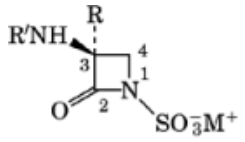
Kirk-Othmer Encyclopedia of Chemical Technology.

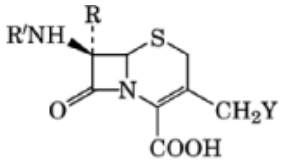
MONOBACTAMS

1. Monobactams

β-Lactam antibiotics are one of the best established classes of antimicrobial agents for the treatment of infectious diseases. Since the discovery of penicillin in 1929 and cephalosporin C in 1955, a large number of semisynthetic analogues has become available. In addition, more sophisticated screening and isolation techniques have afforded a variety of new naturally-occurring β-lactams such as the cephamycins, clavulanic acid, nocardicin, and the carbapenems. In late 1979 another entirely new class of β-lactams, characterized by the 2-oxoazetidine-1-sulfonic acid moiety (**1**, R = H or OCH_3, R′ = acyl), was discovered (1, 2). To distinguish this class of monocyclic, bacterially-produced β-lactam antibiotics from the bicyclic cephalosporins and cephamycins (**2**, R = H or OCH_3, R′ = γ-linked α-aminoadipoyl), the term monobactam was introduced (1).



(**1**)



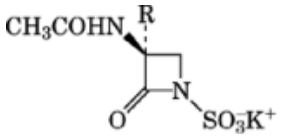
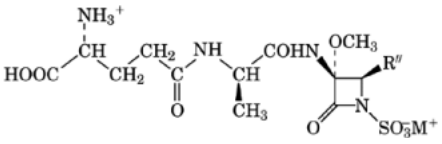
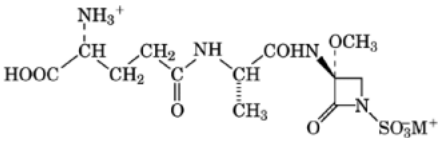
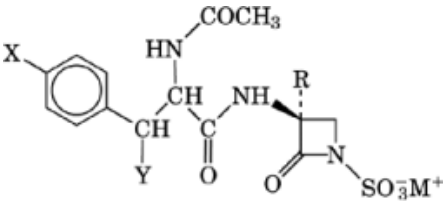
(**2**)

By screening strains of bacteria that were specifically responsive to β-lactam antibiotics, monobactams, varying in substitution at the C-3 position, were identified. The structures of the compounds along with their producing organisms are shown in Table 1. More recently, the 4β-methyl analogue (**4**, R″ = CH_3) of SQ 26,445/sulfazecin (**4**, R″ = H) was isolated using a differential antibacterial assay (3).

1.1. Structure Determination and Biological Properties

The structure and absolute stereochemistry of SQ 26,445 (sulfazecin) (**4**, R″ = H) was unequivocally established by x-ray crystallography (4) and that of SQ 26,180 (**3**, R = OCH_3) was determined by synthesis from a homochiral precursor of defined structure. Starting with 7-aminocephalosporanic acid (7-ACA), stereospecific introduction of the methoxyl group and degradation of the thiazine ring ultimately afforded the corresponding

Table 1. Naturally Occurring Monobactams

Organism	Monobactam	CAS Registry Number	Molecular formula	Structure number	Structure
Chromobacterium violaceum	SQ 26,180	[79720-08-4]	$C_6H_{10}N_2O_6S{\cdot}K$	(**3**, R = OCH_3)	
Gluconobacter oxydans	SQ 26,445	[77912-79-9]	$C_{12}H_{20}N_4O_9S$	(**4**, R″ = H)	
Pseudomonas acidophila	sulfazecin	[77912-79-9]	$C_{12}H_{20}N_4O_9S$	(**4**, R″ = H)	
Pseudomonas mesoacidophila	isosulfazecin	[77900-75-5]	$C_{12}H_{20}N_4O_9S$	**(5)**	
Agrobacterium radiobacter				**(6)**	
		[79720-13-1]	$C_{15}H_{19}N_3O_7S{\cdot}Na$	(**6**, X = H, Y = H, R = OCH_3, M = Na)	
		[79720-14-2]	$C_{15}H_{19}N_3O_8S{\cdot}K$	(**6**, X = OH, Y = H, R = OCH_3, M = K)	
		[81919-28-0]	$C_{14}H_{17}N_3O_7S{\cdot}K$	(**6**, X = OH, Y = H, R = H, M = K)	
		[79720-16-4]	$C_{15}H_{19}N_3O_{12}S{\cdot}2Na$	(**6**, X = $OSO_3^-Na^+$, Y = OH, R = OCH_3, M = Na)	
		[79720-17-5]	$C_{15}H_{19}N_3O_{15}S_3{\cdot}3Na$	(**6**, X = $OSO_3^-Na^+$, Y = $OSO_3^-Na^+$, R = OCH_3, M = Na)	

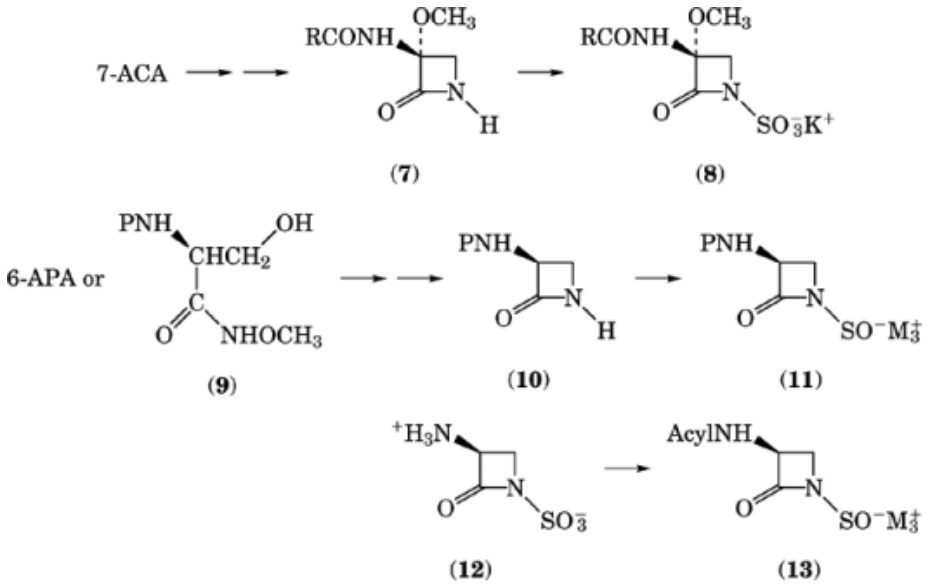
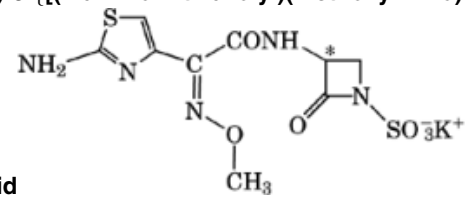


Fig. 1. Synthesis of C-3 side chain analogues of monobactams, where P is an amine protecting group and other terms are defined in text.

Table 2. Biological Activity of (*S*)- and (*R*)-3-{[(2-amino-4-thiazolyl)(methoxyimino)-acetyl]amino}-2-oxo-1-azetidinesulfonic acid



Absolute configuration[a]	CAS Registry Number	Molecular formula	MIC, μg/mL[b]
(*S*)-	[78626-19-4]	$C_9H_{11}N_5O_6S_2$	<0.05
(*R*)-	[78612-61-0]	$C_9H_{11}N_5O_6S_2$	>100

[a] At C-3.
[b] For *P. rettgeri*.

N-1 unsubstituted azetidinone. Subsequent sulfonation confirmed the structure of SQ 26,180 (**8**, R = CH_3) (Fig. 1) (5). The structure and absolute configuration of the nonmethoxylated naturally-occurring monobactam (**6**, X = OH, Y = R = H, M = K) was also proven by total synthesis (6). The monobactams **(1)** correspond in configuration to the naturally-occurring cephalosporins and cephamycins **(2)**. And, as is the case with regard to bicyclic β-lactams, monobactams of opposite absolute configuration to those encountered in nature are inactive. The enantiomeric pair of monobactams shown in Table 2 provide a striking illustration of this specificity.

In penicillins, incorporation of a 6α-methoxyl substituent increases chemical stability three- to fivefold as measured by basic hydrolysis of the β-lactam moiety (7). Similar substitution of a 7α-methoxyl substituent in cephalosporins also results in a modest increase in chemical stability. In contrast, methoxylated monobactams

having simple side chains are much less chemically stable than the corresponding nonmethoxylated compounds. However, the methoxylated monobactams show an increased stability to β-lactamases when compared to their unsubstituted analogues. this relationship is demonstrated by comparing the V_{max}, the rate at saturating substrate concentration for SQ 26,180 (**3**, R = OCH_3) with that of the nonmethoxylated counterpart SQ 26,396 [79720-10-8] (**3**, R = H), $C_5H_8N_2O_5SK$, against the clinically important RTEM lactamase (8). The relative V_{max} (RTEM) are <0.01 and 12, respectively compared to cephaloridine at 100. Similarly, increased β-lactamase stability was also observed for the 7-methoxy-cephalosporins (9).

The monobactams, like pencillins and cephalosporins, interfere with the synthesis of bacterial cell walls. β-Lactam antibiotics bind to a series of penicillin-binding proteins (PBPs) on the cytoplasmic membrane and their antibacterial effect is believed to result from inhibition of a subset of these PBPs known as peptidoglycan transpeptidases. These enzymes are responsible for cross-linking adjacent peptide strands within the peptidoglycan structure of the cell wall, thus improving the integrity of the bacterial protective coat. Aztreonam [78110-38-0], $C_{13}H_{17}N_5O_8S_2$, a clinically efficacious monobactam, specifically binds to PBP-3 thereby preventing septation and subsequently leading to cell death (10).

The biosynthetic origin of monobactams has been elucidated by fermentation experiments using radioactively labeled amino acids (qv). The monocyclic ring of the naturally-occurring C-4 unsubstituted monobactams is derived from serine. Similar techniques have shown that the methyl moiety of the methoxyl group in 3α-methoxylated monobactams is derived from methionine (11).

All of the naturally-occurring monobactams discovered as of this writing have exhibited poor antibacterial activity. However, as in the case of the penicillins and cephalosporins, alteration of the C-3 amide side chain led to many potent new compounds (12). Furthermore, the monobactam nucleus provides a unique opportunity to study the effect of structural modifications at the N-1 and C-4 positions of the azetidinone ring on biological activity. In contrast to the bicyclic β-lactams, these positions on the monocyclic ring system are readily accessible by synthesis.

1.2. Synthesis

Initial syntheses employed the sulfonation of an N-1 unsubstituted azetidinone as the key step. The natural product SQ 26,180 (**3**, R = OCH_3) (**8**, R = CH_3) as well as other methoxylated monobactams were synthesized, starting from either 7-aminocephalosporanic acid [957-68-6] (7-ACA) or 6-aminopenicillanic acid [551-16-6] (6-APA), via sulfonation of the N-1 unsubstituted intermediates (**7** or **10**) as shown in Figure 1 (5, 13). Subsequently, many more C-3 side-chain analogues were prepared by this method. Examples include the sulfonation of the N-1 unsubstituted azetidinones (**10**) that carry protecting groups on the nitrogen atom attached to C-3. 3-Aminomonobactamic acid [79720-18-6] (3-AMA) (**12**), $C_3H_6N_2O_4S$, was readily formed by removal of the protecting group and could subsequently be acylated to afford a wide variety of side-chain monobactams (**13**). 6-APA proved to be a readily available source for 3-AMA (14, 15) which alternatively could be prepared from the *O*-methylhydroxamate (**9**) of L-serine (16).

A second, conceptually distinct chiral synthesis of monobactams was developed from β-hydroxy amino acids. As shown in Figure 2, cyclization of the acylsulfamate of an amino-protected *O*-mesylserine derivative (**14**, R = H) leads directly to the monobactam (**15**). This methodology was also applied to the synthesis of 4α- (**15**, R = CH_3) and 4β-methyl monobactams from L-threonine and allothreonine, respectively (17). The 3(*S*)-(*trans*)-3-amino-4(*S*)-methylmonobactamic acid [80082-65-1] (**16**, R = CH_3), $C_4H_8N_2O_4S$, is the key intermediate to aztreonam [78110-38-0] (**17**). $C_{13}H_{17}N_5O_8S_2$, the first monobactam to achieve the status of a clinically useful antibiotic. A similar synthesis was utilized for the preparation of carumonam [89638-04-8] **(19)**, $C_{12}H_{14}N_6O_{10}S_2$, the second monobactam to undergo clinical trials. The absolute stereochemistry at the C-3 and C-4 positions, as well as the functionality on the C-4 position, were controlled by starting with either L-threonate or L-(+)-tartaric acid. Thus the appropriately substituted acyl sulfamate **(18)**, analogous to (**14**), was obtained for conversion to carumonam **(19)** (18).

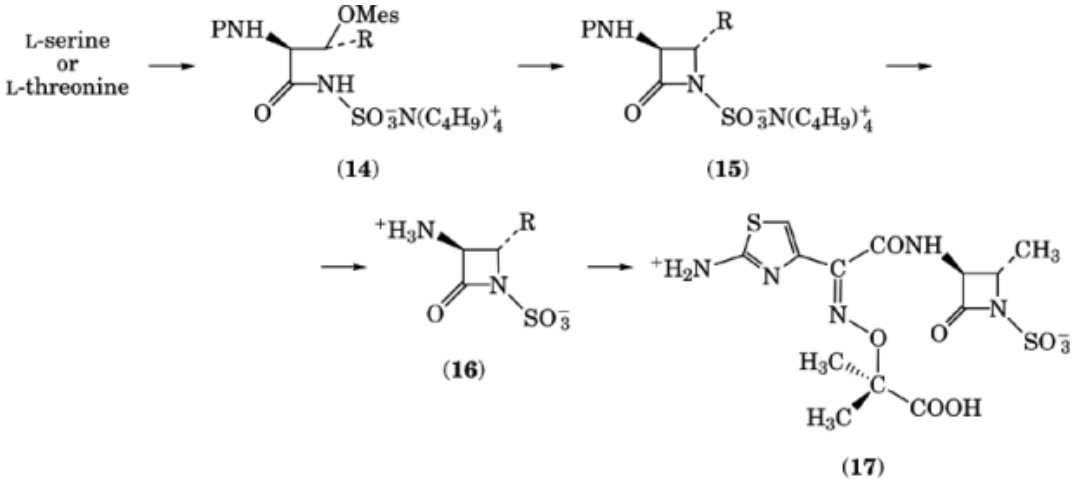
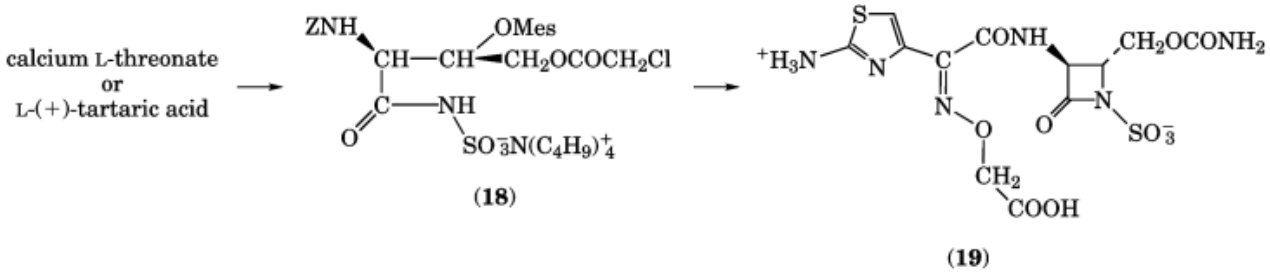


Fig. 2. Synthesis of clinically useful monobactams where R= H, CH_3; P is an amino protecting group, and Mes=mesyl is methanesulfonyl.



Although the activity of methoxylated monobactams could be improved by appropriate side-chain modifications, difficulty of synthesis and poor chemical stability focused attention on the nonmethoxylated analogues. Both high intrinsic activity and excellent β-lactamase stability are exhibited by monobactams that combine C-3 aminothiazole oxime side chains and 4-alkyl, 4-alkenyl, and 4-alkynyl groups (19).

1.2.1. Aztreonam

Aztreonam (**17**) is a totally synthetic compound having an antibacterial spectrum that is unique among β-lactam antibiotics. It exhibits potent and specific activity against a wide range of both β-lactamase-producing and nonproducing aerobic gram-negative bacteria, including *Pseudomonas aeruginosa*, but displays minimal inhibition against anaerobic and gram-positive aerobic, bacteria, eg, staphylococci and streptococci. When tested against strains of *Enterobacteriaceae*, aztreonam inhibited 50% of these isolates at concentrations <0.3 μg/mL (20). Particularly sensitive were members of *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus mirabilis*, *Morganella morganii*, *Serratia marcescens*, and many *Providencia sp.*. The activity of aztreonam is comparable to that of the aminoglycosides and third generation cephalosporins against these *Enterobacteriaceae*. Similarly, 50% of strains of ampicillin-sensitive and resistant *Haemophilis influenzae* and *Neisseria gonorrhoeae* were inhibited at <0.1 μg/mL.

Most dramatic were results with strains of *Pseudomonas aeruginosa* where aztreonam inhibited 50% of the strains at 4 μg/mL, and 90% at 16 μg/mL. Cessation of growth is not the sole consequence of exposure of these organisms to aztreonam as the inhibitory concentration is also generally the lethal concentration.

Aztreonam does not inhibit the majority of strains of *Acinetobacter sp.* and many strains of *Pseudomonas sp.*, eg, *P. maltophilia* and *P. cepacia*. Also little or no activity was seen against *Bacteroides fragilis*. Minimal inhibitory concentrations for the anaerobic and gram-positive aerobic bacteria were usually >100 μg/mL.

Many of the gram-negative isolates tested were multiresistant βlactamase-producing strains that are becoming more common and are gaining increasing clinical importance, particularly in nosocomial infections. Generally resistant to first- and second-generation cephalosporins and most of the penicillins, these organisms elaborate a diverse array of β-lactamases targeted at destroying β-lactam antibiotics. Aztreonam exhibits a high degree of resistance to enzymatic hydrolysis by most of the common β-lactamases (21). Particularly insidious are the plasmid-mediated TEM β-lactamases carried by a wide range of gram-negative bacteria. The plasmid-mediated genes responsible for enzyme production pass freely among the *Enterobacteriaceae*, pseudomonads, *Neisseria gonorrhoeae*, and *Haemophilus influenzae*. Other β-lactamases capable of this same promiscuous behavior are the OXA, SHV, and PSE enzymes. Aztreonam, similar to many of the third-generation aminothiazoleoxime cephalosporins, such as cefotaxime and ceftizoxime, is stable to these β-lactamases. In contrast, compounds like cephaloridine and cefoperazone are readily hydrolyzed. Aztreonam, cefotaxime, and ceftizoxime are also generally stable to the chromosomally mediated β-lactamases, which are widely distributed among specific gram-negative bacteria, but are not transferable. These enzymes, which may coexist in the same organisms with a plasmid-mediated β-lactamase, are found in organisms such as *Klebsiella*, *Providencia*, *Enterobacter*, *Serratia*, *Proteus*, and *Bacteriodes*. The only enzyme in this group showing any appreciable destruction of aztreonam is the relatively uncommon K-1 β-lactamase produced by some strains of *Klebsiella oxytoca*.

In vivo, aztreonam shows excellent pharmacokinetic properties. One hour after parenteral administration of a 1 g dose, serum levels are 45–50 μg/mL. After a 2 g dose, levels are 90 μg/mL at 1 h and there is no apparent accumulation of the drug after multiple dosing. Aztreonam is excreted primarily by the kidneys with two-thirds of an administered dose eliminated unchanged in the urine. Renal secretion, glomerular filtration, and nonrenal mechanisms are involved in the elimination of aztreonam. Serum half-life for elimination is 1.7 h. Aztreonam penetrates well into peritoneal fluid, pleural fluid, and blister fluid. Furthermore, aztreonam undergoes biliary secretion with peak biliary concentrations greater than 40 μg/mL at 2.5 h.

In the clinic, aztreonam has demonstrated efficacy in urinary tract infections (UTI), lower respiratory tract infections, bacteremias, skin and soft tissue infections, intraabdominal and pelvic infections, leukopenic patients, and bone and joint infections resulting from susceptible gram-negative pathogens, allowing for selective therapy or combination therapy if mixed pathogens are involved. Aztreonam markedly decreases the fecal aerobic gram-negative bacteria, selectively eradicating pathogenic bacteria from the intestinal tract without significantly altering the gut anaerobes or impairing their colonization-resistance function.

Overall, aztreonam appears to be a safe agent having toxicity side effects similar to those of other β-lactams. The safety profile suggests that aztreonam may be useful as a replacement for aminoglycoside therapy (22). The biological properties of aztreonam have been extensively reviewed (23).

1.3. Alternative N-1 Activating Groups

β-Lactam antibiotics exert their antibacterial effects via acylation of a serine residue at the active site of the bacterial transpeptidases. Critical to this mechanism of action is a reactive β-lactam ring having a proximate anionic charge that is necessary for positioning the ring within the substrate binding cleft (24).

All of the naturally-occurring monobactams and aztreonam are characterized by the presence of the N-1 sulfonate group, which serves a dual function. The electron withdrawing sulfonate moiety renders the β-lactam ring more reactive toward nucleophilic attack and at the same time provides the anionic charge necessary for binding. A variety of monobactam subclasses bearing N-1 activating groups having the necessary physical properties for antibacterial activity are listed in Table 3 (25).

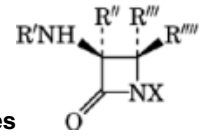
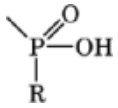
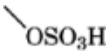
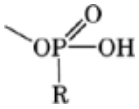
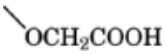
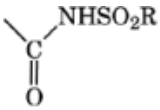
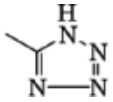


Table 3. Biologically Active Monobactam Subclasses

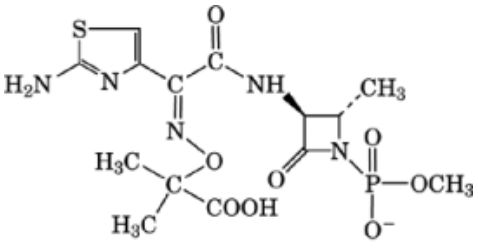
Subclass	Substituent, X
monobactams	
monophosphams	
monosulfactams	
monophosphatams	
monoxacetams (oxamazins)	
monocarbams	
tetrazole-activated monobactams	

1.3.1. Monophosphams

The similar tetrahedral geometry and bond lengths of tetracoordinate phosphorus(V) compared to those of sulfur(VI) suggested that phosphonic and phosphinic acid groups might act as biososteres for the sulfonic acid moiety in the parent monobactams. The 2-oxoazetidine-1-phosphonates and phosphinates (monophosphams) were prepared, as shown in Figure 3**a**, by lithiation of the 3-acylamino-2-azetidinones (**20**) followed by treatment with a phosphoryl or phosphonyl halide to yield the phosphonylated, or phosphinylated, azetidinones (**21**) or (**23**). Cleavage of these esters using trimethylsilyl bromide [2857-97-8] in the presence of bis-trimethylsilylacetamide [10416-59-8] (BSA), which serves as an acid scavenger, affords the diacidic and

monoacidic monophosphams (**22**) and (**24**), respectively (26, 27). Alternatively, Figure 3**b** reaction of lithiated azetidinones with phosphoryl dichlorides yields azetidinone-1-phosphonyl chlorides (**26**), which are solvolyzed to give the monoacidic ester (**27**) or aminolized to ultimately afford the phosphonamide (**29**). Cleavage of the urethane protecting groups by catalytic hydrogenation, when R = $C_6H_5CH_2O$, or by trifluororacetic acid, when R = *t*-C_4H_9O, gives 3-aminomonophosphamic acids. Coupling using [*N, N′*-dicyclohexylcarbodiimide (DCC)-*N*-hydroxybenzotriazole] or mixed anhydride with carboxylic acids formed 3-acylamino derivatives for screening (27).

The monophosphams are less intrinsically active, but more stable to β-lactamases, than their monobactam counterparts. SQ 27,327 [84486-64-6] **(30)**, $C_{14}H_{20}N_5O_8PS$, the most active monophospham, is generally two- to eightfold less active against aerobic gram-negative organisms than is its sulfonic acid analogue, aztreonam (**17**). SQ 27,327, however, is substantially more stable to the K-1 β-lactamase than aztreonam and this is reflected in the >250 − fold difference in antimicrobial activity against the enzyme-producing *Klebsiella* strain (Table 4) (27).



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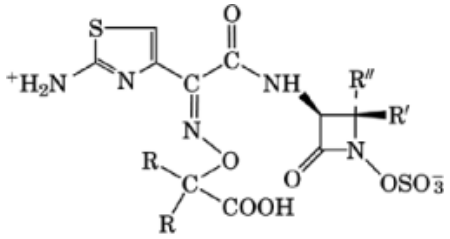

(**31**)

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1.3.2. Monosulfactams

The mosulfactams are a class of monobactams activated by an *N*-1-OSO^-;$_3$ group (28) resulting in good intrinsic activity against gram-negative bacteria. However, a C-4 unsubstituted (**31**, R = CH_3, R′ = R″ = H) [109895-38-7] and the monomethylated [87577-16-0] (**31**, R = CH_3, R′ = H, R″ = CH_3), $C_{13}H_{17}N_5O_9S_2$ and [102572-29-2] (**31**, R = CH_3, R′ = CH_3, R″ = H), $C_{13}H_{47}N_5O_9S_2$, monosulfactams have poor chemical and β-lactamase stability. Dimethylation at the C-4 position (**31**, R = R′ = R″ = CH_3) [109885-32-7] improves the chemical and β-lactamase stability remarkably, while maintaining a high degree of antibacterial activity. Furthermore, removal of the *gem*-dimethyl groups from the C-3 side chain of this C-4 dimethyl compound minimally affects β-lactamase stability (29), but affords an orally-absorbed monosulfactam SQ 30,213 [102507-71-1] (tigemonam) (**31**, R = H, R′ = R″ = CH_3), $C_{12}H_{15}N_5O_9S_2$ (30). The high degree of oral absorption (80% in mice) is atypical for monobactams. The oral absorption and efficacy of tigemonam in animals have been confirmed in humans in Phase I and Phase II clinical studies (31).

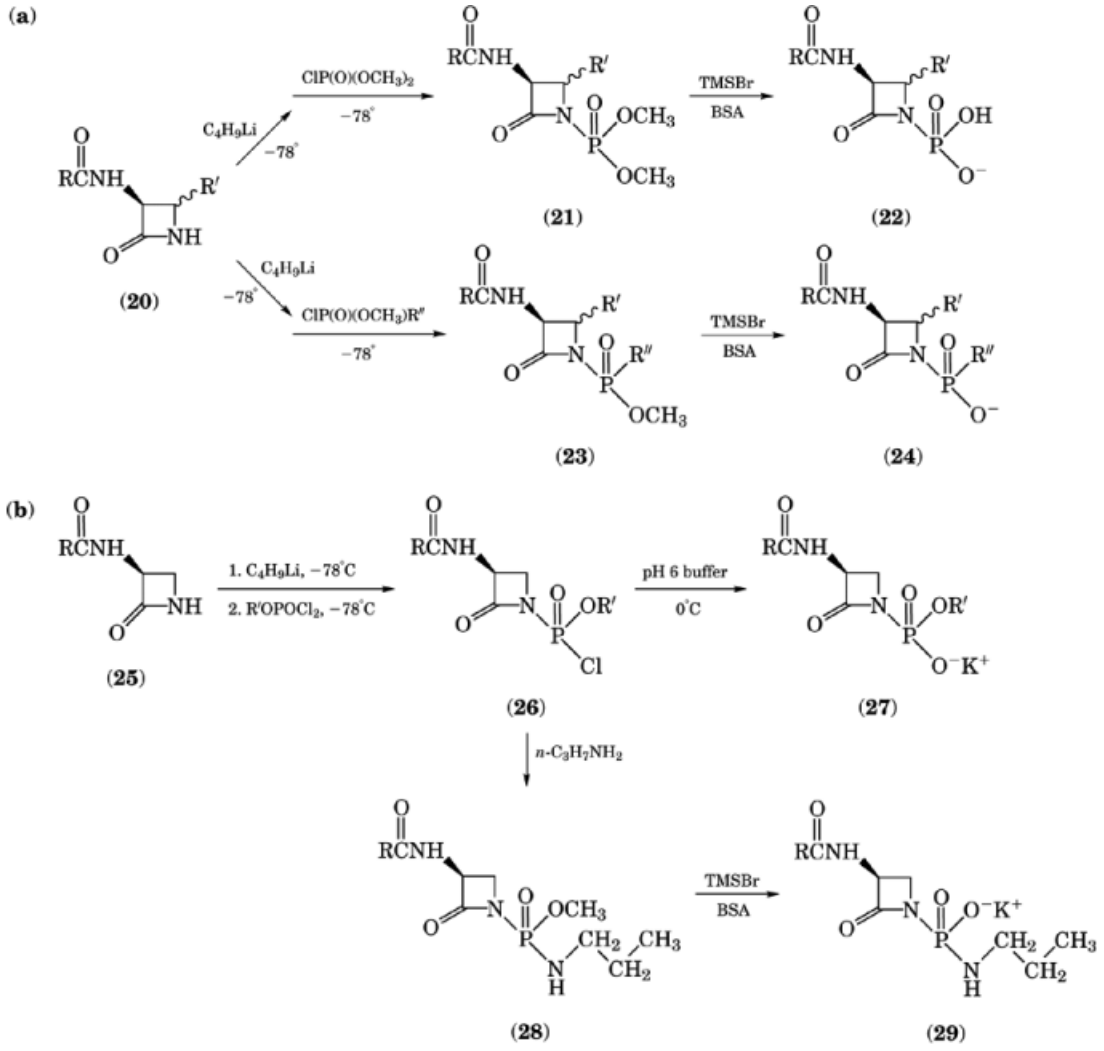
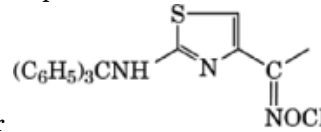


Fig. 3. Synthesis of diacidic and monoacidic N-1 azetidinone phosphonates and phosphinates where TMSBr is trimethylsilyl bromide; BSA is bis-trimethylsilylacetamide; (**a**) $_{R=}$ $C_6H_5CH_2O$, t-C_4H_9O, or  $_{R'=}$ H or CH_3; and $_{R''=}$ CH_3 or C_6H_5; and (**b**) $_{R=}$ t-C_4H_9O or $C_6H_5CH_2O$; and $_{R'=}$ CH_3, CH_2CH_2F, CH_2CF_3, n-C_4H_9, or C_6H_5.

Syntheses of the C-4 unsubstituted and monomethylated monosulfactams utilized hydroxamates derived from β-hydroxy amino acids such as serine or theronine, as precursors to the β-lactam ring (28). By analogy, synthesis of C-4 dimethylated monobactams (Fig. 4) required a protected form of (*S*)-β-hydroxyvaline [102509-19-7] (**33**). This unusual amino acid was prepared by reaction of the dianion of Boc-glycine benzyl ester [54244-89-8] (**32**) with acetone. Cleavage of the benzyl ester afforded the Boc-protected racemic valine [102507-13-1], $C_{10}H_{19}NO_5$, which was resolved as its phenethylamine salt to give Boc-(*S*)-β-hydroxyvaline.

Table 4. Biological Activities of Aztreonam and N-1 Substituted Analogues

Gram-negative organisms	SC #[b]	MIC, μg/mL[a] Aztreonam Low[c]	Aztreonam High[c]	SQ 27,327 Low[c]	SQ 27,327 High[c]	Tigemonam Low[c]	Tigemonam High[c]	SQ 28,112 Low[c]	SQ 28,112 High[c]
E. coli TEM+	10,404	0.1	0.2	0.4	0.8	0.4	0.8	0.4	0.8
E. coli TEM−	10,439	0.1	0.2	0.8	0.8	0.8	0.8	0.8	0.8
Ent. cloacae P99+	10,435	12.5	50	6.3	50	6.3	50	6.3	50
Ent. cloacae P99−	10,441	0.1	0.2	0.8	1.6	0.8	1.6	0.8	1.6
K. aerogenes K1+	10,436	100	>100	0.4	0.4	0.4	0.4	>100	0.4
K. aerogenes K1−	10,440	<0.05	0.1	0.4	0.8	0.4	0.8	0.8	0.8
K. pneumo.	11,066	0.4	0.8	1.6	1.6	0.8	1.6	3.1	25
C. freundii	10,204	0.1	25	0.4	3.1	0.2	0.4	0.8	50
Prot. rettgeri	8,217	<0.05	<0.05	0.2	0.2	<0.05	<0.05	<0.05	0.4
Prot. vulgaris	10,951B	<0.05	0.2	0.4	0.4	<0.05	<0.05	0.4	1.6
Ser. marcescens	9,782	0.2	0.2	1.6	3.1	0.8	1.6	0.8	3.1
Ps. aeruginosa	8,329	3.1	12.5	>100	>100	50	>100	3.1	12.5
Ps. aeruginosa	9,545	0.4	0.8	6.3	6.3	1.6	1.6	0.8	1.6

[a] Minimum Inhibitory Concentration measured by dilution of test compound in agar inoculated with microorganism.
[b] Squibb culture number.
[c] Low and high refer to the number of colony forming units (CFU) used to inoculate the media. Comparing MICs at low (10^4) and high (10^6) CFU, a large increase in MIC at the high inoculum level vs a β-lactamase producer is an indicator of instability via enzymatic hydrolysis.

Coupling *O*-benzylhydroxylamine with amino acid (**33**) afforded the hydroxamate (**34**). Selective sulfonation of the hydroxyl group of (**34**) and subsequent heating of the resulting sulfate (**35**) [107982-89-8] in the presence of base effected cyclization at the sterically hindered carbon atom and gave the β-lactam intermediate (**36**) [10250-25-5]. The final steps, benzyl cleavage, sulfonation, and coupling of the side chain, yielded tigemonam (**38**) (**31**, R = H, R′ = R″ = CH_3), and related analogues (32).

As shown in Table 4, tigemonam is also relatively inactive vs gram-positive bacteria and anaerobes, yet exhibits a high degree of activity against gram-negative organisms as well as resistance to hydrolysis by β-lactamases. The activity of tigemonam is cidal against the *Enterobacteriaceae*, *Hemophilus influenzae*, and *N. gonorrhoeae* but unlike aztreonam, the activity is weak against the pseudomonads (33, 34).

1.3.3. Monophosphatams

The preparation of monophosphatams, the phosphorus analogues of the monosulfactams, is exemplified by the reaction shown in Figure 5 (35). Phosphorylation of the *N*-hydroxyazetidinone [80542-48-9] (**39**) using methylphosphonic or methylphosphoric dichlorides in the presence of 2,6-lutidine or triethylamine at low temperatures gave the chloro intermediates (**40**), which were hydrolyzed *in situ* at pH 3–4 to give the stable potassium salts (**41**) in from 12 to 40% yield. Removal of the *t*-butoxycarbonyl group under acidic conditions at $-10^{\circ}C$ for 1 h gave the amine salts (**42**) in good yield. The desired acyl side chains were then coupled to

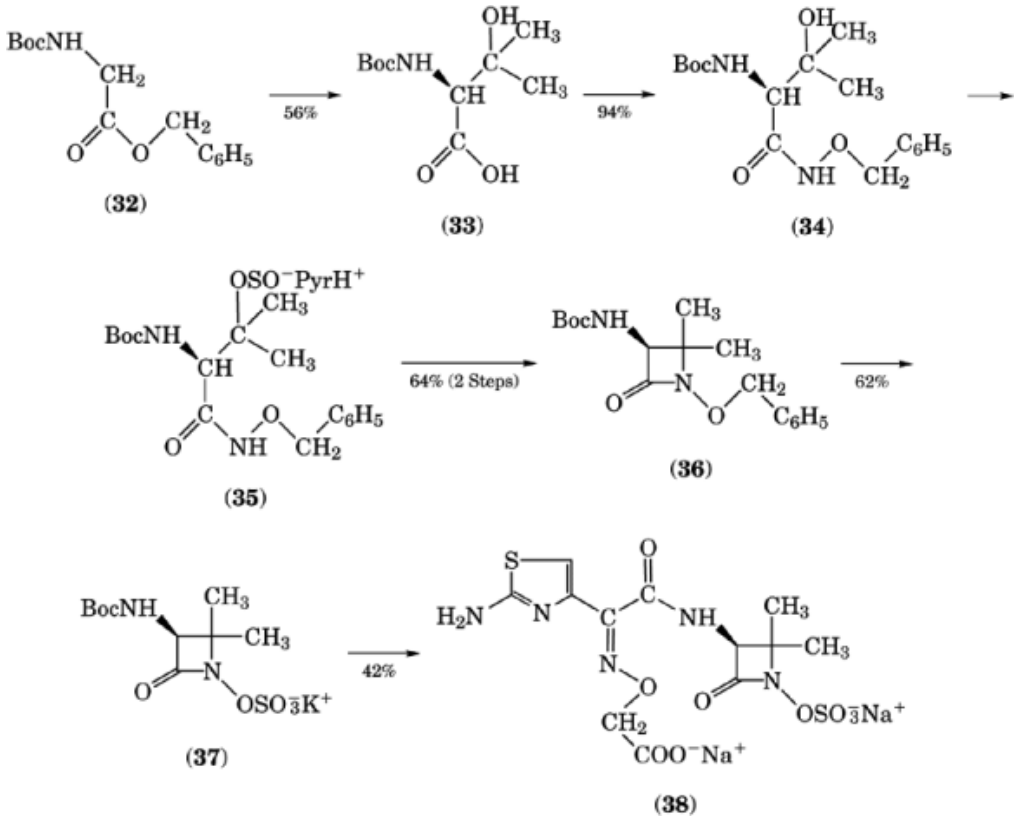


Fig. 4. Synthesis of tigemonam where Boc is the *t*-butoxycarbonyl group; $PyrH^+$ is the pyridinium ion; and the numbers given are percent yield of product.

the amino group using an activated ester of the side chain to afford various acyl derivatives such as SQ 28,112 [91603-73-5] (**43**), $C_{14}H_{20}N_5O_8PS$.

The biological activity of SQ 28,112 (**43**), shown in Table 4, is typical of the monophosphatams which possess the same spectrum of activity as aztreonam, but have comparatively lower intrinsic activity and β-lactamase stability.

1.3.4. Monoxacetams

Once the structural criteria for ring activation and binding of monobactams to the transpeptidases were established, researchers set out to determine if an oxygen atom could provide the requisite β-lactam ring reactivity. It was first necessary to insulate the anionic binding moiety from the oxygen atom by a "spacer" group, and using glycolate structures appended to the *N*-1 position of the β-lactam ring, structures **(44)** were synthesized (36). These 3-acylamino-1-azetidinyloxyacetic acids are known variously as monoxacetams and as oxamazins.

The oxyacetic acid residue of the monoxacetams is bioisosteric with the sulfate moiety of the monosulfactams. In addition, the presence of the carboxylate moiety provided opportunity for the preparation of esters that could act as orally absorbed prodrugs. Based on structure-activity relationships, SQ 82,291 **(45)** [90898-90-1], $C_{12}H_{15}N_5O_6S$, the nonacidic methoxime side chain of which was necessary to maintain oral absorption of the prodrugs, was prepared. SQ 82,291 has a high, specific affinity for the PBP-3 transpeptidase of gram-negative

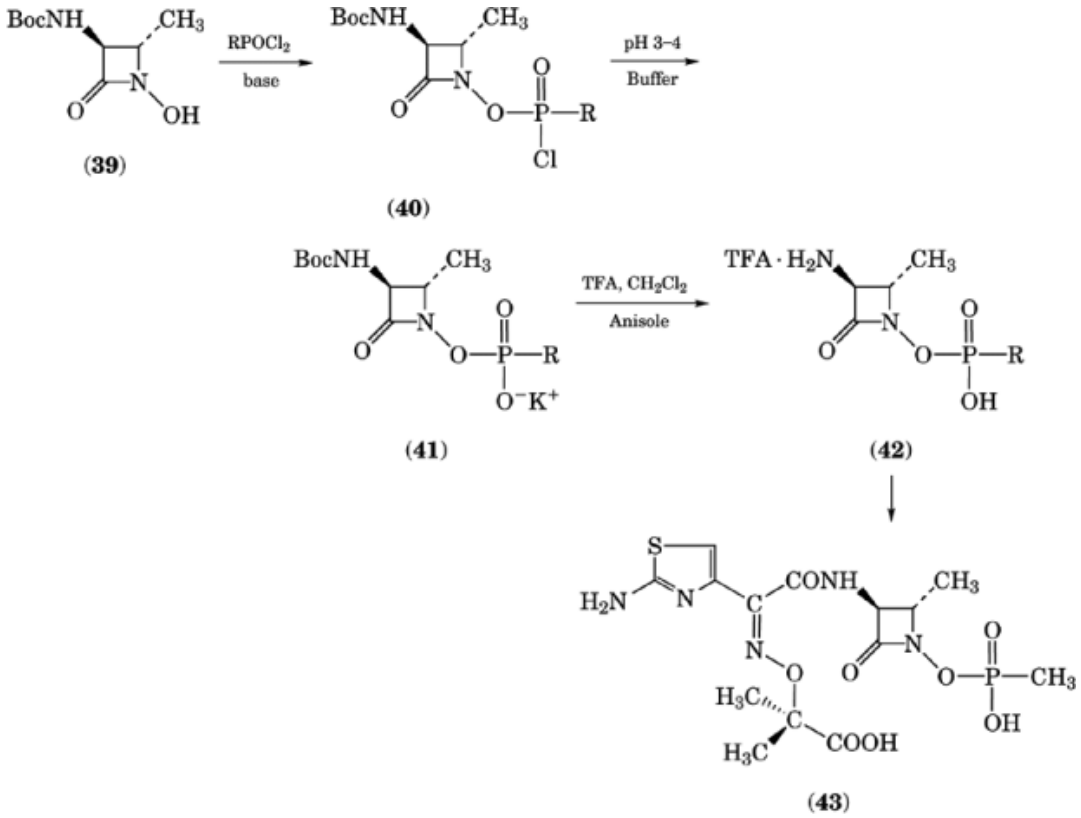
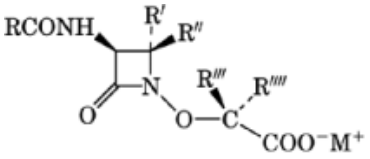
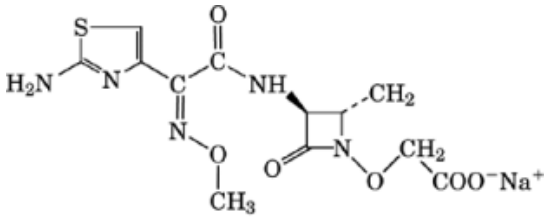


Fig. 5. Synthesis of monophosphatams where Boc is *t*-butoxycarbonyl; R is either CH_3 or OCH_3; and TFA is trifluoroacetic acid.

bacteria. However, it lacks the isobutyric acid moiety of aztreonam (**17**) on the oxime residue and whereas the activity of SQ 82,291 vs the *Enterobacteriaceae* was maintained, antipseudomonal activity was significantly diminished (37).

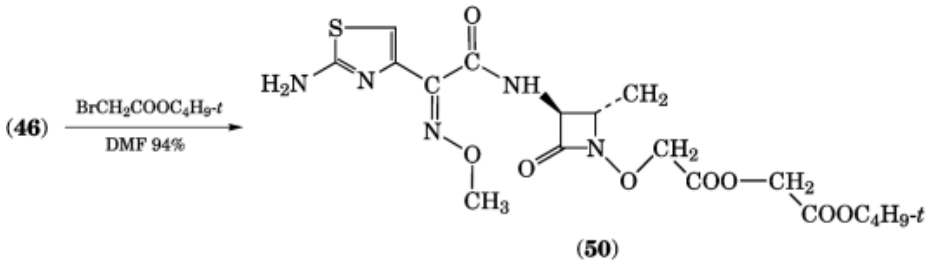




(45)

To synthesize the monoxacetam structures (Fig. 6), alkylation of *N*-protected 1-hydroxyazetidinones **(46)** with the appropriate haloacetic acid derivatives provided (**47**). Alternatively, (**47**) could be prepared from the acyclic hydroxamate ester (**48**). Deprotection of (**47**) furnished the zwitterionic intermediate (**49**) [90849-16-4], $C_6H_{10}N_2O_4$, which subsequently underwent acylation using the C-3 aminothiazole oxime side chain to afford SQ 82,291 **(45)** also known as oximonam (37).

The first oral monobactam to reach clinical trials was SQ 82,531 [90850-05-8] (gloximonam), **(50)**, $C_{18}H_{25}N_5O_8S$, the *t*-butyl glycolate ester of oximonam. This prodrug ester could be formed in high yield by direct alkylation of SQ 82,291 or by carrying the ester from the earlier synthetic stages (**47** or **48**, R′ = $-CH_2COOt\text{-}C_4H_9$). On a multikilogram scale, the overall yield via the route from *N*-hydroxyazetidinone **(46)** was ca 25% (38).



(50)

Gloximonam [90850-05-8] **(50)** was selected for clinical studies because of its desirable physical properties such as crystallinity and solubility and because it was orally absorbed in mice, rats, and monkeys, thus resulting in therapeutic serum levels of the active oximonam. Gloximonam was orally efficacious in a variety of murine gram-negative systemic infections (39) and in Phase I clinical studies gloximonam was well-absorbed. However, further clinical development was discontinued once it was determined that in humans the β-lactam ring underwent significant hydrolysis.

The *N*-aza (40) and *N*-thia (41) analogues of the monoxacetams have also been prepared. Within the nitrogen series, the imino acids [121142-86-7] (**51**, X = N=CHCOOH, R = H, CH_3) the imine stereochemistry of which is unknown, have moderate, notably less than oximonam anti-gram-negative activity. Both the *N*-glycyl [121142-76-5] (**51**, X = $NHCH_2COOH$, R = H) and *N*-thioacetic acid [102652-83-5] **(52)** derivatives were devoid of any significant biological activity.

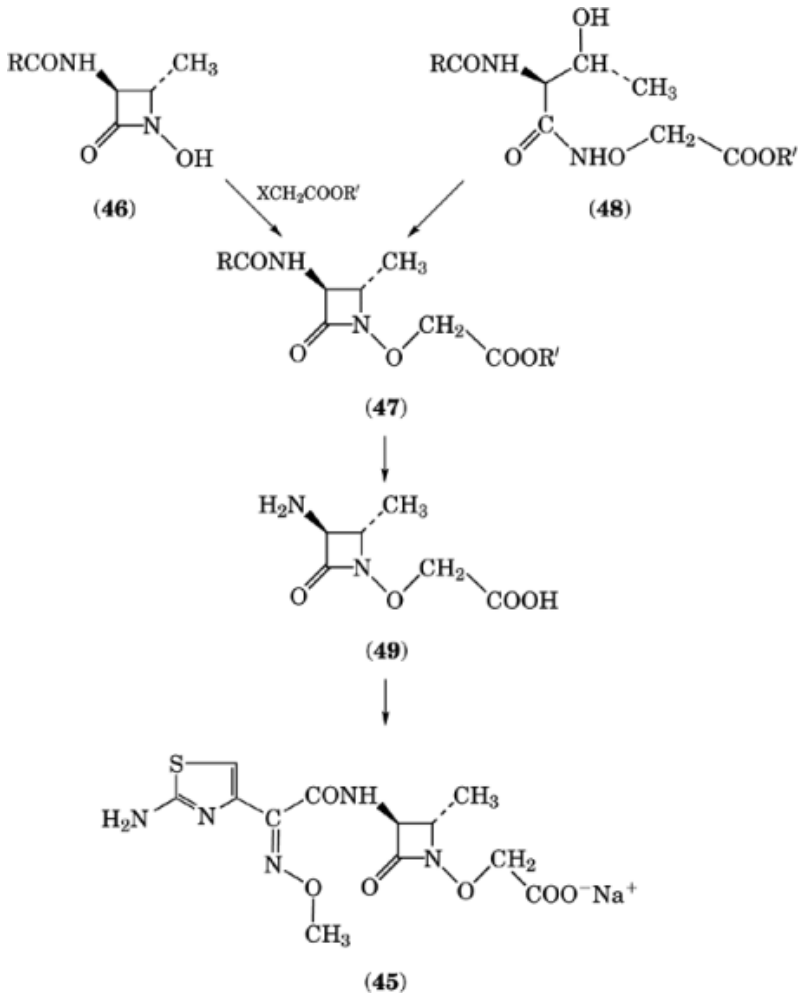
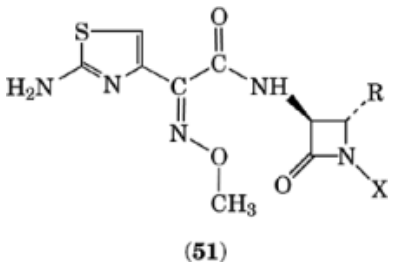
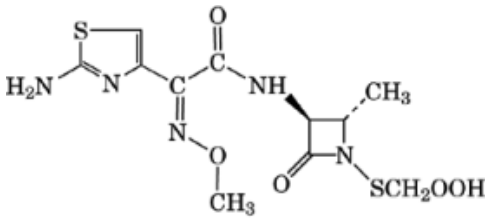


Fig. 6. Synthesis of monoxacetams.



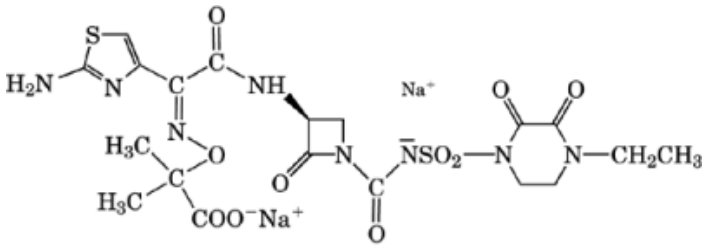


(52)

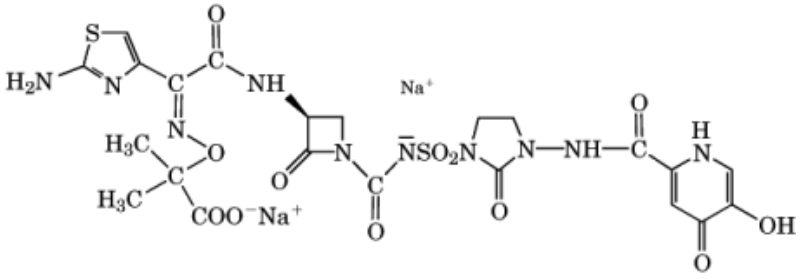
1.3.5. Monocarbams

The reaction sequence for monocarbam preparation is shown in Figure 7. Simple carboxyl *N*-1 substitution, representing one of the simplest carbonyl activating groups, led, as expected, to a carbamic acid that was hydrolytically unstable. Several monocarbams (**55**) and (**56**), having acidic sulfonylaminocarbonyl activating groups, were produced (42). The monocarbams (**55**) were prepared by sequential acylation of 3-aminoprotected 2-azetidinones (**53**) using alkyl or aryl isocyanates, removal of the protecting group, and acylation with activated side-chain acids. Reaction of (**53**) and chlorosulfonyl isocyanate (CSI) afforded the key intermediate (**54**) from which the aminosulfonyl derivatives (**56**) were prepared.

Monocarbams of the general formula (**56**), in which the terminal nitrogen is incorporated in a heterocyclic group, represent a class of potent β-lactamase stable antibacterials which are specifically active against gram-negative microorganisms. SQ 82,228 [109895-39-8] **(57)**, featuring an *N*-ethylpiperazinedione terminus, is one of the most active (42), displaying favorable pharmacokinetic properties in experimental animals and good efficacy when tested parenterally in model infections.



(57)



(58)

Continued efforts to improve the activity of the monobactams against nonfermenting gram-negative rods such as *Pseudomonas aeruginosa*, led to the discovery of SQ 83,360 [104393-00-2] **(58)**, $C_{22}H_{24}N_{10}O_{12}S_2 \cdot 2Na$, a 3-hydroxy-4-pyridone containing monocarbam. The enhanced activity of SQ 83,360 is a direct consequence

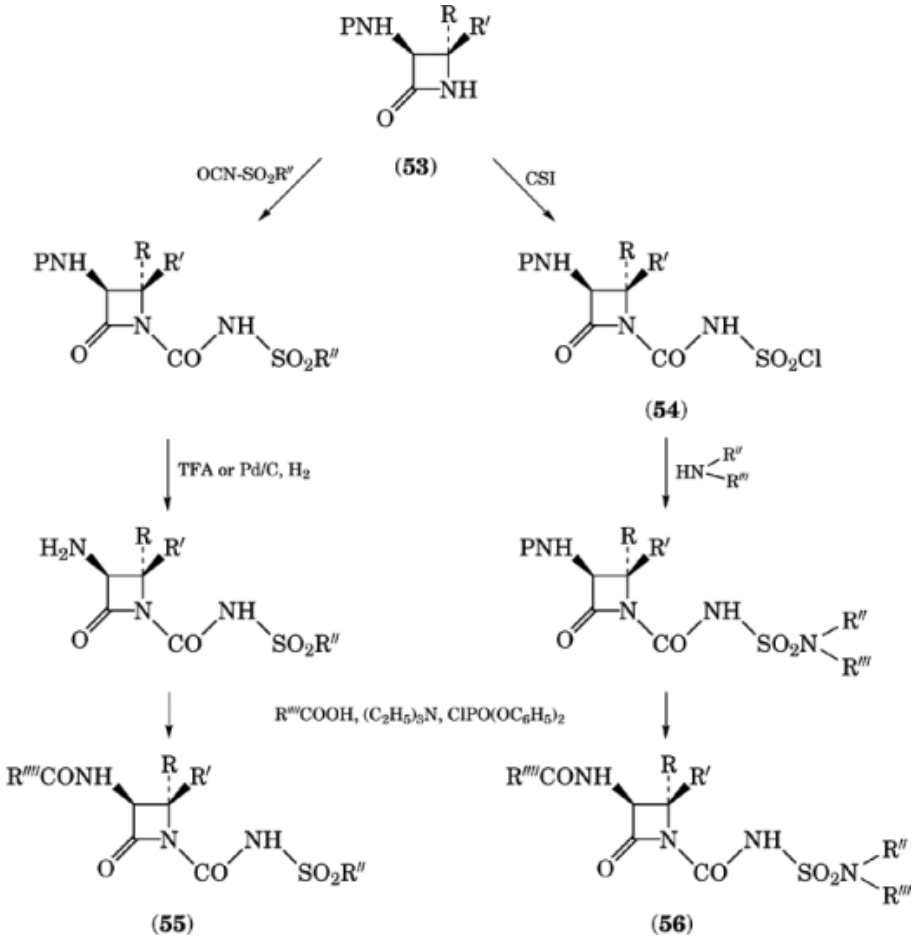


Fig. 7. Synthesis of monocarbams where P is an amino protecting group and CSI is chlorosulfonyl isocyanate. TFA is trifluoroacetic acid.

of the hydroxypyridone moiety, a catechol surrogate recognized by the bacterial *ton* B-dependent ion transport pathway. Uptake of SQ 83,360 by this mechanism increases the concentration of drug in the periplasmic space where the transpeptidases are located (43).

The synthesis of SQ 83,360 is shown in Figure 8. The heterocyclic terminus (**60**) [112333-97-8] of the *N*-1 substituent was constructed in five steps from kojic acid [501-30-4] (**59**). After persilylation, the reaction of intermediate (**60**) and the chlorosulfonyl isocyanate adduct of Z-protected 3-amino-2-azetidinone to afford monocarbam [112333-98-9] (**61**) occurred. Hydrogenolysis of the Z-group formed (**62**) [12335-07-6], followed by coupling and deprotection of the side chain in (**63**) [112334-01-7] to yield SQ 83,360 [108319-07-9] **(58)** in the acid form. A ready supply of Z-azetidinone [80082-81-1], $C_{11}H_{12}N_2O_3$, used in the synthesis of SQ 83,360, was obtained starting with the mesylate of Z-protected serine amide (17). The azetidinone was formed in 53–57% overall yield (44). SQ 83,360 **(58)** demonstrated exceptional activity *in vitro* as can be seen in Table 5. Against numerous clinical *Enterobacteriaceae* strains, SQ 83,360 was similar to aztreonam, yet against nonfermenting gram-negative bacilli, particularly *Ps. aeruginosa*, SQ 83,360 was significantly superior.

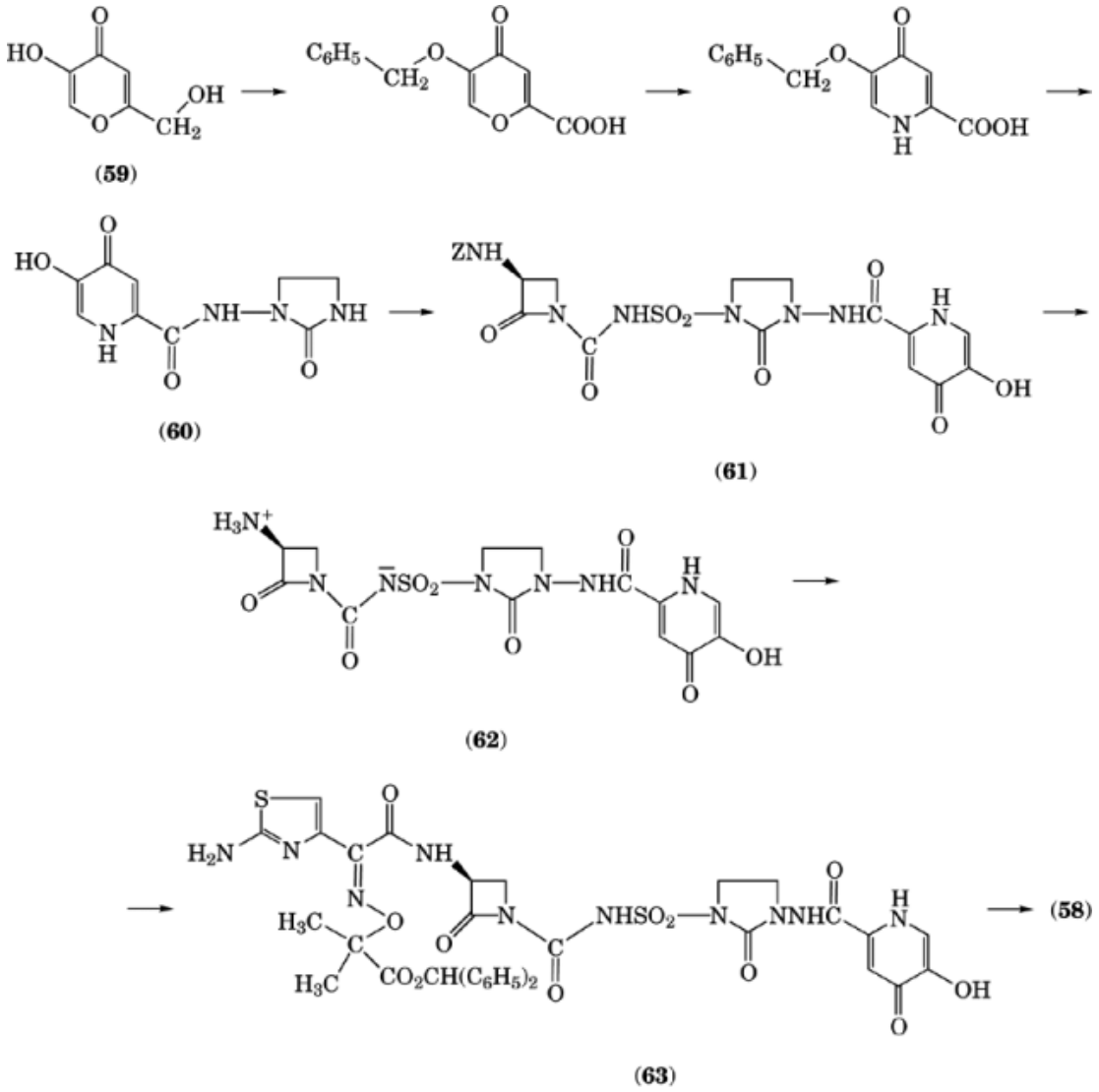


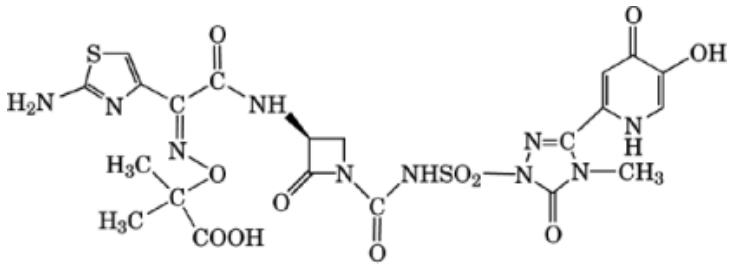
Fig. 8. Synthesis of SQ 83,360.

Since the discovery of SQ 83,360, compounds such as U-78,608 [123444-35-9] **(64)** having different linker groups between the hydroxypyridone group and the sulfonyl residue have been reported. U-78,608 and SQ 83,360 have similar *in vitro* and *in vivo* activity (45).

Table 5. Comparative *In Vitro* Activity of SQ 83,360 and Aztreonam

Organism[b]	Number of strains	MIC_{90}[a]	
		SQ 83,360	Azetreonam
Enterobacteriaceae	236	<0.03 – 1.0	<0.03 – 12.5
Ps. aeruginosa	35	0.05	15.3
Pseudomonas sp.	58	6.3	90.4
Acinetobacter sp.	45	14.7	46.0

[a] Minimum inhibitory concentration for 90% of the strains used. μg/mL
[b] All are nonfermentors except the *Enterobacteriaceae*.

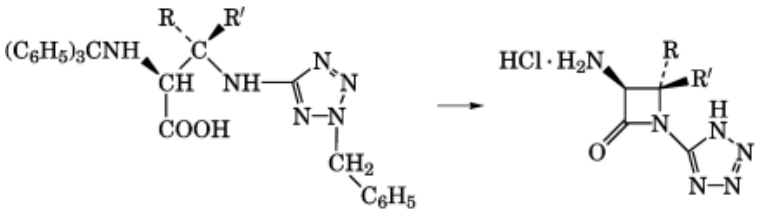


(**64**)

1.3.6. Tetrazole-Activated Monobactams

The tetrazole moiety is one of the closest isosteres to a carboxylic acid, having similar electron-withdrawing capability and acidity. The syntheses of homochiral tetrazole derivatives (**65**) from serine and threonine have been reported (46). As shown in Figure 9, cyclization of amides (**66**, R = H) [90208-25-6] and (**66**, R = CH_3) [90181-42-3] was effected using an intramolecular Mitsunobu reaction. Deprotection afforded the amine salts (**67**, R = H) [90181-47-8] and (**67**, R = CH_3) [90181-49-0], which were then coupled to afford the side-chain analogues (**65**, R′ = CH_3 or $C(CH_3)_2COOH$). Using similar methodology, other C-4 substituted monobactams were prepared whereby the configuration of R in structure (**65**) was either cis or trans and R = CH_2F, $COOCH_3$, CH_2OH, or CH_2OCONH_2 (47).

Alternatively, various 4-substituted derivatives have been prepared via synthesis of amino acid **(68)** by reaction of the anion formed from protected glycine and an appropriately substituted Schiff base.



(**68**) (**69**)

Cyclization of **(68)** and deprotection then yield the monobactam nucleus **(69)** which may be coupled with various C-3 side chains (48). Direct alkylation of the *N*-unsubstituted azetidinone **(70)** using fluorotetrazole [93607-94-4], $C_8H_7FN_4$, also produced **(69)** after deprotection of **(71)** (49).

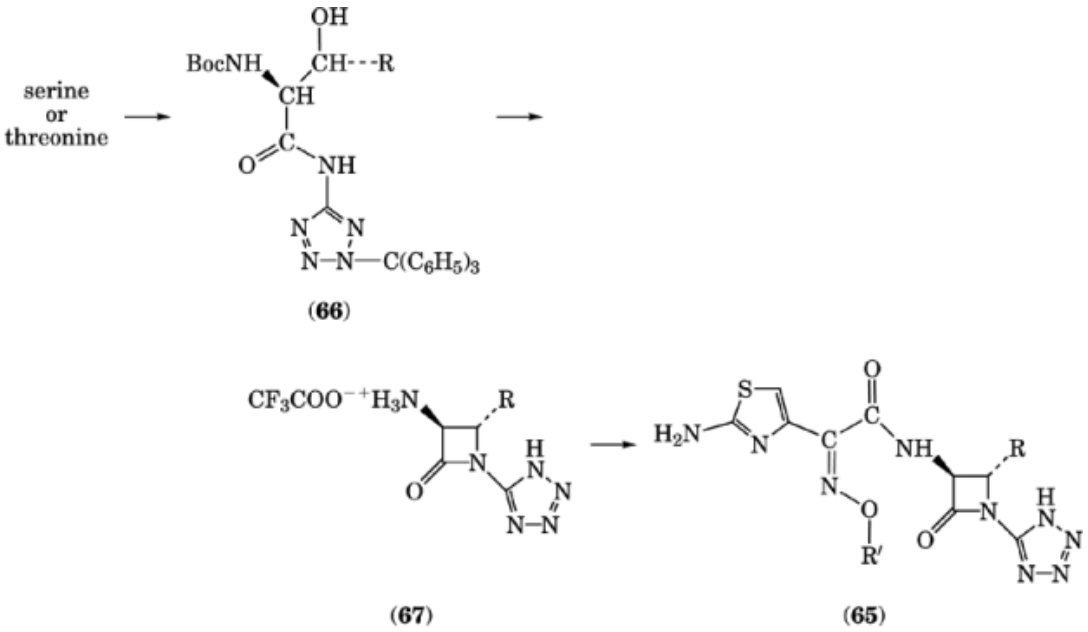
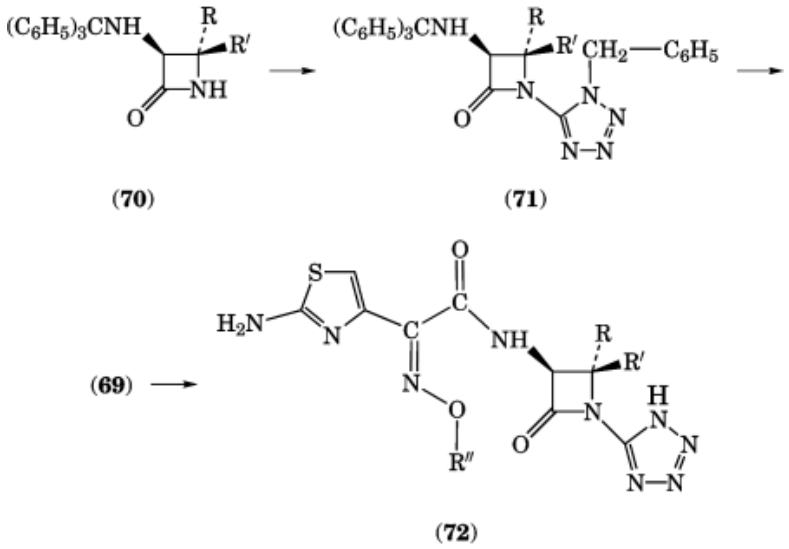
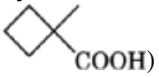


Fig. 9. Synthesis of *N*-(tetrazolyl-5-yl) azetidinones where Boc is *t*-butoxycarbonyl; $_{R=}$ H or CH**3**; and $_{R'=}$ CH_3 or $C(CH_3)_2COOH$.



Structure **(72)** exemplifies the C-4 derivatives prepared by these routes. From this class of monobactams, RU-44790 [110012-78-7] (**72**, R = H, R′ = CH_2F, R″ = ) was identified as having improved β-lactamase stability and activity vs gram-negative bacilli similar to that of aztreonam (50).

1.4. Economic Aspects

Two monobactams were in clinical use as of 1990. Aztreonam (**17**) manufactured by Bristol-Myers Squibb, has the worldwide trademark of Azactam. Aztreonam has received regulatory approval for use in humans in 67 countries and has pending filings in seven additional countries. The global experience with aztreonam and its clinical acceptance signifies that the monobactams are recognized as an important new class of antibacterial agents. Carumonam, manufactured by Takeda, received approval for human usage in 1988 in Japan. Carumonam **(19)** has the trademark, Amasulin.

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