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# A Synthesis of New 3-Dialkoxyphosphinylmethyl and 3-Dihydroxyphosphinylmethyl Cephalosporins

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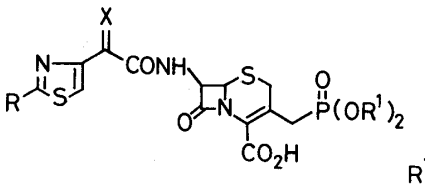
The syntheses and the antibacterial activities of new 3-dimethoxyphosphinylmethyl and 3-dihydroxyphosphinylmethyl cephalosporins I-(*Z*), II-(*Z*), III-(*Z*) and III-(*E*), possessing the chloromethylene or methoxyimino substituent at the α-position to the 7-(2-aminothiazol-4-yl)acetamido or 7-(thiazol-4-yl)acetamido moiety of the cephem nucleus, are described. The key steps of these syntheses were the Michaelis–Arbusov reaction of the 3-halomethylcephem **1** with trimethyl phosphite and the dealkylation reactions of both the dimethoxyphosphinyl group and the *p*-methoxybenzyl ester of **7a**,**b**-(*Z*) by treatment with bromotrimethylsilane to afford **9a**,**b**-(*Z*).

**Keywords**——3-dimethoxyphosphinylmethyl cephalosporin; 3-dihydroxyphosphinylmethyl cephalosporin; Michaelis–Arbusov reaction; trimethyl phosphite; 3-chloromethylcephem; (*Z*)-3-chloro-2-(2-formylaminothiazol-4-yl)propenoic acid; (*Z*)-2-(2-formylaminothiazol-4-yl)-2-methoxyiminoacetic acid; (*E*)-3-chloro-2-(thiazol-4-yl)propenoic acid; bromotrimethylsilane; *N*-methyl-*N*-trimethylsilyltrifluoroacetamide

Phosphonic acid derivatives have interesting biological activities.[1] There have been extensive studies on the introduction of a dialkoxy- or dihydroxy-substituted phosphinyl moiety, $-P(O)(OR)_2$, into β-lactams such as penicillins,[2] cephalosporins,[3] carbapenems,[4] and monobactams.[5]

Continuing our efforts to develop novel cephalosporins possessing the (*Z*)-2-(2-aminothiazol-4-yl)-3-chloropropenamido group at the 7-position on the cephem nucleus,[6] we focused our attention on the introduction of a substituted phosphinyl moiety as a hydrophilic function into the 3-methyl group of these cephems. We report here the syntheses and the antibacterial activities of new (*Z*)-2-(2-aminothiazol-4-yl)-3-chloropropenamido (I-(*Z*)), (*Z*)-2-(2-aminothiazol-4-yl)-2-methoxyiminoacetamido (II-(*Z*)) and 3-chloro-2-(thiazol-4-yl)-propenamido (III-(*Z*) and (*E*)) cephalosporins substituted with the 3-dialkoxyphosphinylmethyl or 3-dihydroxyphosphinylmethyl group (Fig. 1).

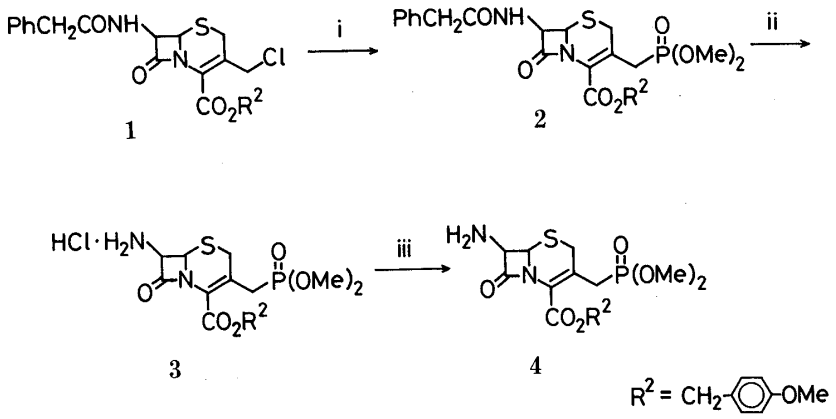
The Michaelis–Arbusov reaction of the 3-chloromethylcephem **1**[7] with 2 molar equivalents of trimethyl phosphite in the presence of sodium iodide in acetone at room temperature gave the 3-dimethoxyphosphinylmethylcephem **2** in moderate yield.[8] Cleavage of the 7-




$R^1$ = Me and H

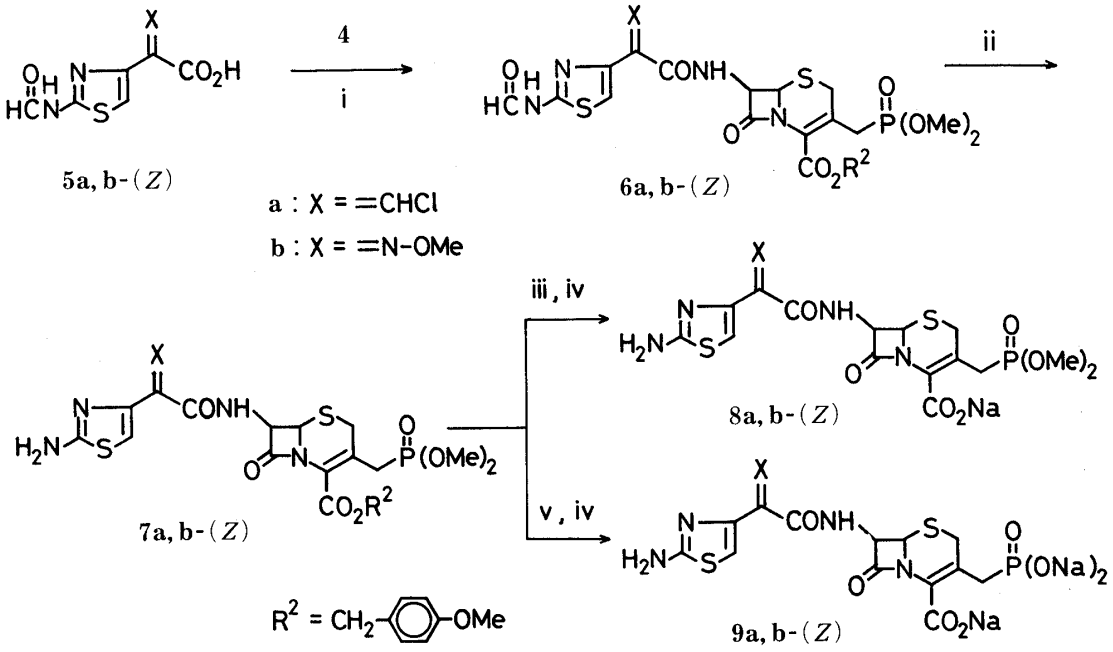
I-(*Z*): R = $NH_2$, X = =CHCl
II-(*Z*): R = $NH_2$, X = =N-OMe
III-(*Z*) and (*E*): R = H, X = =CHCl

Fig. 1




i) NaI, $P(OMe)_3$ ii) $PCl_5$–pyridine, MeOH iii) aq. $NaHCO_3$

Chart 1



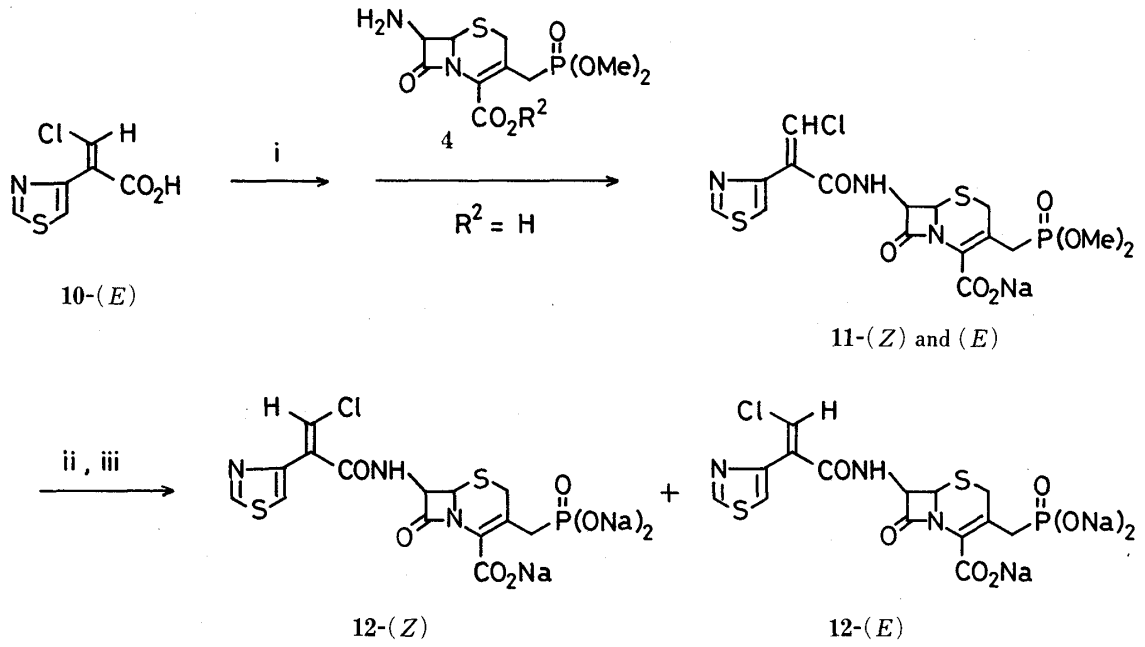

i) DCC, (HOBT) ii) MeOH–HCl iii) $CF_3CO_2H$ iv) aq. $NaHCO_3$ v) MSTFA–$Me_3SiBr$

Chart 2

phenylacetamido group of **2** by the imino-chloride method[9] afforded the 7-amino-3-dimethoxyphosphinylmethylcephem hydrochloride **3** in 89% yield. The free amine **4** was obtained in 89% yield by neutralization of the salt **3** with aqueous sodium hydrogen carbonate. Although the synthesis of the 7-thienylacetamido cephem substituted with a 3-dimethoxyphosphinylmethyl group was disclosed in a patent,[8] there is no precedent for these 7-aminocephem derivatives **3** and **4**, which are key intermediates for the synthesis of various 7-acylated cephems (see Chart 1).

Thus, the following semisynthetic cephalosporins were prepared by acylation of **4**, as outlined in Charts 2 and 3.

First, the condensation of the amine **4** with (*Z*)-3-chloro-2-(2-formylaminothiazol-4-yl)propenoic acid (**5a**)[6a] was carried out using *N*,*N*′-dicyclohexylcarbodiimide (DCC) as the condensing agent to give the cephem **6a**-(*Z*) in 86% yield. The N-formyl group of **6a**-(*Z*) was




i) $POCl_3$–DMF, $\Delta$ ii) a) HCl b) MSTFA–$Me_3SiBr$ iii) aq. $NaHCO_3$

Chart 3

removed by treatment with concentrated hydrochloric acid to give **7a**-(*Z*) in 72% yield. Cleavage of the *p*-methoxybenzyl group of **7a**-(*Z*) with trifluoroacetic acid, followed by neutralization with aqueous sodium hydrogen carbonate, purification by reversed-phase column chromatography and lyophilization afforded **8a**-(*Z*) in 93% yield.

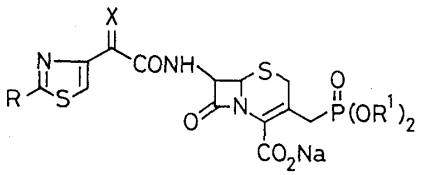
For the transformation of the dimethoxyphosphinyl derivative **8a**-(*Z*) into the desired dihydroxyphosphinyl cephem **9a**-(*Z*), we expected that iodotrimethylsilane[10] could be useful for the dealkylation reaction. Treatment of the acid form of **8a**-(*Z*) with iodotrimethylsilane in acetonitrile at 0 °C to room temperature was unsuccessful due to the high reactivity of the iodosilane. The Glaxo research group[11] has reported a facile dealkylation method for dialkoxyphosphinyl groups in connection with the synthesis of 3-dihydroxyphosphinylcarbamoyloxymethyl cephems by using bromotrimethylsilane[12] and trimethylsilyl urethane as an acid scavenger.[13] We applied this method for the dealkylation of **7a**-(*Z*) by replacing the silyl urethane with *N*-methyl-*N*-trimethylsilyltrifluoroacetamide (MSTFA). Treatment of **7a**-(*Z*) with bromotrimethylsilane in the presence of MSTFA afforded **9a**-(*Z*) in 50% yield after purification by reversed-phase chromatography and lyophilization, thus indicating that the dealkylation reactions of both the dimethoxyphosphinyl group and the *p*-methoxybenzyl ester could be attained in a single operation.

We planned next to synthesize the oxyimino cephalosporins **8b**-(*Z*) and **9b**-(*Z*). A search of the literature indicated, surprisingly, that these compounds having the 3-dialkoxyphosphinylmethyl or 3-dihydroxyphosphinylmethyl group are unknown, although various other cephem derivatives substituted with the (*Z*)-2-(2-aminothiazol-4-yl)-2-methoxyiminoacetamido group at the 7-position are known.

Condensation reaction of (*Z*)-2-(2-formylaminothiazol-4-yl)-2-methoxyiminoacetic acid (**5b**-(*Z*))[14] with the amine **4** using DCC proceeded to give **6b**-(*Z*) in 79% yield. The yield of **6b**-(*Z*) was improved to 96% by using DCC and HOBT (1-hydroxybenzotriazole) as condensing agents. Stepwise removal of the protective groups of **6b**-(*Z*) employing procedures similar to those described for **6a**-(*Z*) gave **8b**-(*Z*) (82% overall yield from **6b**-(*Z*)) and **9b**-(*Z*) (49% overall yield from **6b**-(*Z*)).

Finally, we intended to synthesize the α-chloromethylenethiazolylacetamido cephalos-

TABLE I. Antibacterial Activities (MIC, μg/ml) of 3-Dimethoxyphosphinylmethyl and 3-Dihydroxyphosphinylmethyl Cephalosporins



| X | R | $R^1$ | No. | *S. aureus* 209-P | *E. coli* NIHJ JC-2 | *K. pneumoniae* 8045 | *P. vulgaris* 6897 | *E. cloacae* F1510 |
|---|---|---|---|---|---|---|---|---|
| =CHCl | $NH_2$ | Me | **8a**-(*Z*) | 3.13 | 3.13 | 12.5 | 0.39 | 0.39 |
| =CHCl | $NH_2$ | Na | **9a**-(*Z*) | >100 | 100 | 100 | 25 | >100 |
| =N–OMe | $NH_2$ | Me | **8b**-(*Z*) | 12.5 | 0.39 | 1.56 | 0.01 | 0.1 |
| =N–OMe | $NH_2$ | Na | **9b**-(*Z*) | >100 | 100 | 50 | 12.5 | >100 |
| =CHCl | H | Na | **12**-(*Z*) | >100 | >100 | >100 | >100 | >100 |

MIC: minimum inhibitory concentration.

porins **12**-(*Z*) and **12**-(*E*). (*E*)-3-Chloro-2-(thiazol-4-yl)propenoic acid (**10**-(*E*))[15] was activated with Vilsmeier reagent and isomerized by heating as described in our preceding paper, and then treated with sodium 7-amino-3-dimethoxyphosphinylmethyl-3-cephem-4-carboxylate (derived from the *p*-methoxybenzyl ester **4**). Usual work-up followed by reversed-phase chromatography gave **11**-(*Z*) and **11**-(*E*) in 16% and 35% yields, respectively. The dealkylation reactions of the dimethoxyphosphinyl groups of **11**-(*Z*) and **11**-(*E*) afforded the desired **12**-(*Z*) and **12**-(*E*) in 27% and 63% yields, respectively.

The antibacterial activities of the synthesized 3-dimethoxyphosphinylmethyl and 3-dihydroxyphosphinylmethyl cephalosporins are summarized in Table I.

All compounds were less active than recently commercialized cephalosporins against the microorganisms tested. The 3-dihydroxyphosphinylmethyl cephems **9a**-(*Z*) and **9b**-(*Z*) were unexpectedly less active than the corresponding 3-dimethoxyphosphinylmethyl cephems **8a**-(*Z*) and **8b**-(*Z*). Antibacterial activities of substituted phosphinyl cephem derivatives have rarely been reported, though it was stated in a patent that a 3-diethoxyphosphinyl cephem is less active than the corresponding 3-dihydroxyphosphinyl derivative against *S. subtilis*.[3b] Antibacterial activities of α-oxyimino cephems **8b**-(*Z*) and **9b**-(*Z*) were generally higher than those of α-chloromethylene cephems **8a**-(*Z*) and **9a**-(*Z*) against gram-negative bacteria.

## Experimental

The instruments and experimental techniques were generally as described in the preceding paper. Additionally, $^{31}$P-nuclear magnetic resonance ($^{31}$P-NMR) spectra were recorded on a Varian XL-100A spectrometer at 40.5 MHz with 85% phosphoric acid as the external standard.

***p*-Methoxybenzyl 3-Dimethoxyphosphinylmethyl-7-phenylacetamido-3-cephem-4-carboxylate (2)**——Sodium iodide (0.160 g, 1.1 mmol) was added to a suspension of *p*-methoxybenzyl 3-chloromethyl-7-phenylacetamido-3-cephem-4-carboxylate (**1**)[7] (0.486 g, 1.0 mmol) in acetone (10 ml), and the reaction mixture was stirred for 1 h at room temperature. After addition of trimethyl phosphite (0.29 ml, 2.2 mmol), the reaction mixture was left for 5 d at that temperature. Filtration with filter paper (No. 5C, Toyo) and concentration of the filtrate *in vacuo* gave a residue. This residue was subjected to preparative thin layer chromatography to give **2** (0.304 g, 54%). mp 150—151 °C (AcOEt–$Et_2O$). IR (neat): 3275, 1780, 1725, 1680, 1515, 1360, 1305, 1250, 1175, 1160, 1050, 1030, 825 cm$^{-1}$. $^1$H-NMR ($CDCl_3$) $\delta$: 3.01 (1H, d of ABd, $J_{P-H}$=22.9 and $J_{H-H}$=14.6 Hz), 3.45 (1H, ABd, $J_{H-H}$=18.2 and $J_{P-H}$=3.0 Hz), 3.47 (1H, ABd, $J_{P-H}$=22.9 and $J_{H-H}$=14.6 Hz), 3.62 (1H, d of ABd, $J_{H-H}$=18.2 and $J_{P-H}$=5.2 Hz), 3.63 (2H, br s), 3.667 (3H, d, $J_{P-H}$=11.0 Hz), 3.671 (3H, d, $J_{P-H}$=11.0 Hz), 3.79 (3H, s), 4.93 (1H, dd, $J_{H-H}$=4.8 and $J_{P-H}$=1.0 Hz), 5.14 (1H, ABd, $J$=11.8 Hz), 5.20 (1H, ABd, $J$=11.8 Hz), 5.77 (1H, dd, $J$=9.1 and 4.8 Hz), 6.57 (1H, d, $J$=9.1 Hz, NH), 6.87

(2H, AA′BB′, $J$=8.7 Hz), 7.32 (2H, AA′BB′, $J$=8.7 Hz), 7.26—7.37 (5H, m). $^{31}$P-NMR ($CDCl_3$): 26.1 ppm. MS $m/z$ (relative intensity %): 560 ($M^+$, 2.5), 440 (20), 439 (88), 396 (12), 395 (47), 387 (15), 386 (76), 385 (15), 368 (10), 367 (34), 303 (11), 264 (15), 249 (14), 222 (19), 221 (13), 220 (75), 176 (22), 122 (56), 121 (100). *Anal.* Calcd for $C_{26}H_{29}N_2O_8PS$: C, 55.71; H, 5.21; N, 5.00; S, 5.72. Found: C, 55.46; H, 5.16; N, 4.90; S, 6.09.

***p*-Methoxybenzyl 7-Amino-3-dimethoxyphosphinylmethyl-3-cephem-4-carboxylate Hydrochloride (3)**——Compound **2** (10.0 g, 0.018 mol) was added to a mixture of phosphorus pentachloride (11.22 g, 0.054 mol) and pyridine (4.4 ml, 0.054 mmol) in dichloromethane (110 ml) at 0 °C, and the reaction mixture was stirred for 1.5 h. Methanol (100 ml) was added under cooling at −50 °C, then the temperature of the reaction mixture was allowed to rise to −10 °C immediately, and was kept between −15 and −10 °C for 2 h. Colorless crystals precipitated. After addition of water (20 ml) to this suspension, the solvent was removed and diethyl ether (50 ml) was added. The precipitated crystals were collected by filtration and dried over phosphorus pentoxide to give **3** (7.589 g, 89%) as colorless prisms. mp 172—173 °C (dec.) (MeOH–$Et_2O$). IR (KBr): 1775, 1715, 1610, 1515, 1395, 1260, 1215, 1050, 840 cm$^{-1}$. $^1$H-NMR (DMSO-$d_6$) $\delta$: 3.30 (1H, d of ABd, $J_{P-H}$=22.9 and $J_{H-H}$=14.4 Hz), 3.49 (1H, d of ABd, $J_{P-H}$=24.3 and $J_{H-H}$=14.4 Hz), 3.59 (3H, d, $J_{P-H}$=11.0 Hz), 3.60 (3H, d, $J_{P-H}$=11.0 Hz), 3.64 (1H, d of ABd, $J_{P-H}$=4.2 and $J_{H-H}$=17.5 Hz), 3.74 (1H, d of ABd, $J_{P-H}$=3.1 and $J_{H-H}$=17.5 Hz), 3.75 (3H, s), 5.12 (1H, d, $J$=4.9 Hz), 5.14 (1H, ABd, $J$=12.1 Hz), 5.20 (1H, ABd, $J$=12.1 Hz), 5.25 (1H, d, $J$=4.9 Hz), 6.93 (2H, AA′BB′, $J$=8.7 Hz), 7.45 (2H, AA′BB′, $J$=8.7 Hz), 9.1 (3H, br s, $NH_3Cl$). MS $m/z$ (relative intensity %): 442 ($M^+$ − HCl, 4), 387 (7), 386 (40), 368 (8), 293 (7), 249 (8), 222 (13), 122 (11), 121 (100), 96 (9). *Anal.* Calcd for $C_{18}H_{23}N_2O_7PS \cdot HCl$: C, 45.15; H, 5.05; N, 5.85; Cl, 7.40. Found: C, 44.95; H, 5.16; N, 5.83; Cl, 7.22.

***p*-Methoxybenzyl 7-Amino-3-dimethoxyphosphinylmethyl-3-cephem-4-carboxylate (4)**——Compound **3** (6.00 g, 0.013 mmol) was dissolved in water (50 ml) and neutralized with saturated aqueous sodium hydrogen carbonate, extracted with ethyl acetate, and then dried over sodium sulfate. Removal of the organic solvent *in vacuo* gave **4** (4.93 g, 89%) as a colorless amorphous solid. $^1$H-NMR ($CDCl_3$) $\delta$: 1.9 (2H, br s, $NH_2$), 2.6—4.3 (4H, m), 3.70 (6H, d, $J$=11.0 Hz), 3.80 (3H, s), 4.73 (1H, br d, $J$=8 Hz), 4.96 (1H, br d, $J$=8 Hz), 5.23 (2H, br s), 6.91 (2H, d, $J$=8.8 Hz), 7.40 (2H, d, $J$=8.8 Hz).

The free amine **4** obtained here was successively used in the following condensation reactions.

***p*-Methoxybenzyl 7-[(*Z*)-3-Chloro-2-(2-formylaminothiazol-4-yl)propenamido]-3-dimethoxyphosphinylmethyl-3-cephem-4-carboxylate (6a-(*Z*))**——DCC (2.56 g, 12.4 mmol) was added to a solution of **4** (3.28 g, 7.4 mmol) and (*Z*)-3-chloro-2-(2-formylaminothiazol-4-yl)propenoic acid (**5a**-(*Z*))[6a] (2.24 g, 4.6 mmol) in tetrahydrofuran (THF) (60 ml), and the reaction mixture was stirred at room temperature for 2 h. After filtration of the reaction mixture, the filtrate was concentrated. The residue was purified by silica gel column chromatography to give **6a**-(*Z*) (4.16 g, 86%) as pale yellow prisms after recrystallization. mp 121—123 °C (AcOEt–$Et_2O$). IR (KBr): 1785, 1720, 1680, 1550, 1510, 1360, 1245, 1030, 825 cm$^{-1}$. $^1$H-NMR ($CDCl_3$) $\delta$: 3.09 (1H, d of ABd, $J_{P-H}$=22.9 and $J_{H-H}$=14.7 Hz), 3.52 (1H, d of ABd, $J_{P-H}$=4.5 and $J_{H-H}$=15.7 Hz), 3.55 (1H, d of ABd, $J_{P-H}$=24.6 and $J_{H-H}$=14.7 Hz), 3.70 (3H, d, $J_{P-H}$=11.0 Hz), 3.71 (3H, d, $J_{P-H}$=11.0 Hz), 3.75 (1H, d of ABd, $J_{P-H}$=5.4 and $J_{H-H}$=15.7 Hz), 3.82 (3H, s), 5.14 (1H, dd, $J_{P-H}$=1.0 and $J_{H-H}$=4.7 Hz), 5.22 (1H, ABd, $J$=12.0 Hz), 5.23 (1H, ABd, $J$=12.0 Hz), 5.95 (1H, dd, $J$=8.3 and 4.7 Hz), 6.85 (1H, s), 6.90 (2H, AA′BB′, $J$=8.7 Hz), 7.09 (1H, s), 7.35 (2H, AA′BB′, $J$=8.7 Hz), 7.92 (1H, d, $J$=8.3 Hz, NH), 8.51 (s, 1H), 10.15 (br s, 1H, NHCHO). MS (SIMS) $m/z$: 657 ($M^+$+1). *Anal.* Calcd for $C_{25}H_{26}ClN_4O_9PS_2 \cdot H_2O$: C, 44.48; H, 4.18; Cl, 5.25; N, 8.30. Found: C, 44.75; H, 4.13; Cl, 5.50; N, 8.32.

***p*-Methoxybenzyl 7-[(*Z*)-2-(2-Aminothiazol-4-yl)-3-chloropropenamido]-3-dimethoxyphosphinylmethyl-3-cephem-4-carboxylate (7a-(*Z*))**——Concentrated hydrochloric acid (1.8 ml) was added to a solution of **6a**-(*Z*) (3.09 g, 4.7 mmol) in methanol (47 ml) at 0 °C. The mixture was stirred for 5 h at room temperature, the precipitate was collected by filtration and the filtrate was concentrated under reduced pressure. The resulting precipitate was collected again. The combined precipitate was neutralized with aqueous sodium hydrogen carbonate and extracted with ethyl acetate. Work-up of the organic extract in the usual manner gave **7a**-(*Z*) (2.13 g, 72%). mp 108—110 °C (AcOEt–$Et_2O$). IR (KBr): 1780, 1715, 1660, 1250, 1170, 1050, 1015, 820 cm$^{-1}$. $^1$H-NMR (DMSO-$d_6$) $\delta$: 3.11 (1H, d of ABd, $J_{P-H}$=22.6 and $J_{H-H}$=14.6 Hz), 3.47 (1H, d of ABd, $J_{P-H}$=24.0 and $J_{H-H}$=14.6 Hz), 3.58 (3H, d, $J_{P-H}$=11.0 Hz), 3.59 (3H, d, $J_{P-H}$=11.0 Hz), 3.59 (1H, d of ABd, $J_{P-H}$=4.0 and $J_{H-H}$=18.0 Hz), 3.64 (1H, d of ABd, $J_{P-H}$=5.0 and $J_{H-H}$=18.0 Hz), 3.75 (3H, s), 5.13 (1H, ABd, $J$=12.1 Hz), 5.20 (1H, ABd, $J$=12.1 Hz), 5.23 (1H, dd, $J_{P-H}$=1.0 and $J_{H-H}$=4.8 Hz), 5.77 (1H, dd, $J$=8.0 and 4.8 Hz), 6.40 (1H, s), 6.84 (1H, s), 6.94 (2H, AA′BB′, $J$=8.7 Hz), 7.17 (2H, br s, $NH_2$), 7.36 (2H, AA′BB′, $J$=8.7 Hz), 9.59 (1H, d, $J$=8.0 Hz, NH). MS (SIMS) $m/z$: 629 ($M^+$+1). *Anal.* Calcd for $C_{24}H_{26}ClN_4O_8PS_2$: C, 45.83; H, 4.17; Cl, 5.64; N, 8.91. Found: C, 45.61; H, 4.47; Cl, 5.67; N, 8.58.

**Sodium 7-[(*Z*)-2-(2-Aminothiazol-4-yl)-3-chloropropenamido]-3-dimethoxyphosphinylmethyl-3-cephem-4-carboxylate (8a-(*Z*))**——Compound **7a**-(*Z*) (1.21 g, 1.8 mmol) was dissolved in trifluoroacetic acid (5 ml). The solution was stirred at room temperature for 2 h, then concentration *in vacuo* and trituration with diethyl ether gave 7-[(*Z*)-2-(2-aminothiazole-4-yl)-3-chloropropenamido]-3-dimethoxyphosphinylmethyl-3-cephem-4-carboxylic acid (0.840 g, 96%) as yellow crystals, which were neutralized with saturated aqueous sodium hydrogen carbonate and purified by reversed-phase silica gel column chromatography (Lobar RP-8 column). Lyophilization of the eluate gave **8a**-(*Z*) (280 mg, 29%). IR (KBr): 1760, 1660, 1600, 1530, 1400, 1365, 1230, 1180, 1050, 1030, 820 cm$^{-1}$. $^1$H-NMR (DMSO-$d_6$) $\delta$: 3.01 (1H, d of ABd, $J_{P-H}$=22.1 and $J_{H-H}$=14.7 Hz), 3.30 (1H, d of ABd, $J_{P-H}$=2.7 and $J_{H-H}$=

17.3 Hz), 3.52 (1H, d of ABd, $J_{P-H}=5.0$ and $J_{H-H}=17.3$ Hz), 3.60 (3H, d, $J_{P-H}=10.8$ Hz), 3.61 (3H, d, $J_{P-H}=10.8$ Hz), 3.89 (1H, d of ABd, $J_{P-H}=21.6$ and $J_{H-H}=14.7$ Hz), 5.01 (1H, d, $J=4.7$ Hz), 5.53 (1H, dd, $J=8.0$ and 4.7 Hz), 6.41 (1H, s), 6.82 (1H, s), 7.16 (2H, br s, $NH_2$), 9.48 (1H, d, $J=8.0$ Hz, NH). MS (SIMS) *m/z*: 531 ($M^+ + 1$).

**7-[(*Z*)-2-(2-Aminothiazol-4-yl)-3-chloropropenamido]-3-dihydroxyphosphinylmethyl-3-cephem-4-carboxylic Acid Trisodium Salt (9a-(*Z*))**——MSTFA (0.80 ml, 4.3 mmol) and bromotrimethylsilane (2.0 ml, 15 mmol) were added to a suspension of **7a**-(*Z*) (292 mg, 0.46 mmol) in dichloromethane (6 ml) at 0 °C. After being stirred at 40 °C for 1 h, the reaction mixture was poured into water (100 ml) and neutralized with saturated aqueous sodium hydrogen carbonate. The mixture was washed with dichloromethane (3 lots of 50 ml) and then with ethyl acetate (2 lots of 50 ml). The aqueous layer was concentrated *in vacuo* and subjected to reversed-phase chromatography on a Diaion HP-20 column. Lyophilization of the eluate gave **9a**-(*Z*) (125 mg, 50%) as a colorless amorphous solid. IR (KBr): 1755, 1660 (sh), 1600, 1535, 1405, 1370, 1160, 1060 $cm^{-1}$. $^1$H-NMR ($D_2O$) $\delta$: 2.61 (1H, d of ABd, $J_{P-H}=21.8$ and $J_{H-H}=14.3$ Hz), 2.69 (1H, d of ABd, $J_{P-H}=20.2$ and $J_{H-H}=14.3$ Hz), 3.41 (1H, d of ABd, $J_{P-H}=2.6$ and $J_{H-H}=18.0$ Hz), 3.63 (1H, d of ABd, $J_{P-H}=4.6$ and $J_{H-H}=18.0$ Hz), 5.13 (1H, d, $J=4.5$ Hz), 5.70 (1H, d, $J=4.5$ Hz), 6.58 (1H, s), 6.88 (1H, s). MS (SIMS) *m/z*: 547 ($M^+ + 1$). *Anal.* Calcd for $C_{14}H_{11}ClN_4Na_3O_7PS_2 \cdot H_2O$: C, 29.77; H, 2.32; N, 9.92. Found: C, 29.39; H, 2.80; N, 9.70.

***p*-Methoxybenzyl 3-Dimethoxyphosphinylmethyl-7-[(*Z*)-2-(2-formylaminothiazol-4-yl)-2-methoxyiminoacetamido]-3-cephem-4-carboxylate (6b-(*Z*))**——DCC (2.15 g, 10.4 mmol) and HOBT (1.11 g, 8.2 mmol) were added to a solution of 4 (3.30 g, 7.5 mmol) and (*Z*)-2-(2-formylaminothiazol-4-yl)-2-methoxyiminoacetic acid (**5b**-(*Z*))[14] (1.88 g, 8.2 mmol) in THF (140 ml). The reaction mixture was stirred at room temperature for 2 h. The same work-up as described for **6a**-(*Z*) gave **6b**-(*Z*) (4.7 g, 96%). mp 127—129 °C (AcOEt–$Et_2O$). IR (KBr): 1790, 1725, 1695 (sh), 1685, 1550, 1250, 1040 $cm^{-1}$. $^1$H-NMR (DMSO-$d_6$) $\delta$: 3.11 (1H, d of ABd, $J_{P-H}=22.6$ and $J_{H-H}=14.7$ Hz), 3.47 (1H, d of ABd, $J_{P-H}=23.9$ and $J_{H-H}=14.7$ Hz), 3.58 (3H, d, $J_{P-H}=11.0$ Hz), 3.59 (3H, d, $J_{P-H}=11.0$ Hz), 3.59 (1H, d of ABd, $J_{P-H}=3.0$ and $J_{H-H}=18.4$ Hz), 3.67 (1H, d of ABd, $J_{P-H}=5.4$ and $J_{H-H}=18.4$ Hz), 3.75 (3H, s), 3.89 (3H, s), 5.12 (1H, ABd, $J=12.1$ Hz), 5.20 (1H, ABd, $J=12.1$ Hz), 5.22 (1H, dd, $J_{P-H}=1.0$ and $J_{H-H}=4.7$ Hz), 6.93 (2H, AA′BB′, $J=8.6$ Hz), 7.35 (2H, AA′BB′, $J=8.6$ Hz), 7.42 (1H, s), 8.52 (1H, s), 9.71 (1H, br d, $J=8.4$ Hz, NH), 12.63 (1H, br s, NHCHO). MS (SIMS) *m/z*: 654 ($M^+ + 1$). *Anal.* Calcd for $C_{25}H_{28}N_5O_{10}PS_2$: C, 45.87; H, 4.47; N, 10.70; S, 9.80. Found: C, 45.84, H, 4.40; N, 10.64; S, 9.87.

DCC (1.14 g, 5.5 mmol) was added to a solution of **4** (1.64 g, 3.7 mmol) and **5b**-(*Z*) (1.02 g, 4.5 mmol) in THF (30 ml), and the reaction mixture was stirred at room temperature for 2 d. The same work-up as described above gave **6b**-(*Z*) in 79% yield.

***p*-Methoxybenzyl 7-[(*Z*)-2-(2-Aminothiazol-4-yl)-2-methoxyiminoacetamido]-3-dimethoxyphosphinylmethyl-3-cephem-4-carboxylate (7b-(*Z*))**——A solution of **6b**-(*Z*) (2.548 g, 3.9 mmol) in methanol (30 ml) was treated with concentrated hydrochloric acid (1.2 ml). After being stirred at room temperature for 5 h, the reaction mixture was concentrated *in vacuo*. Water (200 ml) was added to the residue. The resulting solution was neutralized with saturated aqueous sodium hydrogen carbonate and then extracted with ethyl acetate. Usual work-up gave **7b**-(*Z*) (2.06 g, 85%). mp 123—125 °C (Acetone–AcOEt–$Et_2O$). IR (KBr): 3450, 3350, 1785, 1725, 1680, 1615, 1540, 1520, 1245, 1155 (sh), 1130, 820 $cm^{-1}$. $^1$H-NMR (DMSO-$d_6$) $\delta$: 3.10 (1H, d of ABd, $J_{P-H}=22.5$ and $J_{H-H}=14.7$ Hz), 3.47 (1H, d of ABd, $J_{P-H}=24.0$ and $J_{H-H}=14.7$ Hz), 3.58 (1H, d of ABd, $J_{P-H}=4.0$ and $J_{H-H}=18.1$ Hz), 3.58 (3H, d, $J_{P-H}=10.9$ Hz), 3.59 (3H, d, $J_{P-H}=10.9$ Hz), 3.66 (1H, d of ABd, $J_{P-H}=5.0$ and $J_{H-H}=18.1$ Hz), 3.75 (3H, s), 3.83 (3H, s), 5.13 (1H, ABd, $J=12.0$ Hz), 5.19 (1H, dd, $J_{P-H}=1.0$ and $J_{H-H}=4.8$ Hz), 5.20 (1H, ABd, $J=12.0$ Hz), 5.76 (1H, dd, $J=8.3$ and 4.8 Hz), 6.74 (1H, s), 6.93 (2H, AA′BB′, $J=8.7$ Hz), 7.23 (2H, br s, $NH_2$), 7.35 (2H, AA′BB′, $J=8.7$ Hz), 9.61 (1H, d, $J=8.3$ Hz, NH). MS (SIMS) *m/z*: 626 ($M^+ + 1$). *Anal.* Calcd for $C_{24}H_{28}N_5O_9PS_2$: C, 46.08; H, 4.51; N, 11.19; S, 10.25. Found: C, 45.73; H, 4.76; N, 10.80; S, 9.81.

**Sodium 7-[(*Z*)-2-(2-Aminothiazol-4-yl)-2-methoxyiminoacetamido]-3-dimethoxyphosphinylmethyl-3-cephem-4-carboxylate (8b-(*Z*))**——Compound **7b**-(*Z*) (0.447 g, 0.72 mmol) was dissolved in trifluoroacetic acid (2 ml). After being stirred at room temperature for 2 h, the reaction mixture was concentrated *in vacuo*. Trituration of the residue with diethyl ether gave 7-[(*Z*)-2-(2-aminothiazol-4-yl)-2-methoxyiminoacetamido]-3-dimethoxyphosphinylmethyl-3-cephem-4-carboxylic acid (0.430 g, 97%) as yellow crystals, which were neutralized with saturated aqueous sodium hydrogen carbonate and purified by reversed-phase silica gel column chromatography (Lobar RP-8 column). Lyophilization of the eluate gave **8b**-(*Z*) (185 mg, 49%). IR (KBr): 1770, 1670, 1605, 1540, 1400, 1365, 1185, 1040, 810 $cm^{-1}$. $^1$H-NMR (DMSO-$d_6$) $\delta$: 2.99 (1H, d of ABd, $J_{P-H}=22.0$ and $J_{H-H}=14.7$ Hz), 3.28 (1H, d, of ABd, $J_{P-H}=2.6$ and $J_{H-H}=17.4$ Hz), 3.52 (1H, d of ABd, $J_{P-H}=5.0$ and $J_{H-H}=17.4$ Hz), 3.60 (3H, d, $J_{P-H}=10.7$ Hz), 3.61 (3H, d, $J_{P-H}=10.7$ Hz), 3.83 (3H, s), 3.90 (1H, d of ABd, $J_{P-H}=21.5$ and $J_{H-H}=14.7$ Hz), 4.97 (1H, d, $J=4.6$ Hz), 5.51 (1H, dd, $J=8.3$ and 4.6 Hz), 6.74 (1H, s), 7.22 (2H, br s, $NH_2$), 9.50 (1H, d, $J=8.3$ Hz, NH). MS (SIMS) *m/z*: 528 ($M^+ + 1$).

**7-[(*Z*)-2-(2-Aminothiazol-4-yl)-2-methoxyiminoacetamido]-3-dihydroxyphosphinylmethyl-3-cephem-4-carboxylic Acid Trisodium Salt (9b-(*Z*))**——MSTFA (0.52 ml, 2.8 mmol) and bromotrimethylsilane (0.35 ml, 2.7 mmol) were added to a solution of **7b**-(*Z*) (0.266 g, 4.3 mmol) in dichloromethane (10 ml), and the reaction mixture was stirred at room temperature for 3 h. The reaction was so slow that the same amount of bromotrimethylsilane was added and the mixture was stirred at 40 °C for 5 h. The same work-up as described for **9a**-(*Z*) gave **9b**-(*Z*) (0.134 g, 57%) as a

colorless amorphous solid. mp >195 °C (dec.). IR (KBr): 1760, 1660, 1600, 1540, 1410, 1390, 1370, 1050 $cm^{-1}$. $^1$H-NMR ($D_2O$) $\delta$: 2.43 (1H, d of ABd, $J_{P-H}$=20.0 and $J_{H-H}$=14.2 Hz), 2.81 (1H, d of ABd, $J_{P-H}$=20.0 and $J_{H-H}$=14.2 Hz), 3.35 (1H, ABd, $J$=17.8 Hz), 3.76 (1H, d of ABd, $J_{P-H}$=3.8 and $J_{H-H}$=17.8 Hz), 3.88 (3H, s), 5.14 (1H, d, $J$=4.4 Hz), 5.64 (1H, d, $J$=4.4 Hz), 6.94 (1H, s). MS (SIMS) $m/z$: ($M^+$+1), 522 ($M^+$+1−Na+$H^+$), 500 ($M^+$+1−2Na+$H^+$), 478 ($M^+$+1−3Na+$H^+$). *Anal.* Calcd for $C_{14}H_{13}N_5Na_3O_8PS_2 \cdot 5H_2O$: C, 26.55; H, 3.66; N, 11.06; S, 10.12. Found: C, 26.32; H, 3.44; N, 10.66; S, 10.50.

**Sodium 7-[3-Chloro-2-(thiazol-4-yl)propenamido]-3-dimethoxyphosphinylmethyl-3-cephem-4-carboxylate (11-(*Z*) and 11-(*E*))**——THF (1.3 ml) and (*E*)-3-chloro-2-(thiazol-4-yl)propenoic acid (**10**-(*E*))[15] (250 mg, 1.3 mmol) were added to Vilsmeier reagent [prepared from phosphorus oxychloride (0.13 ml, 1.3 mmol) and dimethylformamide (DMF) (0.20 ml, 2.6 mmol)] at 0 °C. The reaction mixture was stirred at 40 °C for 1.5 h. In parallel, *p*-methoxybenzyl 7-amino-3-dimethoxyphosphinylmethyl-3-cephem-4-carboxylate (**4**) (426 mg, 0.89 mmol) was dissolved in trifluoroacetic acid (3 ml). After being stirred at room temperature for 1 h, the reaction mixture was concentrated *in vacuo*. Trituration of the residue with ethyl acetate gave 7-amino-3-dimethoxyphosphinylmethyl-3-cephem-4-carboxylic acid (**4**; R=H) trifluoroacetic acid salt. The collected precipitate was neutralized with sodium hydrogen carbonate in the mixture of water (2.6 ml) and acetone (2.6 ml). To this solution, the above activated carboxylic acid solution was added dropwise at 0 °C, adjusting the pH value of the reaction mixture to 7. The whole was stirred for 15 min, then the organic solvent was removed under reduced pressure, and the pH value was adjusted again to 7. Chromatography on a Lobar RP-8 column and lyophilization gave **11**-(*Z*) (73 mg, 16%) and **11**-(*E*) (161 mg, 35%), each as a colorless amorphous solid.

**11**-(*Z*): $^1$H-NMR (DMSO-$d_6$) $\delta$: 2.7—4.1 (4H, m), 3.63 (6H, d, $J$=11.0 Hz), 5.08 (1H, d, $J$=5.0 Hz), 5.63 (1H, dd, $J$=8.3 and 5.0 Hz), 7.31 (1H, s), 7.56 (1H, d, $J$=1.8 Hz), 9.18 (1H, d, $J$=1.8 Hz), 9.52 (1H, d, $J$=8.3 Hz).

**11**-(*E*): $^1$H-NMR (DMSO-$d_6$) $\delta$: 2.8—4.2 (4H, m), 3.62 (6H, d, $J$=11.0 Hz), 5.00 (1H, d, $J$=4.8 Hz), 5.56 (1H, dd, $J$=8.4 and 4.8 Hz), 7.36 (1H, s), 8.11 (1H, d, $J$=1.8 Hz), 9.23 (1H, d, $J$=1.8 Hz), 9.35 (1H, d, $J$=8.4 Hz).

**7-[(*Z*)-3-Chloro-2-(thiazol-4-yl)propenamido]-3-dihydroxyphosphinylmethyl-3-cephem-4-carboxylic Acid Trisodium Salt (12-(*Z*))**——Compound **11**-(*Z*) (73 mg, 0.14 mmol) was carefully acidified with concentrated hydrochloric acid to pH 3, and then extracted with ethyl acetate. The residue from the extract was dissolved in dichloromethane (2.0 ml). After the addition of MSTFA (0.11 ml, 0.58 mmol) and bromotrimethylsilane (0.13 ml, 0.98 mmol), the reaction mixture was stirred at 40 °C for 1 h. Water (3 ml) was added. The separated aqueous layer was neutralized, washed with dichloromethane and chromatographed on a Lobar RP-8 column. Lyophilization of the eluate afforded **12**-(*Z*) (20 mg, 27%) as a colorless amorphous solid. IR (KBr): 1755, 1660, 1595, 1550, 1410, 1360, 1070, 970 $cm^{-1}$. $^1$H-NMR ($D_2O$) $\delta$: 2.62 (1H, d of ABd, $J_{P-H}$=20.5 and $J_{H-H}$=14.4 Hz), 2.87 (1H, d of ABd, $J_{P-H}$=20.5 and $J_{H-H}$=14.4 Hz), 3.48 (1H, d of ABd, $J_{P-H}$=2.2 and $J_{H-H}$=17.9 Hz), 3.85 (1H, d of ABd, $J_{P-H}$=4.1 and $J_{H-H}$=17.9 Hz), 5.29 (1H, d, $J$=4.6 Hz), 5.83 (1H, d, $J$=4.6 Hz), 7.27 (1H, s), 7.66 (1H, d, $J$=1.9 Hz), 9.03 (1H, d, $J$=1.9 Hz). MS (SIMS) $m/z$: 532 ($M^+$+1).

**7-[(*E*)-3-Chloro-2-(thiazol-4-yl)propenamido]-3-dihydroxyphosphinylmethyl-3-cephem-4-carboxylic Acid Trisodium Salt (12-(*E*))**——The same procedure as described above was applied to **11**-(*E*) (93 mg, 0.19 mmol), MSTFA (0.22 ml, 1.2 mmol), and bromotrimethylsilane (0.25 ml, 2 mmol) to afford **12**-(*E*) (105 mg, 63%) as a colorless amorphous solid. IR (KBr): 1755, 1670, 1600, 1540, 1415, 1370, 1285, 1160, 1065, 880, 820 $cm^{-1}$. $^1$H-NMR ($D_2O$) $\delta$: 2.73 (1H, d of ABd, $J_{P-H}$=22.0 and $J_{H-H}$=14.5 Hz), 2.81 (1H, d, of ABd, $J_{P-H}$=20.2 and $J_{H-H}$=14.5 Hz), 3.52 (1H, d of ABd, $J_{P-H}$=2.9 and $J_{H-H}$=17.9 Hz), 3.73 (1H, d of ABd, $J_{P-H}$=4.6 and $J_{H-H}$=17.9 Hz), 5.19 (1H, d, $J$=4.6 Hz), 5.75 (1H, d, $J$=4.6 Hz), 7.45 (1H, s), 8.07 (1H, d, $J$=2.0 Hz), 9.08 (1H, d, $J$=2.0 Hz). MS (SIMS) $m/z$: 532 ($M^+$+1).

## References and Notes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