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A stereoselective synthesis of an α-substituted α-amino acid as a substructure for the construction of myriocin

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ABSTRACT

A synthetic route to the protected quaternary α-amino acid **2** with a hydroxylated side chain has been achieved. The key transformations are the diastereoselective substrate-controlled epoxidation of allylic alcohol **4**; a highly stereoselective oxidation–reduction protocol, and the excellent regioselective isomerization of the oxazolidinone ring to give an oxazinanone skeleton in derivative **3**. The carboxylic acid **2** obtained represents the polar substructure, which is present in myriocin **1**.

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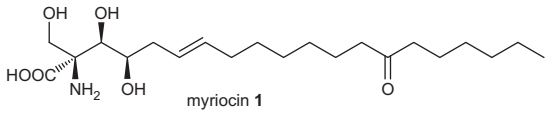
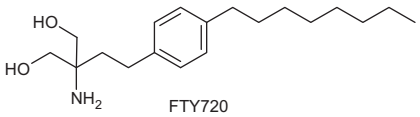
1. Introduction

(+)-Myriocin **1** (also known as thermozymocidine or ISP-I, Fig. 1) is a natural α-substituted α-amino acid, which was initially isolated from two fungal sources as an antifungal agent.[1] In 1994, Fujita et al.[2] isolated the same molecule from the fermentation broth of *Isaria sinclairii* (ATCC 24400) and showed that the immunosuppressive activity of **1** was approximately 10–100 times more potent than that of cyclosporin A.[2] Myriocin **1** has also been reported to have an inhibitory activity[3] against serine palmitoyltransferase (SPT),[4] a key enzyme, which catalyses the first step of the biosynthesis of the sphingolipids. Due to this activity, the antiatherosclerotic properties[5] of **1** have been also examined. Moreover, myriocin **1** have been utilized as a lead compound[6] for the development of novel immunosuppressive agents such as FTY720[7] (Fig. 1), which was approved as a new treatment for multiple sclerosis.

The structure of **1** was established as 2-amino-3,4-dihydroxy-2-hydroxymethyl-14-oxoeicos-6-enoic acid;[1b,c] its absolute configuration was assigned by Payette and Just.[9b,c] Both, the interesting structure of myriocin, which possesses an α-substituted α-amino acid motif, as well as its remarkable bioactivity, have prompted a number of synthetic studies; several total syntheses[8] of **1** and its analogues[8c,e,j,9] have been developed from various starting materials, especially from carbohydrates. From the structural point of view, the polar fragment of myriocin **1** contains an α-substituted serine scaffold linked to two stereogenic centres bearing hydroxyl groups and its construction is certainly the most difficult part of any total synthesis. Prompted by this fact, and in a continuation



myriocin **1**



FTY720

Figure 1. Structures of myriocin **1** and the unnatural immunosuppressant FTY720.

of our previous success in the preparation of substructures possessing a densely functionalized quaternary stereocentre,[10] we decided to attempt the synthesis of the polar head group of **1**. Herein we report the full details of our strategy based on the retrosynthetic plan outlined in Scheme 1.

2. Results and discussion

Our analysis, as illustrated in Scheme 1, envisaged that structure **2**, which possesses three requisite asymmetric centres, could be obtained from the highly functionalized alcohol **3** via an oxidation stereoselective reduction sequence and the base promoted isomerization of an oxazolidinone ring to an oxazinanone. Compound **3** was planned to be constructed from allylic alcohol **4** through an epoxidation and the reductive regioselective ring-opening of an epoxide. For the preparation of derivative **4**, the known furanose **5**[11b] was used as the appropriate starting material.

As shown in Scheme 2, the synthesis of allylic alcohol **4** started with the known and highly functionalized 3-*C*-hydroxymethyl-α-

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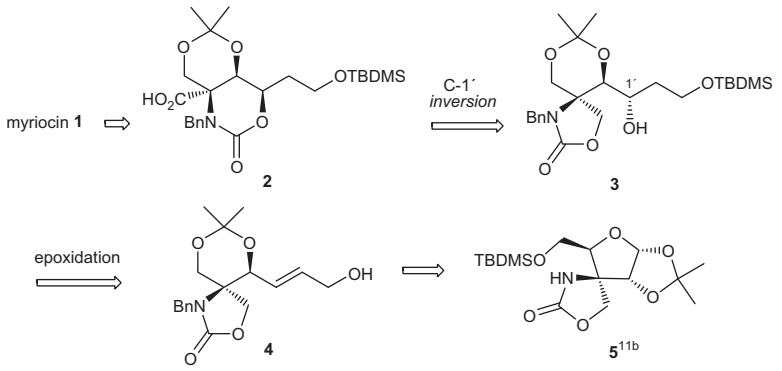
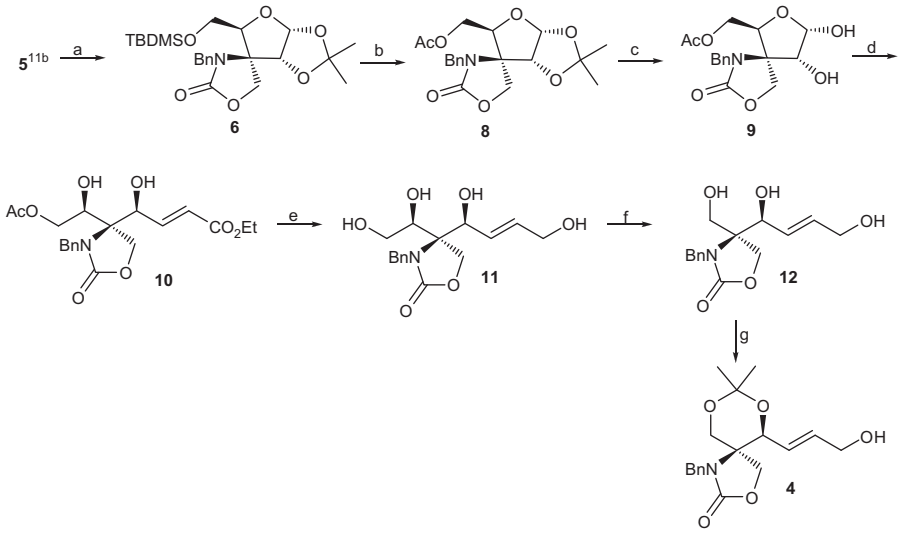


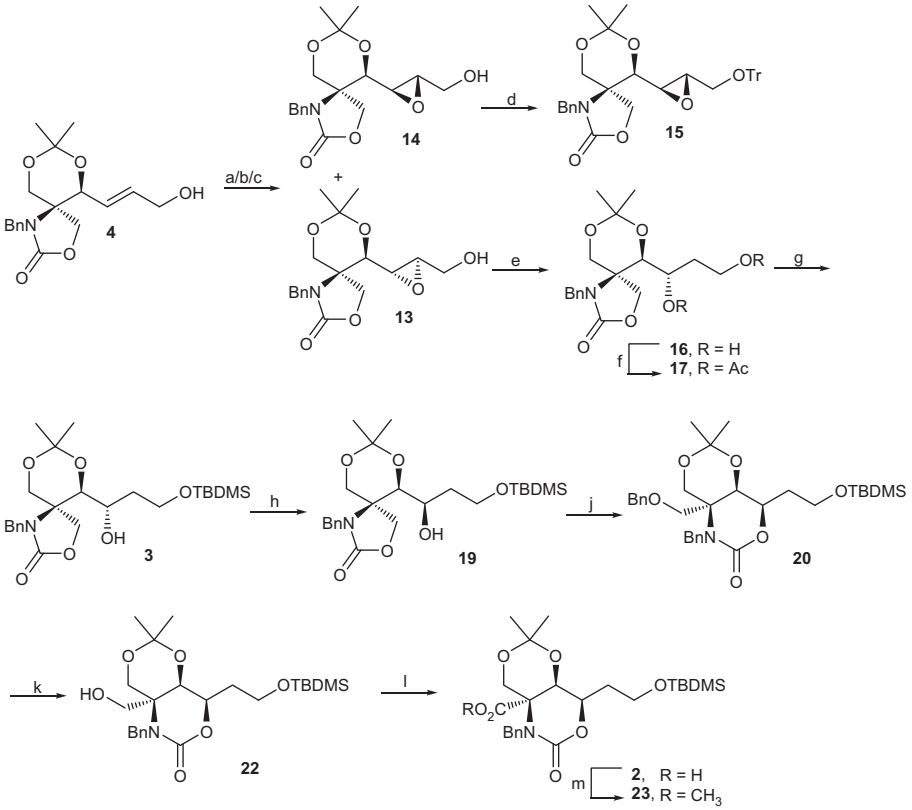
Scheme 1. Retrosynthetic analysis.



Scheme 2. Reagents and conditions: (a) NaH, BnBr, TBAI, 0 °C→rt, 94%; (b) (i) TBAF, THF, 0 °C→rt, **7**, 99%; (ii) Ac_2O, pyridine, DMAP, rt, 98%; (c) TFA/H_2O (85:15), rt, 83%; (d) $Ph_3P{=}CHCO_2Et$, $PhCO_2H$, CH_2Cl_2, rt, 68%; (e) DIBAL-H, CH_2Cl_2, −78 °C, 88%; (f) (i) $NaIO_4$, CH_3OH/H_2O, rt; (ii) $NaBH_4$, EtOH, 0 °C→rt, 64% (after two steps); (g) 2,2-DMP, CSA, 40 °C, 86%.

D-xylofuranose **5**,[11b] which was achieved on a multigram scale according to the combined literature protocols.[10b,11] Exposure of **5** to benzyl bromide in dry DMF using sodium hydride and catalytic amounts of tetrabutylammonium iodide resulted in the formation of the completely protected derivative **6** in 94% yield (Scheme 2). After routine deprotection of **6** with TBAF, the alcohol **7** (99%) obtained was converted into acetate **8** in 98% yield by treatment with Ac_2O in dry pyridine. The acetonide group in **8** was removed by acid hydrolysis (TFA/H_2O) to afford crystalline α-D-xylofuranose derivative **9** as the sole product (83%, Scheme 2); its structure and anomeric configuration were determined by NMR spectroscopic analysis, including NOE experiments. Wittig reaction of **9** with a stabilized ylide, $Ph_3P{=}CHCO_2Et$, (CH_2Cl_2, benzoic acid) furnished (*E*)-α,β-unsaturated ester **10** exclusively in 68% yield (Scheme 2). The observed coupling constant in **10** (J = 15.6 Hz) clearly proved the *trans*-configuration of the C=C bond. Reduction of both ester groups in **10** with DIBAL-H in CH_2Cl_2 at −78 °C gave the corresponding tetrol **11** (88%), which after rapid ^{1}H NMR analysis was used immediately in the next reaction. Oxidative cleavage of **11** with $NaIO_4$ in CH_3OH/H_2O (1:1), followed by reduction ($NaBH_4$, EtOH) of the resultant aldehyde group afforded compound **12** in 64% yield after two reaction steps (Scheme 2). Treatment of **12** with 2,2-DMP and catalytic amounts of CSA resulted in the formation of the corresponding isopropylidene derivative **4** (86%). With allylic alcohol **4** in hand, we were now in a position to explore the crucial step, the stereoselective epoxidation. Initially, exposure of **4** to *m*-CPBA in CH_2Cl_2 produced the corresponding epoxy alcohols **13** and **14** in 85% yield (Scheme 3) as a readily separable mixture of diastereoisomers (**13**:**14** = 84:16 ratio, determined by ^{1}H NMR spectroscopy). On the other hand, the epoxidation of the C=C bond in **13** employing Sharpless asymmetric conditions (SAE)[12] was unsuccessful and afforded an inseparable mixture of the allylic alcohol **4** and the corresponding epoxides **13** and **14** [**13**:**14** = ~80:20 ratio for both auxiliaries used L-(+)-DET and D-(−)-DET, as determined by ^{1}H NMR]. After 5 days, the reaction mixture still contained approximately 70–80% of the starting compound **4** (judged by ^{1}H NMR) and thus it could not be utilized preparatively. These results suggest that the stereochemical outcome of the epoxidation is controlled by the highly hindered substrate and cannot be overridden by asymmetric conditions.

In order to rationalize the observed stereoselectivity in the epoxidation, high-level density functional theory (DFT) calculations, which included electron correlation effects, were carried out. Geometries of the transition states were optimized using B3LYP/6-31G(d,p) with a JAGUAR 7.7 programme.[13] The nature of the vacuum B3LYP transition states was verified with frequency calculations, yielding only one large imaginary frequency. Harmonic zero-point energy corrections at B3LYP/6-31G(d,p) obtained from the frequency calculations of the vacuum transition states were applied to the transition-state energies. Single-point energies



Scheme 3. Reagents and conditions: (a) *m*-CPBA, CH_2Cl_2, 0 °C→rt, 85%; (b) L-(+)-DET (0.3 equiv), TBHP (1.4 equiv), Ti(OPr-*i*)$_4$ (0.25 equiv), CH_2Cl_2, 4 Å MS, −18 °C; (c) D-(−)-DET (0.12 equiv), TBHP (1.5 equiv), Ti(OPr-*i*)$_4$ (0.1 equiv), CH_2Cl_2, 4 Å MS, 18 °C; (d) TrCl, Et_3N, DMAP, CH_2Cl_2, rt, 56%; (e) Red-Al, THF, 0 °C, 62%; (f) Ac_2O, pyridine, DMAP, rt, 74%; (g) TBDMSCl, Et_3N, DMAP, DMF, rt, 65%; (h) (i) IBX, CH_3CN, reflux, **18**, 93%; (ii) L-selectride, THF, −78 °C, 94%; (j) NaH, BnBr, DMF, 0 °C→rt, 89%; (k) (i) H_2, $Pd(OH)_2$/C, EtOH, 10 atm, rt, **21**; (ii) TBDMSCl, imidazole, CH_2Cl_2, 76% (after two steps); (l) (i) IBX, CH_3CN, reflux; (ii) $NaClO_2$, CH_3CN/*t*-BuOH/2-methylbut-2-ene (4:4:1), NaH_2PO_4, 0 °C, 72% (after two steps); (m) CH_3I, K_2CO_3, DMF, rt, 65%.

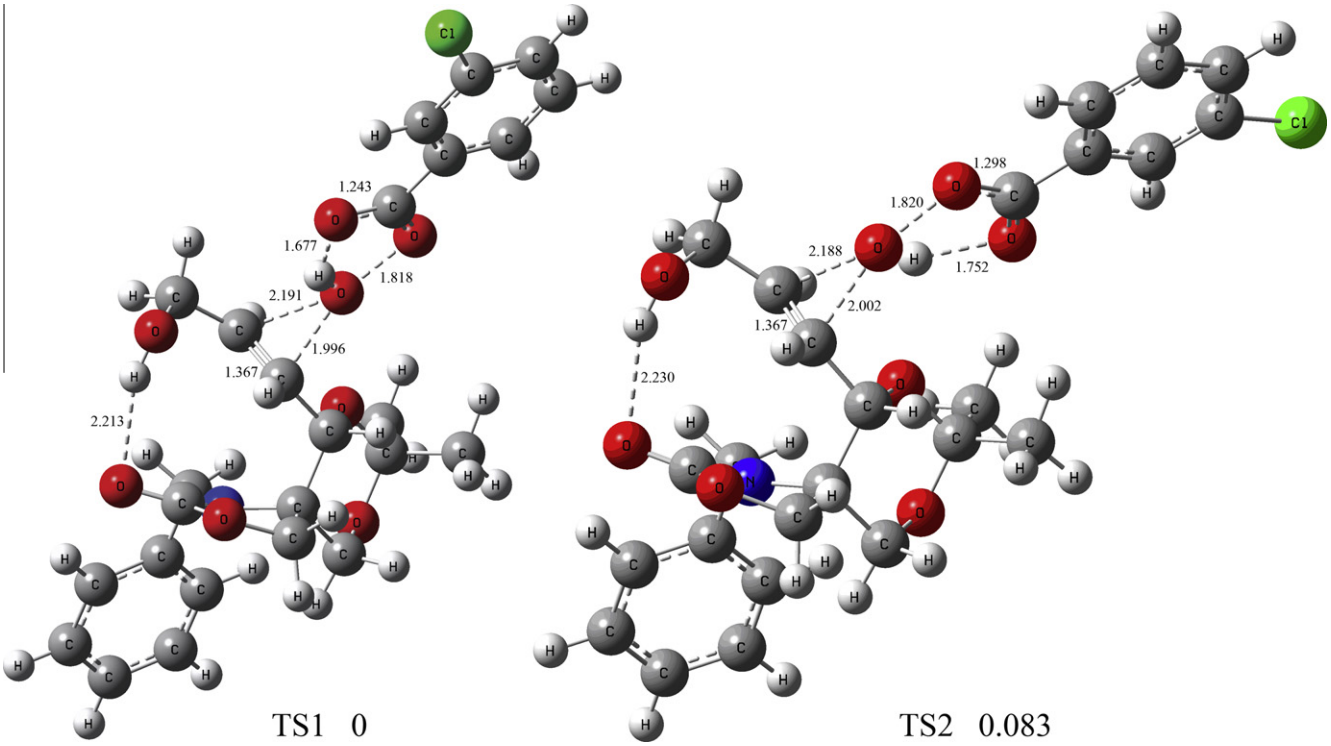


Figure 2. Transition structures TS1 and TS2 for the rearrangement **4**→**13**. Relative energies of transition states (in kcal/mol) and bond distances (in Å) are shown.

were computed with the B3LYP density functional method and the cc-pvTZ basis set. The solvent effect was taken into account via a single-point calculation in a dielectric continuum representing dichloromethane as the solvent. A standard Poisson–Boltzmann continuum solvation model was applied as implemented in JAGUAR 7.7.[13] The epoxidation of allylic alcohol **4** to (2′*R*,3′*R*)-**13** occurs via transition states TS1 and TS2 with relative free energies 0 and 0.083 kcal/mol (Fig. 2); for the conversion of **4** into **14** we have found other two transition states, TS3 and TS4, with relative free energies 1.285 and 0.225 kcal/mol (Fig. 3).

The process is concerted but asynchronous, and calculated geometries were in good agreement with the published results.[14] From the calculations, for the pathway **4**→TS1→**13**, the activation energy was found to be 0.225 kcal/mol lower than for the pathway **4**→TS4→**14**. Thus, the predicted diastereomeric ratio of epoxides **13**:**14** at 0 °C was 60:40. These results are in relatively good agreement with the experimental data (**13**:**14** = 84:16), with the correct prediction of epoxide **13** as the predominant diastereoisomer. These results provide an initial step in understanding the epoxidation; the observed diastereoselectivity seems to depend on many factors that are still to be explored.

The relative stereochemistry of the isolated epoxides **13** and **14** was assigned by X-ray analysis of the tritylated derivative **15** (56%, Scheme 3), which was prepared during our search for the optimal crystalline derivate appropriate for X-ray analysis. This compound was obtained from the minor epoxy alcohol **14** by the reaction of triphenylmethyl chloride in the presence of Et_3N and DMAP and afforded crystals suitable for X-ray measurements. From the crystallographic structure shown in Figure 4, it can be seen that the minor diastereoisomer **14** possesses a (2′*S*,3′*S*)-configuration.

Having incorporated the third stereogenic centre in the key intermediate **13** and determined its stereochemistry (2′*R*,3′*R*), we next focused our attention on the finalization of the synthesis of the polar part of **1**. On the basis of our synthetic plan (Scheme 1), the major epoxy alcohol **13** was reduced with Red-Al[15] in THF at 0 °C to give rise to the desired diol **16** (62%, Scheme 3) with excellent regioselectivity (the second possible isomer was not detected, as determined by ^1H NMR analysis of the crude reaction mixture). The lower 62% yield for this step was most likely due to the formation of some minor unidentified products. Due to the overlap of many proton signals in **16**, it was not possible to determine all of the coupling constants and therefore compound **16** was characterized in the form of diacetate **17** (Ac_2O, pyridine, DMAP, 74%, Scheme 3). The unmasked hydroxyl group in the resulting diol **16** was selectively silylated (TBDMSCl, Et_3N, DMAP and DMF) to afford **3** in 65% yield (Scheme 3). The stereochemistry of the C-1′ (*S*)-configured secondary alcohol **3** was inverted through an oxidation–stereoselective reduction sequence. Thus, the IBX[16] mediated oxidation of **3** resulted in the formation of ketone **18** (93%). Its reduction with the sterically demanding L-selectride in THF at −78 °C gave the requisite C-1′ (*R*)-configured alcohol **19** as a single diastereomer in 94% yield (Scheme 3). Having successfully established the stereochemistry at the C-2 position in **19**, our next task was to develop a suitable synthetic protocol for the base-induced opening of the oxazolidinone ring in **19**, followed by an oxidation of the released hydroxymethyl moiety. In order to avoid problems with the protection of the secondary hydroxyl group in **19**, we turned our attention to the isomerization[17] of **19** to **20**. Interestingly, compound **19** in the presence of sodium hydride and benzyl bromide was converted into the corresponding oxazinanone derivative **20** exclusively in 89% yield (Scheme 3). In order to complete our synthesis, the obtained molecule **20** was then subjected to hydrogenolysis in EtOH using 20% $Pd(OH)_2/C$ (Pearlman's catalyst);[18] however this removed both the *O*-benzyl and TBDMS group

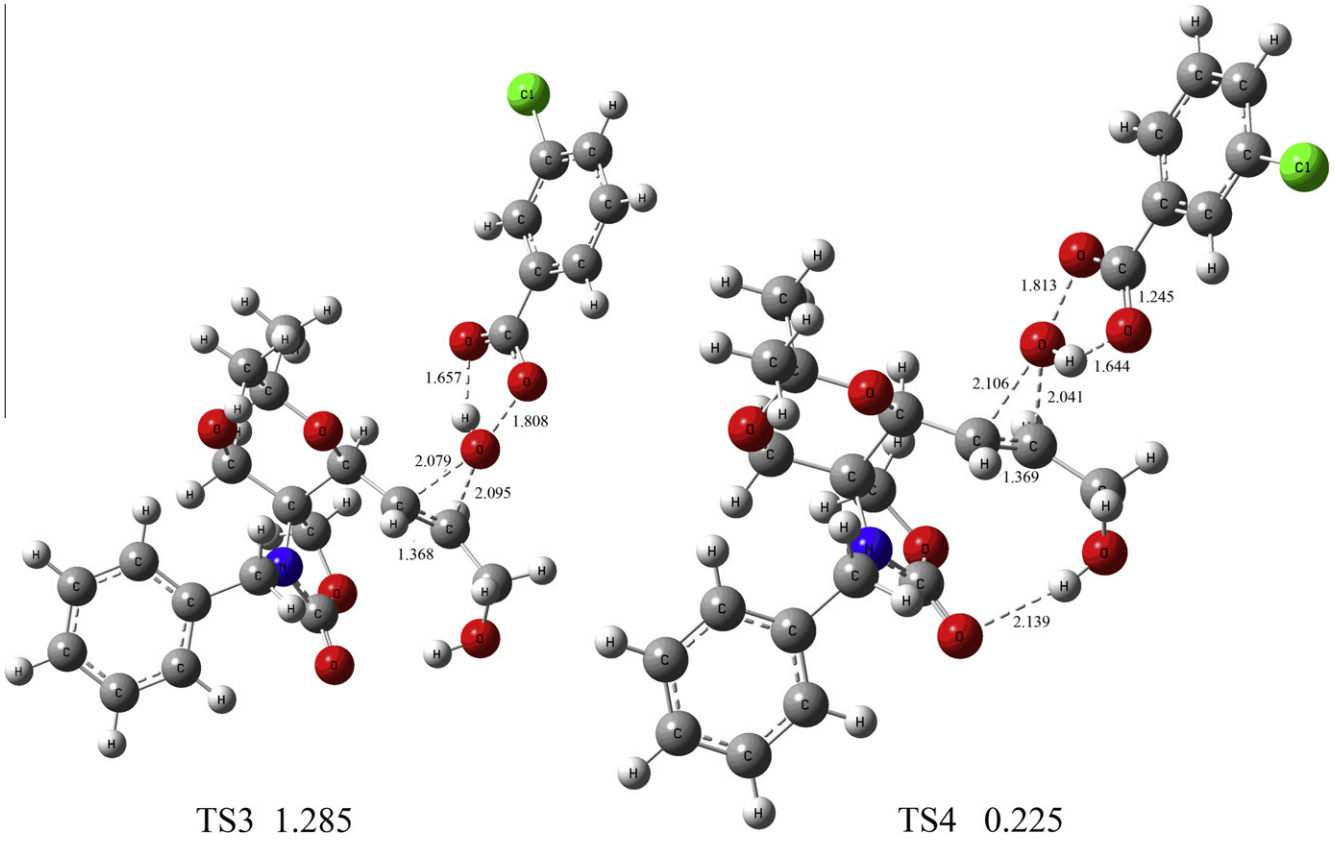


Figure 3. Transition structures TS3 and TS4 for the rearrangement **4**→**14**. Relative energies of transition states (in kcal/mol) and bond distances (in Å) are shown.

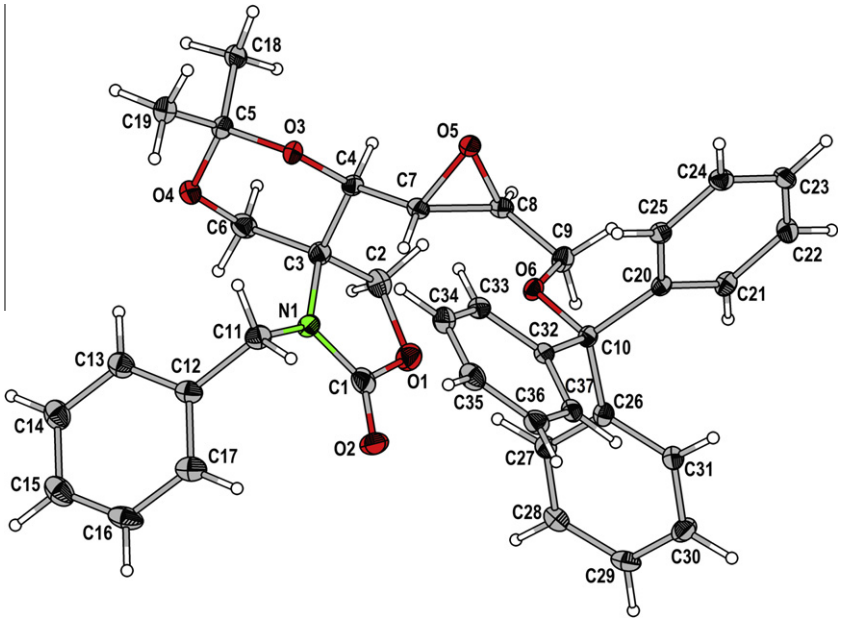


Figure 4. ORTEP structure of compound **15** showing the crystallographic numbering.

to furnish compound **21**. This product, after a short purification through a pad of silica gel and a rapid ^{1}H NMR analysis, was selectively protected as a *tert*-butyldimethylsilyl ether using TBDMSCl and imidazole in dry CH_2Cl_2 to produce **22** in 76% yield after a two-step sequence (Scheme 3). Finally, its oxidation with IBX in acetonitrile followed by $NaClO_2$ treatment provided the protected amino acid **2** (72%), which possesses the correct stereochemistry at all of the stereogenic centres. As an additional confirmation of structure, we transformed **2** to its ester **23** according to standard procedure[19] (CH_3I, K_2CO_3, DMF, Scheme 3).

3. Conclusions

In conclusion, a stereoselective approach towards the protected α-substituted α-amino acid **2**, representing the polar head group of the natural immunosuppressant myriocin **1**, has been developed. The key transformations of our strategy were the implementation of a hydroxyl-bearing asymmetric centre into **4** using a diastereoselective substrate-controlled epoxidation, oxidation highly stereoselective reduction sequence, and the regioselective isomerization of an oxazolidinone moiety into an oxazinanone in **3** in order to establish the structure with the free hydroxymethyl functionality as the source of the carboxylic acid group. The final acid **2** has the protected primary hydroxyl group as a tool for the construction of a non-polar side chain with C=O and C=C functionalities at the required positions via a Wittig reaction or by using Grubb's catalyst-mediated olefin cross metathesis.

4. Experimental

4.1. General methods

All commercial reagents were used in the highest available purity from Aldrich, Fluka, Merck or Acros Organics without further purification. Solvents were dried and purified before use according to standard procedures. For flash column chromatography on silica gel, Kieselgel 60 (0.040–0.063 mm, 230–400 mesh, Merck) was used. Solvents for flash chromatography (hexane, ethyl acetate, methanol, dichloromethane) were distilled before use. Thin layer chromatography was run on Merck silica gel 60 F_{254} analytical plates; detection was carried out with either ultraviolet light (254 nm), or spraying with a solution of phosphomolybdic acid, a basic potassium permanganate solution, or a solution of concentrated H_2SO_4, with subsequent heating. ^{1}H NMR and ^{13}C NMR spectra were recorded in $CDCl_3$, CD_3OD on a Varian Mercury Plus 400 FT NMR (400.13 MHz for ^{1}H and 100.6 MHz for ^{13}C) or on a Varian Premium Compact 600 (599.87 MHz for ^{1}H and 150.84 MHz for ^{13}C) spectrometers using TMS as internal reference. For ^{1}H δ are given in parts per million (ppm) relative to TMS (δ = 0.0) and for ^{13}C relative to $CDCl_3$ (δ = 77.0), CD_3OD (δ = 49.05). The multiplicity of the ^{13}C NMR signals concerning the ^{13}C–^{1}H coupling was determined by the DEPT method. Chemical shifts (in ppm) and coupling constants (in Hz) were obtained by first-order analysis; assignments were derived from COSY and H/C correlation spectra. Infrared (IR) spectra were measured with a Nicolet Avatar 330 FT-IR spectrometer as KBr pellets and expressed in ν values (cm^{-1}). Optical rotations were measured on a P-2000 Jasco polarimeter and reported as follows: $[\alpha]_D$ (c in grams per 100 mL, solvent). Melting points were recorded on a Kofler hot block and are uncorrected. Small quantities of reagents (μL) were measured with appropriate syringes (Hamilton). All reactions were performed under an atmosphere of nitrogen, unless otherwise noted.

4.2. (3a*R*,4′*S*,5*S*,6a*R*)-3′-Benzyl-5-{[(*tert*-butyldimethylsilyl) oxy]-methyl}-2,2-dimethyldihydro-3a*H*-spiro[furo[2,3-*d*] [1,3]dioxole-6,4′-oxazolidin]-2′-one 6

To a solution of **5**[11b] (6.35 g, 17.7 mmol) in dry DMF (43.5 mL), which was pre-cooled to 0 °C, NaH (1.06 g, 44.2 mmol of a 60% dispersion in mineral oil) was added and the reaction mixture was stirred at the same temperature for 10 min. Then, tetrabutylammonium iodide (66 mg, 0.18 mmol) was added followed by dropwise addition of benzyl bromide (2.54 mL, 21.2 mmol). The resulting mixture was stirred at 0 °C for 10 min and at room temperature for another 20 min. The excess hydride was decomposed with methanol (1.5 mL), and the mixture was partitioned between ice water (100 mL) and Et_2O (75 mL). The aqueous phase was extracted with an additional portion of Et_2O (75 mL), the combined organic layers were dried over Na_2SO_4, stripped of solvent, and the residue was subjected to flash chromatography on silica gel

hexane/EtOAc (7:1) to afford 7.47 g (94%) of crystalline compound **6**. Mp 105–106 °C, $[\alpha]_D^{25} = +155.3$ (*c* 0.18, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$): δ 0.07 (s, 6H, 2× CH_3), 0.89 (s, 9H, 3× CH_3), 1.12 (s, 3H, CH_3), 1.45 (s, 3H, CH_3), 3.71 (dd, 1H, $J_{H,H}$ = 10.4 Hz, $J_{5,H}$ = 8.3 Hz, CH_2O), 3.94 (dd, 1H, $J_{H,H}$ = 10.4 Hz, $J_{5,H}$ = 4.8 Hz, CH_2O), 4.05 (dd, 1H, $J_{5,H}$ = 8.3 Hz, $J_{5,H}$ = 4.8 Hz, H-5), 4.16 (d, 1H, $J_{6a,3a}$ = 3.8 Hz, H-3a), 4.17 (d, 1H, $J_{H,H}$ = 15.8 Hz, NCH_2Ph), 4.26 (d, 1H, $J_{5',5'}$ = 9.5 Hz, H-5′), 4.54 (d, 1H, $J_{5',5'}$ = 9.5 Hz, H-5′), 4.87 (d, 1H, $J_{H,H}$ = 15.8 Hz, NCH_2Ph), 5.64 (d, 1H, $J_{6a,3a}$ = 3.8 Hz, H-6a), 7.28–7.37 (m, 5H, Ph). ^{13}C NMR (100 MHz, $CDCl_3$): δ −5.7 (CH_3), −5.6 (CH_3), 18.2 (C_q), 25.8 (4× CH_3), 26.3 (CH_3), 46.8 (NCH_2Ph), 60.1 (CH_2O), 64.7 (C-5′), 70.3 (C-4′), 81.9 (C-5), 83.6 (C-3a), 104.0 (C-6a), 112.2 (C-2), 127.7 (2× CH_{Ph}), 128.0 (CH_{Ph}), 128.8 (2× CH_{Ph}), 137.5 (C_i), 158.7 (C=O). Anal. Calcd for $C_{23}H_{35}NO_6Si$: C, 61.44; H, 7.85; N, 3.12. Found: C, 61.50; H, 7.80; N, 3.15.

4.3. (3a*R*,4′*S*,5*S*,6a*R*)-3′-Benzyl-5-(hydroxymethyl)-2,2-dimethyldihydro-3a*H*-spiro[furo[2,3-*d*][1,3]dioxole-6,4′-oxazolidin]-2′-one 7

To a solution of **6** (7.46 g, 16.6 mmol) in dry THF (166 mL) was added dropwise a 1 M solution of TBAF (16.6 mL, 16.6 mmol) in THF at 0 °C. The resulting reaction mixture was stirred for a further 10 min at 0 °C and then at room temperature for 20 min. The solvent was evaporated in vacuo, and the residue was partitioned between EtOAc (70 mL) and water (35 mL). The aqueous phase was washed with further portions of EtOAc (2 × 30 mL). The organic layers were combined, dried over Na_2SO_4 and stripped of solvent. The crude product was chromatographed on silica gel hexane/EtOAc (1:1) to give 5.51 g (99%) of compound **7** as a colourless oil. $[\alpha]_D^{25} = +154.0$ (*c* 0.18, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$): δ 1.12 (s, 3H, CH_3), 1.45 (s, 3H, CH_3), 3.83 (dd, 1H, $J_{H,H}$ = 11.5 Hz, $J_{5,H}$ = 5.9 Hz, CH_2O), 3.94 (dd, 1H, $J_{H,H}$ = 11.5 Hz, $J_{5,H}$ = 6.3 Hz, CH_2O), 4.12–4.19 (m, 3H, H-5, NCH_2Ph, H-3a), 4.23 (d, 1H, $J_{5',5'}$ = 9.8 Hz, H-5′), 4.58 (d, 1H, $J_{5',5'}$ = 9.8 Hz, H-5′), 4.92 (d, 1H, $J_{H,H}$ = 15.9 Hz, NCH_2Ph), 5.67 (d, 1H, $J_{6a,3a}$ = 3.7 Hz, H-6a), 7.29–7.39 (m, 5H, Ph). ^{13}C NMR (100 MHz, $CDCl_3$): δ 25.8 (CH_3), 26.3 (CH_3), 46.8 (NCH_2Ph), 59.6 (CH_2O), 64.7 (C-5′), 70.2 (C-4′), 82.4 (C-5), 83.5 (C-3a), 104.1 (C-6a), 112.4 (C-2), 127.6 (2× CH_{Ph}), 128.2 (CH_{Ph}), 128.9 (2× CH_{Ph}), 137.3 (C_i), 158.7 (C=O). Anal. Calcd for $C_{17}H_{21}NO_6$: C, 60.89; H, 6.31; N, 4.18. Found: C, 60.95; H, 6.27; N, 4.15.

4.4. (3a*R*,4′*S*,5*S*,6a*R*)-5-(Acetoxymethyl)-3′-benzyl-2,2-dimethyldihydro-3a*H*-spiro[furo[2,3-*d*][1,3]dioxole-6,4′-oxazolidin]-2′-one 8

To a solution of **7** (5.51 g, 16.4 mmol) in dry pyridine (126 mL) were successively added Ac_2O (2.33 mL, 24.6 mmol) and DMAP (0.20 g, 1.64 mmol) at room temperature. The resulting mixture was stirred at the same temperature for 30 min, then poured into ice water (550 mL) and extracted with CH_2Cl_2 (3 × 90 mL). The combined organic layers were dried over Na_2SO_4, the solvent was evaporated, and the residue was purified by flash chromatography on silica gel hexane/EtOAc (2:1) to afford 6.08 g (98%) of crystalline compound **8**. Mp 144–145 °C, $[\alpha]_D^{25} = +123.2$ (*c* 0.21, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$): δ 1.12 (s, 3H, CH_3), 1.44 (s, 3H, CH_3), 2.11 (s, 3H, CH_3CO), 4.16 (d, 1H, $J_{H,H}$ = 15.8 Hz, NCH_2Ph), 4.17 (d, 1H, $J_{5',5'}$ = 10.0 Hz, H-5′), 4.17 (d, 1H, $J_{6a,3a}$ = 3.8 Hz, H-3a), 4.19 (dd, 1H, $J_{5,H}$ = 6.8 Hz, $J_{5,H}$ = 5.3 Hz, H-5), 4.25 (dd, 1H, $J_{H,H}$ = 12.0 Hz, $J_{5,H}$ = 5.3 Hz, CH_2O), 4.30 (dd, 1H, $J_{H,H}$ = 12.0 Hz, $J_{5,H}$ = 6.8 Hz, CH_2O), 4.59 (d, 1H, $J_{5',5'}$ = 10.0 Hz, H-5′), 4.94 (d, 1H, $J_{H,H}$ = 15.8 Hz, NCH_2Ph), 5.69 (d, 1H, $J_{6a,3a}$ = 3.8 Hz, H-6a), 7.29–7.39 (m, 5H, Ph). ^{13}C NMR (100 MHz, $CDCl_3$): δ 20.7 (CH_3), 25.7 (CH_3), 26.3 (CH_3), 46.8 (NCH_2Ph), 60.8 (CH_2O), 64.4 (C-5′), 70.3 (C-4′), 80.0 (C-5), 83.3 (C-3a), 104.2 (C-6a), 112.5 (C-2), 127.6 (2× CH_{Ph}), 128.3 (CH_{Ph}), 128.9 (2× CH_{Ph}), 137.1 (C_i), 158.3 (C=O), 170.3 (C=O). Anal. Calcd for $C_{19}H_{23}NO_7$: C, 60.47; H, 6.14; N, 3.71. Found: C, 60.48; H, 6.15; N, 3.76.

4.5. (5*R*,6*S*,8*S*,9*R*)-6-(Acetoxymethyl)-1-benzyl-8,9-dihydroxy-3,7-dioxa-1-azaspiro [4.4]nonan-2-one 9

Compound **8** (6.04 g, 16.0 mmol) was treated with a mixture of 85:15 TFA/H_2O (130 mL) for 8.5 h at room temperature. The solvent was evaporated in vacuo, and the residue was purified through a short column of silica gel hexane/EtOAc (1:1) to yield 4.48 g (83%) of crystalline compound **9**. Mp 162–164 °C, $[\alpha]_D^{25} = +49.1$ (*c* 0.36, CH_3OH). ^1H NMR (400 MHz, CD_3OD): δ 2.06 (s, 3H, CH_3), 4.04 (d, 1H, $J_{9,8}$ = 4.6 Hz, H-9), 4.18–4.21 (m, 2H, CH_2O), 4.23–4.27 (m, 2H, H-4, H-6), 4.37 (d, 1H, $J_{H,H}$ = 16.3 Hz, NCH_2Ph), 4.73 (d, 1H, $J_{4,4}$ = 9.8 Hz, H-4), 4.75 (d, 1H, $J_{H,H}$ = 16.3 Hz, NCH_2Ph), 5.35 (d, 1H, $J_{9,8}$ = 4.6 Hz, H-8), 7.25–7.41 (m, 5H, Ph). ^{13}C NMR (100 MHz, CD_3OD): δ 20.6 (CH_3), 47.6 (NCH_2Ph), 62.9 (CH_2O), 68.3 (C-4), 72.1 (C-5), 75.1 (C-9), 80.9 (C-6), 97.0 (C-8), 128.2 (2× CH_{Ph}), 128.8 (CH_{Ph}), 129.8 (2× CH_{Ph}), 139.0 (C_i), 160.8 (C=O), 172.1 (C=O). Anal. Calcd for $C_{16}H_{19}NO_7$: C, 56.97; H, 5.68; N, 4.15. Found: C, 56.92; H, 5.71; N, 4.18.

4.6. Ethyl (4*S*,2*E*)-4-{(4′*R*)-4′-[(1″*S*)-2″-acetoxy-1″-hydroxyethyl]-3′-benzyl-2′-oxooxazolidin-4′-yl}-4-hydroxybut-2-enoate 10

To a solution of **9** (4.48 g, 13.3 mmol) in dry CH_2Cl_2 (119 mL) were successively added benzoic acid (0.16 g, 1.33 mmol) and stabilized ylide, $Ph_3P{=}CHCO_2Et$, (8.33 g, 23.9 mmol), and the resulting mixture was stirred at room temperature. After the starting material was completely consumed (3 h, judged by TLC), the reaction was stopped and the solvent was removed under reduced pressure. The residue was purified by flash chromatography on silica gel hexane/EtOAc (1:2) to give 3.68 g (68%) of crystalline ester **10**. Mp 111–112 °C, $[\alpha]_D^{25} = +167.5$ (*c* 0.23, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$): δ 1.26 (t, 3H, *J* = 7.1 Hz, CH_3), 2.13 (s, 3H, CH_3CO), 3.29 (m, 1H, OH), 3.91–3.97 (m, 2H, H-1″, H-5′), 4.03 (dd, 1H, $J_{2'',2''}$ = 12.0 Hz, $J_{2'',1''}$ = 6.8 Hz, H-2″), 4.14 (d, 1H, $J_{5',5'}$ = 9.4 Hz, H-5′), 4.16 (q, 2H, *J* = 7.1 Hz, CH_2), 4.31 (dd, 1H, $J_{2'',2''}$ = 12.0 Hz, $J_{2'',1''}$ = 2.5 Hz, H-2″), 4.65 (d, 1H, $J_{H,H}$ = 15.2 Hz, NCH_2Ph), 4.75 (td, 1H, $J_{4,3}$ = 4.3 Hz, $J_{4,OH}$ = 4.3 Hz, $J_{4,2}$ = 1.9 Hz, H-4), 4.82 (d, 1H, $J_{H,H}$ = 15.2 Hz, NCH_2Ph), 6.18 (dd, 1H, $J_{3,2}$ = 15.6 Hz, $J_{4,2}$ = 1.9 Hz, H-2), 6.87 (dd, 1H, $J_{3,2}$ = 15.6 Hz, $J_{4,3}$ = 4.3 Hz, H-3), 7.28–7.39 (m, 3H, Ph), 7.47–7.52 (m, 2H, Ph). ^{13}C NMR (100 MHz, $CDCl_3$): δ 14.1 (CH_3), 20.8 (CH_3), 46.3 (NCH_2Ph), 60.8 (CH_2), 64.7 (C-5′), 65.3 (C-2″), 68.9 (C-4′), 69.2 (C-4), 72.4 (C-1″), 124.2 (C-2), 128.4 (2× CH_{Ph}), 128.4 (CH_{Ph}), 129.3 (2× CH_{Ph}), 137.9 (C_i) 142.6 (C-3), 159.0 (C=O), 166.0 (C=O), 171.3 (C=O). Anal. Calcd for $C_{20}H_{25}NO_8$: C, 58.96; H, 6.18; N, 3.44. Found: C, 59.00; H, 6.14; N, 3.48.

4.7. (4*R*)-3-Benzyl-4-[(1′*S*,2′*E*)-1′,4″-dihydroxybut-2′-enyl]-4-[(1″*S*)-1″,2″-dihydroxyethyl]oxazolidin-2-one 11

Diisobutylaluminum hydride (37.6 mL of a 1.2 M toluene solution) was added dropwise to a solution of ester **10** (3.68 g, 9.03 mmol) in dry CH_2Cl_2 (76.5 mL) at −78 °C. The reaction mixture was stirred at the same temperature for 1 h and then quenched by the slow addition of cooled acetone (15 mL). To the resulting solution was added solid $Na_2SO_4.10H_2O$ (45 g) at 0 °C. After being stirred for 1.5 h at room temperature, the insoluble parts were filtered off and washed with a mixture of 5:1 CH_2Cl_2/MeOH (480 mL). The evaporation of solvents afforded 2.57 g (88%) of the crude compound **11** as a colourless oil, which was used immediately in the next step without further purification to avoid

problems connected with its hydrophilic nature. ^{1}H NMR (400 MHz, CD_3OD): δ 3.55 (dd, 1H, $J_{2'',2''}$ = 11.7 Hz, $J_{2'',1''}$ = 5.5 Hz, H-2″), 3.68 (dd, 1H, $J_{2'',2''}$ = 11.7 Hz, $J_{2'',1''}$ = 3.0 Hz, H-2″), 3.94 (dd, 1H, $J_{2'',1''}$ = 5.5 Hz, $J_{2'',1''}$ = 3.0 Hz, H-1″), 4.01–4.07 (m, 3H, H-4′, H-5, H-1′), 4.35–4.39 (m, 2H, H-4′, H-5), 4.57 (d, 1H, $J_{H,H}$ = 15.6 Hz, NCH_2Ph), 4.64 (d, 1H, $J_{H,H}$ = 15.6 Hz, NCH_2Ph), 5.68 (dd, 1H, $J_{3',2'}$ = 15.4 Hz, $J_{2',1'}$ = 6.4 Hz, H-2′), 5.94 (dt, 1H, $J_{3',2'}$ = 15.4 Hz, $J_{4',3'}$ = 5.1 Hz, $J_{4',3'}$ = 5.1 Hz, H-3′), 7.18–7.23 (m, 1H, Ph), 7.24–7.31 (m, 2H, Ph), 7.41–7.47 (m, 2H, Ph). Anal. Calcd for $C_{16}H_{21}NO_6$: C, 59.43; H, 6.55; N, 4.33. Found: 59.21; H, 6.73; N, 4.20.

4.8. (4*R*)-3-Benzyl-4-[(1′*S*,2′*E*)-1′,4′-dihydroxybut-2′-enyl]-4-(hydroxymethyl)oxazolidin-2-one 12

A solution of **12** (2.57 g, 7.95 mmol) in a mixture of 1:1 CH_3OH/H_2O (29 mL) was treated with solid $NaIO_4$ (2.04 g, 9.54 mmol). The resulting mixture was stirred at room temperature for 30 min and then diluted with CH_2Cl_2 (20 mL). The insoluble materials were removed by filtration, the solvent was evaporated, and the residue was immediately used in the next step without further purification. To a solution of the crude aldehyde (2.31 g, 7.93 mmol) in EtOH (61.0 mL), which was pre-cooled to 0 °C, was added $NaBH_4$ (0.36 g, 9.52 mmol). The mixture was stirred for 10 min at 0 °C and then for an additional 20 min at room temperature. The reaction was quenched by neutralization with Amberlite IR-120 (H^+), the solid parts were filtered off, the solvent was evaporated, and the residue was subjected to flash chromatography on silica gel CH_2Cl_2/MeOH (9:1) to give 1.49 g (64%) of compound **12** as a colourless oil. $[\alpha]_D^{25} = +31.1$ (*c* 0.29), CH_3OH). ^{1}H NMR (400 MHz, CD_3OD): δ 3.50 (d, 1H, $J_{H,H}$ = 11.7 Hz, CH_2O), 3.56 (d, 1H, $J_{H,H}$ = 11.7 Hz, CH_2O), 4.01 (m, 2H, 2× H-4′), 4.10 (d, 1H, $J_{5,5}$ = 8.7 Hz, H-5), 4.20 (d, 1H, $J_{2',1'}$ = 6.8 Hz, H-1′), 4.28 (d, 1H, $J_{5,5}$ = 8.7 Hz, H-5), 4.47 (d, 1H, $J_{H,H}$ = 15.8 Hz, NCH_2Ph), 4.55 (d, 1H, $J_{H,H}$ = 15.8 Hz, NCH_2Ph), 5.61 (dd, 1H, $J_{3',2'}$ = 15.4 Hz, $J_{2',1'}$ = 6.8 Hz, H-2′), 5.87 (dt, 1H, $J_{3',2'}$ = 15.4 Hz, $J_{4',3'}$ = 5.0 Hz, $J_{4',3'}$ = 5.0 Hz, H-3′), 7.19–7.25 (m, 1H, Ph), 7.26–7.31 (m, 2H, Ph), 7.39–7.43 (m, 2H, Ph). ^{13}C NMR (100 MHz, CD_3OD): δ 46.3 (NCH_2Ph), 62.8 (C-4′), 63.2 (CH_2O), 67.5 (C-5), 69.7 (C-4), 72.3 (C-1′), 128.3 (C-2′), 128.5 (CH_{Ph}), 129.1 (2× CH_{Ph}), 129.4 (2× CH_{Ph}), 135.1 (C-3′), 139.6 (C_i), 162.1 (C=O). Anal. Calcd for $C_{15}H_{19}NO_5$: C, 61.42; H, 6.53; N, 4.78. Found: C, 61.45; H, 6.50; N, 4.80.

4.9. (5*R*,6*S*)-1-Benzyl-6-[(*E*)-3′-hydroxyprop-1′-enyl]-8,8-dimethyl-3,7,9-trioxa-1-azaspiro [4.5]decan-2-one 4

To a solution of **12** (1.49 g, 5.08 mmol) in 2,2-dimethoxypropane (20.8 mL) was added CSA (0.23 g, 0.99 mmol) and the resulting mixture was stirred at 40 °C. After the starting material was completely consumed (1 h, judged by TLC), the reaction was stopped and allowed to cool to room temperature. The solvent was evaporated in vacuo, and the residue was partitioned between CH_2Cl_2 (35 mL) and a saturated $NaHCO_3$ solution (46 mL). The organic layer was dried over Na_2SO_4, the solvent was evaporated, and the residue was purified by flash chromatography on silica gel hexane/EtOAc (1:2) to yield 1.46 g (86%) of crystalline alcohol **4**. Mp 132–134 °C, $[\alpha]_D^{25} = +18.9$ (*c* 0.28, CH_3OH). IR (KBr): ν_{max} 3417, 3005, 2920, 2854, 1714, 1439, 1352, 1265, 1178, 1059, 972, 945, 854, 702. ^{1}H NMR (400 MHz, CD_3OD): δ 1.46 (s, 3H, CH_3), 1.49 (s, 3H, CH_3), 3.76 (d, 1H, $J_{10,10}$ = 12.7 Hz, H_{10}), 3.80 (d, 1H, $J_{4,4}$ = 9.6 Hz, H-4), 3.94 (d, 1H, $J_{10,10}$ = 12.7 Hz, H-10), 4.06–4.09 (m, 2H, 2× H-3′), 4.12 (d, 1H, $J_{4,4}$ = 9.6 Hz, H-4), 4.53 (dd, 1H, $J_{6,1'}$ = 6.2 Hz, $J_{6,2'}$ = 1.2 Hz, H-6), 4.74 (d, 1H, $J_{H,H}$ = 15.7 Hz, NCH_2Ph), 4.91 (d, 1H, $J_{H,H}$ = 15.7 Hz, NCH_2Ph), 5.65 (ddt, 1H, $J_{2',1'}$ = 15.5 Hz, $J_{6,1'}$ = 6.2 Hz, $J_{3',1'}$ = 1.7 Hz, $J_{3',1'}$ = 1.7 Hz, H-1′), 6.08 (dtd, 1H, $J_{2',1'}$ = 15.5 Hz, $J_{3',2'}$ = 5.0 Hz, $J_{3',2'}$ = 5.0 Hz, $J_{6,2'}$ = 1.2 Hz, H-2′), 7.19–7.25 (m, 1H, Ph), 7.26–7.32 (m, 2H, Ph), 7.38–7.44 (m, 2H, Ph). ^{13}C NMR (100 MHz, CD_3OD): δ 19.0 (CH_3), 29.4 (CH_3), 48.3 (NCH_2Ph), 60.0 (C-5), 62.9 (C-3′), 66.7 (C-10), 68.5 (C-4), 77.8 (C-6), 100.7 (C-8), 124.4 (C-1′), 128.2 (CH_{Ph}), 128.8 (2× CH_{Ph}), 129.3 (2× CH_{Ph}), 137.6 (C-2′), 139.8 (C_i), 161.2 (C=O). Anal. Calcd for $C_{18}H_{23}NO_5$: C, 64.85; H, 6.95; N, 4.20. Found: C, 64.87; H, 6.92; N, 4.18.

4.10. (5*R*,6*R*)-1-Benzyl-6-[(2′*R*,3′*R*)-3′-(hydroxymethyl)oxiran-2′-yl]-8,8-dimethyl-3,7,9-trioxa-1-azaspiro[4.5]decan-2-one 13 and (5*R*,6*R*)-1-benzyl-6-[(2′*S*,3′*S*)-3′-(hydroxymethyl)oxiran-2′-yl]-8,8-dimethyl-3,7,9-trioxa-1-azaspiro[4.5]decan-2-one 14

To a solution of **4** (1.30 g, 3.90 mmol) in dry CH_2Cl_2 (40 mL), which was pre-cooled to 0 °C, was added *m*-chloroperoxybenzoic acid (57-86%, Aldrich) (1.99 g, 11.53 mmol) in three portions at 10 min intervals. The reaction mixture was stirred for 1 h at 0 °C and for another 26 h at room temperature. Then, the resulting solution was treated with saturated aqueous $NaHCO_3$ (75 mL) and $Na_2S_2O_3$ (75 mL). The organic layer was dried over Na_2SO_4, the solvent was evaporated, and the residue was chromatographed on silica gel hexane/EtOAc (1:2) to afford 0.97 g (71%) of **13** and 0.19 g (14%) of **14** as colourless oils. Diastereoisomer **13**: $[\alpha]_D^{25} = +88.5$ (*c* 0.19, $CHCl_3$). IR (KBr): ν_{max} 3437, 2993, 1743, 1437, 1385, 1263, 1144, 1014, 858, 698. ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.45 (s, 3H, CH_3), 1.53 (s, 3H, CH_3), 2.31 (m, 1H, OH), 3.02 (dd, 1H, $J_{6,2'}$ = 4.9 Hz, $J_{3',2'}$ = 2.1 Hz, H-2′), 3.22 (ddd, 1H, $J_{3',CH2O}$ = 4.3 Hz, $J_{3',CH2O}$ = 3.3 Hz, $J_{3',2'}$ = 2.1 Hz, H-3′), 3.64 (dd, 1H, $J_{H,H}$ = 12.8 Hz, $J_{3',CH2O}$ = 4.3 Hz, CH_2O), 3.73 (d, 1H, $J_{4,4}$ = 9.4 Hz, H-4), 3.76–3.85 (m, 4H, 2× H-10, H-6, CH_2O), 4.21 (d, 1H, $J_{4,4}$ = 9.4 Hz, H-4), 4.79 (d, 1H, $J_{H,H}$ = 15.3 Hz, NCH_2Ph), 4.93 (d, 1H, $J_{H,H}$ = 15.3 Hz, NCH_2Ph), 7.24–7.35 (m, 3H, Ph), 7.44–7.51 (m, 2H, Ph). ^{13}C NMR (100 MHz, $CDCl_3$): δ 18.4 (CH_3), 28.5 (CH_3), 47.2 (NCH_2Ph), 52.8 (C-2′), 55.6 (C-3′), 57.5 (C-5), 61.2 (CH_2O), 65.5 (C-10), 66.8 (C-4), 75.3 (C-6), 99.7 (C-8), 127.5 (CH_{Ph}), 128.0 (2× CH_{Ph}), 128.4 (2× CH_{Ph}), 138.1 (C_i), 158.6 (C=O). Anal. Calcd for $C_{18}H_{23}NO_6$: C, 61.88; H, 6.64; N, 4.01. Found: C, 61.91; H, 6.58; N, 4.02.

Diastereoisomer **14**: $[\alpha]_D^{25} = +79.1$ (*c* 0.18, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.45 (s, 3H, CH_3), 1.54 (s, 3H, CH_3), 3.10 (dd, 1H, $J_{6,2'}$ = 5.4 Hz, $J_{3',2'}$ = 2.3 Hz, H-2′), 3.21 (ddd, 1H, $J_{3',CH2O}$ = 3.9 Hz, $J_{3',CH2O}$ = 2.7 Hz, $J_{3',2'}$ = 2.3 Hz, H-3′), 3.69–3.78 (m, 5H, CH_2O, 2× H-10, H-6, H-4), 3.90 (dd, 1H, $J_{H,H}$ = 12.9 Hz, $J_{3',CH2O}$ = 2.7 Hz, CH_2O), 4.04 (d, 1H, $J_{4,4}$ = 9.6 Hz, H-4), 4.80 (d, 1H, $J_{H,H}$ = 15.5 Hz, NCH_2Ph), 4.92 (d, 1H, $J_{H,H}$ = 15.5 Hz, NCH_2Ph), 7.25–7.35 (m, 3H, Ph), 7.44–7.48 (m, 2H, Ph). ^{13}C NMR (100 MHz, $CDCl_3$): δ 18.4 (CH_3), 28.4 (CH_3), 47.4 (NCH_2Ph), 53.3 (C-2′), 54.7 (C-3′), 57.6 (C-5), 61.0 (CH_2O), 64.8 (C-10), 67.6 (C-4), 76.9 (C-6), 99.7 (C-8), 127.5 (CH_{Ph}), 127.9 (2× CH_{Ph}), 128.5 (2× CH_{Ph}), 138.2 (C_i), 158.2 (C=O). Anal. Calcd for $C_{18}H_{23}NO_6$: C, 61.88; H, 6.64; N, 4.01. Found: C, 61.86; H, 6.61; N, 3.95.

4.11. (5*R*,6*R*)-1-Benzyl-6-[(2′*S*,3′*S*)-3′-(trityloxymethyl)oxiran-2′-yl]-8,8-dimethyl-3,7,9-trioxa-1-azaspiro[4.5]decan-2-one 15

To a solution of **14** (49 mg, 0.14 mmol) in dry CH_2Cl_2 (0.81 mL) were successively added Et_3N (29.5 μL, 0.21 mmol), triphenylmethyl chloride (59 mg, 0.21 mmol) and DMAP (3.4 mg, 0.028 mmol). The resulting mixture was stirred for 92 h at room temperature, the solvent was evaporated, and the residue was subjected to flash chromatography on silica gel hexane/EtOAc (3:1) to give 46 mg (56%) of crystalline derivative **15**. Mp 173–175 °C, $[\alpha]_D^{25} = +75.3$ (*c* 0.25, $CHCl_3$). IR (KBr): ν_{max} 2991, 2920, 1751, 1448, 1385, 1261, 1142, 1052, 883, 764, 700. ^{1}H NMR (600 MHz, $CDCl_3$): δ 1.42 (s, 3H, CH_3), 1.54 (s, 3H, CH_3), 2.95 (dd, 1H, $J_{6,2'}$ = 5.4 Hz, $J_{3',2'}$ = 2.2 Hz, H-2′), 3.11 (ddd, 1H, $J_{3',CH2O}$ = 5.7 Hz, $J_{3',CH2O}$ = 2.6 Hz, $J''_{3,2'}$ = 2.2 Hz, H-3′) 3.16 (dd, 1H, $J_{H,H}$ = 11.1 Hz,

$J_{3',CH2O}$ = 5.7 Hz, CH_2O), 3.41 (dd, 1H, $J_{H,H}$ = 11.1 Hz, $J_{3',CH2O}$ = 2.6 Hz, CH_2O), 3.62 (d, 1H, $J_{6,2'}$ = 5.4 Hz, H-6), 3.69 (d, 1H, $J_{10,10}$ = 12.7 Hz, H-10), 3.70 (d, 1H, $J_{4,4}$ = 9.5 Hz, H-4), 3.71 (d, 1H, $J_{10,10}$ = 12.7 Hz, H-10), 3.90 (d, 1H, $J_{4,4}$ = 9.5 Hz, H-4), 4.77 (d, 1H, $J_{H,H}$ = 15.5 Hz, NCH_2Ph), 4.91 (d, 1H, $J_{H,H}$ = 15.4 Hz, NCH_2Ph), 7.22–7.26 (m, 4H, Ph), 7.27–7.33 (m, 8H, Ph), 7.42–7.47 (m, 8H, Ph). ^{13}C NMR (150 MHz, $CDCl_3$): δ 18.8 (CH_3), 28.3 (CH_3), 47.3 (NCH_2Ph), 53.0 (C-2′), 53.9 (C-3′), 57.6 (C-5), 63.6 (CH_2O), 65.2 (C-10), 67.5 (C-4), 76.7 (C-6), 86.9 (CPh_3), 99.8 (C-8), 127.1 (3× CH_{Ph}), 127.4 (CH_{Ph}), 127.9 (6× CH_{Ph}), 127.9 (2× CH_{Ph}), 128.4 (2× CH_{Ph}), 128.7 (6× CH_{Ph}), 138.3 (C_i), 143.6 (3× C_i), 157.8 (C=O). Anal. Calcd for $C_{37}H_{37}NO_6$: C, 75.11; H, 6.30; N, 2.37. Found: C, 75.17; H, 6.27; N, 2.39.

4.12. (5*R*,6*R*)-1-Benzyl-6-[(1′*S*)-1′,3′-dihydroxypropyl]-8,8-dimethyl-3,7,9-trioxa-1-azaspiro[4.5]decan-2-one 16

To a solution of **13** (0.82 g, 2.35 mmol) in dry THF (13.0 mL), which was pre-cooled to 0 °C, was added Red-Al (1.81 mL of a 65% solution in toluene). After being stirred at 0 °C for 2.5 h, the reaction mixture was partitioned between ice water (1.1 mL) and Et_2O (26 mL) and then a solid NaF (0.92 g, 21.9 mmol) was added. The resulting suspension was stirred vigorously at room temperature for 1 h and filtered through a small pad of Celite. The solvent was evaporated in vacuo, and the residue was chromatographed on silica gel hexane/EtOAc (1:5) to afford 0.51 g (62%) of compound **16** as a colourless oil. ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.42 (s, 3H, CH_3), 1.44 (s, 3H, CH_3), 1.55–1.65 (m, 1H, H-2′), 1.98–2.06 (m, 1H, H-2′), 3.67 (d, 1H, $J_{10,10}$ = 12.4 Hz, H-10), 3.67–3.82 (m, 4H, H-3′, H-6, H-4, H-10), 3.85–3.99 (m, 2H, H-3′, H-1′), 4.57 (d, 1H, $J_{4,4}$ = 8.6 Hz, H-4), 4.73 (d, 1H, $J_{H,H}$ = 15.4 Hz, NCH_2Ph), 4.90 (d, 1H, $J_{H,H}$ = 15.4 Hz, NCH_2Ph), 7.23–7.28 (m, 1H, Ph), 7.29–7.34 (m, 2H, Ph), 7.42–7.46 (m, 2H, Ph). Anal. Calcd for $C_{18}H_{25}NO_6$: C, 61.52; H, 7.17; N, 3.99. Found: C, 61.40; H, 7.26; N, 3.87.

4.13. (5*R*,6*R*)-1-Benzyl-6-[(1′*S*)-1′,3′-diacetoxypropyl]-8,8-dimethyl-3,7,9-trioxa-1-azaspiro[4.5]decan-2-one 17

Using the same procedure as described for the preparation of **8**, compound **16** (21 mg, 0.06 mmol), Ac_2O (17 μL, 0.18 mmol) and DMAP (1.2 mg, 0.01 mmol) in dry pyridine (0.5 mL) yielded after flash chromatography on silica gel hexane/EtOAc (1:1) 19 mg (74%) of derivative **17** as a colourless oil. $[\alpha]_D^{25} = +96.2$ (*c* 0.15, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$): δ 1.44 (s, 3H, CH_3), 1.51 (s, 3H, CH_3), 1.93–2.16 (m, 8H, 2× CH_3, 2× H-2′), 3.64 (d, 1H, $J_{10,10}$ = 12.8 Hz, H-10), 3.69 (m, 2H, H-4, H-10), 3.99 (d, 1H, $J_{6,1'}$ = 7.4 Hz, H-6), 4.06–4.19 (m, 2H, 2× H-3′), 4.33 (d, 1H, $J_{4,4}$ = 9.1 Hz, H-4), 4.63 (d, 1H, $J_{H,H}$ = 15.3 Hz, NCH_2Ph), 4.91 (d, 1H, $J_{H,H}$ = 15.3 Hz, NCH_2Ph), 5.07 (dt, 1H, $J_{6,1'}$ = 7.4 Hz, $J_{2',1'}$ = 7.4 Hz, $J_{2',1'}$ = 3.7 Hz, H-1′), 7.24–7.34 (m, 3H, Ph), 7.43–7.48 (m, 2H, Ph). ^{13}C NMR (100 MHz, $CDCl_3$): δ 18.8 (CH_3), 20.9 (CH_3), 20.9 (CH_3), 28.1 (CH_3), 29.8 (C-2′), 47.1 (NCH_2Ph), 57.6 (C-5), 60.3 (C-3′), 66.4 (C-10), 68.1 (C-4), 69.5 (C-1′), 76.3 (C-6), 99.8 (C-8), 127.5 (CH_{Ph}), 128.0 (2× CH_{Ph}), 128.4 (2×CH_{Ph}), 138.1 (C_i), 157.9 (C=O), 169.8 (C=O), 171.0 (C=O). Anal. Calcd for $C_{22}H_{29}NO_6$: C, 60.68; H, 6.71; N, 3.23. Found: C, 60.71; H, 6.68; N, 3.19.

4.14. (5*R*,6*R*)-1-Benzyl-6-{(1′*S*)-3′-[(*tert*-butyldimethylsilyl)oxy]-1′-hydroxypropyl}-8,8-dimethyl-3,7,9-trioxa-1-azaspiro[4.5]decan-2-one 3

To a solution of **16** (0.46 g, 1.31 mmol) in dry DMF (1.44 mL) were successively added Et_3N (0.28 mL, 1.99 mmol), *tert*-butyldimethylsilyl chloride (0.29 g, 1.92 mmol) and DMAP (0.16 g, 1.31 mmol). The resulting mixture was stirred at room temperature for 1 h, then poured into ice water (15 mL) and extracted with Et_2O (2 × 20 mL). The organic layers were combined, dried over Na_2SO_4, the solvent was evaporated, and the residue was purified by flash chromatography on silica gel hexane/EtOAc (2:1) to give 0.40 g (65%) of compound **3** as a colourless oil. $[\alpha]_D^{25} = +39.2$ (*c* 0.16, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.08 (s, 3H, CH_3), 0.09 (s, 3H, CH_3), 0.90 (s, 9H, 3× CH_3), 1.42 (s, 3H, CH_3), 1.45 (s, 3H, CH_3), 1.55–1.67 (m, 1H, H-2′), 1.87–1.96 (m, 1H, H-2′), 3.66 (d, 1H, $J_{4,4}$ = 8.4 Hz, H-4), 3.67 (d, 1H, $J_{10,10}$ = 12.7 Hz, H-10), 3.67 (d, 1H, $J_{6,1'}$ = 8.6 Hz, H-6), 3.70–3.81 (m, 2H, H-10, H-3′), 3.84–4.00 (m, 2H, H-3′, H-1′), 4.55 (d, 1H, $J_{4,4}$ = 8.4 Hz, H-4), 4.71 (d, 1H, $J_{H,H}$ = 15.4 Hz, NCH_2Ph), 4.91 (d, 1H, $J_{H,H}$ = 15.4 Hz, NCH_2Ph), 7.19–7.34 (m, 3H, Ph), 7.45–7.50 (m, 2H, Ph).^{13}C NMR (100 MHz, $CDCl_3$): δ −5.6 (CH_3), −5.5 (CH_3), 18.1 (C_q), 19.0 (CH_3), 25.9 (3× CH_3), 28.1 (CH_3), 34.5 (C-2′), 47.0 (NCH_2Ph), 58.2 (C-5), 63.1 (C-3′), 67.3 (C-10), 68.5 (C-4), 72.3 (C-1′), 77.4 (C-6), 99.3 (C-8), 127.2 (CH_{Ph}), 127.9 (2× CH_{Ph}), 128.3 (2× CH_{Ph}), 138.6 (C_i), 159.1 (C=O). Anal. Calcd for $C_{24}H_{39}NO_6Si$: C, 61.90; H, 8.44; N, 3.01. Found: C, 61.87; H, 8.41; N, 3.06.

4.15. (5*R*,6*R*)-1-Benzyl-6-{3′-[(*tert*-butyldimethylsilyl)oxy] propanoyl}-8,8-dimethyl-3,7,9-trioxa-1-azaspiro[4.5]decan-2-one 18

o-Iodoxybenzoic acid (0.35 g, 1.25 mmol) was added to a solution of **3** (0.29 g, 0.62 mmol) in CH_3CN (5.5 mL), and the resulting mixture was stirred at reflux. After the starting material was completely consumed (75 min, judged by TLC), the reaction was stopped and allowed to cool to room temperature. The solid parts were filtered off, the solvent was evaporated, and the residue was chromatographed on silica gel hexane/EtOAc (2:1) to afford 0.27 g (93%) of compound **18** as a colourless oil. $[\alpha]_D^{25} = +43.7$ (*c* 0.15, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.04 (s, 3H, CH_3), 0.05 (s, 3H, CH_3), 0.87 (s, 9H, 3× CH_3), 1.46 (s, 3H, CH_3), 1.55 (s, 3H, CH_3), 2.31 (dt, 1H, $J_{2',2'}$ = 17.5 Hz, $J_{3',2'}$ = 6.2 Hz, $J_{3',2'}$ = 6.2 Hz, H-2′), 2.77 (dt, 1H, $J_{2',2'}$ = 17.5 Hz, $J_{3',2'}$ = 6.6 Hz, $J_{3',2'}$ = 6.6 Hz, H-2′), 3.68 (d, 1H, $J_{10,10}$ = 12.6 Hz, H-10), 3.76 (d, 1H, $J_{10,10}$ = 12.6 Hz, H-10), 3.80 (d, 1H, $J_{4,4}$ = 8.8 Hz, H-4), 3.67–3.75 (m, 1H, H-3′), 3.82–3.85 (m, 1H, H-3′), 4.19 (s, 1H, H-6), 4.54 (d, 1H, $J_{4,4}$ = 8.8 Hz, H-4), 4.62 (d, 1H, $J_{H,H}$ = 15.5 Hz, NCH_2Ph), 4.75 (d, 1H, $J_{H,H}$ = 15.5 Hz, NCH_2Ph), 7.21–7.35 (m, 3H, Ph), 7.36–7.42 (m, 2H, Ph). ^{13}C NMR (100 MHz, $CDCl_3$): δ −5.4 (CH_3), −5.4 (CH_3), 18.3 (C_q), 18.9 (CH_3), 25.9 (3× CH_3), 27.7 (CH_3), 41.9 (C-2′), 46.9 (NCH_2Ph), 58.0 (C-3′), 58.5 (C-5), 67.4 (C-10), 68.1 (C-4), 78.8 (C-6), 100.4 (C-8), 127.4 (CH_{Ph}), 128.1 (2× CH_{Ph}), 128.3 (2× CH_{Ph}), 137.9 (C_i), 158.5 (C=O), 206.4 (C-1′). Anal. Calcd for $C_{24}H_{37}NO_6Si$: C, 62.17; H, 8.04; N, 3.02. Found: C, 62.21; H, 8.01; N, 3.06.

4.16. (5*R*,6*R*)-1-Benzyl-6-{(1′*R*)-3′-[(*tert*-butyldimethylsilyl) oxy]-1′-hydroxypropyl}-8,8-dimethyl-3,7,9-trioxa-1-azaspiro[4.5]-decan-2-one 19

L-Selectride (0.78 mL of a 1 M solution in THF) was added dropwise to a solution of **18** (0.18 g, 0.39 mmol) in dry THF (9.24 mL), which was pre-cooled to −78 °C, and the reaction mixture was stirred at −78 °C for 40 min. After the cautious addition of ice water (1.6 mL), the mixture was allowed to warm to room temperature and stirring was continued for another 30 min. The resulting mixture was extracted with EtOAc (2 × 10 mL), the combined organic layers were dried over Na_2SO_4, stripped of solvent, and the residue was chromatographed on silica gel hexane/EtOAc (2:1) to give 0.17 g (94%) of compound **19** as a colourless oil. $[\alpha]_D^{25} = +103.2$ (*c* 0.35, $CHCl_3$). ^{1}H NMR (400 MHz, $CDCl_3$): δ 0.08 (s, 3H, CH_3), 0.08 (s, 3H, CH_3), 0.90 (s, 9H, 3× CH_3), 1.45 (s, 3H, CH_3), 1.55 (s, 3H, CH_3), 1.68–1.75 (m, 1H, H-2′), 1.86–1.94 (m, 1H, H-2′), 2.75 (d, 1H, $J_{1',OH}$ = 5.4 Hz, OH), 3.65–3.73 (m, 3H, 2× H-10, H-4), 3.75 (d, 1H, $J_{6,1'}$ = 3.1 Hz, H-6), 3.76–3.97 (m, 2H, 2× H-3′), 4.05–4.10 (m,

1H, H-1′), 4.11 (d, 1H, $J_{4,4}$ = 9.4 Hz, H-4), 4.73 (d, 1H, $J_{H,H}$ = 15.6 Hz, NCH_2Ph), 4.86 (d, 1H, $J_{H,H}$ = 15.6 Hz, NCH_2Ph), 7.25−7.35 (m, 3H, Ph), 7.46−7.48 (m, 2H, Ph). ^{13}C NMR (100 MHz, $CDCl_3$): δ −5.5 (CH_3), −5.4 (CH_3), 18.3 (C_q), 18.8 (CH_3), 25.9 (3× CH_3), 28.4 (CH_3), 36.4 (C-2′), 47.8 (NCH_2Ph), 57.8 (C-5), 60.9 (C-3′), 66.4 (C-10), 67.4 (C-4), 68.2 (C-1′), 77.6 (C-6), 99.9 (C-8), 127.2 (CH_{Ph}), 127.9 (2× CH_{Ph}), 128.3 (2× CH_{Ph}), 138.8 (C_i), 158.3 (C=O). Anal. Calcd for $C_{24}H_{39}NO_6Si$: C, 61.90; H, 8.44; N, 3.01. Found: C, 61.92; H, 8.39; N, 2.99.

4.17. (4a*S*,8*R*,8a*R*)-5-Benzyl-8-{2′-[(*tert*-butyldimethylsilyl) oxy]-ethyl}-4a-(benzyloxymethyl)-2,2-dimethyltetrahydro-[1,3] dioxino[5,4-*d*][1,3]oxazin-6(4*H*)-one 20

At first, NaH (15 mg, 0.63 mmol of a 60% dispersion in mineral oil) was added to a solution of **19** (0.10 g, 0.21 mmol) in dry DMF (2.0 mL), which was pre-cooled to 0 °C. The resulting mixture was stirred for 1 h at 0 °C and then benzyl bromide (31 μL, 0.26 mmol) was added, and stirring continued for 10 min at 0 °C and at room temperature for another 23 h. The mixture was partitioned between ice water (10 mL) and Et_2O (10 mL), and the aqueous phase was extracted with further portions of Et_2O (2 × 10 mL). The organic layers were combined, dried over Na_2SO_4, stripped of solvent and the residue was subjected to flash chromatography on silica gel hexane/EtOAc (5:1) to give 106 mg (89%) of compound **20** as a colourless oil. $[\alpha]_D^{25} = +43.8$ (*c* 0.15, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$): δ 0.05 (s, 3H, CH_3), 0.05 (s, 3H, CH_3), 0.88 (s, 9H, 3× CH_3), 1.35 (s, 6H, 2× CH_3), 1.74–1.83 (m, 1H, H-1′), 2.03–2.12 (m, 1H, H-1′), 3.28 (d, 1H, $J_{H,H}$ = 10.1 Hz, CH_2O), 3.34 (d, 1H, $J_{H,H}$ = 10.1 Hz, CH_2O), 3.65 (d, 1H, $J_{4,4}$ = 12.4 Hz, H-4), 3.73 (d, 1H, $J_{4,4}$ = 12.4 Hz, H-4), 3.73–3.84 (m, 2H, 2× H-2′), 4.01 (d, 1H, $J_{8,8a}$ = 1.7 Hz, H-8a), 4.18 (d, 1H, $J_{H,H}$ = 12.1 Hz, OCH_2Ph), 4.27 (d, 1H, $J_{H,H}$ = 12.1 Hz, OCH_2Ph), 4.27 (d, 1H, $J_{H,H}$ = 16.2 Hz, NCH_2Ph), 4.80 (ddd, 1H, $J_{8,1'}$ = 8.3 Hz, $J_{8,1'}$ = 4.8 Hz, $J_{8,8a}$ = 1.7 Hz, H-8), 4.90 (d, 1H, $J_{H,H}$ = 16.2 Hz, NCH_2Ph), 7.14−7.35 (m, 10H, Ph). ^{13}C NMR (100 MHz, $CDCl_3$): δ −5.4 (2× CH_3), 18.3 (C_q), 21.0 (CH_3), 25.9 (3×CH_3), 26.3 (CH_3), 33.3 (C-1′), 47.0 (NCH_2Ph), 58.9 (C-2′), 60.1 (C-4a), 62.7 (C-4), 67.6 (C-8a), 71.0 (CH_2O), 72.1 (C-8), 73.4 (OCH_2Ph), 99.9 (C-2), 127.0 (CH_{Ph}), 127.1 (2× CH_{Ph}), 127.7 (2×CH_{Ph}), 128.0 (CH_{Ph}), 128.3 (2× CH_{Ph}), 128.5 (2× CH_{Ph}), 137.0 (C_i), 138.4 (C_i), 154.8 (C=O). Anal. Calcd for $C_{31}H_{45}NO_6Si$: C, 66.99; H, 8.16; N, 2.52. Found: C, 67.03; H, 8.12; N, 2.49.

4.18. (4a*S*,8*R*,8a*R*)-5-Benzyl-8-(2′-hydroxyethyl)-4a-(hydroxymethyl)-2,2-dimethyltetrahydro-[1,3]dioxino[5,4-*d*] [1,3]oxazin-6(4*H*)-one 21

To a solution of **20** (56 mg, 0.10 mmol) in dry EtOH (2.0 mL) was added $Pd(OH)_2/C$ (28 mg, 20% by weight) in one portion and the resulting mixture was stirred under H_2 (10 atm) at room temperature. After 15 h, no starting material was detected (TLC) in the mixture, which was then filtered through a small pad of silica gel. Evaporation of the solvent afforded 35 mg (99%) of compound **21** as a colourless oil, which was used immediately in the next step without further purification. ^1H NMR (400 MHz, $CDCl_3$): δ 1.36 (s, 3H, CH_3), 1.39 (s, 3H, CH_3), 1.78–1.88 (m, 1H, H-1′), 2.10–2.17 (m, 1H, H-1′), 2.56 (m, 1H, OH), 3.51 (d, 1H, $J_{H,H}$ = 12.4 Hz, CH_2O), 3.58 (d, 1H, $J_{H,H}$ = 12.4 Hz, CH_2O), 3.62 (d, 1H, $J_{4,4}$ = 12.6 Hz, H-4), 3.76 (d, 1H, $J_{4,4}$ = 12.6 Hz, H-4), 3.76–3.84 (m, 1H, H-2′), 3.85–3.93 (m, 1H, H-2′), 4.05 (m, 1H, H-8a), 4.31 (d, 1H, $J_{H,H}$ = 16.3 Hz, NCH_2Ph), 4.86–4.93 (m, 1H, H-8), 4.94 (d, 1H, $J_{H,H}$ = 16.3 Hz, NCH_2Ph), 7.25−7.38 (m, 5H, Ph). Anal. Calcd for $C_{18}H_{25}NO_6$: C, 61.52; H, 7.17; N, 3.99. Found: C, 61.43; H, 7.08; N, 4.05.

4.19. (4a*S*,8*R*,8a*R*)-5-Benzyl-8-{2′-[(*tert*-butyldimethylsilyl) oxy]-ethyl}-4a-(hydroxymethyl)-2,2-dimethyltetrahydro-[1,3] dioxino[5,4-*d*][1,3]oxazin-6(4*H*)-one 22

To a solution of **21** (35 mg, 99.6 μmol) in dry CH_2Cl_2 (1.26 mL) that was pre-cooled to 0 °C, were successively added imidazole (20 mg, 0.29 mmol) and *tert*-butyldimethylsilyl chloride (20 mg, 0.13 mmol). After being stirred at 0 °C for 20 min, the solvent was evaporated and the residue was chromatographed on silica gel hexane/EtOAc (2:1) to yield 35 mg (76%) of compound **22** as a colourless oil. $[\alpha]_D^{25} = +44.8$ (*c* 0.66, $CHCl_3$). ^1H NMR (400 MHz, $CDCl_3$): δ 0.06 (s, 3H, CH_3), 0.07 (s, 3H, CH_3), 0.89 (s, 9H, 3× CH_3), 1.37 (s, 3H, CH_3), 1.39 (s, 3H, CH_3), 1.77–1.85 (m, 1H, H-1′), 2.05–2.13 (m, 1H, H-1′), 3.53 (m, 2H, CH_2OH), 3.66 (d, 1H, $J_{4,4}$ = 12.3 Hz, H-4), 3.71–3.77 (m, 1H, H-2′), 3.78 (d, 1H, $J_{4,4}$ = 12.3 Hz, H-4), 3.80–3.85 (m, 1H, H-2′), 4.03 (d, 1H, $J_{8,8a}$ = 1.5 Hz, H-8a), 4.11 (d, 1H, $J_{H,H}$ = 16.2 Hz, NCH_2Ph), 4.80 (ddd, 1H, $J_{8,1'}$ = 6.8 Hz, $J_{8,1'}$ = 5.0 Hz, $J_{8,8a}$ = 1.5 Hz, H-8), 5.12 (d, 1H, $J_{H,H}$ = 16.2 Hz, NCH_2Ph), 7.23−7.35 (m, 5H, Ph). ^{13}C NMR (100 MHz, $CDCl_3$): δ −5.4 (CH_3), −5.4 (CH_3), 18.2 (C_q), 21.3 (CH_3), 25.9 (4× CH_3), 33.0 (C-1′), 46.7 (NCH_2Ph), 58.8 (C-2′), 61.4 (C-4a), 62.6 (C-4), 64.5 (CH_2OH), 68.0 (C-8a), 72.4 (C-8), 100.1 (C-2), 126.9 (2× CH_{Ph}), 127.5 (CH_{Ph}), 128.8 (2× CH_{Ph}), 138.7 (C_i), 155.3 (C=O). Anal. Calcd for $C_{24}H_{39}NO_6Si$: C, 61.90; H, 8.44; N, 3.01. Found: C, 61.93; H, 8.39; N, 3.04.

4.20. (4a*S*,8*R*,8a*R*)-5-Benzyl-8-{2′-[(*tert*-butyldimethylsilyl) oxy]-ethyl}-2,2-dimethyl-6-oxohexahydro-[1,3]dioxino[5,4-*d*] [1,3]-oxazin-4a-carboxylic acid 2

Using the same procedure as described for the preparation of **18**, compound **22** (35 mg, 75.1 μmol), and *o*-iodoxybenzoic acid (32 mg, 0.11 mmol) in CH_3CN (1.0 mL) afforded, after 30 min, the crude aldehyde (35 mg), which was used immediately in the next reaction without further purification. A solution of $NaClO_2$ (65 mg, 0.72 mmol) and $NaH_2PO_4.2H_2O$ (81 mg, 0.52 mmol) in water (0.36 mL) was added to the solution of aldehyde (35 mg, 75.1 μmol) in a mixture of 4:4:1 CH_3CN/*t*-butanol/2-methylbut-2-ene (1.62 mL) at 0 °C. The resulting mixture was stirred at 0 °C for 1 h, after which the solvent was removed under reduced pressure, and the residue was chromatographed through a short silica gel column CH_2Cl_2/CH_3OH (9:1) to give 26 mg (72%) of compound **2** as a colourless oil. $[\alpha]_D^{25} = +24.3$ (*c* 0.06, CH_3OH). ^1H NMR (400 MHz, CD_3OD): δ 0.03 (s, 3H, CH_3), 0.04 (s, 3H, CH_3), 0.86 (s, 9H, 3× CH_3), 1.30 (s, 3H, CH_3), 1.44 (s, 3H, CH_3), 1.73–1.82 (m, 1H, H-1′), 1.91–2.01 (m, 1H, H-1′), 3.62–3.81 (m, 2H, 2× H-2′), 3.91 (d, 1H, $J_{4,4}$ = 13.2 Hz, H-4), 4.26 (d, 1H, $J_{H,H}$ = 16.8 Hz, NCH_2Ph), 4.34–4.37 (m, 2H, H-8a, H-4), 4.59–4.64 (m, 1H, H-8), 4.91 (d, 1H, $J_{H,H}$ = 16.8 Hz, NCH_2Ph), 7.11−7.15 (m, 1H, Ph), 7.18−7.22 (m, 2H, Ph), 7.32−7.34 (m, 2H, Ph). ^{13}C NMR (100 MHz, CD_3OD): δ −5.3 (CH_3), −5.2 (CH_3) 19.2 (C_q), 19.5 (CH_3), 26.5 (3× CH_3), 28.7 (CH_3), 34.0 (C-1′), 51.2 (NCH_2Ph), 59.6 (C-2′), 63.0 (C-4), 66.1 (C-4a), 67.5 (C-8a), 73.9 (C-8), 100.4 (C-2), 127.8 (CH_{Ph}), 128.4 (2× CH_{Ph}), 128.9 (2× CH_{Ph}), 139.1 (C_i), 157.0 (C=O), 173.0 (COOH). Anal. Calcd for $C_{24}H_{37}NO_7Si$: C, 60.10; H, 7.78; N, 2.92. Found: C, 60.07; H, 7.73; N, 2.97.

4.21. Methyl (4a*S*,8*R*,8a*R*)-5-benzyl-8-{2′-[(*tert*-butyldimethylsilyl)oxy]ethyl}-2,2-dimethyl-6-oxohexahydro-[1,3]dioxino-[5,4-*d*][1,3]oxazin-4a-carboxylate 23

To a solution of **2** (21 mg, 43.8 μmol) in dry DMF (0.16 mL) were successively added K_2CO_3 (6 mg, 43.4 μmol) and methyl iodide (4.1 μL, 65.9 μmol). After being stirred at room temperature for

45 min, the solvent was evaporated in vacuo, and the residue was purified by flash chromatography on silica gel hexane/EtOAc (3:1) to afford 14 mg (65%) of compound **23** as a colourless oil. $[\alpha]_D^{25} = +70.7$ (*c* 0.20, $CHCl_3$). ^1H NMR ($CDCl_3$, 400 MHz): δ 0.05 (s, 3H, CH_3), 0.06 (s, 3H, CH_3), 0.88 (s, 9H, 3× CH_3), 1.38 (s, 3H, CH_3), 1.44 (s, 3H, CH_3), 1.74–1.83 (m, 1H, H-1′), 2.02–2.10 (m, 1H, H-1′), 3.59 (s, 3H, OCH_3), 3.70 (dt, 1H, $J_{2',2'}$ = 10.4 Hz, $J_{2',1'}$ = 5.3 Hz, $J_{2',1'}$ = 5.3 Hz, H-2′), 3.81 (ddd, 1H, $J_{2',2'}$ = 10.4 Hz, $J_{2',1'}$ = 8.5 Hz, $J_{2',1'}$ = 4.3 Hz, H-2′), 3.92 (d, 1H, $J_{4,4}$ = 12.8 Hz, H-4), 4.14 (d, 1H, $J_{8,8a}$ = 1.6 Hz, H-8a), 4.35 (d, 1H, $J_{4,4}$ = 12.8 Hz, H-4), 4.51 (d, 1H, $J_{H,H}$ = 16.4 Hz, NCH_2Ph), 4.58−4.62 (m, 1H, H-8), 4.78 (d, 1H, $J_{H,H}$ = 16.4 Hz, NCH_2Ph), 7.19−7.24 (m, 1H, Ph), 7.26−7.30 (m, 2H, Ph), 7.33−7.35 (m, 2H, Ph). ^{13}C NMR (100 MHz, $CDCl_3$): δ −5.5 (2× CH_3), 18.2 (C_q), 19.7 (CH_3), 25.8 (3× CH_3), 27.4 (CH_3), 32.9 (C-1′), 49.3 (NCH_2Ph), 53.1 (OCH_3), 58.4 (C-2′), 62.0 (C-4), 64.1 (C-4a), 66.4 (C-8a), 72.2 (C-8), 99.7 (C-2), 127.1 (CH_{Ph}), 127.7 (2× CH_{Ph}), 128.1 (2× CH_{Ph}), 137.0 (C_i), 153.9 (C=O), 170.4 (C=O). Anal. Calcd for $C_{25}H_{39}NO_7Si$: C, 60.82; H, 7.96; N, 2.84. Found: C, 60.79; H, 8.01; N, 2.89.

4.22. X-ray techniques

Single crystals of **15** suitable for X-ray diffraction were obtained from Et_2O by slow evaporation at room temperature. The intensities were collected at 100 K on an Oxford Diffraction Gemini R CCD diffractometer using Mo Kα radiation (λ = 0.71073 Å). Selected crystallographic and other relevant data for the compound **15** are listed in Table 1. The structure was solved by direct methods.[20] All non-hydrogen atoms were refined anisotropically by full-matrix least squares calculations based on F^2.[20] All hydrogen atoms were included in calculated positions as riding atoms, with SHELXL97[20] defaults. The PLATON[21] programme was used for structure analysis and molecular and crystal structure drawings.

Table 1
Crystal data and structure refinement parameters for compound **15**

15	
Empirical formula	$C_{37}H_{37}NO_6$
Formula weight	591.69
Temperature, *T* (K)	100(2)
Wavelength, λ (Å)	0.71073
Crystal system	Orthorhombic
Space group	$P2_12_12_1$
Unit cell dimensions	
a (Å)	9.4578 (3)
b (Å)	10.1086 (3)
c (Å)	32.3811(13)
V (Å^3)	3095.80 (18)
Formula per unit cell, *Z*	4
D_{calcd} (g/cm^3)	1.269
Absorption coefficient, μ (mm^{-1})	0.086
F(000)	1256
Crystal size (mm)	0.649 × 0.1117 × 0.0451
θ Range for data collection (°)	3.50–26.50
Index ranges	$-11 \leqslant h \leqslant 11$
	$-12 \leqslant k \leqslant 12$
	$-40 \leqslant l \leqslant 40$
Independent reflections (*R*int)	6390 (0.1298)
Absorption correction	Analytical
Max. and min. transmission	0.996 and 0.967
Refinement method	Full-matrix least-squares on F^2
Data/restraints/parameters	6390/0/399
Goodness-of-fit on F^2	0.761
Final *R* indices [$I>2\sigma(I)$]	R_1 = 0.0347, wR_2 = 0.0528
R indices (all data)	R_1 = 0.0823, wR_2 = 0.0579
Largest diff. peak and hole (e/Å^{-3})	0.150 and −0.161

4.23. Crystallographic data

Complete crystallographic data for the structural analysis have been deposited with the Cambridge Crystallographic Data Centre, CCDC No. 863786. These data can be obtained free of charge from the Director, Cambridge Crystallographic Data Centre, 12 Union Road, Cambridge, CB2 1EZ, UK (fax: +44 1223 336033; e-mail: deposit@ccdc.cam.ac.uk or via: www.ccdc.cam.ac.uk).

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