SYNTHESIS AND BIOLOGICAL ACTIVITY OF 2-LACTONYL PENEMS

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(Received for publication February 19, 1988)

A series of potent antibacterial agents have been prepared. These agents are penems carrying a lactone ring in the C-2 position. Excellent activity against Gram-positive and Gram-negative organisms — except *Pseudomonas aeruginosa* — was found.

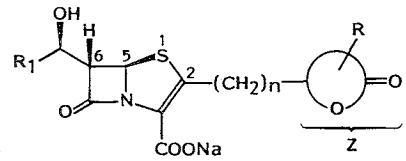
The penems are highly potent broad-spectrum β-lactam antibiotics closely related to the naturally occurring penicillins, cephalosporins and carbapenems. They are not, however, elaborated by microorganisms, but prepared synthetically, preferably by total synthesis[1,2]. Work done mainly by industrial research chemists, and buried to a large extent in the patent literature, resulted in the preparation of penems bearing different substituents, for the most part in the C-2 position. Substituents at C-6 were limited to hydroxymethyl or hydroxyethyl groups; different substitution pattern at C-6 have been shown to result in chemically rather unstable compounds, or in agents lacking any relevant antibacterial activity[3~5].

Limiting the C-6 substituent to the foregoing groups, we started a programme to identify new 2-substituted compounds, distinguishable from the well known 2-aminoalkyl-[6], 2-alkylthio-[7~9], 2-heterocyclyl-[10,11], 2-oxymethyl-[12,13] and 2-oxy-penems[14]. In this paper we report on the preparation and on the biological activity of a series of 2-lactonyl penems of the general formula **1**[15].

Chemistry

The lactonyl penems **1** were synthesized according to WOODWARD's method[1], following the general synthetic pathway depicted in Scheme 1. The common starting material for the synthesis of all penems listed in Table 3 was the silver thiolate **2**[16,17], which after acylation with the appropriate acid chlorides **3** yielded phosphoranes **4**. With compounds of type **4a** (R_1=H; silyl protection on 1'-OH) the two diastereomeric phosphoranes **4** could be separated chromatographically starting with racemic acid chlorides **3**, whereas with compounds of type **4b** (R=CH_3; allyloxycarbonyl protection on 1'-OH) non satisfactory separation was possible, except for the diastereomeric mixture **4b** leading to the penems **30** and **31**. The

Fig. 1. Structures of **1a** and **1b**.

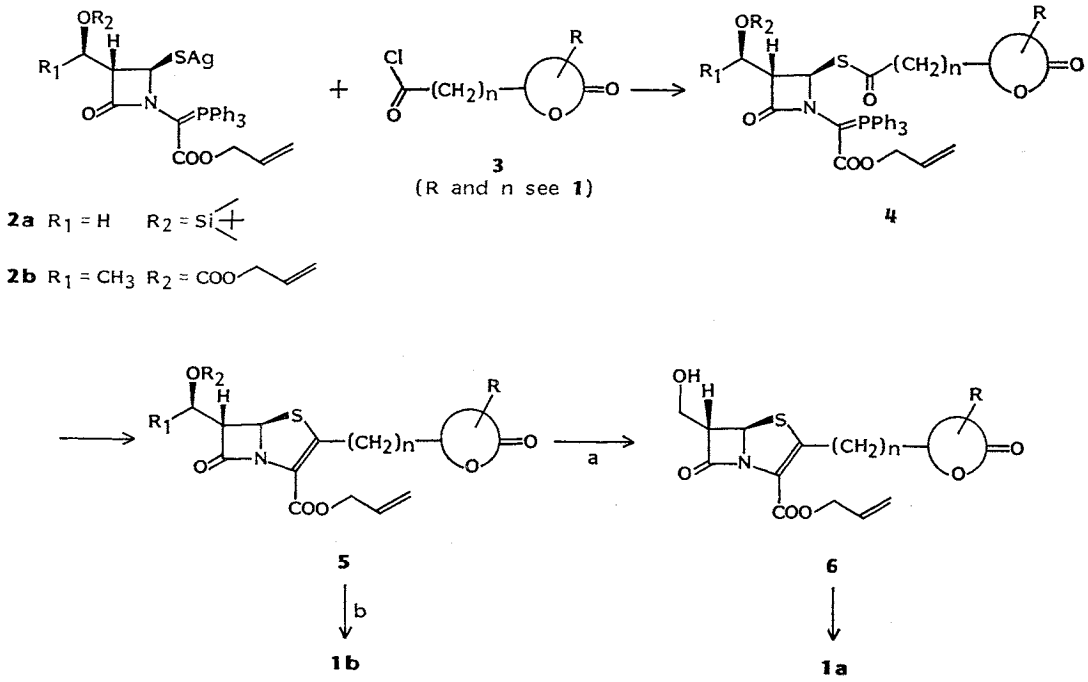


1a R_1 = H

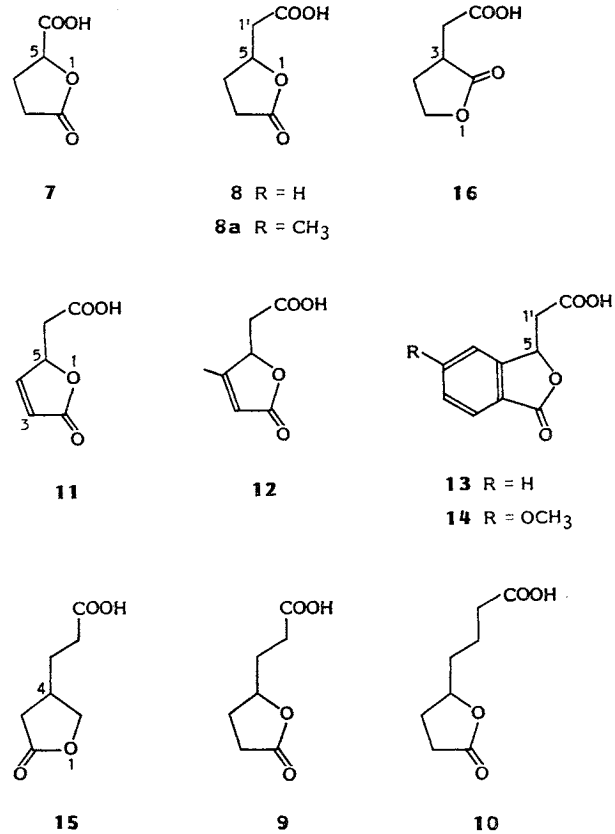
1b R_1 = CH_3

R = H, CH_3, condensed aryl, double bond

Scheme 1. Synthesis of lactonyl penems.



Scheme 2. Lactonic acids 7~16.



penem esters **5** were formed by Wittig cyclization of **4**. Desilylation of **5a** to **6** followed by deblocking of the protective allyloxycarbonyl group in **5b** and in **6** was done by well-established methods.

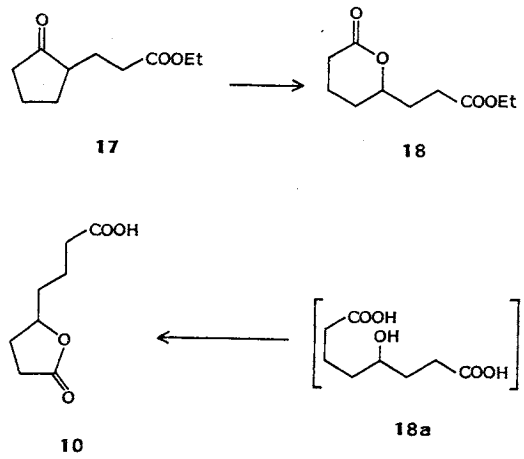
A survey of the lactonic acids **7**~**16** used in this work is shown in Scheme 2. Scheme 3 outlines the synthesis of **10**. Details of the preparation of **7**~**16** are given in the Experimental part.

Absolute Configuration

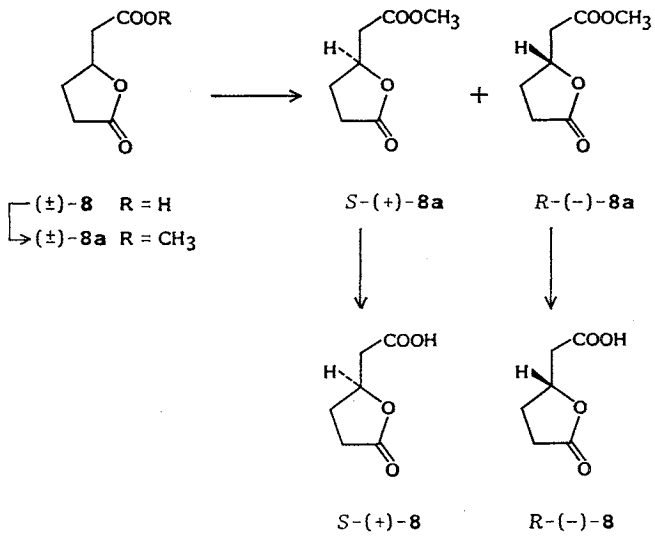
After the penems **26** and **27**, first obtained as a diastereomeric mixture (*ca.* 1:1; MIC values not reported in Table 3) starting from racemic lactonic acid **8**[18] and silver thiolate **2b**, had been found to possess excellent antibacterial properties, we prepared the single diastereomers starting from enantiomerically pure lactonic acid **8**, in order to study their biological activities. According to the literature, compound **8** has been resolved by classical procedures[19], or synthesized by Arndt-Eistert homologation of the butanolide **7** (easily obtained from *S*(+)-glutamic acid by diazotation[20]), or directly from *R*-(−)-β-amino adipinic acid[21].

However, the chemical yield and the optical purity of the above intermediates were poor, and we therefore looked for an alternative approach that would permit the desired compounds to be obtained in optically pure form. The simple 3-step procedure outlined in Scheme 4 proved to be successful: The key step in the preparation of *R*-(−)-**8** and *S*-(+)-**8**, after preparation of the ester (+)-**8a** in 74% yield from (+)-**8** with $(CH_3)_2SO_4$ - K_2CO_3 in acetone, was their chromatographic separation on cellulose triacetate†

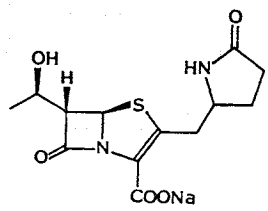
Scheme 3. Synthesis of lactonic acid **10**.



Scheme 4. Preparation of optically active *R*-(−)-**8** and *S*-(+)-**8**.



† For details see ref 22 and Experimental part.



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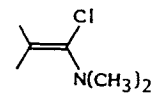

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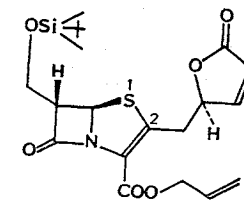

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into optically pure *R*-(−)-**8a** and *S*-(+)-**8a**.

The optical purity of the acids *R*-(−)-**8** and *S*-(+)-**8** (obtained from *R*-(−)-**8a** and *S*-(+)-**8a** by acid hydrolysis) was checked by spectroscopic methods: Addition of chiral shift reagents revealed no signs of enantiomeric impurities, but the racemic mixture of (+)-**8** showed split NMR signals. The enantiomeric excess was therefore estimated to be >98% (for $[\alpha]_D$ values see Table 1). Although **8** can easily be transformed into the corresponding acid chloride **3** by way of pure $SOCl_2$[18], in the case of *R*-(−)-**8** and *S*-(+)-**8** we preferred to use the enaminochloride **20**[23,24], because of the mildness and neutral conditions it affords the corresponding acid chloride. Otherwise the synthesis of **26** and **27** proceeded as outlined in Scheme 1.

Table 1. Data of optically active lactones.

Compound	$[\alpha]_D^{20}$ (°)
S-(+)-**7**	+13.6 (*c* 1.2, MeOH)
	(+10.6 (*c* 5.0, MeOH)[28])
R-(−)-**7**	−14.7 (*c* 1.0, MeOH)
	(−14.3 (*c* 1.19, MeOH)[29])
R-(−)-**8a**	−46.0 (*c* 0.56, EtOH)
	(−36.4 (*c* 0.5, EtOH)[19])
S-(+)-**8a**	+47.2 (*c* 0.54, EtOH)
	(+17.65 (*c* 1, MeOH)[20],
	+28.8 (*c* 1, EtOH)[21])
R-(−)-**8**	−42.5 (*c* 0.52, EtOH)
	(−33.6 (*c* 0.52, EtOH)[19])
S-(+)-**8**	+39.1 (*c* 0.66, EtOH)
	(+17.6 (*c* 0.66, MeOH)[20])

The NMR spectra of the single isomers **26** and **27** revealed quite different signals for the methylene protons at C-2. Whereas the AB quartet of **27** (*R*-configurated side-chain) showed doublets centered at 2.97 and 3.51 ppm, the respective values for **26** (*S*-configurated side-chain) were 3.25 and 3.92 ppm. This pattern was in full agreement with the NMR spectroscopic data of the analogue lactam penem **19** prepared in our laboratory (to be published) and was seen in all chromatographically separable compounds.

At this stage, the NMR spectroscopic data (see Table 2) supported by X-ray analysis (see below) of the crystalline penem ester **21**† indicated the absolute configurations of the side-chain listed in the Tables.

Crystal Structure and Conformation of Penem Ester **21**

Since the absolute configuration of the β-lactam part is given by chemical synthesis, the absolute configuration of **21** is the one shown in the corresponding formula. Bond lengths and angles agree with expected values. The nitrogen atom deviates from the plane of its three substituents by 0.42 Å. The angle between the thiazoline double bond and the ester group is 16°.

† The relative configuration of the side-chain in **21** corresponds to that found in **26**, and indeed both compounds show similar spectroscopic properties with regard to chemical shifts of the methylene protons at C-2. However, because of the changed priority in the CIP nomenclature of the substituents of the chiral center in **29**, the absolute configuration in the side-chain of **21** and therefore also in **29** is *R* and in **26** is *S*.

Table 2. Relevant NMR-data of penems 1[a] (D_2O, δ).

Compound	Configuration	Lactone	5-H_n	4-H_n	3-H_n	1'-H_2
22[b]	*S*	**7**	6.33 (quasi t)	2.81~2.60 (3H, m), 2.21 (1H, m)		
23[b]	*R*	**7**	6.28 (quasi t)	2.80~2.65 (3H, m), 2.30 (1H, m)		
24	*S*	**8**	4.92 (m)	2.43 (m), 2.07 (m)	2.71~2.60 (m)	3.32 (d×d), 3.23 (d×d)
25	*R*	**8**	4.90 (m)	2.45 (m), 2.06 (m)	2.67 (quasi t)	3.50 (d×d), 2.95 (d×d)
26	*S*	**8**	4.89 (m)	2.44 (m), 2.06 (m)	2.70~2.61 (m)	3.32 (d×d), 3.25 (d×d)
27	*R*	**8**	4.92 (m)	2.45 (m), 2.05 (m)	2.67 (quasi t)	3.51 (d×d), 2.97 (d×d)
28	*S*	**11**	5.52 (m)	7.77 (d×d)	6.25 (d×d)	3.68 (d×d), 3.06 (d×d)
29[c]	*R*	**11**	5.54 (m)	7.81 (d×d)	6.23 (d×d)	3.58 (d×d), 3.33 (d×d)
30	*S*	**12**	5.53 (m)		6.00 (quasi s)	3.71 (d×d), 3.16 (d×d)
31[c]	*R*	**12**	5.35 (m)		5.97 (quasi s)	3.62 (d×d), 3.47 (d×d)
33	*S*	**13**	5.98 (t)			4.05 (d×d), 3.26 (d×d)
34	*R*	**13**	6.00 (t)			4.09 (d×d), 3.48 (d×d)
35	*S*	**14**	5.90 (t)			4.00 (d×d), 3.24 (d×d)
36	*R*	**14**	5.92 (t)			4.05 (d×d), 3.42 (d×d)
38	*R,S*	**15**	4.55 (m), 4.19 (m)	2.97 (1H, m), 2.85~2.60 (3H, m), 2.40 (1H, m), 1.88~1.68 (2H, m)		
32	*R,S*	**16**	4.54~4.42 (m), 4.42~4.30 (m)	2.55~2.40 (m), 2.20~2.00 (m)	3.53 (0.5H, d×d), 2.88 (0.5H, d×d), 3.27~3.00 (2H, m)	
37	*R,S*	**9**	[d]		3.00~1.65 (5 complex multiplets)	
39	*R,S*	**10**	4.72 (m)		3.05~1.50 (6 complex multiplets)	

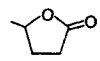
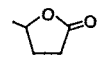
[a] For simplicity, atoms in the lactone side-chains are numbered in this table as shown in Scheme 2.

[b] Contaminated with *ca.* 5~10% of the respective other diastereomer.

[c] Traces of other diastereomer could be seen.

[d] Not visible under H_2O peak.

Table 3. *In vitro* antibacterial activity

n[b]:	0		1		1	
R_1:	CH_3		H		CH_3	
Z:						
Configuration:	*S*	*R*	*S*	*R*	*S*	*R*
Penem:	**22**[c]	**23**[c]	**24**	**25**	**26**	**27**
Staphylococcus aureus 10B	0.05	0.1	0.05	0.05	0.02	0.02
S. aureus 2999 ip	0.05	0.05	0.1	0.05	0.02	0.02
S. aureus A325 (MRSA)	2	2	ne	ne	2	2
Streptococcus pyogenes Aronson	0.05	0.02	0.1	0.2	0.02	0.05
Neisseria meningitidis 1316	0.05	0.05	0.1	0.05	0.02	0.02
Haemophilus influenzae NCTC 4560	2	2	4	4	1	2
Escherichia coli 205	1	0.5	0.2	0.5	0.1	0.5
E. coli 16 R TEM	1	0.5	1	2	0.1	0.5
Klebsiella pneumoniae 327	1	0.5	0.1	1	0.1	0.5
Serratia marcescens 344	8	4	4	4	1	2
Enterobacter cloacae P 99	2	1	2	8	0.5	2
Morganella morganii 1518	8	4	2	2	1	2
Pseudomonas aeruginosa ATCC 12055	>64	>64	>128	>128	>64	>64
Clostridium perfringens 194[d]	0.2	0.1	0.5	0.1	0.2	0.05
Bacteroides fragilis L01[d]	0.1	0.05	0.2	0.1	0.01	0.02

[a] Agar dilution method; DST agar Oxoid, inoculum 10^6 cfu/ml (18 hours incubation at 37°C, MRSA.
[b] See Fig. 1.
[c] For compounds **22** and **23** see ref 34.
[d] Anaerobic incubation.
ne: Not examined.

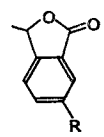
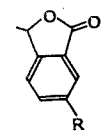
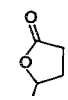
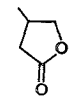
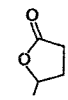
Biological Activity

Lactonyl penems were found to possess potent antibacterial activity against aerobic and anaerobic Gram-positive and Gram-negative bacteria, including methicillin-resistant Staphylococci. They were, however, inactive against *Pseudomonas aeruginosa*. With the exception of compounds bearing a lipophilic condensed aryl side-chain (*e.g.*, **33**, **34**, **35** and **36**), the whole series displayed the same good activity, especially against Gram-positive and, to a slightly lesser extent, also against Gram-negative bacteria, regardless of the length of the spacer $(CH_2)_n$ and the regiochemistry of the side-chain.

This consistency is in sharp contrast to the variable activity seen in other series of penems examined, *e.g.* the 2-heterocyclylmercaptoalkyl derivatives[10].

Surprisingly enough, comparisons of pairs of diastereomers (*e.g.*, **24** and **25**, **28** and **29**, **26** and **27**) revealed only minor differences in antibacterial activity. The diastereomeric penems **26** and **27** displayed the best activity, showing not only a well-balanced antibacterial spectrum, but also good activity *in vivo* (see Tables 3 and 4). Compounds **26** and **27** differed markedly in their stability against human dehydropeptidase-I (DHP-I), in contrast to other members of this penem family, between which no appreciable differences in DHP-stability were found (see Table 5).

of 2-lactonyl penems (MICs in μg/ml)[a].

1 H		1 CH_3		1 CH_3	1 H		1 H		2 CH_3	2 CH_3	3 CH_3
											
R=H		R=CH_3			R=H		R=OCH_3				
S **28**	*R* **29**	*S* **30**	*R* **31**	*R,S* **32**	*S* **33**	*R* **34**	*S* **35**	*R* **36**	*R,S* **37**	*R,S* **38**	*R,S* **39**
0.05	0.1	0.05	0.05	0.02	0.1	0.1	0.1	0.1	0.02	0.01	0.02
0.05	0.1	0.05	0.05	0.02	0.1	0.1	0.2	0.1	0.05	0.02	0.02
16	16	2	2	2	ne	ne	ne	ne	2	2	2
0.2	0.1	0.05	0.02	0.02	0.1	0.1	0.2	0.1	0.02	0.01	0.02
0.05	0.1	0.05	0.05	<0.01	0.2	0.5	0.5	1	<0.01	<0.01	<0.01
4	4	2	2	1	2	4	2	4	0.5	0.5	0.5
1	0.5	1	1	0.1	4	64	16	64	0.5	0.5	2
2	1	2	2	0.1	16	64	64	>128	1	1	4
1	0.2	2	1	0.1	8	64	64	128	0.5	1	4
16	4	8	4	2	64	>128	>128	>128	2	4	8
8	2	4	4	0.5	64	>128	>128	>128	2	4	16
4	2	4	2	2	16	32	16	64	1	1	2
>64	>64	>64	>64	>64	>128	>128	>128	>128	64	128	>64
0.1	0.5	0.05	0.05	0.1	0.1	0.5	0.1	0.2	0.02	0.05	0.05
0.2	0.2	0.05	0.05	0.02	0.5	1	0.5	2	0.02	0.02	0.05

48 hours at 30°C).

Table 4. *In vivo* antibacterial activity of selected penems.

Penems	ED_{50} in mice (mg/kg)[a]		
	Staphylococcus aureus 1098	*Streptococcus pyogenes* Aronson	*Escherichia coli* 2018
26	0.8	0.5	6.5
27	1.9	2.6	6.5
38	6.5	0.3	6.5
39	4.9	1.4	6.5
32	4.9	ne	>30.0

[a] Groups of 5 mice were treated sc twice, 30 minutes and 3 hours after ip infection.
ne: Not examined.

Table 5. Enzymatic[a] stability against the human renal DHP-I in relation to the stereochemistry of the lactone ring.

Penems	**22**	**23**	**24**	**25**	**26**	**27**	**28**	**29**	**35**	**36**	**39**
Absolute configuration[b]	*S*	*R*	*S*	*R*	*S*	*R*	*S*	*R*	*S*	*R*	*R,S*
t/2 DHP-I (hours)	2.27	1.85	2.2	2.6	4.19	8.77	2.93	2.47	2.16	3.42	2.7/3.5

[a] Homogenized human kidney in buffer solution (pH 7; 37°C).
[b] Absolute configuration of the chiral center in the side-chain.

Experimental

IR spectra were obtained using a Perkin-Elmer apparatus Model 983G. The UV spectra were recorded on a Cary 118 instrument. The 360 MHz ^{1}H NMR spectra were recorded on Brucker HX 360 apparatus (TMS 0.0). All mp's are uncorrected.

Lactonic Acid (±)-**9**

To a solution of 3 g (17.22 mmol) 4-oxopimelic acid in 20.7 ml 2 N NaOH was added 0.65 g $NaBH_4$ in 20 ml H_2O at 0°C. After the addition was complete, the solution was stirred for 2 hours at room temp. The reaction mixture was acidified with 4 N HCl, and the solution satd with NaCl and extracted continuously overnight with Et_2O. The solvent was evaporated and the residue chromatographed on silica gel with CH_2Cl_2 - MeOH (9 : 1) as eluent. Yield: 2.42 g (89%); colorless crystal (from Et_2O - hexane); mp 79~80°C (literature[25] 68~70°C); IR (CH_2Cl_2) cm^{-1} 1772, 1712; ^{1}H NMR (DMSO-d_6) δ 4.49 (1H, m, 5-H), 2.54~2.43 (2H, m, 3-H_2), 2.31 (2H, d×t, 2'-H_2), 2.28~2.19 (1H, m) and 1.90~1.74 (3H, m) (1'-H_2 and 4-H_2).

Anal Calcd for $C_7H_{10}O_4$: C 53.16, H 6.37.
Found: C 53.35, H 6.43.

Lactonic Ester (±)-**18**

To an ice-cold solution of 1 g (5.42 mmol) **17**[26] in 10 ml CH_2Cl_2 was added 0.5 g $NaHCO_3$ followed by 1.3 g *m*-CPBA (85%). After stirring overnight at room temp, the reaction mixture was filtered, diluted with CH_2Cl_2 and washed with satd $NaHCO_3$ and 1 N NaOH. The solvent was evaporated and the residue purified by filtration on silica gel in Et_2O - hexane (1 : 1). Further purification was effected by bulb-tube distillation (115°C, 2×10^{-3} mbar), yielding 0.92 g (85%) colorless liquid: IR (CH_2Cl_2) 1728 cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 4.35 (1H, m, 6-H), 4.15 (2H, q) and 1.26 (3H, t) (ester group); 2.65~2.36 (4H, m), 2.04~1.78 (4H, m) and 1.64~1.45 (2H, m) (remaining H's).

Anal Calcd for $C_{10}H_{16}O_4$: C 59.99, H 8.06.
Found: C 59.83, H 8.09.

Preparation of (±)-**10**

A solution of 8.5 g (42.45 mmol) **18** in 27 ml 2 N HCl and 30 ml THF was stirred overnight at 0°C; after stirring for a further hour at 50°C, the reaction mixture was diluted with H_2O (100 ml), saturated with NaCl and continuously extracted overnight with Et_2O. The solvent was evaporated, and the residue chromatographed on silica gel with CH_2Cl_2 - MeOH (98 : 2) as eluent. The yield was 5.4 g (74%) of **10**†: IR (CH_2Cl_2) cm^{-1} 1770, 1711; ^{1}H NMR (CD_3OD) δ 4.54 (1H, m, 5-H), 2.64~2.43 (2H, m), 2.39~2.26 (3H, m), 1.87 (1H, m) and 1.82~1.61 (4H, m) (remaining H's).

Lactone *R*-(−)-**7** and *S*-(+)-**7**

R-(−)-**7** and *S*-(+)-**7** were prepared starting from *R*- and *S*-glutamic acid, respectively, following the procedure described in the literature[27~29].

R-(−)-**7**: IR (CH_2Cl_2) cm^{-1} 1794, 1734; ^{1}H NMR (DMSO-d_6) δ 4.97 (1H, m, 5-H), 2.49 (2H, m) and 2.17 (2H, m) (3-H_2 and 4-H_2); $[\alpha]_D^{20}$ −14.7±1.0° (*c* 1.01, MeOH).

S-(+)-**7**: IR and ^{1}H NMR as for *R*-(−)-**7**; $[\alpha]_D^{20}$ +13.6±0.9° (*c* 1.12, MeOH).

Lactone (±)-**8**

For the preparation of (±)-**8** see refs 18 and 19.

Lactone Ester (±)-**8a**

A solution of 1 g (6.94 mmol) lactonic acid (±)-**8**, 1.3 g (0.99 ml, 10.4 mmol) dimethylsulfate and 1.15 g (8.32 mmol) potassium carbonate were refluxed for 4 hours. The cold solution was filtered through Celite and the solvent evaporated. Filtration on silica gel in Et_2O - hexane (3 : 1) followed by bulb-tube distillation (120°C/0.01 mbar) gave 1.06 g (97%) ester (±)-**8a**: IR (CH_2Cl_2) cm^{-1} 1740, 1780; ^{1}H NMR ($CDCl_3$) δ 4.93 (1H, m, 5-H), 3.74 (3H, s, $COOCH_3$), 2.84 and 2.65 (2H, ABX system,

† As can be seen on TLC, this hydrolysis is clearly a two-step process in which **18a** is an intermediate. As the reaction proceeds, **18a** disappears and only (±)-**10** is isolated (see Scheme 3).

1′-H_2), 2.60 (2H, m), 2.50 (1H, m) and 1.98 (1H, m) (3-H_2 and 4-H_2).

Chromatographic Resolution of (±)-**8a** into *R*-(−)-**8a** and *S*-(+)-**8a** on Cellulose Triacetate

The sorbent cellulose triacetate was prepared by heterogeneous acetylation of microcrystalline cellulose, according to literature procedure[22]. A particle size of 40~70 nm was adjusted by brief milling and sifting. Preparative resolution of the lactone ester (±)-**8a** was performed on a 100×5 cm glass column (Buechi, Uster, Switzerland). This column was slurry-packed with swollen microcrystalline triacetyl-cellulose in 95% ethanol at room temp. A pressure of 4 kg/cm² (flow rate 300 ml/hour) was applied using an Altex 110 A pump, and the eluate was monitored with a polarimeter (241 MC, Perkin-Elmer, Norwalk, CT, U.S.A.) equipped with a 300-μl flow cell (length 10 cm) at 365-nm. Thus 4.2 g of the racemate (±)-**8a** dissolved in 30 ml EtOH - H_2O (95 : 5) was chromatographically resolved on this column. After detection and combination of the fractions each containing the optically pure enantiomer, the solvent (eluent) was evaporated and the residual oil was purified by bulb-tube distillation (80~100°C/0.1 mmHg).

First eluted enantiomer *R*-(−)-**8a**: 1.85 g; $[\alpha]_D^{24}$ −46°; $[\alpha]_{578}^{24}$ −48°; $[\alpha]_{436}^{24}$ −91.7°; $[\alpha]_{365}^{24}$ −140° (*c* 0.564, EtOH).

Second eluted enantiomer *S*-(+)-**8a**: 1.56 g; $[\alpha]_D^{24}$ +47.2°; $[\alpha]_{578}^{24}$ +49.2°; $[\alpha]_{436}^{24}$ +91.5°; $[\alpha]_{365}^{24}$ +137.9° (*c* 0.544, EtOH); (ref 19 $[\alpha]_D^{25}$ −36.4° (*c* 0.5, EtOH), ref 21 $[\alpha]_D^{22}$ +28.8° (*c* 0.4, EtOH)).

Hydrolysis of *S*-(+)-**8a** and *R*-(−)-**8a** to *S*-(+)-**8** and *R*-(−)-**8**

1.56 g (9.87 mmol) *S*-(+)-**8a** suspended in 6.3 ml 2 N HCl was heated for 16 hours at 70°C. The clear, cold solution was evaporated, the residue dissolved in EtOAc and dried over Na_2SO_4. Purification was done by chromatography on silica gel in toluene - EtOAc - formic acid (6 : 2 : 1). Crystallization from EtOAc - hexane gave 850 mg *S*-(+)-**8** as colorless crystals, mp 49~50°C. An additional crop of 143 mg could be obtained from the mother liquor. Total yield 69%.

R-(−)-**8** was prepared in a similar way in 66% yield, mp 49~50°C.

S-(+)-**8**: $[\alpha]_D^{20}$ +39.1° (*c* 0.66, EtOH); IR (CH_2Cl_2) cm^{-1} 1776, 1716; ^1H NMR ($CDCl_3$) δ 4.92 (1H, m, 5-H), 2.88 and 2.72 (2H, ABX system) (1′-H_2), 2.61 (2H, m), 2.50 (1H, m) and 2.0 (1H, m) (3-H_2 and 4-H_2). Addition of chiral shift reagent gave no splitting of signals.

Anal Calcd for $C_6H_8O_4$: C 50.00, H 5.60.
Found: C 49.82, H 5.65.

R-(−)-**8**: $[\alpha]_D^{20}$ −42.5° (*c* 0.52, EtOH); IR (CH_2Cl_2) cm^{-1} 1778, 1717; ^1H NMR ($CDCl_3$) δ 4.90 (1H, m, 5-H), 2.86 and 2.72 (2H, ABX system) (1′-H_2), 2.60 (1H, m), 2.50 (2H, m) and 2.0 (1H, m) (3-H_2 and 4-H_2). Addition of chiral shift reagent gave no splitting of signals.

Anal Calcd for $C_6H_8O_4$: C 50.00, H 5.60.
Found: C 49.61, H 5.89.

Lactonic Acid (±)-**15**

(±)-**15** was synthesized by the method of CEDER and HANSSON[30], who prepared the corresponding optically active methyl ester. However, starting material for the preparation of (±)-**15** was 4-formyl-1-cyclohexene instead of 3-cyclohexene-1-carboxylic acid.

(±)-**15**: IR (CH_2Cl_2) cm^{-1} 1775, 1711; ^1H NMR ($CDCl_3$) δ 4.45 (1H, d×d) and 3.96 (1H, d×d) (5-H_2), 2.73~2.56 (2H, m, 3-H_2), 2.41 (2H, t, 2′-H_2), 2.29~2.15 (1H, m, 4-H), 1.93~1.76 (2H, m, 1′-H_2).

Lactonic Acid (±)-**11**, (±)-**12**, (±)-**13**, (±)-**14** and (±)-**16**

(±)-**11** and (±)-**12** were prepared by the method of PANDELL[31] and ELVIDGE *et al.*[32]. We thank Dr. W. HOYLE, Central Research Laboratories, Ciba-Geigy Manchester, for helpful information regarding the preparation of (±)-**11** and (±)-**12**. Starting material for (±)-**12** was 4-methylcatechol.

Lactonic acids (±)-**13** and (±)-**14** and (±)-**16** were gift from Dr. K. OERTLI and Dr. A. HUXLEY of our company.

Phosphoranes **4a**: General Preparation Method *e.g.* **2b**+*R*-(−)-**8**

To 958 mg (6.65 mmol) acid *R*-(−)-**8** in 20 ml abs CH_2Cl_2 were added at room temp 1.33 ml

(9.97 mmol) enaminochloride **20**. After 1 hour stirring, this acid chloride solution† was added to an ice-cooled, stirred mixture of silver thiolate **2b** (3.09 g, 4.43 mmol) and pyridine (0.55 ml, 6.87 mmol) in 50 ml CH_2Cl_2. After 1 hour stirring at room temp, the reaction mixture was filtered trough Celite, washed with satd $NaHCO_3$ and brine. Purification of the product was done by chromatography on silica gel in toluene - EtOAc (1 : 1) and hexane - EtOAc (1 : 1). Yield 1.18 g phosphorane **4b**: IR (CH_2Cl_2) cm^{-1} 1761, 1694, 1620.

Penem Ester **5**, General Method

Phosphoranes **4** were stirred in toluene at 100°C in the presence of catalytic amounts of BHT (2,6-di-*tert*-butyl-*p*-cresol). After the reaction was complete (TLC monitoring), the solvent was evaporated and the penem esters **5** purified by chromatography (solvent: Toluene - EtOAc or hexane - EtOAc).

Penem Ester **6**, General Desilylation Procedure

To a stirred solution of penem ester **5a** in THF a 5-fold excess of AcOH and 0.1 M tetrabutylammonium fluoride - THF solution (12 equivalents) were added at −70°C by syringe. The cooling bath was removed and the reaction mixture stirred at room temp until starting material could no longer be detected on TLC. The solvent was evaporated *in vacuo*, and the residue diluted with EtOAc, and washed with satd $NaHCO_3$ and brine; after drying (Na_2SO_4) the solution, the solvents was evaporated. Purification was done by column chromatography.

Penems **1a** and **1b**: General Deprotection Procedure Using $Pd(P(C_6H_6)_3)_4$ - Acetylacetone under Argon

To a solution of 1.257 mmol penem ester **5b** (n=1, lactone *R*-(−)-**8**) in 5.5 ml abs THF was added at room temp 3.267 mmol acetylacetone followed by approx 50 mg Pd complex. Stirring was continued for 1.5 hours, after which 3 ml satd $NaHCO_3$ solution was added. After 15 minutes stirring at room temp, the organic solvent was evaporated *in vacuo* and the residue filtered on XAD-2 (elution with H_2O). Yield: 80% lyophilisate.

Deprotection using $SnBu_3H$ instead of acetylacetone works equally well, but the yields are rather lower.

X-Ray Analysis of Compound **21**

Crystal Data: Orthorhombic, space group $P2_12_12_1$, a=6.034(2), b=10.406(3), c=38.117(8) Å, Z=4.

A Philips PW1100 automatic diffractometer was used for data collection with CuKα radiation and graphite monochromator. The intensities of 2488 independent reflections with $\theta<67°$ were measured, of which 2353 were classified as observed with $I<2\sigma$ (I).

The structure was solved by direct methods using the MULTAN78 program[33]. 18 of 20 H-atoms were found from a difference Fourier map, the coordinates of the rest 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 0.048.

Acknowledgments

The authors express their thanks to Mrs. I. MANSO, Mr. J.-C. MANI and Mr. G. PFISTER for their skillfull experimental work, to Mrs. J. GYSIN for the antibacterial tests and to our Physics Department for their spectroscopic support, to Prof. Dr. J. TORHORST, Institute of Pathology, University of Basel, for his kind collaboration.

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† Acid chlorides of lactonic acids **7**~**16** may be prepared alternatively by stirring the acid $SOCl_2$ at 50°C (*e.g.*, **9** and **11**~**16**) or even at reflux temperature (*e.g.*, **7** and **10**) for 1~2 hours.

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