Hydroxamate Approach to the Synthesis of β-Lactam Antibiotics†

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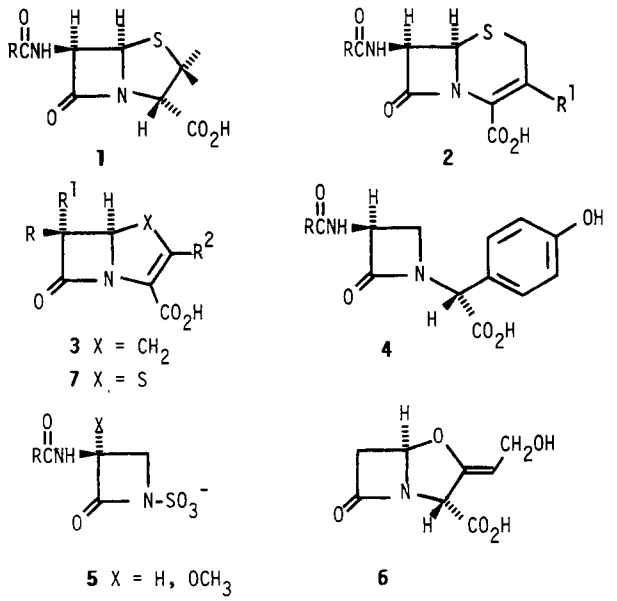
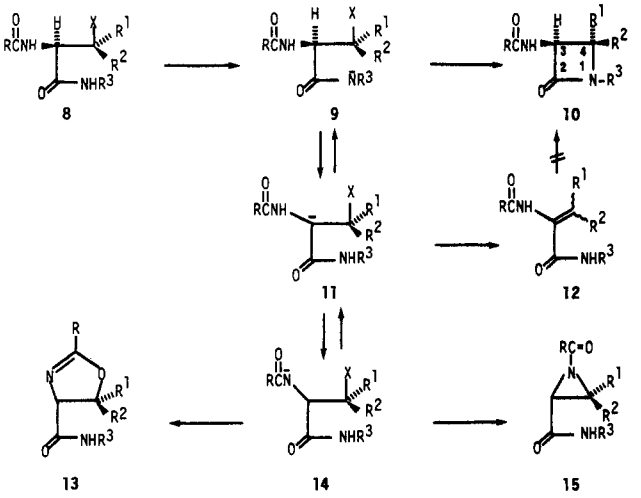
Since the discovery and clinical introduction of penicillin (1) β-lactam antibiotics have played a vital role in maintaining the welfare of the human race.[1] However, intrinsic resistance to these "wonder drugs" among certain pathogenic bacteria, combined with the development of resistance by formerly sensitive strains, primarily by the evolution of β-lactamase enzymes,[2] has encouraged a continued search for even more effective antimicrobial agents. While relatively few of the thousands of chemically modified forms of penicillin have proven to be clinically useful antibiotics, nature has frequently provided clues to help us maintain our antimicrobial defenses.



Another class of natural products, the cephalosporins **2**, are now the most widely used β-lactam antibiotics.[3] Even more recent efforts have resulted in the discovery of still other natural β-lactams including the potent carbapenems **3**,[4] the monocyclic nocardicins **4**[5] and monobactams **5**,[6] and clavulanic acid **6**,[7] a natural β-lactamase inhibitor. The structural diversity of all of these β-lactams has required reconsideration of previously established structure–activity relationships.[8] Consequently, the search for the ultimate antibiotic or, more realistically, for other forms of antibiotics to augment our chemotherapeutic arsenal continues. This paper focuses on our attempts to facilitate this effort.

Marvin Miller was born Jan 29, 1949, in Dickinson, ND. He received his B.S. degree at North Dakota State University and his Ph.D. at Cornell under the direction of Professor G. Marc Loudon. After time as an NIH postdoctoral fellow in Professor Henry Rapoport's group at Berkeley, Dr. Miller joined the faculty at Notre Dame in 1977. His research program involves the synthesis and study of biologically important heteroatom-containing systems (amino acids, peptides, antibiotics, siderophores, and others).

Scheme I



Because of the ease with which derivatives of cephalosporins and penicillins can be chemically rendered antibiotically ineffective, historically little effort was directed toward modification of their core bicyclic structures. However, Woodward's syntheses of the penems **7**[9] provided a clear indication that increasingly powerful organic synthetic methods could lead to the rational design of effective antibiotics. During the subsequent decade, massive industrial and academic effort has been rededicated to the design and synthesis

† Dedicated to my father, Joseph F. Miller, on the occasion of his 70th birthday and to the memory of my father-in-law, Florian B. Krause.

(1) (a) Kammer, R. B. In "Chemistry and Biology of β-Lactam Antibiotics"; Morin, R. B., Gorman, M., Eds.; Academic Press: New York, 1982; Vol 3, Chapter 5. (b) Selwyn, S., Ed. "The β-Lactam Antibiotics: Penicillins and Cephalosporins in Perspective"; Hodder and Stoughton: London, 1980. (c) Sheehan, J. C., Ed. "The Enchanted Ring"; MIT Press: Cambridge, MA, 1982.

(2) (a) Knowles, J. R. *Acc. Chem. Res.* **1985**, *18*, 97. (b) Cimarusti, C. M. *J. Med. Chem.* **1984**, *27*, 247. (c) "Beta-Lactamases"; Hamilton-Miller, J. M. T., Smith, J. T., Eds.; Academic Press: New York, 1979.

(3) (a) "Cephalosporins and Penicillins: Chemistry and Biology"; Flynn, E. H., Ed.; Academic Press: New York, 1972. (b) "β-Lactam Antibiotics"; Salton, M. R. J., Shockman, G. D., Eds.; Acadmemic Press: New York, 1981; Chapter 5.

(4) Johnson, D. B. R.; Schmitt, S. M.; Bouffard, F. A.; Christensen, B. G. *J. Am. Chem. Soc.* **1978**, *100*, 313.

(5) Hashimota, M.; Komori, T.; Kamiya, T. *J. Am. Chem. Soc.* **1976**, *98*, 3023.

(6) (a) Smada, A.; Kitano, K.; Kintaka, K.; Muroe, M.; Asai, M. *Nature* **1981**, *289*, 590. (b) Sykes, R. B.; Cimarusti, C. M.; Bonner, D. P.; Busk, K.; Floyd, D. M.; Georgopapodakou, N. H.; Koster, W. H.; Liu, W. C.; Parker, W. L.; Principe, P. A.; Rathnum, M. L.; Slusarchyk, W. A.; Trejo, W. H.; Wells, J. S. *Nature* **1981**, *291*, 489.

(7) Howarth, T. T.; Brown, A. G.; King, T. J. *J. Chem. Soc., Chem. Commun.* **1976**, 266.

(8) Boyd, D. G. In "Chemistry and Biology of β-Lactam Antibiotics"; Morin, R. B., Gorman M., Eds.; Academic Press: New York, 1982; Vol. 1, Chapter 5.

(9) Woodward, R. B. In "Recent Advances in the Chemistry of β-Lactam Antibiotics"; Elks, J., Ed.; The Chemical Society: London, 1977; pp 167–180.

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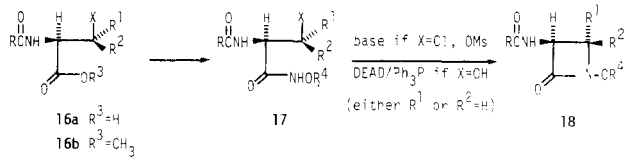
Scheme II



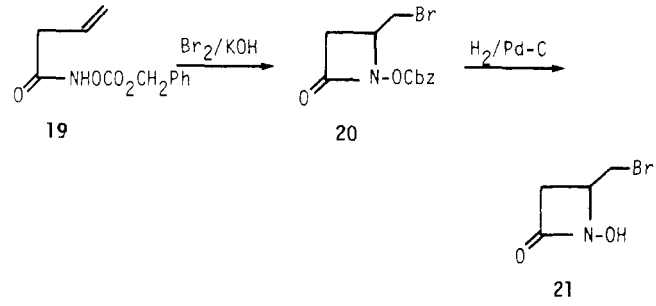
of β-lactam antibiotics.[10] However, of the numerous methods developed for β-lactam synthesis, no single method was compatible with the variety of peripheral substitution and chirality needed for elaboration to any or all β-lactams of current interest.

We thought that synthesis of the 2-azetidinone (β-lactam) ring system by N–C_4 bond closure would be especially attractive because of its biosynthetic analogy[11] and potential ability to use chiral amino acid derivatives or other modified amides with β-leaving groups as starting materials. Although several related N–C_4 bond closures had been devised,[12] the required protection of the peripheral amino acid functionality and chirality, the need for a multistep incorporation of a β-leaving group, or the use of strong base in the cyclization step decreased their utility. Ideally a biomimetic β-lactam synthesis should proceed by direct cyclization without the need for elaborate prior manipulations. While conceptually such a process is represented in Scheme I (**8** → **10**), experimentally it was not feasible. The similarity of the p*K* values of the ultimate C_3–H and the peripheral N–H bonds lead to detrimental proton transfers and subsequent reactions with little desired cyclization. The problem, therefore, became one of differentiating the p*K* values of the three potentially ionizable positions to allow selective ionization to **9**.

N–C_4 Cyclization and Applications

We demonstrated that the required selective ionization could be accomplished by hydroxamic acid based heteroatom activation (Scheme II).[13] Thus, β-halo or β-hydroxy carboxylic acids (**16a**) were converted to the corresponding hydroxamates **17** (p*K* 6–9) by active ester condensation with O-substituted hydroxylamines (H_2NOR^4). Base-initiated cyclization of the β-halo hydroxamates **17** (X = Cl) proceeded cleanly to give the desired *N*-hydroxy β-lactam derivatives **18**, even from chiral β-chloroalanine precursors. Since chiral β-hydroxy acids are much more readily available than β-halo acids, we sought an efficient direct method for the cyclization of the corresponding β-hydroxy hydroxamates. The Mitsunobu reaction (diethyl azodicarboxylate (DEAD)/Ph_3P)[14] served admirably. Reaction of a variety of β-hydroxy hydroxamates **17** (X = OH) with DEAD/Ph_3P provided the desired β-lactams in high yield.[13] *The retention of configuration at C_3 and clean inversion at C_4 during this cyclization implied that essentially any chiral β-lactam could be made by simply choosing the appropriate chiral starting β-hydroxy acid.* Subsequent considerable effort in our laboratories and others has verified this versatility.

Scheme III



The major problems with industrial-scale applications of this N–C_4 cylcization process were the use of expensive reagents, the required chromatographic separation of the products of the Mitsunobu reagents (reduced azodicarboxylates and $Ph_3P{=}O$), and the competitive formation of oxazolines such as **13** (R^3 = OR) when simple acylamino side chains were present. The use of carbamate protecting groups (RCO = Boc, Cbz of **16** → **18**; Scheme II) avoided the oxazoline problem and is usually preferred since the resulting protected 3-amino-substituted β-lactams **18** can later be deprotected and reacylated with any of a variety of desired side chains. Later, taking a lead from the Squibb group,[15] we found that β-mesylates of benzyl hydroxamate of α-acylserine (**17**: X = OMs; R^1 = R^2 = H; R^4 = CH_2Ph) can be directly cyclized under careful conditions (KO-*t*-Bu/DMF/−23 °C) without β-elimination or formation of oxazolines or aziridines.[16] In many cases, we found that the use of expensive O-substituted hydroxylamines and the azodicarboxylates could be avoided by direct hydroxaminolysis of protected amino acid esters **16b** with hydroxylamine itself, followed by in situ acylation and finally substitution of Ph_3P/CCl_4/Et_3N for Ph_3P/DEAD during the cyclization step.[17]

The NH acidity of β,γ-unsaturated *O*-acyl hydroxamates **19** also facilitates direct oxidative cyclization to the corresponding substituted 4-(halomethyl)-*N*-hydroxy-β-lactams 20 (Scheme III).[18]

N–O reduction of the hydroxamate-derived β-lactams was anticipated to provide access to a variety of chiral 3- and 4-substituted N-unsubstituted β-lactams suitable for elaboration to a number of antibiotics that are not available from penicillin degradation products. Finding N–O reduction conditions that were compatible with retention of the β-lactam ring or peripheral functionality and chirality required considerable effort. Eventually an efficient two-step process was developed that first involved deprotection of the hydroxyl group (**18** → **22**) followed by reduction of the free-*N*-hydroxy β-lactam **22** with buffered $TiCl_3$.[19] The overall process

(10) For a nice overview see: Dürckheimer, W.; Blumbach, J.; Latrell, R.; Scheunemann, K. H. *Angew. Chem., Int. Ed. Engl.* **1985**, *14*, 180.

(11) (a) Baldwin, J. E. In "Recent Advances in the Chemistry of β-Lactam Antibiotics"; Brown, A. G., Roberts, S. M., Eds.; Royal Society of Chemistry: London, 1985; Chapter 5. (b) Townsend, C. A.; Brown, A. M. *J. Am. Chem. Soc.* **1983**, *105*, 913.

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(13) (a) Mattingly, P. G.; Kerwin, J. F., Jr.; Miller, M. J. *J. Am. Chem. Soc.* **1979**, *101*, 3983–3985. (b) *Chem. Eng. News* **1979**, Oct 1, pp 25–26. (c) Miller, M. J.; Mattingly, P. G.; Morrison, M. A.; Kerwin, F. G., Jr. *J. Am. Chem. Soc.* **1980**, *102*, 7026.

(14) Mitsunobu, O. *Synthesis* **1981**, 1.

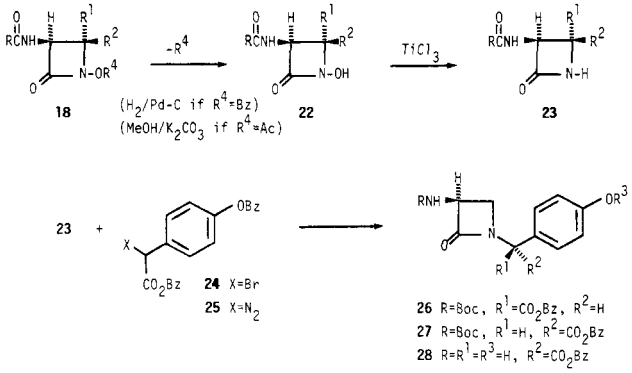
(15) Floyd, D. M.; Fritz, A. W.; Pluscec, J.; Weaver, E. R.; Cimarusti, C. M. *J. Org. Chem.* **1982**, *47*, 5160.

(16) Krook, M. A.; Miller, M. J. *J. Org. Chem.* **1985**, *50*, 1126.

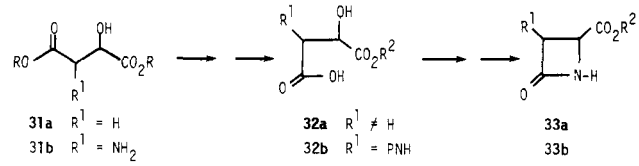
(17) Miller, M. J.; Biswas, A.; Krook, M. A. *Tetrahedron* **1983**, *39*, 2571.

(18) (a) Rajendra, G.; Miller, M. J. *Tetrahedron Lett.* **1985**, 5385. (b) For similar oxidative cyclizations of β,γ-unsaturated *N*-tosylamides, see: Biloski, A. J.; Wood, R. D.; Ganem, B. *J. Am. Chem. Soc.* **1982**, *104*, 3233.

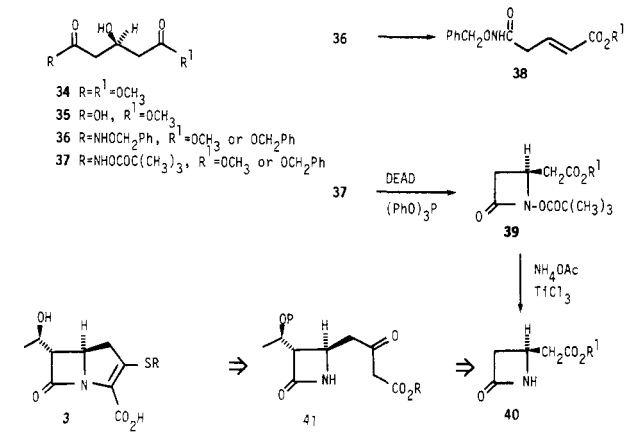
Scheme IV



Scheme V



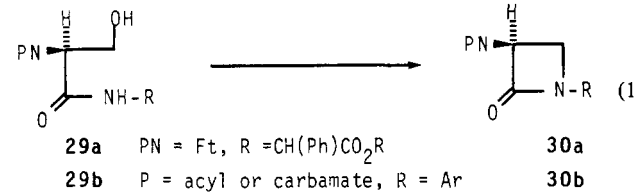
Scheme VI



of conversion of β-hydroxy acids to hydroxamates (**16** → **17**), cyclization (**17** → **18**), and reduction (**18** → **22** → **23**) has been carried out on dozens of different structures and routinely proceeds in excellent overall yields.

With this substitutionally versatile route to chiral N-unsubstituted β-lactams available, we were tempted to claim formal total syntheses of 3-ANA and C_4-substituted analogues. However, close inspection of the literature describing the details of the 3-ANA synthesis from penicillin indicated that the N-alkylation (**23** + **24** → **26**) actually proceeded in very low yield.[21] Alternatively, we found that treatment of the corresponding protected diazophenylacetate **25** with rhodium acetate in the presence of **23** provided a mixture of diasteromers **26** and **27** in 67% yield.[22] Fortunately, the wrong diastereomer **26** was chromatographically separable and isomerized to a new mixture of **26** and **27** with catalytic base, eventually allowing near complete conversion to the protected 3-ANA, **27**. The overall yield of 45% for the conversion of βoc-L-serine (**16**: R = *t*-BuO; $R^1 = R^2 = R^3 = H$) to **27** provided an indication of the practicality of the hydroxamate approach to the synthesis of chiral β-lactams.

Not surprisingly, the novelty of the nocardicins attracted the synthetic interests of several groups.[23] The most straightforward route has been that developed by Townsend.[23d] Following his elegant studies of the biosynthesis of 3-ANA,[11b] he found that treatment of protected L-seryl-D-phenylglycine (**29a**) under modified Mitsunobu conditions provided the 3-ANA nucleus directly (eq 1). These results were quite interesting especially since Bose's studies on the cyclization of β-hydroxy arylamides **29b** indicated that competitive eliminations and other side reactions (Scheme I) often prevail.[24] Our related work also indicated that the efficiency of the direct cyclization of peptides and analogues is variable and depends heavily on the nature of constituents (R of **29**) of the terminal amide.[25]



(1)

29a PN = Ft, R = CH(Ph)CO_2R → **30a**

29b P = acyl or carbamate, R = Ar → **30b**

β-Hydroxy Acid Precursors to β-Lactams

Subsequent to completing our synthesis of 3-ANA, we have attempted to verify that, given the appropriate chiral β-hydroxy acid starting material, the hydroxamate approach is compatible with the synthesis of nearly any β-lactam. In many cases, the key step actually became finding a source of the β-hydroxy acid. Although many suitably substituted β-hydroxy acids (i.e., **16**) were derived from the natural "chiral pool", others were obtained from selected enzyme-mediated hydrolyses, chemical or microbial reductions of β-keto esters, allylic oxidations, and enantioselective aldol condensations. Several of these routes are briefly described.

Diastereoselective alkylation of the dianion of either D- or L-diethyl malates (**31a**: R = Et) followed by bis-saponification and monoesterification provided convenient sources of all the separate optical isomers of β-hydroxy ester **32a**. Subjection of **32a** to the usual hydroxaminolysis followed by cyclization and N–O reduction produced the corresponding 3-alkyl-4-(alkoxycarbonyl)-β-lactams **33a** with complete control of the two chiral centers.[26] Similarly, the availability of all four optical isomers of β-hydroxyaspartic acid (**31b**)

(19) Mattingly, P. G.; Miller, M. J. *J. Org. Chem.* **1980**, *45*, 410. A difficulty with this two-step reduction process was formation of emulsions from Ti^{4+} salts generated during the workup of the $TiCl_3$ reaction. This problem can usually be avoided by addition of chelators such as buffered tartrate before the workup. Alternatively, the Squibb group has reported the effective use of a catalytic amount of $TiCl_3$ with excess Zn to regenerate the Ti^{3+}.[20] The same group also described a direct reduction of the *N*-methoxy-2-azetidinones (**18**, $R^4 = CH_3$) with Na/NH_3.

(20) Cimarusti, C. M.; Denzel, T. H.; Fox, R.; Jacobs, G. A.; Kissick, T. P.; Koster, W. H.; Mineot, J. L.; Mueller, R. H. In "Recent Advances in the Chemistry of β-Lactam Antibiotics"; Brown, A. G., Roberts, S. M., Eds.; Royal Society of Chemistry: London, 1985; Chapter 20.

(21) Kamiya, T. *Spec. Publ.—Chem. Soc.* **1977**, *No. 28*, 29.

(22) Mattingly, P. G.; Miller, M. J. *J. Org. Chem.* **1981**, *46*, 1557. The utility of the $Rh_2(OAc)_4$-mediated N–C bond formation had been previously demonstrated intramolecularly during Merck's synthesis of thienamycin. See ref 28a for an overview.

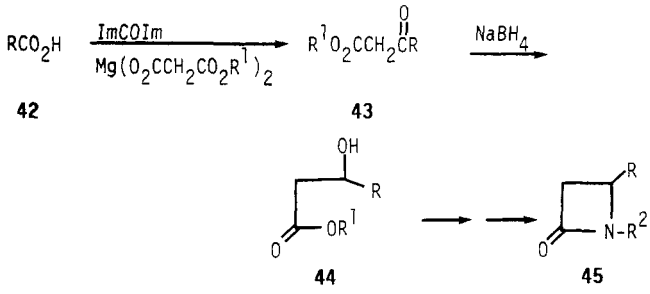
(23) (a) Koppel, G. A.; McShane, L.; Jose, F.; Cooper, R. D. G. *J. Am. Chem. Soc. 1978, 100*, 3933–3935. (b) Wasserman, H. H.; Hlasta, D. L. *Ibid.* **1978**, *100*, 6780. (c) Wasserman, H. H.; Hlasta, D. L.; Tremper, A. W.; Wu, J. S. *Tetrahedron Lett.* **1979**, *549*–552. (d) Townsend, C. A.; Brown, A. M.; Nguyen, L. T. *J. Am. Chem. Soc.* **1983**, *105*, 919.

(24) Bose, A. K.; Saha, D. P.; Manhas, M. S. *J. Org. Chem.* **1981**, *46*, 1229.

(25) Miller, M. J.; Mattingly, P. G. *Tetrahedron*, **1983**, *39*, 2563.

(26) Miller, M. J.; Bajwa, J. S.; Mattingly, P. G.; Peterson, K. *J. Org. Chem.* **1982**, *47*, 4928.

Scheme VII



makes available all the corresponding optically active 3-amino-4-(alkoxycarbonyl)-β-lactams.[27] 4-(Alkoxycarbonyl)-2-azetidinones **33a** and **33b** are useful chiral intermediates for the syntheses of carbapenems and nuclear analogues of penicillins and cephalosporins as well as C-4-substituted monobactams.[28]

Conceptually, chymotrypsin hydrolysis of dimethyl β-hydroxyglutarate was to provide the optically active β-hydroxy acid **35** (Scheme VI) and eventually the β-lactam **40** that is suitable for elaboration to the basic carbapenem framework. Instead, under the usual conditions (DEAD/Ph_3P) attempted cyclization of the corresponding *O*-benzyl hydroxamate **36** produced mainly β,γ-unsaturated hydroxamates **38**, indicating that acidic γ-protons are detrimental to the cyclization process. Subsequent use of the more acidic hydroxamate **37** and modified Mitsunobu conditions [$(PhO)_3P$/DEAD] slowed the cyclization reaction considerably but allowed formation of the β-lactam **39** in 70% yield.[29]

β-Keto ester synthesis by the Masamune/Brooks process,[30] followed by reduction, has also provided a versatile route to a number of required β-hydroxy acids and the corresponding β-lactams (Scheme VII).[30b] The nature of the eventual C_4 substituent of the β-lactam **45** depends simply on the choice of the starting carboxylic acid **42**. The variety of simple and functionalized substituents compatible with the Masamune/Brooks reaction suggests that this will be a substitutionally versatile route to β-lactams. Microbiolgical or enantioselective chemical reduction of the ketones **43** may also make this a chiral route.

We have also used stereo- and enantioselective aldol condensations to provide direct access to highly functionalized β-hydroxy acids (Scheme VIII).[31] Aldol condensation of protected glycine ester **46** with aldehyde **47** provided the racemic protected α-amino-β-hydroxy ester **48** with modest (4:1) threo to erythro selectivity. Fortunately, the major (*S**,*S**)-threo isomer provided the desired protected *cis*-3-amino 4-substituted 2-azetidinone **51** after the hydroxamate-based β-lactam synthesis. Tin-mediated aldol condensation of butyrylthiazolidinethione (**53a**) with aldehyde **54** provided the erythro product **55** cleanly.[31a] This aldol product contained the entire carbon framework needed for the synthesis of the racemic carbapenem, PS-5. Extension to the chiral synthesis of carbapenem intermediates encouraged the development of a highly enantioselective aldol process that utilized a readily available chiral auxillary that could also be easily removed by solvolysis or aminolysis and could be recycled. The *N*-acyl-*4*(R)-(methoxycarbonyl)-1,3-thiazolidine-2-thione (**53b**), derived from cysteine, satisfied all of these requirements.[31b] Formation of the boron enolate of **53** followed by condensation with aldehyde **54** provided the aldol product cleanly in high optical (>93% ee) and chemical yields. Direct hydroxaminolysis of **55b** gave the β-hydroxy hydroxamate **56b**. Cyclization under the usual conditions provided the optically active PS-5 precursor **57**.

Modifications and Rearrangements of *N*-Hydroxy-2-azetidinones

The ready availability of chiral 2-azetidinones by the hydroxamate route prompted further studies of their chemistry. Hydrolysis of *N*-hydroxy-2-azetidinones, followed by N–O reduction, provides a direct route to chiral α-substituted β-amino acids **60** (Scheme IX).[32] The *N*-hydroxy-2-azetidinones themselves are unusually acidic (p*K* 6–7) and often prone to rearrangement to ring-expanded *O*-acyl hydroxylamines **61**.[33] This latter property has been utilized for a novel synthesis of (±)-quisqualic acid (**62**).[34] Simple C_3- or C_4-alkyl-substituted *N*-hydroxy-2-azetidinones can be O-alkylated or acylated and even converted to the corresponding isolable mesylates and tosylates **58** (R^1 = Ms, Ts).[35] However, incorporation of activating groups at either C_3 or C_4 followed by modification of the *N*-hydroxy groups often initiates rearrangements. For example, reaction of 3-(phenylacetmido)-*N*-hydroxy-2-azetidinone (**58b**) with alkyl dichlorophosphates followed by aqueous workup provided the hydroxyimidazolidinone **64**, instead of the expected phosphorylated β-lactam **63**.[36] Participation of the phenylacetamido side chain in the rearrangement was implicated by the fact that when the C_3-amino group was fully protected (**58c**) the phosphorylated β-lactam **65** was obtained nearly quantitatively.

Acidification of the C_4 proton of *N*-hydroxy β-lactams also promotes rearrangement even when the hydoxyl group is converted to a modest leaving group (Scheme X).[37]

Treatment of *N*-hydroxy-2-azetidinones with halomalonates **70** also did not provide the simple alkylated products **71**. Instead, the acidity of **71** intiated a facile rearrangement to the corresponding carbinolamine **74** (Scheme XI). When the starting β-lactam contained

(27) Mattingly, P. G.; Miller, M. J.; Cooper, R. D. G.; Daugherty, B. W. *J. Org. Chem.* **1983**, *48*, 3556.

(28) (a) Salzman, T. N.; Ratcliffe, R. W.; Christensen, B. G.; Bouffard, F. A. *J. Am. Chem. Soc.* **1980**, *102*, 6163. (b) Huffman, W. R.; Holden, K. G.; Buckley, T. F.; Gleason, J. G.; Wu, L. *J. Am. Chem. Soc.* **1977**, *99*, 2352. (c) Kishimoto, S.; Sendai, M.; Hashiguchi, S.; Tomimoto, M.; Satoh, Y.; Matsuo, T.; Kando, M.; Ochiai, M. *J. Antibiot.* **1983**, *36*, 1421.

(29) Morrison, M. A.; Miller, M. J. *J. Org. Chem.* **1983**, *48*, 4421. The utility of **40** for the synthesis of carbapenems has been demonstrated: Shinkai, T.; Liu, T.; Reamer, R. A.; Sletziner, M. *Tetrahedron Lett.* **1982**, *23*, 4899.

(30) (a) Brooks, D. W.; Lu, L. D. L.; Masamune, S. *Angew. Chem., Int. Ed. Engl.* **1979**, *18*, 72. (b) Biswas, A.; Lee, B. H.; Miller, M. J., unpublished results.

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(33) Hirose, T.; Chiba, K.; Mishio, S.; Nakano, J.; Uno, H. *Heterocycles* **1982**, *19*, 1019.

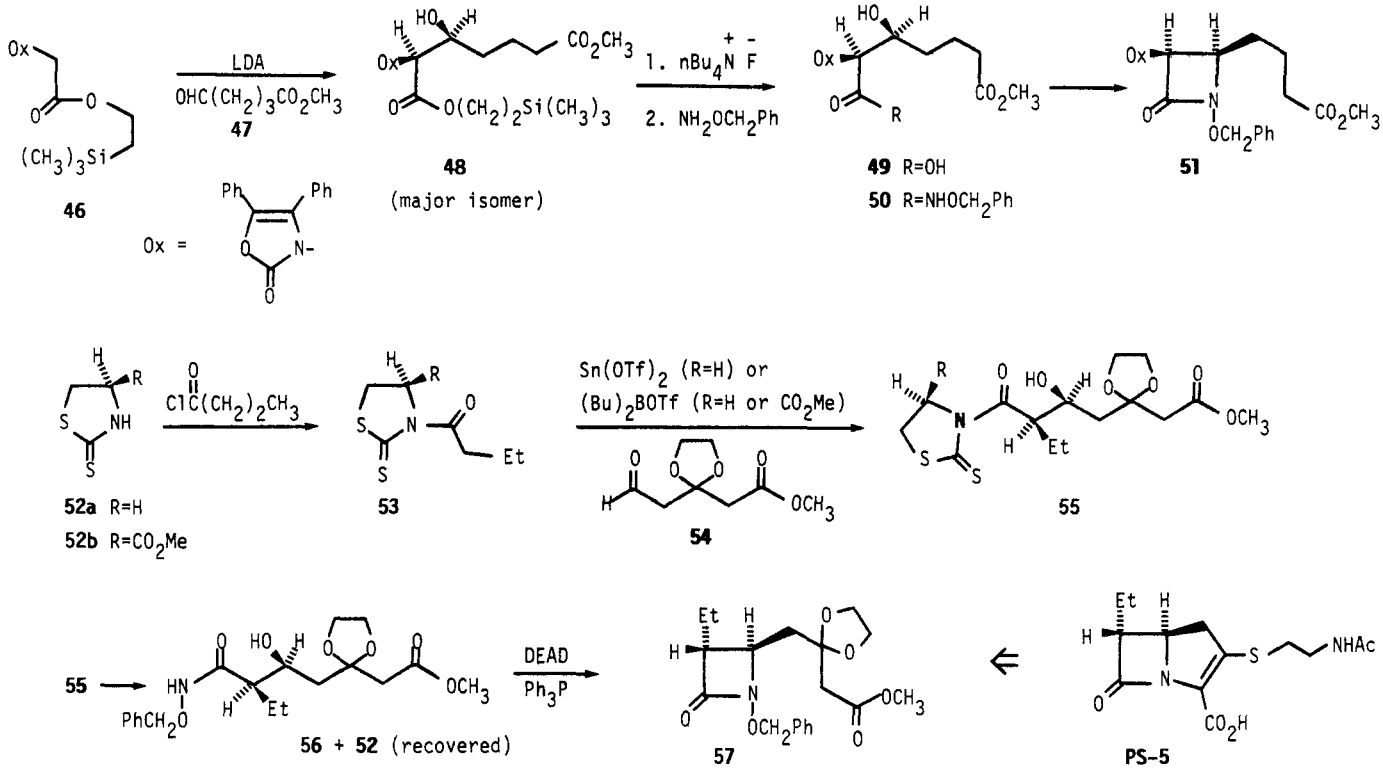
(34) Baldwin, J. E.; Adlington, R. M.; Birch, D. J. *J. Chem. Soc., Chem. Commun.* **1985**, *256*.

(35) Rajendra, G.; Biswas, A.; Miller, M. J., unpublished results.

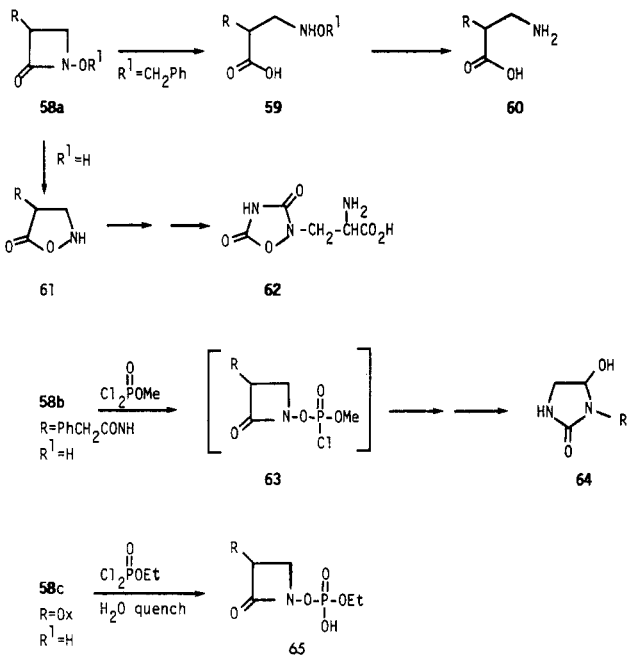
(36) Krook, M. A.; Miller, M. J. *J. Chem. Soc., Chem. Commun.*, in press.

(37) Biswas, A.; Miller, M. J.; Krook, M. A.; Woulfe, S. R. In "Recent Advances in the Chemistry of β-Lactam Antibiotics"; Brown, A. G., Roberts, S. M., Eds.; Royal Society of Chemistry: London, 1985; Chapter 21.

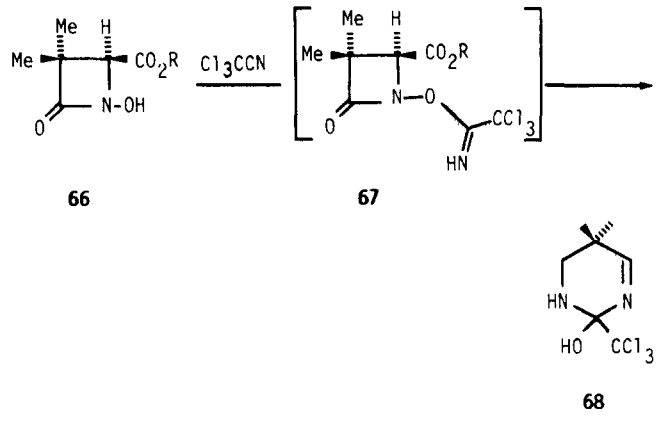
Scheme VIII



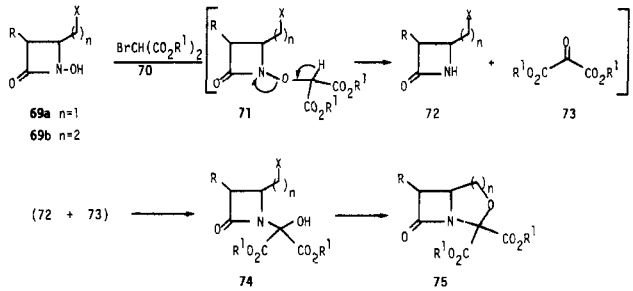
Scheme IX



Scheme X



Scheme XI



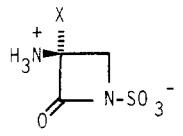
a 4-(halomethyl) or 4-(haloethyl) substituent (**69a, 69b**), this reaction provided direct access to bicyclic β-lactam analogues of isooxapenams **75a** and 3-oxacephams **75b**.[38] Fortunately these rearrangements did not preclude development of novel N-heteroatom containing β-lactams with significant biological activity.

Incorporation of N-Heteroatom Linkages into β-Lactam Antibiotics

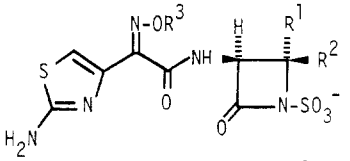
During our syntheses of substituted *N*-hydroxy-2-azetidinones, we noted the high β-lactam carbonyl frequency in their IR spectra and the increased susceptibility of these compounds toward nucleophilic attack at the carbonyl group, relative to N-unsubstituted β-lactams. The intriguing question was whether this heteroatom-induced chemical activation could be used to provide new biologically active compounds. The timely discovery of the monobactams **5**[6b] (sulfazecins)[6a] with their N–SO_3 linkage provided further incentive to explore this concept.

Although the natural monobactams exhibited only modest activity against Gram-negative bacteria, their

(38) Lee, B. H.; Biswas, A.; Miller, M. J. *J. Org. Chem.* **1986**, *51*, 106.



76 X=OCH_3
77 X=H

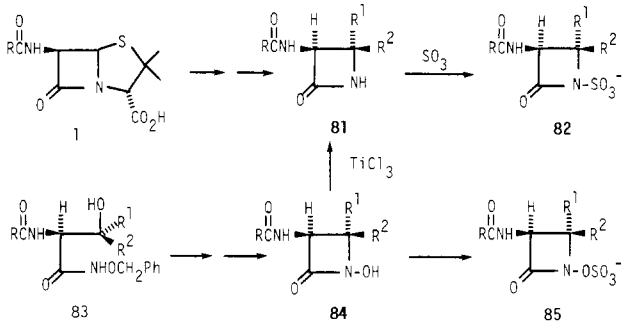


78 aztreonam (R^1=CH_3, R^2=H, R^3=$C(CH_3)_2CO_2H$)
79 carumonam (R^1=H, R^2=CH_2OCONH_2, R^3=CH_2CO_2H)
80 R^1=H, R^2=CH_2F, R^3=CH_3

novelty and structural simplicity coupled with difficulty in obtaining the parent nuclei, 3-methoxy-3-aminomonobactamic acid (**76**, 3-MAMA) or 3-aminomonobactamic acid (**77**, 3-AMA) from biological sources, encouraged significant synthetic interest. While the 3-methoxy group imparts increased β-lactamase stability to the natural monobactams (**5a**: X = OMe), the same group decreases their chemical stability. This fact, plus the inherent difficulties anticipated in synthesis of the corresponding chiral quaternary centers, encouraged the preparation of nonmethoxylated derivatives for further studies of structure–activity relationships. An early route to 3-AMA and the corresponding 3-acylamino derivatives developed by the Squibb group[6b] was based on the classical degradation of penicillin to the novel 3-(acylamino) N-unsubstituted β-lactams **23** previously utilized for the synthesis of the nocardicins (Schemes IV and XII). Subsequent sulfonation of the 3-(acylamino)-2-azetidinones provided direct access to a number of monobactams. However, the inherent incompatability of this method with simple C_4 substitution pointed out the need for alternative syntheses. The hydroxamate approach C_4-substituted N-unsubstituted β-lactams followed by direct N-sulfonation was immediately recognized as a more versatile route to a variety of substituted monobactams (Scheme XII). Significant extensions of this approach and related N–C_4 cyclizations have been elaborated by the Squibb group[20,39] and have resulted in their development of the clinical candidate aztreonam (**78**). Subsequently, other groups have prepared related monobactams (i.e., **79**[40] and **80**[41]) with activity similar to aztreonam against Gram-negative organisms.

The significant antibiotic activity of aztreonam and related monobactams suggests that an activated β-lactam ring with a peripheral ionizable group may be all that is required for biological activity. Thus, it was not inconceivable that our *N*-hydroxy compounds themselves might display antibiotic activity. However, early biological screening of 3-(phenylacetamido)-*N*-hydroxy-2-azetidinone (**84**: R = $PhCH_2$; R^1 = R^2 = H; p*K* ~6.5)[42,43] indicated that it had no significant activity. This result suggested that more appropriate targets might be β-lactams that retain the heteroatom activation but include a spacer before the ionizable group. Thus, one of the first attempts to demonstrate the generality of heteroatom activation was the direct sulfonation of *N*-hydroxy β-lactams (**84** → **85**; Scheme XII).[17,43] Indeed, the resulting monosulfactams **85** are reasonably stable and display even slightly better activity than the corresponding monobactams. Although the initially prepared monosulfactams (**85**: R^1 or R^2 = Me or R^1 = R^2 = H) also appeared to be somewhat more susceptible to inactivation by β-lactamase enzymes,[39b] a recent disclosure indicated that the 4-dimethyl derivative (**85**: R^1 = R^2 = Me) is remarkably β-lactamase stable and orally active.[44]

Scheme XII



Logical structural analogues of the monobactams and monosulfactams include the corresponding carboxylates **86a** or **86b**. However, attempts to prepare a sample of **86a** were frustrated by decarboxylation to form the corresponding N-unsubstituted β-lactam **81**.[39b] Alternatively, the Merck group has reported the synthesis of *N*-(tetrazol-5-yl) β-lactams **86c**.[45] These tetrazolyl analogues of the carboxylates, **86a**, reportedly display moderate to potent antibiotic activity. The massive efforts of the Squibb group have also produced the monophosphams **86d** and monocarbams **86e** that, with appropriate substitution, have activity similar to the corresponding monobactams.[39a,b]

Another noteworthy structural feature of the monobactams **82**, monophosphams **86d**, monocarbams **86e**, and tetrazols **86c** is that, in each case, the ionizable position is only two atoms from the β-lactam nitrogen. All of the more classical antibiotics such as the penicillins **1** and cephalosporins **2** have the ionized carboxylate oxygen positioned three atoms away from the ring nitrogen. The monosulfactams **85** also have the same three-atom spacing from the β-lactam nitrogen to the ionized position. However, the differences in the lengths of C–O and S–O bonds suggest that the actual spatial positioning of the charge in the monosulfactams **85** may be even more distant than in the classical an-

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(40) Kishimoto, S.; Sendai, M.; Hashiguchi, S.; Tommoto, M.; Satoh, Y.; Matsuo, T.; Kondo, M.; Ochiai, M. *J. Antibiot.* **1983**, *36*, 1421.

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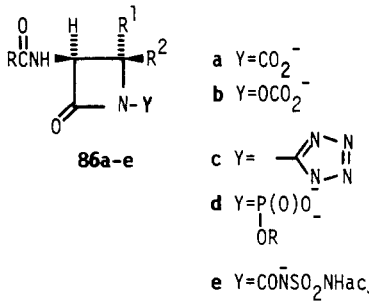
(42) Kindly performed at Eli Lilly and Co.

(43) Gordon, E. M.; Ondetti, M. A.; Pluscec, J.; Cimarusti, C. M.; Bonner, D. P.; Sykes, R. B. *J. Am. Chem. Soc.* **1982**, *104*, 6053.

(44) Koster, W. H.; Slurarchyk, W. A.; Dejneka, T.; Kronenthal, D.; Perri, M. G.; Pilkiewicz, F. G.; Routh, F. L.; Sundeen, J. E.; Weaver, E. R.; Zahler, R. 25th Interscience Conference one Antimicrobial Agents and Chemotherapy, Minneapolis, MN, 1985; Abstract 368.

(45) Andrus, A.; Partridge, B.; Heck, J. V.; Christensen, B. G. *Tetrahedron Lett.* **1985**, *25*, 911.



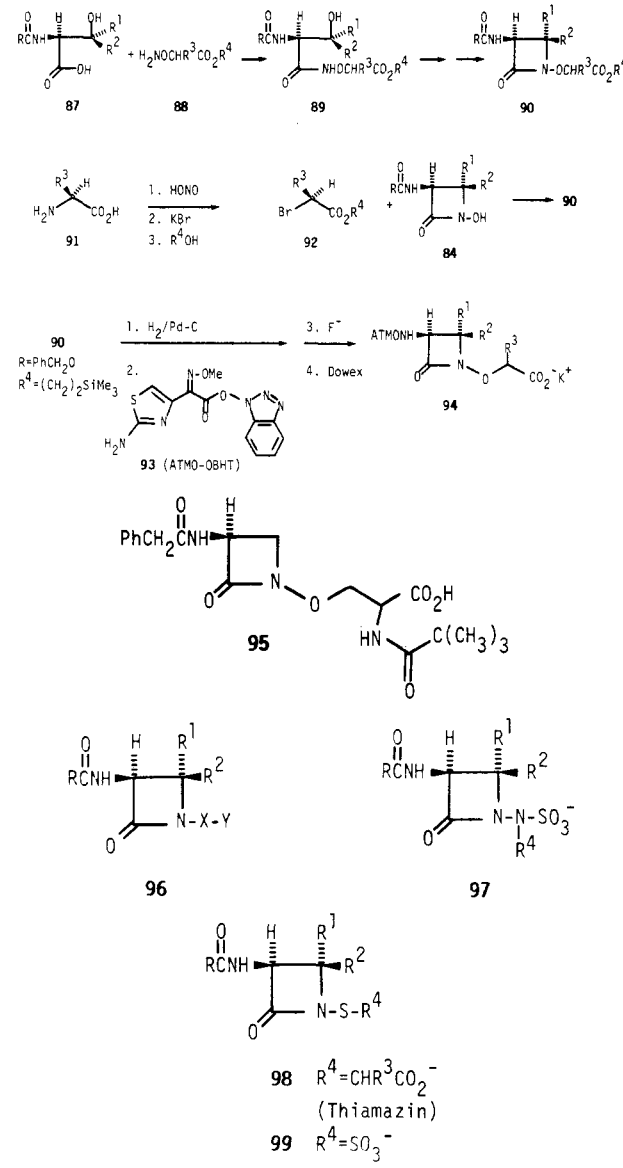
86a-e

tibiotics. The question then arose as to whether the ionizable group could be moved even further from the ring with retention of biological activity if heteroatom or another type of activation of the β-lactam system was maintained.

To test the compatibility of remote positioning of the ionizable group in heteroatom-activated β-lactams, we decided to synthesize examples of [[3(*S*)-(acylamino)-2-oxo-1-azetidinyl]oxy]acetic acids (**90**, oxamazins).[37,46] We found that simple incorporation of aminooxyacetate esters (88) into our usual hydroxamate-based synthetic sequence provided the basic oxamazin nucleus in good yields. However, we also sought a more versatile method that would allow us to prepare chiral alkylated derivatives of the oxyacetic acid portion without requiring the preparation of individual substituted α-aminooxyacetates (88, $R^3 \neq H$). The simple reaction of the potassium salt of *N*-hydroxy-3-(phenylacetamido)-2-azetidinone (84: R = $PhCH_2$; $R^1 = R^2 =$ H) with benzyl bromoacetate (92: R^3 = H; R^4 = $PhCH_2$) demonstrated the utility of a simple direct alkylation approach to the synthesis of oxamazin derivatives **90** (Scheme XIII). In contrast to the previously described attempted alkylation of *N*-hydroxy β-lactams with bromomalonates (Scheme XI), no competitive rearrangements were observed during the alkylation with bromoacetates.

Preliminary antibacterial tests of **90** (R = $PhCH_2$, $R^1 = R^2 = R^3$ = H, $R^4 = K^+$) indicated that it had modest activity against Gram-negative organisms. This result prompted us to replace the phenylacetyl group with the biologically more responsive 2-(2-amino-4-thiazolyl)-2-(*Z*)-(methoxyimino)acetamido (ATMO) side chain (Scheme XIII, **90** → **94**). A variety of substituents (R^3) were also incorporated into the oxyacetic acid portion to determine their effect on the biological activity of the oxamazins. Interestingly, the unsubstituted parent compound **94** ($R^1 = CH_3$; R^2, R^3 = H) was the most biologically active of all of the derivatives prepared. Its activity paralleled closely that of the corresponding monobactam, even though the ionized position of the oxamazin is four atoms from the β-lactam nitrogen. Recently, the Squibb group has also reported the synthesis and study of a number of oxamazins, including orally active ester derivatives.[47] We also prepared and tested an example of a substituted homoxamazin **95**.[48] Unfortunately, **95** has very little antibiotic activity and

Scheme XIII



is only a modest β-lactamase inhibitor. Whether this poor activity is due to the remoteness of the ionizable carboxyl group or the pivalamide substituent has yet to be determined.

If the activity of the oxamazins and other monobactams is primarily due to the heteroatom activation effect, one can imagine development of a number of potential antibiotics with the general structure **96** where X is an activating substituent and Y is an ionizable group. Syntheses of several possible examples of derivatives of **96** are being explored in our laboratory and others. A recent patent[49] has described the synthesis of sulfonated *N*-amino β-lactams **97**, which are aza analogues of the monosulfactams **85**. Though not quantitatively described, forms of **97** apparently display good antibacterial activity.

To help determine whether the heteroatom activation effect on the biological activity of monocyclic β-lactams, generalized by structure **96**, parallels the electronegatively of X, we decided to synthesize the N–S-containing β-lactams **98** and **99**. Treatment of variously substituted β-lactams **100** with substituted thiophthali-

(46) (a) Woulfe, S. R.; Miller, M. J. *Tetrahedron Lett.* **1984**, *25*, 3293. (b) Woulfe, S. R.; Miller, M. J. *J. Med. Chem.* **1985**, *28*, 1447.

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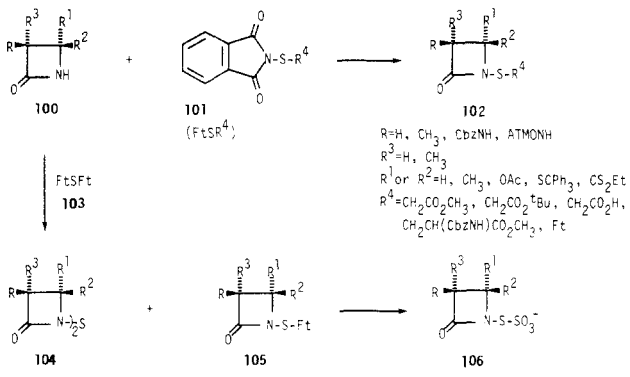
(48) Krook, M. A.; Miller, M. J., unpublished results. We thank Therese Debiak for the β-lactamase studies.

(49) Breuer, H.; Denzel, T. German Patent DE 31 22 795 A1.

Scheme XIV



mides **101** in the presence of a catalytic amount of triethylamine gave the correspondingly substituted *N*-thio-2-azetidinones **102** (Scheme XIV).[50] Specific reaction of 3-acylamino N-substituted β-lactam with *tert*-butyl *S*-phthalimido-α-thioacetate (**101**: R^3 = CH_2CO_2-*t*-Bu) followed by removal of the *tert*-butyl ester provided the first examples of the thiamazins **98** (**102**: R = ATMONH; $R^2 = R^3 = H$; $R^1 = CH_3$; R^4 = CH_2CO_2H). Unfortunately, the biological activity of the thiamazins was not so amazing. They were devoid of antibacterial activity. Whether the lack of activity is simply due to the decreased electronegativity of sulfur relative to the oxygen of the oxamazins has not yet been determined.[51]

Interestingly, reaction of N-unsubstituted β-lactams **100** with bis(phthalimido) sulfur **103** produced either the mono- (**105**) or disubstituted products (**104**) depending on the conditions used.[50b] Reaction of the *S*-phthalimido β-lactams **105** with a variety of nucleophiles also proceeds by direct reaction at sulfur[50b] and should allow elaboration to a number of novel structures. For example, preliminary indications are that we have prepared a sulfur analogue (**106**) of the monosulfactams by simply treating **105** with sodium bisulfite. Again, we were quite surprized to learn from preliminary biological screening of forms of **106** that simple replacement of the oxygen of the monosulfactams **85** with a sulfur atom resulted in complete loss of antibiotic activity, if, as with the thiamazins **98**, we assume that **106** is stable under physiological conditions. The synthesis and study of the monosulfactams **85**, along with the aza (**97**) and thia (**99**, **106**) analogues, completes a simple series for the generalized structure **96** (X = O, NR, S; Y = SO_3^-) and suggests that the order of activity simply parallels the electronegativity (O > N > S ≈ C) of the atom directly attached to the β-lactam nitrogen. Further studies of groups with increased "effective electronegativity" are in progress along with studies related to the "goodness of fit" of these various substrates in appropriate enzyme active sites.

In summary, the hydroxamate-mediated approach to the synthesis of β-lactams by an N–C_4 bond closure has considerable versatility. Depending on the availability of starting chiral β-hydroxy acids, it appears that nearly any corresponding optically active β-lactam can be prepared and elaborated to a variety of natural and unnatural antibiotics. Retention of the N–O bond has also led to further development of the concept of heteroatom-activated β-lactam antibiotics. Thus, the rational design and practical total synthesis of new antibiotics to augment our antimicrobial defense has become possible. However, much more remains to be done!

I am sincerely grateful for the tremendous efforts of my coworkers, most of whom are indicated in the references, and the supportative attitude of my colleagues and staff at Notre Dame (especially Prof. J. P. Freeman for his early encouragement and suggestions) and Eli Lilly. The financial support over the years of the Research Corp., the Petroleum Research Fund, administered by the American Chemical Society, the NIH, an NIH Research Career Development Award, the A. P. Sloan Foundation, Eli Lilly and Co., and Ajinomoto Co. Inc. is also sincerely appreciated. The love, patience, and understanding of my family during these busy times will be forever appreciated.

(50) (a) Woulfe, S. R.; Iwagami, H.; Miller, M. J. *Tetrahedron Lett.* **1985**, *3891*. (b) Woulfe, S. R.; Iwagami, H.; Miller, M. J., submitted for publication.

(51) Several other possibilities are being explored including (a) stability of the thiamazins under physiological conditions, (b) significant restricted rotation about the N–S bond may force the ionizable carboxyl group into an unfavorable position, and (c) increased distance of the carboxyl carbon from other centers on the β-lactam. A recently obtained X-ray structure of **107** indicated that the distances (*a*–*e*) from the carboxyl carbon are consistently larger than the corresponding distances of active antibiotics.[8]