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ENANTIOSELECTIVE TOTAL SYNTHESIS OF THE FUNGICIDE ß-LACTAM ANTIBIOTIC (-)-(2S,5S)-2-(2-HYDROXYETHYL)CLAVAM AND ITS (+)-(2S,5R)-EPIMER.

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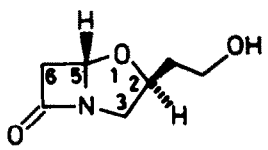
Abstract: Starting from (S)-1,2,4-butanetriol-1,2-acetonide 7 and 4-acetoxy-2-azetidinone 5, the sensitive title compounds 1 and 2 were synthesized. The synthesis confirms the unusual (5S)-configuration of the fungicide natural ß-lactam 1 which previously had been isolated from a Streptomyces strain.

Very recently, Zeeck, Zähner et al. reported on the isolation and structure elucidation of the new ß-lactam antibiotic (-)-2-(2-hydroxyethyl)clavam[2] (1), from cultures of a strain of Streptomyces antibioticus. The rather labile clavam exhibits no ß-lactamase inhibition and only low antibacterial but high antifungal activity. Strong evidence was supported from chiroptical data that the bicyclus 1 has the unusual (S)-configuration at the bridge-head carbon atom and carries a 5ß hydrogen atom unlike to all other ß-lactam antibiotics known in their absolute configuration at that time. In order to confirm the surprising finding unambigously by synthesis of 1 and to give also an access to the remaining three enantiomerically pure stereoisomers 2, ent-1, and ent-2, we undertook the following synthetic study.[3]

In the meantime, for another fungicide clavam, 2-(3-alanyl)clavam (Ro 22-5417)[4,24] (3), the (5S)-configuration was established by spectroscopic data[5] and synthesis[5,6] by the Roche research group. The first known member of this class, (-)-trans-2-(hydroxymethyl)clavam[7] (4) has not been elucidated in its absolute configuration; but by comparison of the optical rotation, its (5S) configuration 4 is to be assumed.



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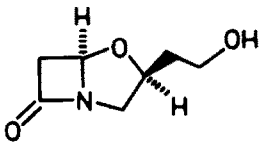

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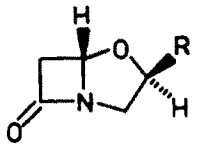


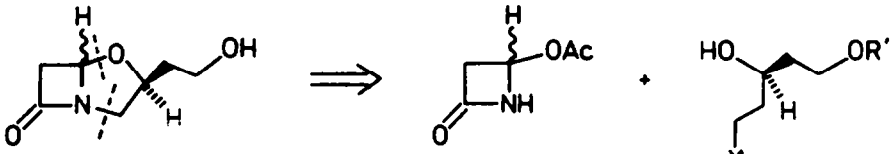
(S)

3 R = CH_2-$CH(NH_2)CO_2H$

4 R = CH_2OH

Synthetic scheme: Although the structure of the ß-lactam 1, from the synthetic point of view, is surprisingly simple, difficulties were expected from its great instability: 1 decomposes rapidly when kept in concentrated solution at room temperature, and also in diluted solutions, it does not survive even slightly basic or acidic reaction conditions.[8] Since 2'-O-protected derivatives of 1 are stable compounds,[2,3] obviously, the decomposition begins with an intramolecular attack of the hydroxy group at the azetidinone ring, presumably at C-5.

For the construction of the clavam skeleton, the Beecham annulation procedure[9] was envisaged, coupling racemic 4-acetoxy-2-azetidinone[10] **5** with a differentiated derivative **6** of (S)-1,2,4-butanetriol across the free 2-hydroxy group, followed by cyclo-alkylation to form a mixture of enantiomerically pure diastereomers **1** and **2**. It is noteworthy to mention, that the use of an enantiomerically pure equivalent for **5** does not offer any advantage because substitutions in the 4-position of 2-azetidinones proceed via planar intermediates, and hence, non-stereospecifically.[11]



1 β-H

2 α-H

5 **6**

The chiral starting material (S)-1,2,4-butanetriol 1,2-acetonide **7** is obtained from (S)-malic acid by the procedure of Corey[12] et al. and is separated from the accompanying 2,4-acetonide via their 3,5-dinitrobenzoates according to A. I. Meyers et al.;[13] or, more conveniently, is prepared via a two-step reduction sequence published recently by S. Saito et al.[14]

For the success of the synthesis, the proper selection of the 2'-O protecting group is crucial: In a preceeding study, a 1 : 1 mixture of the benzyl ethers of **1** and **2** was prepared, neither these nor **1** and **2** turned out to be separable by chromatographic methods.[15] Furthermore the hydrogenolytic deprotection (Pd/C) proceeded very sluggishly with low yield, because protic solvents or catalysts had to be avoided in order to suppress decomposition. Many of the common hydroxyl protecting groups are not compatible with the reaction conditions in several synthetic steps. So we developed the 4-methoxy-3-nitro-benzyl (MNB) group as a new benzyl type protecting group. The reasons are: the reduction of the nitro group to give an amino function offers further possibilities for the diastereomer separation, and, moreover, from a mechanistic study,[16] a more facile hydrogenolysis of +M-substituted benzyl residues was expected to occur.

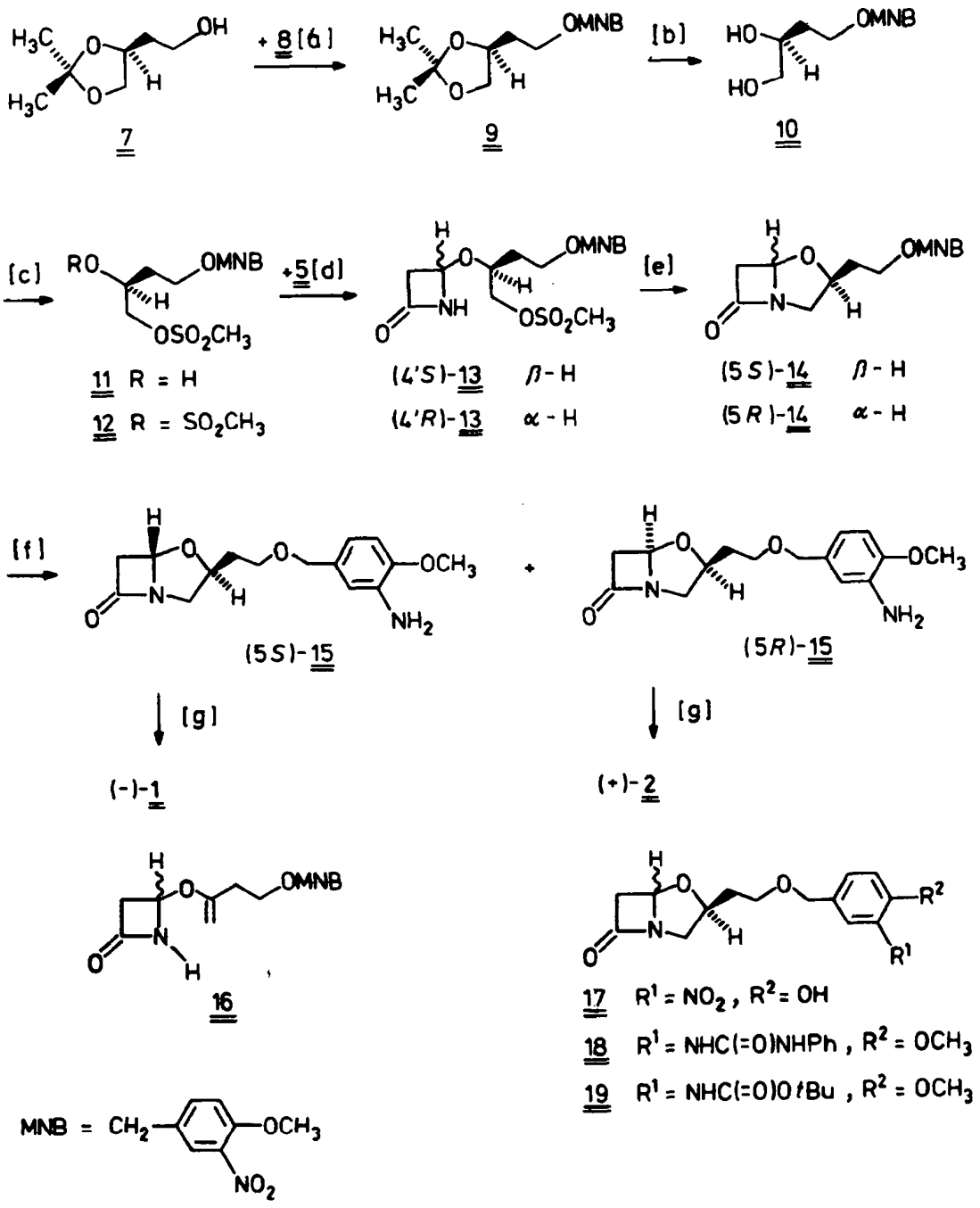
Execution: The (S)-1,2-acetonide **7** was alkylated at the 4-hydroxy group with 4-methoxy-3-nitrobenzyl chloride[17] **8** (Scheme 1) and the crude ether **9** was hydrolyzed to give the diol **10**. The primary mesylate **11** was formed without contamination by the secondary isomer, together with the easily separable dimesylate **12** (7%), on treatment of **10** with methanesulfonyl chloride in dichloromethane/pyridine. For coupling the hydroxyl group of **11** to 4-acetoxy-2-azetidinone (**5**), initially, the benzene solution of **5** and **8** was heated in the presence of zinc oxide[18] to afford the epimeric mixture (S)- and (R)-**13** (5 : 6) with only 17% yield. The yield was enhanced to 70% by application of the palladium catalyzed coupling reaction, recently published by Weigele.[5,6]

The cyclization was accomplished by a modification of the Beecham method,[9] heating **13** in presence of 3 mol-equiv. potassium carbonate and sodium iodide in HMPT. Careful silica gel chromatography yielded the clavams **14** as an epimeric mixture (5S)- and (5R)-**14** in a **4** : 6 ratio with 56% yield, contaminated by 6% of an inseparable unknown isomer. In addition, 1% of the 4'-hydroxy derivative **17** (epimers, 30 : 70, presumably formed by iodide-induced O-demethylation) and 2% of the enol ether **16** (formed by elimination of methanesulfonic acid) were found.

On hydrogenolysis of **14**, the amines **15** (5S : 5R = 30 : 70) were obtained with 85% yield (contaminated by 7% of an unknown isomer). On this stage, (S)- and (R)-**15** could be separated and purified by repeated HPLC on silica gel, using tert-butyl methyl ether/n-heptane/chloroform (3 : 2 : 1) and chloroform/acetonitrile (95 : 5) as eluants. All attemps to separate the more stable nitro compounds **14**, the phenylureas **18** (with phenyl isocyanate, 89%) or the tert-butyl urethanes **19** (25 equiv. di(tert-butyl) dicarbonate, 24 h at 20 °C, 83%) remained unsuccessfully.

Although feasible for simpler model compounds, the hydrogenolytic removal of the 3-amino-

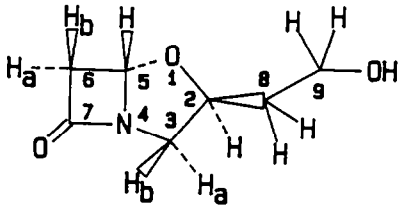
Scheme 1



[a] NaH; 4-methoxy-3-nitrobenzyl chloride, HMPT, 30 °C; 88%; [b] 0.15N aq. p-toluenesulfonic acid/THF, 20 °C; 68%; [c] 1.1 equiv. methanesulfonyl chloride, pyridine, CH_2Cl_2, 5 days 0-5 °C; 67% **11** and 7% **12**; [d] triethylamine, 7 mol-% palladium(II) acetate, benzene, 2 days 25 °C; 70%; or 2 mol-equiv. ZnO, benzene, 55 h at 55 °C; 17%; [e] K_2CO_3/NaI, HMPT, 5 h at 70 °C; 56%; [f] H_2, 10% Pd/C, THF, 20 °C, 1 bar; 85%; [g] HPLC separation; DDQ, CH_2Cl_2/water; 19% **1** or 20% **2**.

4-methoxybenzyl group of **15** or **19** with several Pd-, Pt-, or Ni-catalysts failed. Even in the presence of an equimolar amount (!) Pd/C, the decomposition of clavams **1** and **2** proceeded faster than their liberation. The same is true for the oxydative debenzylation by means of tris(4-bromophenyl)ammonium radical cation hexachloroantimonate, according to Steckhan.[19] Finally, we succeeded by applying of the method of Yonemitsu,[20] using 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) and buffered solution under carefully controlled conditions: From (+)-(S)-**15** the pure (-)-(2S,5S)-clavam **1** was obtained with 19% yield and proved identical with the natural product in all respects, including the optical rotation.[1,21] Similarly, (+)-(R)-**15** gave the (+)-(2S,5R)-epimer **2** with 20% yield, $[\alpha]_D^{23}$ = +125 (c = 0.2 in $CHCl_3$), which also is unstable (presumably attack of OH at C=O).

^{1}H NMR spectra of clavams **1** and **2**: As already recognized by Bentley and Hunt,[9] 2,5-trans-2-alkylclavams exhibit a larger difference in the ^{1}H NMR chemical shift between 3-H_b and 3-H_a than the 2,5-cis analogues (**1**: δ = 1.37; **2**: 0.24 ppm). Table 1 shows the ^{1}H NMR data of **1** and **2**, including some coupling constants, which (in part) were obtained by selective irradiation and spin-tickling experiments.[22] Although, some differences in remote coupling of 3-H_2 are seen, these values are of lower diagnostic value.



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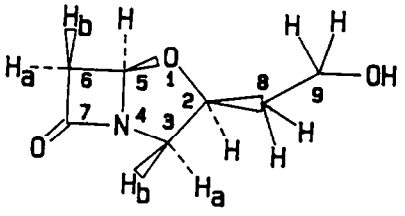

2

Table 1. ^{1}H NMR data ($CDCl_3$, 200 MHz) of **1** and **2** [a]

	1, δ (ppm)	J (Hz)	**2**, δ (ppm)	J (Hz)
2-H	4.28	$J_{2,3a}$ = 7.6, $J_{2,3b}$ = 6.0, $J_{2,8a}$ = 7.6, $J_{2,8b}$ = 5.1	4.49	$J_{2,3a}$ = $J_{2,3b}$ = $J_{2,8a}$ = $J_{2,8b}$ = 6.5-7.0 Hz
3-H_a	2.66	$J_{3,3}$ = 11.7, $J_{3,2}$ = 7.6	3.44	$J_{3,3}$ = 10.6, $J_{3,2}$ = 7.1
3-H_b	4.03	$J_{3,3}$ = 11.7, $J_{3,2}$ = 6.0, $J_{3,5}$ = 0.5	3.20	$J_{3,3}$ = 10.6, $J_{3,2}$ = 6.8, $J_{3,6a}$ = 0.9
5-H	5.35	$J_{5,6b}$ = 2.9, $J_{5,6a}$ = 0.9, $J_{5,3b}$ = 0.5	5.18	$J_{5,6a}$ = 2.7, $J_{5,6b}$ = 0.7
6-H_a	2.87	$J_{6,6}$ = 16.4, $J_{6,5}$ = 0.9	3.27	$J_{6,6}$ = 16.1, $J_{6,5}$ = 2.7, $J_{6,3b}$ = 0.9
6-H_b	3.32	$J_{6,6}$ = 16.4, $J_{6,5}$ = 2.9, $J_{6,3}$ = 0.8	2.88	$J_{6,6}$ = 16.1, $J_{6,5}$ = 0.7
8-H_2	1.86	m	1.90	m
9-H_2	3.83	m	3.81	$J_{9,8}$ = 5.8
OH	1.97	m	1.81	m

[a] Clavam numbering is shown here. For conversion to IUPAC numbering (as used in experimental part), positions 2 and 3 are reversed, 8 and 9 are exchanged for 1' and 2'.

EXPERIMENTAL

LC separations (>1 g) were carried out with "MN-Kieselgel 60", 0.05-0.2 mm (Macherey-Nagel GmbH & Co KG, Düren), or (<1 g) at 1-3 bar on "Silica Woelm 32-63", 0.032-0.063 mm, (Woelm Pharma GmbH & Co, Eschwege). In TLC analyses, spots of β-lactams **1**, **2**, **13**-**19** on treatment with Ehrlich's reagent[23] and heating to 100 °C appear coloured orange to red. HPLC separations were performed on Solvent Delivery System Model 6000 A (Waters Associates Inc., Milford, USA). - Ethyl acetate, pyridine and triethylamine were distilled over CaH_2 prior use; CH_2Cl_2 and $CHCl_3$ were distilled over P_4O_{10}.

(S)-3,4-Isopropylidenedioxy-butyl (4-methoxy-3-nitrobenzyl) ether (**9**): (S)-1,2-Di-O-isopropylidene-1,2,4-butanetriol[13] (**7**) (4.40 g, 30.0 mmol), $[\alpha]_D^{23}$ = -1.83 (c = 10.4, CH_3OH), obtained by lithium alanate reduction of methyl (S)-3,4-di-O-isopropylidene 3,4-dihydroxybutanoate,[14] was dropped to a slurry of sodium hydride (0.79 g, 33 mmol) in dry HMPTA (20 mL) below 30 °C, and stirred for 30 min. 4-Methoxy-3-nitrobenzyl chloride[17] (**8**) (6.05 g, 30.0 mmol) in dry THF (20 mL) were added below 15 °C and stirring was continued for 1.5 h at 30 °C. For work-up, aq. K_2CO_3 soln. 5%, 5mL) was carefully added, the reaction mixture poured into ice water (60 mL) and extracted with diethyl ether (3 x 50 mL). The ether solution was dried (KCl-soln.; solid K_2CO_3) and evaporated. The crude product **9**, 9.20 g (88%), was used for the next step. An analytical sample was purified by LC (silica gel, ether); oil,

$[\alpha]_D^{22}$ = -8.36 (c = 4.5, $CHCl_3$) - IR (KBr): 1535 and 1365 cm^{-1} (NO_2). - ^1H NMR ($CDCl_3$): δ = 1.35 and 1.40 (s, CH_3); 1.89 ("q", J = 6 Hz, 2-H_2), 3.4-4.4 (m, 5 H, 4-H_2, 3-H, 1-H_2); 3.94 (s, OCH_3); 4.46 (s, OCH_2Ar); 7.04 (d), 7.47 (dd), and 7.77 ppm (d) (3 H, aryl).

$C_{15}H_{21}NO_6$ (311.34). Calc. C 57.87 H 6.80. Found C 58.08 H 6.87.

(S)-4-(4-Methoxy-3-nitrobenzyloxy)-1,2-butandiol (**10**): Crude acetonide **9** (8.70 g) and p-toluenesulfonic acid monohydrate (0.6 g) in THF/water (40 + 20 mL) were kept at room temp. for 2.5 days. Potassium carbonate (0.6 g) was added, the THF was rotated off in vacuum and the aq. mixture was extracted with dichloromethane (3 x 40 mL). LC (silica gel, ethyl acetate) afforded 5.09 g **10** (63%, referred to **7**) as a yellow oil, R_F = 0.20.

$[\alpha]_D^{22}$ = +1.37 (c = 2.7, $CHCl_3$), -14.5 (c = 2.6, acetone). - IR (KBr): 3400 (OH), 1530 and 1355 cm^{-1}(NO_2). - ^1H NMR ($CDCl_3$): δ = 1.76 (q, J = 5.8 Hz; 3-H_2); 3.32-3.93 (m, 7 H; OH, 1-H_2, 2-H, 4-H_2); 3.93 (s, OCH_3); 4.44 (s, OCH_2Ar); 7.04, 7.47 and 7.76 ppm (3 H, aryl).

$C_{12}H_{17}NO_6$ (271.27). Calc. C 53.13 H 6.32. Found C 53.22 H 6.26.

(S)-[4-(4-Methoxy-3-nitrobenzyloxy)-2-hydroxybutyl] methanesulfonate (**11**) and (S)-[4-(4-methoxy-3-nitrobenzyloxy)butan-1,2-diyl] bis(methanesulfonate) (**12**): **10** (4.02 g, 17.8 mmol), dry pyridine (1.86 mL, 23.1 mmol), and methanesulfonyl chloride (1.52 mL, 19.5 mmol) in dry dichloromethane (35 mL), were mixed below -10 °C under N_2 and were allowed to stand at 0-5 °C for 5 days. For work-up, dichloromethane (50 mL) and ice-water (50 mL) were added, the organic phase was washed with concd. aq. $CuSO_4$ soln. (2 x 15 mL), water (20 mL) phosphate buffer (pH 7; 10 mL) and brine (10 mL). After drying over Na_2SO_4, evaporation of the solvent in vacuum below 20 °C, the residue (6.88 g) was purified by LC (silica gel, 400 g; ethyl acetate/cyclohexane, 2 : 1), affording the monomesylate **11** (4.24 g, 67%), R_F = 0.15, oil, and the dimesylate **12** (0.61 g, 7%), R_F = 0.24, oil.

$[\alpha]_D^{22}$ = -11.1 (c = 2.8, $CHCl_3$).

11, IR (KBr): 3530 (OH), 1535 and 1350 (NO_2), 1350 and 1178 cm^{-1} (SO_2CH_3). - ^1H NMR ($CDCl_3$): δ = 1.68-2.16 (m, 3-H_2); 3.09 (s, SO_2CH_3); 3.40-3.72 (m, 5 H; 1-H_2, 2-H, 4-H_2); 3.88 (s, OH); 3.98 (s, OCH_3); 4.51 (s, OCH_2Ar); 7.09 (d), 7.53 (dd), and 7.83 (d) (3 H, aryl).

$C_{13}H_{19}O_8NS$ (349.36) Calc. C 44.69 H 5.48 Found C 44.53 H 5.46

12, IR (KBr): 1533 and 1358 (NO_2), 1358 and 1177 cm^{-1} (OSO_2CH_3). - ^1H NMR ($CDCl_3$): δ = 1.88-2.23 (m, 3-H_2); 3.10 and 3.12 (each s, SO_2CH_3); 3.65 (t, J = 5.8 Hz, 4-H_2); 3.99 (s, OCH_3), 4.18-4.62 (m, 2 H, 1-H_2); 4.50 (s, CH_2Ar); 7.08 (d), 7.54 (dd), and 7.83 (d) (3 H, aryl).

$C_{14}H_{21}NO_{10}S_2$ (427.45). Calc. C 39.34 H 4.95. Found C 39.22 H 5.13.

(2S)-4-(4-Methoxy-3-nitrobenzyloxy)-2-[(4RS)-2-oxoazetidin-4-yloxy]butyl methanesulfonate (**13**); - Pd-mediated coupling[5,6]: To the mesylate **11** (4.16 g, 11.9 mmol) and palladium(II)acetate (0.17 g, 0.75 mmol) in anh. benzene (50 mL) under argon, triethylamine (1.20 g, 11.9 mmol) and 4-acetoxy-2-azetidinone[10] **5** (1.54 g, 11.9 mmol) in benzene (15 mL) were added and the reaction mixture stirred at 20-25 °C for 50 h. After 6 h and 24 h, additional **5** (each 0.76 g, 6.0 mmol) and triethylamine (each 0.60 g, 6.0 mmol) in benzene (7 mL) were added. For work-up, the soln. was poured from the tarry residue. The latter was diluted with ethyl acetate (50 mL). A crystalline precipitation was filtered off and washed with ethyl acetate (30 mL). The combined solns. were washed with water (3 x 50 mL) and brine (30 mL), dried over $MgSO_4$, and evaporated in vacuum. The residue was purified by LC (silica gel, 400 g, 0.05-0.2 mm; ethyl acetate) in a cooled column (0-5 °C) yielding 3.49 g (70%) **13**; R_F = 0.18 (red colour on treatment with Ehrlich's reagent[23]), yellow oil, (4'S)-**13** : (4'R)-**13** = 43 : 57 (^1H NMR).

ZnO-mediated coupling: **11** (3.5 mmol), **5** (7.0 mmol), and zinc oxide (0.57 g, 7.0 mmol) in benzene (7 mL) were stirred at 55 °C for 55 h. Work-up with dichloromethane (50 mL), water (20 mL) and acetic acid (0.56 mL), followed by LC yielded 0.236 g (16%) **13** (1 : 1) beside **11** (31%) and 4-(4-methoxy-3-nitrobenzyloxy)-2-azetidinone (9%), R_F = 0.23. -

13, IR (KBr): 3365 (NH), 1770 (C=O), 1533 and 1355 (NO_2), 1350 and 1175 cm^{-1} (OSO_2). - ^1H NMR ($CDCl_3$, 200 MHz): δ = 1.66-1.98 (m, 3-H_2) 2.78-3.26 (m, 3'-H_2); 3.07 and 3.08 (each s, SO_2CH_3); 3.46-4.42 (m, 5 H, 1-H_2, 2-H, and 4-H_2); 4.00 (s, OCH_3); 4.50 (s, CH_2Ar); 5.16 [dd, $J_{4,3c}$ = 4.0 Hz, $J_{4,3t}$ = 1.5 Hz, 4-H of (4'R)-**13**]; 5.29 [dd, $J_{4,3c}$ = 4.0 Hz, $J_{4,3t}$ = 1.8 Hz, 4-H of (4'S)-**13**]; 6.41 and 6.56 (each m, NH) 7.12 and 7.13 (each d); 7.53 and 7.55 (each dd); 7.86 and 7.87 (each d) (3 H, aryl).

$C_{16}H_{22}N_2O_9S$ (418.43). Calc. C 45.93 H 5.30. Found C 45.94 H 5.32

Base-induced cyclization; (3S,5RS)-3-[2-(4-Methoxy-3-nitrobenzyloxy)ethyl]-4-oxa-1-azabicyclo[3.2.0]heptan-7-one [(5R)-**14** and (5S)-**14**]: (2S,4'RS)-**13** (3.48 g, 8.33 mmol, diastereomeric ratio 9 : 7), anh. powdered NaI (3.75 g, 25 mmol), and K_2CO_3 (3.46, 25 mmol) in anh. hexamethyl phosphoric triamide (65 mL) were stirred for 5 h at 70 °C under N_2. For work-up, to the cold reaction mixture tert-butyl methyl ether (500 mL), phosphate buffer solution (pH 7, 100 mL) and ice (100 g) were added. The aqueous soln. was extracted 3 x with tert-butyl methyl ether (100 mL each), the combined ether solns. were washed with water (3 x 100 mL), brine (100 mL), dried over $MgSO_4$ and evapourated in vacuum. LC (silica gel, 200 g; ethyl acetate/cyclohexane, 2 : 1) afforded 1.51 g (56%) **14** [R_F = 0.37; ratio (5S) : (5R) = 40 : 60; ^1H NMR] which are accompanied by 6% of an unseparable unknown isomeric β-lactam X-NO_2.
From the more polar fractions, 25 mg (1%) of the 4'-demethyl derivatives **17** (5S : 5R = 30 : 70), R_F = 0.27, and 50 mg (2%), R_F = 0.19, of the enol ether **16** were isolated. (5RS)-**14**, IR (KBr): 1783 (C=O), 1530 and 1355 cm^{-1} (NO_2). - ^1H NMR ($CDCl_3$, 200 MHz, from the mixture): (5S)-**14**: δ = 1.82-2.04 (m, 1'-H_2); 2.64 (ddd, $J_{2,2}$ = 11.6 Hz, $J_{2,3}$ = 7.3 Hz, $J_{2,6\beta}$ = 0.8 Hz, 2-H_α); 2.85 (d, J = 16.4 Hz, 6-H_α); 3.29 (ddd, $J_{6,6}$ = 16.4 Hz, $J_{6,5}$ = 2.9 Hz, $J_{6,2\alpha}$ = 0.8 Hz, 6-H_β); 3.61 (t, J = 5.9 Hz, 2'-H_2); 3.97 (s, OCH_3); 3.99 (dd, $J_{3,3}$ = 11.6 Hz, $J_{3,2}$ = 6.0 Hz, 2-H_β); 4.30-4.56 (m, 3-H); 4.48 (s, aryl-CH_2); 5.31 (d, J = 2.9 Hz, 5-H); 7.08 (d, J = 8.6 Hz, 5"-H); 7.51 (dd, $J_{6",5"}$ = 8.6 Hz, $J_{6",2"}$ = 2.1 Hz, 6"-H); 7.84 ppm (d, J = 2.1 Hz, 2"-H). (5R)-**14**: δ = 1.82-2.04 (m, 1'-H_2); 2.84 (d, J = 15.7 Hz; 6-H_β); 3.17 (ddd, $J_{2,2}$ = 10.5 Hz, $J_{2,3}$ = 6.7 Hz, $J_{2,6\alpha}$ = 0.6 Hz; 2-H_β); 3.26 (ddd, $J_{6,6}$ = 15.7 Hz, $J_{6,5}$ = 2.5 Hz, $J_{6,2\beta}$ = 0.6 Hz, 6-H_α), 3,43 (dd, $J_{2,2}$ = 10.5 Hz, $J_{2,3}$ = 7.1 Hz, 2-H_α); 3.61 (t, J = 5.9 Hz, 2'-H_2); 3.97 (s, OCH_3); 4.30-4.56 (m, 3-H); 4.48 (s, aryl-CH_2); 5.15 (d, J = 2.5 Hz, 5-H); and aromatic absorptions (see above).

$C_{15}H_{18}N_2O_6$ (322.32). Calc. C 55.90 H 5.63. Found C 55.57 H 5.66.

(3S,5RS)-3-[2-(4-Hydroxy-3-nitrobenzyloxy)ethyl]-4-oxa-1-azabicyclo[3.2.0]heptan-7-one [(5RS)-**17**]: IR (neat): 3285 (OH), 1773 (CO), 1528 and 1320 cm^{-1} (NO_2). - ^1H NMR ($CDCl_3$, 200 MHz, from the diastereomeric mixture), (5S)-**17**: δ = 1.75-2.03 (m, 1'-H_2); 2.58 (dd, $J_{2,2}$ = 12.7 Hz, $J_{2,3}$ = 7.4 Hz, 2-H_α); 2.77 (d, J = 16.1 Hz, 6-H_α); 3.23 (dd, $J_{6,6}$ = 16.1 Hz, $J_{6,5}$ = 2.5 Hz, 6-H_β); 3.54 (t, J = 5.9 Hz, 2'-H_2); 3.95 (dd, $J_{2,2}$ = 12.7 Hz, $J_{2,3}$ = 7.6 Hz, 2-H_β); 4.22-4.53 (m, 3-H); 4.40 (s, aryl-CH_2); 5.26 (d, J = 2.5 Hz, 5-H); 7.09 (d, J = 8.6 Hz, 5"-H); 7.49 (dd, $J_{6,5}$ = 8.6 Hz, $J_{6,2}$ = 2.1 Hz, 6"-H); 8.01 (d, J = 2.1 Hz, 2"-H); 10.46 ppm (b.s, OH). - (5R)-**17**: δ = 1.75-2.03 (m, 1'-H_2);2.77 (d, J = 16.1 Hz; 6-H_β); 3.15 (dd, $J_{2,2}$ = 10.5 Hz, $J_{2,3}$ = 6.4 Hz, 2-H_β); 3.20 (dd, $J_{6,6}$ = 16.1 Hz, $J_{6,5}$ = 2.5 Hz, 6-H_α); 3.36 (dd, $J_{2,2}$ = 10.5 Hz, $J_{2,3}$ = 7.6 Hz, 2-H_α); 3.54 (t, J = 5.9 Hz, 2'-H_2); 4.22-4.53 (m, 3-H); 4.40 (s, aryl-CH_2); 5.08 (d, J = 2.5 Hz; 5-H); and aromatic protons, see above.

$C_{14}H_{16}N_2O_6$ (308.29) Calc. C 54.54 H 5.23. Found H 54.43 H 5.31.

4-[4-(4-Methoxy-3-nitrobenzyloxy)-1-buten-2-yloxy]-2-azetidinone (**16**): IR (neat): 3270 (NH), 1780 (C=O), 1528 and 1355 cm^{-1} (NO_2). - ^1H NMR ($CDCl_3$, 200 MHz): δ = 2.42 (t, J = 6.5 Hz, 3'-H_2); 2.97 (ddd, after D_2O-exchange dd, $J_{3,3}$ = 15.1 Hz, $J_{3,1}$ = 1.3 Hz, $J_{3,4}$ = 1.3 Hz, 3-H_a); 3.28 (ddd, after D_2O-exchange dd, $J_{3,3}$ = 15.1 Hz, $J_{3,4}$ = 3.8 Hz, $J_{3,1}$ = 2.3 Hz, 3-H_b); 3.61 (t, J = 6.5 Hz, 4'-H_2); 3,91 (d, J = 2.8 Hz, 1'-H_{trans}); 4.00 (s, OCH_3); 4,17 (d, J = 2.8 Hz, 1'-H_{cis}); 4.48 (s, aryl-CH_2); 5.47 (dd, $J_{4,3b}$ = 3.8 Hz, $J_{4,3a}$ = 1.3 Hz, 4-H), 6.65-6.84 (m, NH); aryl part: 7.07 (d, J = 8.6 Hz, 5-H); 7.55 (dd, $J_{6,5}$ = 8.6 Hz, $J_{6,2}$ = 2.2 Hz, 6-H); 7.87 ppm (d, J = 2.2 Hz, 2-H).

$C_{15}H_{18}N_2O_6$ (322.22). Calc. C 55.90 H 5.63. Found C 55.99 H 5.79.

(3S,5S)- and (3S,5R)-3-[2-(3-Amino-4-methoxybenzyl)ethyl]-4-oxa-1-azabicyclo[3.2.0]heptan-4-one [(5S)- and (5R)-15]: (5RS)-14 (0.465 g, 1.44 mmol, 40 : 60), accompanied by 6% of X-NO_2, in THF (12 mL) was hydrogenolyzed (1 bar, 10 h) in presence of 10% Pd/C (120 mg). Separation of the catalyst by filtration, evaporation of the solvent, followed by LC (silica gel, 40 g; dichloromethane/tert-butyl methyl ether, 3 : 1) afforded 0.358 g (85%) (5RS)-15 (30 : 70; ^{1}H NMR), accompanied by 7% of X-NH_2 (2 diastereomers).
The isomers (217 mg) were separated by HPLC (Nucleosil, 5 µm, column 250 x 8 mm), 3 mg per run. By using twice tert-butyl methyl ether/n-heptane/dichloromethane (3 : 2 : 1, 2.9 mL/min) as eluant, 36 mg pure (5S)-15 (t_R = 27.2 min), $[\alpha]_D^{20}$ = -105.4 (c = 1.7, $CHCl_3$), and 100 mg of (5R)-15 and X-NH_2 (95 : 5), t_R = 30.6 min, were collected. The second fraction was separated twice with trichloromethane/acetonitrile (95 : 5, 1.5 mL/min) affording 54 mg of pure (5R)-15 (t_R = 25.9 min; X-NH_2 = 26.9 min), $[\alpha]_D^{23}$ = + 99 (c = 1.7, $CHCl_3$). IR (KBr, mixture): 3465 and 3370 (NH_2), 1780 cm^{-1} (C=O). - ^{1}H NMR ($CDCl_3$, 200 MHz); (5S)-15: δ = 1.70-2.03 (m; 1'-H_2); 2.63 (ddd, $J_{2,2}$ = 11.8 Hz, $J_{2,3}$ = 7.6 Hz, $J_{2,6\beta}$ = 0.8 Hz, 2-H_α); 2.84 (dd, $J_{6,6}$ = 16.4 Hz, $J_{6,5}$ = 1.0 Hz, 6-H_α); 3.28 (ddd, $J_{6,6}$ = 16.4 Hz, $J_{6,5}$ = 3.0 Hz, $J_{6,2\alpha}$ = 0.8 Hz, 6-H_β); 3.52 (dd, $J_{2',1'}$ = 6.9 Hz and 5.5 Hz; 2'-H_2); 3.80 (b.s, NH_2); 3.85 (s, OCH_3); 3.97 (ddd, $J_{2,2}$ = 11.8 Hz, $J_{2,3}$ = 6.0 Hz, $J_{2,5}$ = 0.3 Hz, 2-H_β); 4.28-4.44 (m, 3-H); 4.37 (d, J = 1.5 Hz, aryl-CH_2); 5.29 (ddd, $J_{5,6\beta}$ = 3.0 Hz, $J_{5,2\beta}$ = 0.3 Hz, 5-H); 6.63-6.80 ppm (m, 3 aryl-H). - (5R)-15: δ = 1.77-2.06 (m, 1'-H_2); 2.83 (dd, $J_{6,6}$ = 15.9 Hz, $J_{6,5}$ = 0.6 Hz, 6-H_β); 3.14 (ddd, $J_{2,2}$ = 10.5 Hz, $J_{2,3}$ = 6.7 Hz, $J_{2,6\alpha}$ = 0.6 Hz, 2-H_β); 3.23 (ddd, $J_{6,6}$ = 15.9 Hz, $J_{6,5}$ = 2.5 Hz, $J_{6,2\beta}$ = 0.6 Hz, 6-H_α); 3.42 (dd, $J_{2,2}$ = 10.5 Hz, $J_{2,3}$ = 7.2 Hz, 2-H_α); 3.53 (t, J = 6.1 Hz, 2'-H_2); 3.81 (b.s, NH_2); 3.84 (s, OCH_3); 4.36 (s, aryl-CH_2); 4.43 (m, 3-H); 5.12 (dd, $J_{5,6}$ = 2.5 Hz, $J_{5,6\beta}$ = 0.6 Hz, 5-H); 6.63-6.82 ppm (m, 3 aryl-H). - X-NH_2: δ = 5.19 and 5.21 (each d, J = 2.5 Hz).

(-)-(3S,5S)-3-(2-Hydroxyethyl)-4-oxa-1-azabicyclo[3.2.0]heptan-7-one [(-)-1]: (5S)-15 (42 mg, 0.14 mmol) in dichloromethane (5 mL) were added to a suspension of 2,3-dichloro-5,6-dicyano-1.4-benzoquinone (DDQ 37 mg, 0.16 mmol) in dichloromethane/water (0.4 + 0.2 mL) and stirred under Ar at 20 °C for 22 min. For immediate work-up, phosphate buffer (0.5 M, pH 7, 1.5 mL) was added, and the mixture extracted with dichloromethane (5 x 10 mL). The combined solns. were dried over $MgSO_4$, the solvent evaporated and the residue purified by LC (silica gel, 20 g; ethyl acetate) affording 4.2 mg (19%) (-)-1, R_F = 0.27, $[\alpha]_D^{23}$ = -152 (c = 0.2, $CHCl_3$) and 4.7 mg (22%) 3-amino-4-methoxy-benzaldehyde, R_F = 0.52. Under identical conditions, a sample of freshly purified natural (-)-1 exhibited $[\alpha]_D^{23}$ = -154. IR- and ^{1}H NMR-spectra of synthetic (-)-1 proved identical with these of the natural product.

(+)-(3S,5R)-3-(2-Hydroxyethyl)-4-oxa-1-azabicyclo[3.2.0]heptan-7-one [(+)-2]: (5R)-15 (54 mg, 0.18 mmol) and DDQ (47 mg, 0.21 mmol), reaction time 27 min., afforded, as described above, after two LC purifications (silica gel, 8 g; ethyl acetate, R_F = 0.27; tert-butyl methyl ether/dichloromethane, 2 : 1, R_F = 0.12) 5.9 mg (20%) (+)-2, oil; $[\alpha]_D^{23}$ = +125 (c = 0.2, $CHCl_3$). - IR (KBr): 3435 (OH), 1775 cm^{-1} (C=O). - ^{1}H NMR ($CDCl_3$, 200 MHz): δ = 1.54-2.08 (m, OH); 1.84-1.98 (m, 1'-H_2); 2.88 (dd, $J_{6,6}$ = 16.1 Hz, $J_{6,5}$ = 0.7 Hz, 6-H_β); 3.20 (ddd, $J_{2,2}$ = 10.6 Hz, $J_{2,3}$ = 6.8 Hz, $J_{2,6\alpha}$ = 0.9 Hz, 2-H_β); 3.27 (ddd, $J_{6,6}$ = 16.1 Hz, $J_{6,5}$ = 2.7 Hz, $J_{6,2\beta}$ = 0.9 Hz, 6-H_α); 3.44 (dd, $J_{2,2}$ = 10.6 Hz, $J_{2,3}$ = 7.1 Hz, 2-H_α); 3.81 (t, J = 5.8 Hz, 2'-H_2); 4.49 (m, J = 6.7 Hz, 3-H); 5.18 (dd, $J_{5,6\beta}$ = 2.7 Hz, $J_{5,6\alpha}$ = 0.7 Hz, 5-H). ^{13}C NMR ($CDCl_3$): δ = 36.8 (C-1'), 44.1 (C-6), 51.6 (C-3), 60.2 (C-2'), 82.5 (C-2), 84.5 (C-5), 177.8 ppm (C-4).
MS (70 eV, m/e): $C_7H_{11}NO_3$, Calc. 157.0739, Found 157.0739.

(3S,5RS)-3-[2-(3-Phenylureido-4-methoxybenzyloxy)ethyl]-4-oxa-1-azabicyclo[3.2.0]heptan-7-one [(5RS)-18]: (5RS)-15 (22 mg, 0.08 mmol, 6 : 5) in dry dichloromethane (1.5 mL) and phenyl isocyanate (0.012 mL, 0.16 mmol) were stirred at 25 °C for 2 h. Evaporation of the solvent and LC (silica gel, 8 g; tert-butyl methyl ether/dichloromethane, 1 : 3) afforded 28 mg (89%) (5RS)-18 (6 : 5), R_F = 0.57, inseparable yellow oil. - IR (KBr): 3350 (NH), 1783 (lactam-C=O), 1667 cm^{-1} (urea-C=O). - ^{1}H NMR (from the mixture, $CDCl_3$, 100 MHz), (5S)-18: δ = 1.70-2.04 (m, 1'-H_2); 2.62 (dd, $J_{2,2}$ = 12.0 Hz, $J_{2,3}$ = 8.0 Hz, 2-H_α); 3.55 ("t", J = 6.3 Hz, 2'-H_2); 3.80 (s, OCH_3); 3.94 (dd, $J_{2,2}$ = 12.0 Hz, $J_{2,3}$ = 6.0 Hz, 2-H_β); 4.25-4.60 (m, 3-H); 4.40 (s, Ar-CH_2); 5.30 (d, $J_{5,6}$ = 0.5 Hz, 5-H); and others. - (5R)-18: 2.7-3.7 (m, 2-H_2); 5.13 (d, $J_{5,6}$ = 2.5 Hz, 5-H); and others.
$C_{22}H_{25}N_3O_5$ (411.46). Calc. C 64.22 H 6.12. Found C 64.09 H 6.22.

(3S,5RS)-3-[2-(3-N-tert-Butyloxycarbonylamino)-4-methoxy-benzyloxyethyl]-4-oxa-1-azabicyclo[3.2.0]heptan-7-one [(5RS)-19]: (5RS)-15 (49 mg, 0.17 mmol, 1 : 1) and di-tert-butyl dicarbonate (1.0 mL, 4.3 mmol) were kept at 25 °C for 65 h. tert-Butanol and excess dicarbonate were removed by Kugelrohr distillation. The residue was purified by chromatography on silica gel (8 g) using tert-butyl methyl ether/hexanes (1 : 2) as eluant, affording 54 mg (83%) (5RS)-19 (1 : 1), R_F = 0.12, as a colourless oil. No conditions were found for diastereomer separation. - IR (KBr): 3430 (NH), 1780 (lactam-C=O), 1724 cm^{-1} (urethane-C=O). - ^{1}H NMR (from the mixture, $CDCl_3$,

200 MHz), (5S)-**19**: δ = 1.54 (s, CH_3); 1.80-2.02 (m, 1'-H_2), 2.65 (ddd, $J_{2,2}$ = 11.8 Hz, $J_{2,3}$ = 7.5 Hz, $J_{2,6\beta}$ = 0.9 Hz, 2-H_α); 2.85 (d, J = 16.5 Hz, 6-H_α); 3.26 (ddd, $J_{6,6}$ = 16.5 Hz, $J_{6,5}$ = 2.9 Hz, $J_{6,2\alpha}$ = 0.9 Hz, 6-H_β); 3.57 (t, J = 6.6 Hz, 2'-H_2), 3.88 (s, OCH_3); 3.98 (ddd, $J_{2,2}$ = 11.8 Hz, $J_{2,3}$ = 6.2 Hz, $J_{2,5}$ = 0.5 Hz, 2-H_β); 5.31 (dd, $J_{5,6\beta}$ = 0.5 Hz, 5-H); 6.85 (d, J = 8.3 Hz, 5"-H); 6.99 (dd, $J_{6,5}$ = 8.3 Hz, $J_{6,2}$ = 2.2 Hz, 6"-H); 7.13 (b.s, NH); 8.10 ppm (d, J = 2.2 Hz, 2"-H). - (5R)-**19**: δ = 1.54 (s, CH_3); 1.80-2.02 (m, 1'-H_2); 2.85 (d, J = 16.5 Hz, 6-H_β); 3.17 (ddd, $J_{2,2}$ = 10.5 Hz, $J_{2,3}$ = 6.8 Hz, $J_{2,6\alpha}$ = 0.8 Hz, 2-H_β); 3.24 (ddd, $J_{6,6}$ = 16.5 Hz, $J_{6,5}$ = 2.5 Hz, $J_{6,2\beta}$ = 0.8 Hz, 6-H_α); 3.40 (dd, $J_{2,2}$ = 10.5 Hz, $J_{2,3}$ = 7.6 Hz, 2-H_α); 3.57 (t, J = 6.6 Hz, 2'-H_2); 3.88 (s, OCH_3); 5.15 (d, J = 2.5 Hz, 5-H), and others of aryl part.
$C_{20}H_{28}N_2O_6$ (392.46). Calc. C 61.21 H 7.19. Found C 61.40 H 7.31.

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REFERENCES AND FOOTNOTES

1. M. Wanning, H. Zähner, B. Krone, A. Zeeck, Tetrahedron Lett. **22** (1981) 2539.
2. In naming antibiotics, the term clavam replaces the IUPAP name 4-oxa-1-azabicyclo[3.2.0]-heptan-7-one.
3. Part of the Dissertation of T. Hilpert, University of Göttingen, 1986.
4. D. L. Pruess, M. Kellett, J. Antibiot. **36** (1983) 208; R. H. Evans, Jr., H. Ax, A. Jacoby, T. H. Williams, E. Jenkins, J. P. Scannell, ibid. **36** (1983) 213.
5. J.-C. Müller, V. Toome, D. L. Pruess, J. F. Blount, M. Weigele, J. Antibiot. **36** (1983) 217.
6. S. De Bernardo, J. P. Tengi, G. J. Sasso, M. Weigele, J. Org. Chem. **50** (1985) 3457.
7. D. Brown, J. R. Evans, R. A. Fletton, J. Chem. Soc., Chem. Commun. **1979**, 282.
8. A. Zeeck, personal communication.
9. P. H. Bentley, E. Hunt, J. Chem. Soc., Perkin I, **1980**, 2222.
10. K. Clauss, D. Grimm, G. Prossel, Liebigs Ann. Chem. **1974**, 539. - Review: S. Mickel, Aldrichim. Acta **18** (1985) 95.
11. "Non-stereospecific substitution" means, that no correlation is observed between the configurations of educt and product. However, substitutions in 4-acetoxy-2-azetidinones may proceed with essentially complete stereoselectivity if they are governed by a substituent in 3-position.
12. E. J. Corey, H. Niwa, J. Knolle, J. Am. Chem. Soc. **100** (1978) 1942.
13. A. I. Meyers, J. P. Lawson, Tetrahedron Lett. **23** (1982) 4883.
14. S. Saito, T. Hasegawa, M. Inaba, R. Nishida, T. Fujii, S. Nomizu, T. Moriwake, Chemistry Lett. **1984**, 1389.
15. D. Hoppe, unpublished results (1980).
16. A. P. Kieboom, J. F. de Kreuk, H. van Bekkum, J. Catal. **20** (1971) 58; Chem. Abstr. **74** (1971) 99173w.
17. R. Quelet, J. Allard, J. Ducasse, Y. Germain, Bull. Soc. Chim. Fr. [4] **1937**, 1092.
18. Zinc acetate, as used in ref. 9, caused complete decomposition.
19. W. Schmidt, E. Steckhan, Angew. Chem. **91** (1979) 851; Angew. Chem., Int. Ed. Engl. **18** (1979) 802.
20. K. Horita, T. Yoshioka, T. Tanaka, Y. Oikawa, O. Yonemitsu, Tetrahedron **42** (1986) 3021.
21. For natural (-)-**1**, $[\alpha]_D^{23}$ = -141 (c = 0.1, $CHCl_3$) was reported.[1]
22. The ^1H NMR experiments were performed by R. Machinek (University of Göttingen).
23. Ehrlich's reagent is prepared by dissolving 4-(dimethylamino)benzaldehyde (1.0 g) in methanol (75 mL) and aq. hydrochloric acid (36%, 25 mL).
24. Very recently, the isolation of new, structurally related antibiotics (clavamycins) from S. hygroscopicus was reported: H. D. King, J. Langhärig, J. J. Sanglier, J. Antibiot. **39**, 510 (1986); H. U. Naegeli, H.-R. Loosli, A. Nussbaumer, ibid. **39**, 516 (1986).