justed to pH 7.4 with tris(hydroxymethyl)aminomethane, approximately 23 mM Tris base), 5.5 mM glucose, 0.8 mM $MgSO_4$, and 5.4 mM KCl. The tissue was homogenized with 10–12 strokes of a glass-glass homogenizer. The final volume was adjusted to 10 mL and centrifuged at 1000*g* for 15 min. The pellet was washed once with 10 mL of buffer and resuspended in the same volume for binding studies. Incubations were carried out in a total volume of 250 μL containing 50 nM [^{3}H]BTX-B, 1 μM tetrodotoxin, 0.03 mg of scorpion venom, and about 400 μg of the particulate vesicular protein. Incubations for 30 min at 37 °C were terminated by dilution of the reaction mixture with 3 mL of wash buffer and filtration through a Whatman GF/C filter. Filters were washed three times with 3 mL of wash buffer. Filtration was accomplished with a Millipore filtration apparatus for single samples or with a Brandel Cell Harvester (Gaithersburg, MD) to filter sets of 24 samples. The results with both methods were identical. The wash buffer contained the following: 163 mM choline chloride, 5 mM Hepes (adjusted to pH 7.4 with Tris base), 1.8 mM $CaCl_2$, and 0.8 mM $MgSO_4$. Filters were counted in a Beckman scintillation counter using 10 mL of Hydroflour (National Diagnostics). The efficiency of tritium counting was 43%. Specific binding was determined by substracting the nonspecific binding, determined in the presence of 300 μM veratridine, from the total binding of [^{3}H]BTX-B. Specific binding was about 80% of total binding.

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Registry No. BTX-B, 78870-19-6; Na, 7440-23-5; Ca, 7440-70-2; K, 7440-09-7.

Bicyclic and Tricyclic Analogues of Anthramycin

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As analogues of pyrrolo[2,1-*c*][1,4]benzodiazepine antitumor antibiotics, such as anthramycin and tomaymycin, several benzo[1,4]diazepine imines and carbinolamine ethers were prepared and tested in vivo against P388 leukemia. Two different synthetic approaches, namely, a reduction of an aromatic nitro group with a concomitant cyclization and a reduction of a lactam, were employed to generate an imine or a carbinolamine moiety. Bicyclic analogues **6a′**, **6f**, and **6g** were found to be active, indicating that the pyrroline ring of anthramycin is not an absolute necessity for the antitumor activity. Compound **6g**, 3,4-dihydro-9-hydroxy-4-propargyl-5*H*-1,4-benzodiazepin-5-one, was at least as active as neothramycin although it was 5 times less potent. Among the tricyclic analogues, compounds **5**, **7a**, and **8b** were active against P388 leukemia, and they generally appear to be more potent than bicyclic analogues.

Pyrrolo[2,1-*c*][1,4]benzodiazepine antitumor antibiotics are a unique class of compounds represented by anthramycin (**1**) and tomaymycin (**2**).[1] Neothramycin,[2] one of the newest members of this class, and spadicomycin,[3] an anthramycin sodium hydrosulfite adduct, are currently in clinical trials in Japan. Previously, anthramycin and sibiromycin have been tried clinically with only limited success.[5]

On the molecular level, Hurley and co-workers proposed a possible mechanism of action of these agents.[5] According to this proposal, anthramycin (or other members of this class) fits in the minor groove of DNA, and it is bound by a labile aminal linkage between the N2 of guanine and the C11 of anthramycin. The secondary stabilizing force is provided by the hydrogen bonding of the C9 hydroxy group of anthramycin to the O2 of cytosine. The hydroxy group of tomaymycin, on the other hand, is presumed to be involved in bifurcated hydrogen bonding to the sugar and phosphate oxygens of DNA.[6] In 1979 Lown and Joshua prepared compounds **3**–**5** (Chart I) as models of pyrrolobenzodiazepine antibiotics.[7] They found that while compounds **3** and **5** readily added to nucleophiles (e.g., thiophenol), only **5** produced covalent attachment to DNA as shown by ethidium fluorescent assay.

Chart I

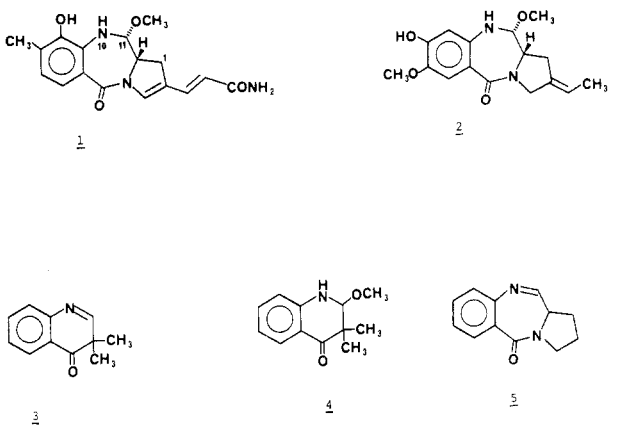
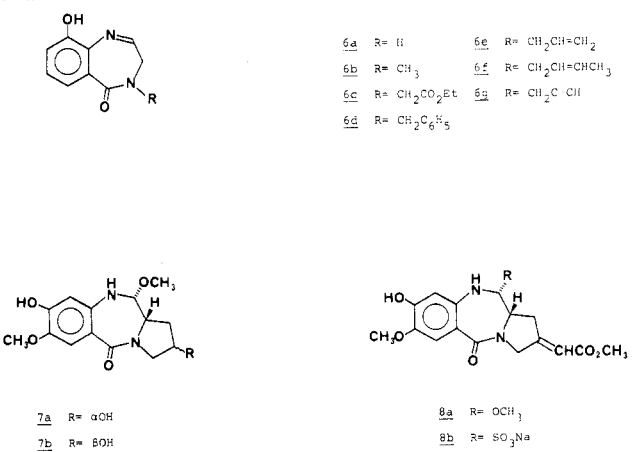


Chart II

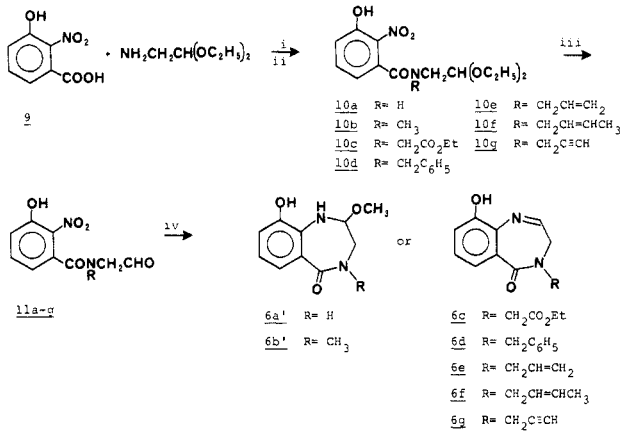


These data and additional published data[8] suggested the possibility of rationally designing new analogues. We

(1) For reviews, see: Hurley, L. H. *J. Antibiot.* **1977**, *30*, 349. Artico, M. *Boll. Chim. Farm.* **1980**, *119*, 445, 505. Hurley, L. H.; Thurston, D. E. *Pharm. Res.* **1984**, 52.
(2) *Pharma Projects* **1983**, K1A ak10.
(3) *SCRIP* **1982**, *690* and *961*, 18.
(4) Korman, S.; Tendler, M. D. *J. New Drugs* **1965**, *5*, 275. Gause, G. F.; Dudnik, Y. V. *Adv. Antimicrob. Antineoplast. Chemother., Proc. Int. Congr. Chemother., 7th, 1971* **1972**, *2*, 87.
(5) Hurley, L. H.; Petrusek, R. *Nature (London)* **1979**, *282*, 529. Hurley, L. H.; Roken, J. S.; Petrusek, R. L. *Biochem. Pharmacol.* **1980**, *29*, 1307.
(6) Arora, S. K. *J. Antibiot.* **1981**, *34*, 462.
(7) Lown J. W.; Joshua, A. V. *Biochem. Pharmacol.* **1979**, *20*, 2017.

0022-2623/85/1828-0388$01.50/0

Scheme I[a]



[a] i, DCC; ii, NaH/RX (method A) or LDA/RX (method B); iii, H^+; iv, H_2/Pd-C/MeOH (method C) or Fe/HOAc/MeOH (method D).

postulated that synthetic analogues might be antitumor active as long as they contain the following features: (1) an imine or carbinolamine functionality at N10–C11, (2) a benzo[1,4]diazepine structure; (3) any substituents to provide secondary stabilizing forces.

We subsequently chose structures **6a–g**, **7a,b**, and **8a,b** as our target molecules (Chart II). Compounds **6a–g** lacked the third (i.e., pyrroline) ring of anthramycin. In the naturally occurring pyrrolobenzodiazepine antibiotics, the significance of the third ring and its C2 substituent was not clear. It was thought that these bicyclic analogues might shed some light on this question. These new structures would contain an imine or a carbinolamine ether moiety at the position corresponding to N10–C11 of anthramycin. They would also contain a phenolic hydroxy group which might participate in hydrogen bonding. Having the same substitution pattern as tomaymycin and neothramycin, tricyclic structures **7a,b** and **8a,b** could be considered as analogues thereof. While our work was in progress, compound **7a** was discovered as a natural product and named chicamycin.[9]

Chemistry. Since the N10–C11 imine or carbinolamine group was quite labile, these functionalities were generated in the last step of the synthetic sequence. The synthesis of the simplest analogue (**6a**) began with the coupling of commercially available 3-hydroxy-2-nitrobenzoic acid[10] and aminoacetaldehyde diethyl acetal (Scheme I). Generation of the aldehyde functionality and reduction of the nitro group gave cyclized carbinolamine ether **6a'**. This type of cyclization had been employed in the synthesis of neothramycin.[11]

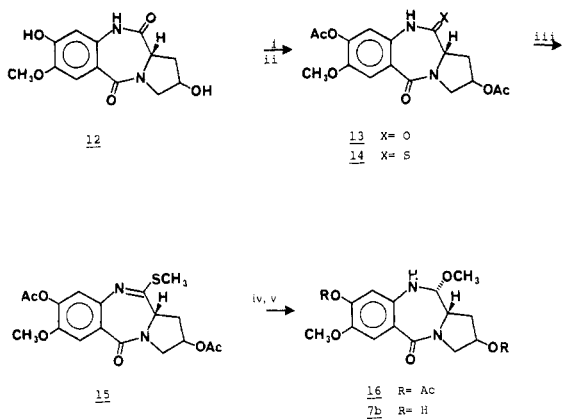
The N4-substituted analogues were prepared by alkylation of the amide nitrogen in **10a**. The alkylated amides were shown by NMR to be a mixture of rotational isomers at the amide bond and they were generally obtained as an oil. The results of alkylation are shown in Table I. Table II summarizes the results of the next two steps—hydrolysis of the acetal and reduction of the nitro group with a concomitant cyclization. When the substituent contained unsaturation, the final reduction of the aromatic nitro group was carried out with use of iron and acetic acid.

Table I. Alkylation of **10a**

no.	RX	method[a]	%	formula	anal.[b]
10b	CH_3I	A	77	$C_{14}H_{20}N_2O_6$	C, H, N
10c	$BrCH_2CO_2Et$	B	52	$C_{17}H_{24}N_2O_8$	C, H, N
10d	$C_6H_5CH_2Br$	B	71	$C_{20}H_{24}N_2O_6$	C[c], H
10e	$CH_2{=}CHCH_2Br$	A	87	$C_{16}H_{22}N_2O_6$	C,[d] H, N
10f	$CH_3CH{=}CHCH_2Br$	A	83	$C_{17}H_{24}N_2O_6$	C,[e] H, N
10g	$CH{\equiv}CCH_2Br$	A	91	$C_{16}H_{20}N_2O_6$	C, H, N

[a] See Scheme I. [b] Analyses shown are correct ±0.4% unless otherwise noted. [c] C: calcd, 61.85; found, 61.42. [d] C: calcd, 56.80; found, 57.25. [e] C: calcd, 57.14; found, 56.72.

Scheme II[a]



[a] i, Ac_2O-Py; ii, $(p\text{-}CH_3OC_6H_4PS_2)_2$; iii, CH_3I/K_2CO_3; iv, Al-Hg; v, K_2CO_3/MeOH.

Since the imine and the carbinolamine ether structures were interconvertible,[1] the final product was obtained in either form depending on the isolation method. That is, if a methanol-containing solvent was used in the silica gel chromatography, a carbinolamine methyl ether was generally obtained; whereas if non-methanol containing solvent mixtures and/or a short contact time with silica gel was used, an imine product was obtained.

Among the tricyclic analogues, syntheses of **2**, **5**, and **7a** have been previously described by us.[11] For the synthesis of **7b** the same intermediate as **7a** was utilized. This intermediate **12** in Scheme II was readily prepared in two steps from 5-methoxy-2-nitro-4-[(4-nitrobenzoyl)oxy]-benzoic acid and *trans*-4-hydroxy-L-proline methyl ester. The C11 amide group of **12** was reduced to carbinolamine ether **16** by using the new method we developed recently.[12] The stereochemistry at C11 was determined as shown since there was no coupling observed in the NMR between the C11 hydrogen and the C11a hydrogen.[13] Compound **7b** was recently synthesized independently by Tozuka et al.[14]

The synthesis of compound **8b** is shown in Scheme III. A Wittig reaction of the ketone **17**[11] in refluxing toluene gave compound **18**. In refluxing THF, on the other hand, the same Wittig reaction gave a small amount of **20** in addition to **18**. The double bond geometry of **18** was not determined, but its NMR indicated that it was essentially a single isomer.

In this sequence the same reduction method as in **7b** was employed. In the deprotection step of **21** an ester exchange

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(11) Miyamoto, M.; Kondo, S.; Naganawa, H.; Maeda, K.; Chno, M.; Umezawa, H. *J. Antibiot.* **1977**, 340.

(12) Kaneko, T.; Wong, H.; Doyle, T. W. *Tetrahedron Lett.* **1983**, *24*, 5165. Kaneko, T.; Wong, H.; Doyle, T. W. *J. Antibiot.* **1984**, *37*, 300.

(13) Tozuka, Z.; Takaya, T. *J. Antibiot.* **1983**, *36*, 142.

(14) Tozuka, Z.; Yazawa, H.; Murata, M., Takaya, T. *J. Antibiot.* **1983**, *36*, 1699.

Table II. Reductive Cyclization of **11a–g**

no.	reduction[a] method	yield, %	formula	anal.[b]	NMR: C-2H (solvent)
6a′	C	30	$C_{10}H_{12}N_2O_3 \cdot CH_3OH$	C, H	4.82 (m) ($CDCl_3$)
6b′	C	37	$C_{11}H_{14}N_2O_3$	C,[c] H, N	5.00 (t, J = 10 Hz) (CD_3OD)
6c	C	45	$C_{13}H_{14}O_4 \cdot 0.5H_2O$	C, H[d]	8.25 (t, J = 4 Hz) ($CDCl_3$)
6d	C	34	$C_{16}H_{14}N_2O_2 \cdot CH_3OH$	C, H, N	7.79 (t, J = 5 Hz) ($CDCl_3$)
6e	D	30	$C_{12}H_{12}N_2O_2 \cdot 0.75CH_3OH$	C, H, N	8.26 (t, J = 4 Hz) ($CDCl_3$)
6f	D	28	$C_{13}H_{14}N_2O_2 \cdot H_2O$	C, H, N	8.12 (t, J = 5 Hz) ($CDCl_3$)
6q	D	34	$C_{12}H_{10}N_2O_2 \cdot 0.75H_2O$	C, H, N	8.22 (t, J = 5 Hz) (CD_3COCD_3)

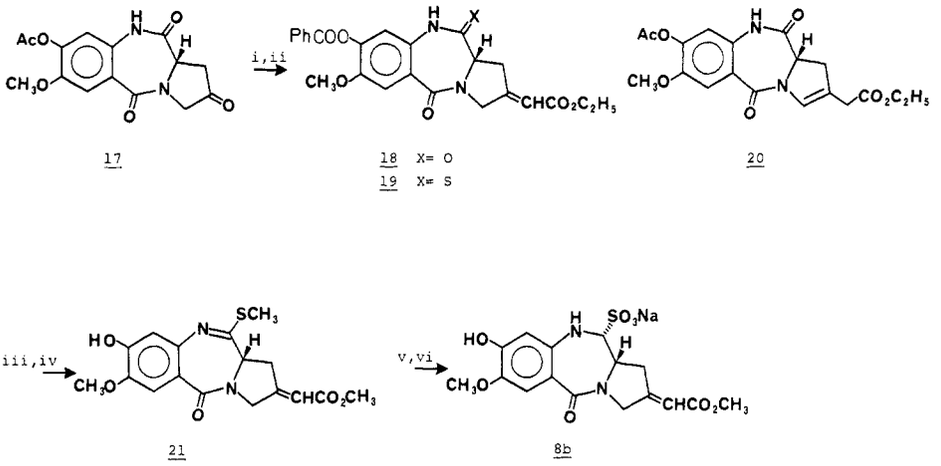
[a] See Scheme I. [b] Analyses shown are correct ±0.4% unless otherwise noted. [c] C: calcd, 59.45; found, 58.97. [d] H: calcd, 5.57; found, 6.01.

Table III. P388 Activity of Analogues

no.	dose schedule	max T/C[a]	OD, mg/kg	ref[b] compd	ref T/C	ref O/D
2	qd 1 → 9	188	0.2			
5	qd 1 → 5	133	8	A	206	0.2
6a	d 1, 5, 9	125	16	N	138	4
6b	d 1	122	16	N	122	8
6c	d 1, 5	106	40[c]	N	111	4
6d	qd 1 → 9	100	16[c]	N	133	3
6e	d 1	117	80	N	128	16
6f	qd 1 → 5	138	20[c]	N	150	2
6g	qd 1 → 5	162	16	N	150	3
7a	qd 1 → 5	144	6.4	A	189	0.3
7b	qd 1 → 5	100	16	A	150	0.3
8b	qd 1 → 5	131	6	A	250	0.3
16	qd 1 → 5	100	16	A	150	0.3

[a] T/C ≥125 is considered active. [b] A = anthramycin methyl ether; N = neothramycin. [c] Highest dose tested.

Scheme III[a]



[a] i, Ph_3PCHCO_2Et; ii, $(p\text{-}CH_3OC_6H_4PS_2)_2$; iii, CH_3I/K_2CO_3; iv, K_2CO_3/MeOH; v, Al-Hg; vi, $NaHSO_3$.

also took place and the final product was obtained as a methyl ester. The carbinolamine methyl ether (**8a**) turned out to be too unstable for isolation, thus the aluminum amalgam reduction product was directly converted to a bisulfite adduct and the product was isolated as such.

Biology. The in vivo activity of the analogues against P388 lymphocytic leukemia is listed in Table III. In this table the activity of the reference compound, i.e., neothramycin or anthramycin methyl ester, in the same test is listed along side. The tests were carried out according to the published procedure.[15]

Due to their relatively low activity (even neothramycin, one of our reference compounds, gives only modest activity), it is difficult to define any structure–activity relationships. It is gratifying, however, to see that the simplest bicyclic compound (**6a**) exhibits activity, even though it is low. Among the N-substituted bicyclic analogues, *N*-propargyl compound **6g** is quite active, giving a T/C value higher than neothramycin, even though it is 5 times less potent. These data indicate that the third ring is not an absolute necessity for antitumor activity. This ring and its C2 substituent may contribute to the compound's potency. Intermediates **10a–g** were not active in the same tests (data not shown), and intermediates **11a–g** were not tested because of their limited stability.

Among the tricyclic analogues, the simplest compound, **5**, lacking any hydrogen-bonding groups, still showed some activity. The corresponding amine (i.e., 1,2,3,10,11,11a-hexahydro-5*H*-pyrrolo[2,1-*c*][1,4]benzodiazepin-5-one) was inactive and this again points out the significance of the imine or carbinolamine moiety. It is of interest to note that the C2α hydroxy compound (**7a**) was active whereas the C2β hydroxy isomer was not under the conditions tested. Intermediates **12** and **14** were inactive and the rest of the intermediates were not tested in the in vivo P388 assay.[16]

(15) Geran, R. I.; Greenberg, N. H.; MacDonald, M. D.; Schumacher, A. M.; Abbott, B. J. *Cancer Chemother. Rep., Part 3* **1972**, *3*, 8.

(16) Intermediate **13** gave a T/C of 144 at 24 mg/kg. In view of the inactivity of oxotomaymycin[17] and **12**, compound **13** seems to be an exception.

Comparison of compound **2** with compounds **7a,b** indicated that for activity and potency the ethylidene group at C2 was more beneficial than a hydroxy group. The bisulfite adduct (**8b**) was also active and potent, and the advantages of a bisulfite adduct were its water solubility and its increased stability. For unstable analogues in this class, the conversion to bisulfite adducts would appear to be a reasonable alternative.[18]

This series of compounds illustrates that it is possible to design and synthesize antitumor active benzo[1,4]diazepene imines or carbinalamine ethers. It warrants further study on attaching groups to the basic skeleton to increase affinity to DNA. The resulting compounds might thus have increased potency and activity.

Experimental Section

Melting points were determined on a Thomas-Hoover capillary apparatus and are not corrected. NMR spectra were obtained on a Varian XL-100 or Bruker 360 spectrometer with tetramethylsilane as internal standard. IR spectra were obtained on a Beckman 4240 spectrophotometer. Elemental analyses were performed by the Analytical Department of these laboratories. The high-resolution mass spectra were obtained at Schrader Analytical Laboratories, Detroit, MI. Preparations of **6a, 6e, 10a–c** are cited as representative procedures for bicyclic analogues.

[*N*-(3-Hydroxy-2-nitrobenzoyl)amino]acetaldehyde Diethyl Acetal (10a). To a solution of dicyclohexylcarbodiimide (5.57 g, 27 mmol) in 100 mL of THF were added at 0 °C 3-hydroxy-2-nitrobenzoic acid (4.58 g, 25 mmol) and aminoacetaldehyde diethyl acetal (3.33 g, 25 mmol). The resulting solution was stirred overnight at room temperature. After the usual workup the residue was filtered through a short silica gel column and crystallized from CH_2Cl_2 and Et_2O to give 7.45 g (79%) of the title compound: mp 118–120 °C; NMR ($CDCl_3$) δ 1.23 (t, 6 H, J = 7 Hz), 3.52–3.86 (m, 6 H), 4.70 (t, 1 H, J = 5 Hz), 6.04 (br s, 1 H), 6.99 (dd, 1 H, J = 7, 2 Hz), 7.21 (dd, 1 H, J = 8, 2 Hz), 7.55 (dd, 1 H, J = 9, 7 Hz), 10.50 (br s, 1 H); IR (KBr) 3290, 1645, 1580, 1535, 1470, 1365, 1335, 1315 cm^{-1}. Anal. ($C_{13}H_{18}N_2O_6$) C, H, N.

1,2,3,4-Tetrahydro-9-hydroxy-2-methoxy-5*H*-1,4-benzodiazepin-5-one (6a') (Method C). Compound **10a** (540 mg, 2 mmol) was dissolved in 10 mL of acetone and 1 mL of 10% aqueous HCl solution was added. After 6 h of stirring at room temperature, the solution was extracted with EtOAc to give 421 mg (94%) of aldehyde **11a**: NMR (acetone-d_6) δ 4.10 (d, 2 H, J = 7 Hz), 7.05–7.90 (m, 3 H), 8.15 (br s, 1 H), 9.57 (s, 1 H). This crude aldehyde (321 mg, 1.43 mmol) was dissolved in 12 mL of MeOH and hydrogenated at 10 psi in the presence of 32 mg of 10% Pd/C. After a usual workup the residue was chromatographed on silica gel (5% MeOH–CH_2Cl_2) and the major product was crystallized from MeOH to give 90 mg (30%) of the title compound: mp 94–97 °C; NMR ($CDCl_3$) δ 3.30 (m, 1 H), 3.38 (s, 3 H), 3.68 (m, 1 H), 4.82 (m, 1 H), 5.65 (d, 1 H, J = 4 Hz), 6.70 (t, 1 H, J = 8 Hz), 6.94 (dd, 1 H, J = 8, 2 Hz), 7.25 (dd, 1 H, J = 8, 2 Hz), 7.52 (br s, 1 H), 9.42 (br s, 1 H); IR (KBr) 3380, 3310, 1625, 1575, 1520, 1470, 1455, 1253, 1190 cm^{-1}. Anal. ($C_{10}H_{12}N_2O_3 \cdot CH_3OH$) C, H.

[*N*-(3-Hydroxy-2-nitrobenzoyl)-*N*-methylamino]acetaldehyde Diethyl Acetal (10b) (Method A). THF (20 mL) was added at 0 °C to a mixture of compound **10a** (1.04 g, 4 mmol) and 50% NaH oil dispersion (442 mg, 9.2 mmol). After the mixture was stirred at 1 h at room temperature, MeI (852 mg, 6 mmol) was added and stirring was continued overnight. After the usual workup, silica gel chromatography (1% MeOH–CH_2Cl_2) gave 958 mg (77%) of the title compound as a slightly yellow oil: NMR ($CDCl_3$) δ 1.30 (t, 6 H, J = 7 Hz), 2.97 (s, 3 H), 3.45–3.95 (m, 6 H), 4.91 (t, 1 H, J = 5 Hz), 6.85 (dd, 1 H, J = 7, 2 Hz), 7.20 (dd, 1 H, J = 8, 2 Hz), 7.60 (dd, 1 H, J = 8, 7 Hz); IR (film) 3320, 1645, 1604, 1580, 1533, 1360, 1298, 1059 cm^{-1}. Anal. ($C_{14}H_{20}N_2O_6$) C, H, N.

[*N*-[(Ethoxycarbonyl)methyl)]-*N*-(3-hydroxy-2-nitrobenzoyl)amino]acetaldehyde Diethyl Acetal (10c) (Method B). To a solution of lithium diisopropylamide (4.2 mmol) in 5 mL of THF was added at −78 °C a solution of **10a** (600 mg, 2 mmol) in 8 mL of THF. After the mixture was stirred for 15 min, a solution of ethyl bromoacetate (367 mg, 2.2 mmol) in 5 mL of THF was added. Stirring was continued for 0.5 h at −78 °C and 1.5 h at room temperature. After the usual workup, silica gel chromatography gave 400 mg of the title compound as a colorless oil in 52% yield: NMR ($CDCl_3$) δ 0.92–1.35 (m, 9 H), 3.10–4.42 (m, 10 Hz), 4.78 (t, 1 H, J = 5 Hz), 6.73–7.63 (m, 3 H), 9.48 (br s, 1 H); IR (KBr) (film) 3240, 1740, 1650, 1542, 1480, 1350, 1200, 1060, 1027 cm^{-1}. Anal. ($C_{17}H_{24}N_2O_8$) C, H, N.

3,4-Dihydro-9-hydroxy-4-allyl-5*H*-1,4-benzodiazepin-5-one (6e) (Method D). Compound **10e** was converted to aldehyde **11e** in a similar manner as **10a**. This crude aldehyde (260 mg, 1 mmol) was dissolved in 5 mL of MeOH. After addition of 200 mg of iron powder and 420 mg of acetic acid, the mixture was refluxed for 2 h. The reaction mixture was diluted with water and EtOAc and then filtered through Celite. The organic layer was washed with brine and dried over Na_2SO_4. The residue obtained was chromatographed on TLC (10% MeOH–CH_2Cl_2) to give 65 mg (30%) of the title compound as an amorphous solid: NMR ($CDCl_3$) δ 3.88 (d, 2 H, J = 4 Hz), 4.40 (dt, 2 H, J = 6, 1 Hz), 5.28–5.51 (m, 2 H), 5.78–6.20 (m, 1 H), 7.33 (dd, 1 H, J = 8, 2 Hz), 7.46 (t, 1 H, J = 8 Hz), 7.22 (dd, 1 H, J = 8, 2 Hz), 8.26 (t, 1 H, J = 4 Hz); IR (KBr) 3383, 1615, 1575, 1492, 1273, 1225 cm^{-1}. Anal. ($C_{12}H_{12}N_2O_2 \cdot 0.75CH_3OH$) C, H, N.

(2*R*,11a*S*)-2,8-Diacetoxy-1,2,3,10,11,11a-hexahydro-7-methoxy-5*H*-pyrrolo[2,1-*c*][1,4]benzodiazepine-5,11-dione (13). A mixture of compound **12**[11] (1.00 g, 3.60 mmol) and 2.5 mL of acetic anhydride in 15 mL of pyridine was stirred at room temperature for 2.5 h. After the usual workup, silica gel chromatography (1% MeOH–CH_2Cl_2) gave 840 mg (76%) of the title compound as an amorphous solid: mp 116–118 °C; NMR ($CDCl_3$) δ 2.08 (s, 3 H), 2.36 (m, 4 H), 3.10 (ddd, 1 H, J = 14, 7, 6 Hz), 3.75 (dd, 1 H, J = 13, 4 Hz), 3.93 (s, 3 H), 4.18 (d, 1 H, J = 13 Hz), 4.32 (t, 1 H, J = 7 Hz), 5.42 (m, 1 H), 6.78 (s, 1 H), 7.60 (s, 1 H), 8.71 (br s, 1 H); IR (KBr) 3470, 1750, 1704, 1649, 1622, 1513, 1440, 1250, 1202 cm^{-1}. Anal. ($C_{17}H_{18}N_2O_7 \cdot 0.5H_2O$) C, H, N.

(2*R*,11a*S*)-2,8-Diacetoxy-1,2,3,10,11,11a-hexahydro-7-methoxy-11-thioxo-5*H*-pyrrolo[2,1-*c*][1,4]benzodiazepin-5-one (14). A mixture of **13** (610 mg, 1.68 mmol) and the Lawesson reagent[19] (340 mg, 0.842 mmol) in 40 mL of benzene was refluxed for 1 h. The residue obtained after evaporation of the solvent was chromatographed on silica gel (1% MeOH–CH_2Cl_2) to give 550 mg (86%) of the title compound as an amorphous solid: mp 120–122 °C; NMR ($CDCl_3$) δ 2.09 (s, 3 H), 2.37 (m, 4 H), 3.62 (d t, 1 H, J = 13, 6 Hz) 3.72 (dd, 1 H, J = 12, 5 Hz), 4.10 (br d, 1 H, J = 12 Hz), 4.44 (dd, 1 H, J = 8, 6 Hz), 5.48 (m, 1 H), 6.86 (s, 1 H), 7.64 (s, 1 H), 9.64 (br s, 1 H); IR (KBr) 3420, 1776, 1745, 1649, 1618, 1505, 1436, 1870, 1250, 1225, 1197 cm^{-1}. Anal. ($C_{17}H_{18}N_2O_6S \cdot 0.75H_2O$) C, H, N.

(2*R*,11a*S*)-2,8-Diacetoxy-7-methoxy-11-(methylthio)-1,2,3,11a-tetrahydro-5*H*-pyrrolo[2,1-*c*][1,4]benzodiazepin-5-one (15). A mixture of **14** (1.30 g, 3.44 mmol), MeI (0.975 g, 6.87 mmol), and K_2CO_3 (1.82 g) in 20 mL of THF was stirred at room temperature for 18 h. The reaction mixture was filtered and the solvent was evaporated to give 1.34 g (100%) of crude title compound: NMR ($CDCl_3$) δ 2.08 (s, 3 H), 2.24–2.60 (m, 8 H), 2.92 (ddd, 1 H, J = 14, 7, 6 Hz), 3.78 (dd, 1 H, J = 13, 4 Hz), 3.94 (s, 3 H), 4.32 (t, 1 H, J = 7 Hz), 5.48 (m, 1 H), 7.02 (s, 1 H), 7.62 (s, 1 H).

(2*R*,11*R*,11a*S*)-2,8-Diacetoxy-7,11-dimethoxy-1,2,3,11a-tetrahydro-5*H*-pyrrolo[2,1-*c*][1,4]benzodiazepin-5-one (16). A solution of **15** (420 mg, 1.07 mmol) in 10% aqueous THF was treated at 0 °C for 18 h with aluminum amalgam (prepared from 289 mg of aluminum foil). The reaction mixture was filtered through Celite and treated with 0.1 N $HgCl_2$ solution in MeOH. The residue obtained after evaporation of the solvent was chro-

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matographed at 5 °C on silica gel TLC (CH_2Cl_2–EtOAc–MeOH, 150:138:12) to give 210 mg (58%) of the title compound as an amorphous solid: mp 88–90 °C; NMR ($CDCl_3$) δ 2.07 (s, 3 H), 2.37 (s, 3 H), 2.57 (dd, 1 H, *J* = 8, 3 Hz), 3.70–4.18 (m, 6 H), 5.42 (m, 1 H), 7.09 (s, 1 H), 7.66 (s, 1 H), 7.75 (d, 1 H, *J* = 4 Hz); IR (KBr) 1770, 1743, 1640, 1610, 1510, 1437, 1237, 1210, 1160 cm^{-1}; $[\alpha]^{24}_D$ +172° (*c* 0.09, MeOH); mass spectrum calcd for $C_{17}H_{18}N_2O_6$ 346.1160, found 346.1139.

(2*R*,11*R*,11a*S*)-2,8-Dihydroxy-7,11-dimethoxy-1,2,3,10,11,11a-hexahydro-5*H*-pyrrolo[2,1-*c*][1,4]benzodiazepin-5-one (7b). To a solution of **16** (210 mg, 0.61 mmol) in 2 mL of MeOH was added at 0 °C 5 mL of K_2CO_3 saturated MeOH. After 1 h of stirring at 0 °C, the solvent was evaporated and the residue was redissolved in 5 mL of water. It was carefully neutralized with 10% HCl solution and washed with CH_2Cl_2. Water was then removed under reduced pressure and the residue was chromatographed on silica gel TLC (EtOAc–MeOH, 85:15) to give 60 mg (37%) of the title compound as an amorphous solid: mp 138–140 °C; NMR (pyridine-d_5) δ 2.22–2.54 (m, 2 H), 3.23 (s, 3 H), 3.78 (s, 3 H), 3.90 (m, 1 H), 4.04–4.52 (m, 3 H), 4.76 (d, 1 H, *J* = 6 Hz), 6.88 (s, 1 H), 7.86 (d, 1 H, *J* = 6 Hz), 8.14 (s, 1 H); IR (KBr) 3360, 1598, 1509, 1440, 1268, 1210, 1065 cm^{-1}; $[\alpha]^{24}_D$ +65° (*c* 0.076, MeOH); mass spectrum calcd for $C_{14}H_{18}N_2O_5 \cdot CH_3OH$ 272.1160, found 272.1135.

(11a*S*)-8-(Benzoyloxy)-2-[(ethoxycarbonyl)methylene]-2,3,5,10,11,11a-hexahydro-7-methoxy-1*H*-pyrrolo[2,1-*c*][1,4]benzodiazepine-5,11-dione (18). A mixture of **17** (1.14 g, 3 mmol)[11] and (carbethoxymethylene)triphenylphosphorane (1.61 g, 4.6 mmol) in 150 mL of toluene was refluxed under argon for 11 h. After the usual workup, silica gel chromatography (20% EtOAc–CH_2Cl_2) gave 850 mg (63%) of the title compound: mp 210–213 °C; NMR ($CDCl_3$) δ 1.28 (t, 3 H, *J* = 7 Hz), 3.32 (m, 2 H), 3.87 (s, 3 H), 4.19 (q, 2 H, *J* = 7 Hz), 4.42 (m, 2 H), 4.91 (m, 1 H), 5.87 (br s, 1 H), 6.92 (s, 1 H), 7.45–7.75 (m, 4 H), 8.22 (dd, 1 H, *J* = 8, 2 Hz), 8.50 (br s, 1 H); IR (KBr) 3240, 1750, 1710, 1648, 1514, 1495, 1435, 1265, 1250, 1226 cm^{-1}. Anal. ($C_{24}H_{22}N_2O_7$) C, H, N.

(11a*S*)-8-(Benzoyloxy)-2-[(ethoxycarbonyl)methylene]-2,3,5,10,11,11a-hexahydro-7-methoxy-11-thioxo-1*H*-pyrrolo[2,1-*c*][1,4]benzodiazepin-5-one (19). A mixture of **18** (340 mg, 0.76 mmol) and the Lawesson reagent (170 mg, 0.42 mmol) in 40 mL of benzene was refluxed under argon for 45 min. The residue obtained after evaporation of the solvent was chromatographed on silica gel (0.5% MeOH–CH_2Cl_2) to give 278 mg (79%) of the title compound: mp 227–228 °C; NMR ($CDCl_3$) δ 1.31 (t, 3 H, *J* = 7 Hz), 3.13 (m, 2 H), 3.91 (s, 3 H), 4.22 (q, 2 H, *J* = 7 Hz), 4.50 (dd, 1 H, *J* = 9, 2 Hz), 4.82 (m, 2 H), 6.00 (m, 1 H), 6.98 (s, 1 H), 7.44–7.72 (m, 4 H), 8.22 (dd, 1 H, *J* = 8, 2 Hz), 9.49 (b s, 1H); IR (KBr) 3430, 1753, 1720, 1653, 1503, 1440, 1382, 1267, 1220, 1160 cm^{-1}. Anal. ($C_{24}H_{22}N_2O_6S \cdot 0.5H_2O$) C, H, N, S.

(11a*S*)-8-Hydroxy-7-methoxy-2-[(methoxycarbonyl)methylene]-11-(methylthio)-2,3,5,11a-tetrahydro-1*H*-pyrrolo[2,1-*c*][1,4]benzodiazepin-5-one (21). A mixture of **19** (548 mg, 1.18 mmol), MeI (1.68 g, 11.8 mmol), and K_2CO_3 (1.63 g) in 25 mL of THF was stirred at room temperature overnight. The residue obtained after filtration and evaporation of the solvent was redissolved in 10 mL of MeOH and treated with 5 mL of K_2CO_3 saturated MeOH solution at 0 °C for 30 min. The solution was then carefully neutralized and the precipitate was collected to give 400 mg (94%) of crude title compound: NMR ($CDCl_3$) δ 2.42 (s, 3 H), 3.33 (m, 2 H), 3.74 (s, 3 H), 3.95 (s, 3 H), 4.45 (m, 2 H), 4.72 (m, 1 H), 5.89 (m, 1 H), 6.82 (s, 1 H), 7.48 (s, 1 H); IR (KBr) 3430, 1740, 1610, 1596, 1507, 1440, 1280, 1212 cm^{-1}; CI (CH_4) mass spectrum 363 (100%, $M^+ + 1$).

Bisulfite Addition Compound of 8-Hydroxy-7-methoxy-2-[(methoxycarbonyl)methylene]-2,3,5,11a-tetrahydro-1*H*-pyrrolo[2,1-*c*][1,4]benzodiazepin-5-one (8b). A solution of **21** (350 mg, 0.97 mmol) in 25 mL of 10% aqueous THF was treated at 0 °C with aluminum amalgam (prepared from 310 mg of aluminum foil) for 13 h. The reaction mixture was filtered through Celite and added to an aqueous (2 mL) solution of sodium bisulfite (614 mg, 5.90 mmol). After the mixture was stirred at 0 °C for 4 h, most of the THF was removed under reduced pressure and the residual aqueous layer was washed alternately with EtOAc and CH_2Cl_2. Water was removed under reduced pressure and the residue was chromatographed with use of C_{18} reversed phase silica gel (THF) and silica gel (30% MeOH–EtOAc) to give 97 mg (24%) of the title compound: mp >168 °C not well-defined; NMR (D_2O) δ 2.85–3.80 (m, 2 H), 3.66 (s, 3 H), 3.87 (s, 3 H), 4.18–4.56 (m, 2 H), 4.80 (m, 1 H), 6.00 (m, 1 H), 6.65 (s, 1 H), 7.21 (s, 1 H); IR (KBr) 3435, 1734, 1608, 1492, 1440, 1410, 1273, 1210 cm^{-1}; mass spectrum calcd for $C_{16}H_{17}N_2O_8SNa \cdot NaSO_3H$ 316.1045, found 316.1013.

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Registry No. 1, 4803-27-4; (±)-**6a**′, 94295-76-8; (±)-**6b**′, 94295-77-9; **6c**, 94295-78-0; **6d**, 94295-79-1; **6e**, 94295-80-4; **6f**, 94295-81-5; **6g**, 94295-82-6; **7a**, 89675-37-6; **7b**, 89300-13-0; **8b**, 94295-83-7; **9**, 602-00-6; **10a**, 94295-84-8; **10b**, 94295-85-9; **10c**, 94295-86-0; **10d**, 94295-87-1; **10e**, 94295-88-2; **10f**, 94295-89-3; **10g**, 94295-90-6; **11a**, 94295-91-7; **11b**, 94295-92-8; **11c**, 94295-93-9; **11d**, 94324-72-8; **11e**, 94295-94-0; **11f**, 94295-95-1; **11g**, 94324-73-9; **12**, 89675-35-4; **13**, 94347-10-1; **14**, 94295-96-2; **15**, 94295-97-3; **16**, 94295-98-4; **17**, 89625-01-4; **18**, 94295-99-5; **19**, 94296-00-1; **21**, 94296-01-2; $NH_2CH_2CH(OEt)_2$, 645-36-3; $BrCH_2C(O)OEt$, 105-36-2; $EtOC(O)CH{=}PPh_3$, 1099-45-2.