183. Are the Known Δ^2-Cephems Inactive as Antibiotics because of an Unfavourable Steric Orientation of their 4α-Carboxylic Group? Synthesis and Biology of Two Δ^2-Cephem-4β-carboxylic Acids

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Two representatives of the yet unknown type of Δ^2-cephem-4β-carboxylic acids were prepared. Contrarily to the prediction based on the activity model of *Cohen*, both acids proved inactive as antibiotics. Possible reasons for this discrepancy are briefly discussed.

Introduction. – It has been suggested by *Tipper* and *Strominger* (1965) [1] that the antibiotic activity of penicillins is based on their ability to acylate, by virtue of their reactive β-lactam grouping, the transamidases involved in the biosynthesis of the bacterial cell wall [2] [3].

This chemical interpretation of the mode of action was subsequently extended to other known β-lactam antibiotics and was accepted by chemists active in this area as working hypothesis for the conception of new potential antibiotics. Along these lines, a great variety of compounds with chemically activated β-lactam moieties was synthesized leading to the discovery of some β-lactam systems of high interest; the penems are a typical example of the fruits of this effort.

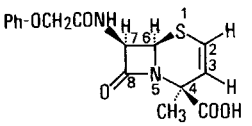
However, failures of this hypothesis were also reported revealing serious deficiencies of the reactivity model. Systematic analyses showed that the chemical reactivity of the β-lactam cannot be simply correlated with the biological properties of the molecules. *Frère et al.* [4] studied a wide range of β-lactam compounds and measured not only their chemical reactivity in alkaline hydrolysis but also the kinetic parameters of their interaction with various β-lactamases and peptidases. As a conclusion of these studies, it has been suggested that the primary parameter which governs the biological activity is the goodness of fit with the enzyme and not the chemical reactivity of the β-lactam.

In 1983, *Cohen* [5] analyzed the three-dimensional (3-D) features of a set of representative, biologically active and inactive β-lactam structures and concluded that highly specific 3-D recognition sites may be involved in the enzymes in their recognition of the antibiotics. Thus, a lack of antibacterial activity can be the result of a non-recognition rather than of an insufficient reactivity of the β-lactam system. According to this model, *e.g.*, the biological inactivity of Δ^2-cephems may not be primarily the result of a low reactivity of their β-lactam grouping – in fact, as shown by *Frère et al.* [4], the liabilities for hydrolysis of Δ^2- and Δ^3-cephems are comparable – but rather a consequence of a misfit with the enzyme due to the unfavourable sterical location of the α-oriented

carboxylic group. In 4β-epimers, on the other hand, the acid function would fit very well with the 3-D requirements of the model of *Cohen*, and he postulated antibiotic activities for such compounds.

However, no Δ^2-cephem acids with a β-orientation of the carboxylic group are known in the literature. Therefore, to check the correctness of the above mentioned challenging postulate, we have now prepared two Δ^2-cephem-4β-carboxylic acids and, for comparison of the biological activities, also their α-oriented counterparts.

Synthesis of 4α-Methyl-7β-(2-phenoxyacetamido)-Δ^2-cephem-4β-carboxylic Acid and of its C(4) Epimer. – The fact that all known Δ^2-cephem-4-carboxylic acids belong to the 4α-series suggests a greater thermodynamic stability of these compounds as compared to the – up to now hypothetic – 4β-epimers[1]). Therefore, to exclude any isomerizations at the C(4) center, we decided to fix the configuration at this position by substituting a Me group for the H-atom, *i.e.*, to synthesize the two epimeric 4-methyl-Δ^2-cephem acids **1** and **2**.



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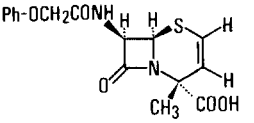

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For the synthesis of both compounds, the 3-hydroxy-Δ^3-cephem ester **3**, an intermediate in the synthesis of the antibiotic *Oraspor*, was chosen as starting material [7]. By treatment of **3** with an excess of MeI in acetone in the presence of K_2CO_3, two C(4)-methylated products **4** and **5** were mainly formed and isolated in 57 and 16.5% yield, respectively, after chromatographic separation. The structure assignment followed unequivocally from ^{1}H-NMR analyses and NOE measurements and was later confirmed by the transformation products of both compounds.

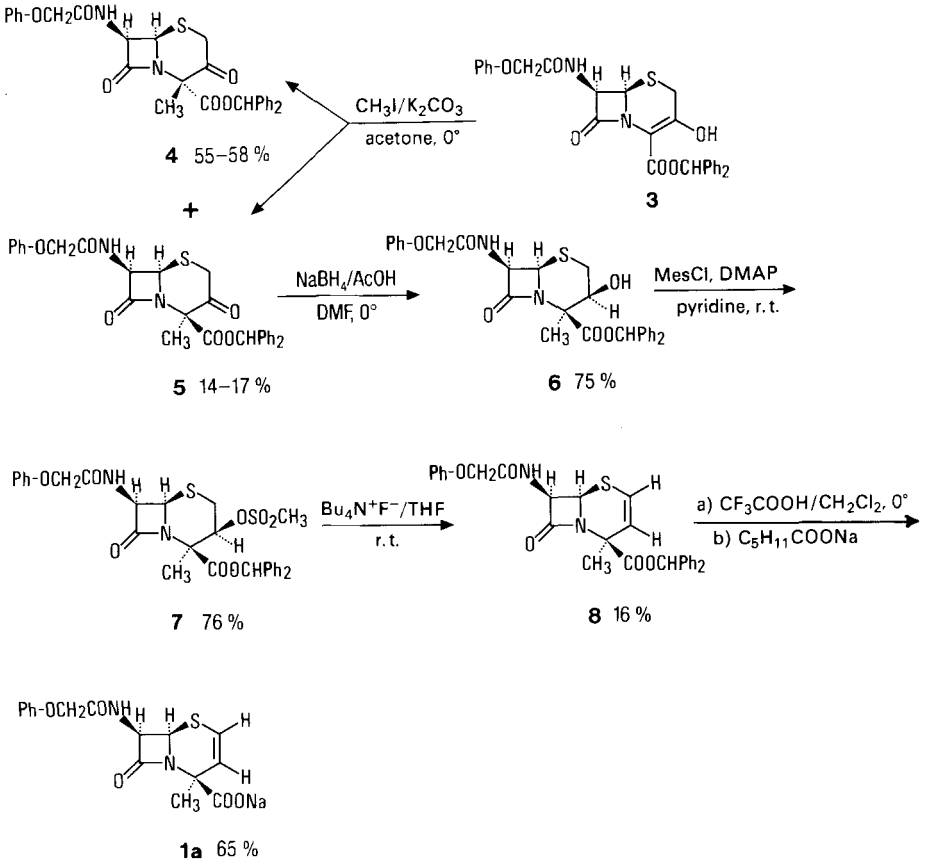
The minor, crystalline methylation product **5** with its β-oriented ester grouping was transformed into the sodium salt of 4α-methyl-7β-(2-phenoxyacetamido)-Δ^2-cephem-4β-carboxylic acid (**1a**) in the following way (*cf. Scheme 1*). A $NaBH_4$ reduction of **5** in DMF in the presence of AcOH afforded the 3β-hydroxy derivative **6** (70–75% isolated yield); it was accompanied by a small amount of the 3α-isomer **9** and by variable amounts of the β-lactam-free lactone **10**. The latter compound is a secondary product formed from the alcohol **6**, the axial OH group of which being extremely well located for an intramolecular interaction with the β-lactam-carbonyl moiety.

The above mentioned trend of the alcohol **6** for intramolecular ring-opening of the β-lactam moiety created problems in its transformation to the mesylate **7**. Finally, the formation of the undesired lactone **10** was substantially suppressed by using a large excess (6–10 equiv.) of mesyl chloride in pyridine in the presence of 4-(dimethylamino)pyridine (DMAP) as catalyst (yield of **7**: 76%).

Unexpected problems of a different kind were met in the conversion of the mesylate **7** to the Δ^2-cephem ester **8**, this in spite of the axial position of the mesyloxy grouping

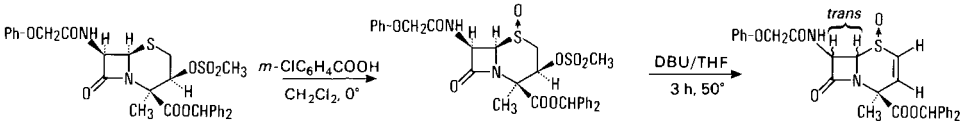
[1]) According to molecular-mechanics calculations [6], the 4α-carboxylic acids are more stable than their 4β-epimers by *ca.* 1 kcal/mol of energy difference.

Scheme 1

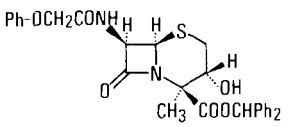


favourable for such a *trans*-elimination reaction. Of the various examined elimination methods, only the use of anhydrous tetrabutylammonium fluoride in THF was partially successful allowing the isolation of **8** in yields of 15–20%; especially with extended reaction times, two other products prevailed: an isomer of **8** with *trans*-oriented β-lactam H-atoms, and the tricyclic keto sulfonate **11**, the latter a result of a primary deprotonation of **7** on the mesylate Me group[2]).

[2]) An attempted preparation of the corresponding *tosylate* from the alcohol **6** failed owing to the above mentioned, easy β-lactam ring-opening of **6** to the lactone **10**. An additional acidification at C(2) for deprotonation by transforming the mesylate **7** into the S-*oxide* **12** resulted, with DBU in THF in the elimination step in a complete *cis* → *trans* isomerization on the β-lactam moiety (*cf.* **13**).



7 **12** **13**



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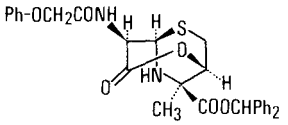

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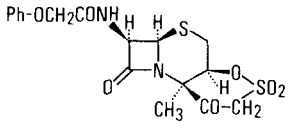

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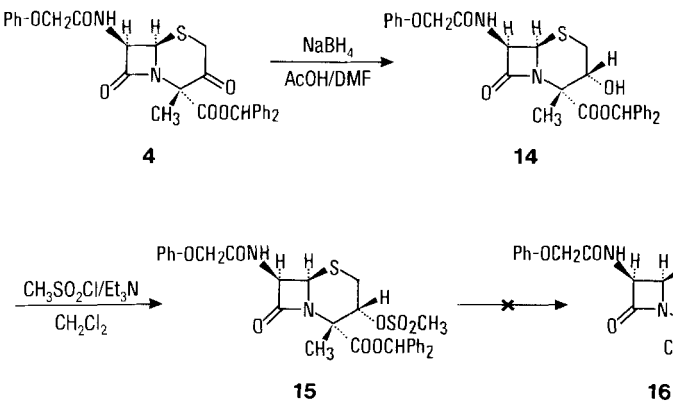
Finally, treatment of the benzhydryl ester **8** with CF_3COOH (in CH_2Cl_2) and an exchange of the free acid thus formed with sodium hexanoate afforded the sodium salt **1a** of the desired 4β-carboxylic acid. Its structure was confirmed, in addition to other spectroscopic evidence, by a strong NOE intensity enhancement for the H-atom at C(6) upon irradiation of CH_3 at C(4)[3]).

For the preparation of the – also unknown – 4β-methyl-Δ^2-cephem acid **2** with the α-oriented carboxylic group, a similar scheme as described for **1a** was considered, the starting material this time being the major methylation product of **3**, namely the 4β-methyl-3-oxocepham ester **4** *(Scheme 2)*.

Scheme 2



4 14

15 16

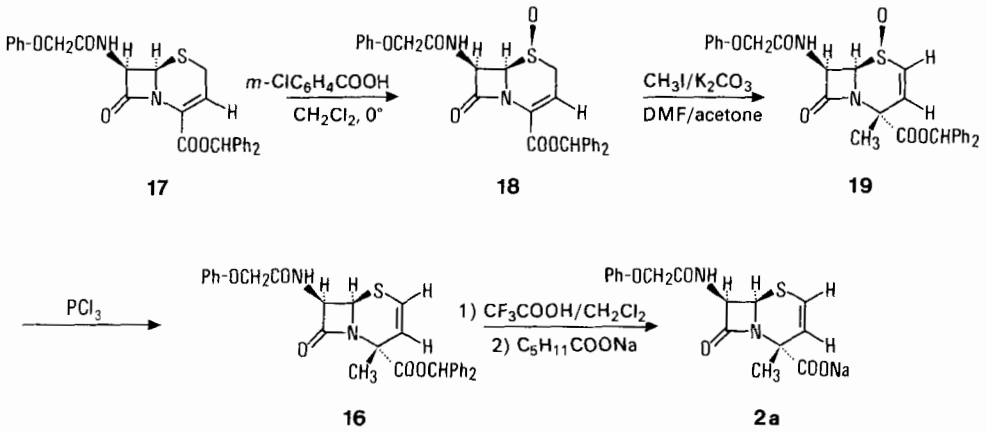
However, in contrast to the reduction of ketone **5** to the axial alcohol **6**, a similar $NaBH_4$ reduction of the epimeric ketone **4** (in AcOH/DMF) gave the equatorial, 3α-oriented alcohol **14** and the following mesylation the equatorial mesylate **15**[4]). All attempts to eliminate the elements of methansulfonic acid from the latter compound led only to intractable decomposition products and no Δ^2-cephem ester **16** could be identified.

On the other hand, a shown in *Scheme 3*, the ester **16** was easily prepared starting from the benzhydryl ester **17** of 7β-(2-phenoxyacetamido)-Δ^3-cephem-4-carboxylic acid [7a] [8]. The Δ^3-cephem *S*-oxide **18**, prepared from **17** by treatment with 1 equiv. of *m*-chloro-

[3]) For the isomeric salt **2a** (see below), no NOE was observed.

[4]) A conformational analysis of the 4-methyl ketones **4** and **5** reveals, that the most stable conformation of **4** has the six-membered ring in a chair and that of **5** in a distorted boat form, the Me and the ester groups on C(4) being – in both cases – equatorial and axial, respectively. The nucleophilic attack of the hydride reagent on the C(3) carbonyl takes place from the face opposite to that where the ester grouping is located and leads, with **4**, to the formation of the equatorial 3α-alcohol **14** and, in the case of **5**, to the axial 3β-alcohol **6**.

Scheme 3



17 **18** **19**

16 **2a**

perbenzoic acid (CH_2Cl_2, 0°), was methylated with an excess of MeI in acetone/DMF in the presence of K_2CO_3 to give exclusively the 4β-methyl-Δ^2-cephem derivative **19**. Deoxygenation of the *S*-oxide grouping in **19** by PCl_3 then afforded the ester **16**. This nicely crystalline compound suited well for an X-ray analysis which confirmed, among other features, the β-orientation of the Me group on C(4) (*cf. Figure* and *Exper. Part*). For

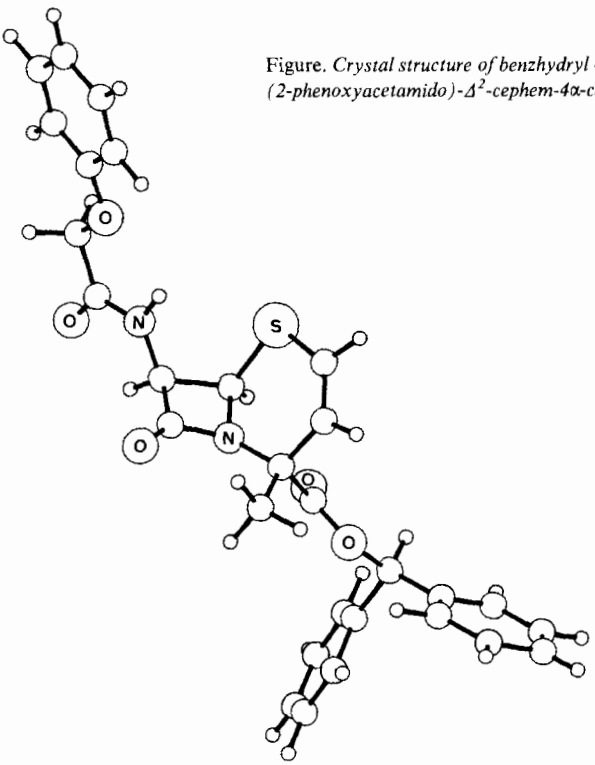
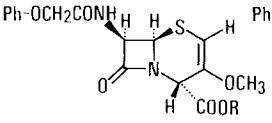


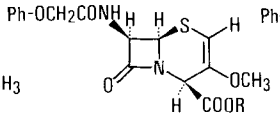
Figure. *Crystal structure of benzhydryl 4β-methyl-7β-(2-phenoxyacetamido)-Δ²-cephem-4α-carboxylate* (**16**)

biological tests, the ester **16** was converted – with CF_3COOH and sodium hexanoate – to the sodium salt **2a** of the corresponding acid.

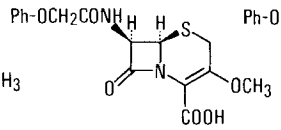
Synthesis of 3-Methoxy-7β-(2-phenoxyacetamido)-Δ²-cephem-4β-carboxylic Acid and its 4α-Epimer. – In the synthesis of 3-methoxy-substituted cephems, a minor by-product was detected in one of the steps which was isomeric with the Δ²-cephem-4α-carboxylate **20** and was identified – mainly by ^{1}H-NMR analysis[5]) – as the 4β-epimer **21** [9].



20 R = $CHPh_2$
22 R = H



21 R = $CHPh_2$
23 R = H



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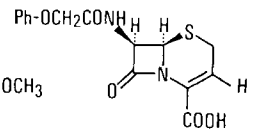

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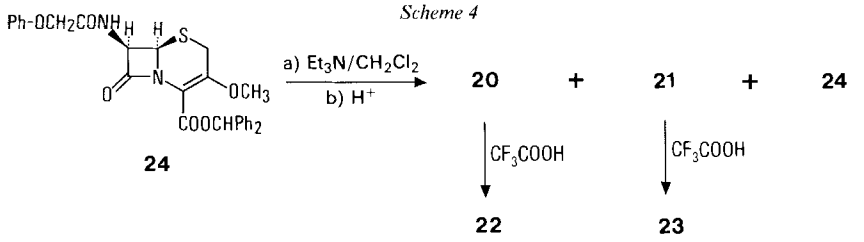
In this observation, we recognized a chance to prepare a pair of Δ²-cephem-4-carboxylic acids without any further substitution on C(4), *i.e.* the acids **22** and **23** which would be true isomers of the biologically active Δ³-cephem-4-carboxylic acid **25**. The geometrical features of the acid **23** should fully comply with the demands of the model of *Cohen*.

Equilibration of the 3-methoxy-Δ³-cephem ester **24** [7a] in CH_2Cl_2 with Et_3N afforded, after acidic quenching, a mixture of **24** (*ca.* 23%) and of the two Δ²-cephem esters **20** (*ca.* 75%) and **21** (*ca.* 2%; *cf. Scheme 4*). Both Δ²-cephem esters were isolated in pure form by column chromatography and by crystallization. Separate removal of the benzhydryl group (CF_3COOH/CH_2Cl_2) then gave the corresponding acids **22** and **23**. Unfortunately, the 4β-carboxylic acid **23** – the most important compound for the planned biological testing – crystallized from all solvents tried in very fine needles unsuitable for an X-ray structure determination; however, its structure as well as that of the isomeric acid **22** and of the corresponding esters **21** and **20** follow unequivocally from their ^{1}H-NMR analyses.

Scheme 4



Biological Results and Discussion. – Both pairs of Δ²-cephem-4-carboxylic acids **1**/**2** and **22**/**23** were tested for biological activity in an agar dilution test; the Δ³-cephem-4-carboxylic acids **25** and **26** were also included in the tests as standards.

As shown in *Table 1*, the hopes for a prominent biological activity of the Δ²-cephem-4β-carboxylic acids (see **1a** and **23**) as postulated by *Cohen* were not fulfilled. The

[5]) A long-distance coupling (over 5 bonds) with $J \approx 1$ Hz is observed in **21** between H–C(4α) and H–C(7α); such a coupling is absent in the ^{1}H-NMR spectra of **20** and of the known Δ²-cephem-4α-carboxylic acids with β-oriented H–C(4).

Table 1. *Biological Activity of Δ^2-Cephem-4-carboxylic Acids* **1**, **2**, **22**, *and* **23** *in Agar Dilution Test*[a])

	1a	**2a**	**26**	**23**	**22**	**25**
Staphylococcus aureus 10 B	> 128	32	0.2	> 128	16	1
Staphylococcus epidermidis R 13	> 128	16	0.2	> 128	8	1
Streptococcus pyogenes Aronson	> 128	64	1	128	32	< 1
Streptococcus pneumoniae III/84	> 128	32	2	128	32	2
Neisseria meningitidis 1316	> 128	128	2	> 128	128	4
Neisseria gonorrhoeae 1317/4	> 128	64	2	> 128	32	4
Haemophilus influenzae NCTC 4560	> 128	> 128	4	> 128	128	4
Clostridium perfringens 194 anaerob	> 128	32	1	32	4	2
Bacteroides fragilis L 01 anaerob	> 128	> 128	32	> 128	> 128	128
Escherichia coli 205	> 128	> 128	32	> 128	> 128	> 128
Klebsiella pneumoniae 327	> 128	> 128	32	> 128	> 128	> 128

[a]) DST agar as test medium; inoculum 10^4/drop, incubation temp. 37°; *MIC* in μg/ml.

4-methyl-substituted compound **1a** proved entirely inactive against all microorganisms used in the test, and the 4-unsubstituted 4β-acid **23** displayed only a marginal activity against three strains. In both cases, the activity of the 4β-oriented acid was inferior to that of the Δ^3-cephem acids **26** and **25**, and even to that of the very weakly active Δ^2-cephem-4α-carboxylic acids (see **2a** and **22**). The relatively high stability of the Δ^2-acids as determined at 37° in a biological buffer at pH 7.4 excludes an explanation of the observed, more or less negative results by decomposition of the substances in the early stages of the biological test.

Thus, it has to be concluded that an activity model based solely on a good fit of the β-lactam compound with the active site of the transpeptidase fails to explain the above mentioned biological results. On the other hand, we feel it incorrect to reject, because of the biological inactivity of the two β-oriented Δ^2-cephem acids, the conclusions about geometrical requirements for antibacterial activity of β-lactam antibiotics as presented in the quoted paper by *Cohen* [5]. Considering this failure of the steric model on the one hand, and the difficulties encountered with the reactivity model on the other hand, we now tend to believe – this somewhat contrarily to the conclusions of *Frère et al.* [4] – that for a good antibiotic activity a subtle combination of both features, namely a 3-D recognition *and* a sufficient chemical reactivity of the β-lactam, are necessary. The latter condition is obviously not fulfilled in the otherwise geometrically well suited Δ^2-cephem-4β-carboxylic acids.

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Experimental Part

General. R_f values: *Merck* silica gel *60* F_{254} TLC plates. M.p.: *Kofler;* uncorrected. IR spectra: absorptions in cm^{-1}, ^{1}H-NMR (400.1 MHz) and ^{13}C-NMR (100.6 MHz): *Bruker WM 400* spectrometer; some spectra were recorded on *Bruker AM 360* and on *Varian HA 100B* spectrometers; chemical shifts as δ values in ppm with respect to tetramethylsilane as internal reference ($\delta = 0$ ppm), coupling constants J in Hz. MS: *Varian CH 7* spectrometer; FAB measurements: *ZAB-HF* spectrometer of *VG Analytical.*

Benzhydryl 4-Methyl-3-oxo-7β-(2-phenoxyacetamido)cepham-4-carboxylates[6]) (**4** and **5**). In a soln. of 20.66 g (40 mmol) of benzhydryl 3-hydroxy-7β-(2-phenoxyacetamido)-Δ^3-cepham-4-carboxylate (**3**) [7] and 60 ml (0.96 mol) of MeI in 240 ml of acetone, 22.12 g (160 mmol) of pulverized K_2CO_3 were suspended at 0° and stirred under Ar in an ice/H_2O bath for 15 h. The K^+ salts were filtered off, the filtrate was evaporated and the residue dissolved in CH_2Cl_2 and washed with 8% aq. $NaHCO_3$ soln. The crude product as obtained by evaporation of the org. phase was chromatographed on 700 g of silica gel (*Merck*, 230–400 mesh ASTM). After several, almost empty fractions eluted with toluene, the major 4α-carboxylate **4** was eluted in many fractions with toluene/AcOEt 19:1 (11.0 g), followed, after several mixed fractions (1.6 g), by the minor 4β-carboxylate **5** (4.06 g) eluted with toluene/AcOEt 19:1 and 4:1. Crystallization of the latter from CH_2Cl_2/Et_2O gave 2.86 g of pure **5**; the mother liquors were combined with the above mentioned mixed fractions and rechromatographed on 10 prep. TLC plates (20 × 100 × 0.2 cm) with toluene/AcOEt 2:1, giving 1.11 g of pure **4** and 0.64 g of pure **5**, increasing the total yield of pure **4** to 12.11 g (57.1%) and that of **5** to 3.50 g (16.5%).

Benzhydryl 4β-Methyl-3-oxo-7β-(2-phenoxyacetamido)cepham-4α-carboxylate[6]) (**4**). Amorphous foam. R_f (toluene/AcOEt 1:1) 0.53. $[\alpha]_D^{20} = +162.3 \pm 0.9°$ (c = 1.132, $CHCl_3$). IR (CH_2Cl_2): 3400, 1780, 1728, 1696, 1600, 1515, 1496, 1440, 1350, 1230, 1178, 1140, 1080, 1060, 950. ^{1}H-NMR (400 MHz, $CDCl_3$): 7.37–6.91 (*m*, 17 H); 5.37 (*dd*, J = 8.0, 4.5, 1 H); 5.25 (*d*, J = 4.5, 1 H); 4.59 (*m*, 2 H); 3.41 (*d*, J = 15, 1 H); 3.09 (*d*, J = 15, 1 H); 1.98 (*s*, 3 H). Anal. calc. for $C_{29}H_{26}N_2O_6S$ (530.60): C 65.65, H 4.94, N 5.28, O 18.09, S 6.04; found: C 65.84, H 5.14, N 5.14, O 17.99, S 5.78.

Benzhydryl 4α-Methyl-3-oxo-7β-(2-phenoxyacetamido)cepham-4β-carboxylate[6]) (**5**). M.p. 151–153° (CH_2Cl_2/Et_2O). R_f (toluene/AcOEt 1:1) 0.46. $[\alpha]_D^{20} = +266.9 \pm 1.0°$ (c = 0.961, $CHCl_3$). IR (CH_2Cl_2): 3414, 1783, 1732, 1698, 1600, 1515, 1496, 1442, 1349, 1239, 1177, 1083, 1062, 957. ^{1}H-NMR (400 MHz, $CDCl_3$): 7.37–6.93 (*m*, 16 H); 6.87 (*s*, 1 H); 5.33 (*dd*, J = 7.5, 4.0, 1 H); 5.04 (*d*, J = 4.0, 1 H); 4.59 (*m*, 2 H); 3.70 (*d*, J = 14, 1 H); 2.93 (*d*, J = 14, 1 H); 1.75 (*s*, 3 H); irradiation at 1.75 (CH_3)→NOE at 5.04 (H–C(6)). Anal. calc. for $C_{29}H_{26}N_2O_6S$ (530.60): C 65.65, H 4.94, N 5.28, O 18.09, S 6.04; found: C 65.63, H 5.03, N 5.17, O 18.13, S 5.81.

Benzhydryl 3β-Hydroxy-4α-methyl-7β-(2-phenoxyacetamido)cepham-4β-carboxylate[6]) (**6**). To a soln. of 4.43 g (8.35 mmol) of **5** in 40 ml of DMF and 10.5 ml of AcOH, 397 mg (10.5 mmol) of $NaBH_4$ were added in small portions within 20 min while stirring under Ar in an ice/H_2O bath. After a total of 60 min of stirring at 0–5°, the mixture was evaporated at 40° under the vacuum of an oil pump and the residue in CH_2Cl_2 washed with H_2O and sat. brine; the aq. parts were reextracted with CH_2Cl_2. The crude product (4.75 g) obtained by evaporation of the combined org. extracts was chromatographed on 200 g of *Merck* silica gel *60* with toluene/AcOEt 9:1. After several fractions containing mainly the minor **9** (see below), a total of 3.34 g (75.1%) of pure **6** was collected as amorphous foam, enclosing trace amounts of solvents (*e.g.* toluene), even after drying for several days at 30°/0.1 mbar. R_f (toluene/AcOEt 1:1) 0.39. $[\alpha]_D^{20} = +95.7 \pm 1.0°$ (c = 1.023, $CHCl_3$). IR (CH_2Cl_2): 3550, 3405, 3052, 1779, 1730, 1692, 1599, 1517, 1494, 1440, 1349, 1226, 1156, 1081, 961. ^{1}H-NMR (400 MHz, $CDCl_3$): 7.60 (*d*, J = 9, 1 H); 7.40–6.89 (*m*, 16 H); 5.69 (*dd*, J = 9, 4.5, 1 H); 5.08 (*d*, J = 4.5, 1 H); 4.54 (*s*, 2 H); 4.06 (*dd*, J = 4.5, 1.5, 1 H); 3.33 (*dd*, J = 14.5, 1.5, 1 H); 2.87 (*dd*, J = 14.5, 4.5, 1 H); 1.46 (*s*, 3 H); irradiation at 1.46→NOE at 5.08, 4.06, and 3.33. Anal. calc. for $C_{29}H_{28}N_2O_6S$ (532.61): C 65.40, H 5.30, N 5.26, S 6.02; found: C 64.88, H 5.50, N 4.99, S 5.83.

In another experiment with 6.15 g of **5**, the early fractions (0.30 g) of the chromatography of the crude product were rechromatographed on *Merck* silica gel plates in toluene/AcOEt 2:1, yielding 107 mg of *benzhydryl 3α-hydroxy-4α-methyl-7β-(2-phenoxyacetamido)cepham-4β-carboxylate*[6]) (**9**) as a minor product. Amorphous foam. R_f (toluene/AcOEt 1:1) 0.44. IR (CH_2Cl_2): 3550, 3400, 1775, 1730, 1690, 1598, 1518, 1492, 1450–1417 (br.), 1360–1320 (br.), 1221, 1182, 1061, 990. ^{1}H-NMR (400 MHz, $CDCl_3$): 7.42–6.90 (*m*, 17 H); 5.64 (*dd*, J = 9, 4, 1 H); 5.02 (*d*, J = 4, 1 H); 4.52 (*s*, 2 H); 4.36 (*dd*, J = 10, 4, 1 H); 3.00 (*dd*, J = 13, 10, 1 H); 2.67 (*dd*, J = 13, 4, 1 H); 1.52 (*s*, 3 H); irradiation at 1.52→NOE at 5.02, 4.36, and 3.00.

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[6]) The systematic names of the parent cepham-, Δ^2-cephem-, and Δ^3-cephem-4-carboxylic acids are *8-oxo-5-thia-1-azabicyclo[4.2.0]octane-*, *8-oxo-5-thia-1-azabicyclo[4.2.0]oct-3-ene-*, and *8-oxo-5-thia-1-azabicyclo-[4.2.0]oct-2-ene-2-carboxylic acid*, respectively.

Benzhydryl 3β-(Methansulfonyloxy)-4α-methyl-7β-(2-phenoxyacetamido)cepham-4β-carboxylate[6] (**7**). To a soln. of 2.16 g (4.05 mmol) of **6** and of 488 mg (4 mmol) of 4-(dimethylamino)pyridine in 30 ml of pyridine, 3.1 ml (*ca.* 40 mmol) of MesCl were slowly added while stirring under Ar at r.t. After a total of 3 h, the mixture (with a crystalline precipitate) was evaporated under high vacuum and the residue in CH_2Cl_2 washed with 8% aq. $NaHCO_3$ soln. and H_2O; the aq. parts were reextracted with CH_2Cl_2. The residue obtained by evaporation of the combined org. parts (3.2 g) was chromatographed on 100 g of *Merck* silica gel (230–400 mesh ASTM) with toluene/AcOEt 9:1. After several small fractions containing impurities, a total of 1.49 g (61%) of **7** was collected as a solid foam enclosing solvent residues, even after prolonged drying under high vacuum. R_f (toluene/AcOEt 2:1) 0.31. IR (CH_2Cl_2): 3405, 3055, 1791, 1744, 1696, 1600, 1519, 1496, 1454, 1442, 1360, 1240, 1179, 1156, 1084, 1064, 1020, 964, 914. ^{1}H-NMR (400 MHz, $CDCl_3$): 7.60 (*d*, *J* = 9, 1 H); 7.48–6.97 (*m*, 15 H); 6.87 (*s*, 1 H); 5.75 (*dd*, *J* = 9, 4, 1 H); 5.10 (*d*, *J* = 4, 1 H); 5.03 (*dd*, *J* = 4, 1, 1 H); 4.54 (*s*, 2 H); 3.41 (*dd*, *J* = 14, 1, 1 H); 3.22 (*dd*, *J* = 14, 4, 1 H); 2.74 (*s*, 3 H); 1.46 (*s*, 3 H). Anal. calc. for $C_{30}H_{30}N_2O_8S_2$ (610.70): C 59.00, H 4.95, N 4.59, O 20.96, S 10.50; found: C 57.27, H 4.76, N 4.51, O 19.84, S 9.90, Cl 3.30; the analysis fits if 4.0% of residual CH_2Cl_2 is considered: calc. C 57.23, H 4.84, N 4.41, O 20.13, S 10.08, Cl 3.30.

In another mesylation experiment, 608 mg (1.14 mmol) of **6** in 8.5 ml of CH_2Cl_2 was treated at r.t. with 133 µl (*ca.* 1.71 mmol) of MesCl in the presence of 277 mg (2.74 mmol) of Et_3N. Workup after 4 h by washing with 8% aq. $NaHCO_3$ soln. and H_2O and prep. TLC of the crude product (*Merck* silica gel *60* plates) with toluene/AcOEt 2:1 afforded only 217 mg (31.1%) of **7**; however, 254 mg of another, less mobile compound, *benzhydryl 8-methyl-3-oxo-4-(2-phenoxyacetamido)-2-oxa-6-thia-9-azabicyclo[3.2.2]nonane-8-carboxylate* (**10**), was also isolated. Amorphous foam. R_f (toluene/AcOEt 2:1) 0.20. IR (CH_2Cl_2): 3400, 1740, 1690, 1598, 1585, 1508, 1493, 1438, 1382, 1370, 1220, 1120, 1081, 1060, 1000, 912. ^{1}H-NMR (400 MHz, $CDCl_3$): 7.42–6.90 (*m*, 17 H); 5.36 (*dd*, *J* = 5, 1, 1 H); 5.10 (*t*, *J* = 2, 1 H); 4.50 (*s*, 2 H); 4.47 (*d*, *J* = 1, 1 H); 3.27 (*m*, 2 H); 1.63 (*s*, 3 H).

The lactone **10** was also detected (by NMR) as a by-product in the $NaBH_4$ reduction of **5** to **6** (see above).

Benzhydryl 4α-Methyl-7β-(2-phenoxyacetamido)-Δ²-cephem-4β-carboxylate[6] (**8**). A soln. of 6.81 g (21.6 mmol) of $Bu_4NF \cdot 3\ H_2O$ in 60 ml of THF was dehydrated by standing overnight in a refrigerator (+5°) with molecular sieves (*Merck* No. 5708; 0.4 nm, pearl form). Of the resulting soln., 30 ml were added to a soln. of 2.20 g (3.6 mmol) of **7** in 72 ml of abs. THF, and the mixture thus obtained was stirred under Ar at r.t. for 3 h. Thereafter, it was diluted with CH_2Cl_2 and washed twice with a buffer soln. (pH 8.0); the aq. parts were reextracted with CH_2Cl_2. The crude product obtained by evaporation of the CH_2Cl_2 (2.2 g) was chromatographed on 100 g of *Merck* silica gel (230–400 mesh ASTM) using toluene/AcOEt 19:1. First, a mixture (254 mg) of **8** and of its 6,7-*trans* isomer was eluted, followed by 926 mg of unchanged **7**. The mixture of **8** and its isomer was rechromatographed on 12 *Merck* anal. silica-gel plates in toluene/AcOEt 2:1, yielding 47 mg (4.4%) of the *trans*-isomer and 170 mg (15.8%, based on consumed **7**) of **8**.

8: Solid foam, enclosing solvent residues, even after extended drying at 30°/0.1 mbar. R_f (toluene/AcOEt 2:1) 0.38. $[\alpha]_D^{20} = -46.8 \pm 1.1°$ (*c* = 0.923, $CHCl_3$). IR (CH_2Cl_2): 3409, 3051, 1780, 1744, 1696, 1600, 1518, 1496, 1442, 1372, 1229, 1131, 1084. ^{1}H-NMR (400 MHz, $CDCl_3$): 7.35–6.84 (*m*, 17 H); 6.43 (*d*, *J* = 10, 1 H); 5.77 (*d*, *J* = 10, 1 H); 5.61 (*dd*, *J* = 9, 4.5, 1 H); 5.06 (*d*, *J* = 4.5, 1 H); 4.54 (*m*, 2 H); 1.61 (*s*, 3 H). MS (160°): 514 ($M^{+\cdot}$), 324, 303, 275, 167, 165, 152, 112, 107. Anal. calc. for $C_{29}H_{26}N_2O_5S$ (514.60): C 67.69, H 5.09, N 5.44, S 6.23; found: C 66.33, H 5.10, N 5.25, S 5.94; calc. for 3.17% of enclosed CH_2Cl_2: C 66.33, H 5.00, N 5.26, S 6.03.

*6,7-*trans-*Isomer of* **8** (absolute configuration at C(6) and C(7) unknown): Solid foam. R_f (toluene/AcOEt 2:1) 0.47. IR (CH_2Cl_2): 3400, 1773, 1740, 1691, 1598, 1520, 1495, 1377, 1230, 1130, 1117, 1080. ^{1}H-NMR (400 MHz, $CDCl_3$): 7.35–6.92 (*m*, 16 H); 6.81 (*s*, 1 H); 6.48 (*d*, *J* = 10, 1 H); 5.71 (*d*, *J* = 10, 1 H); 4.80 (*dd*, *J* = 7, 2, 1 H); 4.76 (*d*, *J* = 2, 1 H); 4.52 (*s*, 2 H); 1.71 (*s*, 3 H).

*1,4aα,5,7α,8,9a-Hexahydro-9aα-methyl-1,8-dioxo-7β-(2-phenoxyacetamido)-2*H*,6aα*H*-azeto[2,1-b][1,2]oxathiazino[5,6-d][1,3]thiazine 3,3-Dioxide* (**11**). In the above described preparation of **8**, the polar by-product **11** was isolated in one case by prep. TLC of the crude product. Starting with 183 mg of **7**, 30 mg of **11** were obtained from 5 *Merck* silica-gel plates (20 × 20 × 0.05 cm) with toluene/AcOEt 2:1. Amorphous foam. R_f (toluene/AcOEt 2:1) 0.10. IR (CH_2Cl_2): 3400, 1767, 1740, 1695, 1597, 1510, 1492, 1380, 1358, 1225, 1179, 1160, 1062, 980. ^{1}H-NMR (400 MHz, $CDCl_3$): 7.47 (*d*, *J* = 9, 1 H); 7.34–6.99 (*m*, 5 H); 5.64 (*dd*, *J* = 9, 4.5, 1 H); 5.12 (*d*, *J* = 15, 1 H); 5.10 (*d*, *J* = 4.5, 1 H); 4.58 (*m*, 2 H); 4.49 (*dd*, *J* = 4, 1.5, 1 H); 4.36 (*d*, *J* = 15, 1 H); 3.51 (*dd*, *J* = 15.5, 1.5, 1 H); 3.24 (*dd*, *J* = 15.5, 4, 1 H); 1.48 (*s*, 3 H); irradiation at 1.48→NOE at 5.10, 4.49, and 3.51. ^{13}C-NMR (100.6 MHz, $CDCl_3$): 188.3; 168.9; 163.8; 156.5; 129.7; 122.2; 114.6; 71.0; 66.7; 61.3; 60.3; 57.5; 54.4; 26.7; 16.7.

Sodium 4α-Methyl-7β-(2-phenoxyacetamido)-Δ²-cephem-4β-carboxylate[6] (**1a**). To a soln. of 170 mg (0.33 mmol) of **8** and of 53 mg (0.5 mmol) of anisole in 1.7 ml of CH_2Cl_2, 252 µl (3.3 mmol) of CF_3COOH were added at 0–5°. The resulting mixture was stirred under Ar in an ice/H_2O bath for 2.5 h. Thereafter, it was diluted with toluene and evaporated, and the residue was once more evaporated with CH_2Cl_2/toluene (removal of CF_3COOH).

Trituration of the residue with MeOH and filtration removed a small amount of a white precipitate. The filtrate was evaporated and the residue redissolved in 1.2 ml of MeOH. Then, 0.2 ml of 3M sodium hexanoate in MeOH were added. On addition of Et_2O to the resulting soln., **1a** precipitated. After filtration, it was washed with Et_2O to give 79 mg (64.6%) of a white, microcrystalline material. IR (KBr): 3428 (br.), 3050, 1754, 1680, 1625, 1535, 1496, 1442, 1387, 1355, 1236, 1085. ^{1}H-NMR (400 MHz, CD_3OD): 7.29–6.96 (*m*, 5 H); 6.28 (*d*, *J* = 10, 1 H); 5.83 (*d*, *J* = 10, 1 H); 5.48 (*d*, *J* = 4, 1 H); 5.04 (*d*, *J* = 4, 1 H); 4.59 (*s*, 2 H); 1.50 (*s*, 3 H); irradiation at 1.50→NOE at 5.83 and 5.04. FAB-MS (thioglycerol matrix): 371 ([*M* + H]$^+$) and several cluster ions, *e.g.* 393 ([*M* + H + Na]$^+$), 479 ([*M* + thioglycerol + H]$^+$), 501 ([479 + Na]$^+$), 763 ([*M* + *M* + Na]$^+$).

Stability tests: *a)* According to ^{1}H-NMR, a 0.74% soln. of **1a** in a D_2O-phosphate buffer, 'pD' 7.4, remained unchanged at r.t. for 12 days. At 37°, a soln. of **1a** in D_2O was unchanged after 24 h. *b)* $t_{1/2}$ at 37°: 50 h in a biological buffer soln., pH 7.4; 2 h in a phosphate buffer soln., pH 3.0; 50 h in human plasma (HPLC evidence).

Benzhydryl 3α-Hydroxy-4β-methyl-7β-(2-phenoxyacetamido)cepham-4α-carboxylate[6] (**14**). Reduction of 6.25 g (11.8 mmol) of **4** in a way similar to that described above for the preparation of **6** (60 ml of DMF, 15 ml of AcOH, 840 mg of $NaBH_4$; 90 min at 0°) gave, after chromatography of the crude product on 300 g of *Merck* silica gel *60* with toluene/AcOEt 19:1, 3.48 g (55.5%) of **14**. It crystallized from CH_2Cl_2/pentane in white needles including some CH_2Cl_2. M. p. (65°→)71°. R_f (toluene/AcOEt 1:1) 0.52. IR (CH_2Cl_2): 3406, 3053, 1772, 1695, 1599, 1520, 1495, 1441, 1366, 1236, 1173, 1133, 1062, 956. ^{1}H-NMR (400 MHz, $CDCl_3$): 7.43–6.92 (*m*, 17 H); 5.55 (*dd*, *J* = 9.5, 4.5, 1 H); 4.86 (*d*, *J* = 4.5, 1 H); 4.54 (*m*, 2 H); 3.83 (*m*, 1 H); 3.45 (*d*, *J* = 10, 1 H); 2.78 (*m*, 2 H); 2.01 (*s*, 3 H). FAB-MS: 533 ([*M* + H]$^+$). Anal. calc. for $C_{29}H_{28}N_2O_6S$ (532.61): C 65.40, H 5.30, N 5.26, O 18.02, S 6.02; found: C 64.44, H 5.20, N 5.31, O 17.93, S 6.12, Cl 1.10.

Benzhydryl 3α-(Methansulfonyloxy)-4β-methyl-7β-(2-phenoxyacetamido)cepham-4α-carboxylate[6] (**15**). To a soln. of 1.06 g (2.0 mmol) of **14** and 0.67 ml (4.8 mmol) of Et_3N in 15 ml of CH_2Cl_2, 0.23 ml (3 mmol) of mesyl chloride were added at 0° and stirred at r.t. for 75 min. Thereafter, the mixture was diluted with CH_2Cl_2 and successively washed with H_2O and aq. $NaHCO_3$ soln.; the aq. parts were reextracted with CH_2Cl_2. The crude product as obtained by evaporation of the combined org. parts was chromatographed on 30 g of *Merck* silica gel *60* with toluene/AcOEt 9:1. A total of 1.04 g (85.2%) of **15** was collected in several fractions as a solid foam retaining traces of residual solvents (*e.g.* toluene), even after extensive drying under high vacuum. R_f (toluene/AcOEt 2:1) 0.43. $[\alpha]_D^{20}$ = +77 ± 1° (*c* = 1.035, $CHCl_3$). IR (CH_2Cl_2): 3406, 3055, 1777, 1734, 1696, 1600, 1519, 1495, 1441, 1351, 1238, 1210, 1178, 1133, 1082, 949, 871, 821. ^{1}H-NMR (400 MHz, $CDCl_3$): 7.42–6.93 (*m*, 17 H); 5.55 (*dd*, *J* = 9.5, 4.5, 1 H); 5.32 (*d*, *J* = 4.5, 1 H); 4.84 (*dd*, *J* = 10, 3.5, 1 H); 4.56 (*m*, 2 H); 3.45 (*dd*, *J* = 13.5, 10, 1 H); 2.98 (*dd*, *J* = 13.5, 3.5, 1 H); 2.75 (*s*, 3 H); 2.06 (*s*, 3 H); irradiation at 2.06→NOE at 4.88; irradiation at 3.45→NOE at 2.98 and 5.32. FAB-MS: 611 ([*M* + H]$^+$). Anal. calc. for $C_{30}H_{30}N_2O_8S_2$ (610.70): C 59.00, H 4.95, N 4.59, S 10.50; found: C 59.65, H 5.18, N 4.39, S 10.29.

4α-[(Benzhydryloxy)carbonyl]-4β-methyl-7β-(2-phenoxyacetamido)-Δ^2-cephem 1-Oxide[6]) (**19**). To a soln. of 10.0 g (*ca.* 20 mmol) of benzhydryl 7β-(2-phenoxyacetamido)-Δ^3-cephem-4-carboxylate (**17**) [7a] [8] in 200 ml of CH_2Cl_2, 4.22 g of 90% *m*-chloroperbenzoic acid were added in several portions, while stirring in an ice/H_2O bath. After a total of 90 min, the resulting mixture (with precipitated *m*-chlorobenzoic acid) was diluted with more CH_2Cl_2 and successively washed with H_2O, 8% aq. $NaHCO_3$ soln., and sat. brine; the aq. parts were reextracted with CH_2Cl_2. Evaporation of CH_2Cl_2 from the combined org. parts left a crystalline residue which, after washing with Et_2O, afforded 9.5 g (92%) of white crystals of *4-[(benzhydryloxy)carbonyl]-7β-(2-phenoxyacetamido)-Δ^3-cephem-4-carboxylate 1-oxide*[6]) (**18**). This material was used in the next step without any further purification. R_f (toluene/AcOEt 1:1) 0.19. IR (CH_2Cl_2): 3400, 1791, 1720, 1687, 1635, 1595, 1505, 1485, 1390, 1220, 1160, 1100, 1077, 1055, 1038, 980, 960.

To **18** (13.3 g, 25.7 mmol) in 75 ml of DMF and 625 ml of MeCN, 20.6 ml of MeI were added and stirred with 3.6 g of finely pulverized K_2CO_3 at r.t. After 15 h, more MeI (10 ml) was added and stirring was continued for another 6 h. The resulting brown mixture was evaporated and the residue partitioned between AcOEt and H_2O; the org. phase was washed with sat. brine, and all aq. parts were reextracted with AcOEt. Evaporation of AcOEt from the combined org. parts left a brown foam (15 g) which was chromatographed on 250 g of *Merck* silic gel *60*. After several fractions with toluene/AcOEt 9:1, **19** was eluted with toluene/AcOEt 4:1 and 3:1 as a colourless foam (6.0 g, 43.9%). R_f (toluene/AcOEt 1:1) 0.35. IR (CH_2Cl_2): 3360, 1770, 1730, 1680, 1591, 1503, 1482, 1365, 1315, 1204, 1110, 1071, 1052, 1020. ^{1}H-NMR (360 MHz, $CDCl_3$): 8.18 (*d*, *J* = 10.2, 1 H); 7.41–6.72 (*m*, 18 H); 6.09 (*dd*, *J* = 10.2, 5, 1 H); 4.58 (*AB*, *J* = 15, 2 H); 4.52 (*d*, *J* = 5, 1 H); 2.13 (*s*, 3 H).

Benzhydryl 4β-Methyl-7β-(2-phenoxyacetamido)-Δ^2-cephem-4α-carboxylate[6]) (**16**). To a soln. of 5.5 g (10.37 mmol) of **19** in 45 ml of DMF, 1.82 ml (20.86 mmol) of PCl_3 were slowly added at −10° and stirred under Ar at −10° for another 15 min. Then, the mixture was poured on crashed ice, the product extracted into AcOEt, and the org. part successively washed with 8% aq. $NaHCO_3$ soln., H_2O, and sat. brine; all aq. parts were reextracted with

AcOEt. Evaporation of the combined org. extracts, finally under high vacuum, afforded 5.2 g of a brown residue which was chromatographed on 150 g of *Merck* silica gel *60* using toluene/AcOEt 9:1. The title ester was collected in several fractions as a solid foam (4.2 g, 78.7%), crystallizing from AcOEt on addition of petroleum ether. M.p. 98–102° (AcOEt/petroleum ether). R_f (toluene/AcOEt 1:1) 0.59. IR (CH_2Cl_2): 3400, 1775, 1740, 1694, 1599, 1518, 1494, 1440, 1382, 1245, 1228, 1124, 1082, 1065. ^{1}H-NMR (100 MHz, $CDCl_3$): 7.46–6.80 (*m*, 17 H); 6.3 (*d*, J = 10.1, 1 H); 5.82 (*d*, J = 10.1, 1 H); 5.7 (*dd*, J = 9, 5, 1 H); 5.14 (*d*, J = 5, 1 H); 4.56 (*s*, 2 H); 1.94 (*s*, 3 H). Anal. calc. for $C_{29}H_{26}N_2O_5S$ (514.60): C 67.69, H 5.09, N 5.44, S 6.23; found: C 67.84, H 5.25, N 5.59, S 6.22.

Crystal-Structure Analysis of **16**. Crystal Data: Monoclinic, space group C_2, a = 17.997(6), b = 10.074(5), c = 15.918(6) Å, β = 91.22(5)°, Z = 4. A *Philips PW1100* automatic diffractometer was used for data collection with MoK_α radiation and graphite monochromator. The intensities of 4462 independent reflections with $\theta < 30°$ were measured of which 3653 were classified as observed with $I > 2\sigma(I)$. The structure was solved by direct methods using the MULTAN 78 program [10]. From a difference *Fourier* map, 21 of 26 H-atoms were found, the coordinates of the remaining were calculated assuming tetrahedral geometry. The structure was refined by full matrix least squares calculations with anisotropic (isotropic for H-atoms) thermal parameters to a final R value of 0.081.

Table 2. *Positional Parameters and their Estimated Standard Deviations for Non-H-Atoms of* **16**

Atom	x	y	z	Atom	x	y	z
S(1)	0.3016(1)	0.3790(3)	0.0432(1)	C(20)	0.2370(4)	0.6867(7)	–0.3415(4)
C(2)	0.3177(5)	0.5521(9)	0.0365(6)	C(21)	0.2901(6)	0.735(1)	–0.3980(6)
C(3)	0.3427(4)	0.6203(8)	–0.0291(5)	C(22)	0.2882(7)	0.679(1)	–0.4792(7)
C(4)	0.3627(4)	0.5623(7)	–0.1123(4)	C(23)	0.2359(7)	0.583(1)	–0.5076(7)
N(5)	0.3738(3)	0.4183(5)	–0.1040(3)	C(24)	0.1854(6)	0.540(1)	–0.4506(6)
C(6)	0.3233(4)	0.3253(8)	–0.0623(5)	C(25)	0.1863(6)	0.593(1)	–0.3712(6)
C(7)	0.3876(4)	0.2204(7)	–0.0650(5)	N(26)	0.4087(3)	0.1537(6)	0.0101(4)
C(8)	0.4366(4)	0.3361(7)	–0.0929(4)	C(27)	0.4053(8)	0.0204(5)	0.0116(4)
C(9)	0.4350(5)	0.6271(8)	–0.1424(6)	O(28)	0.3914(3)	–0.0510(6)	–0.0484(5)
C(10)	0.2957(4)	0.5806(7)	–0.1740(4)	C(29)	0.4251(4)	–0.0434(7)	0.0954(4)
O(11)	0.2486(3)	0.4997(6)	–0.1863(4)	O(30)	0.4509(3)	0.0542(5)	0.1524(3)
O(12)	0.2965(2)	0.7033(5)	–0.2058(3)	C(31)	0.4768(4)	0.0126(8)	0.2287(5)
C(13)	0.2301(4)	0.7424(7)	–0.2533(4)	C(32)	0.5051(5)	0.106(1)	0.2812(5)
C(14)	0.2262(4)	0.8915(7)	–0.2556(4)	C(33)	0.5320(5)	0.074(1)	0.3617(6)
C(15)	0.2864(4)	0.9732(8)	–0.2472(4)	C(34)	0.5282(5)	–0.055(1)	0.3897(6)
C(16)	0.2760(5)	1.1109(8)	–0.2539(5)	C(35)	0.4992(5)	–0.149(1)	0.3373(5)
C(17)	0.2069(5)	1.1710(9)	–0.2660(6)	C(36)	0.4718(4)	–0.1144(9)	0.2570(5)
C(18)	0.1499(6)	1.081(1)	–0.2734(7)	O(37)	0.5016(3)	0.3536(6)	–0.1068(3)
C(19)	0.1580(5)	0.9464(9)	–0.2670(6)				

Table 3. *Positional Parameters and their Estimated Standard Deviations for H-Atoms of* **16**

Atom	x	y	z	Atom	x	y	z
H(38)	0.305(4)	0.614(9)	0.089(5)	H(51)	0.328(5)	0.808(9)	–0.374(5)
H(39)	0.349(5)	0.724(9)	–0.020(5)	H(52)	0.327(5)	0.713(9)	–0.525(5)
H(40)	0.382(4)	0.134(9)	–0.106(5)	H(53)	0.234(5)	0.539(9)	–0.565(5)
H(41)	0.271(5)	0.306(9)	–0.095(5)	H(54)	0.145(5)	0.465(9)	–0.472(5)
H(42)	0.450(4)	0.587(9)	–0.202(5)	H(55)	0.148(5)	0.563(9)	–0.325(5)
H(43)	0.479(5)	0.609(9)	–0.097(5)	H(56)	0.421(5)	0.206(9)	0.066(5)
H(44)	0.426(5)	0.734(9)	–0.148(5)	H(57)	0.468(5)	–0.116(9)	0.087(5)
H(45)	0.182(5)	0.701(9)	–0.223(5)	H(58)	0.376(5)	–0.091(9)	0.121(5)
H(46)	0.339(5)	0.930(9)	–0.239(5)	H(59)	0.509(5)	0.205(9)	0.259(5)
H(47)	0.324(5)	1.175(9)	–0.247(5)	H(60)	0.555(5)	0.148(9)	0.402(5)
H(48)	0.199(5)	1.265(9)	–0.271(5)	H(61)	0.547(5)	–0.083(9)	0.448(5)
H(49)	0.095(5)	1.119(9)	–0.285(5)	H(62)	0.496(5)	–0.250(9)	0.358(5)
H(50)	0.110(5)	0.883(9)	–0.274(5)	H(63)	0.450(5)	–0.189(9)	0.216(5)

Table 4. *Bond Distances* [Å] *in* **16**[a])

S(1)–C(2)	1.771(7)	C(10)–O(12)	1.335(9)	C(23)–C(24)	1.37(2)
S(1)–C(6)	1.817(7)	O(12)–C(13)	1.453(8)	C(24)–C(25)	1.37(2)
C(2)–C(3)	1.34(1)	C(13)–C(14)	1.50(1)	N(26)–C(27)	1.344(9)
C(3)–C(4)	1.45(1)	C(13)–C(20)	1.52(1)	C(27)–O(28)	1.219(9)
C(4)–N(5)	1.47(1)	C(14)–C(15)	1.36(1)	C(27)–C(29)	1.52(1)
C(4)–C(9)	1.54(1)	C(14)–C(19)	1.35(1)	C(29)–O(30)	1.410(9)
C(4)–C(10)	1.55(1)	C(15)–C(16)	1.40(1)	O(30)–C(31)	1.356(9)
N(5)–C(6)	1.471(9)	C(16)–C(17)	1.39(1)	C(31)–C(32)	1.35(1)
N(5)–C(8)	1.409(9)	C(17)–C(18)	1.37(1)	C(31)–C(36)	1.36(1)
C(6)–C(7)	1.57(1)	C(18)–C(19)	1.36(2)	C(32)–C(33)	1.40(1)
C(7)–C(8)	1.53(1)	C(20)–C(21)	1.41(1)	C(33)–C(34)	1.38(2)
C(7)–N(26)	1.417(9)	C(20)–C(25)	1.39(1)	C(34)–C(35)	1.36(2)
C(8)–O(37)	1.208(9)	C(21)–C(22)	1.41(2)	C(35)–C(36)	1.40(1)
C(10)–O(11)	1.189(9)	C(22)–C(23)	1.42(2)		

[a]) Numbers in parentheses are estimated standard deviations in the least significant digits.

Sodium 4β-Methyl-7β-(2-phenoxyacetamido)-Δ²-cephem-4α-carboxylate[6]) (**2a**). To a soln. of 1.03 g (2 mmol) of **16** and of 318 mg (2.94 mmol) of anisole in 10 ml of CH_2Cl_2, 1.5 ml of CF_3COOH were added, while cooling the mixture in an ice/H_2O bath. After 2.5 h at 0–5° and 0.5 h at r.t., toluene was added and the mixture evaporated, finally once more with toluene/$CHCl_3$ 1:1 and with Et_2O. The residue thus obtained (1.3 g) was dissolved in a small volume of MeOH, and 1.3 ml of 3M sodium hexanoate in MeOH was added. Addition of Et_2O at 0° caused precipitation of **2a** as a white, microcrystalline powder which was washed with Et_2O (540 mg, 73%). IR (nujol): 3400 (br.), 1750 (br.), 1680 (br.), 1600, 1530, 1495, 1390, 1354, 1240, 1211, 1065. ^{1}H-NMR (360 MHz, D_2O): 7.43–7.00 (*m*, 5 H); 6.27 (*d*, *J* = 10.2, 1 H); 5.95 (*d*, *J* = 10.2, 1 H); 5.38 (*d*, *J* = 5, 1 H); 5.12 (*d*, *J* = 5, 1 H); 4.76 (*s*, 2 H); 1.76 (*s*, 3 H).

Stability tests: *a)* According to ^{1}H-NMR, a 0.74% soln. of **2a** in a D_2O-phosphate buffer, 'pD' 7.4, remained unchanged at r.t. over 12 days. *b)* $t_{½}$ at 37°: 50 h in a biological buffer soln., pH 7.4; 50 h in a phosphate buffer, pH 3.0; 50 h in human plasma (all HPLC evidence).

Benzhydryl 3-Methoxy-7β-(2-phenoxyacetamido)-Δ²-cephem-4α- (**20**) *and -4β-carboxylate*[6]) (**21**). A soln. of 13.26 g (25 mmol) of benzhydryl 3-methoxy-7β-(2-phenoxyacetamido)-Δ³-cephem-4-carboxylate (**24**) [7a] in 100 ml of CH_2Cl_2 containing 101 mg (1.0 mmol) of Et_3N was stirred at r.t. for 6 h. The resulting equilibration mixture was washed 3 times with 100 ml each of 0.01N aq. HCl and the org. part evaporated to a foam (13 g). A part (8.0 g) of this residue was chromatographed on 220 g of *Merck* silica gel *60* with toluene/AcOEt/AcOH 90:10:0.5. Successively, **20** (5.69 g, 71%) and **21** (0.16 g, 2.0%) were eluted followed by the unchanged **24** (1.75 g, 22%). No other products were detected under these mild isomerization conditions.

For the preparation of a larger amount of **21**, 350 g of **24** were isomerized as above. From the crude product, most of **20** and **24** were crystallized by addition of 3 l of Et_2O to the oily material. The crystals were filtered off, the mother liquor – enriched in **21** – was evaporated and the residue (16 g) chromatographed as described above, yielding 6.37 g (1.8%) of pure **21**.

Benzhydryl 3-Methoxy-7β-(2-phenoxyacetamido)-Δ²-cephem-4α-carboxylate[6]) (**20**). M.p. 121–122° (EtOH/toluene 4:1). R_f (toluene/AcOEt 4:1) 0.44. $[\alpha]_D^{20} = +281 \pm 1.0°$ (*c* = 1.042, dioxane). IR (CH_2Cl_2): 3404, 3056, 1785, 1747, 1696, 1600, 1519, 1495, 1440, 1324, 1209, 1163, 1083, 1064, 988, 838. ^{1}H-NMR (100 MHz, $CDCl_3$): 7.58–6.86 (*m*, 17 H); 5.70 (*dd*, *J* = 9, 5, 1 H); 5.26 (*d*, *J* = 5, 1 H); 5.12 (*d*, *J* = 1, 1 H); 5.0 (*d*, *J* = 1, 1 H); 4.55 (*s*, 2 H); 3.52 (*s*, 3 H). ^{13}C-NMR (25.2 MHz, $CDCl_3$; only relevant resonances): 87.7 (C(2), 1 C); 143.3 (C(3), 1 C); 51.8 (C(4), 1 C); 53.8 (C(6), 1 C); 58.9 (C(7), 1 C); 55 (CH_3O, 1 C). Anal. calc. for $C_{29}H_{26}N_2O_6S$ (530.60): C 65.65, H 4.94, N 5.28, S 6.04; found: C 65.76, H 4.90, N 5.43, S 5.93.

Benzhydryl 3-Methoxy-7β-(2-phenoxyacetamido)-Δ²-cephem-4β-carboxylate[6]) (**21**). Amorphous foam. R_f (toluene/AcOEt 4:1) 0.27. $[\alpha]_D^{20} = -3.6 \pm 0.9°$ (*c* = 1.133, dioxane). IR (CH_2Cl_2): 3407, 3051, 1785, 1750, 1695, 1600, 1518, 1495, 1440, 1209, 1177, 1116, 1083, 1064, 1005, 837. ^{1}H-NMR (360 MHz, $CDCl_3$): 7.38–6.86 (*m*, 18 H); 5.63 (*ddd*, *J* = 10, 5, 2, 1 H); 5.32 (*s*, 1 H); 5.06 (*d*, *J* = 5, 1 H); 4.61 (*d*, *J* = 2, 1 H); 4.54 (*s*, 2 H); 3.50 (*s*, 3 H). ^{13}C-NMR (90.5 MHz, $CDCl_3$; only relevant resonances): 88.9 (C(2), 1 C); 147.9 (C(3), 1 C); 56.4 (C(4), 1 C); 57.3 (C(6), 1 C); 57.8 (C(7), 1 C); 55.3 (CHO_3, 1 C). FAB-MS: 531 ($[M + H]^+$). Anal. calc. for $C_{29}H_{26}N_2O_6S$ (530.60): C 65.65, H 4.94, N 5.28, S 6.04; found: C 65.76, H 5.07, N 5.12, S 5.80.

3-Methoxy-7β-(2-phenoxyacetamido)-Δ²-cephem-4α-carboxylic Acid[6] (**22**). To a stirred soln. of 13.26 g (25.0 mmol) of **20** and 6.0 g (55.6 mmol) of anisole in 25 ml of CH_2Cl_2, 44.5 g (390 mmol) of CF_3COOH was added at 0° within 10 min. After another 45 min of stirring at 0°, the product was precipitated by adding 200 ml of $(i\text{-}Pr)_2O$ and cooling. The crude **22** was filtered off, washed with $(i\text{-}Pr)_2O$, and dried *i.v.* at r.t. (8.95 g). The product was suspended in 80 ml of CH_2Cl_2, filtered, and crystallized from i-PrOH affording 5.96 g (65%) of pure **22** as white crystals. M.p. 178–179° (dec.). R_f (CH_2Cl_2/MeOH/AcOH 75:5:0.5) 0.33. $[\alpha]_D^{20} = 327.3 \pm 0.9°$ (c = 1.081, dioxane). IR (KBr): 3305, 1768, 1725, 1689, 1623, 1597, 1535, 1488, 1443, 1330, 1230, 1205, 1165, 1063, 1015, 763, 698. ^{1}H-NMR (360 MHz, (D_6)DMSO): 13.5 (br. *s*, 1 H); 9.08 (*d*, J = 10, 1 H); 7.35–6.92 (*m*, 15 H); 5.55 (*s*, 1 H); 5.50 (*dd*, J = 10, 5, 1 H); 5.17 (*d*, J = 5, 1 H); 4.70 (*s*, 1 H); 4.65 (*AB*, J = 15, 2 H); 3.55 (*s*, 3 H). ^{13}C-NMR (90.5 MHz, (D_6)DMSO): 88.2 (C(2), 1 C); 143.5 (C(3), 1 C); 51.8 (C(4), 1 C); 53.4 (C(6), 1 C); 59.5 (C(7), 1 C); 164.5 (C(8), 1 C); the signals due to the side chains are not listed. FAB-MS: 365 ($[M + H]^+$). Anal. calc. for $C_{16}H_{16}N_2O_6S$ (364.37): C 52.74, H 4.43, N 7.69, S 8.80; found: C 52.61, H 4.45, N 7.72, S 8.83.

Stability tests: $t_{1/2}$ at 37°: 100 h in a biological buffer, pH 7.4; 9 h in human plasma (both HPLC evidence).

3-Methoxy-7β-(2-phenoxyacetamido)-Δ²-cephem-4β-carboxylic Acid[6] (**23**). As for **22** with 4.61 g (8.7 mmol) of **21**, 2.04 g (18.9 mmol) of anisole in 9 ml of CH_2Cl_2, and 15.4 g (135 mmol) of CF_3COOH. The crude **23** was precipitated by slow addition of 215 ml of $(i\text{-}Pr)_2O$ while keeping the temp. below 5°. The white-yellow crystals (2.50 g) obtained by filtration were recrystallized from CH_2Cl_2 yielding 2.15 g (68%) of **23** as small, white crystals. M.p. 161–162° (dec.). R_f (CH_2Cl_2/MeOH/AcOH 75:25:0.5) 0.33. $[\alpha]_D^{20} = +50.3 \pm 0.9°$ (c = 1.077, dioxane). IR (KBr): 3315, 1777, 1752, 1675, 1617, 1600, 1542, 1495, 1400, 1210, 1175, 1004, 756, 690. ^{1}H-NMR (360 MHz, $CDCl_3/CD_3OD$): 7.38–6.97 (*m*, 5 H); 5.63 (*dd*, J = 5, 2, 1 H); 5.45 (*s*, 1 H); 5.10 (*d*, J = 5, 1 H); 4.55 (*d*, J = 2, 1 H); 3.63 (*s*, 3 H); 4.60 (*s*, masked by the HOD signal, 2 H). ^{13}C-NMR (90.5 MHz, (D_6)DMSO): 87.7 (C(2), 1 C); 145.9 (C(3), 1 C); 55.8 (C(4), 1 C); 55.5 (C(6), 1 C); 57.8 (C(7), 1 C); 166.2 (C(8), 1 C); the remaining signals are not listed. FAB-MS: 365 ($[M + H]^+$). Anal. calc. for $C_{16}H_{16}N_2O_6S$ (364.37): C 52.74, H 4.43, N 7.69, S 8.80; found: C 52.03, H 4.48, N 7.72, S 8.40.

Stability tests: $t_{1/2}$ at 37°: > 100 h in a biological buffer, pH 7.4; 50 h in human plasma (both HPLC evidence).

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