30052) for financial support. We thank also Professor Gilbert Stork for providing experimental details for preparation of the allylic–homoallylic dihalide used in Scheme III[8b] and Dr. Edward Asirvatham for a helpful suggestion concerning use of pentenolide sulfoxide **5b**. We thank Dr. Alan Chalk of the Givaudan Corporation for a sample of natural (−)-β-vetivone and Professor Alex Nickon of this department for temporary use of his GC equipment.

Chiral Synthesis of Bicyclomycin and Diastereomeric Stereoselectivity of the Key Aldol Condensation

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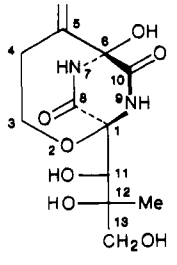
Optically pure bicyclomycin (**1**) was synthesized via aldol condensation of racemic 7,9-bis(*p*-methoxybenzyl)-5-methylene-6-[(*tert*-butyldimethylsilyl)oxy]-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (**2**) with 2,3-di-*O*-isopropylidene-2-*C*-methyl-L-glyceraldehyde (**3**). The major condensation product **4** was then N-de(*p*-methoxybenzyl)ated and O-deisopropylidenated simultaneously with CAN and O-de(*tert*-butyldimethylsilyl)ated with Bu_4NF under finely optimized conditions, respectively, to give **1**. The structures of three other diastereomers of **4** were elucidated through comparison with the products of the aldol condensation of optically pure **2** and **3**. The compounds (+)-**2** and (−)-**2** were prepared by diastereomeric separation of the synthetic precursor of **2**, i.e., 5,6-dihydroxy-7,9-bis(*p*-methoxybenzyl)-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione as its (−)-MTPA ester, followed by the previously established four-step conversion. The stereoselectivity of the aldol condensation was explained by the chair conformation-like transition states.

Bicyclomycin (**1**), an antibiotic isolated from cultures of *Streptomyces sapporonensis*[1] and *S. aizunensis*[2] has unique antibacterial activity[3] against some Gram-negative microorganisms and has been produced by a Japanese pharmaceutical company. The relative[4] and absolute[5] structure of **1** was established by X-ray analysis. The most remarkable structural characteristic of **1** is a highly oxidized, bicyclic 2,5-piperazinedione (BPD) framework, which has prompted many strategies for the total synthesis of **1**. Three groups have reported the synthesis of racemic[6] and chiral (78% ee[7] and 100% ee[8]) bicyclomycin. In this paper, we report the details of the preliminary communication[8] on the chiral synthesis of **1**.



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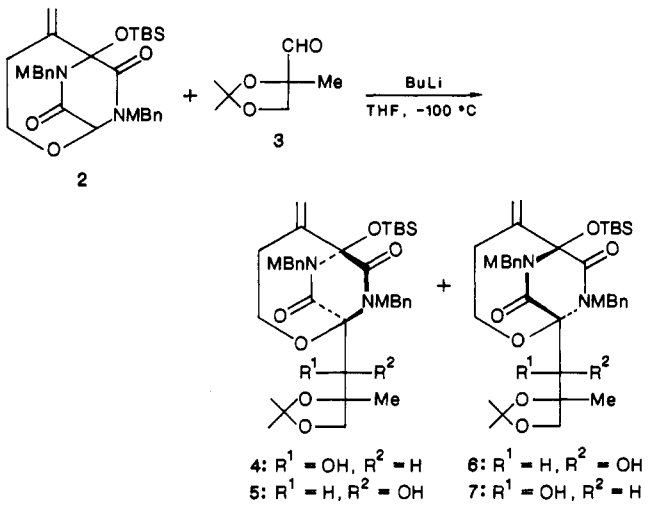
Results and Discussion

Aldol Condensation of 2 with 3. In the course of these total syntheses,[6-8] the C–C coupling of the BPD bridgehead carbanion with the branched-chain aldehyde is a common key step. In our synthesis, 7,9-bis(*p*-methoxybenzyl)-5-methylene-6-[(*tert*-butyldimethylsilyl)oxy]-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (**2**)[9] was chosen as the BPD derivative and 2,3-di-*O*-isopropylidene-2-*C*-methyl-L-glyceraldehyde (**3**)[10] as the carbonyl component for the

Scheme I



2 **3**

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0022-3263/88/1953-6035$01.50/0

Table I. ^{1}H NMR Spectral Data of Diastereomeric Aldol Condensation Products at 500 MHz

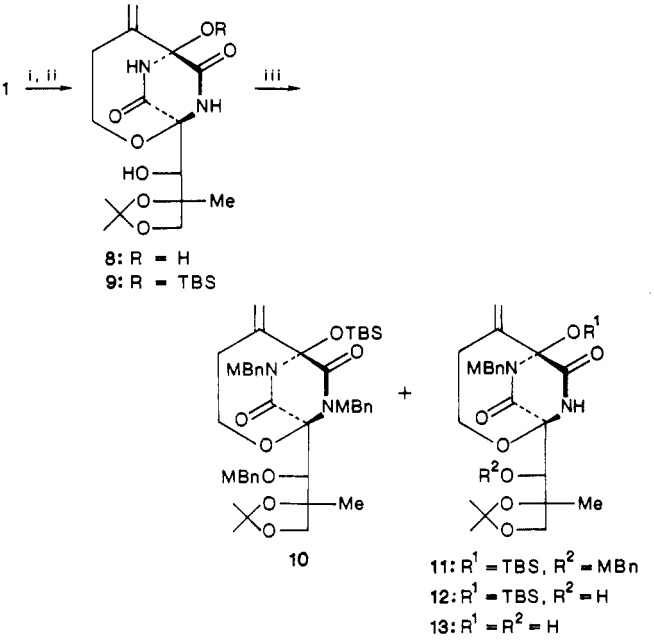
protons	4	5	6	7
H-11	4.64 d (9.8)	4.62	4.34 d (11.1)	5.08 br s
C_{11}-OH	6.40 d	3.17 (6.4)	5.00 d (11.1)	5.10
C_{12}-CH_3	0.99 s	0.89 s	1.06 s	1.20 s
H-13a	3.78 d (8.9)	3.71 d (8.8)	3.37 d (8.3)	(3.7) [3.72 d (8.9)][b]
H-13b	4.06 d	4.13 d (8.5)	3.99 d (8.3)	(3.8)
H-3a	2.97 m	3.20 br	3.22 dd (9.4, 13.3)	3.45 br
H-3b	3.41 ddd (3.2, 6.3, 13.5)	3.60 br	3.70 dd (7.7, 13.3)	(4.13 br)
H-4a, H-4b	1.76–1.84 m	1.95 br	1.94 dd (9.4, 16.6)	2.05
		2.10 br	2.07 dd (7.7, 16.6)	2.25 br
H-5′a	4.88 s	5.00 s	4.97 s	5.08 br s [5.03 br s][b]
H-5′b	5.44 d (0.6)	5.50 s	5.48 s	5.54 s[a]
MBn OCH_3	3.77 s	3.77 s	3.76 s	3.76 br s[a]
	3.78 s	3.79 s	3.79 s	3.80 s
CH_2	4.40, 4.63 AB q (14.0)	4.49, 4.63 AB q (14.3)	4.55, 4.65 AB q (14.2)	4.54,[a] 4.58 AB q (14.5)
	4.58, 5.08 AB q (15.3)	4.63, 5.26 AB q (15.3)	4.64, 4.69 AB q (15.0)	4.37,[b] 5.10
aromatic	6.77 d, 7.29 d (8.6)	6.78 d, 7.23 d (8.6)	6.76 d, 7.33 d (8.6)	6.78,[d] 7.43 d[a] (6.8)
	6.79 d, 7.48 d (8.9)	6.83 d, 7.40 d (8.4)	6.82 d, 7.41 d (8.6)	6.87 br s, 7.16 br s [6.86 d, 7.18 d (7.9)][b]
Isp CCH_3	1.36 s	1.26 s	1.21 s	1.39 s
	1.41 s	1.38 s	1.34 s	1.49 br s[a]
TBS $SiCH_3$	0.28 s	0.18 s	0.19 s	0.11 br s[a] [0.13 s][b]
	0.45 s	0.41 s	0.44 s	0.38 br s[a]
CCH_3	0.92 s	0.85 s	0.90 s	0.80 br s[a] [0.82 s][b]

[a] These peaks become remarkably sharpter at 45 °C. [b] Clearly shifted signals at the elevated temperature.

condensation. Formation of the carbanion of **2** with butyllithium followed by aldol condensation with **3** at −100 °C gave a mixture of diastereomers, which showed on silica gel TLC (ether) three spots with R_f values, 0.61, 0.56, and 0.51. The approximate ratio of these three spots was 3:1:3 on the basis of the isolated yields. The component with the highest R_f value proved to have the same configurations, i.e., 1*S*, 6*R*, and 11*S* (**4**), as those of natural bicyclomycin by chemical conversion of **1** into **4** as described in the next paragraph. The configurations of the component with the lowest mobility were ascertained to be 1*R*, 6*S*, and 11*R* (**6**), by NOE experiments (see below). The middle spot proved to be a mixture of two minor components having 1*S*, 6*R*, and 11*R* (**5**) and 1*R*, 6*S*, and 11*S* (**7**) configurations, respectively. In order to elucidate the stereochemistry of these three unnatural diastereomers, the coupling reactions of **3** with chiral **2**, which reduces the possible number of diastereomers, was helpful. The ratio of four diastereomers, **4**, **5**, **6**, and **7**, was estimated by the intensity of 500-MHz ^{1}H NMR signals utilizing the differences in chemical shifts of Si-methyl and C_{12}-methyl groups (Table I) to be 15:2:13:3.

Preparation of 4 from Natural 1. Treatment of **1** with 2-methoxypropene and pyridinium *p*-toluenesulfonate (PPTS) gave 12,13-*O*-isopropylidene derivative **8** quantitatively. The aminal hydroxyl group of **8** was selectively silylated in *N*,*N*-dimethylformamide (DMF) with chloro-*tert*-butyldimethylsilane and imidazole to afford the 6-*O*-*tert*-butyldimethylsilyl derivative **9** in 96% yield. The secondary hydroxyl group at C-11 may be sterically hindered and could not be silylated. This free hydroxyl group can be easily confirmed by a doublet at 4.02 in the ^{1}H NMR spectrum, which is characteristic for the bi-

Scheme II[a]



[a] Reagents and conditions: (i) $Me_2C(OMe)_2$, PPTS, Me_2CO, room temperature; (ii) TBSCl, imidazole, DMF, room temperature; (iii) MBnBr, NaH, NaH, DMF, 0 °C.

cyclomycin derivatives with a free hydroxyl at C-11. Methoxybenzylation of **9** with 3.5 equiv of benzyl bromide and sodium hydride in DMF gave the N^7,N^9,*O*-tris(*p*-methoxybenzyl) derivative **10** and the N^7,*O*-bis(*p*-methoxybenzyl) derivative **11**, in 37% and 50% yield, respectively. Oxidative removal of the *O*-(*p*-methoxybenzyl) group of **10** with 2,3-dichloro-5,6-dicyanobenzoquinone afforded, in 80% yield, **4**, which was identical with one of the major diastereomers obtained by the coupling of **2** with **3**. A similar selective O-de(*p*-methoxybenzyl)ation of **11** gave **12** in 82% yield.

Aldol Condensation with Chiral BPD. Chiral BPD (+)-**2** and (−)-**2** were prepared via optical resolution of 5,6-dihydroxy-7,9-bis(*p*-methoxybenzyl)-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (a racemic mixture of **14** and **19**) as the (*S*)-(−)-2-methoxy-2-phenyl-3,3,3-trifluoropropionic acid[11] [(*S*)-(−)-MTPA] esters **15** and **20**, followed

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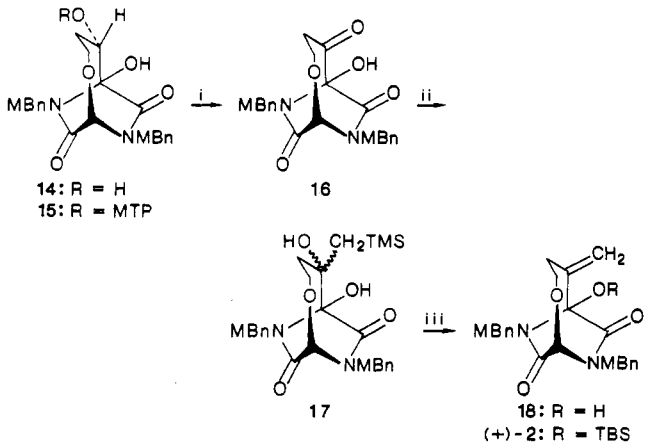
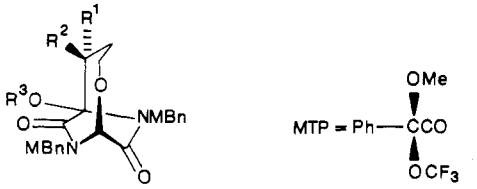
(10) Yoshimura, J.; Hara, K.; Yamaura, M.; Mikami, K.; Hashimoto, H. *Bull. Chem. Soc. Jpn.* **1982**, *55*, 933.

Scheme III[a]



[a] Reagents and conditions: (i) DMSO, $(COCl)_2$, CH_2Cl_2, −78 °C; (ii) $TMSCH_2MgCl$, Et_2O, −30 °C; (iii) $(CF_3CO)_2O$, DMAP, CH_2Cl_2, room temperature—Bn_4NCl, KF, CH_3CN, 40 °C—MeOH.

by Swern oxidation and modified Peterson olefination as reported previously.[9] A separable mixture of diastereomers[12] **15** and **20** was prepared by treatment of a racemic mixture of **14** and **19** with the acid chloride[11] of (*S*)-(−)-MTPA in the presence of 4-(dimethylamino)pyridine in 90% yield. Chemical conversions described later identified the diastereomer with the lower melting point (107.5–108 °C) as **15** (1*R*,6*S*) and the other diastereomer (mp 174.5 °C) as **20** (1*S*,6*R*). A conventional deacylation of **15** and **20** with sodium methoxide gave an enantiomeric pair, **14** and **19**, respectively. Enantiomers **14** and **19** were



19: R^1 = OH, R^2 = R^3 = H
20: R^1 = OMTP, R^2 = R^3 = H
21: R^1, R^2 = O, R^3 = H
22: R^1, R^2 = H, CH_2TMS, R^3 = H
23: R^1, R^2 = CH_2, R^3 = H
(−)-**2**: R^1, R^2 = CH_2, R^3 = TBS

converted to (1*S*,6*R*)-(+)-**2** and (1*R*,6*S*)-(−)-**2** by methylenation as described above and *tert*-butyldimethylsilylation. It is noteworthy that the loss of asymmetry at C-5 reversed the sign of optical rotation. Thus, **14** ($[\alpha]_D$ −45.1°) was converted into **16** ($[\alpha]_D$ +47.0°) and then into (+)-**2** ($[\alpha]_D$ +15.9°), while **19** ($[\alpha]_D$ +46.1°) was transformed into **21** ($[\alpha]_D$ −47.4°) and then into (−)-**2** ($[\alpha]_D$ −15.5°).

The coupling of each chiral BPD, (+)-**2** and (−)-**2**, with **3** was performed in the same manner as described for racemic BPD **2**. In the case of (+)-**2**, a 4:1 mixture of **4** and **5** was obtained in 64% yield, while in the case of (−)-**2**, a 2.2:1 mixture of **6** and **7** was obtained in a similar yield. The second major product with the lowest R_f value in the coupling of racemic **2** with **3** and the major product in the coupling of (−)-**2** with **3** proved to be identical and have

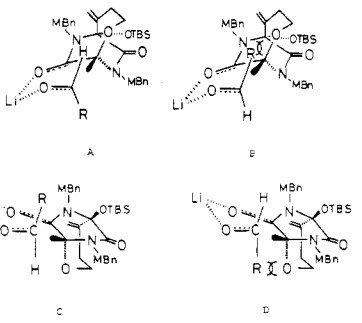


Figure 1.

Figure 2.

Table II. NOE Experiments[a] of Compounds 4 and 6

compds	NOE observed[b]
4	H-11 ⇄ H-13b, 11-OH ⇄ 12-CH_3, C_{11}-OH ⇄ H-13b
6	H-11 ⇄ 12-CH_3, H-11 ⇄ H-13b, 11-OH ⇄ H-13b, 11-OH ⇄ Ispb

[a] The following protons were irradiated: H-11, 11-OH, 12-CH_3, H-13a, H-13b, and Ispb. [b] In all compounds additional NOE was observed between the following protons: H-11 ⇄ 11-OH, 12-CH_3 ⇄ H-13a, 12-CH_3 ⇄ Ispa, H-13a ⇄ H-13b, H-13b ⇄ Ispb.

1*S*,6*R*, and 11*R* configurations (**6**). The structure of **6** was ascertained by NOE experiments as shown in the next paragraph. Therefore, the minor components **5** and **7** were deduced to have 1*S*,6*R*,11*R* and 1*R*,6*S*,11*S* configurations, respectively.

These stereochemical results can be explained by the six-membered cyclic transition state accepted generally for the aldol-type reactions of lithium enolates with carbonyl compounds.[13] In the case of the coupling between **3** and (+)-**2**, if a coordination of lithium ion to the carbonyl oxygen of (+)-**2** adjacent to the carbanion and to that of **3** is assumed, two slightly flattened chair conformation-like transition states 1A and 1B (Figure 1) are plausible. In transition state 1B a steric repulsion between the alkyl group (R) of **3** and the BPD ring is expected and transition state 1A is favored, that is, the carbanion attacks the carbonyl from its *re* face to give **4** predominantly. In the case of the coupling between **3** and (−)-**2**, the transition states 1C and 1D (Figure 1) are plausible, where the former is favored by a similar steric argument to give **6** predominantly. Thus, it was found that the stereoselectivity in the addition of the BPD carbanions to the chiral aldehyde **3** changed, depending on the absolute configuration of the BPD ring.

Configuration of C-11 in 6. The NOE experiments of the configurationally determined **4** and the undetermined **6** revealed the configuration of C-11 in **6**. Only the results indicative of the rotational conformation around the C_{11}–C_{12} bond are listed in Table II. In the case of **4**, which has the 11*S* configuration, the NOEs among H-11, C_{11}-OH, and H-13b together with those between H-11 and Ispb and C_{11}-OH and C_{12}-CH_3 supported the conformer depicted in Figure 2A. In the case of **6**, the situation was totally interchanged with respect to H-11 and C_{11}-OH, proving the configuration of C-11 to be *R* as shown Figure 2B.[14]

(11) Dale, J. A.; Dull, D. L.; Mosher, H. S. *J. Org. Chem.* **1969**, *34*, 2543.

(12) Because the ^1H NMR data of a racemic mixture composed of **14** and **19** indicate a single component, the configuration of C-5 was tentatively depicted based only on the mechanistic deduction for the previous dihydroxylation step.[9]

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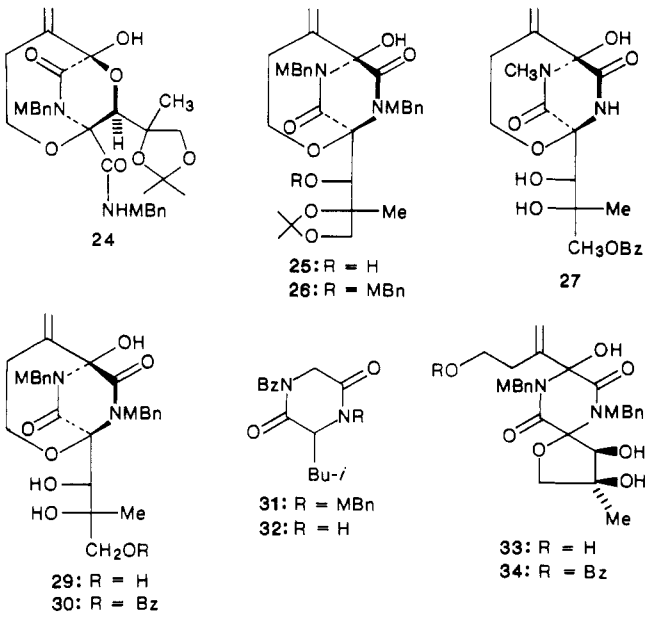
Table III. Treatment of Bicyclomycin Derivatives with Tetrabutylammonium Fluoride

entry	substr	Bu_4NF_4 equiv	Bu_4NF_4 concn, M	temp, °C	time, h	solvent	product (yield, %)
1	**4**	3.0	0.43	25	2	THF	**24** (93)
2	**4**	1.2	0.03	25	6	THF	**24** (90)
3	**4**	1.2[a]	0.02	0	5	CH_2Cl_2	**24** (26), **25** (69)
4	**25**	1.3	0.02	25	0.5	THF	**24** (93)
5	**9**	1.4	0.06	25	0.5	CH_2Cl_2	**8** (89)
6	**9**	1.4[a]	0.06	0	1.5	THF	**8** (95)
7[b]	**12**	3.0	0.27	25	0.25	THF	**13** (61)
8	**12**	1.5	0.04	25	1.5	THF	**13** (91)
9[c]	**28**	1.3[a]	0.03	0	0.5	THF	**1** (90)

[a] Added in three portions with 30-min intervals. [b] With longer reaction time (3 h) no characterizable products could be obtained. [c] The same amount of tetrabutylammonium fluoride was used in a concentration of 0.37 M at 25 °C gave **1** in about 60% yield after 0.5 h.

Chart I



Rearrangement of N-Alkylated Bicyclomycin Derivatives in the Presence of Tetrabutylammonium Fluoride. An attempted O-desilylation of **4** with tetrabutylammonium fluoride gave, instead of the desired **25**, rearranged product **24** in good yield. The structure of **24** was ascertained by a singlet due to H-8 at δ 4.31 instead of a doublet of the corresponding proton (H-11) in the starting material. This result prompted us to examine the conditions of the intramolecular rearrangement, which may place new hurdles to the total synthesis of bicyclomycin. Several related derivatives (**4**, **9**, **12**, **25**, and **28**) were treated with tetrabutylammonium fluoride in tetrahydrofuran or dichloromethane. The results summarized in Table III show the following characteristics of this rearrangement. (i) This intramolecular isomerization via ring opening of the aminal structure proceeds only in the case of N^7,N^9-bis(*p*-methoxybenzyl) derivatives, because under the similar conditions monosubstituted or unsubstituted derivatives did not give the corresponding rearranged products (entries 6–9). A similar substituent effect was observed in another kind of ring opening of the aminal structure on O-deisopropylidenation under acidic conditions as described in ref 15. (ii) Treatment of the O-de(*tert*-butyldimethylsilyl)ated derivative **27** with tetrabutylammonium fluoride gave the same rearranged product in high yield (entry 4). However, treatment of **25** with salts such as tetrabutylammonium chloride and potassium fluoride or with bases such as pyridine and triethylamine did not promote the rearrangement at all, and unchanged **25** was recovered in high yields. Thus, in this rearrangement tetrabutylammonium fluoride plays a very characteristic role. In relation to these experiments, it is noteworthy that the same rearrangement was observed[16] on methylation of a N^7-methylbicyclomycin derivative (**27**) with methyl iodide and potassium carbonate in DMF at 40 °C, where, considering the above results, N-methylation seems to occur before the rearrangement.

In conclusion, addition of tetrabutylammonium fluoride in small portions and lower temperature increased the yield of the O-desilylated derivatives (entries 3, 6, and 9).

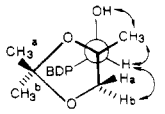
Table IV. N-De(*p*-methoxybenzyl)ation of Bicyclomycin Derivatives

entry	substr	oxidant (equiv)	temp, °C	time, min	product (yield, %)
1	4	CAN[a] (4)	25	25	**28** (49)
2	12	CAN[a] (2.3)	25	25	**28** (54)
3	4	$K_2S_2O_8$[b] (4.3)	60	40	**9** (21)
4	12	$K_2S_2O_8$[b] (2.3)	60	40	**9** (39)

[a] Ceric ammonium nitrate (0.2 M). [b] In the presence of Cu^{2+} and pyridine.

Oxidative N-De(*p*-methoxybenzyl)ation of Bicyclomycin Derivatives. We reported the oxidative deprotection of *N*-*p*-methoxybenzyl groups on the 2,5-piperazinedione derivatives with cerium(IV) ammonium nitrate (CAN).[17] This method was successfully applied to the 6-*O*-(*tert*-butyldimethylsilyl) derivatives of bicyclomycin such as **4** and **12** to give the N-de(*p*-methoxybenzyl)ated and simultaneously O-deisopropylidenated derivative **28** in moderate yields as shown in Table IV. In the cases of bicyclomycin derivatives having free hydroxyl at C-6 such as **13**, **26**, **29**, and **30**, the desired N-de(*p*-methoxybenzyl)ated derivatives were not obtained under the same conditions, presumably due to complex side re-

(14) In relation to these results it is noteworthy that the mono-*N*-methoxybenzylated analogue **12** of **4** seems to exist in a different rotamer shown, suggesting the stabilization due to hydrogen bonding between NH and the C_{12} oxygen. NOE observed: H-11 ↔ 12-CH_3, H-11 ↔ H-13b, 11-OH ↔ 12-CH_3, and Ispb ↔ NH.

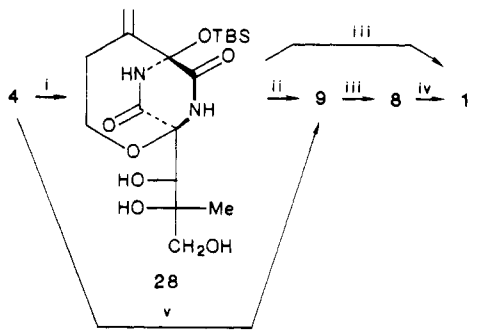


(15) In relation to this deacetalation, a different feature was observed in the case of the corresponding bis-N-(*p*-methoxybenzyl)ated derivative **25**. Treatment of **25** with 70% acetic acid gave spirocyclic compound **33** in 25% yield together with a major O-deisopropylidenated derivative (**29**, 66%). Both isomers were characterized as benzoates (**30** and **34**) of the primary hydroxyl group. The structure of **34** was confirmed by characteristic NMR data reported previously.[9b]

(16) Wacker, O.; Kump, W.; Wuller, B. M. *Tetrahedron Lett.* **1983**, *24*, 5607.

(17) (a) Yoshimura, J.; Yamaura, M.; Suzuki, T.; Hashimoto, H. *Chem. Lett.* **1983**, 1001. (b) Yamaura, M.; Suzuki, T.; Hashimoto, H.; Yoshimura, J.; Okamoto, T.; Shin, C. *Bull. Chem. Soc. Jpn.* **1985**, *58*, 1413.

Scheme IV[a]



[a] Reagents: (i) CAN; (ii) 2-methoxypropene, PPTS; (iii) Bu_4NF; (iv) 70% AcOH; (v) $K_2S_2O_8$, Cu^{2+}.

actions including the rearrangement as described above.

On the other hand, the recently reported oxidant, potassium persulfate,[18] proved to be effective in the presence of copper(II) ion for N-de(*p*-methoxybenzyl)ation of 1-benzyl-3-isobutyl-4-(*p*-methoxybenzyl)-2,5-piperazinedione[17b] (**31**), where 1-benzyl-3-isobutyl-2,5-piperazinedione (**32**) was obtained in 89% yield. However, the deprotection of **4** and **12** under the same conditions gave **9** in lower yield than those with CAN, although de-O-isopropylidenation did not occur in this case.

Conversion of 4 into 1. A few possible combinations of deprotection steps, which converts **4** into **1**, are shown in Scheme IV. When the preliminary communication[8] was reported, the conditions of O-de(*tert*-butyldimethylsilyl)ation with tetrabutylammonium fluoride without the rearrangement could not be optimized. Thus, compound **28** obtained by treatment of **4** with CAN in 49% yield was O-isopropylidenated again with 2-methoxypropene and PPTS to give **9** quantitatively. The compound **9** could also be prepared directly from **4** in 21% yield by oxidative N-de(*p*-methoxybenzyl)ation with sodium persulfate. Although O-desilylation of **4** with tetrabutylammonium fluoride was accompanied or followed by rearrangement, compound **9** did not show such tendency presumably due to steric hindrance of the isopropylidene group, and **8** was obtained in 95% yield. Deacetalation of **8** with 70% acetic acid afforded **1** in 92% yield.[15] Finally, it was found that under optimum conditions (Table III, entry 9) compound **28** could be O-desilylated to give **1** directly in 90% yield.

Experimental Section

General. Melting points and are uncorrected. ^1H NMR spectra were recorded at 100 MHz or 500 MHz in $CDCl_3$ unless otherwise stated. ^{13}C NMR spectra were recorded at 22.6 MHz. Full assignment are given only for the first member of a family of analogues and for certain unobvious cases. Optical rotations were measured on either a Carl Zeiss LEP-Al or a JASCO DIP-4 polarimeter at 20 ± 5 °C. Chromatography was performed on Wakogel C-200, flash chromatography on either Wakogel C-300 (Wako Pure Chem. Ind.) or Kieselgel 60 (230–400 mesh, Merck), and thin layer chromatography on Kieselgel 60.

Aldol Condensation between 6-[(*tert*-Butyldimethylsilyl)oxy]-7,9-bis(*p*-methoxybenzyl)-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (2) and (*S*)-2,3-*O*-Isopropylidene-2-*C*-methylglyceraldehyde (3). (A) With Racemic 2. To a stirred solution of racemic **2** (25 mg, 0.045 mmol) at −100 °C in THF (0.6 mL) were added with stirring 1.6 M BuLi in hexane (56 μL, 0.09 mmol) and after 10 min a solution of **3** (26 mg, 0.18 mmol) in THF (0.6 mL). The solution was stirred at the same temperature for 3 h and at −76 °C for 3 h. The reaction mixture was poured into saturated NH_4Cl, extracted with CH_2Cl_2, and provided the crude product mixture, which was fractionated by flash chromatography with hexane–AcOEt (7:3) to give **4** (10 mg, 32%, amorphous), a mixture of **5** and **7** (3.0 mg, 9.5%, syrup), and **6** (9.5 mg, 30%, syrup). (1*S*,6*R*)-6-[(*tert*-Butyldimethylsilyl)oxy]-1-[(1*S*,2*S*)-2,3-*O*-isopropylidene-2-methyl-1,2,3-trihydroxypropyl]-7,9-bis(*p*-methoxybenzyl)-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (**4**): amorphous, mp 61–63 °C; $[\alpha]_D$ +135° (*c* 1.0, $CHCl_3$); IR (KBr) 3330, 1685, 1645, 1635, 1610, 1585; ^1H NMR (500 MHz) data given in Table I; ^{13}C NMR δ −2.3 (q, SiMe), 19.8 (s, CMe_3), 21.1 (q, 2′-Me), 26.1, 28.1 (each q, CMe_2), 27.0 (q, CMe_3), 34.2 (t, C-4), 44.8, 47.2 (each t, CH_2 in MBn), 55.2 (q, OMe), 65.4 (t, C-3), 75.4 (t, C-3′), 79.7 (d, C-1′), 82.7 (s, C-2′), 85.7, 86.9 (each s, C-1 and C-6), 110.4 (s, CMe_2), 117.4 (t, $=CH_2$), 113.2, 113.4, 130.6, 132.8 (each d, aromatic in MBn), 149.3 (s, C-5), 128.8, 158.9, 159.2 (each s, aromatic in MBn), 167.3, 168.5 (each s, C-8 and C-10). Anal. Calcd for $C_{37}H_{52}N_2O_9Si$: C, 63.77; H, 7.52; N, 4.02. Found: C, 63.67; H, 7.59; N, 4.00.

(1*R*,6*S*)-6-[(*tert*-Butyldimethylsilyl)oxy]-1-[(1*R*,2*S*)-2,3-*O*-isopropylidene-2-methyl-1,2,3-trihydroxypropyl]-7,9-bis(*p*-methoxybenzyl)-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (**6**): syrup; $[\alpha]_D$ +2.1° (*c* 1.1, $CHCl_3$); ^1H NMR data given in Table I; ^{13}C NMR δ 19.7 (CMe_3), 25.6, 25.9, 26.6 (CMe_3), 28.3, 35.0, 44.6, 47.4, 55.3, 64.9, 76.7, 77.7, 82.4, 87.0, 87.4, 110.4, 113.3, 113.8, 118.0, 127.1, 127.7, 130.8, 131.5, 149.2, 158.9, 159.4, 167.0, 168.8. Anal. Calcd for $C_{37}H_{52}N_2O_9Si$: C, 63.77; H, 7.52; N, 4.02. Found: C, 63.52; H, 7.65; N, 3.87.

(B) With (1*S*,6*R*)-(+)-2. The bicyclic 2,5-piperazinedione (+)-**2** was coupled with **3** in the same manner as described above to give **4** and **5** in 51% and 13% yield, respectively. The major product **4** showed the same physical properties as described above, and ^1H NMR data of the minor one (**5**) are shown in Table I.

(C) With (1*R*,6*S*)-(−)-2. The bicyclic 2,5-piperazinedione (−)-**2** was coupled with **3** in the same manner as described above to give **6** and **7** in 44% and 20% yield, respectively. The major product **6** showed the same physical properties as described above and ^1H NMR data of the minor one (**7**) are given in Table I.

Preparation of 4 by O-De(*p*-methoxybenzyl)ation of 10. To a stirred solution of **10** (200 mg, 0.25 mmol) in CH_2Cl_2 (9 mL) were added DDQ (61.5 mg, 0.27 mmol) and water (0.5 mL). The mixture was stirred at room temperature for 2 h, and a syrup obtained by usual workup was purified in the same manner as described above to give **4** (137 mg, 80%).

(1*S*,6*R*)-6-[(*tert*-Butyldimethylsilyl)oxy]-[(1*S*,2*S*)-2-methyl-1,2,3-trihydroxypropyl]-7,9-diaza-2-oxabicyclo-[4.2.2]decane-8,10-dione (28). From 4. To a stirred solution of **4** (70 mg, 0.10 mmol) in acetonitrile (1.6 mL) were added a 1 M aqueous solution of CAN (0.4 mL) and then after 15 min a further portion of the same CAN solution (0.2 mL). The reaction mixture was allowed to an additional 10 min, diluted with water, and extracted with chloroform. A crude and syrupy product mixture obtained by usual workup of the extract was fractionated by flash chromatography with hexane–AcOEt (1:1 → 1:9) to give **28** (20.5 mg, 49%) and **9** (2.0 mg, 4.8%). **28**: mp 99–100 °C (amorphous); $[\alpha]_D$ +37.8° (*c* 0.9, $CHCl_3$); ^1H NMR δ 0.13, 0.29 (s, 6 H), 0.95 (s, 9 H), 1.45 (s, 3 H), 2.0, 2.8 (each br s, 2 H, OH), 2.6–2.8 (m, 2 H), 3.35, 3.77 (AB q, 2 H, J = 9.0 Hz), 3.8–4.1 (m, 2 H), 4.15 (d, J = 7.0 Hz, H-1′), 5.15, 5.60 (each s, 2 H), 6.15, 8.61 (each br s, 2 H); ^{13}C NMR δ −3.5 (q), 18.3 (s), 24.7 (q), 25.9 (q, *t*-Bu), 35.4, 64.5, 67.4, (each t), 71.6 (d), 76.6, 84.0, 87.7 (each s), 117.6 (t), 147.4 (s), 166.4, 169.7 (each s). Anal. Calcd for $C_{18}H_{32}N_2O_7Si$: C, 51.90; H, 7.74; N, 6.73. Found: C, 51.78; H, 7.81; N, 6.62.

From 12. Treatment of **12** (35 mg, 0.06 mmol) with CAN in the same manner as just described above gave **28** (15 mg, 54%).

(1*S*,6*R*)-6-[(*tert*-Butyldimethylsilyl)oxy]-1-[(1*S*,2*S*)-2,3-*O*-isopropylidene-2-methyl-1,2,3-trihydroxypropyl]-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (9). From 28. To a solution of **28** (21 mg, 0.05 mmol) in acetone (1 mL) were added 2-methoxypropene (0.1 mL) and a catalytic amount of PPTS, and the solution was kept at room temperature for 12 h. A syrupy residue obtained by evaporation of the mixture was purified by flash chromatography with hexane–AcOEt (3:2) to give **9** (22.7 mg, quantitative) as an amorphous solid: mp 81–2 °C; $[\alpha]_D$ +58.2° (*c* 1.0, $CHCl_3$); IR (KBr) 3400, 3300, 1700, 1695,

(18) Bose, A. K.; Mauhas, M. S.; Van der Veen, J. M.; Amin, S. G.; Fernandez, I. F.; Gala, K.; Gruska, R.; Kapar, J. C.; Khajavi, M. S.; Kreder, J.; Mukkarilli, L.; Ram, B.; Sugiura, M. L.; Vincent, S. E. *Tetrahedron* **1981**, *37*, 2321.

1640; ^{1}H NMR (100 MHz) δ 0.12, 0.29 (each s, 6 H, SiMe), 0.97 (s, 9 H, *t*-Bu), 1.39 (s, 3 H, Me), 1.47 (s, 6 H, Me), 2.45–2.73 (m, 2 H), 3.77, 4.35 (AB q, 2 H, *J* = 9.0 Hz), 3.84–4.04 (m, 2 H), 4.02 (d, H-1′), 4.35 (br d, 1′-OH), 5.11, 5.59 (each s, 2 H), 6.49, 7.92 (each br s, 2 H, NH); ^{13}C NMR δ −3.6 (q), 18.3 (s), 22.8, 25.8, 26.5, 27.5 (each q), 35.3 (t), 68.8, 73.0 (each t), 75.9 (d), 83.4, 85.2 (each s), 117.2 (s), 110.7 (s), 147.4, 167.5, 168.5 (each s). Anal. Calcd for $C_{21}H_{36}N_2O_7Si$: C, 55.24; H, 7.95; N, 6.14. Found: C, 55.36; H, 8.11; N, 5.94.

From 4. To a solution of **4** (80 mg, 0.12 mmol) in acetonitrile (8 mL) were added $K_2S_2O_8$ (124 mg, 0.5 mmol), anhydrous $CuSO_4$ (14.4 mg, 0.09 mmol), water (5.6 mL), and pyridine (80 μL), and then the mixture was heated at 60 °C for 40 min and after addition of ice–water extracted with CH_2Cl_2. A syrupy mixture obtained by usual workup of the extract was fractionated by flash chromatography to give **9** (11 mg, 21%).

From 12. Treatment of **12** (45 mg, 0.078 mmol) with $K_2S_2O_8$–$CuSO_4$ in the same manner as just described above gave **9** (13.8 mg, 39%).

From 8. To a solution of **8** (760 mg, 2.22 mmol) in DMF (30 mL) were added chloro-*tert*-butyldimethylsilane (410 mg, 2.66 mmol) and imidazole (226 mg, 3.33 mmol). After being stirred for 9 h at room temperature, the mixture was poured into water and extracted with AcOEt. The extract was washed with water, dried, and evaporated to give a syrupy residue, which was purified on a column of silica gel with hexane–AcOEt (1:1) to give **9** (966 mg, 96%).

General Procedure for O-De(*tert*-butyldimethylsilyl)-ation with Bu_4NF. To a solution of the 6-*O*-(*tert*-butyldimethylsilyl) derivative in an appropriate solvent (4–65 mL/1 mmol substrate) was added Bu_4NF (1 M THF solution or solid) according to the conditions and concentration described in Table III, and the mixture was kept at 0 °C or 25 °C for 0.25–6 h. The typical procedures are described for individual compounds.

(1*S*,6*R*)-6-Hydroxy-1-[(1*S*,2*S*)-2,3-*O*-isopropylidene-2-methyl-1,2,3-trihydroxypropyl]-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (8). From 9. To a stirred solution of **9** (20 mg, 0.044 mmol) in CH_2Cl_2 (1.0 mL) at 0 °C was added a 1 M THF solution of Bu_4NF (60 μL, 0.06 mmol) in three portions at intervals of 30 min. A solid residue obtained from the $CHCl_3$ extract of the reaction mixture was purified by flash chromatography with hexane–AcOEt (3:7) to give **8** (14 mg, 95%) as colorless prisms: mp 204 °C dec; $[\alpha]_D$ +71.6° (*c* 1.0, MeOH); IR (KBr) 3525, 3450, 3290, 3200, 1700, 1675; ^{1}H NMR (100 MHz) δ 1.38 (s, 2-Me), 1.44, 1.46 (each s, 6 H, CMe_2), 2.5–2.7 (m, 2 H, H-4), 3.8–4.0 (m, 2 H, H-3), 4.10 (d, *J* = 8.0 Hz, H-1′), 3.77, 4.38 (AB q, 2 H, *J* = 8.0 Hz, H-3′), 4.63 (d, 1′-OH), 5.03 (br s, 6-OH), 5.16, 5.61 (each br s, 2 H, $=CH_2$), 6.94, 8.24 (each br s, 2 H, NH); ^{13}C NMR δ 25.0, 26.8, 28.2 (each q), 36.5 (t), 66.5 (t), 73.1 (t and d), 82.8, 86.4, 88.9 (each s), 11.5 (s), 116.7 (t), 149.1, 168.3, 172.0 (each s). Anal. Calcd for $C_{15}H_{22}N_2O_7$: C, 52.62; H, 6.48; N, 8.18. Found: C, 52.79; H, 6.79; N, 7.95.

From 1. To a suspension of bicyclomycin (**1**) (6.0 g, 19.1 mmol) in dry acetone (180 mL) were added 2-methoxypropene (11 mL, 115 mmol) and a catalytic amount of PPTS, and the mixture was kept overnight at room temperature. The crystalline residue obtained by evaporation of the reaction mixture was washed with CH_2Cl_2 and recrystallized from acetone–CH_2Cl_2 to give **8** (6.45 g, 99%).

(1*S*,6*R*)-6-Hydroxy-1-[(1*S*,2*S*)-2-methyl-1,2,3-trihydroxypropyl]-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (Bicyclomycin, 1). From 8. A solution of **8** (32 mg, 0.094 mmol) in 80% aqueous acetic acid (2 mL) was kept at room temperature for 8 h and evaporated, after addition of silica gel (100 mg), at 5–10 °C under reduced pressure. The residual gel was directly subjected to flash chromatography with $CHCl_3$–MeOH (7:1) to give **1** (27.0 mg, 92%) as colorless prisms: mp 189–191 °C dec [lit. mp 188–189 °C dec,[1a] 170–171 °C dec;[2b] $[\alpha]_D$ +63.2° (*c* 1.0, MeOH [lit.[1a] $[\alpha]^{23}_D$ +63.5° (*c* 1, MeOH)]; ^{1}H NMR (100 MHz in DMSO-d_6) δ 1.15 (s, 3 H), 2.2–2.6 (m, 2 H), 3.1–3.5 (AB q, 2 H, *J* = 12.0 Hz), 3.5–3.9 (m, 2 H), 3.84 (d, H-1′), 4.42 (t, *J* = 5.0 Hz, 3′-OH), 5.08 (s, 2′-OH), 5.14 (d, *J* = 7.5 Hz, 1′-OH), 4.96, 5.28 (each d, *J* = 1.8 Hz, $=CH_2$), 6.68 (s, 6-OH), 8.63, 8.92 (each s, NH); ^{13}C NMR δ 24.8 (q), 36.3, 64.2, 67.6 (each t), 71.4 (d), 78.0, 82.4, 88.7 (each s), 116. 1 (t), 149.9 (s), 167.2, 170.4 (each s).

From 28. Treatment of **28** with Bu_4NF in a similar manner as described for the preparation of **8** from **9** under the conditions given for the entry 9 in Table III afforded **1** in 90% yield.

***p*-Methoxybenzylation of 9.** To an ice-cold suspension of NaH (815 mg, 17.0 mmol) in DMF (20 mL) were added dropwise with stirring a solution of **9** (2.5 g, 5.48 mmol) in DMF (20 mL) and then *p*-methoxybenzyl bromide (3.86 g, 19.2 mmol) during 30 min. After being stirred for 2 h, the mixture was acidified with acetic acid, poured into saturated $NaHCO_3$, and extracted with AcOEt. The residue obtained from the extract was fractionated on a column of silica gel with hexane–AcOEt (7:3) to give **10** and crude **11** in 37% and 50% yield, respectively. The latter compound was characterized as O-de(*p*-methoxybenzyl)ated derivative **12** as described below. (1*S*,6*R*)-6-[(*tert*-Butyldimethylsilyl)-oxy]-1-[(1*S*,2*S*)-2,3-dihydroxy-2,3-*O*-isopropylidene-1-[(*p*-methoxybenzyl)oxy]-2-methylpropyl]-7,9-bis(*p*-methoxybenzyl)-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (**10**): ^{1}H NMR (100 MHz) δ 0.16, 0.42 (each s, 6 H), 0.87 (s, 9 H), 1.24, 1.27, 1.31 (each s, 9 H), 2.1–2.3 (m, 2 H), 3.0–3.2, 3.4–3.6 (each m, 2 H), 3.76 (s, 3 H, OMe), 3.80 (s, 6 H, OMe), 3.72, 4.19 (AB q, 2 H, *J* = 8.2 Hz), 4.3–4.9 (m, 7 H, CH_2 in MBn and H-1′), 5.06, 5.52 (each s, 2 H), 6.72, 6.79, 6.83, 7.23, 7.25, 7.44 (each d, 12 H, *J* = 8.0 Hz, MBn); ^{13}C NMR δ −1.7, −1.3, 19.6, 26.2 (2′-Me), 26.7 (CMe_3), 28.5, 29.3, 35.2, 44.8, 48.4, 55.3 (3 × OMe), 64.7, 71.5 (CH_2 in OMBn), 76.1, 82.3 (C-1′), 84.3 (C-6 and C-2′), 87.1 (C-1), 108.2, 113.3, 113.5, 117.3, 129.6, 131.5, 128.6, 128.8, 130.6, 148.8, 158.6, 158.9, 159.0, 165.2, 169.7.

Selective O-de(*p*-methoxybenzyl)ation of crude **11** (100 mg, 0.14 mmol) with DDQ as described for **4**, followed by flash chromatography on silica gel with hexane–AcOEt (7:3), afforded **12** (68 mg, 82%) as an amorphous solid. (1*R*,6*S*)-6-[(*tert*-Butyldimethylsilyl)oxy]-1-[(1*S*,2*S*)-2,3-*O*-isopropylidene-2-methyl-1,2,3-trihydroxypropyl]-7-(*p*-methoxybenzyl)-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (**12**): mp 65–67 °C; $[\alpha]_D$ +46.1° (*c* 1.0, $CHCl_3$); IR (KBr) 3420, 3310, 1715, 1670, 1640; ^{1}H NMR (500 MHz) δ 0.17, 0.38 (each s, 6 H), 0.85 (s, 9 H), 1.31 (s, 3 H, 2′-Me), 1.43, 1.46 (each s, 6 H), 2.09 (dd, *J* = 9.0, 16.5 Hz, H-4a), 2.42 (dd, *J* = 7.9 Hz, H-4b), 3.72 (dd, *J* = 8.1, 13.4 Hz, H-3a), 3.75 (d, *J* = 8.9 Hz, H-13a), 3.82 (dd, *J* = 9.0 Hz, H-3b), 3.77 (s, 3 H, OMe). 4.04 (d, *J* = 8.2 Hz, H-11), 4.31 (d, H-13b), 4.57, 4.69 (AB q, *J* = 14.5 Hz), 4.67 (d, 1′-OH), 5.10, 5.58 (each s, 2 H, H-5′a,b), 6.77, 7.23 (each d, 4 H, *J* = 8.5 Hz), 7.96 (s, NH); ^{13}C NMR δ −1.9, −2.8, 19.6, 22.9 (2′-Me), 26.6 (CMe_3), 27.7, 29.7 (CMe_2) 35.9, 44.2, 55.1, 64.4, 75.6 (C-3′), 76.4 (C-1′), 83.9, 84.6, 87.3 (C-1, C-6 and C-2′), 110.7, 129.1, 113.5, 117.9, 128.8, 129.1, 147.3, 158.8, 166.4, 167.7. Anal. Calcd for $C_{29}H_{44}N_2O_8Si$: C, 60.39; H, 7.69; N, 4.86. Found: C, 60.32; H, 7.49; N, 4.51.

(1*S*,6*R*)-1-[(1*S*,2*S*)-2,3-*O*-Isopropylidene-2-methyl-1,2,3-trihydroxypropyl]-7-(*p*-methoxybenzyl)-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (13). Treatment of **12** with Bu_4NF in the same manner as described in the general procedure gave **13** in 91% yield under the conditions given for entry 8 in Table III: ^{1}H NMR (100 MHz) δ 1.24, 1.41 (each s, 9 H), 1.9–2.4 (m, 2 H), 3.7–3.9 (m, 2 H), 3.76 (s, 3 H), 4.05 (d, *J* = 8.4 Hz, H-1′), 4.08, 4.29 (AB q, *J* = 8.5 Hz), 4.38, 4.66 (AB q, 2 H, *J* = 13.8 Hz), 4.98, 5.65 (each s, $=CH_2$), 5.12 (d, 1′-OH), 5.16 (s, 6-OH), 6.77, 7.34 (each d, *J* = 8.4 Hz), 8.10 (s, NH); ^{13}C NMR δ 22.3, 26.5, 27.5 (each q), 35.5, 44.6 (each t), 55.2 (q), 64.5, 73.3 (each t), 77.7 (d), 83.7 (s), 84.1 (d), 84.3 (s), 110.8 (s), 113.5 (d), 119.1 (t), 128.5 (s), 130.6 (d), 146.1, 159.0, 167.1, 168.9 (each s).

(1*S*,6*R*)-5-Hydroxy-7,9-bis(*p*-methoxybenzyl)-6-[[(*S*)-2-methoxy-2-phenyl-3,3,3-trifluoropropanoyl]oxy]-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (15) and Its 1*R*,6*S* Isomer (20). To a solution of 5,6-dihydroxy-7,9-bis(*p*-methoxybenzyl)-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (racemic 1:1 mixture of **14** and **19**, 100 mg, 0.23 mmol) in CH_2Cl_2 (5 mL) were added (*R*)-(−)-2-methoxy-2-phenyl-3,3,3-trifluoropropionyl chloride (66.8 mg, 0.26 mmol; prepared[11] in a conventional manner from the corresponding acid and thionyl chloride) and (*N*,*N*-dimethylamino)pyridine (138 mg, 1.3 mmol). After being refluxed for 50 h, the reaction solution was washed with 1 M HCl, saturated $NaHCO_3$, and NaCl, dried, and evaporated to give a crude mixture of **15** and **20** which was separated on a column of silica gel to afford **15** and **20** (each 67 mg, 45%) as crystals.

15: colorless prism (EtOH); mp 107–108 °C; $[\alpha]_D$ −4.0° (*c* 1.1, $CHCl_3$); IR (KBr) 1760, 1700, 1670; ^1H NMR δ 1.6–2.2 (m, 2 H, H-4), 3.04–4.01 (m, H-3), 3.33 (dd, *J* = 9.4, 15.0 Hz, H-3′), 3.56 (d, 3 H, *J* = 1.1 Hz, OMe), 3.76, 3.79 (s, 6 H, OMe in MBn), 4.10, 4.62, 4.35, 4.80 (2 × AB q, 4 H, *J* = 14.3 Hz, CH_2 in MBn), 4.93 (s, OH), 5.18 (s, H-1), 5.28 (dd, 1 H, H-5), 6.74, 6.84, 7.12, 7.23 (each d, 8 H, *J* = 8.0 Hz, aromatic in MBn), 7.1–7.2 (m, 5 H, Ph); ^{13}C NMR δ 31.3 (t, C-4), 46.0, 49.0 (each t, CH_2 in MBn), 55.2 (q, 2 × OMe in MBn), 55.4 (q, OMe), 59.0 (t, C-3), 81.0 (d, C-5), 82.7 (d, C-1), 83.9 (s, C-6), 85.1 (q, C-2′), 113.8, 114.4 (each d, MBn), 123.3 (q, C-3′), 127.5, 128.6, 129.7, 130.5 (each d, MBn), 125.7 (s, Ph), 129.9, 131.6 (each s, MBn), 158.7, 159.8 (each s, MBn), 163.3 (s, C=O), 165.5, 167.8 (each s, C-8 and C-10). Anal. Calcd for $C_{33}H_{33}F_3N_2O_9$: C, 60.18; H, 5.05; N, 4.25. Found: C, 59.91; H, 5.03; N, 4.30.

20: coloress prism (EtOH); mp 174.5 °C; $[\alpha]_D$ −15.1° (*c* 1.1, $CHCl_3$); IR (KBr) 1760, 1690, 1680; ^1H NMR δ 1.67–2.27 (m, 2 H), 3.63–4.03 (m, 1 H), 3.36 (dd, 1 H, *J* = 9.6, 13.9 Hz), 3.59 (d, 3 H, *J* = 1.1 Hz), 3.73, 3.77 (s, 6 H), 3.81, 4.29, 4.57, 4.81 (2 × AB q, 4 H, *J* = 15.0 Hz), 4.92 (s, 1 H), 5.17 (s, 1 H), 5.37 (dd, 1 H, H-5), 6.67, 6.79, 6.99, 7.15 (each d, 8 H, *J* = 8.4 Hz), 7.27–7.67 (m, 5 H). Anal. Calcd for $C_{33}H_{33}F_3N_2O_9$: C, 60.18; H, 5.05; N, 4.25. Found: C, 59.97; H, 4.91; N, 4.39.

(1*S*,6*R*)-5,6-Dihydroxy-7,9-bis(*p*-methoxybenzyl)-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (14). The ester **15** (67 mg, 0.10 mmol) was deacylated conventionally with sodium methoxide in methanol. Purification on silica gel with AcOEt–hexane (3:2) gave **14** (45 mg) as an amorphous powder; $[\alpha]_D$ −45.1° (*c* 0.75, $CHCl_3$). Anal. Calcd for $C_{23}H_{26}N_2O_7$: C, 62.43; H, 5.92; N, 6.33. Found: C, 62.03; H, 6.23; N, 6.05.

(1*R*,6*S*)-5,6-Dihydroxy-7,9-bis(*p*-methoxybenzyl)-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (19). The ester **20** was deacylated in the same manner as described above to yield **19**. Anal. Calcd for $C_{23}H_{26}N_2O_7$: C, 62.43; H, 5.92; N, 6.33. Found: C, 62.13; H, 6.33; N, 6.16.

Preparation of Optically Active 2. Both enantiomers **14** and **19** were converted into (+)-**2** and (−)-**2**, respectively, in the same manner as described[9] for the preparation of racemic **2**.

(1*S*,6*R*)-6-Hydroxy-7,9-bis(*p*-methoxybenzyl)-7,9-diaza-2-oxabicyclo[4.2.2]decane-5,8,10-trione (16): yield 91%; $[\alpha]_D$ +47.0°. Anal. Calcd for $C_{23}H_{24}N_2O_7$: C, 62.72; H, 5.49; N, 6.36. Found: C 62.40; H, 5.52; N, 6.07.

(1*R*,6*S*)-6-Hydroxy-7,9-bis(*p*-methoxybenzyl)-7,9-diaza-2-oxabicyclo[4.2.2]decane-5,8,10-trione (21): $[\alpha]_{20}$ −47.4°. Anal. Calcd for $C_{23}H_{24}N_2O_7$: C, 62.72; H, 5.49; N, 6.36. Found: C, 62.32; H, 5.39; N, 6.02.

(1*S*,6*R*)-6-(*tert*-Butyldimethylsilyl)-7,9-bis(*p*-methoxybenzyl)-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione [(+)-2]: 33% yield (three steps from **16**); $[\alpha]_D$ +15.9° ($CHCl_3$). Anal. Calcd for $C_{30}H_{40}N_2O_6Si$: C, 65.20; H, 7.30; N, 5.07. Found: C, 65.09; H, 7.11; N, 4.77.

(1*R*,6*S*)-6-(*tert*-Butyldimethylsilyl)-7,9-bis(*p*-methoxybenzyl)-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione [(−)-2]: $[\alpha]_D$ −15.5° ($CHCl_3$). Anal. Calcd for $C_{30}H_{40}N_2O_6Si$: C, 65.20; H, 7.30; N, 5.07. Found: C, 65.56; H, 7.30; N, 5.42.

(1*S*,6*R*,8*S*)-6-Hydroxy-8-[(*S*)-1,2-dihydroxy-1,2-*O*-isopropylidene-2-methylethyl]-9-(*p*-methoxybenzyl)-1-[(*p*-methoxybenzyl)carbamoyl]-5-methylene-9-aza-2,5-dioxabicyclo[4.2.2]decan-10-one (24). To a solution of **4** (620 mg, 0.89 mmol) in THF (3.6 mL) at room temperature was added a 1 M solution of Bu_4NF in THF (2.5 mL, 2.7 mmol). After 3 h, the mixture was poured into water and extracted with CH_2Cl_2. A syrupy residue obtained by conventional workup of the extract was purified on a column of silica gel with hexane–AcOEt (7:3) to give **24** (482 mg, 93%) as an amorphous solid: mp 75–76 °C; $[\alpha]_D$ −88.7° (*c* 1.0, $CHCl_3$); IR (KBr) 3400, 1690, 1665; ^1H NMR (100 MHz) δ 1.40 (s, 6 H), 1.44 (s, 3 H), 1.9–2.3 (m, 1 H), 2.6–3.0 (m, 2 H), 3.5–3.8 (m, 1 H), 3.76, 3.81 (each s, 6 H), 3.77, 4.18 (AB q, 2 H, *J* = 9.5 Hz, H-2′), 4.31 (s, H-8), 4.88 (s, OH), 4.10, 5.24 (AB q, 2 H), 4.35, 4.55 (AB q, 2 H), 5.00, 5.45 (each s, 2 H), 6.70, 6.88, 7.09, 7.35 (each d, 8 H); ^{13}C NMR δ 21.9, 26.1, 27.0, 36.1, 43.7, 47.6, 55.1, 55.4, 65.7, 75.9, 80.1 (C-8), 80.8 (C-1′), 92.6, 96.4 (C-1 and C-4), 111.1, 113.7, 114.2, 116.7 (=CH_2), 128.0 (s, MBn), 129.6, 131.3, 147.0, 159.2, 159.3, 164.5, 170.1. Anal. Calcd for $C_{31}H_{38}N_2O_9$: C, 63.90; H, 6.57; N, 4.81. Found: C, 63.45; H, 6.67; N, 4.57.

(1*S*,6*R*)-1-[(1*S*,2*S*)-2,3-*O*-Isopropylidene-2-methyl-1,2,3-trihydroxypropyl]-7,9-bis(*p*-methoxybenzyl)-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (25). Compound **26** was O-de(*p*-methoxybenzyl)ated in the same manner as described in the preparation of **4** from **10** to give **25** in 78% yield: syrup; $[\alpha]_D$ +127° (*c* 1.4, $CHCl_3$); ^1H NMR δ 1.26, 1.36 (each s, 9 H), 1.88–2.12 (m, 2 H, H-4), 2.64–2.98, 3.4–3.68 (each m, 2 H, H-3), 3.77 (s, 6 H), 3.68, 4.11 (AB q, 2 H, *J* = 9.0 Hz), 4.26, 4.54 (AB q, 2 H, *J* = 14.0 Hz), 4.59, 5.08 (AB q, 2 H, *J* = 14.5 Hz), 4.60 (d, *J* = 10.5 Hz), 5.26 (s, OH), 4.95, 5.50 (each s, 2 H), 6.53 (d, *J* = 10.8 Hz, 1′-OH), 6.72, 7.32, 7.39 (each d, 8 H, *J* = 9.0 Hz); ^{13}C NMR δ 26.0, 28.1, 29.3 (each q), 34.0, 45.1, 47.6 (each t), 55.2 (q), 65.6, 75.6 (each t), 80.2 (d), 82.7, 83.9, 86.1, 110.8 (each s), 113.4, 113.5 (each d), 118.6 (t), 127.4, 127.9, (each s), 131.3, 132.7 (each d), 147.7, 159.1, 159.4 (each s), 168.7 (s, C-8 and C-10). Anal. Calcd for $C_{31}H_{38}N_2O_9$: C, 63.90; H, 6.57; N, 4.81. Found: C, 63.65; H, 6.67; N, 4.57.

(1*S*,6*R*)-1-[(1*S*,2*S*)-2,3-Dihydroxy-2,3-*O*-isopropylidene-1-[(*p*-methoxybenzyl)oxy]-2-methylpropyl]-7,9-bis(*p*-methoxybenzyl)-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (26). Treatment of **10** with Bu_4NF in the same manner as described in the general procedure under the conditions given for entry 5 in Table III, followed by purification on a column of silica gel with hexane–AcOEt (4:1), gave **26** in 90% yield: ^1H NMR (100 MHz) δ 1.24 (s, 6 H), 1.37 (s, 3 H), 2.2–2.4 (m, 2 H, H-4), 3.0–3.4 (m, 2 H, H-3), 3.72, 3.74, 3.78 (each s, 9 H), 3.6–3.8 (1 H, H-3′a), 4.28 (AB q, *J* = 8.5 Hz, H-3′b), 4.1–4.9 (m, 7 H, 3 × CH_2 in MBn and H-1′), 5.12 (s, 6-OH), 5.07, 5.59 (each s, 2 H), 7.29, 7.34, 7.37, 7.72, 7.74, 7.76 (each d, 12 H, *J* = 8.0 Hz); ^{13}C NMR δ 26.2, 28.5, 29.3, 34.6, 45.0, 48.6, 55.2, 64.9, 71.9 (t, CH_2 in MBn), 76.2, 82.4 (C-1′), 83.7, 84.0, 84.2 (C-1, C-6, and C-2′), 108.6, 113.4, 113.5, 114.0, 118.3 (=CH_2), 128.2, 128.8, 130.4, 129.6, 130.7, 131.4, 158.8, 159.1, 165.2, 170.4. Anal. Calcd for $C_{39}H_{46}N_2O_{10}$: C, 66.65; H, 6.60; N, 3.99. Found: C, 65.28; H, 7.01; N, 3.97.

O-Deisopropylidenation of 25. A solution of **25** (270 mg, 0.46 mmol) in 80% aqueous acetic acid (5 mL) was kept at room temperature for 33 h. After addition of water conventional extraction of the products with AcOEt followed by fractionation on silica gel with hexane–AcOEt (7:3) gave **29** (165 mg, 66%) and **33** (62 mg, 25%). (1*S*,6*R*)-6-Hydroxy-7,9-bis(*p*-methoxybenzyl)-1-[(1*S*,2*S*)-2-methyl-1,2,3-trihydroxypropyl]-5-methylene-7,9-diaza-2-oxabicyclo[4.2.2]decane-8,10-dione (**29**): ^1H NMR (100 MHz) δ 1.25 (s, 3 H), 1.96–2.20 (m, 2 H, H-4), 2.66–3.80 (m, 4 H), 3.15, 3.46 (AB q, 2 H, H-3′), 3.73, 3.76 (each s, 6 H), 4.61 (d, *J* = 11 Hz, H-1′), 4.29, 4.61 (AB q, 2 H, *J* = 14 Hz), 4.55, 4.93 (AB q, 2 H, *J* = 13.5 Hz), 5.00, 5.56 (each s, 2 H), 5.66 (d, 1 H, 1′-OH), 5.80 (br s, 1 H, 6-OH), 6.70, 7.32, 7.36 (each d, 8 H, *J* = 8.5 Hz). Selective benzoylation of **29** (130 mg, 0.24 mmol) with benzoyl chloride (35.4 mg, 0.25 mmol) and pyridine (57 mg, 0.72 mmol) in 1,2-dichloromethane (5 mL) followed by flash chromatography on silica gel with hexane–AcOEt (1:1) gave monobenzoate **30** (123 mg, 79%): $[\alpha]_D$ +76.3° (*c* 1.8, $CHCl_3$); ^1H NMR δ 1.27 (s, 3 H), 2.0–2.2 (m, 2 H, H-4), 2.28 (br s, 1 H, 2′-OH), 2.70–3.05 (m, 1 H), 3.5–3.7 (m, 1 H), 3.73, 3.76 (each s, 6 H), 4.18 (s, 2 H), 4.69 (d, 1 H, *J* = 10 Hz, H-1′), 4.29, 4.55 (AB q, 2 H, *J* = 13.5 Hz), 4.56, 4.92 (AB q, 2 H, *J* = 16 Hz), 5.00, 5.53 (each s, 2 H), 5.22 (s, 1 H, 6-OH), 5.79 (d, 1 H, 1′-OH), 6.6–6.8 (m, 4 H), 7.2–7.55 (m, 7 H), 7.65–8.0 (m, 2 H); ^{13}C NMR δ 20.9, 29.3 (each q), 34.2 (t), 44.9, 47.2 (each t), 65.2 (t), 71.0 (t), 74.9 (s), 76.0 (d), 84.1, 86.1 (each s), 113.6, 113.7 (each d), 159.0, 159.3, 168.6, 168.8 (each s). Anal. Calcd for $C_{35}H_{38}N_2O_{10}$: C, 65.00; H, 5.92; N, 4.33. Found: C, 65.17; H, 5.97; N, 4.31.

8-[(1*S*,2*S*)-3-Hydroxy-1-methylenepropyl]-3-methyl-6,9-bis(*p*-methoxybenzyl)-3,4,8-trihydroxy-6,9-diaza-1-oxaspiro[4.5]decane-7,10-dione (**33**): ^1H NMR δ 1.26 (s, 3 H), 2.40 (t, 2 H, *J* = 6.0 Hz, H-2′), 3.67 (t, 2 H, H-3′), 3.78 (s, 6 H), 4.08 (br d, 1 H, *J* = 8.0 Hz, H-4), 4.13, 4.29 (AB q, 2 H, *j* = 10 Hz, H-2), 4.32, 4.64 (AB q, 2 H, *J* = 16 Hz), 4.26, 4.76 (AB q, 2 H, *J* = 15.5 Hz), 5.34, 5.70 (each s, 2 H, =CH_2), 3.19, 3.32, 5.22, 6.11 (each br s, 4 H, OH), 6.72, 6.76, 7.10, 7.14 (each d, 8 H, *J* = 8.5 Hz). Selective benzoylation of **33** (18 mg) in the same manner as described above and purification on silica gel with hexane–AcOEt (3:2) gave 3′-benzoate **34** (11 mg, 51%): $[\alpha]_D$ −4.2° (*c* 0.8, $CHCl_3$); ^1H NMR δ 1.28 (s, 3 H), 1.70 (br s, 1 H, 2′OH), 2.59 (t, 2 H, H-4), 3.13 (br

d, 1 H, 1'-OH), 3.74, 3.76 (each s, 6 H), 4.09 (d, 1 H, J = 10.0 Hz, H-1'), 4.14 (t, 2 H, J = 6.0 Hz, H-3), 4.12, 4.41 (AB q, 2 H, J = 7.0 Hz, H-3'), 4.35, 4.72, 4.20, 4.80 (each AB q, J = 14 Hz), 5.20, 5.75 (each s, 2 H, $=CH_2$), 6.06 (s, 1 H, 6-OH), 6.74 (d, 4 H, J = 8.0 Hz), 7.0-7.6 (m, 7 H), 7.9-8.05 (m, 2 H); ^{13}C NMR δ 21.8 (q), 29.7 (t, C-4), 45.4, 46.9 (each t), 55.2 (q), 63.3 (t), 77.1 (s, C-2'), 80.5 (d, C-1'), 81.6 (t, C-3'), 86.5, 96.5 (each s, C-5 and C-8), 117.8 (t), 143.0 (s, C-5), 113.6, 114.2, 128.1, 128.4, 129.5, 132.9 (each d), 128.8, 158.7, 159.0 (each s), 166.0, 166.4, 166.8 (each s). Anal. Calcd for $C_{35}H_{38}N_2O_{10}$: C, 65.00; H, 5.92; N, 4.33. Found: C, 65.23; H, 6.05; N, 4.28.

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Registry No. **1**, 38129-37-2; (±)-**2**, 94807-54-2; (+)-**2**, 117249-39-5; (-)-**2**, 95341-59-6; **3**, 79243-92-8; **4**, 95237-55-1; **5**, 117305-43-8; **6**, 117305-44-9; **7**, 117305-45-0; **8**, 63777-16-2; **9**, 95237-57-3; **10**, 117183-89-8; **11**, 117183-90-1; **12**, 117183-91-2; **13**, 117183-92-3; **14**, 117249-34-0; (±)-**14**, 117249-40-8; **15**, 117183-93-4; **16**, 117249-35-1; **19**, 117249-36-2; **20**, 117249-37-3; **21**, 117249-38-4; **24**, 117201-61-3; **25**, 95694-70-5; **26**, 117183-94-5; **28**, 95237-56-2; **29**, 117183-95-6; **30**, 117183-96-7; **33**, 117183-97-8; **34**, 117183-98-9; (*S*)-(-)-MTPA, 17257-71-5; CH_2=C(OMe)CH_3, 116-11-0; ClSi(*t*-Bu)Me_2, 18162-48-6; 4-$MeOC_6H_4CH_2Br$, 2746-25-0; PhCOCl, 98-88-4.

Reactivity of Quinone Imine and Quinone Diimine Metabolites of the Antitumor Drug Amsacrine and Related Compounds to Nucleophiles

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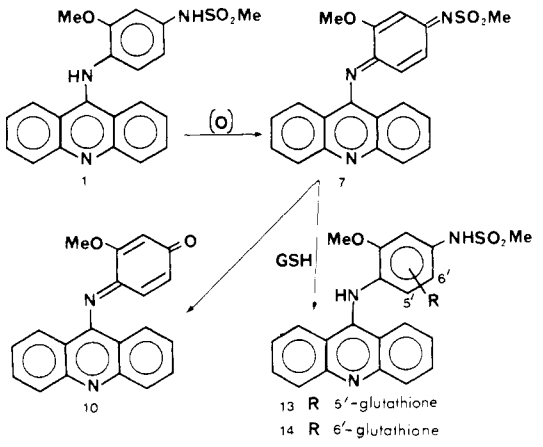
The quinone diimine AQDI (**7**) and the quinone imine AQI (**10**) (products of oxidative metabolism of the clinical antileukemia drug amsacrine (**1**)) and related compounds were prepared, and their reactions with a variety of nucleophiles were studied. Reaction of both quinone diimines and quinone imines with methanethiol gave reduced products resulting from 1,4-addition, while reaction with methylamine and dimethylamine gave almost exclusively quinonoid products, resulting from 1,4-addition followed by reoxidation. These results clarify the mechanism by which certain metabolites of amsacrine are probably formed. Detailed NMR studies of the quinonoid products show that the presence of a bulky group in the 3'-position results in the anilino ring being restricted to one conformer, where the group is distal from the acridine ring.

Recent studies[1,2] on the metabolism of the clinical antileukemic drug amsacrine (**1**) in rats have identified the glutathione 5'-conjugate **13** as the main biliary metabolite. This is postulated to be formed by nucleophilic 1,4-addition to an intermediary quinone diimine **7** (AQDI) (Scheme I). Other metabolites detected include the quinone imine **10**, presumably formed by competing hydrolysis of **7**. Chemical reaction of AQDI with glutathione has been shown to be rapid,[3] resulting in a 60:40 mixture of the 5'-adduct **13** and the isomeric 6'-adduct **14**. Oxidation of amsacrine to AQDI is facile, with an oxidation potential for the reversible two-electron oxidation of 280 mV,[4] and can be accomplished readily by liver microsomes[5] or by reaction with MnO_2.[1] We have also observed the reaction to occur spontaneously under neutral conditions; dilute solutions of amsacrine free base in aqueous methanol slowly produce AQDI, while solutions of the hydrochloride salt do not.

The role of this facile oxidation in the biological activity of amsacrine is not well understood, but is clearly important. Some studies have reported that AQDI is more cytotoxic than amsacrine itself,[5] while rapid cleavage of DNA is observed in the presence of amsacrine, oxygen, and

Scheme I



copper salts, suggesting the possibility of redox cycling of the drug.[6] It is also possible that the quinone diimine could act as an alkylating agent toward biological macromolecules, since the well-studied quinone imines *N*-acetyl-*p*-benzoquinone imine (**19**) and *N*-(4-ethoxyphenyl)-*p*-benzoquinone imine (**20**) (the major oxidative metabolites of the drugs acetaminophen and phenacetin respectively[7,8]) are known to exert their cytotoxic effects via arylation of both glutathione and protein thiols.[9,10]

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