THE PENEMS, A NEW CLASS OF β-LACTAM ANTIBIOTICS

7. SYNTHESIS AND ANTIMICROBIAL ACTIVITY OF 2-HETEROCYCLYLMERCAPTOALKYL DERIVATIVES

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2-Heterocyclylmercaptoalkyl penems were synthesized and their *in vitro* potency was established. The compounds exhibit moderate to strong antibacterial activity against various Gram-positive and Gram-negative bacteria. Their antimicrobial activity is related to the nature of the heterocycle, the length of the hydrocarbon spacer between the 2-position of the penem nucleus and the mercapto group, and the substitution pattern of the C-6 position of the penem skeleton.

In recent years, we have been engaged[1~3] in extensive studies of the influence of the C-2 substituent of the penem nucleus on the antimicrobial activity of the substances. Although a few 2-heterocyclylmercaptomethyl derivatives were described in the 6-(1-*R*-hydroxyethyl) series,[4,5] we decided to examine penem derivatives **1** (Table 1) with the more easily accessible 6-hydroxymethyl group, or with an extended C-2 hydrocarbon side-chain, or both.

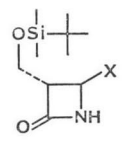
Chemistry

According to WOODWARD's strategy[6], the penem nucleus was obtained by an intramolecular Wittig cyclization of the appropriate phosphoranes **6** and **7**.

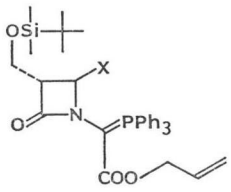
The (5*R*,6*S*)-6-hydroxymethyl-2-methyl derivatives **8a~h** were synthesized (Scheme 1) by two different methods; substitution of the methylsulfonyl leaving group in the azetidinone **2** with tritylmercaptan, formation of the phosphorane by the widely used and classical three-step procedure[7~9] (*i.e.* condensation of the glyoxylate at the NH group of the azetidinone, conversion to the chloride with $SOCl_2$ and subsequent reaction with Ph_3P to the phosphorane), conversion to the silver thiolate **5** with silver nitrate in methanol by analogy with known procedures[10,11] and acylation with chloroacetylchloride provided the key compound **6**. A nucleophilic displacement of the chlorine by thiolates afforded the phosphoranes **7a, d~h**, and subsequent thermolysis the corresponding penems **8a, d~h**. The alternative method is based on the prior cyclization of the key phosphorane **6** to the penem **9**, followed by the substitution of the chlorine to afford compounds **8b, c**.

The (5*R*,6*S*)-6-(1′*R*-hydroxyethyl)penem **8i** was obtained (Scheme 2) in seven steps from the 3-*tert*-butyldimethylsilyloxyethyl-4-*tert*-butylsulfonylazetidinone **10**. The replacement of the *tert*-butylsulfonyl moiety by a tritylthio group (**10**→**11**), construction of the phosphorane **12** by the three-step procedure, and subsequent cleavage of the *S*-trityl bond with silver nitrate in methanol afforded the silver thiolate **13**, acylation of which with 1-methyltetrazol-5-ylacetyl chloride gave the phosphorane **7i**. The Wittig cyclization readily yielded the corresponding penem **8i**.

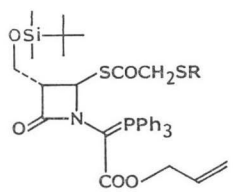
Scheme 1.



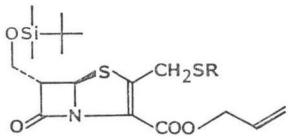
2 X = SO_2CH_3
3 X = $SCPh_3$



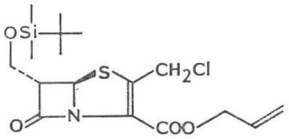
4 X = $SCPh_3$
5 X = SAg
6 X = $SCOCH_2Cl$



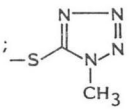
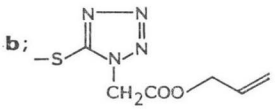
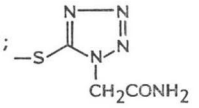
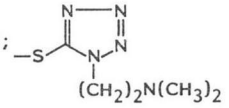
7a, d ~ h

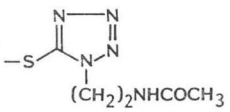
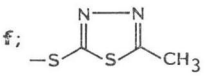
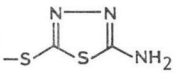
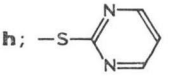


8a ~ h

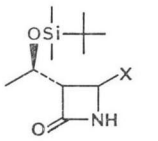


9

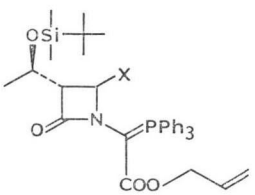
7, 8 SR: **a**;  **b**;  **c**;  **d**; 

e;  **f**;  **g**;  **h**; 

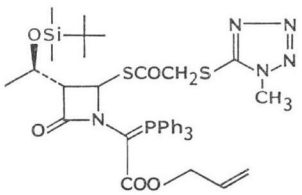
Scheme 2.



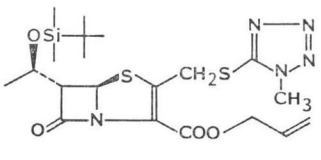
10 X = SO_2- 
11 X = $SCPh_3$



12 X = $SCPh_3$
13 X = SAg

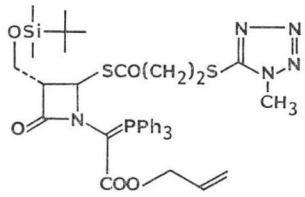


7i

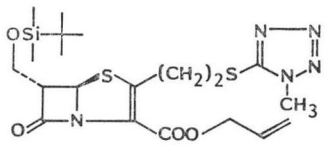


8i

Scheme 3.

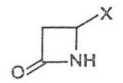


7j



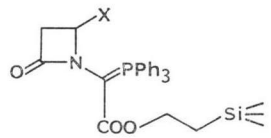
8j

Scheme 4.



14 X = OAc

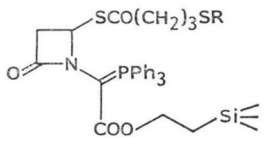
15 X = $SCPh_3$



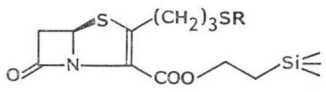
16 X = $SCPh_3$

17 X = SAg

18 X = $SCO(CH_2)_3Cl$

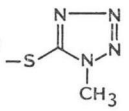


7k, l, m

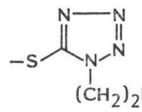


8k, l, m

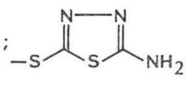
7, 8 SR: **k**;



l;



m;



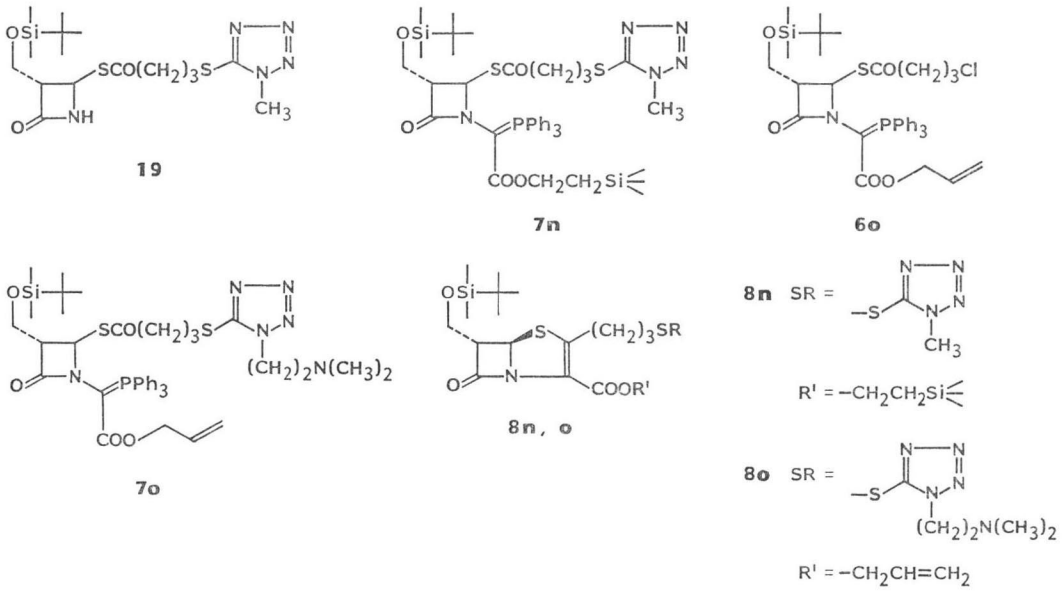
The (5*R*,6*S*)-2-ethyl derivative **8j** (Scheme 3) was obtained from the azetidinone **2**[12] by analogy with the above outlined synthesis by acylation of the previously described silver thiolate **5** with 3-(1-methyltetrazol-5-yl)mercaptopropionyl chloride (**5**→**7j**) and thermolysis of the latter to the penem **8j**.

The (5*R*,6*S*)-6-unsubstituted (5-*R*,*S*)-2-propyl derivatives **8k**～**m** were obtained (Scheme 4) through cyclization of the corresponding phosphoranes **7k, l, m**. The latter are synthesized by the usual multi-step sequence (**14**→**15**→**16**→**17**→**18**→**7k, l, m**) from 4-acetoxyazetidinone[13] **14**.

The 6-hydroxymethyl-2-propyl derivatives **8n, o** are both derived from the azetidinone **2** (Scheme 5). Whereas penem **8n** is obtained by the original WOODWARD procedure (*i.e.* substitution of acetoxy group by the appropriate thiocarboxylate to afford **19**, then the three-step sequence of the phosphorane synthesis to give **7n** and subsequent thermolysis to **8n**), penem **8o** is synthesized by acylation of the silver thiolate **5** with 4-chlorobutanoyl chloride to **6o**, substitution of the chlorine with 1-methyltetrazol-5-yl thiolate (**6o**→**7o**) and subsequent cyclization. The penems **8a**～**j, n, o** were deprotected to the final products **1a**～**j, n, o** by a two-step procedure:

A preliminary deblocking of the *tert*-butyldimethylsilyl group at the C-6 alcoholic function with tetrabutylammonium fluoride and acetic acid. For the 2-methylene derivatives **8a**～**j** this method often

Scheme 5.



afforded small amounts of a by-product identified as the isomeric 2-exomethylidene penams[5]. This problem was solved by inverting the reaction sequence, *i.e.* by deblocking first the hydroxyl-function in the phosphoranes **7** and then cyclizing the latter to the 6-hydroxymethyl (or 6-hydroxyethyl) penems.

Finally an either by Pd catalyzed deprotection of the allyl ester with tributyltinhydride (**8a**~**j**, **o**) or splitting of the trimethylsilylethyl ester with fluoride anions[14] (**8n**).

This last procedure was also used for the deprotection of the C-6 unsubstituted penems **8k**, **l**, **m**.

Antimicrobial Activity

The minimum inhibitory concentration (MIC) values of the 2-heterocyclylthioalkylpenems **1** for selected strains of Gram-positive and Gram-negative bacteria were determined by the agar dilution technique[15].

Table 1 lists the activities of a series of penem compounds with different C-6 substituents and various lengths of the C-2 carbon side-chain terminally substituted by the heterocyclic mercaptans commonly used in the cephalosporin series.

With the exception of the sodium salt of (4-mercaptotetrazol-1-yl)acetic acid (*cf.* **1b**), which possessed only moderate Gram-positive activities, all the heterocyclic substituents examined proved to be equally good. Penem compounds featuring no substituent at the C-6 position, although stable against β-lactamases of the Gram-positive strains, were unstable against the β-lactamases produced by Gram-negative bacteria and, in contrast to C-6 substituted penems, only moderately active against anaerobes.

Despite a slight gain on the Gram-positive side of the spectrum an increase in the length of the C-2 carbon side-chain induces a perceptible loss of activity on the Gram-negative side.

As observed earlier with other compounds in the penem series, and in striking contrast to thienamycin derivatives[16], none of the tested substances showed any significant activity against *Pseudomonas*.

Conclusions

Penem compounds bearing a 2-heterocyclylthioalkyl side-chain displayed a high degree of antibacterial activity, except against *Pseudomonas*. The 6-(1′*R*-hydroxyethyl) and the more easily accessible 6-hydroxymethyl derivatives proved to be about equally active, and in contrast to the 6-unsubstituted penems, stable against β-lactamase-producing strains (Table 1). With increasing length of the C-2 carbon side-chain, Gram-negative activity diminishes.

Experimental

IR spectra were obtained using a Perkin-Elmer apparatus Model 983G (main absorptions given in cm^{-1}). The UV spectra were recorded on a Cary 118 instrument. The 60 MHz ^1H NMR spectra were recorded on a Varian EM 360 A and the 360 MHz spectra on a Brucker HX 360 apparatus. The signals are listed in δ values (TMS 0.0). Melting points are uncorrected.

Antibiotic Susceptibility

All *in vitro* antibacterial activities are given as MIC in μg/ml. MICs were determined by the conventional agar dilution technique[15] using Diagnostic Sensitivity Test agar (Oxoid) enriched with 1% supplement C (Difco) after overnight incubation at 37°C and an inoculum of about 10^8 cfu/ml.

Phosphoranes

General Preparation of Phosphoranes **7a**, **d**～**h**, **k**～**m**, **o**; Phosphorane **7a**: Pyridine (1.32 ml), 4-dimethylaminopyridine (60 mg) and chloroacetyl chloride (0.6 ml) were added successively to an ice-cooled, stirred solution of the silver thiolate **5**, (3.56 g), prepared in four steps by analogy with known procedures[10,11] in abs CH_2Cl_2 (100 ml). After 30 minutes at 0°C the reaction mixture was filtered and after dilution with CH_2Cl_2, washed with aq $NaHCO_3$ and brine, then dried (Na_2SO_4) and evaporated *in vacuo* to give, after column chromatography (toluene - EtOAc, 95: 5), the intermediate phosphorane **6** (2.15 g): IR (CH_2Cl_2) cm^{-1} 1750, 1680, 1615.

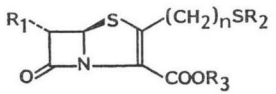
The phosphorane **6** (1.6 g) was dissolved in abs DMF (5 ml) and treated dropwise with a solution of sodium (1-mehyltetrazol-5-yl)thiolate (0.325 g) in abs DMF (5 ml). After 0.75 hour the reaction mixture was poured on ice water, and the insoluble title phosphorane **7a** (1.7 g) was obtained after filtration and drying *in vacuo*: IR (CH_2Cl_2) cm^{-1} 1750, 1670, 1615.

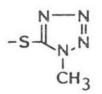
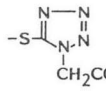
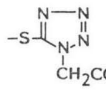
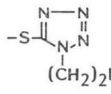
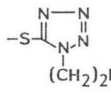
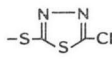
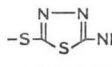
General Preparation of Phosphoranes **7i**, **j**; Phosphorane **7j**: To a solution of the silver thiolate **5** (4.5 g) in abs CH_2Cl_2 (100 ml) at 0°C were added pyridine (1.26 ml), 4-dimethylaminopyridine (20 mg) and, dropwise, a solution of 4-(1-methyltetrazolyl)mercaptopropionyl chloride (1.6 g). After 30 minutes the silver chloride was filtered off and, after dilution with CH_2Cl_2, the solution washed with aq $NaHCO_3$ and brine and finally dried (Na_2SO_4). Evaporation *in vacuo* of the solvent and purification by chromatography (toluene - EtOAc, 4: 1) gave the title compound **7j** (1 g): IR (CH_2Cl_2) cm^{-1} 1750, 1685, 1620.

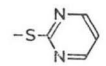
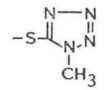
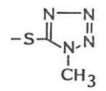
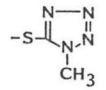
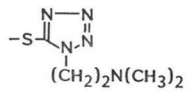
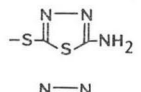
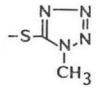
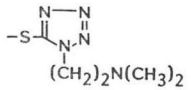
Phosphorane **7n**; Ethyl-4-(1-methyltetrazolylmercapto)butyrate: To a stirred solution of ethyl-4-bromobutyrate (6.44 g) in abs EtOH (60 ml) were added sodium (1-methyltetrazolyl)thiolate (5.34 g). After 30 minutes at 90°C the solvent was evaporated *in vacuo*. The residue was partitioned between H_2O and EtOAc and the separated organic layer was dried over Na_2SO_4. After evaporation of the solvent *in vacuo*, the crude material was purified by column chromatography (SiO_2; eluant: toluene - EtOAc, 1: 1) to afford 7.07 g of the title compound. IR (CH_2Cl_2) cm^{-1} 1730, 1370, 1160, ^1H NMR (60 MHz, $CDCl_3$) 1.27 (3H, t, CH_3), 1.9～2.65 (4H, 2m, $2CH_2$), 3.43 (2H, t, CH_2), 4.0 (3H, s, CH_3), 4.17 (2H, q, CH_2).

4-(1-Methyltetrazolylmercapto)butyric Acid: To a stirred solution of ethyl-4-(1-methyltetrazolylmercapto)butyrate (21.8 g) in THF (478 ml) was added a solution of 1 N NaOH (143 ml). After stirring for 3 hours at room temp, the solution was concentrated *in vacuo* and washed with EtOAc. The aq phase was transferred to an ice-bath, acidified to pH 3 by addition of 4 N HCl (30 ml) and extracted with EtOAc (3×80 ml). The extracts were dried (Na_2SO_4) and evaporated *in vacuo* to leave the pure title compound (17.79 g): IR ($CH_2Cl)_2$ cm^{-1} 3500, 2500, 1700; ^1H NMR (60 MHz, $CDCl_3$) 2.17 (2H, m, CH_2), 2.5 (2H, m, CH_2), 3.47 (2H, t, CH_2), 4.0 (3H, s, CH_3), 8.8 (1H, br s, COOH).

Table 1. *In vitro* antibacterial activity of various 2-heterocyclylthioalkylpenems **1**.



1	*n*	R_1	SR_2	R_3	MIC (μg/ml)							
					S. a. 10 B	*S. a.* 2999i$^+$p$^+$	*S. p.* Aronson	*E. c.* 205	*E. c.* 205 R+TEM	*M. m.* 2359	*P. a.* ATCC 12055	*B. f.* L01
a	1	CH_2OH		Na	0.1	0.2	0.05	0.2	0.2	1	>128	0.2
b	1	CH_2OH		Na	4	4	1	0.2	4	1	64	1
c	1	CH_2OH		Na	0.2	0.5	0.1	0.2	0.5	1	64	0.5
d	1	CH_2OH		Na	0.2	0.2	0.2	0.5	1	2	>128	1
e	1	CH_2OH		Na	0.5	1	0.1	0.2	1	1	>128	0.5
f	1	CH_2OH		Na	0.2	0.2	0.05	0.5	1	2	>128	0.1
g	1	CH_2OH		Na	0.1	0.2	0.05	0.5	1	2	128	0.2

h	1	CH_2OH		Na	0.2	0.2	0.1	1	2	4	128	0.2
i[4]	1	(*R*)-$CH(CH_3)OH$		Na	0.01	0.02	0.05	0.5	0.5	1	>128	0.05
j	2	CH_2OH		Na	0.05	0.05	0.1	1	2	2	>128	0.2
k	3	H		Na	0.05	0.05	0.02	2	>128	2	128	2
l	3	H		H	0.5	0.5	0.1	4	>32	8	> 32	8
m	3	H		H	1	1	0.5	16	> 32	8	> 32	8
n	3	CH_2OH		Na	0.05	0.05	0.1	2	4	2	>128	0.5
o	3	CH_2OH		Na	0.2	0.2	0.1	4	4	4	>32	1

Abbreviation: *S. a.*; *Staphylococcus aureus*, *S. p.*; *Streptococcus pyogenes*, *E. c.*; *Escherichia coli*, *M. m.*; *Morganella morganii*, *P. a.*; *Pseudomonas aeruginosa*, *B. f.*; *Bacteroides fragilis*.

4-(1-Methyltetrazolylmercapto)butyroyl Chloride: To a stirred solution of 4-(1-methyltetrazolylmercapto)butyric acid (2.2 g) in dry benzene (20 ml) were added thionyl chloride (1.03 ml) and DMF (4 drops). After refluxing for 20 minutes, the solvent was evaporated *in vacuo* and the crude title compound (2.1 g) was obtained without further purification: IR (CH_2Cl_2) cm^{-1} 1790, 1390, 1170; ^1H NMR (60 MHz, $CDCl_3$) 2.3 (2H, m, CH_2), 3.17 (2H, t, CH_2), 3.43 (2H, t, CH_2), 3.98 (3H, s, CH_3).

4-(1-Methyltetrazolylmercapto)thiobutyric Acid: A solution of 4-(1-methyltetrazolylmercapto)-butyroyl chloride (0.92 g) in dry CH_2Cl_2 (1.4 ml) was added dropwise to an ice-cooled solution (5.54 ml) of pyridine and H_2S in CH_2Cl_2. (The stock solution is prepared by dissolution of H_2S (6 g) in a pyridine (30 ml) and CH_2Cl_2 (100 ml) solution). After stirring for 1 hour at 0°C under a nitrogen atmosphere, the reaction mixture was diluted with CH_2Cl_2 and acidified with 2 N H_2SO_4. After separation of the organic phase and two further extractions with CH_2Cl_2 the combined organic layers were extracted twice with 10% $NaHCO_3$ (5 ml). The extracts were reacidified with 2 N H_2SO_4 (pH 3) and washed twice with CH_2Cl_2; the combined organic layers were dried and evaporated to give the title compound (0.75 g): IR (CH_2Cl_2) cm^{-1} 2575, 1695; ^1H NMR (60 MHz, $CDCl_3$) 2.2 (2H, m, CH_2), 2.9 (2H, t, CH_2), 3.43 (2H, t, CH_2), 3.97 (3H, s, CH_3), 4.35 (1H, br s, COSH).

(3*S*,4*R*)-3-(*tert*-Butyldimethylsilyloxymethyl)-4-(4-(1-methyltetrazol-5-ylmercapto)butyroylthio)-azetidin-2-one (**19**): To a stirred solution of **2**[12] (0.88 g) in dry THF (6 ml) was added dropwise, at room temp, 4-(1-methyltetrazol-5-ylmercapto)thiobutyric acid (0.72 g) dissolved in 0.33 N NaOH (10 ml). The pH was kept between 9 and 10 by addition of 0.33 N NaOH, and after 3.5 hours the reaction mixture was extracted with CH_2Cl_2. The combined organic extracts were washed with brine, dried (Na_2SO_4) and evaporated *in vacuo*. The title compound (0.7 g) was obtained by chromatography (toluene - EtOAc, 2: 1): IR (CH_2Cl_2) cm^{-1} 3410, 1770, 1680; ^1H NMR (60 MHz, $CDCl_3$) 0.06 (6H, s, $2CH_3$), 0.9 (9H, s, $3CH_3$), 2.23 (2H, m, CH_2), 2.82 (2H, t, CH_2), 3.38 (1H, m, CH), 3.45 (2H, t, CH_2), 3.97 (3H, 2H, s, m, CH_3, CH_2), 5.35 (1H, d, CH_2), 6.5 (1H, br s, NH).

Trimethylsilylethyl-2-[(3*S*,4*R*)-3-(*tert*-butyldimethylsilyloxymethyl)-4-[4-(1-methyltetrazol-5-yl-mercapto)butyroylthio]-2-oxo-azetidin-1-yl]triphenylphosphoranylidene Acetate (**7n**): To the mixture of **19** (3.35 g) and the ethyl-hemiacetal of trimethylsilylglyoxylate (3.42 g) in toluene (60 ml) and DMF (12 ml) were added 4A Molecular Sieves (30 g).

After stirring at 100°C for 8 hours, the mixture was filtered, the solvents evaporated under vacuum and the crude hydroxyl derivative dissolved in THF (35 ml) and subsequently reacted at −15°C over 10 minutes with thionyl chloride (1.04 ml) and $N(Et)_3$ (2.08 ml). After 1 hour at 0°C, the reaction mixture was filtered and evaporated *in vacuo* to leave the corresponding chloride. The latter was dissolved without further purification in dioxane (20 ml) treated with Ph_3P (2.25 g) and 2,6-lutidine (1 ml) and stirred for 18 hours at 50°C. The reaction mixture was finally filtered and the pure title compound **7n** (2.16 g) obtained by column chromatography (toluene - EtOAc, 4: 1): IR (CH_2Cl_2) cm^{-1} 1745, 1680, 1600.

General Preparation of the Penems **8a**, **d**～**o**; Penem **8a**: A stirred solution of phosphorane **7a** (1.7 g) in abs toluene (500 ml) was heated at 90°C for 50 minutes. After evaporation of the solvent *in vacuo* the residue was chromatographed (toluene - EtOAc, 95: 5) to give the title compound **8a** (0.99 g): MP (ether - hexane) 57～59°C; IR (CH_2Cl_2) cm^{-1} 1785, 1700, 1575, 1310; ^1H NMR (360 MHz, DMSO-d_6) 0.07 (6H, s, $2CH_3$), 0.89 (9H, s, $3CH_3$), 3.91 (2H, m, CH_2), 5.25 (1H, m, CH), 5.42 (1H, m, CH), 5.56 (2H, d, CH), 5.95 (1H, m, CH).

General Preparation of the Penems **8b**, **c**; Penem **8c**: A stirred solution of phosphorane **6** (0.341 g) in abs toluene (15 ml) was heated at 90°C for 45 minutes. After evaporation of the solvent *in vacuo*, the crude **9** (IR (CH_2Cl_2) cm^{-1} 1785, 1705, 1580) was dissolved in DMF (1 ml) and treated dropwise with a solution of (1-carbamoylmethyltetrazol-5-yl)thiol (0.087 g) and *N*,*N*-diisopropylethylamine (0.094 ml) in DMF (0.5 ml). The reaction mixture was partitioned between H_2O and EtOAc, and the organic phase separated, washed with H_2O and brine, dried (Na_2SO_4) and evaporated to dryness. Chromatographic purification (EtOAc) yielded the title compound **8c** (0.128 g): IR (CH_2Cl_2) cm^{-1} 3500, 3400, 1785, 1700, 1575; ^1H NMR (360 MHz, $CDCl_3$) 0.055 (6H, s, $2CH_3$), 0.87 (9H, s, $3CH_3$), 3.88 (2H, m, CH_2), 3.95 (1H, m, CH), 4.57, 4.75 (2H, ABq, CH_2), 4.69 (2H, m, CH_2), 5.0 (2H, s, CH_2), 5.25 (1H, m, CH), 5.4 (1H, m, CH), 5.59 (1H, d, CH), 5.91 (1H, m, CH), 5.95, 6.15 (2H, 2br s, $CONH_2$).

Deprotection of the Functional Groups

General Preparation of the Penems **1a**~**j**, **o**; Penem **1j**: To a stirred solution of penem **8j** (0.71 g) in THF (15 ml) were added at −70°C successively AcOH (0.6 ml) and, over a 15-minute period, a 0.1 M tetrabutylammonium fluoride (TBAF) - THF solution (31.3 ml). The cooling bath was removed and after 3.75 hours at room temp the reaction mixture was concentrated *in vacuo*, diluted with EtOAc, washed with aq $NaHCO_3$ and brine and after drying (Na_2SO_4) evaporated to dryness. Column chromatography afforded the intermediate 6-hydroxymethyl derivative: IR (CH_2Cl_2) cm^{-1} 3600, 1780, 1700, 1580; ^1H NMR (360 MHz, $CDCl_3$) 3.22, 3.45 (2H, m, CH_2), 3.56 (2H, m, CH_2), 3.91 (3H, s, CH_3), 3.95 (1H, m, CH), 4.05 (2H, m, CH_2), 4.7 (2H, m, CH_2), 5.26 (1H, m, CH), 5.4 (1H, m, CH), 5.64 (1H, d, CH), 5.94 (1H, m, CH). To a stirred solution of this penem (0.265 g) in abs THF (10 ml) were added at −10°C tetrakis(triphenylphosphine)palladium (15 mg) and tributyltin hydride (0.22 ml). After 20 minutes at the same temp AcOH (0.047 ml) was added and the reaction mixture was concentrated *in*

Table 2. Spectroscopic data of penem compounds **1**.

Compound	^1H NMR (360 MHz, D_2O) δ (ppm)	IR (cm^{-1})			UV $\lambda_{max}^{H_2O}$ nm
		Solvent	β-Lactam	COO^-	
1a	3.92 (2H, m, CH_2), 4.0 (1H, m, CH), 4.12 (3H, s, CH_3), 4.44, 4.57 (2H, ABq, CH_2), 5.61 (1H, d, CH)	KBr	1776	1616	313
1b	3.96 (3H, m, CH_2, CH), 4.47~4.65 (2H, m, CH_2), 5.12 (2H, s, CH_2), 5.6 (1H, d, CH)	DMSO-d_6	1770	1637	310
1c	3.86 (2H, m, CH_2), 3.99 (1H, m, CH), 4.49~4.66 (2H, m, CH_2), 5.37 (2H, s, CH_2), 5.58 (1H, d, CH)	—	—	—	312*
1d	2.6 (6H, s, $2CH_3$), 2.88 (2H, s, CH_2), 3.93 (2H, m, CH_2), 4.0 (1H, m, CH), 4.48, 4.6 (2H, ABq, CH_2), 4.68 (2H, m, CH_2), 5.62 (1H, s, CH)	—	—	—	312
1e	1.93 (3H, s, CH_3), 3.67 (2H, m, CH_2), 3.92 (2H, m, CH_2), 4.0 (1H, m, CH), 4.52, 4.66 (2H, ABq, CH_2), 4.6 (2H, m, CH_2), 5.6 (1H, d, CH)	DMSO-d_6	1773	1619	312
1f	2.76 (3H, s, CH_3), 3.93 (3H, m, CH_2, CH), 4.58 (2H, m, CH_2), 5.58 (1H, d, CH)	DMSO-d_6	1771	1619	310
1g	3.94 (2H, m, CH_2), 3.98 (1H, m, CH), 4.41 (2H, s, CH_2), 5.59 (1H, d, CH)	—	—	—	310
1h	3.94 (2H, m, CH_2), 4.02 (1H, m, CH), 4.48~4.72 (2H, m, CH_2), 5.55 (1H, d, CH), 7.25 (1H, m, CH), 8.62 (2H, m, 2CH)	DMSO-d_6	1769	1619	300
1i	1.29 (3H, d, CH_3), 3.87 (1H, m, CH), 4.12 (3H, s, CH_3), 4.22 (1H, m, CH), 4.44~4.56 (2H, m, CH_2), 5.61 (1H, d, CH)	KBr	1764	1612	310
1j	3.1~3.38 (2H, m, CH_2), 3.55 (2H, m, CH_2), 3.86 (1H, m, CH), 3.93 (2H, m, CH_2), 4.0 (3H, s, CH_3), 5.48 (1H, d, CH)	KBr	1762	1602	305
1k	—	DMSO-d_6	1770	1610	303*
1l	—	—	—	—	306*
1m	—	—	—	—	284*,**
1n	2.0 (2H, m, CH_2), 2.8~3.1 (2H, m, CH_2), 3.35 (2H, m, CH_2), 3.96 (3H, s, CH_3), 4.0 (3H, m, CH_2, CH), 5.6 (1H, d, CH)	DMSO-d_6	1769	1610	303
1o	—	—	—	—	303

* Phosphate buffer, pH 7.4.

** Absorption is mainly due to the presence of the aminothiadiazolemercapto moiety.

vacuo and partitioned between H_2O and EtOAc. The pH of the aq phase was maintained at 8 by addition of $NaHCO_3$. After separation of the organic phase and concentration of the mixture *in vacuo*, column chromatography (XAD-2; elution with H_2O) and lyophilization gave the penem **1j** (0.135 g) (Table 2).

General Preparation of the Penems **k, l, m, n**; Penem **1n**: Trimethylsilylethyl 6-hydroxymethyl-2-(1-methyltetrazol-5-yl)mercaptopropyl penem carboxylate (0.77 g) (obtained from **8n** by cleavage of the *tert*-butyldimethylsilyl ether by the method described above) was dissolved in THF (14 ml) and after cooling at −30°C, treated in several portions with a 0.1 M TBAF - THF solution (67 ml). After 10 minutes the reaction mixture was diluted with EtOAc (90 ml) and H_2O (90 ml). The aq phase was acidified to pH 3 by addition of 4 N HCl and the organic phase separated and extracted twice with 0.05 M $NaHCO_3$. The aq extracts were concentrated *in vacuo* and chromatographed on an OPTI-UPC 12 (Antec AG CH-4431 Bennwil) reversed phase silica gel column. Elution with H_2O and lyophilization of the product containing fractions afforded the title compound **1n** (0.22 g) (Table 2).

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