surements and analyses of the data produced were performed at the Center for Fast Kinetics Research at the University of Texas at Austin. The CFKR is supported jointly by the Biotechnology Research Program of the Division of Research Resources of NIH (Grant No. RR00886) and by the University of Texas at Austin. Additional lifetime measurements were carried out in the laboratory of Prof. David Holden, University of Waterloo, whom we acknowledge for his assistance. The fluorescence lifetime measurement for **11a** by Edinburgh Instruments, Edinburgh, U.K., is also acknowledged.

Studies on the Reactivity of Bicyclomycin with Amines[1]

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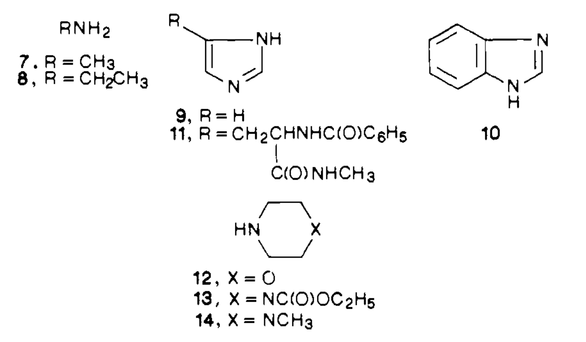
Abstract: The reactivity of bicyclomycin (**1**) under basic conditions has been investigated. Eight different amines were sequentially reacted with **1**. Treatment of bicyclomycin with the primary amines methylamine and ethylamine yielded the ring-cleaved products **15–18**. Correspondingly, use of the heteroaromatic amines imidazole, benzimidazole, and (dl)-N_α-benzoylhistidine methylamide in these experiments led to the formation of a diastereomeric mixture of the ring-opened adducts **19**, **21**, and **22**, respectively. Finally, treatment of **1** with the secondary amines morpholine, ethyl piperazinecarboxylate, and N-methylpiperazine furnished the novel adducts **26–28**, respectively. Analysis of the composite results suggests that a key step in the base-mediated chemical processes is the reversible ring-opening of the C(6)-hemiaminal bond to give the enone **2**. The mechanism of these reactions and the implications of these studies in relation to the mode of action of the antibiotic are discussed.

Bicyclomycin (**1**), a clinically used antibiotic, has received considerable attention in recent years.[2-4] It is a structurally unique cyclic peptide possessing pronounced activity against several strains of Gram-negative bacteria. Most proposals pertaining to the mode of action of **1** have suggested that nucleophilic species present within the peptidoglycan assembly of the bacterial cell wall play a pivotal role in the activation and subsequent binding of the chemotherapeutic agent.[5-9] Both sulfhydryl-containing proteins[5] and amidases[6-8] have been advanced as likely candidates in these transformations. Information in favor of the former species emanated from the pioneering studies of Iseki and co-workers which demonstrated that **1** reacts with methyl mercaptan at basic pH.[5a] This result, coupled with the observation that **1** covalently interacts with inner-membrane proteins of *Escherichia coli*,[5c] led to the notion that the antibacterial activity of **1** is associated with the binding of a nucleophile (i.e., a protein sulfhydryl group) to the terminal olefinic group [C(5)–C(5a)] of the drug. The initially proposed pathway for this transformation is depicted in Scheme I.[6] Alternatively, recent work by Vasquez and co-workers has led to the speculation that amidases play an integral role in the bicyclomycin activation process.[7] Moreover, the close structural correspondence of **1** to the projected structure of the diaminopimelic acid–diaminopimelic acid unit within the peptidoglycan assembly of the cellular membrane has prompted the suggestion by Williams and his group that the drug functions as a competitive substrate for a protease involved in the biosynthesis of the bacterial cell envelope (Scheme II).[6] Key steps in this hypothesis include the enzymic cleavage of the C(9)–N(10) amide bond in **1** to yield **4** and the Michael addition of a second biological nucleophile to the conjugated system **5** to generate **6**.

In light of these mechanistic scenarios, it is surprising that no information exists on the reactivity of bicyclomycin with amines. All previous accounts have focused on sulfur-[5a,8-10] and oxygen-containing[11] nucleophiles. In this paper, we report on the chemical reactivity of **1** with primary, secondary, and heteroaromatic amines, including a functionalized derivative of histidine. Special attention is centered on the interplay of both the type of amine used and the pH of the reaction medium on the product profile. Arguments are advanced that a critical step in the chemical activation of the drug is the reversible ring-opening of the C(6)-hemiaminal bond of bicyclomycin to generate enone **2**. Michael addition of the amine to the α,β-unsaturated system generates a C(5a)-substituted adduct. This species can then be converted into a series of novel rearranged, ring-opened, or ring-cleaved products depending upon the reaction conditions.

Results

(a) Choice of Amines. The reactivity of bicyclomycin with eight different amines was assessed. The primary amines methylamine (**7**) and ethylamine (**8**), and the heteroaromatic amines imidazole



(**9**) and benzimidazole (**10**) were chosen as simple models of

(1) Presented, in part, at the 3rd Chemical Congress of North America, Toronto, Canada, June 1988, Abstract ORGN 287.
(2) Williams, R. M.; Durham, C. A. *Chem. Rev.* **1988**, *88*, 511 and references therein.
(3) Tanaka, N. *Antibiotics (N.Y.)* **1979**, *5*, 18.
(4) (a) Miyoshi, T.; Miyari, N.; Aoki, H.; Kohsaka, M.; Sakai, H.; Imaka, H. *J. Antibiot.* **1972**, *25*, 569. (b) Kamiya, T.; Maeno, S.; Hashimoto, M.; Mine, Y. *Ibid.* **1972**, *25*, 576. (c) Nishida, M.; Mine, Y.; Matsubara, T. *Ibid.* **1972**, *25*, 582. (d) Nishida, M.; Mine, Y.; Matsubara, T.; Goto, S.; Kuwahara, S. *Ibid.* **1972**, *25*, 594.
(5) (a) Someya, A.; Iseki, M.; Tanaka, N. *J. Antibiot.* **1979**, *32*, 402. (b) Tanaka, N.; Iseki, M.; Miyoshi, T.; Aoki, H.; Imanaka, H. *Ibid.* **1976**, *29*, 155. (c) Someya, A.; Iseki, M.; Tanaka, N. *Ibid.* **1978**, *31*, 712.
(6) Williams, R. M.; Armstrong, R. W.; Dung, J.-S. *J. Med. Chem.* **1985**, *28*, 733.
(7) Pisabarro, A. G.; Canada, F. J.; Vasquez, D.; Arriaga, P.; Rodriguez-Tebar, A. J. *J. Antibiot.* **1986**, *34*, 914.
(8) (a) Williams, R. M.; Tomizawa, K.; Armstrong, R. W.; Dung, J.-S. *J. Am. Chem. Soc.* **1985**, *107*, 6419. (b) Williams, R. M.; Tomizawa, K.; Armstrong, R. W.; Dung, J.-S. *Ibid.* **1987**, *109*, 4028.
(9) Abuzar, S.; Kohn, H. *J. Am. Chem. Soc.* **1988**, *110*, 4089.
(10) Kohn, H.; Abuzar, S. *J. Am. Chem. Soc.* **1988**, *110*, 3661.
(11) Kohn, H.; Abuzar, S. *J. Org. Chem.* **1988**, *53*, 2769.

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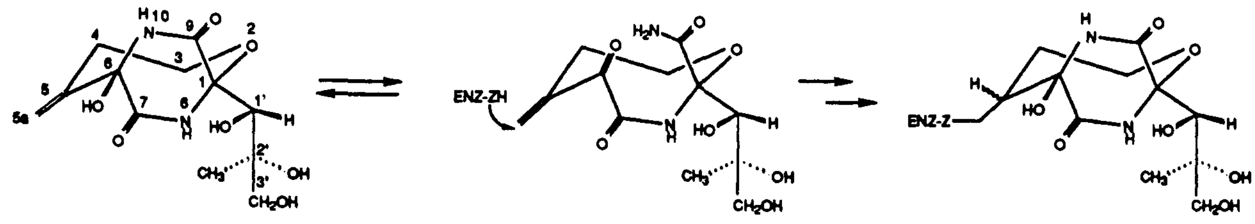
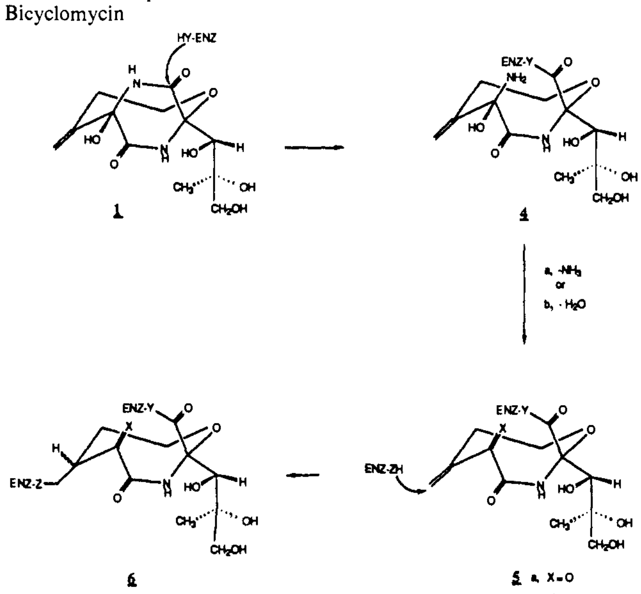
Scheme I. Proposed Mechanism for the Mode of Action of Bicyclomycin



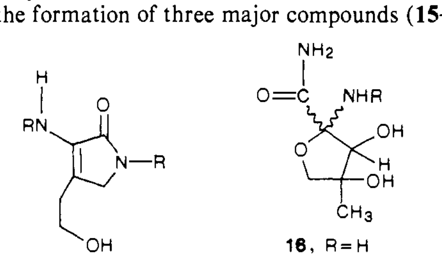
1 2 3

Scheme II. Proposed Mechanism for the Mode of Action of Bicyclomycin



functionalized derivatives of lysine and histidine, respectively. Both of these amino acids may play an important role in the drug activation and binding process. In addition to **9** and **10**, the simple histidine adduct (*dl*)-N_α-benzoylhistidine methylamide (**11**) was incorporated into this study. The pH of these reactions (i.e., amines **7** and **8**, pH 12.5; amines **9**–**11**, pH 9.9–10.6) was governed by the inherent reactivity of the amine[12a] and its basicity (pK_a).[12] The high-pH conditions employed in the primary amine (estimated pK_a = 10.8–12.5[12b]) transformations led us to examine the reactivity of **1** with the weaker secondary amines (estimated pK_a ≈ 8.2–9.8[12b]) morpholine (**12**), ethyl piperazinecarboxylate (**13**), and *N*-methylpiperazine (**14**). The reduced basicity of amines **12**–**14** permitted these reactions to proceed at lower pH values (i.e., pH 8.3–10.8) than those utilized for **7** and **8**. These conditions approximated those utilized in our previous studies of bicyclomycin with thiols.[9]

(b) Primary Amines. Treatment of **1** with excess methylamine (**7**) led to the formation of three major compounds (**15**–**17**) and



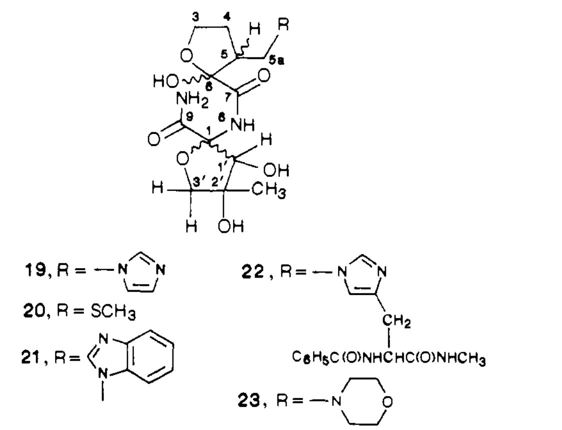
15, R = CH_3
18, R = CH_2CH_3

16, R = H
17, R = CH_3

an unidentified minor product. At the conclusion of the reaction no starting material was observed (TLC analysis). All three products were readily identified on the basis of their observed spectral properties. Lactam **15** displayed three downfield signals in the proton-decoupled ^{13}C NMR spectrum at 116.55, 137.23, and 170.96 ppm, which have been attributed to carbons 4, 3, and 2, respectively. A similar pattern has been reported for 2-(5*H*)-furanones.[11] Inspection of the proton-decoupled ^{13}C NMR spectrum for **16** and **17** indicated that each adduct existed as a pair of diastereomers present in a 1:1 ratio. In agreement with the proposed structures, both compounds exhibited signals in the ^{13}C NMR spectrum between 93 and 95 ppm for the hemiaminal carbon atom.[13] The electron-impact mass spectrum of **16** and **17** displayed prominent ions at *m/e* 132 and 146, respectively. High-resolution mass spectral analyses of these fragments was consistent with the loss of a carbamyl (C(O)NH_2) moiety from the parent ion of each compound.

Reaction of bicyclomycin with excess ethylamine (**8**) gave a comparable result (TLC analysis). Repeated PTLC of the product mixture permitted the isolation of **18** and **16**.

(c) Heteroaromatic Amines. A different product profile was observed for the reaction of bicyclomycin with a slight excess of imidazole (**9**), benzimidazole (**10**), and (*dl*)-N_α-benzoylhistidine methylamide (**11**). TLC analysis of the reaction of **1** with **9** indicated the absence of bicyclomycin and the presence of four adducts (**19a**–**d**) having similar R_f values (R_f 0.38–0.26, 20%



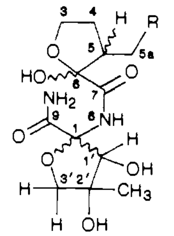
methanol–chloroform). Repeated PTLC (more than four times), permitted the isolation of all four compounds in sufficient quantities to permit their structural elucidation. Compounds **19a**–**d** exhibited a parent ion (M + 1) in the FAB mass spectrum at *m/e* 371, in agreement with the formation of a 1:1 adduct ($C_{15}H_{22}N_4O_7$) between bicyclomycin and imidazole. Inspection of the proton-decoupled ^{13}C NMR spectra for **19a**–**d** in methanol-d_4 indicated that each chromatographic fraction consisted of a single diastereomer. Interestingly, the proton-decoupled ^{13}C NMR spectrum for **19b** in dimethyl-d_6 sulfoxide was considerably more complex than the spectrum observed in methanol-d_4. This phenomenon may reflect the formation of specific conformational isomers in the polar, aprotic solvent dimethyl sulfoxide.[14] Further analysis of the ^{13}C NMR spectra (Table I) for **19a**–**d** revealed

(12) (a) Pearson, R. G.; Sobel, H.; Songstad, J. *J. Am. Chem. Soc.* **1968**, *90*, 319. (b) For a compilation of the pK_a values for the conjugate acids of these amines, see: Perrin, D. D. *Dissociation Constants*; Butterworth: London, 1965.

(13) For related ^{13}C NMR assignments, see: (a) Cortes, S.; Kohn, H. *J. Org. Chem.* **1983**, *48*, 2246. (b) Liao, Z. K.; Kohn, H. *J. Org. Chem.* **1984**, *49*, 3812 and references therein.

(14) Wuthrich, K. *NMR of Proteins and Nucleic Acids*; Wiley: New York, 1986.

Table I. Characteristic ^{13}C NMR Data for Bicyclomycin Ring-Opened Compounds[a]



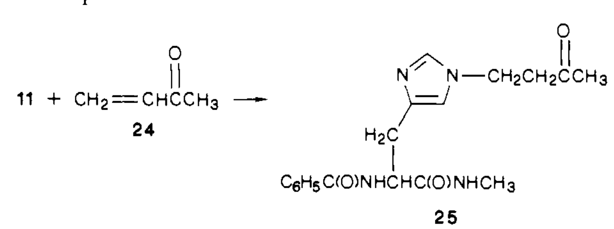
compd	C(1)	C(3)	C(4)	C(5)	C(5a)	C(6)	C(1′)	C(2′)	C(2′)CH_3	C(3′)	C_{Ar}(2″)	C_{Ar}(4″)	C_{Ar}(5″)
19a	94.69	68.99	29.53	47.28[b]	*b*, *c*	108.83	78.75[b]	80.10[b]	21.05	80.65[b]	139.06	129.15	121.20
19b	94.65	68.82	30.05	47.58[b]	*b*, *c*	102.90	78.68	80.06[b]	21.07	81.36[b]	138.68	129.09	120.84
19b[d]	92.38	66.73	27.94	45.50[b]	47.06	101.52	77.18[b]	78.02[b]	20.74	78.55[b]	137.41	128.17	119.40
		67.23	28.63	45.96	47.16		77.22	78.47	20.85	78.73	137.73	128.48	119.99
19c,d	89.69	68.03	30.02	47.82[b]	*b*, *c*	102.85	77.06[b]	78.05[b]	22.55	79.59	138.70	129.11	120.91
	93.50	68.43	30.15			102.94	77.46	78.14	23.19	80.59	138.89		
21a	94.67	69.08	29.40	45.27[b]	47.44	103.07	78.68[b]	80.01[b]	21.06	80.68[b]	145.37 or 143.95		
21b	94.70	68.89	30.14	45.62[b]	*b*, *c*	106.70	78.63[b]	79.99[b]	21.11	81.50[b]	144.91 or 144.09		
21c	89.74	68.80	30.27	45.80[b]	*b*, *c*	106.85	77.08[b]	78.17	23.15	80.64	144.90 or 144.07		
22a	94.60	68.64	29.46	47.34[b]	*b*, *c*	102.77	78.69[b]	80.09[b]	21.13	81.01[b]	138.56, 138.84		
	94.67	68.77	30.02	47.55			79.88	80.84		81.37	138.76, 138.98		
		68.91					79.93						
22b	89.70	68.72	30.08	47.45[b]	*b*, *c*	102.94	77.04[b]	78.05	23.19	80.61[b]	138.58, 138.69		
	89.78	68.92		47.75				78.14	23.25		138.78		
23	89.64	68.55	30.47	43.61	59.37	101.51	76.99	78.68	21.09	80.59			
			30.76	44.04		104.33	77.42	79.58	22.49	81.30			
			31.10				78.03	80.23	23.29				

[a] The number in each entry is the chemical shift value (δ) observed in ppm relative to Me_4Si. All spectra were obtained at 75.5 MHz. The solvent used was CD_3OD unless otherwise indicated. [b] These peaks may be interchanged. [c] The peak for this carbon atom was obscured by the signal for the solvent. [d] The solvent used was DMSO-d_6.

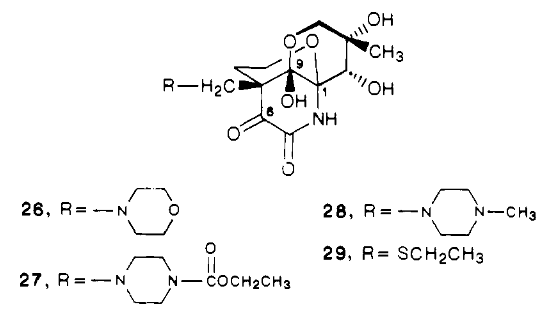
several additional features that proved helpful in the assignment of structure. In particular, the C(6) resonances appeared between 102.83 and 102.94 ppm, while signals for C(1′) and C(3′) were detected between 77.06 and 81.36 ppm. These resonances are significantly downfield from the corresponding signals in **1**,[15] suggesting that extensive reorganization of the bicyclomycin ring system had taken place. The ^{13}C NMR spectra coupled with the ^{1}H and COSY NMR and mass spectral data supported the proposed general structure **19** for all four imidazole products. The precise stereochemical assignment of each individual adduct was not determined. Significantly, compound **19** is analogous to the revised structure **20** for the bicyclomycin–sodium methanethiolate adduct obtained under basic conditions.[10]

A comparable result was observed for the reaction of bicyclomycin with benzimidazole (**10**). Separation of the product mixture by PTLC (two developments) gave three adducts (R_f 0.5–0.4, 20% methanol–chloroform) whose spectral properties were compatible with the proposed ring-opened structure **21**. The ^{13}C NMR spectral data for **21a–c** are listed in Table I and were in excellent agreement with the values observed for the imidazole adducts **19**.

The imidazole-mediated reaction has been extended to the functionalized amino acid derivative (*dl*)-N_α-benzoylhistidine methylamide (**11**). Treatment of **1** with **11** led to the isolation of two distinct fractions **22a** and **22b** (R_f 0.42 and 0.30, 20% methanol–chloroform) after PTLC (two developments). The parent ion observed in the mass spectrum of both materials was consistent with the formation of a 1:1 adduct ($C_{26}H_{34}N_6O_9$). The corresponding ^{13}C NMR spectra for these samples indicated that both **22a** and **22b** existed as a mixture of diastereomers. Significantly, the patterns observed in the ^{1}H and ^{13}C NMR (Table I) spectra for these compounds were in accord with the proposed ring-opened adduct **22**. Close inspection of the proton-decoupled ^{13}C NMR spectrum permitted the assignment of the site of attachment on the imidazole ring nucleus. Previous NMR studies have documented that notable differences exist between the N(1),C(4)- and the N(1),C(5)-substituted compounds.[16] Both **22a** and **22b** exhibited signals in the regions of 118 and 138 ppm for the imidazole ring carbon atoms. This pattern is typical of N(1),C(4) substitution. The regioselectivity of the bicyclomycin–N_α-benzoylhistidine methylamide process was mirrored by the reaction of **11** with 3-buten-2-one (**24**). A single adduct **25** was isolated. The ^{13}C NMR spectrum of this product was in agreement with the proposed N(1),C(4)-imidazole substitution pattern.



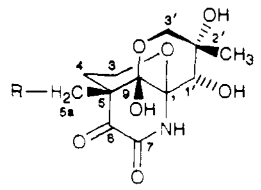
(d) Secondary Amines. Our survey of the reactivity of bicyclomycin with organic bases concluded with the cyclic amines morpholine (**12**), ethyl piperazinecarboxylate (**13**), and *N*-methylpiperazine (**14**). Treatment of **1** with a slight excess of each of these amines under moderately basic conditions (pH 10.2–10.8) yielded the corresponding C(5a)-substituted products **26–28**, respectively, along with trace amounts of unidentified



(15) For a comprehensive study of the NMR spectral properties of bicyclomycin, see: Kohn, H.; Abuzar, S.; Korp, J. D.; Zektzer, A.; Martin, G. E. *J. Heterocycl. Chem.* **1988**, *25*, 1511.

(16) (a) Begtrup, M.; Elguero, J.; Faure, R.; Camps, P.; Estopa, C.; Ilavsky, D.; Fruchier, A.; Marzin, C.; de Mendoza, J. *Magn. Reson. Chem.* **1988**, *26*, 134. (b) Al-Badr, A. A. *Spectrosc. Lett.* **1983**, *16*, 613. (c) Faure, R.; Vincent, E. J. *Heterocycles* **1983**, *9*, 1713.

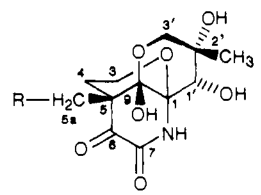
Table II. Characteristic ^{1}H NMR Data for Rearranged Bicyclomycin Compounds **26–28**[a]



compd	C(3)*H*H′	C(3)H*H*′	C(4)*H*H′	C(4)H*H*′	C(5a)*H*H′	C(5a)H*H*′	C(1′)*H*	C(2′)C*H*$_3$	C(3′)*H*H′	C(3′)H*H*′
26	3.50–3.75 (m)	3.94 (dd, *J* = 6.4, 13.6 Hz)	1.41 (dd, *J* = 2.8, 13.6 Hz)	2.07 (app dt, *J* = 6.4, 13.6 Hz)	2.73 (d, *J* = 14.1 Hz)	3.30 (d, *J* = 14.1 Hz)	3.83 (s)	1.16 (s)	3.62 (d, *J* = 12.0 Hz)	4.15 (d, *J* = 12.0 Hz)
27	3.72 (app dt, *J* = 2.6, 13.5 Hz)	3.98 (dd, *J* = 6.3, 13.5 Hz)	1.45 (dd, *J* = 2.6, 13.5 Hz)	2.11 (app dt, *J* = 6.3, 13.5 Hz)	2.80 (d, *J* = 14.3 Hz)	3.30–3.40 (m)	3.87 (s)	1.20 (s)	3.68 (d, *J* = 12.0 Hz)	4.21 (d, *J* = 12.0 Hz)
28	3.66 (app dt, *J* = 2.5, 13.6 Hz)	3.93 (dd, *J* = 6.3, 13.6 Hz)	1.40 (dd, *J* = 2.5, 13.6 Hz)	2.06 (app dt, *J* = 6.3, 13.6 Hz)	2.75 (d, *J* = 14.2 Hz)	3.22–3.38 (m)	3.82 (s)	1.15 (s)	3.62 (d, *J* = 11.9 Hz)	4.14 (d, *J* = 11.9 Hz)

[a] The number in each entry is the chemical shift value (δ) observed in ppm relative to Me_4Si, followed by the multiplicity of the signal and the coupling constant(s) in hertz. All spectra were recorded at 300 MHz, and the solvent used was CD_3OD. The ^{1}H NMR assignments were verified from the corresponding COSY spectrum.

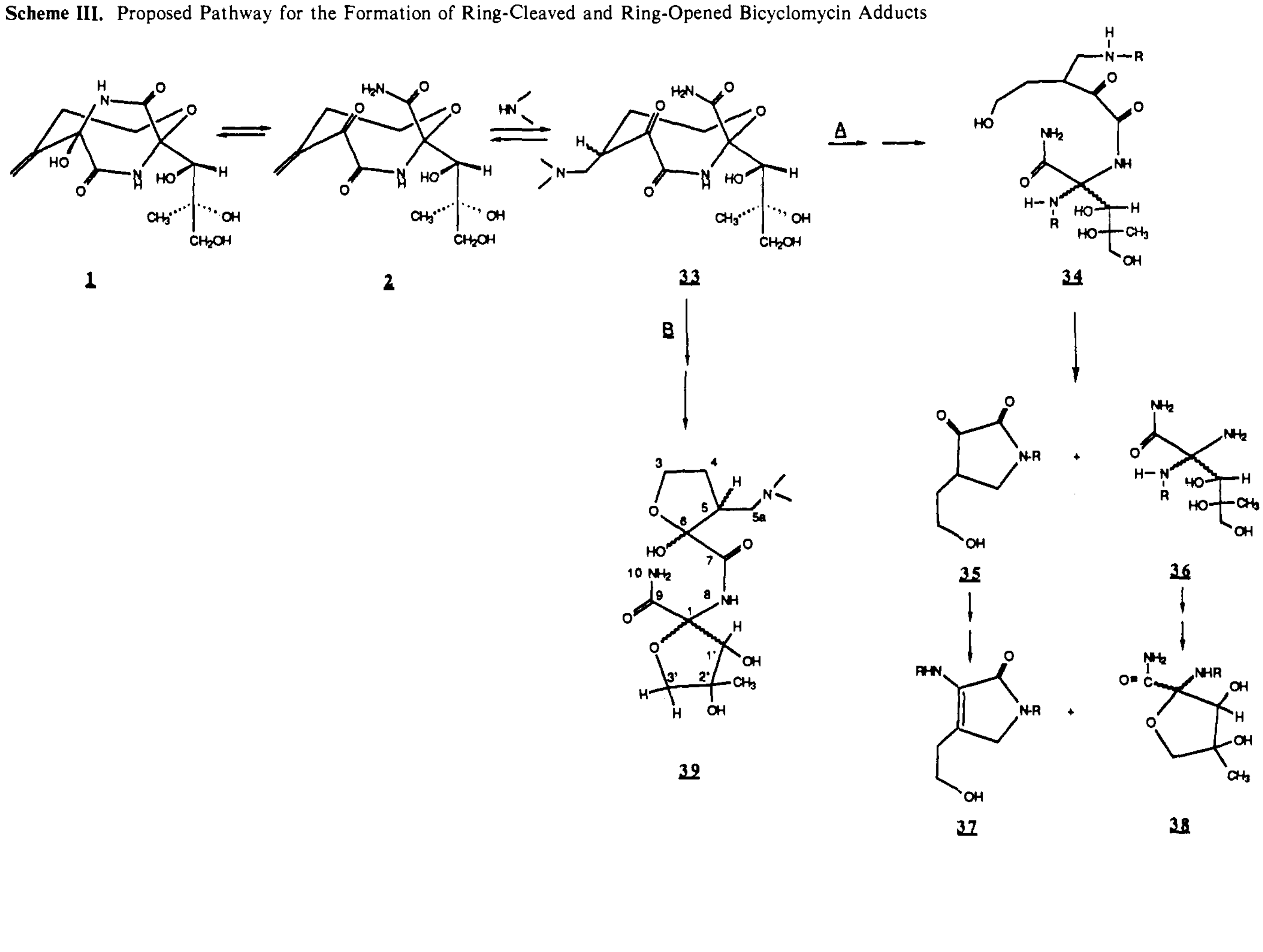
Table III. Characteristic ^{13}C NMR Data for Rearranged Bicyclomycin Compounds **26–28**[a]



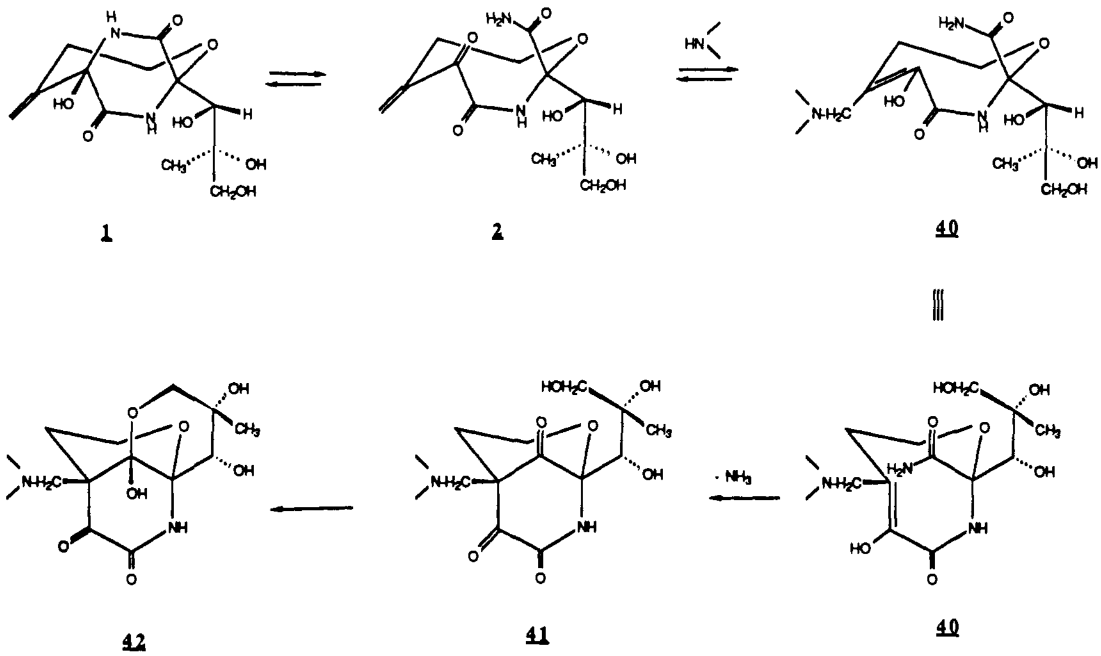
compd	C(1)	C(3)	C(4)	C(5)	C(5a)	C(6)	C(9)	C(1′)	C(2′)	C(2′)*C*H$_3$	C(3′)
26	84.99	60.42	32.77	58.20	54.61	196.33	96.35	70.19[b]	71.87[b]	21.08	72.63[b]
27	84.97	59.85	32.71	58.17	54.68[c]	196.20	96.31	70.22[b]	71.83[b]	21.08	72.66[b]
28	84.97	59.59	32.76	58.20	54.53[d]	196.27	96.31	70.16[b]	71.84[b]	21.07	72.65[b]

[a] The number in each entry is the chemical shift value (δ) observed in ppm relative to Me_4Si. All spectra were obtained at 75.5 MHz. The solvent used was CD_3OD unless otherwise indicated. [b] These peaks may be interchanged. [c] This peak may be interchanged with the signal (55.27 ppm) tentatively assigned for the ethyl piperazinecarboxylate ring. [d] This peak may be interchanged with the signals (55.12 and 56.20 ppm) tentatively assigned for the *N*-methylpiperazine ring.

Scheme III. Proposed Pathway for the Formation of Ring-Cleaved and Ring-Opened Bicyclomycin Adducts

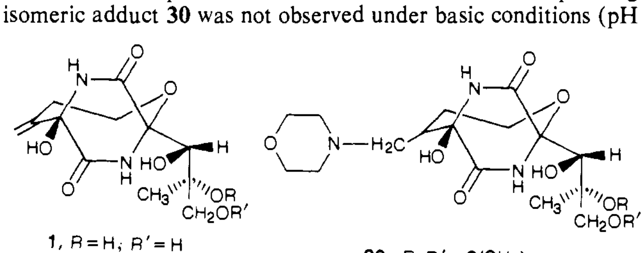


Scheme IV. Proposed Pathway for the Generation of Rearranged Bicyclomycin Adducts



compounds. Reduction of the pH of the solution (pH 8.3) led to decreased amounts of products (TLC analysis). Support for the proposed structural assignments for **26–28** derived from several key spectral observations. In particular, the C(5a)-methylene protons in the ^{1}H NMR spectra (Table II) for compounds **26–28** appeared as a distinct AB pair between δ 2.7 and 3.4. In the ^{13}C NMR spectra (Table III), diagnostic signals were detected at approximately 85, 96, and 196 ppm and have been assigned to carbons 1, 9, and 6, respectively.[17] These chemical shifts compared favorably with the values previously observed for the ethyl mercaptan-bicyclomycin adduct **29** obtained under "neutral-pH" conditions.[9] In the case of **29**, definitive proof of structure was obtained by X-ray crystallographic analysis.[9] Of note, only a single set of signals was observed in the proton-decoupled ^{13}C NMR spectra for **26–28**, indicating that the secondary amine mediated transformations proceeded in a stereoselective manner.

Treatment of bicyclomycin with morpholine under more basic conditions (pH 12.5) led to a dramatically different product profile. Compound **26** was not detected, but a more polar adduct was isolated as a diastereomeric mixture. This compound has been tentatively identified as the ring-opened product **23**. Compatible with this structure, the proton-decoupled ^{13}C NMR spectrum for **23** (Table I) displayed signals at approximately 103, 81, and 78 ppm for carbons C(6), C(1′), and C(3′), respectively.[10] In the case of the morpholine-mediated reactions, the corresponding isomeric adduct **30** was not observed under basic conditions (pH



8.3–12.5). This compound, however, could be prepared by initial conversion of **1** to the 2′,3′-acetonide **31**,[18] followed by treatment with morpholine ("pH" 10.6) to give **32**, and then deprotection with 50% aqueous acetic acid. The proton-decoupled ^{13}C NMR spectrum for **32** displayed a single set of lines providing evidence that the addition of morpholine to the exocyclic methylene group in **31** yielded a single stereoisomer.

Discussion

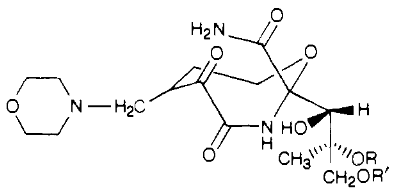
The ring-cleaved (i.e., **15–18**), ring-opened (i.e., **19**, **21**, and **22**) and rearranged (i.e., **26–28**) products produced in the amine-mediated transformations may stem from a *common intermediate*. *We suggest that the key step is the rupture of the hemiaminal group at C(6) to give enone **2***. Subsequent conjugate addition of the amine furnishes **33**. In the case of methylamine and ethylamine (Scheme III, route A), this step can be followed by cleavage of the aminal bond at C(1) and then trapping of the resulting imine by the excess primary amine present in the solution to yield **34**. Intramolecular acyl bond cleavage of the pyruvamide-type (C(7)–N(8)) bond by the amine at C(5a) then gives **35** and **36**, which can cyclize to furnish **37** and **38** (i.e., **15–18**). A similar pathway (Scheme III, route B) is envisioned initially for the imidazole, benzimidazole, and N_α-benzoylhistidine methylamide mediated reactions. In these transformations, however, Michael addition to enone **2** generates the fully substituted amine **33**. This species is not likely to undergo an intramolecular acyl substitution reaction at C(7) in a subsequent step. Accordingly, cleavage of the aminal group at C(1) in these processes ultimately furnishes the bis(tetrahydrofuranyl) derivatives **39** (i.e., **19**, **21**, and **22**). Significantly, formation of **39** is envisioned to proceed with the generation of three *new* chiral centers, thereby accounting for the number of isomeric products isolated in each reaction. Interestingly, the formation of the ring-opened adducts with the heteroaromatic amines (i.e., **1** → **39**) proceeded at lower pH values than the thiolate-induced transformations (i.e., **1** → **20**).[5a,10] This observation suggests that the rupture of the aminal linkage at C(1) is general-base-catalyzed. A related process has been proposed for the cleavage of the C(6)–N(10) bond in **1** upon reaction of bicylcomycin with thiolates.[8]

Enone **2** is also projected as a key intermediate in the formation of the novel rearranged adducts **26–28** (Scheme IV). Conjugate addition of the secondary amine to **2** generates **33** and the corresponding tautomer **40**. Enol **40** is ideally situated to undergo an intramolecular mixed-Claisen reaction to produce **41** and ammonia. Cyclization of **41** in the final step yields the observed hemiketal **42** (i.e., **26–28**). Significantly, the mild conditions employed in the secondary amine mediated transformations should minimize alternative reaction processes (i.e., C(1)–O(2) bond cleavage).

A comparable hypothesis can be invoked to account for the generation of **32** from acetonide **31** and morpholine. In this

(17) The numbering system employed for **1** has been retained for compounds **26–28** and is depicted in Tables II and III.
(18) Kamiya, T.; Maeno, S.; Kitaura, Y. Belgium Patent 847 475.

specific scenario, initial enone formation is followed by formation of the C(5a)-substituted adduct **43**. This species cannot isomerize



43, R, R′ = $C(CH_3)_2$

to the thermodynamically more stable bis(tetrahydrofuranyl) product.[19] Accordingly, closure of the piperazinedione ring regenerates the bicyclomycin-ring skeleton to give **32**.

Conclusions

The amine-mediated bicyclomycin transformations yielded a spectrum of products that have provided useful information concerning the pathway for the chemical activation of **1**. The type of adduct generated hinged upon the amine employed and the pH of the reaction medium. In all cases, C(5a)-functionalized products were produced. Formation of these adducts can be rationalized by initial cleavage of the C(6)-hemiaminal bond of **1** to generate enone **2**. Moreover, under moderate pH conditions, a novel rearrangement (**1** → **42**) of the bicyclomycin ring system was discovered.

These results suggest that bicyclomycin undergoes activation by a chemical-mediated pathway. In this scenario, the initial step is the *reversible* ring opening of the C(6)–N(10) bond to generate enone **2**. The efficiency of the subsequent drug-binding process (i.e., **2** → **33** (**40**)) is expected to be dependent upon the environment (i.e., medium, pH), the biological receptor (nucleophile), and the effective concentration of the nucleophile. The finding that bicyclomycin reacts with secondary amines to yield the novel rearranged adducts **42** may have added biological significance. The proposed intramolecular mixed-Claisen transformation (Scheme IV) generates the highly reactive ring system **41**. Piperidinetrione **41** may be capable of undergoing further transformations (i.e., drug binding) necessary for the mode of action of the antibiotic.

Experimental Section

General Methods. Infrared spectra (IR) were run on a Perkin-Elmer 283, an IBM IR-32, or a Nicolet 10DX FT spectrometer and calibrated against the 1601-cm^{-1} band of polystyrene. Absorption values are expressed in wavenumbers (cm^{-1}). Proton (^{1}H NMR) and carbon (^{13}C NMR) nuclear magnetic resonance spectra were taken on Nicolet NT-300 and General Electric QE-300 NMR instruments. Chemical shifts (δ) are in parts per million (ppm) relative to Me_4Si and coupling constants (J values) are in hertz. Low-resolution electron-impact mass spectral data (MS) were obtained at an ionizing voltage of 70 eV on a Bell and Howell 21-491 mass spectrometer at the University of Texas–Austin. The low-resolution chemical-ionization mass spectral studies conducted at the University of Texas were run on either a Finnegan MAT 4023 or a TSQ-70 instrument, while the low-resolution FAB spectral investigations were conducted on the TSQ-70 instrument. High-resolution electron-impact mass spectra were performed on a CEC 21-110B double-focusing magnetic sector spectrometer at the University of Texas–Austin by Dr. John Chinn. The FAB spectra at the Baylor College of Medicine were performed on a VG ZAB-SEQ instrument by Dr. Simon Gaskell and at the University of Houston on a VG 70 SEQ instrument by Dr. R. B. Freas. The chemical-ionization mass spectral studies at the Baylor College of Medicine were performed by Dr. Simon Gaskell on a VG JS250 instrument. Microanalyses were obtained from Spang Microanalytical Laboratory, Eagle Harbor, MI. pH measurements were determined on a pHM26 meter.

All glassware was dried before use. The solvents and reactants were of the best commercial grade available and were used without further purification unless noted. Thin-layer chromatography and thick-layer chromatography were run on precoated silica gel GHLF microscope slides (2.5 × 10 cm; Analtech No. 21521) or silica gel GHLF (20 × 20 cm; Analtech No. 11187).

Treatment of Bicyclomycin with Methylamine (7). A solution (2 mL, pH 12.5) of 4% aqueous methylamine (**7**) (80 mg, 2.68 mmol) and **1** (50 mg, 0.165 mmol) was stirred at room temperature (15 h). The solvent was removed in vacuo and the crude product was purified by PTLC with 20% methanol–chloroform as the eluent to give three distinct fractions (R_f 0.73 (**15** and an unidentified compound), 0.35 (**17**), 0.20 (**16**); 20% methanol–chloroform). The more mobile component was further purified by PTLC with 3% methanol–chloroform as the eluent (three developments) to yield **15** (R_f 0.70, 20% methanol–chloroform). The following properties were obtained for each isolated compound.

Compound **15** (3.5 mg, 12%): as a semisolid; FTIR (KBr) 1689 cm^{-1}; ^{1}H NMR (CD_3OD) δ 2.66 (t, J = 6.5 Hz, 2 H, C(4)CH_2CH$_2$OH), 2.89 (s, 3 H, NHCH_3), 2.99 (s, 3 H, NCH$_3$), 3.67 (t, J = 6.5 Hz, 2 H, C(4)CH$_2$CH_2OH), 3.80 (s, 2 H, C(5)H$_2$); ^{13}C NMR (CD_3OD) 29.53 (C(4)CH$_2$CH$_2$OH), 31.04 (CH$_3$), 32.54 (CH$_3$), 54.87 (C(5)), 62.43 (C(4)CH$_2$$CH_2$OH), 116.55 (C(4)), 137.23 (C(3)), 170.96 (C(2)) ppm; MS (EI) m/e (relative intensity) 170 (53), 139 (100), 110 (77), 94 (31), 58 (51); M_r (EI) 170.10565 (calcd for $C_8H_{14}N_2O_2$, 170.10553).

Compound **17** (5.4 mg, 17%): oil, FTIR (Nujol) 1666 cm^{-1}; ^{1}H NMR (CD_3OD) δ 1.26 (s, 3 H, CH$_3$), 2.74 (s, 3 H, NHCH_3), 3.71–3.90 (m, 3 H, CH_2, C(OH)H); ^{13}C NMR (CD_3OD) 22.37, 22.47 (CH$_3$), 26.30, 28.16 (NCH$_3$), 77.38, 77.62 (CH$_2$ or C(OH)H or C(OH)CH$_3$), 77.52, 80.31 (C(OH)CH$_3$ or C(OH)H or CH$_2$), 79.55, 85.19 (C(OH)H, or C(OH)CH$_3$, or CH$_2$), 93.54 (H$_2$NC(O)CNHCH$_3$), 175.32 (CO) ppm; MS (+CI) m/e (relative intensity) 191 [M + 1, 48]$^+$, 174 (40), 146 (11), 131 (13), 117 (100); MS (EI) m/e (relative intensity) 146 (M$^+$ − CONH$_2$, 21), 132 (100), 127 (29), 83 (34), 58 (99); M_r (EI) 146.08193 (calcd for $C_6H_{12}NO_3$, M$^+$ − CONH$_2$, 146.08172), 132.06626 (calcd for $C_5H_{10}NO_3$, M$^+$ − CONH$_2$ − CH$_2$, 132.06607).

Compound **16** (6.5 mg, 22%): as a semisolid; FTIR (KBr) 1684 cm^{-1}; ^{1}H NMR (CD_3OD) δ 1.28 (s, 3 H, CH$_3$), 3.75–3.91 (m, 3 H, CH$_2$, C(OH)H); ^{13}C NMR (CD_3OD) 21.40, 22.52 (CH$_3$), 77.36, 77.45 (CH$_2$ or C(OH)CH$_3$ or C(OH)H), 77.55, 78.05 (C(OH)CH$_3$ or C(OH)H or CH$_2$), 79.61, 85.21 (C(OH)H or CH$_2$ or C(OH)CH$_3$), 93.46 (H$_2$NC(O)CNH$_2$), 177.62 (CO) ppm; MS (EI) m/e (relative intensity) 132 (M$^+$ − CONH$_2$, 100), 74 (85), 73 (84); M_r (EI) 132.06613 (calcd for $C_5H_{10}NO_3$, M$^+$ − CONH$_2$, 132.06607).

Treatment of Bicyclomycin with Ethylamine (8). A solution (pH 12.5) of **1** (25 mg, 0.082 mmol) in 4% aqueous ethylamine (**8**) (1 mL, 0.88 mmol) was stirred at room temperature (15 h). The solvent was removed in vacuo and the residue was purified by PTLC with 20% methanol–chloroform as the eluent to give two distinct fractions (R_f 0.70 (**18** and an unidentified compound), 0.20 (**16**); 20% methanol–chloroform). The more mobile fraction was further purified by PTLC with 3% methanol–chloroform as the eluent (three developments) to yield **18** (R_f 0.70, 20% methanol–chloroform).

Compound **18**: (2.5 mg, 15%) as a semisolid; ^{1}H NMR (CD_3OD) δ 1.15 (t, J = 7.0 Hz, 3 H, CH$_2$CH_3), 1.16 (t, J = 7.1 Hz, 3 H, CH$_2$CH_3), 2.62 (t, J = 6.6 Hz, 2 H, C(4)CH_2CH$_2$OH), 3.24 (q, J = 7.0 Hz, 2 H, CH_2CH$_3$), 3.45 (q, J = 7.1 Hz, 2 H, CH_2CH$_3$), 3.67 (t, J = 6.6 Hz, 2 H, C(4)CH$_2$CH_2OH), 3.83 (s, 2 H, C(5)H$_2$); ^{13}C NMR (CD_3OD) 13.84 (CH$_3$), 15.91 (CH$_3$), 31.18 (C(4)CH$_2$CH$_2$OH), 38.27 (CH$_2$CH$_3$), 40.69 (CH$_2$CH$_3$), 52.28 (C(5)), 62.11 (C(4)CH$_2$$CH_2$OH), 117.21 (C(3)), 135.79 (C(4)), 170.40 (C(2)) ppm; MS (EI) m/e (relative intensity) 198 (24), 167 (53), 124 (100), 96 (42), 82 (30), 68 (26), 56 (24); M_r (EI) 198.13698 (calcd for $C_{10}H_{18}N_2O_2$, 198.13683).

Compound **16** (2.0 mg, 14%): as a semisolid; ^{1}H NMR (CD_3OD) δ 1.28 (s, 3 H, CH$_3$), 3.75–3.91 (m, 3 H, CH$_2$, C(OH)H).

Treatment of Bicyclomycin with Imidazole (9). A solution of **1** (50 mg, 0.165 mmol) and **9** (17 mg, 0.25 mmol) in water (5 mL) was stirred at room temperature (15 h) at pH 10.5. The solvent was removed in vacuo at 40 °C and the residue was purified by PTLC with 25% methanol–chloroform as the eluent to give a mixture of **19a–d** (43 mg). This mixture was further purified by PTLC with 15% methanol–chloroform (four developments) as the eluent to give the following compounds.

Compound **19a**: yield, 8 mg (13%) as a semisolid; R_f 0.38 (20% methanol–chloroform); FTIR (KBr) 1685, 1500 cm^{-1}; ^{1}H NMR (CD_3OD) δ 1.28 (s, 3 H, CH$_3$), 1.91–2.01 (m, 2 H, C(4)HH′), 2.82–2.89 (m, 1 H, C(5)H), 3.90–3.96 (m, 2 H, C(3′)HH′, C(3)HH′), 4.12–4.22 (m, 2 H, C(3)HH', C(5a)HH′), 4.25–4.33 (m, 3 H, C(3′)HH', C(5a)HH', C(1′)H), 6.92 (br s, 1 H, C(5″)H), 7.14 (br s, 1 H, C(4″)H), 7.64 (br s, 1 H, C(2″)H). The ^{1}H NMR assignments were confirmed by the corresponding COSY experiment. ^{13}C NMR (CD_3OD) 21.05 (CH$_3$), 29.63 (C(4)), 47.28 (C(5a)), 68.99 (C(3)), 78.75 (C(1′) or C(2′) or C(3′)), 80.10 (C(2′) or C(1′) or C(3′)), 80.65 (C(3′) or C(2′) or C(1′)), 94.69 (C(1)), 102.83 (C(6)), 121.20 (C(5″)), 129.15 (C(4″)), 139.06 (C(2″)), 172.43 (C(7) or C(9)), 173.06 (C(9) or C(7)) ppm. The signal for the C(5a) carbon resonance was confirmed by the reverse-detected

(19) Maag, H.; Blount, J. F.; Coffen, D. L.; Steppe, T. V.; Wong, F. *J. Am. Chem. Soc.* **1978**, *100*, 6786.

^1H–^{13}C heteronuclear shift correlation experiment.[20] The peak for the C(5) carbon is presumed to reside beneath the signal for the solvent. MS (+FAB) 371 $[M + 1]^+$; M_r (+FAB) 371.1589 (three determinations) (calcd for $C_{15}H_{23}N_4O_7$, $[M + 1]^+$ 371.1567).

Compound **19b**: yield, 6 mg (10%) as a semisolid; R_f 0.34 (20% methanol–chloroform); FTIR (KBr) 1684, 1512 cm^{-1}; ^1H NMR (CD_3OD) δ 1.28 (s, 3 H, CH_3), 1.93–1.97 (m, 2 H, C(4)HH′), 2.68–2.73 (m, 1 H, C(5)H), 3.88–3.99 (m, 2 H, C(3))*H*H′, C(3′)*H*H′), 4.02–4.18 (m, 2 H, C(3)H*H′*, C(5a)*H*H′), 4.21–4.33 (m, 2 H, C(3′)H*H′*, C(5a)H*H′*), 4.50 (s, 1 H, C(1′)H), 6.88 (br s, 1 H, C(5″)H), 7.07 (br s, 1 H, C(4″)H), 7.60 (br s, 1 H, C(2″)H). The ^1H NMR assignments were confirmed by the corresponding COSY experiment. ^{13}C NMR (CD_3OD) 21.07 (CH_3), 30.05 (C(4)), 47.58 (C(5) or C(5a)), 68.82 (C(3)), 78.68 (C(1′) or C(2′) or C(3′)), 80.06 (C(2′) or C(1′) or C(3′)), 81.36 (C(3′) or C(2′) or C(1′)), 94.65 (C(1)), 102.88 (C(6)), 120.84 (C(5″)), 129.09 (C(4″)), 138.68 (C(2″)), 172.66 (C(7) or C(9)), 172.72, 172.99 (C(9) or C(7)) ppm. An unattributed signal at 68.99 (68.82) ppm was observed. The peak for the C(5) or C(5a) carbon is presumed to reside beneath the signal for the solvent. ^{13}C NMR (DMSO-d_6) 20.74, 20.85 (CH_3), 27.94, 28.63 (C(4)), 45.50, 45.96 (C(5) or C(5a)), 47.06, 47.16 (C(5a) or C(5)), 66.73, 67.23 (C(3)), 77.18, 77.22, 78.02, 78.47, 78.55, 78.73 (C(1′), C(2′), C(3′)), 92.38 (C(1)), 101.52 (C(6)), 119.40, 119.99 (C(5″)), 128.17, 128.48 (C(4″)), 137.41, 137.73 (C(2″)), 168.91, 169.31 (C(7) or C(9)), 170.55, 170.66 (C(9) or C(7)) ppm. MS (+FAB) 371 $[M + 1]^+$.

Compounds **19c** and **19d**: yield, 13 mg (21%) as a semisolid; R_f 0.28, 0.26 (20% methanol–chloroform); ^1H NMR (CD_3OD) δ 1.28, 1.33 (s, 3 H, CH_3), 1.85–2.04 (m, 2 H, C(4)HH′), 2.59–2.69 (m, 1 H, C(5)H), 3.75–4.20 (m, 6 H, C(3)HH′, C(3′)HH′, C(5a)*H*H′, C(1′)H), 4.35–4.50 (m, 1 H, C(5a)H*H′*), 6.94 (br s, 1 H, C(5″)H), 7.16 (br s, 1 H, C(4″)H), 7.68 (br s, 1 H, C(2″)H); ^{13}C NMR (CD_3OD) 22.55, 23.19 (CH_3), 30.02, 30.15 (C(4)), 47.82 (C(5) or C(5a)), 68.03, 68.43 (C(3)), 77.06, 77.46 (C(1′) or C(2′) or C(3′)), 78.05, 78.14 (C(2′) or C(1′) or C(3′)), 79.59, 80.59 (C(3′) or C(2′) or C(1′)), 89.69, 93.50 (C(1)), 102.85, 102.94 (C(6)), 120.91 (C(5″)), 129.11 (C(4″)), 138.70, 138.89 (C(2″)), 172.92, 172.99 (C(7) or C(9)), 175.34, 177.60 (C(9) or C(7)) ppm. The peak for the C(5) or C(5a) carbon is presumed to reside beneath the signal for the solvent. The binary mixture (6 mg) was further purified by PTLC with 20% methanol–chloroform as the eluent (five developments) to give pure **19c** and **19d**.

Compound **19c**: yield, 1 mg as a semisolid; R_f 0.28 (20% methanol–chloroform); FTIR (KBr) 1684, 1506 cm^{-1}; ^1H NMR (CD_3OD) δ 1.35 (s, 3 H, CH_3), 1.85–2.03 (m, 2 H, C(4)HH′), 2.60–2.73 (m, 1 H, C(5)H), 3.88–4.16 (m, 6 H, C(3)HH′, C(3′)HH′, C(1′)H, C(5a)*H*H′), 4.49 (dd, *J* = 3.06, 13.3 Hz, 1 H, C(5a)H*H′*), 6.96 (br s, 1 H, C(5″)H), 7.17 (br s, 1 H, C(4″)H), 7.70 (br s, 1 H, C(2″)H). The ^1H NMR assignments were confirmed by the corresponding COSY experiment. MS (+FAB) 371 $[M + 1]^+$; M_r (+FAB) 371.1576 (three determinations) (calcd for $C_{15}H_{23}N_4O_7$, $[M + 1]^+$ 371.1567).

Compound **19d**: yield, 2 mg as a semisolid; R_f 0.26 (20% methanol–chloroform); FTIR (KBr) 1684, 1508 cm^{-1}; ^1H NMR (CD_3OD) δ 1.28 (s, 3 H, CH_3), 1.88–2.04 (m, 2 H, C(4)HH′), 2.68–2.71 (m, 1 H, C(5)H), 3.77–4.18 (m, 6 H, C(3)HH′, C(3′)HH′, C(1′)H, C(5a)*H*H′), 4.35–4.50 (m, 1 H, C(5a)H*H′*), 6.94 (br s, 1 H, C(5″)H), 7.15 (br s, 1 H, C(4″)H), 7.67 (br s, 1 H, C(2″)H); MS (+FAB) 371 $[M + 1]^+$.

Treatment of Bicyclomycin with Benzimidazole (10). A solution of **1** (50 mg, 0.165 mmol) and **10** (20 mg, 0.185 mmol) in tetrahydrofuran–water (1:3) (5 mL) was stirred at room temperature (20 h) at "pH" 10.6. The solvent was removed in vacuo and the residue was dissolved in methanol (5 mL). The insoluble materials were filtered off and the filtrate was concentrated and purified by PTLC with 15% methanol–chloroform (two developments) as the eluent to give the following compounds.

Compound **21a**: yield, 4.5 mg (7%) as a semisolid; R_f 0.50 (20% methanol–chloroform); FTIR (KBr) 1685, 1500 cm^{-1}; ^1H NMR (CD_3OD) δ 1.13 (s, 3 H, CH_3), 1.98–2.18 (m, 2 H, C(4)HH′), 3.00–3.15 (m, 1 H, C(5)H), 3.80–4.00 (m, 3 H, C(3)*H*H′, C(3′)*H*H′, C(1′)H), 4.05–4.25 (m, 2 H, C(3)H*H′*, C(3′)H*H′*), 4.37–4.51 (m, 1 H, C(5a)*H*H′), 4.53–4.60 (m, 1 H, C(5a)H*H′*), 7.24–7.35 (m, 2 H, C(5″,6″)H), 7.58–7.65 (m, 2 H, C(4″,7″)H), 8.14 (s, 1 H, C(2″)H). The ^1H NMR assignments were confirmed by the corresponding COSY experiment. ^{13}C NMR (CD_3OD) 21.06 (CH_3), 29.40 (C(4)), 45.27 (C(5) or C(5a)), 47.44 (C(5) or C(5a)), 69.08 (C(3)), 78.68 (C(1′) or C(2′) or C(3′)), 80.01 (C(2′) or C(1′) or C(3′)), 80.68 (C(3′) or C(2′) or C(1′)), 94.67 (C(1)), 103.07 (C(6)), 111.71 (C(7″)), 120.10 (C(4″)), 123.63 (C(5″)), 124.48 (C(6″)), 134.81 (C(7a″)), 143.95 (C(2″) or C(3a″)), 145.37 (C(3a″) or C(2″)), 172.16 (C(7) or C(9)), 173.13 (C(9) or C(7)) ppm; MS (–CI) 420 $[M]^-$; MS (+FAB) 421 $[M + 1]^+$; MS (–FAB) 420 $[M]^-$.

Compound **21b**: yield, 3.25 mg (5%) as a semisolid; R_f 0.45 (20% methanol–chloroform); FTIR (KBr) 1684, 1506 cm^{-1}; ^1H NMR (CD_3OD) δ 1.29 (s, 3 H, CH_3), 1.96–2.09 (m, 2 H, C(4)HH′), 2.88–2.93 (m, 1 H, C(5)H), 3.88–4.12 (m, 2 H, C(3)*H*H′, C(3′)*H*H′), 4.12–4.19 (m, 2 H, C(3)H*H′*, C(3′)H*H′*), 4.26–4.38 (m, 1 H, C(5a)*H*H′), 4.46 (s, 1 H, C(1′)H), 4.55–4.64 (m, 1 H, C(5a)H*H′*), 7.25–7.33 (m, 2 H, C(5″,6″)H), 7.56–7.65 (m, 2 H, C(4″,7″)H), 8.16 (s, 1 H, C(2″)H). The ^1H NMR assignments were confirmed by the corresponding COSY experiment. ^{13}C NMR (CD_3OD) 21.11 (CH_3), 30.14 (C(4)), 45.62 (C(5) or C(5a)), 68.89 (C(3)), 78.63 (C(1′) or C(2′) or C(3′)), 79.99 (C(2′) or C(1′) or C(3′)), 81.50 (C(3′) or C(1′) or C(2′)), 94.70 (C(1)), 106.70 (C(6)), 111.51 (C(7″)), 120.16 (C(4″)), 123.47 (C(5″)), 124.25 (C(6″)), 134.97 (C(7a″)), 144.09 (C(2″) or C(3a″)), 144.91 (C(3a″) or C(2″)) ppm. The peak for the C(5) or C(5a) carbon is presumed to reside beneath the signal for the solvent. The peaks for the C(7) and C(9) carbons could not be detected. MS (–CI) 420 $[M]^-$; MS (+FAB) 421 $[M + 1]^+$; MS (–FAB) 420 $[M]^-$, 419 $[M - 1]^-$.

Compound **21c**: yield, 5 mg (7%) as a semisolid; R_f 0.40 (20% methanol–chloroform); FTIR (KBr) 1684, 1501 cm^{-1}; ^1H NMR (CD_3OD) δ 1.33 (s, 3 H, CH_3), 1.82–1.88 (m, 1 H, C(4)*H*H′), 2.02–2.09 (m, 1 H, C(4)H*H′*), 2.75–2.86 (m, 1 H, C(5)H), 3.80–4.00 (m, 4 H, C(3)*H*H′, C(1′)H, C(3′)HH′), 4.11–4.18 (m, 1 H, C(3)H*H′*), 4.29–4.37 (m, 1 H, C(5a)*H*H′), 4.64–4.77 (m, 1 H, C(5a)H*H′*), 7.26–7.31 (m, 2 H, C(5″,6″)H), 7.64–7.66 (m, 2 H, C(4″,7″)H), 8.22 (s, 1 H, C(2″)H). The ^1H NMR assignments were confirmed by the corresponding COSY experiment. ^{13}C NMR (CD_3OD) 23.15 (CH_3), 30.27 (C(4)), 45.80 (C(5) or C(5a)), 68.80 (C(3)), 77.08, 78.08, 78.17, 80.64, (C(1′), C(2′), C(3′)), 89.74 (C(1)), 106.86 (C(6)), 111.73 (C(7″)), 120.07 (C(4″)), 123.49 (C(5″)), 124.31 (C(6″)), 134.96 (C(7a″)), 144.07 (C(2″)) or C(3a″)), 144.90 (C(3a″) or C(2″)), 173.00 (C(7) or C(9)), 175.40 (C(7) or C(9)) ppm. An unattributed signal at 78.08 (77.08) ppm was observed. The peak for the C(5) or C(5a) carbon is presumed to reside beneath the signal for the solvent. MS (–CI) 420 $[M]^-$, 419 $[M - 1]^-$; MS (+FAB) 421 $[M + 1]^+$; MS (–FAB) 420 $[M]^-$, 419 $[M - 1]^-$.

N_α-Benzoylhistidine Methylamide (11). N_α-Benzoylhistidine methyl ester[21] (100 mg, 0.32 mmol) was added to a 40% methylamine (**7**) solution (2 mL) and heated at reflux for 6 h. The solvent was removed and the residue was purified by PTLC with 20% methanol–chloroform as the eluent to give the title compound; yield, 50 mg (57%); mp 206 °C (ethanol); R_f 0.50 (20% methanol–chloroform); FTIR (KBr) 1644 cm^{-1}; ^1H NMR (CD_3OD) δ 2.71 (s, 3 H, NHC*H*$_3$), 3.04–3.22 (m, 2 H, CH_2), 4.79 (t, *J* = 6.73 Hz, 1 H, CH), 6.89 (s, 1 H, C(5)H), 7.40–7.50 (m, 3 H, C(3′, 4′, 5′)H), 7.60 (s, 1 H, C(2)H), 7.80 (d, *J* = 7.40 Hz, 2 H, C(2′, 6′)H); ^{13}C NMR (CD_3OD) 26.39 ($NHCH_3$), 30.50 (CH_2), 55.62 (CH), 118.02 (C(5)), 128.44 (C(2′, 6′) or C(3′, 5′)), 129.47 (C(3′, 5′) or C(2′, 6′)), 132.82 (C(4′)), 135.13 (C(1′), C(4)), 136.31 (C(2)), 169.99 (C_6H_5CONH), 174.17 ($CONHCH_3$) ppm.

Anal. Calcd for $C_{14}H_{16}N_4O_2$: C, 61.76; H, 5.88; N, 20.59. Found: C, 61.88; H, 6.00; N, 20.53.

(*dl*)-N_α-Benzoyl-*N*-1-(3-oxobut-1-yl)histidine Methylamide (25). To a solution of **11** (20 mg, 0.073 mmol) in tetrahydrofuran–water (1:3, 1 mL), **24** (10.3 mg, 0.146 mmol) was added and the "pH" of the solution was raised to 10.00. This solution was stirred at room temperature (15 h) and then the solvent was removed in vacuo. The residue was purified by PTLC with 10% methanol–chloroform to give the title compound: yield, 13.5 mg (54%) as a semisolid; R_f 0.70 (15% methanol–chloroform); FTIR (KBr) 1709, 1651 cm^{-1}; ^1H NMR (DMSO-d_6) δ 2.01 (s, 3 H, $COCH_3$), 2.56 (br s, 3 H, NHC*H*$_3$), 2.86–2.91 (m, 4 H, CHC*H*$_2$, $COCH_2$), 4.04 (t, *J* = 6.45 Hz, 2 H, NCH_2), 4.53–4.57 (m, 1 H, C*H*CH$_2$), 6.87 (s, 1 H, C(5)H), 7.45–7.56 (m, 4 H, C(3′, 4′, 5′)H, NH), 7.84–7.87 (m, 3 H, C(2)H, C(2′, 6′)H), 8.64 (d, *J* = 7.42 Hz, 1 H, N*H*CH); ^{13}C NMR (DMSO-d_6) 25.69 ($NHCH_3$), 29.84 (CO*C*H$_3$), 30.30 (CH*C*H$_2$), 40.69 (CO*C*H$_2$), 43.66 (N*C*H$_2$CH$_2$), 53.81 (*C*HCH$_2$), 116.46 (C(5)), 127.39 (C(2′,6′) or C(3′, 5′)), 128.20 (C(3′, 5′) or C(2′, 6′)), 131.26 (C(4′)), 134.20 (C(1′)), 136.83 (C(2)), 137.86 (C(4)), 166.00 (C_6H_5CONH), 171.62 ($CONHCH_3$), 206.24 ($COCH_3$) ppm; MS (+CI) 343 $[M + 1]^+$; M_r (EI) 342.17013 (calcd for $C_{18}H_{22}N_4O_3$, 342.16919).

Treatment of Bicyclomycin with N_α-Benzoylhistidine Methylamide (11). Bicyclomycin (25 mg, 0.082 mmol) was dissolved in tetrahydrofuran–water (1:3) (2.5 mL) and **11** (24 mg, 0.088 mmol) was added. The "pH" of the solution was raised to 9.90 and the reaction mixture was stirred (20 h) at room temperature. The solvent was removed in vacuo and the residue was purified by TLC with 15% methanol–chloroform (two developments) as the eluent to give the following compounds.

Compound **22a**: yield, 8.5 mg (18%) as a semisolid; R_f 0.42 (20% methanol–chloroform); FTIR (KBr) 1678 cm^{-1}; ^1H NMR (CD_3OD) δ

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1.26, 1.30, 1.31, 1.32 (s, 3 H, C(2')CH_3), 1.80–2.10 (m, 2 H, C(4)HH'), 2.60–2.67 (m, 1 H, C(5)H), 2.72, 2.75, 2.77 (s, 3 H, NHCH_3), 2.98–3.19 (m, 2 H, CHCH_2), 3.74–4.55 (m, 7 H, C(3)HH', C(3')HH', C(5a)HH', C(1')H), 4.73–4.77 (m, 1 H, CHCH$_2$), 6.75, 6.90, 6.94 (s, 1 H, C(5'')H), 7.42–7.59 (m, 4 H, C(2'')H, C(3''', 4''', 5''')H), 7.79–7.83 (m, 2 H, C(2''', 6''')H). The ^{1}H NMR assignments were confirmed by the corresponding COSY experiment. ^{13}C NMR (CD_3OD) 21.13 (C(2')CH_3), 26.43, 27.51 (NHCH_3), 29.46, 30.02 (C(4)), 31.42 (CHCH_2), 47.34, 47.55 (C(5) or C(5a)), 55.47, 55.64 (CHCH$_2$), 68.64, 68.77, 68.91 (C(3)), 78.69, 79.88, 79.93, 80.09, 80.84, 81.01, 81.37 (C(1'), C(2'), C(3')), 94.60, 94.67 (C(1)), 102.77 (C(6)), 118.71, 118.94 (C(5'')), 128.49(C(2''', 6''') or C(3''', 5''')), 129.55 (C(2''', 6''') or C(3''', 5''')), 132.91 (C(4''')), 135.09 (C(1''')), 138.56, 138.76, 138.84, 138.98 (C(2''), C(4'')), 169.84, 169.90, 172.49, 172.71, 172.93, 173.09, 173.67, 174.12, 174.25, 174.33 (CO) ppm. The peak for the C(5) or C(5a) carbon is presumed to reside beneath the signal for the solvent. MS (+FAB) 575 [M + 1]$^+$.

Compound **22b**: yield, 5 mg (11%) as a semisolid; R_f 0.30 (20% methanol–chloroform); FTIR (KBr) 1686 cm^{-1}; ^{1}H NMR (CD_3OD) δ 1.32, 1.33 (s, 3 H, C(2')CH_3), 1.66–2.10 (m, 2 H, C(4)HH'), 2.49–2.67 (m, 1 H, C(5)H), 2.72 (s, 3 H, NHCH_3), 2.95–3.12 (m, 2 H, CHCH_2), 3.64–4.21 (m, 6 H, C(3)HH', C(3')HH', C(5a)HH', C(1')H), 4.27–4.39 (m, 1 H, C(5a)HH'), 4.71–4.87 (m, 1 H, CHCH$_2$), 6.96, 6.97 (s, 1 H, C(5'')H), 7.44–7.52 (m, 3 H, C(3''', 4''', 5''')H), 7.58 (br s, 1 H, C(2'')H), 7.64–7.82 (m, 2 H, C(2''', 6''')H). The ^{1}H NMR assignments were confirmed by the corresponding COSY experiment. ^{13}C NMR (CD_3OD) 23.19, 23.25 (C(2')CH_3), 26.39, 26.55 (NHCH_3), 30.08 (C(4)), 31.50 (CHCH_2), 47.45, 47.75 (C(5) or C(5a)), 55.60, 55.82 (CHCH$_2$), 68.72, 68.92 (C(3)), 77.04, 78.05, 78.14, 80.61 (C(1'), C(2'), C(3')), 89.70, 89.78 (C(1)), 102.94 (C(6)), 118.77, 118.83 (C(5'')), 128.46 (C(2''', 6''') or C(3''', 5''')), 129.54 (C(2''', 6''') or C(3''', 5''')), 132.84 (C(4''')), 135.18 (C(1''')), 138.58, 138.69, 138.78 (C(2''), C(4'')), 169.87, 172.82, 173.86, 174.27 (CO) ppm. The peak for the C(5) or C(5a) carbon is presumed to reside beneath the signal for the solvent. MS (+FAB) 575 [M + 1]$^+$.

Treatment of Bicyclomycin with Morpholine (12). A 1% aqueous solution (1.25 mL, pH 10.2) of **12** (12.36 mg, 0.142 mmol) and **1** (25 mg, 0.083 mmol) was stirred at room temperature (20 h). The solvent was removed in vacuo and the residue was purified by PTLC (silica gel) with 10% methanol–chloroform as the eluent to yield **26** (7.2 mg, 23%) as a semisolid: R_f 0.50 (10% methanol–chloroform); FTIR (KBr) 1740, 1699 cm^{-1}; ^{1}H NMR (CD_3OD) δ 1.16 (s, 3 H, C(2')CH_3), 1.41 (dd, J = 2.8, 13.6 Hz, 1 H, C(4)HH'), 2.07 (app dt, J = 6.4, 13.6 Hz, 1 H, C(4)HH'), 2.50–2.80 (br s, 4 H, N(CH_2)$_2$), 2.73 (d, J = 14.1 Hz, 1 H, C(5a)HH'), 3.30 (d, J = 14.1 Hz, 1 H, C(5a)HH'), 3.50–3.75 (m, 5 H, O(CH_2)$_2$, C(3)HH'), 3.62 (d, J= 12.0 Hz, 1 H, C(3')HH'), 3.83 (s, 1 H, C(1')H), 3.94 (dd, J = 6.4, 13.6 Hz, 1 H, C(3)HH'), 4.15 (d, J = 12.0 Hz, 1 H, C(3')HH'). The ^{1}H NMR assignments were confirmed by the corresponding COSY experiment. ^{13}C NMR (CD_3OD) 21.08 (C(2')CH_3), 32.77 (C(4)), 54.61 (C(5a)), 56.07 (N(CH_2CH$_2$)$_2$O), 58.20 (C(5)), 60.42 (C(3)), 68.08 (N(CH$_2$$CH_2$)$_2$O), 70.19 (C(1') or C(2') or C(3')), 71.87 (C(2') or C(1') or C(3')), 72.63 (C(3') or C(2') or C(1')), 84.99 (C(1)), 96.35 (C(9)), 160.15 (C(7)), 196.33 (C(6)) ppm; M_r (+CI) 373.158 78 (calcd for $C_{16}H_{25}N_2O_8$, [M + 1]$^+$, 373.161 09).

Treatment of Bicyclomycin with Ethyl Piperazinecarboxylate (13). The preceding reaction was repeated with a 1% aqueous solution (2 mL, pH 10.6) of **13** (35 mg, 0.136 mmol) and **1** (25 mg, 0.083 mmol). The solvent was removed in vacuo and the crude material was purified by PTLC with 7% methanol–chloroform as the eluent (two developments) to yield 6.0 mg (16%) of **27** as a semisolid: R_f 0.50 (10% methanol–chloroform); FTIR (KBr) 3495, 1734, 1701 cm^{-1}; ^{1}H NMR (CD_3OD) δ 1.20 (s, 3 H, C(2')CH_3), 1.28 (t, J = 7.1 Hz, 3 H, CH$_2$$CH_3$), 1.45 (dd, J = 2.6, 13.5 Hz, 1 H, C(4)HH'), 2.11 (app dt, J = 6.3, 13.5 Hz, 1 H, C(4)HH'), 2.50–2.90 (br s, 4 H, N(CH_2CH$_2$)$_2$NCO$_2$CH$_2$CH$_3$), 2.80 (d, J = 14.3 Hz, 1 H, C(5a)HH'), 3.30–3.40 (m, 5 H, C(5a)HH', N(CH$_2$$CH_2$)$_2NCO_2CH_2CH_3$), 3.68 (d, J = 12.0 Hz, 1 H, C(3')HH'), 3.72 (app dt, J= 2.6, 13.5 Hz, 1 H, C(3)HH'), 3.87 (s, 1 H, C(1')H), 3.98 (dd, J = 6.3, 13.5 Hz, 1 H, C(3)HH'), 4.13 (q, J = 7.1 Hz, 2 H, CH_2CH$_3$), 4.21 (d, J = 12.0 Hz, 1 H, C(3')HH'). The ^{1}H NMR assignments were confirmed by the corresponding COSY experiment. ^{13}C NMR (CD_3OD) 14.87 (CH$_2$$CH_3$), 21.08 (C(2')$CH_3$), 32.71 (C(4)), 44.83 (N(CH$_2$$CH_2$)$_2NCO_2CH_2CH_3$), 54.68 (C(5a) or N($CH_2CH_2$)$_2NCO_2CH_2CH_3$), 55.27 (N($CH_2CH_2$)$_2NCO_2CH_2CH_3$ or C(5a)), 58.17 (C(5)), 59.85 (C(3)), 62.83 (NCO$_2$$CH_2CH_3$), 70.22 (C(1') or C(2') or C(3')), 71.83 (C(2') or C(1') or C(3')), 72.66 (C(3') or C(2') or C(1')), 84.97 (C(1)), 96.31 (C(9)), 156.95 (NCO_2CH$_2$CH$_3$), 160.16 (C(7)), 196.20 (C(6)) ppm; M_r (EI) 443.189 90 (calcd for $C_{19}H_{29}N_3O_9$, 443.190 38).

Treatment of Bicyclomycin with N-Methylpiperazine (14). With use of the previous procedure described for the reaction of **1** with **12**, **1** (25 mg, 0.083 mmol) was added to a 1% aqueous solution (2 mL, pH 10.8) of **14** (20 mg, 0.2 mmol) at room temperature. The solvent was removed in vacuo and the residue purified by PTLC with 10% methanol–chloroform as the eluent to yield **28** (3.5 mg, 11%) as a semisolid: R_f 0.50 (10% methanol–chloroform); FTIR (KBr) 1740, 1697 cm^{-1}; ^{1}H NMR (CD_3OD) δ 1.15 (s, 3 H, C(2')CH_3), 1.40 (dd, J = 2.5, 13.6 Hz, 1 H, C(4)HH'), 2.06 (app dt, J = 6.3, 13.6 Hz, 1 H, C(4)HH'), 2.22 (s, 3 H, NCH$_3$), 2.40–3.00 (br s, 8 H, N($CH_2$$CH_2$)$_2NCH_3$), 2.75 (d, J = 14.2 Hz, 1 H, C(5a)HH'), 3.22–3.38 (m, C(5a)HH', CD_3OD), 3.62 (d, J = 11.9 Hz, 1 H, C(3')HH'), 3.66 (app dt, J = 2.5, 13.6 Hz, 1 H, C(3)HH'), 3.82 (s, 1 H, C(1')H), 3.93 (dd, J = 6.3, 13.6 Hz, 1 H, C(3)HH'), 4.14 (d, J = 11.9 Hz, 1 H, C(3')HH'). The ^{1}H NMR assignments were confirmed by the corresponding COSY experiment. ^{13}C NMR (CD_3OD) 21.07 (C(2')CH_3), 32.76 (C(4)), 45.84 (NCH$_3$), 54.53 (N(CH_2CH$_2$)NCH$_3$ or C(5a) or N(CH$_2$$CH_2$)$_2NCH_3$), 55.12 (N($CH_2CH_2$)$_2NCH_3$ or C(5a) or N(CH$_2$$CH_2$)$_2NCH_3$), 56.20 (N($CH_2CH_2$)$_2NCH_3$ or C(5a) or N(CH$_2$$CH_2$)NCH$_3$), 58.20 (C(5)), 59.59 (C(3)), 70.16 (C(1') or C(2') or C(3')), 71.84 (C(2') or C(3') or C(1')), 72.65 (C(3') or C(1') or C(2')), 84.97 (C(1)), 96.31 (C(9)), 160.17 (C(7)), 196.27 (C(6)) ppm; MS (+CI) 386 [M + 1]$^+$.

Treatment of Bicyclomycin with Morpholine (12) at pH 12.5. To a solution of **1** (50 mg, 0.165 mmol) in water (4 mL, pH 12.5) was added a 1% aqueous morpholine (**12**) solution (1.7 mL, 0.195 mmol) and the pH of the solution was raised to 12.5. The mixture was stirred at room temperature (3 h), and then the solvent was removed in vacuo. The crude product was purified by preparative TLC with 20% methanol–chloroform (two developments) as the eluent to give compound **23**: yield, 5 mg (8%) as a semisolid; R_f 0.40 (20% methanol–chloroform); FTIR (KBr) 1684 cm^{-1}; ^{1}H NMR (CD_3OD) δ 1.33 (s, 3 H, CH$_3$), 1.76–2.04 (m, 1 H, C(4)HH'), 2.10–2.36 (m, 1 H, C(4)HH'), 2.36–2.97 (m, 7 H, C(5)H, C(5a)HH', N(CH_2CH$_2$)$_2$O), 3.47–4.15 (m, 9 H, C(3)HH', C(3')HH', C(1')H, N(CH$_2$$CH_2$)$_2$O). The ^{1}H NMR assignments were confirmed by the corresponding COSY experiment. ^{13}C NMR (CD_3OD) 21.09, 22.49, 23.29 (CH$_3$), 30.47, 30.76, 31.10 (C(4)), 43.61, 44.04 (C(5)), 54.97 (N(CH_2CH$_2$)$_2$O), 59.37 (C(5a)), 67.78 (N(CH$_2$$CH_2$)$_2$O), 68.55 (C(3)), 76.99, 77.42, 78.03, 78.68, 79.58, 80.23, 80.59, 81.30 (C(1'), C(2'), C(3')), 89.64 (C(1)), 101.51, 104.33 (C(6)), 175 (CO) ppm. The remaining carbonyl signal was not detected. MS (+FAB) 390 [M + 1]$^+$.

Treatment of 2',3'-Bicyclomycin Acetonide (31) with Morpholine (12). Compound **31** (12 mg, 0.035 mmol) was stirred in a 1% morpholine (**12**) (20 mg, 0.23 mmol) tetrahydrofuran–water (1:1) solution (2 mL, "pH" 10.6) at room temperature (15 h). The solvent was removed in vacuo and the crude product was purified by PTLC with 10% methanol–chloroform as the eluent to obtain 8.2 mg of **32** (55%): mp 135–140 °C; R_f 0.70 (15% methanol–chloroform); FTIR (KBr) 1684 cm^{-1}; ^{1}H NMR (CD_3OD) δ 1.35 (s, 3 H, C(2')CH_3), 1.45 (s, 6 H, C(CH_3)$_2$), 1.51–1.66 (m, 1 H, C(4)HH'), 1.80–1.95 (m, 1 H, C(4)HH'), 2.24–2.42 (m, 2 H, C(5a)HH', C(5)H), 2.42–2.58 (br s, 2 H, N(CHH'CH$_2$)$_2$O), 2.63–2.84 (m, 3 H, C(5a)HH', N(CHH'CH$_2$)$_2$O), 3.62–3.78 (m, 5 H, C(3')HH', N(CH$_2$$CH_2$)$_2$O), 3.83–3.94 (m, 1 H, C(3)HH'), 4.02–4.15 (m, 1 H, C(3)HH'), 4.08 (s, 1 H, C(1')H), 4.45 (d, J = 8.4 Hz, 1 H, C(3')HH'); ^{13}C NMR (CD_3OD) 24.89 (C(2')CH_3), 26.81 (CCH_3), 28.40 (CCH_3), 31.88 (C(4)), 45.23 (C(5)), 54.24 (N(CH_2CH$_2$)$_2$O), 61.48 (C(5a)), 66.03 (C(3)), 67.80 (N(CH$_2$$CH_2$)$_2$O), 73.17 (C(1') or C(3')), 73.27 (C(3') or C(1')), 85.48 (C(6)), 86.37 (C(2')), 89.48 (C(1)), 111.73 (C(CH$_3$)$_2$), 165.78 (C(7)), 171.69 (C(9)) ppm; MS (+FAB) 430 [M + 1]$^+$.

Conversion of Acetonide 32 to 30. Acetonide **32** (6 mg, 0.014 mmol) was dissolved in 50% aqueous acetic acid (1 mL) and the solution was heated at 60 °C (90 min). The solvent was removed in vacuo and the crude mixture was purified by PTLC with 15% methanol–chloroform as the eluent to yield 3.5 mg (64%) of **30** as a semisolid: R_f 0.40 (15% methanol–chloroform); FTIR (KBr) 1686 cm^{-1}; ^{1}H NMR (CD_3OD) δ 1.32 (s, 3 H, C(2')CH_3), 1.54–1.72 (m, 1 H, C(4)HH'), 1.78–1.95 (m, 1 H, C(4)HH'), 2.23–2.38 (m, 2 H, C(5)H, C(5a)HH'), 2.38–2.58 (br s, 2 H, N(CHH'CH$_2$)$_2$O), 2.58–2.84 (m, 3 H, C(5a)HH', N(CHH'CH$_2$O)$_2$), 3.56 (d, J = 11.4 Hz, 1 H, C(3')HH'), 3.62–3.80 (m, 5 H, C(3')HH', N(CH$_2$$CH_2$)$_2$O), 3.80–3.91 (m, 1 H, C(3)HH'), 4.02 (s, 1 H, C(1')H), 3.98–4.13 (m, 1 H, C(3)HH'). The ^{1}H NMR assignments were confirmed by the corresponding COSY experiments. ^{13}C NMR (CD_3OD) 24.19 (C(2')CH_3), 31.89 (C(4)), 45.42 (C(5)), 54.31 (N(CH_2CH$_2$)$_2$O), 61.37 (C(5a)), 65.41 (C(3)), 67.88 (N(CH$_2$$CH_2$)$_2$O), 68.50 (C(3')), 72.38 (C(1')), 78.09 (C(2')), 85.46 (C(6)), 90.14 (C(1)), 168.83 (C(7)), 172.10 (C(9)) ppm; MS (+FAB) 390 [M + 1]$^+$; M_r (+FAB) 390.1876 (three determinations) (calcd for $C_{16}H_{28}N_3O_8$, [M + 1]$^+$ 390.1876).

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Organic Reactions Catalyzed by Copper-Loaded Polymers. Reactivity vs Polymer Structure

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Abstract: Four types of polymers were constructed: P-M, P-M-H_1, P-H_2-M, and P-H_2-M-H_1, where P = polystyrene, M = metal (Cu^{2+}), and H = hydrocarbon chain (H_1 = 14 carbons and H_2 = six carbons). Thus, P-M is devoid of an aliphatic hydrocarbon, whereas P-M-H_1 has a metal interposed between the polymer and a long hydrocarbon chain. With both, however, the metal resides near the polymer backbone. In contrast, P-H_2-M and P-H_2-M-H_1 have a six-carbon spacer separating the metal and polymer. The latter also possesses a 14-carbon outer chain, so that the copper is situated between two hydrocarbon regions. Of the four polymeric types, P-H_1-M was found to be the most active in catalyzing the hydrolysis of nerve-agent simulants. Thus, 4-nitrophenyl diphenyl phosphate is rapidly hydrolyzed ($t_{1/2}$ = 2.7 min) with 1.0 mM polymer-bound Cu^{2+} at pH = 8.0 (25.0 °C). The reactions display saturation kinetics and operate via a turnover mechanism. In addition to the rate studies, six synthetically useful copper-promoted reactions (including a Diels-Alder cyclization, an epoxide opening, and an aryl iodide hydrolysis) were examined. Five of these manifest higher yields and shorter reaction times with the metallopolymer as opposed to an equivalent amount of conventional copper salt. Easy reaction workup is another virtue of the polymer-catalyzed processes.

Chemical-warfare agents, such as nerve gas and mustard, owe their potency to a high reactivity toward nucleophiles in body tissues. Consequently, any strategy for chemical defense against these loathsome materials requires the development of compounds for which the agents have an even greater affinity. Notable progress along these lines has appeared recently from the laboratories of Moss and ourselves.[1,2] Moss found that iodosobenzoates destroy phosphorus(V) compounds related to nerve agents.[1] We, on the other hand, exploited "metallomicelles" to inactivate the deadly neurotoxins.[2] Turnover mechanisms with 10^5–10^6 rate enhancements were achieved. The present article is dedicated to rendering nerve agents and their simulants impotent via hydrolyses catalyzed by copper-loaded polymers. Rate studies revealed how the surfaces of the new "metallopolymer" systems interact with small molecules. In addition, the polymers were examined for their ability to promote six synthetically useful organic reactions.[3]

Four types of polymers were constructed (Figure 1): P-M, P-M-H_1, P-H_2-M, and P-H_2-M-H_1, where P = polystyrene, M = metal (Cu^{2+}), and H = hydrocarbon chain (H_1 = 14 carbons, and H_2 = six carbons). Thus, P-M is devoid of hydrocarbon, whereas P-M-H_1 has a metal interposed between the polymer surface and a long hydrocarbon chain. With both polymers, however, the metal resides near the polymer backbone. In contrast, P-H_2-M and P-H_2-M-H_1 have a six-carbon spacer separating the metal and polymer. The latter also possesses a 14-carbon outer chain, so that the metal is situated between two hydrocarbon regions.

The polymers were implanted with copper owing to the known ability of this metal to catalyze the hydrolysis of phosphorus(V) compounds.[4] There was, of course, also good reason for incorporating hydrocarbon tails into the polymers. If one is to achieve a high level of catalysis, substrates must bind to the polymer prior to the actual chemistry. Contiguous hydrocarbon tails can provide a means for attracting hydrophobic substrates to the polymer surfaces similar to the action of surfactant chains in micelles. This is not speculation. Many years ago, Cordes et al.[5] showed that small organic molecules bind hydrophobically to poly-4-vinylpyridine quaternized with dodecyl bromide (a "polysoap"). No one has, however, yet investigated the catalytic consequences of embedding Cu^{2+} within nonpolar regions of polymer surfaces.

The catalytic activity of the polymer systems was tested with two substrates: 4-nitrophenyl isopropylphenylphosphinate (I) and 4-nitrophenyl diphenyl phosphate (II). These compounds were selected because: (a) They are easily handled "simulants" of the more relevant but also more toxic nerve agents such as GD. (b) Considerable work, including our metallomicelle experiments,[2] has been carried out on the substrates, so that there exists a large body of data with which to judge the efficacy of the polymeric catalysts.[6] (c) The hydrolysis of the substrates, in contrast to GD, can be monitored spectrophotometrically. (d) Since the

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