.

Pharmacological Method. Spontaneously Hypertensive Rat Assay. All compounds were evaluated for antihypertensive activity in spontaneously hypertensive rats (Okamoto-Aoki strain) at an oral screening dose of 50 mg/kg. Systolic blood pressures were determined by tail-cuff plethysmography predose on day 1 (zero time) and 2 h postdose on day 3. Details of the method are described by Buggy et al.[5] The test compounds were suspended in distilled water with Tween 80 and administered orally at 50 mg/kg unless otherwise indicated. Animals were dosed every day. Four animals per drug were used in the preliminary screen.

Acknowledgment. The authors express their appreciation to Marc N. Agnew and Anastasia Rizwaniuk for spectral data and to Susan Raite and Joan Galbraith for performing pharmacological assays. We also gratefully acknowledge June D. Baird-Strupczewski and Ann Van Dine for library research and Grace M. Naumovitz for assistance in preparation of the manuscript.

Registry No. (±)-1, 80806-39-9; (±)-**2a**, 88057-41-4; (±)-**2b**, 80805-99-8; (±)-**2c**, 88057-42-5; (±)-**2d**, 88057-43-6; (±)-**2e**, 88057-44-7; (±)-**2f**, 80806-00-4; (±)-**4a**, 88057-56-1; (±)-**4a**·HCl, 80806-15-1; (±)-**4b**, 88057-57-2; (±)-**4b**·HCl, 80806-23-1; (±)-**4c**, 88057-58-3; (±)-**4c**·HCl, 88057-45-8; (±)-**4d**, 88057-59-4; (±)-**4d**·HCl, 88057-46-9; (±)-**4e**, 88057-60-7; (±)-**4e**·HCl, 88057-47-0; (±)-**4f**, 88057-61-8; (±)-**4f**·HCl, 88057-48-1; (±)-**4g**, 88057-62-9; (±)-**4g**·HCl, 88057-49-2; (±)-**4h**, 80806-26-4; (±)-**4i**, 88057-50-5; (±)-**4j**·HI, 88057-51-6; (±)-**4k**, 88057-52-7; (±)-**4l**, 88057-53-8; (±)-**4m**, 88057-54-9; (±)-**4n**·HI, 88057-55-0; (±)-**4j**, 88057-63-0; CS_2, 75-15-0; 1-methylpiperazine, 109-01-3; morpholine, 110-91-8.

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A Cyclic Angiotensin Antagonist: [1,8-Cysteine]angiotensin II

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[Cys(*S*-acetamidomethyl)[1,8]]angiotensin II has been synthesized by the solid-phase method and purified by carboxymethylcellulose chromatography and reversed-phase HPLC. Treatment of this peptide with iodine gave the cyclic octapeptide [Cys[1,8]]angiotensin II, which was isolated by Sephadex G-25 chromatography and reversed-phase HPLC. Comparison of the circular dichroism spectra of the open-chain and cyclic peptides, respectively, demonstrated the existence of a disulfide bond with right-handed chirality in the cyclic peptide. In the isolated rat uterus assay, the open-chain peptide was found to be a potent antagonist of angiotensin II, having approximately 10% of the activity of [Sar[1], Ile[8]]angiotensin II, and the cyclic peptide had about 10% of the antagonist potency of its open-chain synthetic precursor.

The design of potent in vivo antagonists of biologically active peptides is desirable for clarification of the physiological functions of hormones and for therapeutic application in certain disease states. Conformational restriction of peptides via cyclization has proved to be a very valuable tool in the design of peptide hormone analogues that have resistance to metabolic degradation.[1-3] Several magnetic resonance studies have suggested a near-cyclic conformation for angiotensin II (ANG II, Asp-Arg-Val-Tyr-Ile-His-Pro-Phe).[4-6] Cyclization of ANG II with water-soluble carbodiimide has resulted in an inactive product,[7] possibly because the C-terminal α-carboxyl group was derivatized in the cyclization step. We have synthesized a cyclic analogue of ANG II in which the C-terminal carboxyl group remains free, by substituting Cys for the N-terminal and C-terminal amino acids of the molecule. These substitutions would be anticipated to produce an angiotensin antagonist, since the presence of a straight or branched side chain at the C terminus of ANG II is known to abolish intrinsic activity without severely influencing receptor binding affinity. In general, substitution of the N-terminal amino acid of ANG II appears to influence only the rate of metabolic degradation of the molecule and has a minor influence on receptor binding.[8]

Results and Discussion

The synthesis of the cyclic peptide [Cys[1,8]]ANG II generally followed expectations, although the yields of intermediate and final products, both from the synthesis

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