A Novel Oral Carbapenem CS-834: Chemical Stability of Pivaloyloxymethyl Esters of Carbapenems and Cephalosporins in Phosphate Buffer Solution

Sir:

Carbapenems are a new type of β-lactam antibiotic. They have a broad antimicrobial spectrum and potent bactericidal activity against both Gram-positive and Gram-negative organisms.[1,2] They are highly stable against hydrolysis by β-lactamases. Because of these attractive features, both academic and industrial scientists all over the world have made tremendous efforts to develop synthetic carbapenems for clinical use. Imipenem,[3] panipenem[4] and meropenem[5] have successively been launched on the market and several compounds are under clinical evaluation.[6~8] However, most of the compounds are developed for injectable use, rather than oral administration. Chemical and biological instability have been recognized as serious problems for the development of orally active carbapenems.

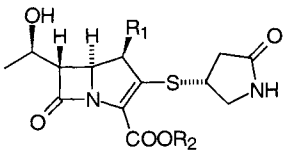
On the other hand, orally active cephalosporins have been widely investigated, and many compounds have been developed for clinical use. Most of these compounds are ester prodrugs that are enzymatically converted to their parent acids after absorption from the intestinal tract. But their chemical stability is limited by characteristic degradation through Δ^3–Δ^2 isomerization of cephem double bond.[9] Recently, we reported that penem esters have higher stability than cephalosporin esters because the penem double bond can not be isomerized.[10]

Here, we have compared the chemical stability of a novel oral 1β-methyl carbapenem CS-834, its 1-H derivative and cephalosporin esters.[11,12] It was found that 1β-methyl and 1-H carbapenem esters are more stable than cephalosporin esters in neutral phosphate buffer. The difference in the degradation mechanisms of these β-lactams is discussed.

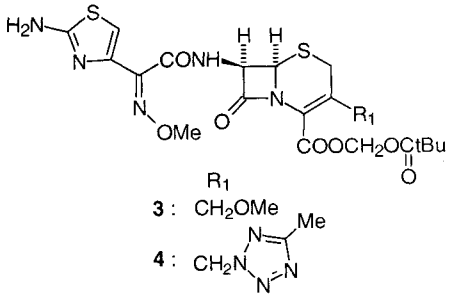
We have selected four test compounds, 1β-methyl carbapenem **1** (CS-834), 1-H carbapenem **2**, 3-methoxymethyl cephalosporin **3** and 3-tetrazolylmethyl cephalosporin **4**, as shown in Scheme 1.[11~14] The ester part of compounds **1**~**4** is fixed to a pivaloyloxymethyl group that is a standard promoiety of ester-type prodrugs for oral use. Cephalosporins **3** and **4** have a 2-(2-aminothiazol-3-yl)-2-methoxyiminoacetamide group at the C-7 position that is common among the third generation cephalosporins. Carbapenems **1** and **2** are newly synthesized compounds directed towards development as orally active carbapenems.[11,12]

Compounds **1**~**4** were dissolved in phosphate buffer at pH 6.86 and their degradation was followed by HPLC. The initial concentration of each ester was 50 μg/ml. Results are shown in Figure 1. Compounds **1**~**4** apparently decomposed according to first order kinetics. The degradation rate constants of esters **1**~**4** were 0.031, 0.084, 0.132 and 0.837 hr^{-1}, respectively. Contrary to

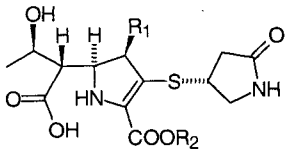
Scheme 1.



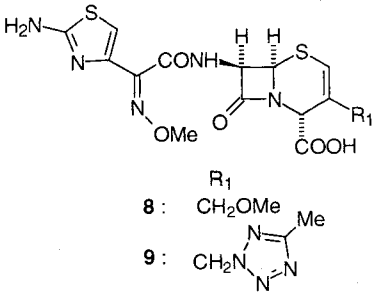
	R_1	R_2
1:	Me	$CH_2OCOtBu$
2:	H	$CH_2OCOtBu$
5:	Me	H
6:	H	H



	R_1
3:	CH_2OMe
4:	



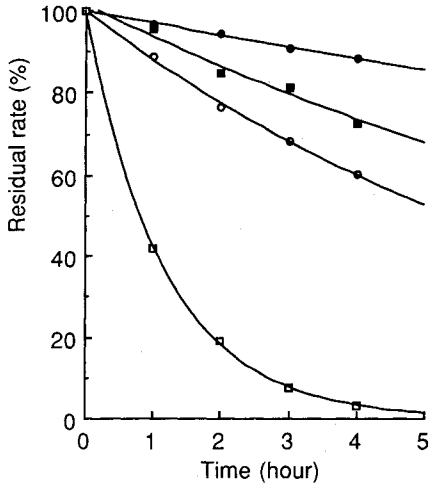
	R_1	R_2
7:	Me	$CH_2OCOtBu$



	R_1
8:	CH_2OMe
9:	

Fig. 1. Chemical stability of carbapenem and cephalosporin esters **1**~**4** in a phosphate buffer solution of pH 6.86 at 37°C.

● 1-Me carbapenem **1**, ■ 1-H carbapenem **2**, ○ cephalosporin **3**, □ cephalosporin **4**.



the conventional view that carbapenems are unstable even under neutral conditions, both carbapenems **1** and **2** showed higher stability than cephalosporins **3** and **4**.

1β-Methyl carbapenem **1** was more stable than 1-H carbapenem **2**, probably due to the preferable contribution of the 1β-methyl group which sterically protects the β-lactam ring against hydrolysis.[15] However, the contribution of the 1β-methyl group is not significant in the chemical hydrolysis although enzymatic degradation of carbapenems with dehydropeptidase-I is dramatically improved by the 1β-methyl group.[2,12]

Degradation of carbapenem esters **1** and **2** did not yield their parent acids **5** and **6**, respectively. This means that the degradation mechanism of carbapenem esters does not involve simple hydrolysis of the ester moiety. They are likely to decompose through hydrolysis of the β-lactam ring, which is highly activated by fusion with a five-membered ring.[16,17]

The initial hydrolysis product **7** from 1β-methyl carbapenem ester **1**, which eluted a little faster than ester **1** on HPLC analysis, was isolated from the degradation mixture. Product **7** was characterized by UV absorption (λ_{max} 330 nm, S–C=C–COOC), ^{1}H-NMR (δ 1.06 ppm, pivaloyl group) and MS (m/z 459, $[M+H]^+$) spectroscopy. 1-H Carbapenem **2** degrades in the same manner, but a little faster than 1β-methyl carbapenem **1**.

As reported in previous papers, on the other hand, degradation products from cephalosporin esters **3** and **4** were Δ^2-cephalosporin acids **8** and **9**, which are produced through Δ^3–Δ^2 isomerization of the cephem double bond followed by hydrolysis of the Δ^2-cephem esters.[9,18] The sulphur atom at the C-1 position helps the isomerization by increasing the acidity of the proton at the C-2 position which is removed as the initial step of the isomerization. Hydrolysis of the β-lactam ring was negligible.

The isomerization of carbapenem double bond probably is not involved in the degradation mechanism, because the acidity of the proton at the C-1 position is not high enough to promote the isomerization under neutral conditions.

We have investigated orally active β-lactam antibiotics, in which our attention has been focused on chemical and biological stability, in order to attain effective bioavailability. The reference compound, 3-methoxymethyl cephalosporin **3**, used in this experiment has high stability among cephalosporin esters. Degradation through Δ^3–Δ^2 isomerization seems to be a fatal problem for the development of ester prodrugs of cephalosporins.[19] This characteristic mechanism of degradation has not been observed in the case of penem and carbapenem esters. Hydrolysis of the β-lactam ring in carbapenems has been less of a problem than expected under neutral conditions. The ester-type prodrugs of carbapenems could be a new promising area for development of potent orally active β-lactam antibiotics.[11,12]

In conclusion, we have found a distinct difference in the degradation mechanisms of carbapenem and cephalosporin esters. Carbapenem esters decompose *via* hydrolysis of the β-lactam ring, and they are more stable, even in the case of 1-H carbapenem, than cephalosporin esters which decompose through Δ^3–Δ^2 isomerization of the cephem double bond.

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