+ P (4.0 mg from day 6 through 30), (4) Ec (0.4 mg from day 1 through 30) + 4FE2 (20 μg from day 6 through 30) + P (4.0 mg from day 6 through 30), and (5) Ec (0.4 mg from day 1 through 8) + P (4.0 mg from day 6 through 30).

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Structure–Activity Relationships of Synthetic Antibiotic Analogues of Anisomycin[1,2]

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A general synthetic sequence was used to synthesize a series of analogues of anisomycin, and the biological activities of the new synthetic analogues as antiprotozoals, antifungals, and antibacterials were evaluated. The synthetic antibiotics included 3β-acetoxy-4α-hydroxy-2β-(*p*-methylbenzyl)pyrrolidine (**1b**), 3β-acetoxy-2β-benzyl-4α-hydroxypyrrolidine (**1c**), 3β-acetoxy-4α-hydroxy-2β-(*m*-methoxybenzyl)pyrrolidine (**1d**), 3β-acetoxy-4α-hydroxy-2β-(*o*-methoxybenzyl)pyrrolidine (**1e**), 3β-acetoxy-4α-hydroxy-2β-(α-methyl-*p*-methoxybenzyl)pyrrolidine (**1f**), and 3β-acetoxy-4α-hydroxy-2β-(α-phenyl-*p*-methoxybenzyl)pyrrolidine (**1g**). The anisomycin analogues showed activity against protozoa and fungi, but this activity was restricted primarily to the *p*-methylbenzyl and benzyl analogues **1b** and **1c**. The activities dropped dramatically as the methoxy substituent was moved to the meta or ortho positions of the benzyl group (**1d** and **1e**) or a methyl or phenyl group was attached at the α-benzyl carbon (**1f** and **1g**).

Anisomycin (**1a**),[3] which exhibits a remarkably selective inhibition of peptide chain elongation on 60S eukaryotic ribosomes,[4] has become a valuable tool in molecular biology. Because of this mode of action, anisomycin exhibits selective activity against several strains of fungi and protoza.[5] The antibiotic has been shown to be useful in clinical trials for the treatment of amoebic dysentery[6] and vaginitis[7] and in field applications as a plant fungicide.[8] In an effort to search for a more effective anisomycin antibiotic and to establish structure–activity relationships, we recently developed an efficient, stereospecific total synthesis of (±)-anisomycin (**1a**) and demonstrated the utility of the synthesis by preparing two closely related analogues, **1b** and **1c**.[1b] Herein, this general synthetic sequence was used to prepare new anisomycins, **1d–g**, and subject the entire series **1a–g** to biological evaluation.

Chemistry. The first phase of the synthesis was to elaborate the entire carbon skeleton of anisomycin, which is embodied in the 2-benzylpyrroles **2a–g**, by utilizing our tandem alkylation–reduction techniques.[1a] The second phase of the synthetic sequence was the stereospecific synthesis of the *syn*-epoxides **7a–g**. The last phase then involved the regioselective, stereospecific ring opening of these *syn*-2-benzyl-3-pyrrolidine epoxides **7a–g** and subsequent selective manipulations to convert the resultant 3β,4α-dihydroxy-2β-benzylpyrrolidines **8a–g** by a protection–acetylation–deprotection sequence to the anisomycins **1a–g**. The entire synthesis is outlined in Scheme I.

In our original total synthesis of anisomycin (**1a**) and the analogues **1b** and **1c**, the benzyloxycarbonyl (Cbz) group had been used to protect the secondary amine group at two crucial stages of the synthesis.[1b] N-Protection was necessary during the generation of the halohydrin **5** and during the selective acylation of the *trans*-diol **9**, and for these purposes the *N*-Cbz group performed this task admirably. However, when the *N*-Cbz protecting group was employed in the **d–g** series, the protecting group could *not* be removed by catalytic hydrogenation from the *N*-Cbz derivatives of the *syn*-2-benzyl-3-pyrrolidine epoxides **7d–g**. We subsequently discovered, to our chagrin, that this is a rather general phenomenon for a sterically hindered Cbz protecting group. Medium-pressure (45 psi) catalytic hydrogenation or HBr/HOAc conditions destroyed the epoxide. A procedure,[9] of limited success, was to reflux the *N*-Cbz derivatives (R_2 = Cbz) of the *syn*-epoxides **7e** and **7g** in triethylsilane containing a catalytic amount of $PdCl_2$ and Et_3N to yield the corresponding deprotected *syn*-epoxides **7e** and **7g** in ca. 70% yields. The procedure failed for the *N*-Cbz derivatives (R_2 = Cbz) of the *syn*-epoxides **7d** and **7f**. The best solution to this problem was to change to 2,2,2-trichloroethoxycarbonyl (TCE) as the N-protecting group, which could be removed

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Table I. Antiprotozoal Activities of Anisomycin and Analogues[a]

	concn, μg/mL, for % inhibn											
	Trichomonas vaginalis				*Trichomonas foetus*				*Entamoeba histolytica*			
compd	100%	>95%	95-25%	<25%	100%	>95%	95-25%	<25%	100%	>95%	95-25%	<25%
(−)-1a	2	0.5	0.25		4	1	0.5	0.25	2	1		0.5
(±)-1a	2	1	0.5	0.25	8	2		1	4	2	0.5	0.25
(±)-1b	8	4	2		16	4	1	0.5	4	2	1	
(±)-1c	8	4	2		32	16	4	2	16	2		
(±)-1d	32	16	8	4		16		8	32	16		8
(±)-1e				32				32				32
(±)-1f				32				32				32
(±)-1g			32	16			32	16				32
metronidazole	0.5	0.25	0.12	0.06	0.5		0.25	0.12	0.25	0.12		
clotrimazole	32		8	4					32	16	8	4

[a] See Experimental Section for details.

efficiently with zinc in THF–HOAc to yield the *syn*-epoxides **7d–g** (ca. 85%).

The second major problem developed in the **d–g** series when we realized that neither the *N*-Cbz group nor the *N*-TCE group could be removed from the protected anisomycin analogues of **10** (R_2 = Cbz or TCE) at the end of the syntheses. The N-protection was required during the previous step to realize selective sequencial acylation of the *trans*-diols **8a–g**. The protecting groups, allyloxycarbonyl[10] and 9-fluorenylmethoxycarbonyl (Fmoc),[11] proved satisfactory and could, in turn, be removed under nonhydrolytic conditions. We selected the bulkier Fmoc group, since its presence on the secondary amine of the *trans*-diols **9d–g** made the subsequent desired acylation of the α-hydroxy group at C-4 with 2,2,2-trichloroethyl chloroformate more selective. After acetylation of the β-hydroxy group at C-3, the TCE group was removed with zinc (THF–HOAc) and then the *N*-Fmoc group with 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU)[12] in THF to complete the syntheses of the anisomycin analogues **1d–g**.

In the synthesis of the last anisomycin analogue **1g** in this series, the regioselectivity of the trans opening[13] of the *syn*-epoxide **7g** was lost, and a 1:1 mixture of *trans*-diols was formed. It is known that the regioselectivity is lost with the bulky N-protected *syn*-epoxide **6a**,[1b,14] and it is now apparent that a bulky phenyl group on the α-benzyl carbon also has the same effect. In this case the mixture of the products **1g** and 4β-acetoxy-3α-hydroxy-2β-(α-phenyl-*p*-methoxybenzyl)pyrrolidine (**11**), after carrying the diol mixture through the sequence, was conveniently separated at the end of the synthesis.

Biological Results and Discussion

In vitro antiprotozoal activities of the anisomycin series **1a–g** are summarized in Table I, and the antifungal and antibacterial activities are summarized in Tables II and III . As might be expected,[5] the anisomycin analogues showed activity against protozoa and fungi, but this activity was restricted primarily to the *p*-methylbenzyl and benzyl analogues **1b** and 1c. The activities dropped dramatically as the methoxy substituent was moved to the meta or ortho positions of the benzyl group (**1d** and **1e**) or a methyl or phenyl group was attached at the α-benzyl carbon (**1f** and **1g**).

Table II. Antifungal and Antibacterial Activities (Tube Dilution) of Anisomycin and Analogues[a,b]

	minimum inhibitory concn, μg/mL			
compd	*S.c.*[c]	*T.m.*[c]	*C.a.*[c]	*E.f.*[c]
(−)-1a	0.25	2	4	16
(±)-1a	0.5	4	8	32
(±)-1b	1	4	8	16
(±)-1c	2	16	32	32
(±)-1d	8	>32	>32	>32
(±)-1e	>32	>32	>32	>32
(±)-1f	>32	>32	>32	>32
(±)-1g	>32	>32	>32	>32
metronidazole	>32	>32	>32	>32
clotrimazole	0.06	0.06	4	0.06

[a] See Experimental Section for details. [b] Compounds 1a–g were inactive (>32) for the following bacteria: *Actinomyces israelii, Clostridium novyi, Propionibacterium acnes, Bacteroides fragilis, Staphylococcus aureus, Streptococcus pyogenes C, Bacillus subtilis, Mycobacterium fortuitum, Streptomyces griseus, Nocardia asteroides, Escherichia coli, Salmonella schottmuelleri,* and *Pseudomonas aeruginosa.* [c] Fungi: *S.c.* = *Saccharomyces cerevisiae; T.m.* = *Trichophyton mentagrophytes; C.a.* = *Candida albicans; E.f.* = *Epidermophyton floccosum.*

Numerous compounds of diverse chemical structure have modes of action similar to anisomycin for inhibiting protein synthesis by binding to eukaryotic ribosomes.[4] In an attempt to correlate structure with the ability to inhibit protein synthesis, it has been proposed that the essential structural feature required for this biological activity is a secondary amine group adjacent to an asymmetric carbon (*R* configuration) that is linked through a methylene to a sterically unhindered six-membered ring.[15] The structure–activity relationships reported herein tend to support the Grollman hypothesis.

Experimental Section

General Comments. The synthesis of the 2-benzylpyrroles **2a–g**[1a] and the (±)-anisomycins 1a–c have been described.[1b] Benzyl chloroformate, *N*-bromoacetamide (recrystallized from CH_2Cl_2, mp 110–111 °C), 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU), sodium trifluoroacetate, and 2,2,2-trichloroethyl chloroformate were from Aldrich Chemical Co. 9-Fluorenylmethyl 4,6-dimethyl-2-pyrimidylthiocarbonate was prepared by the general method of Nagasawa et al.[16] *N*-Iodosuccinimide was from

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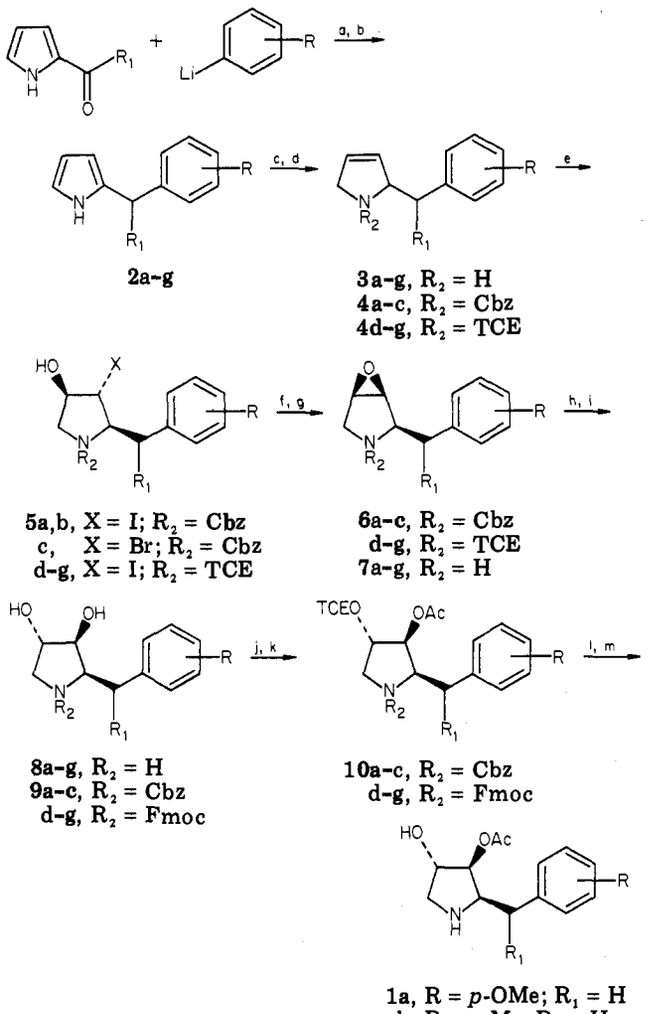
Scheme I[a]



[a] (a) Et_2O; (b) Li-NH_3/NH_4Cl; (c) Zn/aq HCl/EtOH; (d) $C_6H_5CH_2OCOCl$ or CCl_3CH_2OCOCl in toluene/2 N NaOH; (e) NIS or NBA/aq $HClO_4$/THF; (f) 5-10% KOH (EtOH); (g) H_2/10% Pd-C/MeOH or Zn/HOAc-THF, then 2 N NaOH; (h) CF_3CO_2H/CF_3CO_2Na/120 °C; (i) $C_6H_5CH_2OCOCl$ or 9-fluorenylmethyl 4,6-dimethyl-2-pyrimidylthiocarbonate in 10% Na_2CO_3/THF; (j) CCl_3CH_2OCOCl/pyridine/CH_2Cl_2; (k) Ac_2O/pyridine; (l) Zn/HOAc-THF, then 2 N NaOH; (m) H_2/10% Pd-C/MeOH or DBU/THF.

Alfa Products. Solvents were removed in vacuo with either a house vacuum line system (88 torr) or a vacuum pump (5 torr) attached to a rotary evaporator. TLC were performed on Analtech silica gel GF plates (10 × 20 cm, 250 μm) with Et_2O–hexane (1:1) for compounds **4d–g** and **6d–g**, MeOH–$CHCl_3$ (1:4) for compounds **3d–g** and **7d–g**, and toluene–$CHCl_3$–MeOH–28% NH_4OH (40:10:10:1) for compounds **9d–g** and **1d–g**. Purification of products by column chromatography was accomplished on silica gel (Grace, grade 62, 60–200 mesh). The assigned structures of all isolated products were consistent with the spectral data. Six of the intermediate secondary amines **3d,f,g**, **6e,f**, and **7g**, which were oils or amorphous solids, did not give completely satisfactory elemental analyses in spite of repeated purifications. However, analyses of the four N-derived secondary amines **4d,f,g** and **6g** were satisfactory, as were the analyses of **1e,f**, the direct conversion products of the *N*-TCE epoxides **6e** and **6f**, respectively.

Melting points (uncorrected) were determined with a Fisher-Johns apparatus. Refractive indexes were determined with an American Optical ABBE Model 10450 refractometer. Specific rotations were determined with a Rudolph Research Model Autopol III automatic polarimeter. The IR spectra were determined with a Perkin-Elmer Model 180 infrared spectrophotometer. The UV spectra were determined with a Cary Model 118 ultraviolet spectrophotometer. All NMR spectra were determined in $CDCl_3$, and the chemical shifts are expressed in δ values (ppm) relative to a Me_4Si internal standard. The ^1H NMR spectra were determined at 79.5 MHz with a Varian Associates Model CFT-20 Fourier transform NMR spectrometer, at 90 MHz with a Varian Associates Model EM 390 NMR spectrometer, at 100 MHz with a Varian Associates Model XL-100 and a JEOL Model JNM-PS-FT-100 Fourier transform NMR spectrometers, and at 360 MHz with a Bruker Model WH 360 Fourier transform NMR spectrometer. The ^{13}C NMR spectra were determined at 20 MHz with a Varian Associates Model FT-80A and at 25.2 MHz with a Varian Associates Model XL-100 Fourier transform NMR spectrometers, and noise (broadband proton) decoupled spectra were collected. The mass spectra were determined with a Varian Associates Model MAT CH-5 single-focusing mass spectrometer with an SS-100C data system.

2-(*m*-Methoxybenzyl)-3-pyrroline (3d). Reduction of 6.00 g (32.1 mmol) of 2-(*m*-methoxybenzyl)pyrrole (**2d**)[1a] as described for **3a**[1b] yielded 3.24 g (17.14 mmol, 53%) of **3d** as a colorless oil: n^{22}_D 1.5605; IR (film) 3450–3100, 3075, 3000, 2940, 2840, 1610, 1590, 1500, 1475, 1460, 1445, 1270, 1160, 1055, 790, 710 cm^{-1}; UV (MeOH) λ_{max} 220 nm (ε 8400, sh), 272 (2055), 279 (1920); ^1H NMR (79.5 MHz) δ 7.36–7.02 (1 H, complex m), 6.94–6.59 (3 H, complex m), 5.84 (1 H, d with further fine splitting, J = 8 Hz), 5.74 (1 H, d with further fine splitting, J = 8 Hz), 4.40–4.03 (1 H, broad m), 3.80 (3 H, s) superimposed on a complex multiplet at 3.90–3.60 (2 H), 2.72 (2 H, d, J = 7 Hz), 2.06 (1 H, broad s, exchanges with D_2O); mass spectrum, m/z (relative intensity) 189 (M^+, 1), 188 (5), 187 (30), 186 (17), 156 (9), 121 (8), 91 (9), 80 (36), 70 (15), 68 (100), 43 (23), 41 (14), 39 (10). Anal. ($C_{12}H_{15}NO$) H; C, N: calcd, 76.15, 7.40; found, 74.49, 6.20.

2-(*o*-Methoxybenzyl)-3-pyrroline (3e). Reduction of 10.50 g (56.2 mmol) of 2-(*o*-methoxybenzyl)pyrrole (**2e**)[1a] as described for **3a**[1b] yielded 6.70 g (35.4 mmol, 65%) of **3e** as a colorless oil: n^{23}_D 1.5610; IR (film) 3450–3100, 3060, 3025, 3000, 2940, 2920, 2840, 1605, 1590, 1500, 1470, 1440, 1250, 1120, 1060, 1040, 760 cm^{-1}; UV (MeOH) λ_{max} 215 nm (ε 15950), 270 (1900), 277 (1760); ^1H NMR (79.5 MHz) δ 7.30–6.97 (2 H, complex m), 6.97–6.67 (2 H, complex m), 5.80 (1 H, d with further fine splitting, J = 8 Hz), 5.70 (1 H, d with further fine splitting, J = 8 Hz), 4.40–4.03 (1 H, broad m), 3.80 (3 H, s) superimposed on a complex m at 3.90–3.60 (2 H), 2.78 (2 H, d, J = 7 Hz), 2.49 (1 H, broad s, exchanges with D_2O); mass spectrum, m/z (relative intensity) 189 (M^+, 1), 188 (13), 187 (100), 186 (30), 172 (20), 170 (14), 156 (22), 154 (19), 115 (10), 91 (17), 81 (21), 80 (99), 68 (63), 43 (25). Anal. ($C_{12}H_{15}NO$·HCl) C, H, N.

2-(α-Methyl-*p*-methoxybenzyl)-3-pyrroline (3f). Reduction of 12.70 g (63.2 mmol) of 2-(α-methyl-*p*-methoxybenzyl)pyrrole (**2f**)[1a] as described for **3a**[1b] yielded 7.10 g (35.0 mmol, 55%) of **3f** as a colorless oil: IR (film) 3500–3050, 3065, 2970, 2935, 2835, 1610, 1580, 1520, 1460, 1250, 1180, 1040, 835 cm^{-1}; UV (MeOH) λ_{max} 224 nm (ε 9200), 275 (1385), 282 (1200); ^1H NMR (79.5 MHz) δ 7.12 (2 H, d with further fine splitting, J = 9 Hz), 6.82 (2 H, d with further fine splitting, J = 9 Hz), 5.80 (1 H, d with further fine splitting, J = 8 Hz), 5.53 (1 H, d with further fine splitting, J = 8 Hz), 4.03 (1 H, broad s, exchanges with D_2O) superimposed on 4.28–3.90 (1 H, broad m), 3.79 (3 H, s) superimposed on a complex multiplet at 3.90–3.63 (2 H), 2.77 (1 H, quintet, J = 7 Hz), 1.36 (3 H, d, J = 7 Hz); mass spectrum, m/z (relative intensity) 203 (M^+, 0.7), 202 (1), 201 (3), 186 (9), 136 (10), 135 (10), 121 (7), 91 (8), 68 (100), 41 (18). Anal. ($C_{13}H_{17}NO$·HCl) H; C, N: calcd, 65.13, 5.84; found, 63.07, 5.21.

2-(α-Phenyl-*p*-methoxybenzyl)-3-pyrroline (3g). Reduction of 3.70 g (14.1 mmol) of 2-(α-phenyl-*p*-methoxybenzyl)pyrrole (**2g**)[1a] as described for **3a**[1b] yielded 2.15 g (8.11 mmol, 58%) of **3g** as an amorphous solid: IR (film) 3600–3100, 3060, 3010, 2950, 2930, 2900, 2840, 1610, 1580, 1510, 1250, 1180, 1035, 705 cm^{-1}; UV (MeOH) λ_{max} 229 nm (ε 14550), 261 (1210), 268 (1455), 277 (1770), 283 (1500); ^1H NMR (79.5 MHz) δ 7.19 (5 H, superficial s) superimposed on a complex m at 7.4–7.1 (2 H), 6.74 (2 H, dd, J = 9 and 2 Hz), 5.76 (1 H, dd, J = 6 and 2 Hz), 5.53 (1 H, dt, J =

Table III. Antifungal and Antibacterial Activities (Agar Diffusion) of Anisomycin and Analogues[a,b]

compd	inhibitory zone, mm diameter								
	S.c.[c]	T.m.[c]	C.a.[c]	E.f.[c]	A.i.[d]	C.n.[d]	P.a.[d]	B.f.[d]	S.p.[d]
(−)-1a	13	10	±[e]	8	0	0	0	10	0
(±)-1a	10	10	0	8	0	0	0	0	0
(±)-1b	0	10	0	8	0	0	0	0	0
(±)-1c	±	10	0	8	0	0	0	0	0
(±)-1d	0	10	0	8	10	15	12	10	0
(±)-1e	0	0	0	0	0	13	±	0	0
(±)-1f	0	0	0	0	0	8	0	24	0
(±)-1g	0	0	0	10	10	20	18	0	13
metronidazole	0	0	0	8	0	25	0	20	0
clotrimazole	17	37	20	30	18	20	25	25	18

[a] See Experimental Section for details. [b] Compounds 1a-g were inactive (0) for the following bacteria: *Staphylococcus aureus, Bacillus subtilis, Mycobacterium fortuitum, Streptomyces griseus, Nocardia asteroides, Escherichia coli, Salmonella schottmuelleri,* and *Pseudomonas aeruginosa.* [c] Fungi: *S.c. = Saccharomyces cerevisiae; T.m. = Trichophyton mentagrophytes; C.a. = Candida albicans; E.f. = Epidermophyton floccosum.* [d] Bacteria: *A.i. = Actinomyces israelii; C.n. = Clostridium novyi; P.a. = Propionibacterium acnes; B.f. = Bacteroides fragilis; S.p. = Streptococcus pyogenes C.* [e] Indicates questionable activity.

6 and 2 Hz), 4.88–4.53 (1 H, broad m), 3.75 (3 H, s) superimposed on complex m at 3.95–3.52 (3 H), 1.85 (1 H, broad s, exchanges with D_2O); mass spectrum, *m/z* (relative intensity) 265 (M^+, 4), 264 (7), 263 (23), 262 (8), 198 (18), 197 (25), 186 (27), 165 (14), 156 (13), 154 (12), 153 (16), 152 (11), 91 (15), 70 (18), 69 (22), 68 (100), 43 (18), 41 (34). Anal. ($C_{18}H_{19}NO$) H, N; C: calcd, 81.47; found, 80.22.

1-[(Trichloroethoxy)carbonyl]-2-(*m*-methoxybenzyl)-3-pyrroline (4d). To a cooled (3–5 °C, ice–water bath), stirred solution of 4.00 g (21.2 mmol) of **3d** in 53 mL of toluene and 53 mL of 2 N NaOH was added 3.2 mL (23.2 mmol) of 2,2,2-trichloroethyl chloroformate. After the mixture was stirred for 1 h, the organic phase was separated, washed with H_2O, dried ($MgSO_4$), filtered, and concentrated at vacuum pump pressure. Following column chromatography (50 g of silica gel, 2 to 10% Et_2O in hexane), 6.09 g (16.7 mmol, 79%) of **4d** was obtained as a colorless oil: n^{21}_D 1.5470; IR (film) 3000, 2950, 2870, 2835, 1725, 1600, 1585, 1490, 1415, 1330, 1260, 1130, 1055, 770, 720 cm^{-1}; UV (MeOH) λ_{max} 223 nm (ϵ 24 180, sh), 272 (2175), 279 (2095); ^1H NMR (79.5 Hz) δ 7.32–7.02 (1 H, complex m), 6.88–6.60 (3 H, complex m), 5.74 (1 H, d, J = 8 Hz), 5.64 (1 H, d, J = 8 Hz), 4.81 (2 H, s) superimposed on 5.0–4.6 (1 H, broad m), 4.45–4.13 (1 H, complex m), 4.13–3.70 (1 H, complex m), 3.75 (3 H, s), 3.47–3.09 (1 H, complex m), 3.02–2.59 (1 H, complex m, apparent dt, J = 13 and 8 Hz); mass spectrum, *m/z* (relative intensity) 367 (M^+, 0.2), 365 (M^+, 0.6), 363 (M^+, 0.4), 246 (28), 244 (75), 242 (72), 216 (11), 135 (18), 133 (31), 131 (34), 122 (34), 121 (21), 112 (35), 95 (18), 91 (23), 68 (100). Anal. ($C_{15}H_{16}NO_3Cl_3$) C, H, N, Cl.

1-[(Trichloroethoxy)carbonyl]-2-(*o*-methoxybenzyl)-3-pyrroline (4e). Treatment of 1.00 g (5.30 mmol) of **3e** as described for **4d** yielded 1.63 g (4.50 mmol, 85%) of **4e** as a colorless oil: n^{23}_D 1.5430; IR (film) 3025, 2950, 2875, 2830, 1725, 1650, 1500, 1420, 1330, 1250, 1060, 760, 720 cm^{-1}; UV (MeOH) λ_{max} 222 nm (ϵ 8055), 272 (2570), 279 (2420); ^1H NMR (79.5 MHz) δ 7.30–6.96 (2 H, complex m), 6.96–6.62 (2 H, complex m), 5.66 (1 H, d, J = 8 Hz), 5.56 (1 H, d, J = 8 Hz), 4.82 (2 H, s) superimposed on 5.06–4.70 (1 H, broad m), 4.24 (1 H, d with further fine splitting, J = 14 Hz), 3.87 (1 H, dd, J = 14 and 5 Hz) on which is superimposed singlets at 3.80 and 3.79 (3 H), 3.33 (1 H, dd, J = 13 and 4 Hz), 2.88 (1 H, dd, J = 13 and 7 Hz); mass spectrum, *m/z* (relative intensity) 367 (M^+, 0.4), 365 (M^+, 1), 363 (M^+, 1), 246 (44), 244 (100), 242 (100), 216 (22), 133 (48), 113 (51), 122 (86), 112 (44), 95 (22), 91 (54), 68 (84). Anal. ($C_{15}H_{16}NO_3Cl_3$) C, H, N, Cl.

1-[(Trichloroethoxy)carbonyl]-2-(α-methyl-*p*-methoxybenzyl)-3-pyrroline (4f). Treatment of 3.00 g (14.8 mmol) of **3f** as described for **4d** yielded 4.40 g (11.6 mmol, 79%) of **4f** as a colorless oil: IR (film) 3000, 2965, 2910, 2870, 2835, 1730, 1615, 1520, 1420, 1330, 1255, 1185, 1130, 715 cm^{-1}; UV (MeOH) λ_{max} 224 nm (ϵ 12 655), 274 (1665), 281 (1425); ^1H NMR (79.5 MHz) δ 7.2–6.6 (4 H, complex m), 6.0–5.3 (2 H, complex m), 5.1–4.6 (3 H, complex m), 4.38 (1 H, d with further fine splitting, J = 16 Hz), 4.08 (1 H, d with further fine splitting, J = 16 Hz), singlets at 3.79 and 3.74 (3 H) superimposed on multiplet at 4.0–3.0 (1 H), doublets at 1.32 and 1.12 (3 H, J = 7 Hz); mass spectrum, *m/z* (relative intensity) 379 (M^+, 0.4), 377 (M^+, 0.4), 344 (3), 342 (5), 246 (30), 244 (86), 242 (85), 230 (33), 187 (19), 136 (100), 135 (100), 133 (51), 131 (55), 121 (37), 112 (66), 105 (93), 103 (43), 95 (37), 91 (64), 79 (68), 77 (54), 68 (100), 67 (50). Anal. ($C_{16}H_{18}NO_3Cl_3$) C, H, N, Cl.

1-[(Trichloroethoxy)carbonyl]-2-(α-phenyl-*p*-methoxybenzyl)-3-pyrroline (4g). Treatment of 5.30 g (20.0 mmol) of **3g** as described for **4d** yielded 8.20 g (18.6 mmol, 93%) of **4g** as a colorless oil: IR (film) 3080, 3060, 3025, 3000, 2950, 2910, 2865, 2730, 1720, 1610, 1520, 1415, 1330, 1250, 1180, 1130, 1035, 840, 805, 760, 710 cm^{-1}; UV (MeOH) λ_{max} 225 nm (ϵ 13 735), 268 (1425), 276 (1660), 283 (1365); ^1H NMR (100 MHz) δ 7.4–7.0 (7 H, complex m), 7.0–6.7 (2 H, complex m), 6.0–5.7 (2 H, m), 5.5–5.3 (1 H, m), 5.1–4.6 (3 H, complex overlapping multiplets), 4.22 (1 H, d with further fine splitting, J = 16 Hz), singlets at 3.81 and 3.80 (3 H), 3.56 (1 H, d with further splitting, J = 16 Hz); mass spectrum, *m/z* (relative intensity) 439 (M^+, 0.1), 406 (0.4), 404 (0.5), 292 (5), 244 (24), 242 (25), 198 (70), 197 (100), 182 (12), 165 (21), 153 (24), 133 (16), 131 (17), 112 (15), 95 (11), 91 (11), 68 (46). Anal. ($C_{21}H_{20}NO_3Cl_3$) C, H, N, Cl.

1-[(Trichloroethoxy)carbonyl]-2β-(*m*-methoxybenzyl)-3,4β-epoxypyrrolidine (6d). To a cooled (3–5 °C, ice–water bath), stirred solution of 6.09 g (16.7 mmol) of **4d** in 120 mL of THF was added dropwise a solution of 4.8 g of 70% perchloric acid in 12 mL of H_2O, followed by the slow addition of 13.0 g (57.8 mmol) of crystalline *N*-iodosuccinimide over a 1-h period. During an additional hour of stirring the temperature was allowed to slowly rise to ambient, and then a saturated Na_2SO_3 solution was added until the mixture on testing gave a negative peroxide test with KI–starch test paper. The mixture was then extracted with Et_2O, and the organic phase separated, dried ($MgSO_4$), and concentrated at reduced pressure (88 torr). After a solution containing the concentrate (**5d**) in 150 mL of 5% KOH (EtOH) was stirred for 30 min, water was added, and the solution was extracted with CH_2Cl_2. The organic phase was separated, dried ($MgSO_4$), and filtered, and the filtrate was concentrated at reduced pressure (88 torr). Following column chromatography (60 g of silica gel, 5 to 20% Et_2O in hexane), 5.21 g (13.7 mmol, 82%) of **6d** was obtained as an amorphous solid: IR (film) 3040, 2995, 2945, 2875, 2855, 1720, 1585, 1515, 1420, 1390, 1320, 1120, 865, 815, 760, 710 cm^{-1}; UV (MeOH) λ_{max} 230 nm (ϵ 8280), 273 (1455), 280 (1495), 290 (9100, sh); ^1H NMR (79.5 MHz) δ 7.71 (1 H, d, J = 8 Hz), 7.42–6.69 (2 H, complex multiplets), 6.55 (1 H, dd, J = 8 and 3 Hz), 4.81 (2 H, broad s, $w_{1/2}$ = 6 Hz), two singlets at 3.82 and 3.79 (3 H) superimposed on 4.4–3.3 (6 H, complex overlapping multiplets), 3.1–2.0 (1 H, m); mass spectrum, *m/z* (relative intensity) 383 (M^+, 1), 381 (M^+, 4), 379 (M^+, 4), 262 (10), 260 (31), 258 (33), 234 (11), 232 (7), 158 (21), 156 (33), 133 (20), 131 (21), 127 (23), 121 (70), 105 (17), 91 (100), 86 (50), 84 (45), 77 (17), 65 (22), 56 (25), 42 (16). Anal. ($C_{15}H_{16}NO_4Cl_3$) C, H, N, Cl.

1-[(Trichloroethoxy)carbonyl]-2β-(*o*-methoxybenzyl)-3,4β-epoxypyrrolidine (6e). Treatment of 6.00 g (16.5 mmol) of **4e** as described for **6d** yielded 3.80 g (10.0 mmol, 61%) of **6e**

as an amorphous solid: IR (film) 3010, 2950, 2875, 2835, 1720, 1600, 1590, 1490, 1420, 1395, 1330, 1255, 1120, 1035, 760, 720 cm^{-1}; UV (MeOH) λ_{max} 233 nm (ϵ 8360), 272 (1575), 279 (1720), 292 (960); ^1H NMR (79.5 MHz) δ 7.52 (1 H, dd with further fine splitting, J = 10 and 2 Hz), 7.25 (1 H, apparent t with further fine splitting, J = 8 Hz), 6.91 (1 H, apparent t with further fine splitting, J = 7 Hz), 6.63 (1 H, d, J = 8 Hz), two overlapping singlets at 4.81 and 4.72 (2 H), two overlapping singlets at 3.84 and 3.70 (3 H) superimposed on 4.4–3.3 (6 H, complex overlapping multiplets), 3.1–2.5 (1 H, complex m); mass spectrum, m/z (relative intensity) 383 (M^+, 0.3), 382 (0.3), 381 (M^+, 1), 380 (0.6), 379 (M^+, 1), 378 (0.3), 358 (6), 262 (31), 260 (96), 258 (100), 247 (30), 217 (18), 133 (50), 131 (53), 127 (58), 121 (23), 97 (18), 95 (25), 91 (32), 90 (57), 84 (63), 77 (22), 56 (25). Anal. ($C_{15}H_{16}NO_4Cl_3$) H, N; C, Cl: calcd, 47.33, 27.94; found, 46.88, 27.24.

1-[(Trichloroethoxy)carbonyl]-2β-(α-methyl-*p*-methoxybenzyl)-3,4β-epoxypyrrolidine (6f). Treatment of 4.00 g (10.6 mmol) of **4f** as described for **6d** yielded 2.60 g (6.6 mmol, 62%) of **6f** as a colorless oil: IR (Nujol) 3045, 3000, 2955, 2880, 2835, 1725, 1615, 1585, 1520, 1420, 1400, 1330, 1255, 1180, 1130, 1040, 885, 835, 765, 720 cm^{-1}; UV (MeOH) λ_{max} 224 nm (ϵ 11 820), 275 (1650), 282 (1460); ^1H NMR (79.5 MHz) two overlapping doublets at δ 7.33 and 7.25 (2 H, J = 8 Hz), two overlapping doublets at 6.91 and 6.82 (2 H, J = 8 Hz), 4.75 (2 H, s), two overlapping singlets at 3.80 and 3.79 (3 H) superimposed on 4.2–3.3 (6 H, complex overlapping multiplets), two overlapping doublets at 1.42 and 1.33 (3 H, J = 7 Hz); mass spectrum, m/z (relative intensity) 399 (M^+, 0.1), 397 (M^+, 0.9), 395 (M^+, 3), 393 (M^+, 3), 246 (5), 136 (28), 135 (100), 105 (22), 103 (9), 91 (11), 79 (8), 77 (8). Anal. ($C_{16}H_{18}NO_4Cl_3$) C, H, N; Cl: calcd, 26.95; found, 27.46.

1-[(Trichloroethoxy)carbonyl]-2β-(α-phenyl-*p*-methoxybenzyl)-3,4β-epoxypyrrolidine (6g). Treatment of 7.80 g (17.7 mmol) of **4g** as described for **6d** yielded 4.80 g (10.5 mmol, 59%) of **6g** as a colorless oil: IR (film) 3060, 3035, 3000, 2950, 2875, 2835, 1720, 1615, 1520, 1420, 1390, 1310, 1255, (M^+, 1130, 1040, 875, 850, 820, 760, 710 cm^{-1}; UV (MeOH) λ_{max} 227 nm (ϵ 13 525), 262 (1090), 268 (1350), 276 (1560), 283 (1310); ^1H NMR (79.5 MHz) δ 7.23 (5 H, superficial s) superimposed on 7.58–7.06 (2 H, complex m), two overlapping doublets at 6.86 and 6.74 (2 H, J = 8 Hz), 4.65 (2 H, broad s, $w_{1/2}$ = 5 Hz) superimposed on a broad multiplet at 4.84–4.44 (1 H), singlets at 3.80 and 3.75 (3 H) superimposed on 4.3–3.1 (5 H, broad complex overlapping multiplets); mass spectrum, m/z (relative intensity) 459 (M^+, 0.1), 457 (M^+, 0.5), 455 (M^+, 0.5), 439 (0.5), 437 (0.5), 308 (2), 198 (18), 197 (100), 165 (7), 153 (7), 119 (10), 105 (20), 97 (8), 91 (17), 85 (14), 71 (24), 69 (15), 57 (43), 56 (14), 55 (20), 43 (35), 41 (25). Anal. ($C_{21}H_{20}NO_4Cl_3$) C, H, N, Cl.

2β-(α-Phenyl-*p*-methoxybenzyl)-3,4β-epoxypyrrolidine (7g). To a stirred solution containing 4.60 g (10.1 mmol) of **6g** in 100 mL of HOAc–THF (1:4) was added 20 g of zinc powder. After stirring for 1 h, the slurry was filtered, and the filtrate was adjusted to pH 9 with 2 N NaOH. The solution was then extracted with CH_2Cl_2, and the organic phase was separated, dried ($MgSO_4$), and concentrated at reduced pressure (88 torr). Following column chromatography (40 g of silica gel, 5% MeOH in CH_2Cl_2), 2.35 g (8.36 mmol, 83%) of **7g** was obtained as a colorless oil: IR (film) 3340, 3030, 3000, 2950, 2900, 2850, 2830, 1700 (wk), 1615, 1585, 1520, 1455, 1250, 1180, 1040, 700 cm^{-1}; UV (MeOH) λ_{max} 229 nm (ϵ 12 930), 262 (1145), 268 (1450), 276 (1650), 273 (1430); ^1H NMR (79.5 Hz) δ 7.27 (5 H, superficial s) superimposed on 7.4–7.0 (2 H, complex m), 6.81 (2 H, d with further fine splitting, J = 9 Hz), 3.95 (1 H, d, J = 11 Hz), 3.75 (3 H, s) superimposed on 3.66 (1 H, d, J = 11 Hz), 3.53 (1 H, d, J = 3 Hz), 3.21 (1 H, d, J = 3 Hz) superimposed on 3.18 (1 H, d, J = 13 Hz), 2.79 (1 H, d, J = 13 Hz), 1.55 (1 H, s, exchanges with D_2O); mass spectrum, m/z (relative intensity) 282 (1), 281 (M^+, 3), 263 (2), 213 (3), 198 (26), 197 (86), 182 (10), 165 (18), 153 (16), 103 (12), 84 (100), 75 (13), 69 (19), 56 (19), 43 (18). Anal. Calcd for $C_{18}H_{19}NO_2$: C, 76.84; H, 6.81; N, 4.98. Found: C, 73.78; H, 6.32; N, 4.47.

3β-Acetoxy-4α-hydroxy-2β-(*m*-methoxybenzyl)pyrrolidine (1d). Treatment of 5.21 g (13.7 mmol) of **6d** as described for **7g** yielded 2.20 g (10.7 mmol, 78%) of **7d** as a colorless oil. A solution of 1.50 g (7.30 mmol) of **7d** and 1.50 g of sodium trifluoroacetate in 50 mL of trifluoroacetic acid was cautiously lowered into a hot oil bath (ca. 120 °C) so that boiling started immediately.[17] After 16 h of refluxing, the solution was allowed to cool and the solvent was then removed at reduced pressure (88 torr). Tetrahydrofuran (50 mL) was added to the residue, and a solution of 10% Na_2CO_3 was added to adjust the solution pH 9. Crystalline 9-fluorenylmethyl 4,6-dimethyl-2-pyrimidylthiocarbonate[16] (3.30 g, 9.11 mmol) was slowly added to the mixture, which was then heated at 45 °C for 1 h. After cooling, the mixture was extracted with CH_2Cl_2. The organic phase was separated, dried ($MgSO_4$), and filtered, and the filtrate was concentrated at reduced pressure (88 torr). Following column chromatography on 150 g of silica gel and eluting with toluene–CH_2Cl_2–MeOH–MeOH saturated with NH_3 gas (32:8:3:1) yielded *N*-[(fluorenylmethoxy)-carbonyl]-3β,4α-dihydroxy-2β-(*m*-methoxybenzyl)pyrrolidine (**9d**). To a cooled (3–5 °C, ice–water bath) solution of the entire sample of **9d** in 50 mL of CH_2Cl_2 containing 5 mL of pyridine was added (dropwise, 5 min) 1.00 mL (7.29 mmol) of 2,2,2-trichloroethyl chloroformate. After 30 min, the CH_2Cl_2 was removed at reduced pressure (88 torr), and the residue was treated in situ with a solution of 20 mL of pyridine containing 1.50 mL (15.9 mmol) of acetic anhydride. After 18 h the solution was poured onto ice and extracted with CH_2Cl_2. The organic phase was separated, dried ($MgSO_4$), and filtered, and the filtrate was concentrated at reduced pressure (88 torr). The residue (**10d**) was dissolved in 50 mL of acetic acid–THF (1:4), and 7.5 g of zinc powder was added. After stirring for 1 h, the slurry was filtered, and the filtrate was adjusted to pH 9 with 2 N NaOH. The solution was then extracted with CH_2Cl_2, and the organic phase was concentrated at reduced pressure (88 torr). To the residue was added 20 mL of THF, followed by 2.00 mL (13.0 mmol) of DBU;[12] after the mixture was stirred for 1 h, the THF was removed at reduced pressure (88 torr). Column chromatography of the residue on 150 g of silica gel and elution with toluene–CH_2Cl_2–MeOH–MeOH saturated with NH_3 gas (32:8:3:1) yielded 680 mg (2.57 mmol, 35%) of **1d** as a white solid, which crystallized from EtOAc: mp 102–103 °C; IR (film) 3340, 3600–3025, 3000, 2945, 2875, 2840, 1740, 1605, 1590, 1495, 1460, 1440, 1380, 1250, 1160, 1040, 965, 790, 700 cm^{-1}; UV (MeOH) λ_{max} 216 nm (ϵ 7530), 272 (2140); ^1H NMR (100 MHz) δ 7.36–7.10 (1 H, complex m), 6.90–6.70 (3 H, complex m), 4.76 (1 H, dd, J = 5 and 1.5 Hz), 4.20 (1 H, ddd, J = 7, 5, and 1.5 Hz), 3.79 (3 H, s), 3.45 (1 H, dd, J = 11.5 and 6.5 Hz) superimposed on 3.7–3.3 (1 H, complex m), 2.84 (2 H, d, J = 6 Hz) superimposed on 2.74 (1 H, dd, J = 11 and 5 Hz), 2.50 (2 H, broad s, exchanges with D_2O), 2.14 (3 H, s); mass spectrum, m/z (relative intensity) 266 (M^+ + 1, 0.2), 162 (2), 144 (30), 126 (12), 121 (9), 91 (11), 84 (100), 43 (16). Anal. ($C_{14}H_{19}NO_4$) C, H, N.

3β-Acetoxy-4α-hydroxy-2β-(*o*-methoxybenzyl)pyrrolidine (1e). Treatment of 3.80 g (10.0 mmol) of **6e** as described for **7g** yielded 1.33 g (6.50 mmol, 65%) of **7e** as a colorless oil. A solution of 1.30 g (6.30 mmol) of **7e** and 1.30 g of sodium trifluoroacetate in 50 mL of trifluoroacetic acid was cautiously lowered into a hot oil bath (ca. 120 °C) so that boiling started immediately.[17] After 16 h of refluxing, the solution was allowed to cool, and the solvent was then removed at reduced pressure (88 torr). Tetrahydrofuran (50 mL) was added to the residue, and a solution of 10% Na_2CO_3 was added to adjust the solution to pH 9. Crystalline 9-fluorenylmethyl 4,6-dimethyl-2-pyrimidylthiocarbonate[16] (2.90 g, 8.00 mmol) was slowly added to the mixture, which was then heated at 45 °C for 1 h. After cooling, the mixture was extracted with CH_2Cl_2. The organic phase was separated, dried ($MgSO_4$), and filtered, and the filtrate was concentrated at reduced pressure (88 torr). Following column chromatography on 150 g of silica gel and elution with toluene–CH_2Cl_2–MeOH–MeOH saturated with NH_3 gas (32:8:3:1) yielded *N*-[(fluorenylmethoxy)-carbonyl]-3β,4α-dihydroxy-2β-(o-methoxybenzyl)pyrrolidine (**9e**). To a cooled (3–5 °C, ice–water bath) solution of the entire sample of **9e** in 50 mL of CH_2Cl_2 containing 5 mL of pyridine was added (dropwise, 5 min) 0.95 mL (6.90 mmol) of 2,2,2-trichloroethyl chloroformate. After 30 min, the CH_2Cl_2 was removed at reduced pressure (88 torr), and the residue was treated in situ with a solution of 20 mL or pyridine containing 1.50 mL (15.9 mmol) of acetic anhydride. After 18 h the solution was poured onto ice and extracted with CH_2Cl_2. The organic phase was separated, dried ($MgSO_4$), and filtered, and the filtrate was concentrated

(17) See ref 1b, footnote 19.

at reduced pressure (88 torr). The residue (10e) was dissolved in 50 mL of acetic acid–THF (1:4) and 6.5 g of zinc powder was added. After the mixture was stirred for 1 h, the slurry was filtered, and the filtrate was adjusted to pH 9 with 2 N NaOH. The solution was then extracted with CH_2Cl_2, and the organic phase was concentrated at reduced pressure (88 torr). To the residue was added 20 mL of THF, followed by 2.00 mL (13.0 mmol) of DBU;[12] after the mixture was stirred for 1 h, the THF was removed at reduced pressure (88 torr). Column chromatography of the residue on 150 g of silica gel and elution with toluene–CH_2Cl_2–MeOH–MeOH saturated with NH_3 gas (32:8:3:1) yielded 450 mg (1.70 mmol, 27%) of 1e as an off-white amorphous solid: IR (film) 3330, 3600–3025, 3005, 2940, 2840, 1740, 1605, 1590, 1500, 1470, 1440, 1380, 1250, 1035, 760 cm^{-1}; UV (MeOH) λ_{max} 221 nm (ϵ 7410), 271 (2240), 277 (2140); ^1H NMR (100 MHz) δ 7.3–7.0 (2 H, complex overlapping multiplets), 7.0–6.7 (2 H, complex overlapping multiplets), 4.76 (1 H, dd, J = 4.5 and 1.5 Hz), 4.19 (1 H, ddd, J = 6, 4.5, and 1.5 Hz), 3.81 (3 H, s) and 3.44 (1 H, dd, J = 11.5 and 7 Hz) are superimposed on 3.92–3.28 (1 H, complex m), 2.95–2.62 (3 H, complex overlapping multiplets), 2.54 (2 H, broad s, exchanges with D_2O), 2.14 (3 H, s); mass spectrum, m/z (relative intensity) 266 (M^+ + 1, 0.3), 162 (1), 150 (2), 144 (27), 126 (11), 121 (8), 91 (17), 84 (100), 43 (20). Anal. ($C_{14}H_{19}NO_4$) C, H, N.

3β-Acetoxy-4α-hydroxy-2β-(α-methyl-*p*-methoxybenzyl)pyrrolidine (1f). Treatment of 2.40 g (6.09 mmol) of **6f** as described for **7g** yielded 1.10 g (5.02 mmol, 82%) of **7f** as a colorless oil. A solution of 1.50 g (6.80 mmol) of **7f** and 1.50 g of sodium trifluoroacetate in 50 mL of trifluoroacetic acid was cautiously lowered into a hot oil bath (ca. 120 °C) so that boiling started immediately.[17] After 16 h of refluxing, the solution was allowed to cool, and the solvent was then removed at reduced pressure (88 torr). Tetrahydrofuran (50 mL) was added to the residue, and a solution of 10% Na_2CO_3 was added to adjust the solution to pH 9. Crystalline 9-fluorenylmethyl 4,6-dimethyl-2-pyrimidylthiocarbonate[16] (2.77 g, 7.65 mmol) was slowly added to the mixture, which was then heated at 45 °C for 1 h. After cooling, the mixture was extracted with CH_2Cl_2. The organic phase was separated, dried ($MgSO_4$), and filtered, and the filtrate was concentrated at reduced pressure (88 torr). Column chromatography on 150 g of silica gel and elution with toluene–CH_2Cl_2–MeOH–MeOH saturated with NH_3 gas (32:8:3:1) yielded *N*-fluorenylmethoxycarbonyl-3β,4α-dihydroxy-2β-(α-methyl-*p*-methoxybenzyl)pyrrolidine (**9f**). To a cooled (3–5 °C, ice–water bath) solution of the entire sample of **9f** in 50 mL of CH_2Cl_2 containing 5 mL of pyridine was added (dropwise, 5 min) 0.85 mL (6.20 mmol) of 2,2,2-trichloroethyl chloroformate. After 30 min, the CH_2Cl_2 was removed at reduced pressure (88 torr), and the residue was treated in situ with a solution of 20 mL of pyridine containing 1.50 mL (15.9 mmol) of acetic anhydride. After 18 h the solution was poured onto ice and extracted with CH_2Cl_2. The organic phase was separated, dried ($MgSO_4$), and filtered, and the filtrate was concentrated at reduced pressure (88 torr). The residue (**10f**) was dissolved in 50 mL of acetic acid–THF (1:4), and 6.5 g of zinc powder was added. After the mixture was stirred for 1 h, the slurry was filtered, and the filtrate was adjusted to pH 9 with 2 N NaOH. The solution was then extracted with CH_2Cl_2, and the organic phase was concentrated at reduced pressure (88 torr). To the residue was added 20 mL of THF, followed by 2.00 mL (13.0 mmol) of DBU;[12] after the mixture was stirred for 1 h, the THF was removed at reduced pressure (88 torr). Column chromatography of the residue on 150 g of silica gel and elution with toluene–CH_2Cl_2–MeOH–MeOH saturated with NH_3 gas (32:8:3:1) yielded 700 mg (2.50 mmol, 36%) of a 1:1 mixture of two diastereomers (at the α-benzyl carbon) of **1f**. The first diastereomer eluted from the column (ca. 18%) was not completely pure, while the second (ca. 18%) was obtained as a white solid, which crystallized from EtOAc: mp 172–173 °C; IR (KBr) 3325, 3550–3000, 2965, 2925, 2875, 1740, 1620, 1520, 1470, 1455, 1380, 1240, 1100, 1045, 980, 900, 850, 830, 790, 740 cm^{-1}; UV (MeOH) λ_{max} 223 nm (ϵ 10515), 275 (2225), 282 (1865); ^1H NMR (100 MHz) δ 7.08 (2 H, d with further fine splitting, J = 8.8 Hz), 6.79 (2 H, d with further fine splitting, J = 8.8 Hz), 4.31 (1 H, dd, J = 4 and 0.7 Hz), 4.08 (1 H, ddd, J = 6, 4, and 1.5 Hz), 3.77 (3 H, s), two overlapping dd at 3.43 (1 H, J = 11 and 6 Hz) and 3.41 (1 H, J = 10 and 4 Hz), 2.81 (1 H, dd, J = 11 and 4 Hz) superimposed on 3.08–2.65 (1 H, complex m), 2.35 (2 H, broad s, exchanges with D_2O), 2.03 (3 H, s), 1.33 (3 H, d, J = 6.9 Hz); mass spectrum, m/z (relative intensity) 280 (0.2), 279 (M^+, 0.2), 162 (1), 144 (30), 135 (10), 126 (14), 121 (5), 105 (5), 91 (5), 84 (100), 43 (13). Anal. ($C_{15}H_{21}NO_4$) C, H, N.

3β-Acetoxy-4α-hydroxy-2β-(α-phenyl-*p*-methoxybenzyl)pyrrolidine (1g) and 4β-Acetoxy-3α-hydroxy-2β-(α-phenyl-*p*-methoxybenzyl)pyrrolidine (11). A solution of 2.00 g (7.11 mmol) of **7g** and 2.00 g of sodium trifluoroacetate in 50 mL of trifluoroacetic acid was cautiously lowered into a hot oil bath (ca. 120 °C) so that boiling started immediately.[17] After 16 h of refluxing, the solution was allowed to cool, and the solvent was then removed at reduced pressure (88 torr). Tetrahydrofuran (50 mL) was added to the residue, and a solution of 10% Na_2CO_3 was added to adjust the solution to pH 9. Crystalline 9-fluorenylmethyl 4,6-dimethyl-2-pyrimidylthiocarbonate[16] (3.20 g, 8.70 mmol) was slowly added to the mixture, which was then heated at 45 °C for 1 h. After cooling, the mixture was extracted with CH_2Cl_2. The organic phase was separated, dried ($MgSO_4$), filtered, and concentrated at reduced pressure (88 torr). Following column chromatography on 150 g of silica gel and elution with toluene–CH_2Cl_2–MeOH–MeOH saturated with NH_3 gas (32:8:3:1) yielded a 1:1 *N*-fluorenylmethoxycarbonyl *trans*-diol mixture. To a cooled (3–5 °C, ice–water bath) solution of the entire sample in 50 mL of CH_2Cl_2 containing 5 mL of pyridine was added (dropwise, 5 min) 1.00 mL (7.29 mmol) of 2,2,2-trichloroethyl chloroformate. After 30 min, the CH_2Cl_2 was removed at reduced pressure (88 torr), and the residue was treated in situ with a solution of 20 mL of pyridine containing 2.00 mL (21.2 mmol) of acetic anhydride. After 18 h the solution was poured onto ice and extracted with CH_2Cl_2. The organic phase was separated, dried ($MgSO_4$), and filtered, and the filtrate was concentrated at reduced pressure (88 torr). The residue was dissolved in 50 mL of acetic acid–THF (1:4), and 10.0 g of zinc powder was added. After the mixture was stirred for 1 h, the slurry was filtered, and the filtrate was adjusted to pH 9 with 2 N NaOH. The solution was then extracted with CH_2Cl_2, and the organic phase was concentrated at reduced pressure (88 torr). To the residue was added 20 mL of THF, followed by 2.00 mL (13.0 mmol) of DBU;[12] after the mixture was stirred for 1 h the THF was removed at reduced pressure (88 torr). Column chromatography of the residue on 150 g of silica gel and elution with toluene–CH_2Cl_2–MeOH–MeOH saturated with NH_3 gas (32:8:3:1) yielded first 500 mg (1.44 mmol, 20%) of 11 as a colorless oil and then 500 mg (1.44 mmol, 20%) of **1g** as a white amorphous solid. Compound 11: IR (film) 3325, 3600–3000, 3055, 3025, 3000, 2930, 2830, 1730, 1610, 1580, 1515, 1450, 1370, 1245, 1180, 1035, 755, 700 cm^{-1}; UV (MeOH) λ_{max} 230 nm (ϵ 13715), 261 (1545), 268 (1790), 276 (1840), 283 (1465); ^1H NMR (79.5 MHz) δ 7.27 (5 H, broad s) superimposed on 7.47–7.09 (2 H, m), 6.79 (2 H, d with further fine splitting, J = 9 Hz), 4.88 (1 H, superficial quintet, J = 3 Hz), 3.75 (3 H, s) superimposed on 4.28–3.50 (3 H, complex overlapping multiplets), 3.50–2.95 (1 H, m), 2.74 (1 H, dd, J = 13 and 3 Hz), singlets at 2.05 and 2.04 (3 H) superimposed on 2.15–1.75 (2 H, broad s, exchanges with D_2O); mass spectrum, m/z (relative intensity) 342 (M^+ + 1, 0.2), 226 (2), 197 (10), 165 (8), 153 (6), 144 (43), 126 (7), 84 (100), 43 (11). Anal. ($C_{20}H_{23}NO_4$) H, N; C: calcd, 70.36; found, 69.49. Compound **1g**: IR (film) 3350, 3500–3000, 3065, 3030, 2930, 2875, 2860, 1725, 1670, 1615, 1520, 1460, 1380, 1310, 1250, 1040, 975, 880, 840, 740, 710 cm^{-1}; UV (MeOH) λ_{max} 230 nm (ϵ 11965), 261 (1300), 268 (1465), 276 (1515), 283 (1300); ^1H NMR (100 MHz) δ 7.26 (5 H, s) superimposed on 7.5–7.1 (2 H, complex m), 6.95–6.70 (2 H, complex m), 4.55 (1 H, m, $w_{1/2}$ = 5 Hz), 4.11 (1 H, d, J = 2.3 Hz) superimposed on 4.28–4.04 (2 H, complex m), 3.75 and 3.74 (3 H, s), 3.46 (1 H, dd, J = 11 and 7 Hz), 2.80 (1 H, dd, J = 11 and 5 Hz), 2.02 and 1.98 (3 H, s) superimposed on 2.08 (2 H, broad s, exchanges with D_2O); mass spectrum, m/z (relative intensity) 342 (M^+ + 1, 0.4), 197 (13), 165 (9), 153 (9), 144 (71), 126 (26), 84 (100), 56 (7), 55 (7), 43 (18). Anal. ($C_{20}H_{23}NO_4$) C, H, N.

Antimicrobial and Antiprotozoal Activity. Bacterial Strains. *Staphylococcus aureus, Bacillus subtilus, Mycobacterium fortuitum, Streptomyces griseus, Nocardia asteroides, Escherichia coli, Salmonella schottmuelleri,* and *Pseudomonas aeruginosa* were grown in Mueller–Hinton broth and tested in broth or agar at 37 °C, except for *S. griseus*, which was grown

and tested at 28 °C. *Streptococcus pyogenes C, Actinomyces israelii, Clostridium novyi, Propionibacterium acnes*, and *Bacteriodes fragilis* were grown in thioglycolate broth and tested in this broth or on brain–heart infusion agar, anaerobically at 37 °C.

Fungal Strains. *Candida albicans, Saccharomyces cerevisiae, Trichophyton mentagrophytes*, and *Epidermophyton floccosum* were grown in Sabouraud's dextrose broth and tested in broth or agar at 37 °C, except for *T. mentagrophytes* and *E. floccosum*, which were grown and tested at 28 °C.

Protozoal Strains. *Trichomonas vaginalis* was grown in Tripticase yeast maltose basal medium (TYM) 721, *T. foetus* in TYM 359, and *Entamoeba histolytica* (J 190) in modified Boeck Drbohlav medium, all at 37 °C.

Preparation of Drugs. Stock solutions of the anisomycins **1a–g** were prepared at a concentration of 1 mg/mL for agar diffusion and at 3.2 mg/mL for tube dilution testing. Me_2SO–EtOH (1:9) was used to solubilize the compounds.

Determination of Antibacterial and Antifungal MIC and Zone Size. Agar diffusion (zone size, millimeter diameter) and tube dilution (minimum inhibitory concentrations, MICs) tests were done in the appropriate media cited above. Inocula for seeding agar plates and tubes were obtained from 24-h bacteria and yeast cultures, 48-h anaerobe cultures, and 4–5 day dermatophyte cultures. Inocula for seeding agar plates was used as a 1:100 dilution in agar. Inocula for MICs was 0.05 mL of undiluted culture for *S. pyogenes C, A. israelii, C. novyi, P. acnes*, and *B. fragilis*; a 1:10 dilution in saline for *T. mentagrophytes* and *E. floccosum*; a 1:100 dilution in saline for *M. fortuitum, S. griseus*, and *N. asteroides*; and a 1:1000 dilution in saline for the rest of the organisms. For agar diffusion tests, a solution containing 0.05 mg of the drug was absorbed onto paper disks, placed on the agar, and incubated. For MICs (which used ca. 3 mL of broth/tube), the drug concentrations ranged from 32 to 0.06 μg/mL with twofold serial dilutions. The concentration of Me_2SO–EtOH used showed no inhibitory effect on any of the test organisms. Zone sizes and MICs were determined after incubation for 24 h for bacteria and 48 h for fungi. The MIC was defined as the lowest concentration of drug at which no visible bacterial or fungal growth was observed.

Determination of Antiprotozoal Activity. Protozoa were grown in appropriate media cited above for 48 h and then pooled. Tubes containing 10 mL of media and drug concentrations ranging from 32 to 0.06 μg/mL were inoculated with 0.6×10^6 organisms and incubated for 48 h. Drug-treated tubes were compared with untreated controls and scored, based on microscopic examination through the tube, as either 100, >95, 95 to 25%, or <25% inhibition.

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Registry No. (–)-**1a**, 22862-76-6; (±)-**1a**, 21497-40-5; (±)-**1b**, 82892-50-0; (±)-**1c**, 82892-51-1; (±)-**1d**, 84109-88-6; (±)-**1e**, 84109-89-7; (±)-**1f** (isomer 1), 84109-90-0; (±)-**1f** (isomer 2), 84110-19-0; **1g**, 84109-91-1; **2d**, 79499-36-8; **2e**, 79499-35-7; (±)-**2f**, 84110-05-4; (±)-**2g**, 84110-07-6; (±)-**3d**, 84109-92-2; (±)-**3e**, 84109-93-3; (±)-**3e**·HCl, 84110-04-3; **3f**, 84109-94-4; **3f**·HCl, 84110-06-5; **3g**, 84109-95-5; (±)-**4d**, 84109-96-6; (±)-**4e**, 84109-97-7; **4f**, 84109-98-8; **4g**, 84117-58-8; (±)-**5d**, 84110-08-7; (±)-**6d**, 84109-99-9; (±)-**6e**, 84110-00-9; **6f**, 84110-01-0; **6g**, 84110-02-1; (±)-**7d**, 84110-09-8; (±)-**7e**, 84110-13-4; **7f**, 84110-16-7; **7g**, 84110-03-2; (±)-**9d**, 84110-11-2; (±)-**9e**, 84110-14-5; **9f**, 84110-17-8; (±)-**10d**, 84110-12-3; (±)-**10e**, 84110-15-6; **10f**, 84110-18-9; **11**, 84117-57-7; 2,2,2-trichloroethyl chloroformate, 17341-93-4; *N*-iodosuccinimide, 516-12-1; 9-fluorenylmethyl 4,6-dimethyl-2-pyrimidylthiocarbonate, 84110-10-1.

Quantitative Structure–Activity Relationships for 2-[(Phenylmethyl)sulfonyl]pyridine 1-Oxide Herbicides

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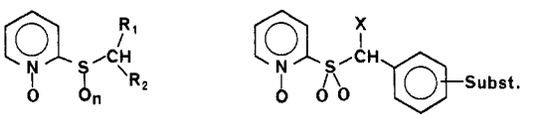
Phenyl-substituted analogues of 2-[(phenylmethyl)sulfonyl]pyridine 1-oxide preemergent herbicides were examined in order to determine quantitative relationships between structure and activity against the following three weed species: switch grass (*Panicum virgatum* L.), barnyard grass (*Echinochloa crusgalli* L. Beauv.), and green foxtail (*Setaria viridis* L. Beauv.). Analogues were chosen to provide maximum parameter orthogonality. Regression analysis yielded structure–activity relationships wherein the most significant substituent parameters associated with herbicidal activity were found to be the partition coefficient (π), the molar refractivity (MR), and two indicator variables, Z (denoting the presence of an α-methyl group) and H (denoting an ortho substituent capable of hydrogen bonding). For green foxtail, the structure–activity relationship was found to be: $-\log ED50 = 0.43\pi - 0.052MR + 0.50H + 0.24Z + 0.61$, where ED50 is expressed in moles per acre. The regression equations were found to explain 79–93% of the bioactivity for the three weed species studied. It was further shown that these equations represent the best possible correlations within the limitations of the biological data.

For some time we have been interested in demonstrating the utility of QSAR (quantitative structure–activity relationships) in our crop-protection research programs. This study was undertaken to prove the utility of in vivo whole plant biodata as a resource for meaningful and predictive QSAR equations.

The class of preemergent herbicides of general formula 1 was recently patented.[1,2] Highly active members of the series are effective control agents for a wide variety of weed



1 n = 1,2; R_1 = H, CH_3; R_2 = alkyl, aryl

2 X = H, CH_3

species, e.g., yellow nutsedge (*Cyperus esculentus* L.), barnyard grass (*Echinochloa crusgalli* L. Beauv.), switch grass (*Panicum virgatum* L.), and Johnson grass (*Sorgum halepense* L. Pets.). Although many subclasses were synthesized to fully explore structure–activity dependencies, the present study was directed to the structural

(1) H. L. Plant and A. R. Bell, U.S. Patent 3960542 (1976).
(2) H. L. Plant, J. W. Zukel, and A. R. Bell, U.S. Patent 4019893 (1977).

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