

STRUCTURE-ACTIVITY RELATIONSHIPS OF 6-(HETEROCYCLYL)-
METHYLENE PENAM SULFONES; A NEW CLASS
OF β -LACTAMASE INHIBITORS

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6-(Heterocyclyl)methylene penam sulfones (**1**) are effective β -lactamase inhibitors and potent ampicillin and cefazolin potentiators against both Gram-positive and Gram-negative β -lactamase producing bacteria. Several of these analogs having a π -deficient 2-heteroaryl substituent attached to the C₆-methylene position showed better inhibitory activity than clavulanic acid, Ro 15-1903, 6 β -bromopenicillanic acid, and sulbactam against a variety of β -lactamases. The compounds were devoid of any antibacterial activity, but in combination with ampicillin or cefazolin, exhibited synergistic activity at least equal to clavulanic acid, Ro 15-1903, 6 β -bromopenicillanic acid or sulbactam against β -lactamase producing strains. Structure-activity relationships for a number of compounds are described. The structure-activity relationships can be rationalized by an enzyme inhibition mechanism which we have previously proposed on the basis of methanolysis of 6-(2-pyridyl)methylene penam sulfone (**1a**). Two synthetic routes to prepare compounds of structural type **1** *via* either a Wittig reaction or an aldol condensation are reported. β -Lactamase inhibition and MIC data are presented.

Many bacteria become resistant to β -lactam antibiotics by producing β -lactamases. Research directed at overcoming this problem of resistance has resulted in two clinically useful β -lactamase inhibitors; clavulanic acid (**2**)¹⁾ and sulbactam (**3**)²⁾. In our efforts to further improve sulbactam activity, we were led to the preparation of structures of type **1** wherein a heterocyclylmethylene group is attached to C(6) of the penam sulfone nucleus. We now report that several of these compounds are very potent β -lactamase inhibitors and synergize with ampicillin or cefazolin against resistant strains. The structure-activity relationships in this series of compounds indicate that a π -deficient 2-heteroaryl group in the C(6) side chain and the sulfone nucleus are important for potent activity.

Chemistry

The syntheses of 6-(heterocyclyl)methylene penam sulfones (**1**) were accomplished *via* either a Wittig reaction or an aldol condensation.

Route A

The first synthetic route *via* a Wittig reaction starting from 6 α -hydroxypenicillanic acid³⁾ is outlined in Scheme 1. Swern oxidation⁴⁾ of allyl 6 α -hydroxypenicillanate (**5**) gave crude allyl 6-oxopenicillanate (**6**)⁵⁾ which was used directly in the next step without further purification. Reaction of **6** with

a freshly prepared Wittig reagent **10** at -78°C gave the *Z* olefin **7** predominantly. However, when basic Wittig reagents (e.g. $\text{R}=\text{CH}_2\text{N}(\text{CH}_3)_2$) were employed, the reaction failed and resulted in the loss of the β -lactam ring, even at -78°C . Oxidation of the olefin **7** with 2 eq of *m*-chloroperbenzoic acid, followed by deallylation using the method of JEFFREY and McCOMBIE⁹⁾ gave the final 6-(substituted)methylene penam sulfones (**1**).

Compounds **1o** ($\text{R}=\text{CH}_2\text{OH}$) and **1p** ($\text{R}=\text{CH}=\text{NOCH}_3$) were prepared starting from 6-formylmethylene penam sulfone (**9m**) and their syntheses are shown in Scheme 2.

Deblocking of diallylpenam sulfone (**9j** or **9k**) with 2 eq of potassium 2-ethylhexanoate in the presence of a catalytic amount of $\text{Pd}(\text{PPh}_3)_4$ and PPh_3 gave a mixture of hydroxypyridyl (**1q** or **1r**) and allyloxypyridyl (**1j** or **1k**) analogs as shown in Scheme 3. The mixture of two components was separated by C_{18} medium pressure column chromatography.

Compound **1s** ($\text{R}=\text{2-pyridyl } N\text{-oxide}$) was prepared by oxidation of **9a** with *m*-chloroperbenzoic acid, followed by deblocking.

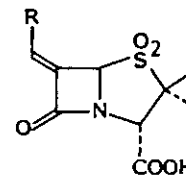
Route B

The aldol condensation route is outlined in Scheme 4. Treatment of allyl 1,1-dioxo-6 α -bromopenicillanate (**13**) with 1 eq of methylmagnesium bromide at -78°C , followed by quenching with an aldehyde at -78°C afforded **11** as a roughly 2:1 mixture of (6 α ,8*R*) and (6 α ,8*S*) diastereomers, respectively. Acylation of the mixture of isomers **11**, followed by elimination gave an olefin sulfone **9**. We found that acetylation of **11** with acetic anhydride in the presence of 1 eq of pyridine, followed by 1,5-diazabicyclo[4.3.0]non-5-ene (DBN) elimination gave **9** in the best yields. It is noteworthy that elimination of a roughly 2:1 mixture of 8*R* and 8*S* isomers of π -deficient 2-heteroaryl analogs (e.g. **12c**, **12d**, **12a'**, **12c'** and **12d'**) gave surprisingly the *Z* isomer **9** exclusively. Other compounds of structure type **12** gave **9** as a mixture of *Z* and *E* isomers. Deblocking of the allyl group of **9** gave the final potassium salt **1**.

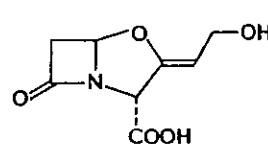
Results and Discussion

In Vitro Activity

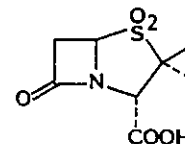
The 6-(heterocyclyl)methylene penam sulfones in combination with ampicillin or cefazolin, produce a synergistic effect against both penicillinase and cephalosporinase producing organisms (Table 1). The best compounds were equivalent to or better than clavulanic acid, 6 β -bromopenicillanic acid (**4**)⁷⁾, Ro 15-1903 (**8n**)^{8,9)}, and sulbactam in synergizing with ampicillin against penicillinase producing strains of *Staphylococcus aureus* 01A400, *Klebsiella pneumoniae* 53A079, and *Haemophilus influenzae*



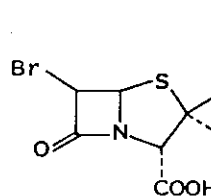
CP-65,372 (**1a**) $\text{R} = \text{2-Pyridyl}$
CP-68,146 (**1a'**) $\text{R} = \text{2-Thiazolyl}$



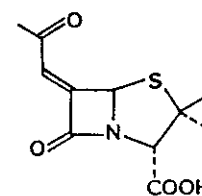
Clavulanic acid (**2**)



Sulbactam (**3**)

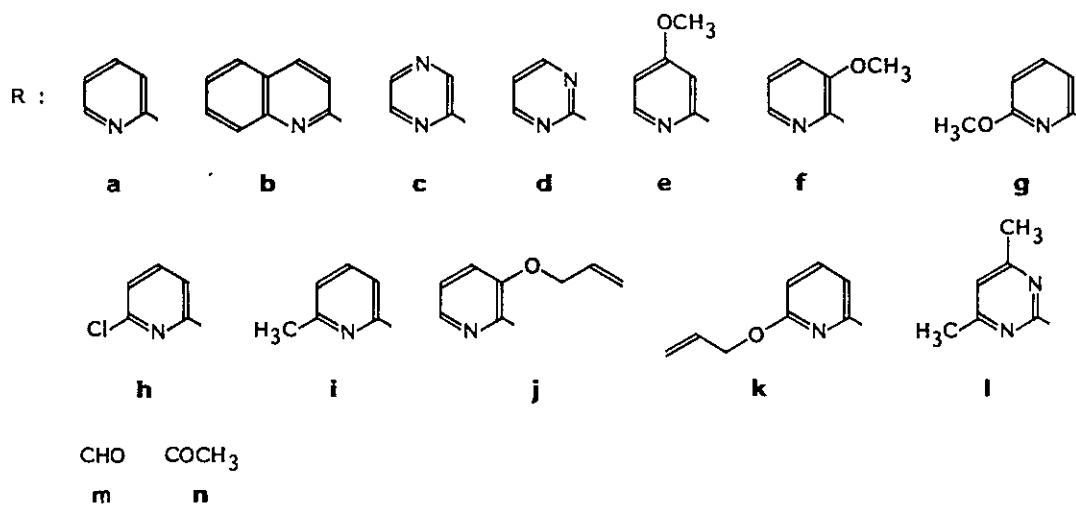
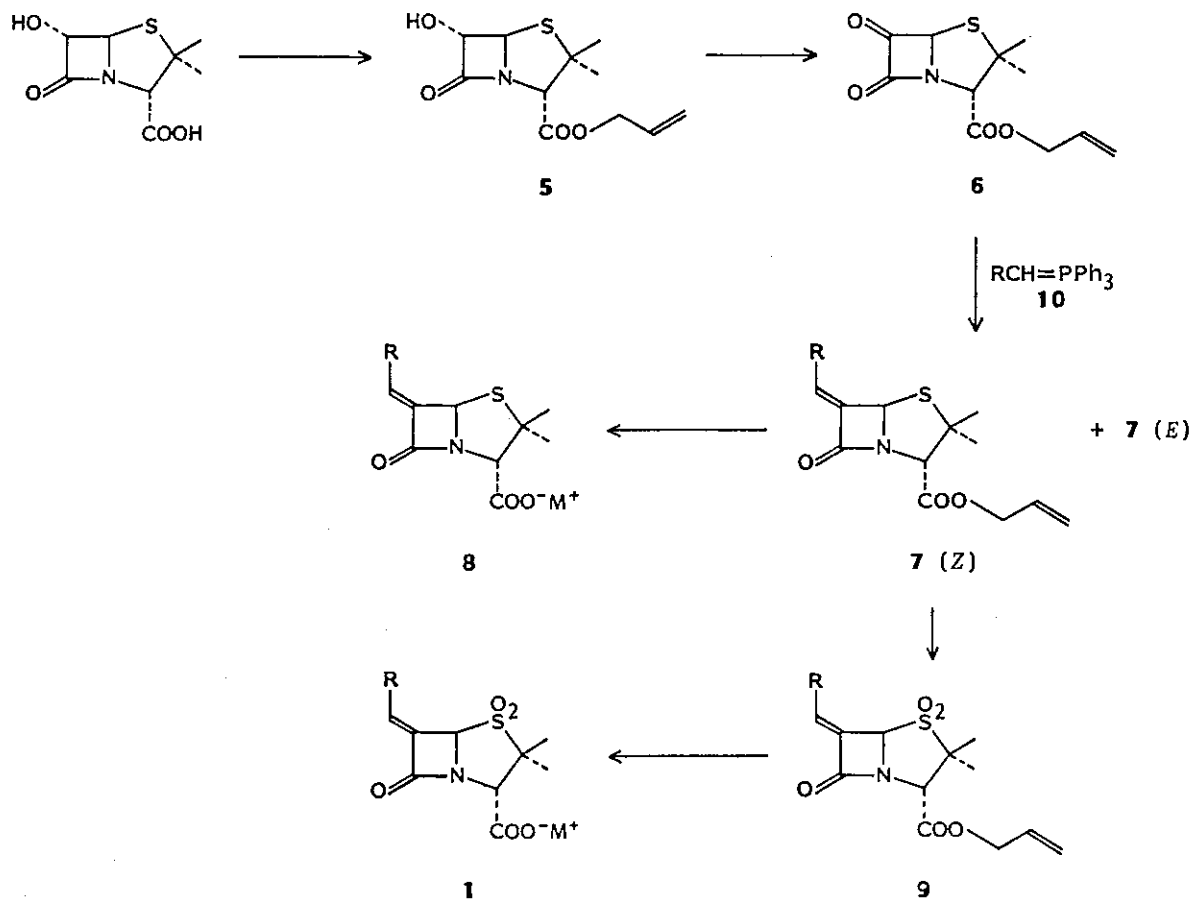


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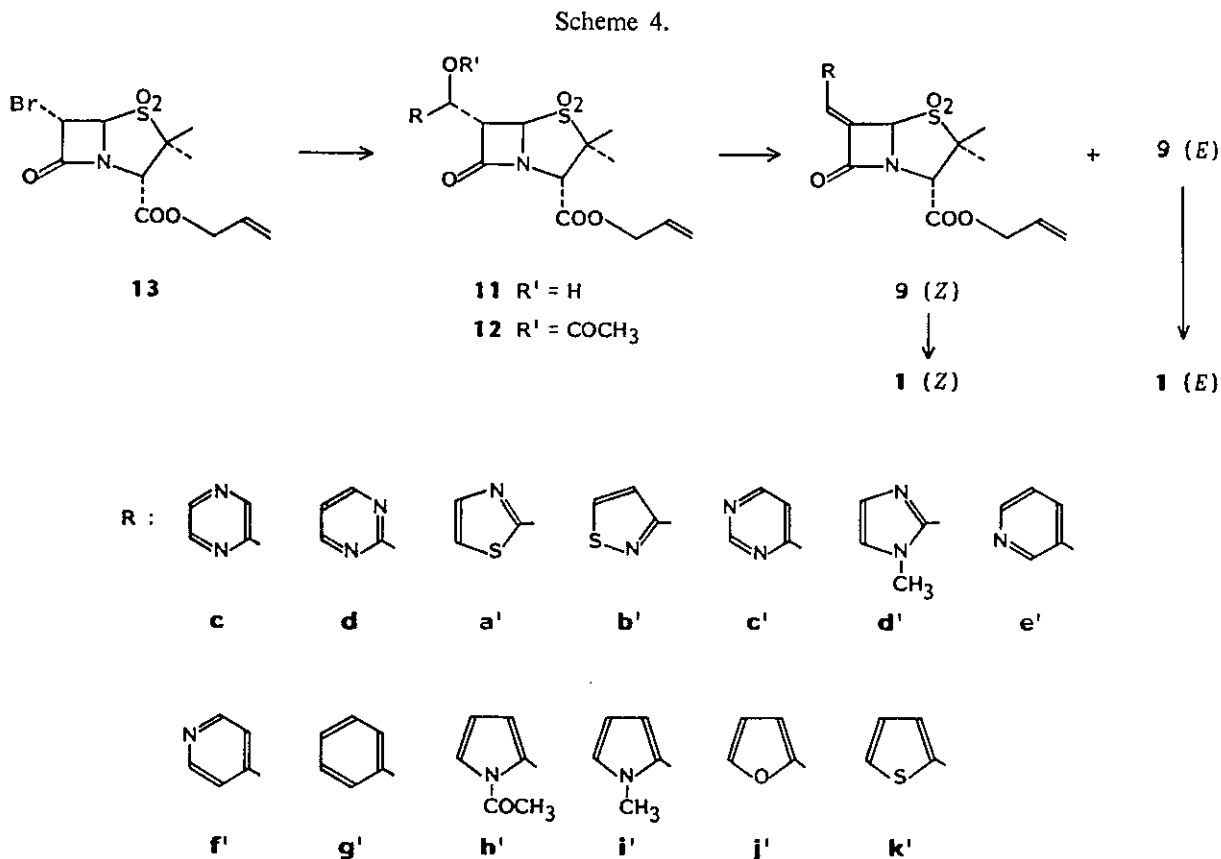
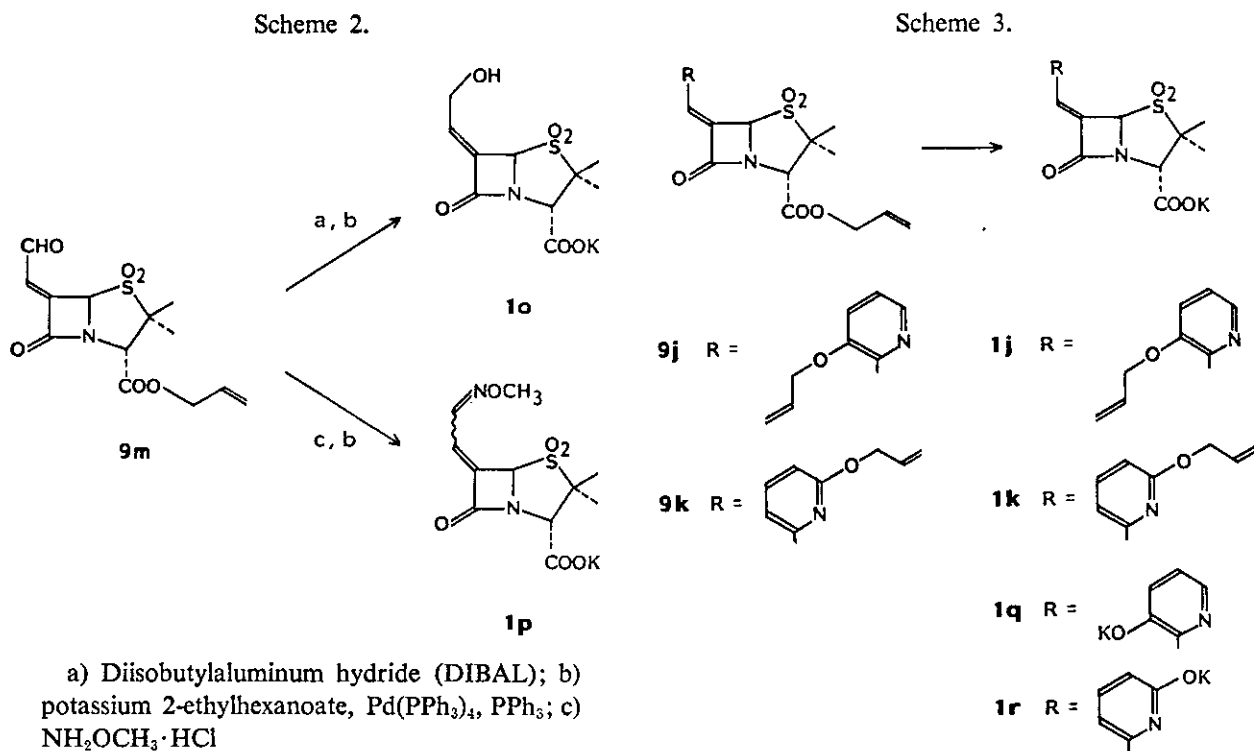


Ro 15-1903 (**8n**)

Scheme 1.



54A042 (see compound **1a**, **1q** and **1a'** in Table 1). When combined with amoxicillin and tested against recent amoxicillin-resistant clinical isolates of *S. aureus* (25 strains) and *H. influenzae* (30 strains), compound **1a'** was of equal potency to clavulanic acid against *H. influenzae* strains and 8 times better against *S. aureus* strains (Table 2). Against inducible cephalosporinase-producing organisms, several compounds combined with ampicillin were as potent as sulbactam/ ampicillin and 6 β -bromopenicillanic acid (strains 63A095, 67B009, 97A001 and 52A104 in Table 1), and much more potent than clavulanic acid and Ro 15-1903. In addition, several compounds (**1a**, **1f** and **1b'**) produced a synergistic effect



against *Pseudomonas aeruginosa* 52A104 (Table 1).

Sulbactam and the most potent of the new inhibitors in series 1 also demonstrated a synergistic effect in combination with cefazolin (compound 1a and 1a' presented in Table 3 as examples) against both penicillinase (51A129) and cephalosporinase producing organisms (67B009, 63A095 and 97A001).

Table 1. MIC^a data of a 1:1 combination of an inhibitor with ampicillin.


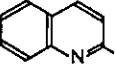


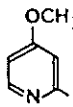
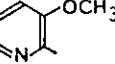
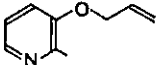
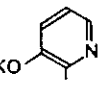






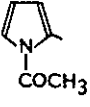



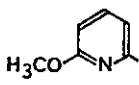
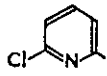
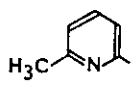
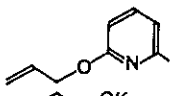
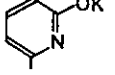
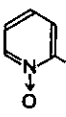
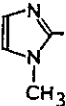

Compound number	R	Isomer	MIC ($\mu\text{g/ml}$)							
			<i>S. a.</i> 01A400	<i>E. c.</i> 51A129	<i>H. inf.</i> 54A042	<i>K. pne.</i> 53A079	<i>S. mar.</i> 63A095	<i>E. clo.</i> 67B009	<i>M. mor.</i> 97A001	<i>P. aer.</i> 52A104
Ampicillin alone			50	>200	>200	100	>200	200	>200	>200
1:1 Ampicillin plus inhibitor										
2	(Clavulanic acid)		0.39	12.5	0.39	12.5	50	100	>100	>100
3	(Sulbactam)		0.78~1.56	100	0.78	6.25	12.5	50	6.25	100
4	(6 β -Bromopenicillanic acid)		0.39	25	0.78	3.12	3.12	50		>100
8n	(Ro 15-1903)		0.78	50	0.78	12.5	25	>100	100	>100
1a		Z	0.39	25	<0.2	6.25	12.5	25	3.12	100
1b		Z	0.78	>100		12.5	25	50	25	>100
1c		Z	0.39	25		3.12	6.25	100	6.25	>100
1d		Z	0.39	50		6.25	6.25	50	3.12	>100
1e		Z	0.78	25		6.25	12.5	50	6.25	>100
1f		Z	1.56	100		12.5	25	25	3.12	100
1j		Z	1.56	50		6.25	12.5	50	6.25	>100
1q		Z	0.39	25		6.25	25	50	12.5	>100
1a'		Z	0.39	50	0.39	3.12	25	100	3.12	>100
1b'		Z	0.78	100	0.39	12.5	25	>100	12.5	100
		E	0.78	100	0.39	12.5	25	>100	12.5	100
1c'		Z	1.56	100		12.5	50	>100	25	>100
1e'		Z	50	>100		50	50	>100	100	>100
1f'		Z:E=	12.5	>100		50	50	100	50	>100
1g'		Z	25	>100		50	50	>100	25	>100
1o	CH ₂ OH	Z	6.25	100		25	25	50	12.5	>100
1p	CH=NOCH ₃	(Z,E)+ (Z,Z)	25	>100		50	>100	>100	50	>100
1h'		Z	25	>100		50	50	>100	50	>100

Table 1. (Continued)

Compound number	R	Isomer	MIC ($\mu\text{g/ml}$)							
			<i>S. a.</i> 01A400	<i>E. c.</i> 51A129	<i>H. inf.</i> 54A042	<i>K. pne.</i> 53A079	<i>S. mar.</i> 63A095	<i>E. clo.</i> 67B009	<i>M. mor.</i> 97A001	<i>P. aer.</i> 52A104
1i'		Z	12.5	>100		50	50	100	50	>100
1j'		Z:E= 4:1	6.25	>100		25	25	>100	25	>100
1k'		Z	12.5	>100		50	50	100	25	>100
1g		Z	12.5	>100		25	100	50	25	>100
1h		Z	50	>100		50	>100	>100	25	>100
1i		Z	12.5	>100		25	50	100	25	>100
1k		Z	12.5	>100		25	100	>100	50	>100
1r		Z	6.25	>100		25	50	>100	>100	>100
1s		Z	12.5	>100		50	>100	>100	100	>100
1d'		Z	3.12	100		12.5	25	50	6.25	>100
8a		Z	3.12	>100		100	>100	>100	100	>100

* Concentration of both components; i.e. MIC=0.39 $\mu\text{g/ml}$ inhibitor plus 0.39 $\mu\text{g/ml}$ ampicillin.

Organisms abbreviation: *S. a.*; *Staphylococcus aureus*, *E. c.*; *Escherichia coli*, *H. inf.*; *Haemophilus influenzae*, *K. pne.*; *Klebsiella pneumoniae*, *S. mar.*; *Serratia marcescens*, *E. clo.*; *Enterobacter cloacae*, *M. mor.*; *Morganella morganii*, *P. aer.*; *Pseudomonas aeruginosa*.

Table 2. MIC data of inhibitors with amoxicillin.

Compound	Isomer	MIC ₉₀ ($\mu\text{g/ml}$) ^a	
		<i>Haemophilus influenzae</i> (30 strains)	<i>Staphylococcus aureus</i> (25 strains)
Amoxicillin alone		>16	>16
Inhibitor		Inhibitor+amox	Inhibitor+amox
CP-68,146 (1a')		0.25+4	0.031+4
CP-71,126 (1b')	Z	0.125+4	0.5+4
Clavulanic acid		0.25+4	0.25+4

* Combination of inhibitor plus 4 $\mu\text{g/ml}$ amoxicillin (amox); 4 $\mu\text{g/ml}$ is the approximate blood level of amoxicillin from 250 mg dose in humans.

Table 3. MIC data of a 1:1 combination of an inhibitor with cefazolin.

	MIC ($\mu\text{g/ml}$)						
	<i>S. a.</i> 01A400	<i>E. c.</i> 51A129	<i>K. pne.</i> 53A079	<i>E. clo.</i> 67B009	<i>S. mar.</i> 63A095	<i>M. mor.</i> 97A001	<i>P. aer.</i> 52A104
Compound alone							
CP-65,372 (1a)	>200	>200	>200	>200	>200	>200	>200
CP-68,146 (1a')	>200	>200	>200	>200	>200	>200	>200
Sulbactam (3)	200	100	25	50	100	100	>200
Clavulanate (2)	25	100	100	>200	200	200	>200
Ro 15-1903 (8n)	>200	>200	>200	>200	>200	>200	>200
Cefazolin	0.78	50	1.56	>200	>200	>200	>200
1:1 Combination of compound plus cefazolin (cef)							
1a+cef	0.39	3.12	1.56	50	25	25	>100
1a'+cef	0.39	12.5	0.78	>100	50	12.5	>100
Sulbactam+cef	0.39	12.5	1.56	50	25	12.5	>100
Clavulanate+cef	0.39	3.12	1.56	100	200	100	>100
Ro 15-1903+cef	0.78	6.25	1.56	>100	>100	>100	>100

Organisms abbreviation: See Table 1.

However, none of these new inhibitors, clavulanic acid, Ro 15-1903, or sulbactam synergized with cefazolin against *P. aeruginosa* (Table 3). Ro 15-1903 in combination with cefazolin demonstrated a synergistic effect only against *Escherichia coli* 51A129, whereas clavulanic acid in combination with cefazolin demonstrated a synergistic effect against *E. coli* 51A129 and *Enterobacter cloacae* 67B009 (Table 3).

β -Lactamase Inhibitory Activity

The inhibitors were tested against cell free β -lactamase preparations, including a penicillinase from *S. aureus*, a Type III plasmid-mediated broad spectrum penicillinase from *E. coli*, and a Type Ia inducible chromosomal cephalosporinase from *E. cloacae*. Active inhibitors were further tested against a known TEM-1 plasmid mediated penicillinase and a Type Id cephalosporinase from *P. aeruginosa*. Ro 15-1903 inhibited the penicillinases better than clavulanic acid and sulbactam (Table 4). Many of the experimental compounds were equivalent to Ro 15-1903 in inhibiting penicillinases (1a, 1e, 1q and 1a'). In general, the 6-(heterocyclyl)methylene penam sulfones are also potent cephalosporinase inhibitors. Compounds 1a, 1b, 1f and 1j produced ~90% or greater inhibition of the *E. cloacae* and *P. aeruginosa* enzymes compared to less than 10% inhibition for sulbactam (Table 4).

For a more quantitative determination of β -lactamase inhibitory activity, IC_{50} 's of several of the more potent β -lactamase inhibitors were determined with a TEM-1 β -lactamase (Table 5). IC_{50} values were determined with and without preincubation of the inhibitor with the TEM-1 β -lactamase, and the results indicate that all inhibitors tested demonstrate an irreversible mode of inhibition. The relative potency of the β -lactamase inhibitors are: CP-68,146 (1a') > Ro 15-1903 > 6 β -bromopenicillanic acid > clavulanic acid > sulbactam. There is not a great difference in the inhibitory activity between the latter two compounds (Table 5). These results are consistent with the *in vitro* synergy potency data in Tables 1 and 2.

Structure-activity Relationships

We have previously described¹⁰⁾ a hypothesis for the enzyme inhibition mechanism of 1a on the

Table 4. β -Lactamase inhibitory activity.


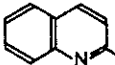



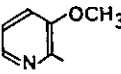
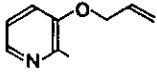
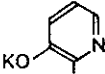










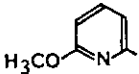
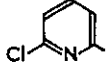
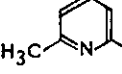
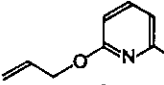
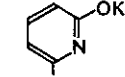
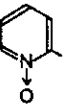
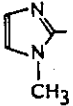


Compound number	R	Isomer	Enzyme inhibition (%) ^a				
			<i>S. a.</i> 01A400	<i>E. c.</i> 51A129	TEM-1 51A560	<i>E. clo.</i> 67B009	<i>P. aer.</i> 52A104
2 (Clavulanate)			85	81			
3 (Sulbactam)			46	40	42	9	4
8n (Ro 15-1903)			97	100		16	
1a		Z	90	100	100	89	91
1b		Z	64	95	93	93	94
1c		Z	91	100	100	18	58
1d		Z	92	100	97	63	
1e		Z	100	100		77	
1f		Z	48	100	92	96	
1j		Z	84	98	97	94	93
1q		Z	95	100		64	
1a'		Z	97	96	99	28	99
1b'		Z	92	88			
1c'		Z	87	96	83	19	60
1e'		Z	13	5			
1f'		Z:E=7:3	1	24			
1g'		Z	0	28			
1o	CH ₂ OH	Z	0	29			
1p	CH=NOCH ₃	(Z,Z)+(Z,E)	2	23		0	
1h'		Z	24	14		54	
1i'		Z	18	14			
1j'		Z:E=4:1	11	41		8	
1k'		Z	3	37		19	

Table 4. (Continued)

Compound number	R	Isomer	Enzyme inhibition (%) ^a				
			<i>S. a.</i> 01A400	<i>E. c.</i> 51A129	TEM-1 51A560	<i>E. clo.</i> 67B009	<i>P. aer.</i> 52A104
1g		Z	26	28		73	
1h		Z	1	7			
1i		Z	16	62		95	
1k		Z	11	26		47	
1r		Z	22	93		13	
1s		Z	0	18		27	
1d'		Z	39	85		77	
1b'		E	45	20			
8a		Z	26	0			

^a The percent inhibition at the enzyme level was obtained at the following indicated concentrations (μM) for inhibitor [I] and substrate [S]:

Staphylococcus aureus (*S. a.*) 01A400; [I]/[ampicillin]=8/32.

Escherichia coli (*E. c.*) 51A129; [I]/[ampicillin]=1/32.

Plasmid TEM-1 51A560; [I]/[ampicillin]=1/32.

Enterobacter cloacae (*E. clo.*) 67B009; [I]/[benzylpenicillin]=8/32.

Pseudomonas aeruginosa (*P. aer.*) 52A104; [I]/[benzylpenicillin]=8/32.

Table 5. Determination of IC_{50} of β -lactamase inhibitors.

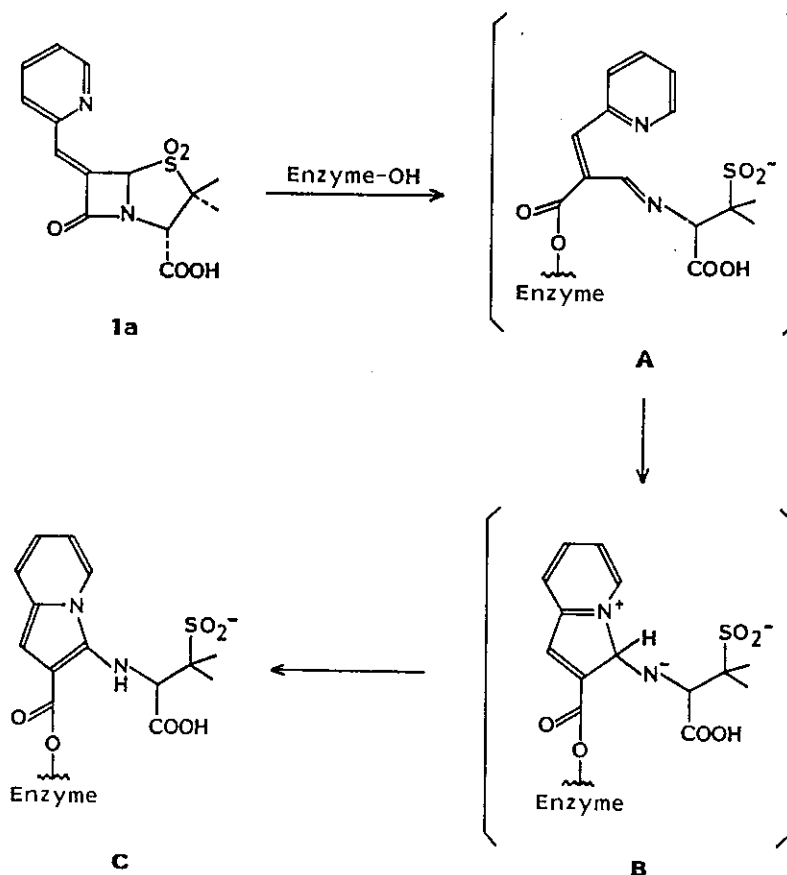
Compound	IC_{50} (μM) TEM-1 β -lactamase	
	No preincubation ^a	15 minutes preincubation ^b
Clavulanic acid	9.8	0.19
Sulbactam	3.0	0.31
6 β -Bromopenicillanic acid	8.6	0.047
CP-68,146 (1a')	0.09	0.0027
Ro 15-1903	0.32	0.016

^a β -Lactamase inhibitor added to substrate and enzyme at zero time.

^b β -Lactamase inhibitor preincubated 15 minutes with enzyme prior to addition of substrate.

basis of its chemical reaction with sodium methoxide (Scheme 5). We proposed that after bimolecular interaction between a β -lactamase and compound 1a, an aromatic acyl-enzyme ester C is obtained. This conjugated acyl-enzyme ester having an electron-donating amino group at the *ortho* position is stable and the acyl-enzyme ester is expected to be resistant to hydrolysis to regain active enzyme. We

Scheme 5.



proposed that **C** is a stable aromatic acyl-enzyme intermediate which is probably responsible for the potent β -lactamase inhibitory activity of **1a**; presumably the other compounds in the series can form a similar intermediate. Many compounds in series **1** were prepared to test our hypothetical mechanism of enzyme inactivation. As a result, the structure-activity relationships can be rationalized by the proposed mechanism.

The following structural features are considered to be important for a compound to exhibit inhibitory activity: A) A 2-heteroaryl group attached to the C(6)-methylene site; B) a π -deficient heteroaryl group; C) no steric congestion around the N lone pair; D) a penam sulfone nucleus; E) the *Z* isomer of the 6-(2-heteroaryl)methylene penam sulfone. These features of the observed structure-activity relationships are rationalized by the proposed mechanism of enzyme inactivation as follows:

A) A 2-Heteroaryl Group Attached to the C(6)-Methylene Site

All the 2-heteroaryl analogs shown in Table 4 are capable of forming the acyl-enzyme intermediate **C**; all analogs show potent inhibitory activity. 3- or 4-Pyridyl analogs (**1e'** or **1f'**) which cannot give an aromatic acyl-enzyme intermediate **C** possess only weak activity compared to that of the corresponding 2-pyridyl analog (**1a**). Compound **1g'**, **1o** and **1p** which are not 2-heteroaryl type analogs all have poor activity (Table 4).

B) π -Deficient Heteroaryl Ring

All the compounds in Table 4 which possess a π -deficient 2-heteroaryl side chain at the C(6)-methylene site show good activity. On the contrary, π -excessive 2-heteroaryl analogs (*e.g.* **1h'** ~ **1k'**) which should not rearrange to an enzymatically inactive form like **C** show moderate to weak activity (Table 4).

C) Steric Effect

In general, the biological activity decreases in the presence of a sterically hindering substituent on the pyridine nucleus. A substituent next to the N of the pyridyl ring (e.g. **1g**, **1h**, **1i**, **1k** and **1r**) produces especially severe steric hindrance, which prevents the N atom from approaching the imine group (see Scheme 5, A→C). Furthermore, the 2-pyridyl *N*-oxide analog (**1s**), where the N lone pair is blocked, shows poor inhibitory activity.

D) Sulfone Nucleus

The proposed mechanism of enzyme inactivation requires the sulfone to act as a leaving group in order to generate intermediate C. This requirement is illustrated by the fact that replacing the sulfone in **1a** with a sulfide, a poorer leaving group, afforded compound **8a** which is only weakly active.

E) Geometry

According to the proposed mechanism, the *Z* isomer can form the enzymatically inactive form C more rapidly than the corresponding *E* isomer due to the ease of cyclization. Indeed, the *Z* isomer possesses better inhibitory activity than the corresponding *E* isomer (see the inhibitory activity of **1b'** (*Z*) and relative to that of **1b'** (*E*) shown in Table 4), although both isomers showed a similar synergistic activity. It is possible that the synergistic activity of the *E* isomer may be due to its isomerization during overnight incubation.

Conclusion

In conclusion, we have demonstrated that penam sulfone analogs bearing a π -deficient 2-heteroarylmethylene group at the C(6) position possess potent β -lactamase inhibitory activity. The structure-activity relationships observed in the series **1** are consistent with our proposed mechanism for irreversible inhibition.

Experimental

IR spectra were recorded on a Perkin-Elmer model 283B spectrophotometer. NMR spectra were recorded on a Varian T-60 or Bruker 250 spectrometer using TMS as internal standard. Analytical HPLC was carried out with a Waters Associates Instrument using Waters μ Bondapak C18 column. X-Ray analyses were studied by Professor J. BORDNER at North Carolina State University.

Chemicals

Sulbactam and 6 β -bromopenicillanic acid¹¹⁾ were prepared in the Pfizer laboratories. Lithium clavulanate was kindly supplied by Beecham Pharmaceuticals. The sodium salt of Ro 15-1903 was prepared in our laboratory (see **8n** in the experimental section). Other antibiotics were obtained commercially.

In Vitro Assays

Cell free extracts of β -lactamase producing organisms were prepared as described previously²⁾. *E. coli* TEM-1 (Type IIIa) β -lactamase was purchased from Calbiochem-Behring, San Diego, CA. For the percent inhibitory data in Table 4, the rate of hydrolysis was determined by the micro-iodometric assay of ZIMMERMANN and ROSSELET¹²⁾ which is a modification of the NOVICK¹³⁾ micro-iodometric assay; incubations were at 37°C for 10 minutes¹⁴⁾ after a 10 minutes preincubation of inhibitor with the β -lactamase enzyme. For the determination of IC₅₀ values, the initial rate of hydrolysis was determined by continuous monitoring of absorbance for 10 minutes in a Perkin-Elmer Lambda-3 Spectrophotometer (Norwalk, CT) with the optical chamber heated to 37°C, and wavelength at 623 nm. Experiments were done with and without a 15-minute preincubation of inhibitor with the TEM-1 β -lactamase.

Minimal inhibitor concentrations (MICs) were determined on brain heart infusion agar (Scott Laboratory Inc., Fiskeville, Rhode Island) as the basal medium²³ by the method of ERICSSON and SHERRIS¹³ using the multiple inoculator described by STEERS *et al.*¹⁶. Synergy was defined as occurring when the MIC of each component in the combination was one-fourth or less than its MIC as a single agent.

Allyl 6 α -Hydroxypenicillanate (5)

A solution of 85 g (0.39 mol) of 6 α -hydroxypenicillanic acid²³ in 300 ml of DMF was treated with 34 ml (0.39 mol) of allyl bromide and 57 ml (0.41 mol) of triethylamine and the mixture was stirred at room temperature for 15 hours. The mixture was quenched with water and extracted with ethyl ether. The combined ether layers were neutralized with saturated sodium bicarbonate solution, washed with water, dried (MgSO₄) and concentrated *in vacuo* to afford 43 g (43%) of crude product. The crude material was purified by silica gel column chromatography (chloroform - EtOAc, 9:1) to yield 22.75 g (23%) of **5**. ¹H NMR (60 MHz, CDCl₃) δ 1.42 (3H, s), 1.60 (3H, s), 4.45 (1H, s), 4.5~5.0 (3H, m), 5.2~6.2 (4H, m).

Allyl 6-Oxopenicillanate (6)

A mixture of 2.84 ml (0.04 mol) of DMSO, 3.67 ml (0.026 mol) of trifluoroacetic anhydride and 50 ml of methylene chloride was stirred at -78°C for 10 minutes. A solution of 5.14 g (0.02 mol) of allyl 6 α -hydroxypenicillanate (**5**) in 10 ml of methylene chloride was added at -78°C, and the resulting mixture was stirred at that temperature for 40 minutes. Triethylamine (7.24 ml, 0.052 mol) was added and the mixture was gradually warmed to room temperature and quenched with water. After extracting with methylene chloride, the organic layer was washed with water, dried (MgSO₄) and the solvent was evaporated *in vacuo* to give **6** as a yellow oil, 5.1 g (100%). ¹H NMR (60 MHz, CDCl₃) δ 1.60 (6H, s), 4.75 (2H, m), 4.82 (1H, s), 5.1~6.3 (3H, m), 5.82 (1H, s).

Allyl 6-(Z)-(2-Pyridyl)methylenepenicillanate (7a)

A mixture of 2.64 g (6.8 mmol) of 2-picolyltriphenylphosphonium chloride and 0.265 g (6.8 mmol) of sodium amide in 6 ml of dry THF was stirred at room temperature for 30 minutes. The resulting brown suspension was cooled to -78°C and a solution of 1.8 g (7.0 mmol) of allyl 6-oxopenicillanate (**6**) in 4 ml of dry THF was added in one portion and the mixture was stirred at -78°C for 3 minutes. The reaction was quenched by addition of saturated ammonium chloride solution and extracted with EtOAc and the combined organic layers were washed with water, dried (MgSO₄) and concentrated *in vacuo* to give 3.3 g of red oil. The oil was purified by silica gel column chromatography to yield 1.35 g (60%) of **7a** as a yellow oil. ¹H NMR (250 MHz, CDCl₃) δ 1.50 (3H, s), 1.58 (3H, s), 4.57 (1H, s), 4.65 (2H, d), 5.15~6.15 (3H, m), 6.17 (1H, d), 6.87 (1H, d), 7.2~7.4 (2H, m), 7.6 (1H, m), 8.62 (1H, dd); ¹³C NMR (250 MHz, CDCl₃) δ 168.7, 167.5, 152.1, 149.9, 146.7, 136.3, 131.1, 125.9, 124.0, 123.2, 119.1, 70.5, 70.0, 65.8, 62.8, 33.0, 26.0.

Compounds (**7b**~**7n**) were obtained from **6** and the appropriate Wittig reagent **10** by procedure similar to that described for **7a**.

7b: Z isomer, 80% yield; ¹H NMR (CDCl₃) δ 1.5 (3H, s), 1.62 (3H, s), 4.6 (1H, s), 4.7 (2H, d), 5.1~6.2 (3H, m), 6.35 (1H, d), 7.0 (1H, d), 7.2~8.2 (6H, m).

7c: Z isomer, 50% yield; ¹H NMR (CDCl₃) δ 1.5 (3H, s), 1.6 (3H, s), 4.55 (1H, s), 4.67 (1H, s), 5.0~6.2 (3H, m), 6.15 (1H, d), 6.95 (1H, d), 8.4~8.8 (3H, m).

7d: Z isomer, 31% yield; ¹H NMR (CDCl₃) δ 1.5 (3H, s), 1.6 (3H, s), 4.6 (1H, s), 4.7 (2H, m), 5.1~6.3 (3H, m), 6.2 (1H, d), 7.0 (1H, d), 7.0~7.35 (1H, m), 8.8 (2H, d).

7e: Z isomer, 23% yield; ¹H NMR (CDCl₃) δ 1.5 (3H, s), 1.7 (3H, s), 3.8 (3H, s), 4.55 (1H, s), 4.7 (2H, m), 5.1~6.2 (3H, m), 6.2 (1H, s), 6.5~7.0 (3H, m), 8.5 (1H, d).

7f: Z isomer, 35% yield; ¹H NMR (CDCl₃) δ 1.5 (3H, s), 1.6 (3H, s), 3.85 (3H, s), 4.55 (1H, s), 4.7 (2H, d), 5.1~6.1 (3H, m), 6.2 (1H, d), 7.0~7.5 (3H, m), 8.2 (1H, t).

7g: Z isomer, 36% yield; ¹H NMR (CDCl₃) δ 1.5 (3H, s), 1.6 (3H, s), 3.95 (3H, s), 4.65 (3H, m), 5.1~6.2 (3H, m), 6.25 (1H, d), 6.6~7.0 (3H, m), 7.3~7.7 (1H, m).

7h: Z isomer, 20% yield; ¹H NMR (CDCl₃) δ 1.45 (3H, s), 1.55 (3H, s), 4.65 (2H, d), 5.2~6.1 (3H, m), 6.15 (1H, d), 6.75 (1H, d), 7.05~7.75 (3H, m).

7i: *Z* isomer, 27% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.5 (3H, s), 1.6 (3H, s), 2.6 (3H, s), 4.6 (1H, s), 4.65 (2H, m), 5.1~6.2 (3H, m), 6.2 (1H, d), 6.85 (1H, d), 7.0~7.7 (3H, m).

7j: *Z* isomer, 47% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.54 (3H, s), 1.62 (3H, s), 4.6~5.0 (5H, m), 5.0~6.2 (6H, m), 6.7~7.2 (3H, m), 7.4~7.8 (1H, m).

7k: *Z* isomer, 31% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.5 (3H, s), 1.6 (3H, s), 4.4~4.8 (5H, m), 5.1~6.3 (6H, m), 6.2 (1H, d), 7.15 (2H, d), 7.4 (1H, d), 8.15 (1H, t).

7l: *Z* isomer, 24% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.55 (3H, s), 1.65 (3H, s), 2.55 (6H, s), 4.65 (1H, s), 4.75 (2H, d), 5.1~6.1 (3H, m), 6.25 (1H, d), 6.9 (1H, s), 7.0 (d, 1H).

7m: The reaction was carried out with formylmethylene triphenylphosphorane in benzene and stirred at room temperature for 10 minutes. A mixture of *Z* and *E* isomers was obtained. Only the *Z* isomer was isolated in 45% yield. $^1\text{H NMR}$ (CDCl_3) δ 1.50 (3H, s), 1.60 (3H, s), 4.52 (1H, s), 4.62 (2H, d), 5.1~6.0 (3H, m), 5.90 (1H, d), 6.80 (1H, d), 9.73 (1H, d).

7n: The compound was prepared by the method as described for **7m**. 70% of the *Z* isomer and 4.3% of the *E* isomer were isolated. $^1\text{H NMR}$ of the *Z* isomer (CDCl_3) δ 1.52 (3H, s), 1.62 (3H, s), 2.40 (3H, s), 4.57 (1H, s), 4.7 (2H, m), 5.1~6.2 (3H, m), 6.0 (1H, d), 6.5 (1H, d); $^1\text{H NMR}$ of the *E* isomer (CDCl_3) δ 1.50 (3H, s), 1.60 (3H, s), 2.6 (3H, s), 4.57 (1H, s), 4.68 (2H, m), 5.1~6.2 (2H, m), 5.77 (1H, s), 6.0 (1H, s).

Sodium 6-(*Z*)-(2-Pyridyl)methylenepenicillanate (**8a**)

A mixture of 120 mg (0.38 mmol) of allyl 6-(*Z*)-(2-pyridyl)methylenepenicillanate (**7a**), 20 mg of tetrakis(triphenylphosphine)palladium and 20 mg of triphenylphosphine dissolved in 3 ml of EtOAc was treated with a solution of 0.5 M sodium 2-ethylhexanoate in EtOAc (0.76 ml, 0.38 mmol) was added at room temperature under nitrogen. The mixture was stirred for 2 hours and the precipitate was collected by filtration, washed with EtOAc and dried *in vacuo* to give 57 mg (48%) of **8a** as a yellow solid. $^1\text{H NMR}$ (D_2O) δ 1.55 (6H, s), 4.33 (1H, s), 6.17 (1H, d), 7.03 (1H, d), 7.18~8.07 (3H, m), 8.57 (1H, m); IR (KBr) cm^{-1} 3433, 1756 (β -lactam CO), 1605.

Compound **8n** (Ro 15-1903) was prepared by the method described for **8a** in 43% yield as its sodium salt. $^1\text{H NMR}$ (250 MHz, D_2O) δ 1.52 (3H, s), 1.57 (3H, s), 2.4 (3H, s), 4.4 (1H, s), 6.1 (1H, s), 6.7 (1H, s); IR (KBr) cm^{-1} 3413, 1762 (β -lactam CO), 1607.

Allyl 1,1-Dioxo-6-(*Z*)-(2-pyridyl)methylenepenicillanate (**9a**)

To a solution of 2.5 g (7.58 mmol) of allyl 6-(*Z*)-(2-pyridyl)methylenepenicillanate (**7a**) in 25 ml of methylene chloride was added 3.20 g (15.16 mmol) of 85% *m*-chloroperbenzoic acid and the mixture was stirred under nitrogen for 3 hours at room temperature. After quenching with saturated sodium thiosulfate solution and water, the organic layer was separated. The organic layer was washed with saturated sodium bicarbonate solution and with water, dried (MgSO_4) and the solvent was removed under reduced pressure to give 2.8 g yellow oil. The oil was purified by silica gel column chromatography (hexane - EtOAc, 7:3) to yield 1.7 g (62%) of **9a** as white crystals. $^1\text{H NMR}$ (CDCl_3) δ 1.48 (3H, s), 1.63 (3H, s), 4.45 (1H, s), 4.73 (2H, d), 5.1~6.2 (3H, m), 5.77 (1H, d), 7.27 (1H, d), 7.1~8.1 (3H, m), 8.6 (1H, m); $^{13}\text{C NMR}$ (CDCl_3) δ 168.1, 166.9, 150.3, 136.8, 132.8, 130.7, 126.0, 124.6, 119.9, 72.0, 66.6, 64.3, 63.2, 20.4, 18.5; IR (KBr) cm^{-1} 3437, 1783 (β -lactam CO), 1759, 1586.

Compounds (**9b**~**9m**) were also obtained from **7a**~**7m** by the method described for **9a**.

9b: 56% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.5 (3H, s), 1.6 (3H, s), 4.45 (1H, s), 5.1~6.1 (3H, m), 5.8 (1H, d), 7.3~8.5 (7H, m).

9c: 40% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.56 (3H, s), 1.7 (3H, s), 4.6 (1H, s), 4.85 (2H, m), 5.2~6.2 (3H, m), 5.8 (1H, d), 7.4 (1H, d), 8.5~9.0 (3H, m).

9d: 30% yield; yellow solid, $^1\text{H NMR}$ (CDCl_3) δ 1.45 (3H, s), 1.65 (3H, s), 4.5 (1H, s), 4.75 (2H, m), 5.2~6.3 (3H, m), 5.75 (1H, s), 7.1~7.5 (2H, m), 8.9 (2H, d).

9e: 99% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.5 (3H, s), 1.7 (3H, s), 3.9 (3H, s), 4.5 (1H, s), 4.75 (2H, m), 5.2~6.3 (3H, m), 5.7 (1H, d), 6.7~7.0 (2H, m), 7.2 (1H, d), 8.5 (1H, d).

9f: 47% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.4 (3H, s), 1.6 (3H, s), 3.8 (3H, s), 4.4 (1H, s), 4.65 (2H, d), 5.3~6.2 (3H, m), 5.8 (1H, d), 7.4 (1H, d), 8.35 (1H, t).

9g: 56% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.55 (3H, s), 1.7 (3H, s), 4.1 (3H, s), 4.55 (1H, s), 4.8 (2H, d),

5.2~6.0 (3H, m), 6.7~7.8 (5H, m).

9h: 34% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.5 (3H, s), 1.6 (3H, s), 4.45 (1H, s), 4.7 (2H, d), 5.1~6.3 (3H, m), 5.75 (1H, d), 7.1~7.8 (4H, m).

9i: 53% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.5 (3H, s), 1.64 (3H, s), 2.64 (3H, s), 4.48 (1H, s), 4.74 (2H, m), 5.3~5.5 (2H, m), 5.8 (1H, d), 7.1~7.2 (2H, m), 7.25 (1H, d), 7.6 (1H, t); IR (KBr) cm^{-1} 3433, 1795 (β -lactam CO), 1756, 1728, 1688, 1588.

9j: 47% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.5 (3H, s), 1.6 (3H, s), 4.5 (1H, s), 4.7 (2H, d), 4.95 (2H, d), 5.05~5.55 (3H, m), 5.7 (1H, d), 5.7~6.5 (3H, m), 6.6~7.2 (3H, m), 7.3~7.7 (1H, t).

9k: 54% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.45 (3H, s), 1.6 (3H, s), 4.4~4.8 (5H, m), 5.1~6.3 (6H, m), 5.65 (1H, d), 7.15 (2H, d), 7.7 (1H, d), 8.15 (1H, t).

9l: 49% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.5 (3H, s), 1.6 (3H, s), 2.55 (6H, s), 4.5 (1H, s), 4.7 (2H, d), 5.0~5.9 (3H, m), 5.7 (1H, d), 6.9 (1H, s), 7.3 (1H, d).

9m: 22% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.42 (3H, s), 1.60 (3H, s), 4.50 (1H, s), 4.70 (2H, d), 5.1~6.0 (3H, m), 5.32 (1H, s), 7.1 (1H, m), 9.73 (1H, d).

Allyl 1,1-Dioxo-6-(Z)-(2-hydroxyethylidene)penicillanate (9o)

To a solution of 190 mg (0.61 mmol) of allyl 1,1-dioxo-6-(Z)-formylmethylenepenicillanate (**9m**) in 4 ml of dry THF was added 0.61 ml (0.61 mmol) of 1 M diisobutylaluminum hydride in hexane at -78°C . The mixture was stirred at -78°C for 10 minutes, quenched with MeOH, stirred at room temperature for 20 minutes and filtered. The filtrate was concentrated *in vacuo* to give 258 mg of crude product which was diluted with water and extracted with chloroform. The organic layer was dried (MgSO_4) and concentrated to afford 160 mg of material which was purified by silica gel column chromatography (chloroform - EtOAc, 4 : 1) to yield 113 mg (60%) of **9o**. $^1\text{H NMR}$ (CDCl_3) δ 1.40 (3H, s), 1.60 (3H, s), 2.60 (1H, br s), 4.3 (2H, m), 4.4 (1H, s), 4.7 (2H, d), 5.1~6.0 (3H, m), 5.25 (1H, d), 6.38 (1H, m).

Allyl 1,1-Dioxo-6-(Z)-(methoxyiminomethylene)penicillanate (9p)

A solution of 210 mg (0.67 mmol) of allyl 1,1-dioxo-6-(Z)-formylmethylenepenicillanate (**9m**) in 5 ml of methylene chloride was treated with 56 mg (0.67 mmol) of methoxyamine HCl and 53 mg (0.67 mmol) of pyridine at room temperature. The mixture was stirred at room temperature for 5 hours, quenched with water and extracted with methylene chloride. The organic layer was dried (Na_2SO_4) and concentrated to give 189 mg (83%) of **9p** as a 1 : 1 mixture of (Z,Z) and (Z,E) isomers. $^1\text{H NMR}$ (CDCl_3) δ 1.60 (3H, s), 1.75 (3H, s), 4.1 (3H, s), 4.6 (1H, s), 4.9 (2H, m), 5.0~6.1 (3H, m), 6.6~7.3 (2H, m), 7.7 (0.5H s), 7.85 (0.5H, s).

Allyl 1,1-Dioxo-6-(Z)-(1-oxo-2-pyridyl)methylenepenicillanate (9s)

A solution of 100 mg (0.286 mmol) of allyl 1,1-dioxo-6-(Z)-(2-pyridyl)methylenepenicillanate (**9a**) in 5 ml of methylene chloride was treated with 120 mg (0.59 mmol) of 85% *m*-chloroperbenzoic acid and stirred at room temperature for 3 days. The mixture was quenched with saturated sodium thiosulfate solution and extracted with methylene chloride. The organic layer was neutralized with saturated bicarbonate solution, washed with water, dried (MgSO_4) and concentrated to give 82 mg of yellow oil. The oil was purified by silica gel column chromatography using EtOAc as eluent to give 22 mg (21%) of **9s** and 14 mg (13%) of a byproduct, 2,3-epoxypropanyl 1,1-dioxo-6-(Z)-(1-oxo-2-pyridyl)methylenepenicillanate, identified by $^1\text{H NMR}$ spectrum. $^1\text{H NMR}$ of **9s** (CDCl_3) δ 1.5 (3H, s), 1.6 (3H, s), 4.45 (1H, d), 4.7 (2H, d), 5.1~6.0 (3H, m), 5.8 (1H, s), 7.1~8.4 (5H, m).

Sodium 1,1-Dioxo-6-(Z)-(2-pyridyl)methylenepenicillanate (1a)

A mixture of 145 mg (0.4 mmol) of allyl 1,1-dioxo-6-(Z)-(2-pyridyl)methylenepenicillanate (**9a**), 20 mg of tetrakis(triphenylphosphine)palladium and 20 mg of triphenylphosphine was dissolved in 2 ml of EtOAc. 0.5 M Sodium 2-ethylhexanoate in EtOAc (0.8 ml, 0.4 mmol) was added and the mixture was stirred at room temperature for 20 minutes. The resulting precipitate was filtered, washed with EtOAc and dried to afford 130 mg (95%) of **1a** as yellow solid. $^1\text{H NMR}$ (D_2O) δ 1.5 (3H, s), 1.6 (3H, s), 4.23 (1H, s), 5.90 (1H, d), 7.1~8.0 (4H, m), 8.57 (1H, m); IR (KBr) cm^{-1} 3454, 1770 (β -lactam CO), 1621.

Compounds (**1b**~**1s**) were prepared from compounds (**9b**~**9s**) using potassium or sodium 2-ethylhexanoate according to the method described for **1a**.

1b: Sodium salt, 76% yield; ^1H NMR (250 MHz, $\text{DMSO-}d_6$) δ 1.45 (3H, s), 1.48 (3H, s), 3.82 (1H, s), 5.96 (1H, s), 7.55~8.55 (7H, m); IR (KBr) cm^{-1} 3437, 1771 (β -lactam CO), 1619.

1c: Potassium salt, 92% yield; ^1H NMR (250 MHz, $\text{DMSO-}d_6$) δ 1.40 (3H, s), 1.45 (3H, s), 3.85 (1H, s), 5.9 (1H, s), 7.53 (1H, s), 8.65 (2H, m), 8.9 (1H, s); ^{13}C NMR ($\text{DMSO-}d_6$) δ 173.3, 172.9, 152.6, 151.5, 150.9, 150.4, 141.3, 129.8, 76.3, 71.7, 69.8, 25.5, 23.8; IR (KBr) cm^{-1} 3406, 1770 (β -lactam CO), 1614.

1d: Potassium salt, 89% yield; ^1H NMR (D_2O) δ 1.6 (3H, s), 1.68 (3H, s), 4.4 (1H, s), 6.1 (1H, s), 7.48 (1H, s), 7.54 (1H, t), 8.88 (2H, d); ^{13}C NMR (D_2O) δ 175.5, 172.5, 163.2, 160.9, 139.9, 132.5, 124.5, 74.6, 68.9, 22.7, 20.9; IR (KBr) cm^{-1} 3439, 1771 (β -lactam CO), 1615.

1e: Potassium salt, 75% yield; ^1H NMR (D_2O) δ 1.8 (3H, s), 1.9 (3H, s), 4.56 (1H, s), 6.22 (1H, s), 7.2 (1H), 7.34 (1H), 7.62 (1H), 8.68 (1H); ^{13}C NMR (D_2O) δ 175.6, 173.7, 169.4, 155.1, 154.6, 134.2, 134.0, 115.9, 113.9, 74.6, 68.7, 64.6, 58.7, 22.7, 20.9; IR (KBr) cm^{-1} 3409, 1763 (β -lactam CO), 1615.

1f: Sodium salt, 55% yield; ^1H NMR (250 MHz, $\text{DMSO-}d_6$) δ 1.42 (3H, s), 1.48 (3H, s), 3.85 (1H, s), 3.95 (3H, s), 5.85 (1H, d), 7.45 (1H, d), 7.45~8.0 (2H, m), 8.22 (1H, m); IR (KBr) cm^{-1} 3454, 1767 (β -lactam CO), 1617.

1g: Potassium salt, 50% yield; ^1H NMR (D_2O) δ 1.6 (3H, s), 1.67 (3H, s), 4.02 (3H, s), 4.38 (1H, s), 6.21 (1H, s), 6.9 (1H, d), 7.19 (1H, d), 7.39 (1H, s); ^{13}C NMR (D_2O) δ 180.5, 179.3, 171.6, 160.4, 155.9, 147.9, 139.2, 129.7, 121.7, 78.2, 73.5, 73.0, 61.7, 27.1, 25.8; IR (KBr) cm^{-1} 3424, 1758 (β -lactam CO), 1620.

1h: Sodium salt, 86% yield; ^1H NMR (250 MHz, $\text{DMSO-}d_6$) δ 1.56 (3H, s), 1.64 (3H, s), 4.34 (1H, s), 6.18 (1H, s), 7.45~8.0 (4H, m); IR (KBr) cm^{-1} 3433, 1771 (β -lactam CO), 1624.

1i: Potassium salt, 76% yield; ^1H NMR (250 MHz, D_2O) δ 1.5 (3H, s), 1.6 (3H, s), 2.5 (3H, s), 4.28 (1H, s), 6.05 (1H, d), 7.2~7.35 (2H, m), 7.4 (1H, d), 7.67 (1H, t); ^{13}C NMR (250 MHz, D_2O) δ 175.7, 174.0, 163.0, 152.5, 140.7, 132.8, 128.3, 126.8, 74.5, 68.5, 68.4, 26.0, 22.4, 20.7; IR (KBr) cm^{-1} 3404, 1784 (β -lactam CO), 1758, 1611.

1j: Potassium salt, 73% yield; ^1H NMR (250 MHz, $\text{DMSO-}d_6$) δ 1.38 (3H, s), 1.44 (3H, s), 3.80 (1H, s), 5.0 (2H, m), 5.2~5.4 (2H, m), 5.9 (1H, d), 6.0~6.2 (1H, m), 6.91 (1H, d), 7.29 (1H, d), 7.33 (1H, d), 7.8 (1H, t); IR (KBr) cm^{-1} 3447, 1767 (β -lactam CO), 1616.

1k: Potassium salt; the reaction gave a mixture of *Z* and *E* isomers (1:1) by deblocking from the single *Z* isomer of the allyl ester; 99% yield; ^1H NMR (250 MHz, $\text{DMSO-}d_6$) δ 1.35 (1.5H, s), 1.4 (1.5H, s), 1.44 (1.5H, s), 1.48 (1.5H, s), 3.7 (1H, s), 4.55 (1H, d), 4.75 (1H, d), 5.15~5.5 (2H, m), 5.67 (0.5H, s), 5.8~6.0 (0.5H, m), 6.75~7.0 (2H, m), 7.4~7.6 (1.5H, m), 8.2 (0.5H, d); IR (KBr) cm^{-1} 3429, 1760 (β -lactam CO), 1613.

1l: Sodium salt, 75% yield; ^1H NMR (250 MHz, D_2O) δ 1.52 (3H, s), 1.60 (3H, s), 2.5 (6H, s), 4.32 (1H, s), 6.16 (1H, d), 7.35 (1H, d), 7.65~7.75 (1H, m); IR (KBr) cm^{-1} 3403, 1781 (β -lactam CO), 1629.

1o: Sodium salt, 53% yield; ^1H NMR (D_2O) δ 1.5 (3H, s), 1.6 (3H, s), 4.24 (1H, s), 4.30 (2H, dd), 5.75 (1H, s), 7.20 (1H, d); IR (KBr) cm^{-1} 3440, 1767 (β -lactam CO), 1674.

1p: Free acid, (*Z,Z*):(*Z,E*)=1:1, 79% yield; ^1H NMR (CDCl_3) δ 1.5 (3H, s), 1.6 (3H, s), 3.98 (3H, d), 4.42 (1H, s), 5.22 (1H, s), 6.5~6.7 (1H, m), 6.9~7.2 (1H, m), 7.7 (0.5H, s), 7.85 (0.5H, s); IR (KBr) cm^{-1} 3513, 1789 (β -lactam CO), 1727, 1709, 1610.

1s: Sodium salt, 97% yield; ^1H NMR (250 MHz, D_2O) δ 1.5 (3H, s), 1.6 (3H, s), 4.32 (1H, s), 5.95 (1H, d), 7.45 (1H, d), 7.5~7.9 (3H, m), 8.37 (1H, m); IR (KBr) cm^{-1} 3455, 1768 (β -lactam CO), 1622.

Dipotassium 1,1-Dioxo-6-(*Z*)-(2-(3-hydroxypyridyl)methylenepenecillanate (**1q**))

A mixture of 90 mg (0.22 mmol) of allyl 1,1-dioxo-6-(*Z*)-(2-(3-allyloxypyridyl)methylenepenecillanate (**9j**), 22 mg of tetrakis(triphenylphosphine)palladium and 22 mg of triphenylphosphine was dissolved in 2 ml of EtOAc. 0.5 M potassium 2-ethylhexanoate in EtOAc (0.88 ml, 0.44 mmol) was added and the mixture was stirred at room temperature for 30 minutes. The resulting precipitate was filtered

and washed with EtOAc to give 90 mg (100%) of brown solid. The brown solid consisted of a 49 : 51 mixture of **1q** and **1j**, respectively by HPLC analysis (MeOH - 0.02 M NH₄OAc, 1 : 1). The solid was then purified by medium pressure reverse-phase C18 column chromatography to yield 15 mg of pure **1q**. ¹H NMR (250 MHz, D₂O) δ 1.52 (3H, s), 1.61 (3H, s), 4.22 (1H, s), 5.97 (1H, s), 7.15 (1H, d), 7.27 (1H, dd), 7.8 (1H, s), 7.94 (1H, d); IR (KBr) cm⁻¹ 3424, 1758 (β-lactam CO), 1620.

Dipotassium 1,1-Dioxo-6-(Z)-(2-(6-hydroxypyridyl)methylenepenicillanate (1r)

1r was prepared by the method as described for **1q**. 92 mg (100% yield) of a 72 : 28 mixture of **1r** : **1k** (HPLC analysis) was obtained. The mixture was separated by medium pressure column chromatography on C18 silica gel (MeOH - water, 30 : 70) to give 24 mg of pure **1r**. ¹H NMR (250 MHz, D₂O) δ 1.52 (3H, s), 1.64 (3H, s), 3.75 (1H, s), 6.15 (1H, s), 6.75 (1H, d), 6.90 (1H, d), 7.35 (1H, s), 7.78 (1H, m); IR (KBr) cm⁻¹ 3420, 1776 (β-lactam CO), 1615.

Allyl 1,1-Dioxo-6α-bromopenicillanate (13)

A solution of 50 g (0.16 mol) of 1,1-dioxo-6α-bromopenicillanic acid¹⁷⁾ in 150 ml of DMF was treated with 13 ml (0.15 mol) of allyl bromide, 1.8 g of sodium bicarbonate, and 22 ml (0.16 mol) of triethylamine and the mixture was stirred at room temperature for 15 hours. The mixture was quenched with water and extracted 3 times with ethyl ether. The combined ether layers were washed with saturated sodium bicarbonate solution, washed with water, dried (Na₂SO₄) and concentrated *in vacuo* to afford 46 g (87%) of **13**.

Allyl 1,1-Dioxo-6α-(2-thiazolyl)hydroxymethylpenicillanate (11a')

Allyl 1,1-dioxo-6α-bromopenicillanate (**13**) (3.52 g, 10 mmol) was dissolved in 90 ml of dry THF and cooled to -78°C. 2.8 M Methylmagnesium bromide in ether (3.5 ml) was added and the mixture was stirred at -78°C for 2 minutes. A solution of thiazole-2-carboxaldehyde (1.13 g, 10 mmol) in 5 ml of dry THF was added and stirring continued at -78°C for 5 minutes. The mixture was poured into saturated ammonium chloride solution, extracted with EtOAc and the organic layer was dried (MgSO₄). Evaporation of solvent *in vacuo* gave an oil. The oil was purified by silica gel column chromatography (MeOH - CHCl₃, 5 : 95) to give 2.13 g (55%) of **11a'** as a 2 : 1 mixture of (6α,8R) and (6α,8S) stereoisomers, respectively. The mixture of isomers was separated by silica gel column chromatography (EtOAc - CHCl₃, 10 : 90) to give 0.511 g of the less polar (6α,8S) isomer, 1.022 g of a mixture of two isomers and 0.426 g of the more polar (6α,8R) isomer. The less polar isomer was recrystallized from EtOAc and the more polar isomer was recrystallized from methylene chloride. The stereochemistry of the less polar isomer was assigned as (6α,8S) on the basis of the X-ray analysis. ¹H NMR of (6α,8S) isomer (250 MHz, CDCl₃) δ 1.40 (3H, s), 1.60 (3H, s), 4.38 (1H, dd), 4.43 (1H, s), 4.67~4.75 (2H, m), 4.76 (1H, d), 5.3~5.5 (2H, m), 5.63 (1H, d), 5.85~6.05 (1H, m), 7.4 (1H, d), 7.8 (1H, d); IR (KBr) cm⁻¹ 3378, 1792 (β-lactam CO), 1739, 1645. ¹H NMR of (6α,8R) isomer (250 MHz, CDCl₃) δ 1.36 (3H, s), 1.60 (3H, s), 4.22 (1H, d), 4.4 (1H, s), 4.65 (2H, m), 4.88 (1H, s), 5.25~5.5 (2H, m), 5.5 (1H, d), 5.8~6.0 (1H, m), 7.35 (1H, d), 7.75 (1H, d); ¹³C NMR (CDCl₃) δ 170.5, 170.0, 166.6, 142.7, 130.7, 120.2, 120.1, 68.2, 66.9, 64.1, 63.1, 63.0, 58.2, 20.2, 18.7; IR (KBr) cm⁻¹ 3388, 1787 (β-lactam CO), 1736.

Compounds (**11c**, **11d**, **11c'**~**11k'**) were prepared employing the appropriate aldehyde in the procedure of example **11a'**.

11c: A 70 : 30 mixture of (8R) : (8S), 56% yield; ¹H NMR (250 MHz, CDCl₃) δ 1.40 (3H, s), 1.57 (3H, two sets of s), 4.25 (1H, m), 4.37 (0.7H, s), 4.42 (0.3H, s), 4.75 (2H, m), 4.8 (0.3H, d), 4.85 (0.7H, d), 5.25~5.5 (3H, m), 5.9 (1H, m), 8.52 (2H, m), 8.84 (1H, m); ¹³C NMR (250 MHz, CDCl₃) δ 170.1, 166.6, 153.7, 144.5, 144.4, 143.8, 143.5, 143.4, 142.9, 130.9, 130.8, 128.8, 120.4, 120.3, 68.2, 66.9, 64.1, 63.3, 63.2, 63.1, 58.2, 20.3, 20.2, 18.8, 18.6.

11d: 57% yield (mixture of isomers); further purification gave 9% yield of pure less polar (6α,8S) isomer and 8% yield of more polar (6α,8R) isomer. (6α,8S) Isomer: ¹H NMR (250 MHz, CDCl₃) δ 1.41 (3H, s), 1.6 (3H, s), 4.45 (1H, s), 4.4~4.8 (4H, m), 5.2~5.6 (3H, m), 5.7~6.3 (1H, m), 7.35 (1H, t), 8.85 (2H, d); (6α,8R) isomer: ¹H NMR (250 MHz, CDCl₃) δ 1.45 (3H, s), 1.6 (3H, s), 4.4 (1H, s), 4.45 (1H, dd), 4.7~4.9 (2H, m), 4.95 (1H, d), 5.2~5.6 (3H, m), 5.7~6.3 (1H, m), 7.35 (1H, t), 8.85

(1H, d).

11c': 37% yield (two isomers); ¹H NMR (CDCl₃) δ 1.4 (3H, s), 1.6 (3H, s), 4.2~4.4 (2H, m), 4.5~5.0 (3H, m), 5.1~6.1 (6H, m), 7.6 (1H, d), 8.87 (1H, d), 9.23 (1H, s).

11d': 25% yield; ¹H NMR (CDCl₃) δ 1.36 (1.5H, s), 1.40 (1.5H, s), 1.60 (1.5H, s), 1.65 (1.5H, s), 3.7 (3H, s), 4.0~4.5 (2H, m), 4.4 (1H, s), 4.5~4.8 (2H, m), 5.0~6.0 (4H, m), 6.7 (1H, s), 6.85 (1H, s).

11e': 33% yield; ¹H NMR (CDCl₃) δ 1.3 (3H, s), 1.55 (3H, s), 4.04 (1H, br s), 4.35 (1H, s), 4.5~4.85 (3H, m), 5.1~6.1 (4H, m), 7.1~7.4 (1H, m), 7.6~8.0 (1H, m), 8.2~7.8 (2H, m).

11f': 35% yield; ¹H NMR (CDCl₃) δ 1.3 (3H, s), 1.55 (3H, s), 4.0 (1H, m), 4.35 (1H, s), 4.4~6.8 (3H, m), 5.1~6.2 (4H, m), 7.2~7.5 (2H, m), 8.2~8.6 (2H, m).

11g': 78% yield; ¹H NMR (CDCl₃) δ 1.4 (3H, s), 1.6 (3H, s), 3.28 (1H, br s), 3.8~4.2 (1H, m), 4.35 (1H, s), 4.45~4.8 (3H, m), 5.0~6.1 (4H, m), 7.3 (5H, s).

11h': 38% yield; ¹H NMR (CDCl₃) δ 1.40 (3H, s), 1.60 (3H, s), 2.50 and 2.60 (3H, s), 4.1~4.4 (1H, m), 4.4 and 4.5 (1H, s), 4.6~5.0 (3H, m), 5.1~6.0 (4H, m), 6.0~7.2 (4H, m).

11i': 32% yield; ¹H NMR (CDCl₃) δ 1.40 (3H, s), 1.62 (3H, s), 3.68 (3H, s), 4.0~4.4 (1H, m), 4.42 (1H, s), 4.5~4.8 (3H, m), 5.0~6.0 (4H, m), 6.0~6.7 (3H, m).

11j': 36% yield; ¹H NMR (CDCl₃) δ 1.4 (0.4H, s), 1.5 (2.6H, s), 1.62 (0.4H, s), 1.7 (2.6H, s), 3.0 (1H, br s), 4.0~4.4 (1H, m), 4.45 (1H, s), 4.5~4.9 (2H, m), 5.1~6.1 (4H, m), 6.3~6.6 (2H, m), 7.45 (1H, m).

11k': 50% yield; ¹H NMR (CDCl₃) δ 1.25 (0.75H, s), 1.35 (2.25H, s), 1.55 (3H, s), 3.3 (1H, br s), 4.05 (1H, dd), 4.3 (1H, s), 4.3~4.8 (3H, m), 5.0~6.2 (4H, m), 6.8~7.4 (3H, m).

Allyl 1,1-Dioxo-6-(2-thiazolyl)methylenepenicillanate (9a')

A) Acetylation: A solution of allyl 1,1-dioxo-6-(2-thiazolyl)hydroxymethylpenicillanate (**11a'**) (500 mg, 1.29 mmol) in 5 ml of THF was treated with acetic anhydride (0.37 ml, 3.88 mmol) and pyridine (0.31 ml, 3.88 mmol) at room temperature. The resulting mixture was stirred at room temperature for 2 hours. The reaction was quenched with water and extracted with methylene chloride. The extracts were dried and concentrated to give 555 mg (100% yield) of **12a'** which was used directly for the next elimination step. ¹H NMR (CDCl₃) δ 1.52 (3H, s), 1.70 (3H, s), 2.35 (3H, s), 4.4~4.6 (2H, m), 4.6~5.0 (3H, m), 5.2~6.4 (3H, m), 6.65 (1H, d), 7.4 (1H, d), 7.8 (1H, d).

B) Elimination: The product from part A (555 mg, 1.29 mmol) was dissolved in 5 ml of methylene chloride and 1,5-diazabicyclo[4.3.0]non-5-ene (DBN) (160 mg, 1.29 mmol) was added. The mixture was stirred at room temperature for 1 hour, quenched with water and extracted with methylene chloride. The extracts were dried (Na₂SO₄) and concentrated to yield a brown oil. The oil was purified through silica gel column chromatography (EtOAc - hexane, 1:1) to give 189 mg (40%) of **9a'** as a yellow oil. ¹H NMR (CDCl₃) δ 1.53 (3H, s), 1.65 (3H, s), 4.33 (1H, s), 4.55 (2H, d), 5.0~5.4 (2H, m), 5.45 (1H, s), 5.4~6.0 (1H, m), 7.1 (1H, m), 7.25 (1H, m), 7.65 (1H, d).

The following compounds (**9**) were prepared from the corresponding hydroxy derivatives **11** by the same procedure as described for **9a'**.

12c: 100% yield (8R:8S=6:4); ¹H NMR (CDCl₃) δ 1.40 (0.6×3H, s), 1.50 (0.4×3H, s), 1.60 (3H, s), 2.3 (3H, s), 4.4 (2H, m), 4.7 (2H, m), 5.0~6.2 (4H, m), 6.42 (0.6H, d), 6.46 (0.4H, d), 8.7 (2H, s), 8.83 (1H, s).

9c: 63% yield (*Z* isomer); ¹H NMR (CDCl₃) δ 1.56 (3H, s), 1.7 (3H, s), 4.6 (1H, s), 4.85 (2H, m), 5.2~6.2 (3H, m), 5.8 (1H, d), 7.4 (1H, d), 8.5~9.0 (3H, m).

12d: 100% yield (8R:8S=1:1); ¹H NMR (CDCl₃) δ 1.4 (0.5×3H, s), 1.45 (0.5×3H, s), 1.62 (3H, s), 2.26 (3H, s), 4.43 (0.5H, s), 4.46 (0.5H, s), 4.45~5.0 (4H, m), 5.2~5.6 (2H, m), 5.7~6.3 (1H, m), 6.4 (0.5H, d), 6.5 (0.5H, d), 7.35 (1H, t), 8.85 (2H, d).

9d: 80% yield (*Z* isomer); ¹H NMR (CDCl₃) δ 1.45 (3H, s), 1.65 (3H, s), 4.5 (1H, s), 4.75 (2H, m), 5.2~6.3 (3H, m), 5.75 (1H, s), 7.1~7.5 (2H, m), 8.9 (2H, d).

9b': 59% yield; ¹H NMR (CDCl₃) δ 2.46 (3H, s), 2.60 (3H, s), 4.46 (1H, s), 5.20~5.42 (2H, m), 5.66 (1H, s), 5.80~6.02 (1H, m), 7.38~7.42 (2H, m), 8.71 (1H, m); IR (KBr) cm⁻¹ 1795 (β -lactam CO).

12c': 100% yield; ¹H NMR (CDCl₃) δ 1.4 (3H, s), 1.6 (3H, s), 2.2 (3H, s), 4.4 (1H, m), 4.6~5.0 (2H, m), 5.0~6.1 (3H, m), 6.3 (1H, d), 7.4 (1H, m), 8.9 (1H, m), 9.2 (1H, m).

9c': 55% yield (*Z* isomer); $^1\text{H NMR}$ (CDCl_3) δ 1.5 (3H, s), 1.6 (3H, s), 4.4 (1H, s), 4.7 (2H, m), 5.0~6.2 (3H, m), 5.7 (1H, d), 7.1 (1H, d), 7.2 (1H, dd), 8.8 (1H, d), 9.2 (1H, d).

12d': 75% yield (1:1 of isomers); $^1\text{H NMR}$ (CDCl_3) δ 1.4 (1.5H, s), 1.5 (1.5H, s), 1.6 (1.5H, s), 1.7 (1.5H, s), 2.2 (3H, s), 3.7 (3H, two sets of s), 4.0~6.0 (8H, m), 6.3~6.5 (1H, m), 6.8 (1H, m), 7.0 (1H, m).

9d': 90% yield (*Z* isomer); $^1\text{H NMR}$ (CDCl_3) δ 1.43 (3H, s), 1.60 (3H, s), 3.65 (3H, s), 4.35 (1H, s), 4.7 (2H, d), 5.0~6.0 (3H, m), 5.65 (1H, d), 6.9 (1H, d), 7.05 (1H, d), 7.15 (1H, m).

12g': 84% yield (two isomers); $^1\text{H NMR}$ (CDCl_3) δ 1.3 and 1.4 (3H, s), 1.6 (3H, s), 2.08 and 2.2 (3H, s), 4.2 (1H, dd), 4.4 (1H, s), 4.5 (1H, d), 4.65 (2H, d), 6.25 (1H, m), 7.3 (5H, m).

9g': 17% of *Z* isomer and 11% of *E* isomer. *Z* isomer: $^1\text{H NMR}$ (CDCl_3) δ 1.45 (3H, s), 1.58 (3H, s), 4.45 (1H, s), 4.75 (2H, d), 5.45 (1H, d), 5.2~6.2 (3H, m), 7.36 (1H, d), 7.45 (5H, s). *E* isomer: $^1\text{H NMR}$ (CDCl_3) δ 1.45 (3H, s), 1.60 (3H, s), 4.45 (1H, s), 4.68 (2H, d), 5.37 (1H, d), 5.1~6.05 (3H, m), 7.35 (1H, d), 7.45 (1H, s).

12h': 75% yield (*Z* isomer); $^1\text{H NMR}$ (CDCl_3) δ 1.4 (3H, s), 1.6 (3H, s), 2.15 (3H, s), 2.55 (3H, s), 4.15~4.3 (1H, dd), 4.4 (1H, s), 4.6~4.8 (3H, m), 5.1~6.0 (3H, m), 6.1~6.6 (2H, m), 6.6~7.4 (2H, m).

9h': 73% yield; $^1\text{H NMR}$ (CDCl_3) δ 1.5 (3H, s), 1.6 (3H, s), 2.55 (3H, s), 4.4 (1H, s), 4.65 (2H, d), 5.0~6.0 (4H, m), 6.3 (1H, t), 6.8 (1H, dd), 7.2 (1H, m), 8.2 (1H, d).

9i': 42% yield of needle crystals; $^1\text{H NMR}$ (CDCl_3) δ 1.5 (3H, s), 1.7 (3H, s), 3.72 (3H, s), 4.5 (1H, s), 4.75 (2H, m), 5.1~6.3 (4H, m), 6.6~7.0 (2H, m), 7.6 (1H, m).

Allyl 1,1-Dioxo-6-(2-furanyl)methylenepenicillanate (9j')

To a solution of 310 mg (0.84 mmol) of allyl 1,1-dioxo-6 α -(2-furanyl)hydroxymethylpenicillanate (**11j'**) in 5 ml of methylene chloride was added 0.14 ml (0.924 mmol) of trifluoromethylsulfonyl chloride and the mixture was then stirred at room temperature for 2 hours. The reaction mixture was quenched with water and extracted with methylene chloride. The organic layer was dried (MgSO_4) and concentrated to give 330 mg of crude product. The crude material was purified by silica gel column chromatography eluting with chloroform to afford **9j'** as a 4:1 mixture of (*Z*) and (*E*) isomers, respectively (HPLC analysis). $^1\text{H NMR}$ (CDCl_3) δ 1.47 (3H, s), 1.61 (3H, s), 4.47 (1H, s), 4.74 (2H, d), 5.1~6.2 (4H, m), 5.52 (1H, dd), 6.8 (1H, m), 7.15 (1H, d), 7.6 (1H, d).

Compounds (**9e'**, **9f'** and **9k'**) were also obtained by the method as described for **9j'**.

9e': The *Z* and *E* isomers were isolated in 22% and 19% yields, respectively. *Z* isomer: $^1\text{H NMR}$ (CDCl_3) δ 1.5 (3H, s), 1.7 (3H, s), 4.5 (1H, s), 4.7 (2H, d), 5.5 (1H, d), 5.1~6.2 (3H, m), 7.3~7.5 (2H, m), 7.7~8.0 (1H, m), 8.53~8.83 (2H, m). *E* isomer: $^1\text{H NMR}$ (CDCl_3) δ 1.48 (3H, s), 1.6 (3H, s), 4.5 (1H, s), 4.7 (2H, d), 5.25 (1H, s), 5.1~6.2 (3H, m), 6.88 (1H, s), 7.2~7.5 (1H, m), 8.43~9.0 (3H, m).

9f': 36% yield (*Z*:*E*=7:3); $^1\text{H NMR}$ (CDCl_3) δ 1.45 (3H, s), 1.62 (3H, s), 4.5 (1H, s), 4.7 (2H, d), 5.2~6.2 (4H, m), 6.75 (0.3H, s), 7.2~7.5 (1.7H, m), 7.6~7.85 (1H, m), 8.5~8.83 (2H, m).

9k': A 23% yield of **9k'** based on 38% of recovered starting material were obtained. $^1\text{H NMR}$ (CDCl_3) δ 1.45 (3H, s), 1.6 (3H, s), 4.4 (3H, s), 4.7 (2H, d), 5.0~6.2 (3H, m), 5.25 (1H, d), 7.0~7.65 (4H, m).

The following compounds were prepared using potassium or sodium 2-ethylhexanoate by the method as described for **1a**.

1a': 85% yield (*Z* isomer), potassium salt; $^1\text{H NMR}$ (250 MHz, $\text{DMSO}-d_6$) δ 1.40 (3H, s), 1.45 (3H, s), 3.80 (1H, s), 5.83 (1H, s), 7.66 (1H, s), 8.04 (2H, m).

1b': A 50% yield of a 7:2 mixture of *Z*:*E* isomers was obtained, which was then separated by C18 reverse phase medium pressure liquid chromatography (MPLC) (acetonitrile - water, 1:9). *Z* isomer, potassium salt; $^1\text{H NMR}$ (D_2O) δ 1.50 (3H, s), 1.60 (3H, s), 4.30 (1H, s), 6.03 (1H, s), 7.6 (1H, s), 7.62 (1H, d), 9.02 (1H, d); IR (KBr) cm^{-1} 1760 (β -lactam CO), 1610. *E* isomer, potassium salt; $^1\text{H NMR}$ (D_2O) δ 1.48 (3H, s), 1.60 (3H, s), 4.33 (1H, s), 5.66 (1H, s), 7.28 (1H, s), 8.2 (1H, d), 8.98 (1H, d).

1c': 92% yield (*Z* isomer), potassium salt; $^1\text{H NMR}$ ($\text{DMSO}-d_6$) δ 1.35 (3H, s), 1.40 (3H, s), 3.75 (1H, s), 5.74 (1H, d), 7.25 (1H, d), 7.55 (1H, dd), 8.9 (1H, d), 9.1 (1H, d); IR (KBr) cm^{-1} 3404,

1777 (β -lactam CO), 1620.

1d': 87% yield (*Z* isomer), potassium salt; ^1H NMR ($\text{DMSO-}d_6$) δ 1.38 (3H, s), 1.45 (3H, s), 3.80 (4H, s), 5.68 (1H, s), 7.15 (1H, s), 7.35 (2H, m); ^{13}C NMR ($\text{DMSO-}d_6$) δ 169.00, 167.9, 141.2, 130.6, 129.9, 124.6, 115.2, 70.55, 66.2, 64.4, 32.5, 20.2, 18.5; IR (KBr) cm^{-1} 3428, 1762 (β -lactam CO), 1614.

1e': 72% yield (*Z* isomer), sodium salt; ^1H NMR (D_2O) δ 1.55 (3H, s), 1.62 (3H, s), 4.23 (1H, s), 6.05 (1H, s), 7.4~8.8 (5H, m); IR (KBr) cm^{-1} 3462, 1788 (β -lactam CO), 1623.

1e': 91% yield (*E* isomer), sodium salt; ^1H NMR (D_2O) δ 1.55 (3H, s), 1.65 (3H, s), 4.30 (1H, s), 5.60 (1H, s), 6.96 (1H, s), 7.10~9.00 (4H, m); IR (KBr) cm^{-1} 3445, 1761 (β -lactam CO), 1623.

1f': 93% yield (*Z*:*E*=7:3), sodium salt; ^1H NMR (D_2O) δ 1.50 (3H, s), 1.60 (3H, s), 4.30 (0.3H, s), 5.55 (0.3H, s), 6.00 (0.7H, d), 6.95 (0.3H, s), 7.2~7.8 (4.7H, m).

1g': 91% yield (*Z* isomer), potassium salt; ^1H NMR (D_2O) δ 1.45 (3H, s), 1.55 (3H, s), 4.17 (1H, s), 5.50 (1H, s), 7.00 (1H, s), 7.27~7.60 (3H, m), 7.60~8.10 (2H, m); IR (KBr) cm^{-1} 3453, 1759 (β -lactam CO), 1619.

1h': 56% yield (*Z* isomer), sodium salt; ^1H NMR (D_2O) δ 1.56 (3H, s), 1.65 (3H, s), 2.68 (3H, s), 4.25 (1H, s), 5.88 (1H, s), 6.50 (1H, t), 7.00 (1H, d), 7.60~8.00 (1H, m), 8.20 (1H, s); IR (KBr) cm^{-1} 3438, 1764 (β -lactam CO), 1622.

1i': 73% yield (*Z* isomer), sodium salt; ^1H NMR (D_2O) δ 1.50 (3H, s), 1.60 (3H, s), 3.65 (3H, s), 4.10 (1H, s), 5.40 (1H, s), 6.10~6.50 (1H, m), 7.00 (2H, br s), 7.2~7.4 (1H, m).

1j': 47% yield (*Z*:*E*=3:1), sodium salt; ^1H NMR (250 MHz, D_2O) δ 1.5 (3H, s), 1.6 (3H, s), 4.25 (1H, s), 5.57 (0.25H, s), 5.9 (0.75H, s), 6.65 (1H, m), 7.0 (1H, d), 7.17 (0.25H, d), 7.36 (0.75H, s), 7.78 (1H, m); IR (KBr) cm^{-1} 3472, 1764 (β -lactam CO), 1621.

1k': 33% yield (*Z* isomer), sodium salt; ^1H NMR (D_2O) δ 1.55 (3H, s), 1.60 (3H, s), 4.2 (1H, s), 5.8 (1H, s), 7.0~7.9 (4H, m); IR (KBr) cm^{-1} 3425, 1773 (β -lactam CO), 1619.

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