SYNTHESIS AND BIOLOGICAL ACTIVITY OF 3-VINYLTHIO- AND 3-VINYLTHIOMETHYLCEPHEM DERIVATIVES

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The synthesis and biological properties of some 3-vinylthio- and 3-vinylthiomethylcephem derivatives are described. Both series possess potent antibacterial activity. Among them, 3-[(*Z*)-2-cyanovinylthiomethyl]cephem derivative was found to have an expanded antibacterial spectrum.

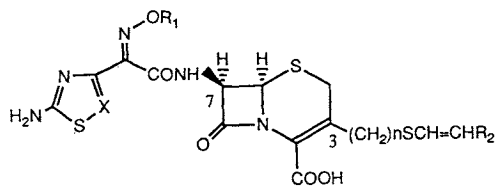
Recently we have studied the synthesis and biological properties of cephalosporins with a 1-hydroxyethyl moiety in the 7 position, which is well-known as the unique side chain of many carbapenem antibiotics.[1,2] We reported that 7α-(1-hydroxyethyl)cephem derivatives, which had an electron-withdrawing group at the 3 position, possessed potent β-lactamase inhibitory activity. This finding of a substituent effect led us to investigate the biological properties of cephalosporin derivatives having other substituents which are characteristic in carbapenem antibiotics.

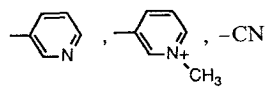
As shown in Fig. 1, we were interested in the vinylthio substituents, which are the C-2 side chains of many natural occurring carbapenem compounds such as *N*-acetyldehydrothienamycin (**3**),[3] AB-110-D (**4**).[4] In cephem compounds, little is known about the substituent effect of vinylthio(methyl) moiety at the C-3 position on the biological property in contrast to well-known heterocyclic thio(methyl)cephem derivatives.[5] Thus, we prepared two new types of derivatives, 3-vinylthiocephem (**1**) and 3-vinylthiomethylcephem (**2**).

Chemistry

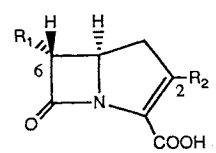
Initially, we attempted to introduce the vinylthio moiety to the C-3 position employing an addition-elimination reaction of lithium vinylthiolate (**7**), which was derived from 2-ethoxy-1,3-oxathiolane (**6**) as described by TANIMOTO and his co-workers,[6] to 3-methylsulfonyloxycephem (**16**).[5] However, this treatment afforded only Δ^2-isomer of **16** and degradation products due to the strongly basic condition. We then employed silver thiolate as described by BATESON and his co-workers.[7] Various silver vinylthiolates **5a**~**5f**†

Fig. 1. Structures of compounds **1**~**4**.



R_1 = Et, Allyl
R_2 = –NHAc, H,  , –CN
X = CH, N

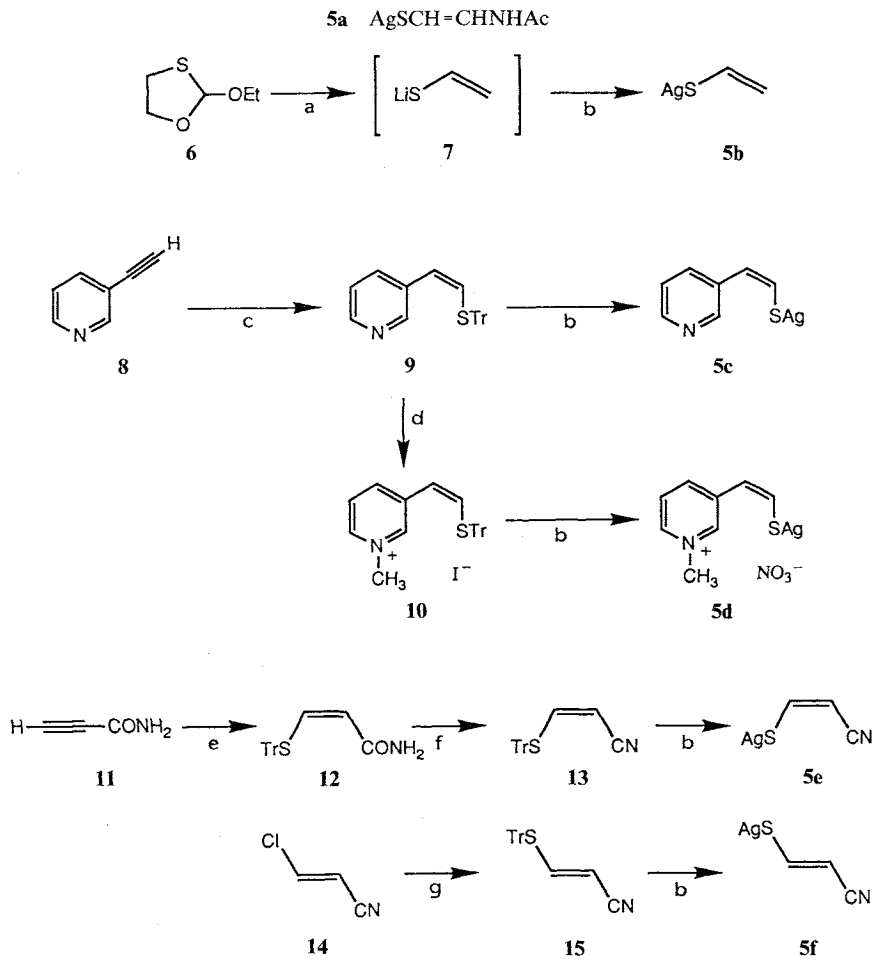
1 n = 0
2 n = 1



R
3 R_1 = –CH(OH)CH_3
R_2 = –SCH=CHNHAc (*E*)
4 R_1 = –CH(OSO_3H)CH_3
R_2 = –SCH=CHNHAc (*Z*)

† As silver salts **5a**~**5f** are explosive, particular attention should be given to these compounds.

Scheme 1. Synthesis of silver salts **5a**~**5f**.

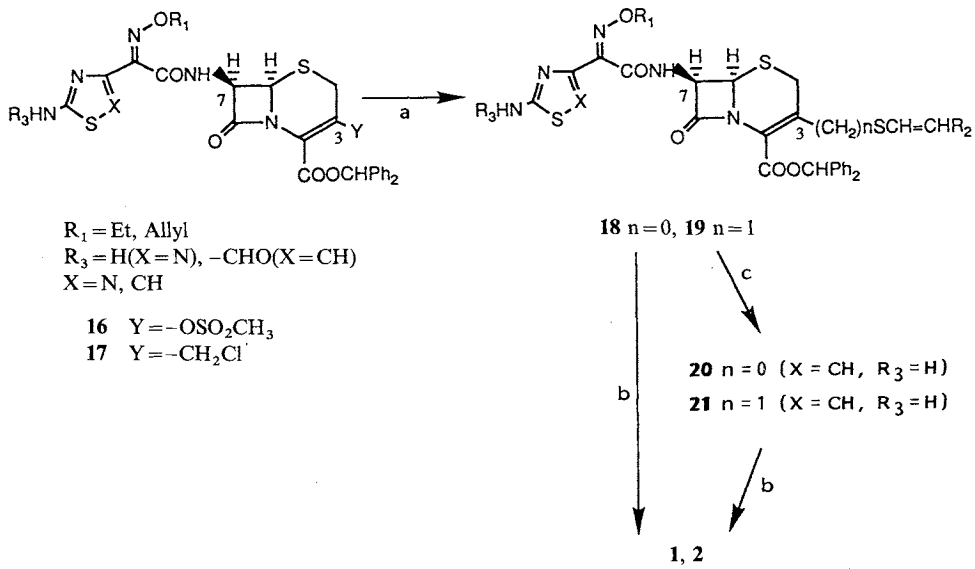


a) LDA, b) $AgNO_3$, c) Ph_3CSH, *tert*-BuOK, d) CH_3I, e) Ph_3CSH, 1 N NaOH, f) PCl_5, g) Ph_3CSH, Et_3N.

were synthesized as shown in Scheme 1. Silver acetamidovinylthiolate (**5a**)[7] was obtained from triphenylmethylthiol and bromoacetaldehyde diethylacetal *via* several steps as *E,Z* mixture (*E*-*Z*, 3:2). Lithium vinylthiolate (**7**) reacted with silver nitrate in the dark to give crude silver vinylthiolate (**5b**). 3-Ethynylpyridine (**8**) was treated with triphenylmethylthiol and potassium *tert*-butoxide to give 3-[(*Z*)-2-(triphenylmethylthio)vinyl]pyridine (**9**) in 60% yield. The *Z* assignment of the vinyl moiety was based upon the observed coupling constant (J_{cis}=12 Hz) in ^1H NMR. Detritylation of **9** with silver nitrate gave silver salt **5c**. Compound **9** reacted with methyl iodide to give 1-methyl-3-pyridinium compound **10** in 90% yield. Compound **10** was also converted to silver salt **5d** in a similar manner. Silver (*Z*)-2-cyanovinylthiolate **5e** was derived from propionamide (**11**) in the same manner as that of **5c**. The (*E*)-isomer **5f** was prepared by the treatment of (*E*)-2-chloroacrylonitrile (**14**) with triphenylmethylthiol and triethylamine followed by reaction with silver nitrate.

As shown in Scheme 2, these silver salts **5a**~**5f** were treated with 3-methylsulfonyloxy cephems **16** or 3-chloromethylcephems **17**[8] and sodium iodide in the dark to give 3-vinylthiocephems **18** (n=0) or 3-vinylthiomethylcephems **19** (n=1), respectively. In these reactions, the treatment of *E,Z* mixture of **5a**

Scheme 2. General synthetic route of 3-vinylthio- and 3-vinylthiomethylcephem derivatives, **1** and **2**.



a) **5a**~**5f**, NaI, b) TFA, anisole, c) conc HCl-methanol.

with **16** afforded only *Z*-isomer of **18**, and the other treatment of **5b**~**5f** with **16** or **17** gave **18** or **19** with retention of the configuration. The protecting groups in **18** and **19** were removed in usual fashion to give **1** and **2**, that is, the *N*-formyl groups were deprotected with concentrated hydrochloric acid in methanol, the diphenylmethyl groups were removed with trifluoroacetic acid and anisole, respectively.

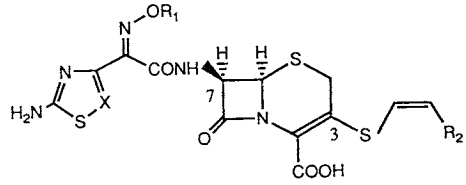
Biological Results and Discussion

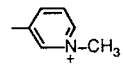
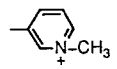
The *in vitro* antibacterial activities of the 3-vinylthiocephem derivatives **1a**~**1e** against selected Gram-positive and Gram-negative bacteria are shown in Table 1. Compound **1a**, which has (*Z*)-2-(acetamido)vinylthio group at the C-3 position, corresponding to the C-2 side chain of carbapenem compound **4**, showed greater activity against *Staphylococcus aureus* 209P JC-1 than ceftizoxime (CZX), which has no substituent at the 3 position, moderate activity against *Escherichia coli* NIHJ JC-2 and *Klebsiella pneumoniae* 12, and weak activity against *Pseudomonas aeruginosa* IAM 1095. The antibacterial spectra of compounds **1b**~**1e** were similar to that of compound **1a**. 3-Pyridinium compounds, **1d-1** and **1d-2** were potent activity against *S. aureus* 209P JC-1, *E. coli* NIHJ JC-2, and *K. pneumoniae* 12, but less active against *P. aeruginosa* IAM 1095 than the corresponding 3-pyridine compounds, **1c-1** and **1c-2**.

The antibacterial spectra of 7β-(2-aminothiazol)cephem compounds (**1c-2** and **1d-2**, X=CH) were similar to those of the corresponding 7β-(5-amino-1,2,4-thiadiazol)cephem compounds (**1c-1** and **1d-1**, X=N).

The MICs of the 3-vinylthiomethylcephem derivatives, **2c**~**2f** are shown in Table 2. The antibacterial spectra of **2c-1** and **2c-2** are similar to that of the 3-vinylthiocephem derivatives, and in comparison between **1c** and **2c**, the vinylthiomethylcephem derivatives, **2c-1** and **2c-2** have twice to eight times less activity against *S. aureus* 209P JC-1, *E. coli* NIHJ JC-2 and *K. pneumoniae* 12 than the corresponding vinylthiocephem derivatives, **1c-1** and **1c-3**. The antibacterial activity of 3-pyridinium compound **2d** had improved activity in comparison to the corresponding 3-pyridine compound **2c-1**.

Table 1. MICs of 3-vinylthiocephem derivatives **1a**~**1e**.

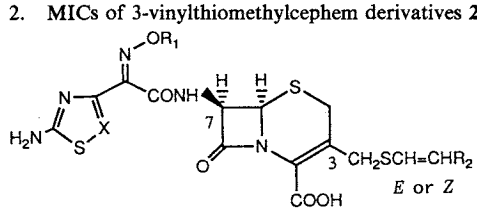


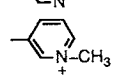
Compound No.	R_1	X	R_2	MIC (μg/ml)			
				S.a.	*E.c.*	*K.p.*	*P.a.*
1a	Et	N	NHAc	0.78	0.39	1.56	25
1b	Et	N	H	1.56	0.39	0.78	12.5
1c-1	Et	N		0.39	0.10	0.78	6.25
1c-2	Et	CH		0.78	0.10	0.78	3.13
1c-3	Allyl	N		0.20	0.20	0.78	12.5
1d-1	Et	N		0.20	0.05	0.39	50
1d-2	Et	CH		0.20	0.05	0.39	12.5
1e	Allyl	N	CN	0.78	0.20	0.78	25
CZX				6.25	≦0.025	≦0.025	50
IPM				≦0.025	0.78	0.10	0.78

Abbreviations: *S.a.*, *Staphylococcus aureus* 209P JC-1; *E.c.*, *Escherichia coli* NIHJ JC-2; *K.p.*, *Klebsiella pneumoniae* 12; *P.a.*, *Pseudomonas aeruginosa* IAM 1095.

[a] Mueller-Hinton agar 10^{-2}; stamp method; 37°C, 18 hours.

Table 2. MICs of 3-vinylthiomethylcephem derivatives **2c**~**2f**.

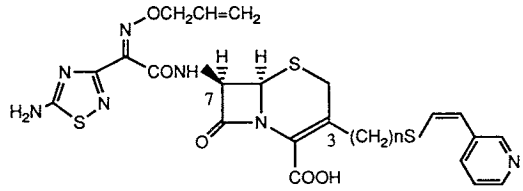


Compound No.	R_1	X	R_2	*E,Z*	MIC (μg/ml)[a]			
					S.a.	*E.c.*	*K.p.*	*P.a.*
2c-1	Et	N		*Z*	0.78	0.78	3.13	6.25
2c-2	Allyl	N		*Z*	0.39	0.78	3.13	3.13
2d	Et	N		*Z*	0.78	0.05	0.20	3.13
2e-1	Allyl	N	CN	*Z*	0.78	0.10	0.78	0.78
2e-2	Allyl	CH	CN	*Z*	0.39	0.39	1.56	1.56
2f	Allyl	N	CN	*E*	0.78	0.10	0.78	3.13
CZX					6.25	≦0.025	≦0.025	50
IPM					≦0.025	0.78	0.10	0.78

Abbreviations: See Table 1.

[a] Mueller-Hinton agar 10^{-2}; stamp method; 37°C, 18 hours.

Table 3. Binding affinities of **1c-3**, **2c-2**, CZX, and IPM for PBPs in *Escherichia coli* NIHJ JC-2.



Compound No.	n	I_{50} (μg/ml)[a]						
		1A	1B	2	3	4	5	6
1c-3	0	0.47	0.22	1.9	0.036	8.5	>25	5.8
2c-2	1	0.015	2.1	>25	0.04	>25	>25	>25
CZX		0.020	0.1	>25	0.012	>25	>25	>25
IPM		0.2	0.6	<0.1	9.8	<0.1	0.3	0.6

[a] Concentration required to inhibit binding of [^{14}C]benzylpenicillin to each protein by 50%.

Interestingly, we found that (*Z*)-2-cyanovinylthiomethyl compounds, **2e-1** and **2e-2** have potent activity against *P. aeruginosa* IAM 1095, while the corresponding (*E*)-isomer **2f** and (*Z*)-cyanovinylthio derivative **1e** have lower activity.

As a result, compound **2e-1** has an expanded antibacterial spectra against both Gram-positive bacteria and Gram-negative bacteria including *P. aeruginosa*, and among 3-vinylthio(methyl)cephem derivatives, (1-methyl-3-pyridinio)vinylthio compounds, **1d-1** and **1d-2** have the most potent activity against *S. aureus* 209P JC-1, *E. coli* NIHJ JC-2, and *K. pneumoniae* 12.

Table 3 showed the effect of the C-3 substituents of cephem derivatives, **1c-3** and **2c-2** upon their affinity for the penicillin-binding proteins (PBPs) of *E. coli* NIHJ JC-2. The 3-vinylthiomethyl compound **2c-2** has strong affinity for PBPs 1A, 1B, and 3, and its affinity pattern is similar to that of CZX. However, the corresponding 3-vinylthiocephem compound **1c-3** has good affinity not only for PBPs 1A, 1B, and 3, but also for PBP 2, which is the primary target for carbapenems such as imipenem (IPM). For the present, it is uncertain how the difference in the affinity profiles between **1c-3** and **2c-2** is reflected in the antibacterial activity although it is a notable feature.

Experimental

MP's were measured with a Yanagimoto micro melting point apparatus and are uncorrected. IR spectra were recorded with a Hitachi 260-10 spectrometer. ^{1}H NMR were recorded using a Hitachi R-90H spectrometer. Chemical shifts (δ) are recorded in ppm from sodium 2,2-dimethyl-2-silapentane-5-sulfonate (DSS) (in D_2O) or TMS (in $CDCl_3$ and DMSO-d_6) as internal standard.

Preparation of Silver Vinylthiolates **5b**~**5f**

Silver Vinylthiolate (**5b**)

To a solution of *N*,*N*-diisopropylamine (3.39 ml, 19.5 mmol) in dry THF (80 ml) was added 1.55 M *n*-butyllithium solution in hexane (12.6 ml) at −60°C in nitrogen atmosphere. The mixture was stirred at 0°C for 30 minutes. To this solution was added a solution of 2-ethoxy-1,3-oxathiolane (**6**)[6] (3 ml, 19.5 mmol) in dry THF (5 ml) at −60~−70°C. After additional 30 minutes at −65°C, the mixture was poured into a solution of silver nitrate (8.46 g, 49.8 mmol) in a mixture of water (20 ml) and methanol (80 ml) at 0°C. After stirring for 30 minutes in the dark, the mixture was adjusted to pH 6.5 with dilute sulfuric acid. The precipitate was collected by filtration, washed with water, methanol, and diethyl ether successively, and dried *in vacuo* to give crude silver salt **5b** (7.41 g) as a brown solid.

3-[(*Z*)-2-(Triphenylmethylthio)vinyl]pyridine (**9**)

To a solution of triphenylmethylthiol (1.41 g, 5.1 mmol) and 3-ethynylpyridine (**8**) (0.5 g, 4.8 mmol) in dry THF (10 ml) was added potassium *tert*-butoxide (571 mg, 5 mmol) at room temperature. The mixture was refluxed for 2 hours. After being cooled to room temperature, the reaction mixture was poured into ice water. The mixture was extracted with ethyl acetate. The extract was washed with saturated aqueous sodium chloride, dried over magnesium sulfate, and evaporated *in vacuo* to give a crystalline powder. The crystalline powder was washed with ethanol and dried to give **9** (1.11 g, 60%): MP 140~141℃; IR (Nujol) cm^{-1} 1590, 1560, 1470, 1450, 1410; ^1H NMR ($CDCl_3$) δ 6.03 (1H, d, J=11 Hz), 6.27 (1H, d, J=11 Hz), 7.13~7.23 (1H, m), 7.27 (15 H, m), 7.90 (1H, dt, J=2 and 8 Hz), 8.40 (1H, dd, J=2 and 5 Hz), 8.63 (1H, d, J=2 Hz).

Silver (*Z*)-2-(3-Pyridinyl)vinylthiolate (**5c**)

To a solution of **9** (690 mg, 1.81 mmol) in a mixture of THF (3 ml), methanol (5 ml), and pyridine (0.147 ml, 1.82 mmol) was added dropwise a solution of silver nitrate (371 mg, 2.18 mmol) in water (20 ml) at room temperature. The mixture was stirred at 0℃ for 1 hour in the dark. The precipitate was collected, washed with methanol, and dried over phosphorus pentoxide to give crude **5c** (487 mg) as a brown solid: IR (Nujol) cm^{-1} 1590, 1580, 1560, 1420.

1-Methyl-3-[(*Z*)-2-(triphenylmethylthio)vinyl]pyridinium Iodide (**10**)

To a solution of **9** (5 g, 13.1 mmol) in dichloromethane (100 ml) was added methyl iodide (8.3 ml, 133 mmol) at room temperature. The mixture was stirred at room temperature for 6 hours. The precipitate was collected by filtration, washed with diethyl ether, and dried *in vacuo* to give **10** (6.64 g, 97%) as a colorless solid: IR (Nujol) cm^{-1} 1570, 1500, 1490, 1445; ^1H NMR (DMSO-d_6) δ 4.33 (3H, s), 6.35 (1H, d, J=12 Hz), 6.55 (1H, d, J=12 Hz), 7.00~7.40 (15H, m), 8.40 (1H, dd, J=5 and 8 Hz), 8.57 (1H, d, J=8 Hz), 8.73 (1H, d, J=5 Hz), 8.90 (1H, s).

Silver (*Z*)-2-(1-Methyl-3-pyridinio)vinylthiolate Nitrate (**5d**)

This compound was derived from compound **10** as described for **5c** from **9**.

(*Z*)-3-Triphenylmethylthioacrylamide (**12**)

To a mixture solution of propionamide (**11**) (1.0 g, 14.5 mmol) in THF and water (1 : 1, 20 ml) was added triphenylmethylthiol (4.2 g, 15.2 mmol) at 0℃. The mixture was stirred at 0~10℃ for 30 minutes and poured into ice water. The precipitate was collected by filtration and dried over phosphorus pentoxide *in vacuo* to give **12** (4.2 g, 84%) as a colorless solid: IR (Nujol) cm^{-1} 3380, 3180, 1640, 1570.

(*Z*)-2-Triphenylmethylthioacrylonitrile (**13**)

To a mixture of **12** (3.0 g, 8.7 mmol) and DMF (40 ml) was added phosphorus pentachloride (3.65 g, 17.5 mmol). The mixture was stirred at 20℃ for 30 minutes, poured into ice water, and extracted with ethyl acetate. The organic layer was washed with saturated aqueous sodium chloride, dried over magnesium sulfate, and evaporated *in vacuo* to give **13** (2.85 g, 77%) as a solid: IR (Nujol) cm^{-1} 2200; ^1H NMR (DMSO-d_6) δ 5.65 (1H, d, J=10 Hz), 6.88 (1H, d, J=10 Hz), 7.00~7.67 (15H, m).

Silver (*Z*)-2-Cyanovinylthiolate (**5e**)

This compound was derived from compound **13** as described for **5c** from **9**.

IR (Nujol) cm^{-1} 2200, 1530.

(*E*)-2-Triphenylmethylthioacrylonitrile (**15**)

To a solution of (*E*)-2-chloroacrylonitrile (**14**) (100 mg, 1.14 mmol) in THF (2.0 ml) was added triphenylmethylthiol (0.332 g, 1.2 mmol) and triethylamine (0.175 ml, 1.26 mmol) at 0℃. The mixture was stirred at room temperature for 16 hours, poured into ice water and extracted with ethyl acetate. The extract was washed with saturated aqueous sodium chloride, dried over magnesium sulfate, and evaporated *in vacuo* to give **15** (0.37 g, 100%) as a solid: IR (Film) cm^{-1} 2210, 1560, 1490, 1440; ^1H NMR (DMSO-d_6) δ 5.26

(1H, d, J=16 Hz), 5.78 (1H, d, J=16 Hz), 7.00~7.30 (15 H, m).

Silver (*E*)-2-Cyanovinylthiolate (**5f**)

This compound was derived from compound **15** as described for **5c** from **9**.

IR (Nujol) cm^{-1} 2210, 1540, 920, 860.

General Procedure for the Synthesis of 3-Vinylthio- and 3-Vinylthiomethylcephem Compounds, **18** and **19**

To a mixture of crude silver salt **5** (20 g) and acetonitrile (90 ml) was added sodium iodide (8.77 g, 58.5 mmol) at room temperature in the dark. The mixture was stirred for 30 minutes at room temperature in dark and cooled to 0℃. To the mixture was added 3-methylsulfonyloxy- or 3-chloromethylcephem, **16**[5] or **17**[8] (9.9 mmol), at 0℃. The mixture was stirred for 30 minutes at 0℃. The precipitate was filtered off. The filterate was evaporated *in vacuo*. The residue was added to a mixture of ethyl acetate and water. After stirring, the organic layer was separated, washed with saturated aqueous sodium chloride, dried over magnesium sulfate, and evaporated *in vacuo*. The residue was purified by silica gel column chromatography to give 3-vinylthio- or 3-vinylthiomethylcephems, **18** or **19**.

General Procedure for Deformylation of Compounds **18** and **19**

To a mixture of **18** or **19** (2.0 mmol, X=CH) in methanol (25 ml) was added conc hydrochloric acid (6.0~8.0 mmol) at room temperature. The mixture was stirred at 30~35℃ for 1 hour. After neutralization with 5% aqueous sodium hydrogen carbonate, the mixture was evaporated *in vacuo* and the residue was added to a mixture of ethyl acetate and water. The organic layer was separated, washed with saturated aqueous sodium chloride, dried over magnesium sulfate, and evaporated *in vacuo*. The residue was purified by silica gel column chromatography to give 7-aminothiazol compounds **20** or **21**.

General Procedure for Deprotection of Diphenylmethyl Group of Compounds, **18~21**

To a mixture of diphenylmethyl ester (**18~21**, 1.0 g) and anisole (1.0 ml) in dichloromethane (2.0 ml) was added TFA (3.0 ml) under ice-cooling. The mixture was stirred at the same temperature for 1 hour and poured into diisopropyl ether (100 ml). The precipitate was collected by filtration and purified by column chromatography (Diaion HP-20; 30 ml, eluent; 2-propanol - water, 1 : 10) followed by freeze-drying to give **1** or **2**.

7β-[(*Z*)-2-(5-Amino-1,2,4-thiadiazol-3-yl)-2-ethoxyiminoacetamido]-3-[(*Z*)-2-(acetamido)vinylthio]-3-cephem-4-carboxylic Acid (**1a**)

MP 125℃ (dec); IR (Nujol) cm^{-1} 3300, 3250, 1770, 1670, 1610; ^{1}H NMR (DMSO-d_6) δ 1.23 (3H, t, J=7 Hz), 2.00 (3H, s), 3.50 and 3.80 (2H, ABq, J=18 Hz), 4.15 (2H, q, J=7 Hz), 5.13 (1H, d, J=5 Hz), 5.33 (1H, d, J=8 Hz), 5.70 (1H, dd, J=5 and 8 Hz), 7.14 (1H, dd, J=8 and 11 Hz), 8.00 (2H, s), 9.40 (1H, d, J=8 Hz), 9.70 (1H, d, J=11 Hz).

7β-[(*Z*)-2-(5-Amino-1,2,4-thiadiazol-3-yl)-2-ethoxyiminoacetamido]-3-vinylthio-3-cephem-4-carboxylic Acid (**1b**)

MP 145℃ (dec); IR (Nujol) cm^{-1} 1765, 1670, 1525; ^{1}H NMR (DMSO-d_6) δ 1.21 (3H, t, J=7 Hz), 3.48 and 3.84 (2H, ABq, J=18 Hz), 4.11 (2H, q, J=7 Hz), 5.12 (1H, d, J=5 Hz), 5.33 (1H, d, J=16 Hz), 5.40 (1H, d, J=10 Hz), 5.72 (1H, dd, J=5 and 8 Hz), 6.54 (1H, dd, J=10 and 16 Hz), 7.98 (2H, s), 9.42 (1H, d, J=8 Hz).

7β-[(*Z*)-2-(5-Amino-1,2,4-thiadiazol-3-yl)-2-ethoxyiminoacetamido]-3-[(*Z*)-2-(3-pyridyl)vinylthio]-3-cephem-4-carboxylic Acid (**1c-1**)

MP 171℃ (dec); IR (Nujol) cm^{-1} 3300, 3250, 1770, 1670, 1610; ^{1}H NMR (DMSO-d_6) δ 1.27 (3H, t, J=7 Hz), 3.66 and 4.12 (2H, ABq, J=18 Hz), 4.20 (2H, q, J=7 Hz), 5.23 (1H, d, J=5 Hz), 5.87 (1H, dd, J=5 and 8 Hz), 6.71 (1H, d, J=11 Hz), 6.89 (1H, d, J=11 Hz), 7.48 (1H, dd, J=5 and 7.5 Hz), 7.95 (1H, dd, J=2 and 7.5 Hz), 8.13 (2H, br s), 8.49 (1H, d, J=5 Hz), 8.68 (1H, br s), 9.58 (1H, d, J=8 Hz).

7β-[(Z)-2-(2-Aminothiazol-4-yl)-2-ethoxyiminoacetamido]-3-[(Z)-2-(3-pyridyl)vinylthio]-3-cephem-4-carboxylic Acid (**1c-2**)

IR (Nujol) cm^{-1} 1770, 1660, 1530; ^1H NMR (DMSO-d_6) δ 1.27 (3H, t, J=7 Hz), 3.50~4.40 (4H, m), 5.27 (1H, d, J=5 Hz), 5.87 (1H, dd, J=5 and 8 Hz), 6.77 (1H, s), 6.80 (1H, d, J=12 Hz), 6.93 (1H, d, J=12 Hz), 7.50~7.83 (1H, m), 7.83~8.50 (3H, m), 8.50~8.90 (2H, m), 9.67 (1H, d, J=8 Hz).

7β-[(Z)-2-Allyloxyimino-2-(2-aminothiazol-4-yl)acetamido]-3-[(Z)-2-(3-pyridyl)vinylthio]-3-cephem-4-carboxylic Acid (**1c-3**)

IR (Nujol) cm^{-1} 1760, 1630, 1530; ^1H NMR (DMSO-d_6) δ 3.62 and 4.17 (2H, ABq, J=18 Hz), 4.67 (2H, m), 4.90~5.60 (2H, m), 5.20 (1H, d, J=5 Hz), 5.67~6.10 (1H, m), 5.90 (1H, dd, J=5 and 8 Hz), 6.80 (1H, s), 7.10~7.60 (1H, m), 7.70~8.00 (1H, m), 8.10 (2H, brs), 8.47 (1H, m), 8.67 (1H, brs), 9.63 (1H, d, J=8 Hz).

7β-[(Z)-2-(5-Amino-1,2,4-thiadiazol-3-yl)-2-ethoxyiminoacetamido]-3-[(Z)-2-(1-methyl-3-pyridinio)-vinylthio]-3-cephem-4-carboxylate (**1d-1**)

IR (Nujol) cm^{-1} 1775, 1670, 1605, 1520; ^1H NMR (DMSO-d_6) δ 1.27 (3H, t, J=7 Hz), 3.20~3.80 (2H, m), 3.90~4.50 (2H, m), 4.37 (3H, s), 5.07 (1H, d, J=5 Hz), 5.60 (1H, dd, J=5 and 8 Hz), 6.65 (1H, d, J=10 Hz), 7.05 (1H, d, J=10 Hz), 7.80~9.00 (5H, m), 9.20 (1H, brs), 9.40 (1H, d, J=8 Hz).

7β-[(Z)-2-(2-Aminothiazol-4-yl)-2-ethoxyiminoacetamido]-3-[(Z)-2-(1-methyl-3-pyridinio)vinylthio]-3-cephem-4-carboxylate (**1d-2**)

IR (Nujol) cm^{-1} 1760, 1650, 1600, 1520; ^1H NMR (DMSO-d_6) δ 1.20 (3H, t, J=7 Hz), 3.00~3.80 (2H, m), 4.13 (2H, q, J=7 Hz), 4.37 (3H, s), 5.07 (1H, d, J=5 Hz), 5.60 (1H, dd, J=5 and 8 Hz), 6.62 (1H, d, J=10 Hz), 6.67 (1H, s), 7.02 (1H, d, J=10 Hz), 7.27 (2H, brs), 7.80~8.20 (1H, m), 8.30~8.60 (1H, m), 8.77 (1H, d, J=6 Hz), 9.27 (1H, brs), 9.43 (1H, d, J=8 Hz).

7β-[(Z)-2-Allyloxyimino-2-(5-amino-1,2,4-thiadiazol-3-yl)acetamido]-3-[(Z)-2-cyanovinylthio]-3-cephem-4-carboxylic Acid (**1e**)

IR (Nujol) cm^{-1} 2200, 1760, 1660, 1605, 1510; ^1H NMR (DMSO-d_6) δ 3.68 and 4.12 (2H, ABq, J=18 Hz), 4.67 (2H, m), 5.00~5.50 (3H, m), 5.60~6.20 (2H, m), 5.90 (1H, d, J=10 Hz), 7.77 (1H, d, J=10 Hz), 8.10 (2H, brs), 9.60 (1H, d, J=8 Hz).

7β-[(Z)-2-(5-Amino-1,2,4-thiadiazol-3-yl)-2-ethoxyiminoacetamido]-3-[(Z)-2-(3-pyridyl)vinylthiomethyl]-3-cephem-4-carboxylic Acid (**2c-1**)

MP 156~160℃ (dec); IR (Nujol) cm^{-1} 3300, 3200, 1770, 1670, 1620, 1580, 1530, 1250, 1230, 1180; ^1H NMR (D_2O) δ 1.23 (3H, t, J=7 Hz), 3.27~3.66 (2H, m), 3.66~4.03 (2H, m), 4.27 (2H, q, J=7 Hz), 5.20 (1H, d, J=5 Hz), 5.87 (1H, d, J=5 Hz), 6.30 (1H, d, J=11 Hz), 6.67 (1H, d, J=11 Hz), 7.10~7.40 (1H, m), 7.60~7.80 (1H, m), 8.07~8.30 (1H, m), 8.30~8.50 (1H, m).

7β-[(Z)-2-Allyloxyimino-2-(5-amino-1,2,4-thiadiazol-3-yl)acetamido]-3-[(Z)-2-(3-pyridyl)vinylthiomethyl]-3-cephem-4-carboxylic Acid (**2c-2**)

IR (Nujol) cm^{-1} 1770, 1670, 1620, 1580, 1530, 1400; ^1H NMR (DMSO-d_6) δ 3.60 (2H, brs), 3.60~4.30 (2H, m), 4.63 (2H, m), 5.00~5.50 (2H, m), 5.20 (1H, d, J=5 Hz), 5.60~6.30 (1H, m), 5.80 (1H, dd, J=5 and 8 Hz), 6.47 (1H, d, J=11 Hz), 6.83 (1H, d, J=11 Hz), 7.47 (1H, m), 7.93 (1H, m), 8.10 (2H, brs), 8.43 (1H, m), 8.63 (1H, m), 9.57 (1H, d, J=8 Hz).

7β-[(Z)-2-(5-Amino-1,2,4-thiadiazol-3-yl)-2-ethoxyiminoacetamido]-3-[(Z)-2-(1-methyl-3-pyridinio)-vinylthiomethyl]-3-cephem-4-carboxylate (**2d**)

IR (Nujol) cm^{-1} 1755, 1660, 1590; ^1H NMR (D_2O) δ 1.30 (3H, t, J=7 Hz), 3.47~4.57 (4H, m), 4.37 (3H, s), 4.73 (2H, q, J=7 Hz), 5.30 (1H, d, J=5 Hz), 5.80 (1H, d, J=5 Hz), 6.50 (1H, d, J=11 Hz), 7.03 (1H, d, J=11 Hz), 7.77~8.13 (1H, m), 8.30~8.80 (3H, m).

7β-[(Z)-2-Allyloxyimino-2-(5-amino-1,2,4-thiadiazol-3-yl)acetamido]-3-[(Z)-2-cyanovinylthiomethyl]-3-cephem-4-carboxylic Acid (**2e-1**)

MP 166℃ (dec); IR (Nujol) cm^{-1} 3250, 2210, 1765, 1670, 1615; ^1H NMR (DMSO-d_6) δ 3.60 (2H, brs), 3.80 and 4.25 (2H, ABq, J=14 Hz), 4.67 (2H, m), 5.20 (1H, d, J=5 Hz), 5.17~6.20 (3H, m), 5.70 (1H, d, J=11 Hz), 5.77 (1H, dd, J=5 and 8 Hz), 7.75 (1H, d, J=11 Hz), 8.10 (2H, brs), 9.50 (1H, d, J=8 Hz).

7β-[(Z)-2-Allyloxyimino-2-(2-aminothiazol-4-yl)acetamido]-3-[(Z)-2-cyanovinylthiomethyl]-3-cephem-4-carboxylic Acid (**2e-2**)

MP 148℃ (dec); IR (Nujol) cm^{-1} 3300, 2220, 1770, 1670, 1620; ^1H NMR (DMSO-d_6) δ 3.53 (2H, brs), 3.60 and 4.20 (2H, ABq, J=13 Hz), 4.57 (2H, m), 5.10~6.20 (4H, m), 5.17 (1H, d, J=5 Hz), 5.67 (1H, d, J=11 Hz), 6.70 (1H, s), 7.70 (1H, d, J=11 Hz), 9.60 (1H, d, J=8 Hz).

7β-[(Z)-2-Allyloxyimino-2-(5-amino-1,2,4-thiadiazol-3-yl)acetamido]-3-[(E)-2-cyanovinylthiomethyl]-3-cephem-4-carboxylic Acid (**2f**)

MP 136℃ (dec); IR (Nujol) cm^{-1} 3300, 2220, 1770, 1675, 1620; ^1H NMR (DMSO-d_6) δ 3.35~4.33 (4H, m), 4.69 (2H, d, J=5 Hz), 5.15~6.20 (4H, m), 5.22 (1H, d, J=5 Hz), 5.75 (1H, d, J=15 Hz), 7.91 (1H, d, J=15 Hz), 8.15 (2H, brs), 9.67 (1H, d, J=8 Hz).

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