A Straightforward, Highly Stereoselective Synthesis of Protected Isostatine Derivatives

Patricia Castejón, Albert Moyano,* Miquel A. Pericàs* and Antoni Riera

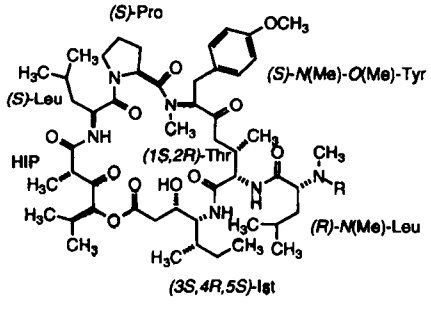
Abstract: A novel strategy for the synthesis of isostatine derivatives has been developed. Contrary to previous approaches to isostatine, a non-proteinogenic amino acid that is an essential component of the didemnins, the present synthesis does not require the intermediacy of the expensive amino acid D-*allo*-isoleucine, the starting material being commercially available enantiopure *(S)*-2-methyl-1-butanol. Steps in the sequence include catalytic asymmetric epoxidation, regioselective titanium-promoted opening of an epoxy alcohol with an ammonia equivalent, stereospecific generation of an *N*-Boc-amino epoxide and its nucleophilic opening by a cyanide anion. Application of this method has permitted the enantioselective preparation of isostatine methyl ester and, for the first time, of isostatine amide, both in fully protected form.

Keywords
aminopolyols · asymmetric aminations · asymmetric epoxidation · asymmetric hydroxylation · didemnins

Introduction

The didemnins (Fig. 1) constitute a class of macrocyclic peptides of marine origin isolated for the first time in 1981 by Rinehart et al.[1] from the tunicate *Trididendrum solidum* during a systematic study of Caribbean marine natural products.

From the structural point of view, all of the didemnins have a common cyclodepsipeptide core, consisting of four codified amino acids ((1*S*,2*R*)-threonine, (*S*)-proline, (*S*)-leucine and (*S*)-*N*-methyl-*O*-methyltyrosine), (2*S*,4*S*)-2,5-dimethyl-4-hydroxy-3-oxohexanoic acid (HIP) and the previously unknown β-hydroxy-γ-amino acid (3*S*,4*R*,5*S*)-4-amino-3-hydroxy-5-methylheptanoic acid or isostatine (Ist; **1**, Fig. 2). The didemnins differ among themselves in the structure of the side chain of the threonine amino group which, in turn, strongly influences the biological properties of these compounds. In fact, although didemnins A and B both possess antiviral and cytotoxic activity, didemnin B is itself a potent immunosuppressive agent in vivo and in vitro,[2] with specific activity against several pathogenic viruses,[3] and was the first marine natural product to enter clinical trials (phases I and II) as a carcinostatic agent.[4] The discovery that the new amino acid isostatine (**1**) and not (3*S*,4*R*)-statine (**2**, Fig. 2) was present in the macrocyclic core of the didemnins[4a, 5] and that it was essential to the bioactivity of didemnin A[5a] spurred the development of synthetic approaches to this compound, the supply of which from renewable sources is clearly insufficient and which has not yet been found in any other class of natural products. Isostatine is structurally related to dolaisoleuine (**3**, Fig. 2), a *N*,*O*-dimethylated diastereomer of **1** that is a component of dolastatin 10, an anti-



didemnin A	R = H
didemnin B	R = *(S)*-Lac-*(S)*-Pro
didemnin C	R = *(S)*-Lac
didemnin D	R = *(S)*-pGlu-(*(S)*-Gln)$_3$-*(S)*-Lac-*(S)*-Pro
didemnin E	R = *(S)*-pGlu-(*(S)*-Gln)$_2$-*(S)*-Lac-*(S)*-Pro
didemnin G	R = CHO
didemnin H	R = *(S)*-pGlu-*(S)*-Gln
didemnin X	R = $nC_7H_{15}CH(OH)$-CH_2CO-(*(S)*-pGlu)$_3$-*(S)*-Lac-*(S)*-Pro
didemnin Y	R = $nC_7H_{15}CH(OH)$-CH_2CO-(*(S)*-pGlu)$_4$-*(S)*-Lac-*(S)*-Pro
didemnin DDB	R = Pyruv-*(S)*-Pro

Fig. 1. The didemnins.

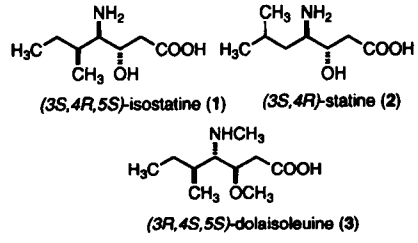


Fig. 2. Absolute configurations of isostatine (**1**), (3*S*,4*R*)-statine (**2**) and dolaisoleuine (**3**).

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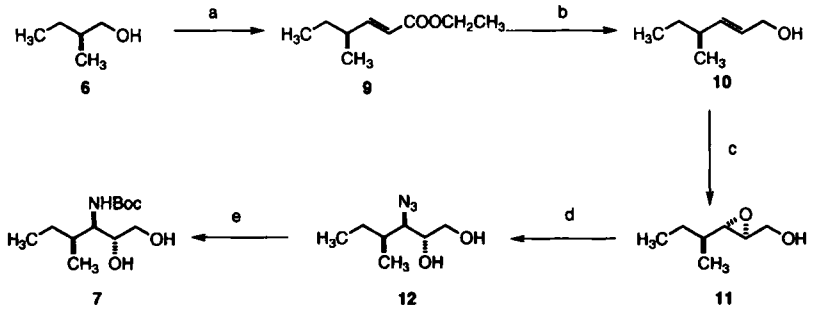
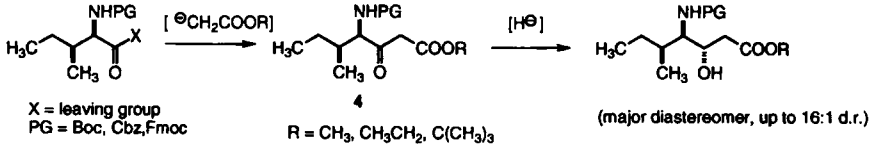
neoplastic pseudopentapeptide isolated in 1984 by Pettit and co-workers[6] from the Indian Ocean sea hare *Dolabella auricularia*.

Only two distinct strategies have been devised up to now to access isostatine or its derivatives:

i) Stereoselective reduction of β-keto esters of general structure **4** obtained through the reaction of properly activated D-*allo*-isoleucine derivatives with ester enolates (Scheme 1).[7] The best reducing agents for this approach are borohydrides ($NaBH_4$, KBH_4, $NaBH_3CN$), affording the major (3*S*,4*R*,5*S*) isomer in variable diastereomeric ratios.

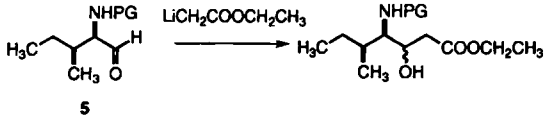


Scheme 4. Reaction conditions and yields: a) aq. NaClO, TEMPO (cat.), KBr (cat.), CH_2Cl_2; $Ph_3PCHCOOCH_2CH_3$; distillation (71%); b) DIBAL-H (2.5 equiv), diethyl ether, −78 °C (96%); c) *tert*-butyl hydroperoxide, CH_2Cl_2, L-(+)-DIPT (cat.), $Ti(OiPr)_4$ (cat.), −20 °C, 5 h (82%, 94:6 diastereomer ratio); d) $Ti(OiPr)_2(N_3)_2$, benzene, 75 °C (100%); e) H_2 (1 atm), 10% Pd/C (cat.), $(Boc)_2O$, ethyl acetate (92%).



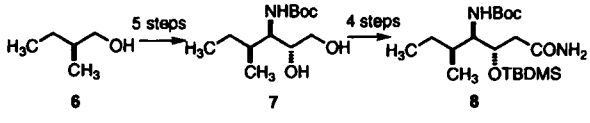
Scheme 1. Synthesis of isostatine by stereoselective reduction of D-*allo*-isoleucine-derived β-keto esters.

ii) Aldol condensation of an *N*-protected D-*allo*-isoleucinal **5** with the lithium enolate of ethyl acetate, which leads to a 1:1.2 mixture of *anti* and *syn* diastereomers (Scheme 2).[5a, 8] Both methods rely on the use as starting material of D-*allo*-isoleucine, an amino acid which is commercially available but expensive (its price being between 60 and 90 times that of L-isoleucine).[9]



Scheme 2. Synthesis of isostatine by aldol condensation of D-*allo*-isoleucinal.

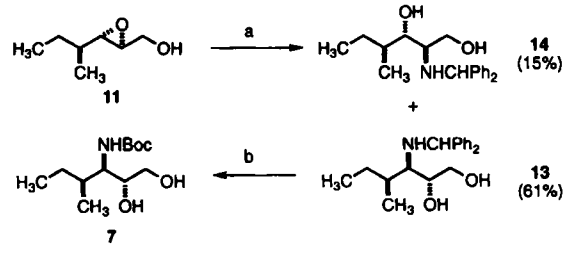
We disclose in this paper a novel, non-amino-acid-based strategy for the preparation of isostatine derivatives, exemplified by the first synthesis of a fully protected, stereochemically pure (3*S*,4*R*,5*S*)-isostatine amide (**8**) from the inexpensive, commercially available (*S*)-2-methyl-1-butanol **6** (Scheme 3). This approach relies upon the ready accessibility of the aminodiol **7**, which should be easily available through a convenient three-step route to highly enantiopure *anti*-*N*-protected-3-amino-1,2-diols from simple allylic alcohols developed in our laboratories.[11] We have also demonstrated that these compounds are exceedingly versatile chiral synthons for the diastereo- and enantioselective preparation of several types of amino acids (α-amino acids,[12a, b] β-amino acids,[12c] α-hydroxy-β-amino acids[12d, e] and β-hydroxy-γ-amino acids[13]) as well as of other pharmaceutically interesting compounds.[14]



Scheme 3. Non-amino-acid-based approach to isostatine amide.

Results and Discussion

The first objective in our proposed strategy was the preparation of the key *N*-Boc-aminodiol intermediate **7** (Scheme 4). To that effect, commercial (*S*)-2-methyl-1-butanol **6** (whose enantiomeric purity was found to be greater than 98%, according to ^1H NMR analyses of the two diastereomeric Mosher esters)[15] was converted to the known ethyl ester **9**[16] in 71% yield by means of TEMPO-catalyzed oxidation to the corresponding aldehyde[17] followed by in situ Wittig reaction with ethyl triphenylphosphoranylacetate. Subsequent low-temperature, DIBAL-H mediated reduction of **9** gave almost quantitatively the (*E*) allyl alcohol **10**,[18] which upon catalytic Sharpless epoxidation[19] with (+)-diisopropyl tartrate as the chiral ligand produced the expected epoxy alcohol **11** with 94% diastereomeric purity. Exposure of **11** to titanium diazidodiisopropoxyde in hot benzene[20] gave the azidodiol **12** with complete regioselectivity and quantitative yield. Finally, catalytic hydrogenation of **12** in ethyl acetate in the presence of an excess of di-*tert*-butyl dicarbonate led directly to the *N*-Boc aminodiol **7** in 92% yield. This important intermediate can thus be obtained from optically pure **6** in five steps and in at least 51% overall yield. Relevant to the scale-up possibilities of the sequence is the fact that **7** could also be prepared through nucleophilic opening of the epoxy alcohol **11** by diphenylmethylamine,[11] a much safer ammonia equivalent than the azide ion (Scheme 5). Thus, the reaction of **11** with a 100% excess of diphenylmethylamine in refluxing 1,2-dichloroethane and in the presence of titanium tetraisopropoxide gave a 4:1 mixture of the regioisomeric benzhydrylaminodiols **13** and **14**, from which the major one, arising from C_3 attack of the amine, could be isolated in 61% yield (based on reacted **11**) after column chromatography of the crude products. As described above for the azide



Scheme 5. Reaction conditions and yields: a) Ph_2CHNH_2 (2 equiv), $Ti(OiPr)_4$ (3 equiv), 1,2-dichloroethane, reflux, 100 h; chromatographic separation (61% **13**, 15% **14**); b) H_2 (1 atm), 20% $Pd(OH)_2/C$ (cat.), $(Boc)_2O$, ethyl acetate (80%).

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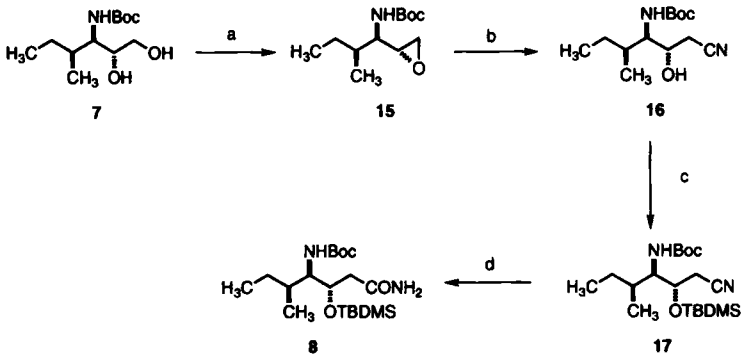
12, one-pot hydrogenolysis–reprotection afforded the *N*-Boc derivative 7 in 80% yield.

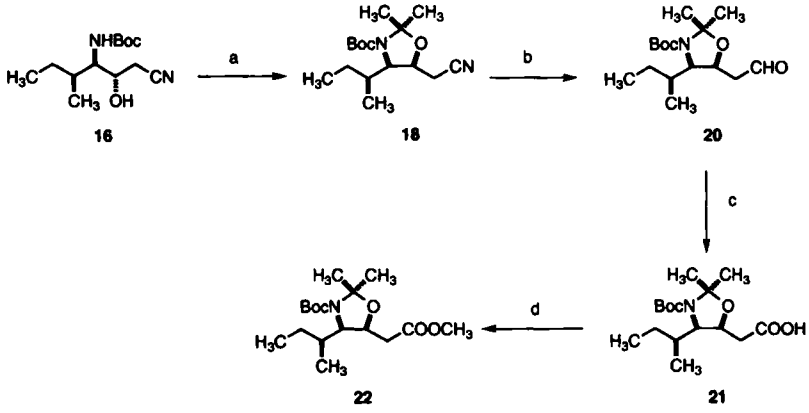
The conversion of 7 into a protected isostatine amide (Scheme 6) commenced with the preparation of the aminoalkyl epoxide 15, which was obtained from 7 in 72% yield as a



Scheme 6. Reaction conditions and yields: a) PPh_3, diethyl azodicarboxylate, $CHCl_3$, reflux (72%); b) acetone cyanohydrin, triethylamine, THF, reflux (68%); c) *tert*-butyldimethylsilyl chloride (3 equiv), imidazole (6 equiv), DMF, RT (87%); d) 1 M NaOH, aq. H_2O_2, aq. *tert*-butyl alcohol, RT (68%).

diastereomerically pure, highly crystalline compound after treatment with triphenylphosphine/diethyl azodicarboxylate in refluxing chloroform, according to the procedure recently reported by us.[21] Next, regioselective epoxide opening effected by acetone cyanohydrin in a basic medium[22] gave the hydroxynitrile 16, arising from the exclusive attack of cyanide anion at the less substituted carbon of the oxirane ring. Protection of the hydroxyl as a *tert*-butyldimethylsilyl ether[23] and hydrogen peroxide-promoted nitrile hydrolysis[24] completed the first totally stereoselective synthesis of an isostatine derivative based on a non-amino-acid starting material.

The versatility of our approach could be also conveniently demonstrated by means of the conversion of the intermediate hydroxynitrile 16 into the known isostatine derivative 22 (Scheme 7). The necessary transformations were patterned upon the lines of our protocol for the enantioselective synthesis of *N*-Boc-*N,O*-oxazolidine-protected *anti*-β-hydroxy-γ-amino acids.[13] Thus, treatment of 16 with 2-methoxypropene under the conditions described by Joullié and co-workers for a related transformation[7a] led to the formation of oxazolidine 18 in 81% yield (based on a 68% conversion of the starting



Scheme 7. Reaction conditions and yields: a) 2-methoxypropene, *p*-toluenesulfonic acid (cat.), DMF, RT (81%); b) DIBAL-H (1.8 equiv), diethyl ether, −40 to −20 °C, 30 min (100%); c) $KMnO_4$, 1 M NaH_2PO_4, *tert*-butyl alcohol, RT (94%); d) excess diazomethane, diethyl ether (95%).

product).[25] The DIBAL-H mediated reduction of the nitrile group gave a quantitative yield, and the resulting aldehyde 20 was directly oxidized to the acid 21 by the procedure of Masamune et al.[26] (94% yield); this acid offers interesting possibilities with regard to the construction of the didemnin core. Finally exposure of 21 to an excess of ethereal diazomethane led to the ester 22,[7a] whose spectroscopic data coincided fully with those of an authentic sample.

Conclusion

We have described concise, highly stereoselective approaches to fully protected isostatine amide 8 and methyl ester 22, which stand out as the first asymmetric syntheses of isostatine derivatives which do not proceed via the uncommon α-amino acid D-*allo*-isoleucine. The key steps of the sequences, which exemplify a general method for the EPC-synthesis of β-hydroxy-γ-amino acids,[13] involve catalytic Sharpless epoxidation[19] of an allylic alcohol, regioselective epoxy-alcohol opening with an ammonia surrogate,[11, 20] stereospecific conversion of the resulting aminodiol into an *N*-Boc-aminoalkyl epoxide[21] and nucleophilic attack at the less substituted oxirane carbon by a cyanide anion. It is also worth noting that the present method is in principle suitable for the preparation of dolaisoleuine 3, by simply changing the enantiomeric series of the tartrate ester employed in the catalytic Sharpless epoxidation step.

Experimental Procedure

Melting points were determined in open-ended capillary tubes on a Büchi–Tottoli apparatus or a Reichert Thermovar Köfler apparatus and are uncorrected. Infrared spectra were measured with a Perkin–Elmer 681 or a Nicolet FT-IR 510 spectrometer by film NaCl or KBr pellet techniques. ^1H and ^{13}C NMR spectra were recorded on a Varian Gemini-200 or Unity-300 spectrometer with tetramethylsilane or chloroform as an internal standard. The multiplicity in ^{13}C NMR spectra was determined by DEPT techniques. Mass spectra were recorded at 70 eV ionizing voltage on a Hewlett–Packard 5890 apparatus. Ammonia was used for chemical ionization (CI). Optical rotations were measured with a Perkin–Elmer 241 MC automatic polarimeter. Elemental analyses were performed by the "Servei d'Anàlisis Elementals del CSIC de Barcelona". THF and diethyl ether used in the reactions were dried by distillation over metallic sodium and benzophenone; dichloromethane, chloroform, 1,2-dichloroethane and DMF were distilled over calcium hydride, and benzene over metallic sodium. All reactions were carried out in oven or flame-dried glassware under an atmosphere of prepurified nitrogen. The course of all of the reactions described could be conveniently monitored by TLC (aluminium plates precoated with silica gel 60 F_{254}, Merck). Silica gel (J. T. Baker, 70–230 mesh) was used for column chromatography.

(4*S*,*E*)-4-Methyl-2-hexenoic acid ethyl ester (9): A dichloromethane solution of (*S*)-2-methylbutanal was prepared according to the procedure described in ref. [17] with the following reagents and quantities: (*S*)-2-methyl-1-butanol (6, 23.38 g, 0.265 mol); anhydrous dichloromethane (115 mL); 2,2,6,6-tetramethylpiperidine-1-oxyl (TEMPO, 0.41 g, 2.65 mmol); aqueous KBr (3.16 g in 14 mL, 26.5 mmol); aqueous NaClO (0.74 M, 394 mL, 0.292 mol). The resulting dichloromethane solution (273 g, approximately 1.3 M with respect to (*S*)-2-methylbutanal as estimated by ^1H NMR) was added to a stirred solution of ethyl triphenylphosphoranylacetate (120 g, 0.346 mol), and the resulting mixture was heated under reflux for 8 h and stirred at room temperature for 12 h. After elimination of the solvent under reduced pressure, hexane (25 mL) was added and the precipitated triphenylphosphine oxide was filtered off and thoroughly washed with dichloromethane. Rotavaporation of the solvents and distillation of the crude oil (82–83 °C, 15 mm Hg) gave 29.45 g (71% overall yield from 6) of the stereochemically pure ester 9 as a colourless oil. $[\alpha]_D^{23} = +33.7°$ ($c = 2.88$ in $CHCl_3$) (ref. [16]: $[\alpha]_D^{20} = +5$ ($c = 0.8$ in $CHCl_3$) [27]); ^1H NMR (200 MHz, $CDCl_3$, 25 °C,

TMS): $\delta = 0.88$ (t, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.05 (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.29 (t, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.41 (m, 2H; CH_2), 2.21 (m, 1H; CH), 4.19 (q, $^3J(H,H) = 7$ Hz, 2H; CH_2), 5.78 (d, $^3J(H,H) = 16$ Hz, 1H; CH), 6.88 (dd, $^3J(H,H) = 16$ Hz, $^3J'(H,H) = 8$ Hz, 1H; CH); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 11.3$ (CH_3), 14.0 (CH_3), 18.7 (CH_3), 28.6 (CH_2), 37.9 (CH), 59.8 (CH_2), 119.5 (CH), 154.0 (CH), 166.5 (Cq); IR (NaCl film): $\tilde{\nu} = 2980$, 2940, 2880, 1725, 1650, 1450, 1360, 985 cm^{-1}; MS (CI): *m/z* (%): 191 (45) $[M+35]^+$, 174 (100) $[M+18]^+$, 157 (7) $[M+1]^+$.

(4*S*,*E*)-4-Methyl-2-hexen-1-ol (10) [18]: To a cold (−78 °C) solution of ethyl ester **9** (0.20 g, 1.28 mmol) in anhydrous diethyl ether (19 mL) a 20% solution of DIBAL-H in hexanes (3.2 mL, 3.2 mmol) was added slowly. After stirring at −78 °C for 1.5 h, methanol (3 mL) was added, and the resulting mixture was allowed to reach room temperature, during which time (15 to 30 min) a precipitate of aluminium oxide was formed. This precipitate was filtered off and washed with dichloromethane. The combined organic solutions were washed with brine and dried over anhydrous magnesium sulfate. Evaporation of the solvents under reduced pressure produced 0.28 g of a crude oil, which was purified by column chromatography on triethylamine-pretreated silica gel (2.5% v/v) with hexane/ethyl acetate mixtures of increasing polarity as eluents to afford 0.14 g (96% yield) of a colourless oil, the (*E*)-allyl alcohol **10**. $[\alpha]_D^{23} = +34.3$ ($c = 2.40$ in $CHCl_3$); ^1H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 0.86$ (t, $^3J(H,H) = 7.5$ Hz, 3H; CH_3), 0.98 (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.32 (m, 2H; CH_2), 1.70 (brs, 1H; OH), 2.05 (m, 1H; CH), 4.09 (d, $^3J(H,H) = 4.4$ Hz, 2H; CH_2), 5.58 (m, 2H; 2 olefinic CH); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 11.5$ (CH_3), 19.7 (CH_3), 29.3 (CH_2), 37.8 (CH), 63.3 (CH_2), 127.2 (CH), 138.3 (CH); IR (NaCl film): $\tilde{\nu} = 3320$ (br), 2960, 2920, 2880, 1660, 1450, 1370, 1070, 100, 960 cm^{-1}; MS (70 eV, EI): *m/z* (%): 113 (32) $[M^+ - 1]$, 97 (54) $[M^+ - 18]$, 83 (100) $[C_6H_{11}]^+$.

(2*S*,3*S*,4*S*)-4-Methyl-2,3-epoxyhexan-1-ol (11) [18, 19a]: To a cold (−20 °C), stirred mixture of 4 Å molecular sieves (powder, 0.65 g, dried before use at 350 °C/1 mm Hg over 6 h), L-(+)-diisopropyl tartrate (0.28 g, 1.21 mmol) and anhydrous dichloromethane (7.1 mL) was added titanium tetraisopropoxide (0.24 mL, 0.81 mmol) through a cannula, followed immediately by a solution of allyl alcohol **10** (1.85 g, 16.2 mmol) in anhydrous dichloromethane (7.1 mL), and after 30 min by a 2.78 M isooctane solution of *tert*-butyl hydroperoxide (11.6 mL, 32.4 mmol) diluted with anhydrous dichloromethane (7.1 mL). The mixture was stirred at −20 °C for 5 h, after which time no starting product could be detected by TLC. A cooled (0 °C) aqueous solution (45 mL) of $FeSO_4 \cdot 7H_2O$ (19.4 mmol) and tartaric acid (1.46 g, 9.73 mmol) was subsequently added, and the temperature was slowly raised to 0 °C. The phases were separated and the aqueous one was extracted twice with dichloromethane. The combined organic phases were cooled to 0 °C and were treated with 4.5 mL of a cool, 30% aqueous NaOH solution saturated with NaCl. The mixture was vigorously stirred for 2 h at the same temperature, transferred to a separating funnel and diluted with water (15 mL). The aqueous layer was extracted twice with diethyl ether, and the combined organic phases were subsequently dried over sodium sulfate. Elimination of the solvents under reduced pressure yielded 2.46 g of a crude oil, which was purified by column chromatography on triethylamine-pretreated silica gel (2.5% v/v) with hexane/ethyl acetate mixtures of increasing polarity as eluents, to afford 1.73 g (82% yield) of a colourless oil, the epoxy alcohol **11**, with a diastereomeric excess of 88% [28]. $[\alpha]_D^{23} = -25.5$ ($c = 2.37$ in $CHCl_3$); ^1H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 0.95$ (t, $^3J(H,H) = 7.5$ Hz, 3H; CH_3), 0.98 (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.33 (m, 2H; CH_2), 1.58 (m, 1H; CH), 2.30 (brs, 1H; OH), 2.78 (dd, $^3J(H,H) = 7$ Hz, $^3J'(H,H) = 2.4$ Hz, 1H; CH), 2.95 (m, 1H; CH), 3.60 (m, 1H; CHH'OH), 3.90 (m, 1H; CHH'OH); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 11.0$ (CH_3), 15.1 (CH_3), 26.9 (CH_2), 36.6 (CH), 57.2 (CH), 60.4 (CH), 61.9 (CH_2); IR (NaCl film): $\tilde{\nu} = 3400$ (br), 2960, 2930, 2870, 1465, 1385, 1230, 1070, 895 cm^{-1}; MS (70 eV, EI): *m/z* (%): 129 (30) $[M^+ - 1]$, 87 (43) $[C_5H_{11}O]^+$, 83 (100) $[C_6H_{11}]^+$.

(2*R*,3*R*,4*S*)-3-Azido-4-methylhexane-1,2-diol (12) [20]: To a refluxing, stirred suspension of titanium diazidodiisopropoxide (1.275 g, 5.1 mmol) in anhydrous benzene (43 mL) a solution of the epoxy alcohol **11** (0.55 g, 4.25 mmol) in anhydrous benzene (21 mL) was added dropwise, and refluxing maintained for 5–10 min more. The resulting solution was allowed to attain room temperature, and the solvent was eliminated under reduced pressure. The residue was taken up in diethyl ether (85 mL) and 5% aqueous sulfuric acid (35 mL) and the mixture stirred for 30–45 min, after which the two layers were completely clear. The phases were separated, the aqueous one extracted with dichloromethane, and the combined organic phases washed with brine. Drying over anhydrous magnesium sulfate and evaporation of the solvents under reduced pressure gave 0.75 g (quantitative yield) of crude azidodiol **12**, which was pure enough for subsequent transformations, but which can be further purified by column chromatography on silica gel with hexane/ethyl acetate mixtures of increasing polarity as eluents. The regioselectivity of the ring opening is higher than 16:1, according to ^{13}C NMR spectroscopy. Colourless oil; $[\alpha]_D^{23} = -34.7$ ($c = 2.0$ in $CHCl_3$); ^1H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 0.95$ (t, $^3J(H,H) = 7$ Hz, 3H; CH_3), 0.97 (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.45 (m, 2H; CH_2), 1.90 (m, 1H; CH), 2.20 (brs, 1H; OH), 2.63 (brs, 1H; OH), 3.46 (m, 1H; CH), 3.72 (m, 2H; CH_2), 3.83 (m, 1H; CH); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 11.6$ (CH_3), 13.7 (CH_3), 27.2 (CH_2), 35.8 (CH), 63.8 (CH_2), 68.0 (CH), 71.3 (CH); IR (NaCl film): $\tilde{\nu} = 3360$ (br), 3050, 2960, 2860, 2100, 1450, 1260, 1070 cm^{-1}; MS (CI): *m/z* (%): 208 (13) $[M+35]^+$, 191 (100) $[M+18]^+$, 174 (13) $[M+1]^+$.

Ring-opening reaction of (2*S*,3*S*,4*S*)-4-methyl-2,3-epoxyhexan-1-ol (11) with benzhydrylamine to give (2*R*,3*R*,4*S*)-3-(benzhydrylamino)-4-methylhexane-1,2-diol (13) and (2*R*,3*S*,4*S*)-2-(benzhydrylamino)-4-methylhexane-1,3-diol (14) [11]: To a stirred solution of epoxy alcohol **11** (0.50 g, 3.84 mmol) and titanium tetraisopropoxide (3.43 mL, 11.5 mmol) in anhydrous 1,2-dichloroethane (27 mL) a solution of benzhydrylamine (1.41 g, 7.68 mmol) in 1,2-dichloroethane (27 mL) was added dropwise. The mixture was stirred under reflux for 100 h, allowed to attain room temperature and treated with 16 mL of a 10% NaOH aqueous solution saturated with NaCl. After stirring at ambient temperature for 12 h, the titanium oxide precipitate was eliminated by filtration through Celite® and thoroughly washed with dichloromethane. The aqueous phase was extracted twice with dichloromethane, and the combined organic phases were subsequently dried over magnesium sulfate. Elimination of the solvents under reduced pressure gave 2.07 g of a crude oil which was subjected to column chromatography on triethylamine-pretreated silica gel (2.5% v/v), with hexane/ethyl acetate mixtures of increasing polarity as eluents. In this way 90 mg (18%) of starting material was obtained, along with 0.59 g of **13** (49%, 61% yield based on reacted **11**) and 0.15 g (13%) of **14**.

(2*R*,3*R*,4*S*)-3-(benzhydrylamino)-4-methylhexane-1,2-diol (13): Colourless oil; $[\alpha]_D^{23} = -16.8$ ($c = 2.27$ in $CHCl_3$); ^1H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 0.75$ (t, $^3J(H,H) = 7$ Hz, 3H; CH_3), 0.93 (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.20 (m, 2H; CH_2), 1.60 (m, 1H; CH), 2.60 (m, 1H; CH), 2.70 (br, 3H; NH +2OH), 3.52–3.72 (m, 3H; CH +CH_2), 4.99 (m, 1H; CH), 7.10–7.40 (m, 10H; aromatic CH); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 12.1$ (CH_3), 14.8 (CH_3), 27.4 (CH_2), 35.8 (CH), 59.9 (CH), 64.9 (CH_2), 65.0 (CH), 71.5 (CH), 127.0 (CH), 127.1 (CH), 127.2 (CH), 127.4 (CH), 128.4 (CH), 143.4 (Cq), 144.0 (Cq); IR (NaCl film): $\tilde{\nu} = 3360$ (br), 3070, 3040, 3010, 2940, 2910, 2860, 1590, 1470, 1450, 1440, 1060, 1020, 730, 690 cm^{-1}; MS (70 eV, EI): *m/z* (%): 252 (15) $[M^+ - C_2H_5O_2]$, 167 (100) $[C_{13}H_{11}]^+$, 83 (4) $[C_6H_{11}]^+$.

(2*R*,3*S*,4*S*)-2-(benzhydrylamino)-4-methylhexane-1,3-diol (14): Colourless oil; $[\alpha]_D^{23} = +16.7$ ($c = 2.78$ in $CHCl_3$); ^1H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 0.69$ (d, $^3J(H,H) = 6$ Hz, 3H; CH_3), 0.86 (t, $^3J(H,H) = 7.5$ Hz, 3H; CH_3), 1.15–1.50 (m, 2H; CH_2), 1.70 (m, 1H; CH), 2.55 (brs, 3H; NH +2OH), 2.70 (m, 1H; CH), 3.44 (dd, $^3J(H,H) = 8$ Hz, $^3J'(H,H) = 4$ Hz, 1H; CH), 3.72 (m, 2H; CH_2), 4.99 (m, 1H; CH), 7.20–7.40 (m, 10H; aromatic CH); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 10.8$ (CH_3), 14.6 (CH_3), 25.4 (CH_2), 36.8 (CH), 56.3 (CH), 60.5 (CH_2), 64.0 (CH), 75.8 (CH), 127.1 (CH), 127.2 (CH), 127.3 (CH), 127.5 (CH), 128.5 (CH), 128.6 (CH), 143.5 (Cq), 143.8 (Cq); IR (NaCl film): $\tilde{\nu} = 3400$ (br), 3080, 3040, 3010, 2945, 2910, 2860, 1590, 1470, 1455, 1440, 1035, 1015, 740, 695 cm^{-1}; MS (70 eV, EI): *m/z* (%): 226 (14) $[M^+ - C_5H_{11}O]$, 167 (100) $[C_{13}H_{11}]^+$, 83 (22) $[C_6H_{11}]^+$.

(2*R*,3*R*,4*S*)-3-(*tert*-Butoxycarbonylamino)-4-methylhexane-1,2-diol (7) from (2*R*,3*R*,4*S*)-3-(benzhydrylamino)-4-methylhexane-1,2-diol (13) [13a]: To a stirred suspension of 20% $Pd(OH)_2$ on carbon (55 mg) in dry ethyl acetate (1.0 mL) under a hydrogen atmosphere a solution of (2*R*,3*R*,4*S*)-3-benzhydrylamino-4-methylhexane-1,2-diol (0.55 g, 1.75 mmol) and di-*tert*-butyl dicarbonate (0.50 g, 2.28 mmol) in ethyl acetate (2.4 mL) was added through a cannula. After stirring for 24 h at room temperature, the reaction mixture was filtered through Celite®, which was subsequently washed thoroughly with dichloromethane. Elimination of the solvents under reduced pressure gave a crude product (0.83 g), which was purified by column chromatography on triethylamine-pretreated silica gel (2.5% v/v) with hexane/ethyl acetate mixtures of increasing polarity as eluents, to give 0.35 g (80% yield) of pure *N*-Boc-aminodiol **7**.

(2*R*,3*R*,4*S*)-3-(*tert*-Butoxycarbonylamino)-4-methylhexane-1,2-diol (7) from (2*R*,3*R*,4*S*)-3-azido-4-methylhexane-1,2-diol (12) [21]: To a stirred suspension of 10% Pd on carbon (75 mg) in dry ethyl acetate (1.3 mL) under a hydrogen atmosphere a solution of (2*R*,3*R*,4*S*)-3-azido-4-methylhexane-1,2-diol (0.74 g, 4.28 mmol) and di-*tert*-butyl dicarbonate (1.21 g, 5.56 mmol) in ethyl acetate (5.9 mL) was added through a cannula. After being stirred for 7 h at room temperature, the reaction mixture was filtered through Celite®, which was subsequently washed thoroughly with dichloromethane. Elimination of the solvents under reduced pressure gave a crude product (1.44 g), which was purified by column chromatography on triethylamine-pretreated silica gel (2.5% v/v) with hexane/ethyl acetate mixtures of increasing polarity as eluents, to give 0.97 g (92% yield) of *N*-Boc-aminodiol **7** as a colourless oil. $[\alpha]_D^{23} = +7.2$ ($c = 2.03$ in $CHCl_3$); ^1H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 0.87$ (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 0.92 (t, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.32 (m, 2H; CH_2), 1.45 (s, 9H; $C(CH_3)_3$), 2.05 (m, 1H; CH), 2.50 (brs, 2H; 2OH), 3.40 (m, 2H; CH_2), 3.58 (m, 1H; CH), 3.66 (m, 1H; CH), 4.64 (brd, 1H; NH); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 11.6$ (CH_3), 12.9 (CH_3), 27.1 (CH_2), 28.2 ($3CH_3$), 33.0 (CH), 53.5 (CH), 63.4 (CH_2), 71.6 (CH), 80.1 (Cq), 157.3 (Cq); IR (NaCl film): $\tilde{\nu} = 3340$ (br), 2950, 2920, 2860, 1690 (br, C=O), 1510, 1450, 1390, 1360, 1240, 1070 cm^{-1}; MS (70 eV, EI): *m/z* (%): 186 (12) $[M^+ - C_2H_5O_2]$, 130 (76) $[M^+ - C_4H_8 - C_2H_5O_2]$, 86 (100) $[C_6H_{12}N]^+$.

(*R*)-1-[(1*R*,2*S*)-1-(*tert*-Butoxycarbonylamino)-2-methylbutyl]oxirane (15) [21]: To a stirred solution of the *N*-Boc aminodiol **7** (0.92 g, 3.72 mmol) and triphenylphos-

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phine (1.02 g, 3.90 mmol) in dry chloroform (30 mL) at room temperature, a solution of diethyl azodicarboxylate (0.68 g, 3.90 mmol) in anhydrous chloroform (1.5 mL) was added dropwise. The resulting mixture was heated to reflux for 48 h, cooled and stripped of solvents by evaporation. The residue was purified directly by column chromatography on triethylamine-pretreated (2.5% v/v) silica gel with hexane/ethyl acetate mixtures of increasing polarity as eluents. This method yielded 0.61 g (72% yield) of the oxirane **15** as a white solid. M.p. 47–48 °C; $[\alpha]_D^{23} = -2.16$ ($c = 1.93$ in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 0.92$ (t, $^3J(H,H) = 7$ Hz, 3H; CH_3), 0.95 (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.30 (m, 2H; CH_2), 1.45 (s, 9H; $C(CH_3)_3$), 1.73 (m, 1H; CH), 2.78 (m, 2H; CH_2), 2.88 (m, 1H; CH), 3.38 (m, 1H; CH), 4.48 (brd, 1H; NH); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 11.5$ (CH_3), 14.1 (CH_3), 26.2 (CH_2), 28.2 ($3CH_3$), 36.6 (CH), 46.1 (CH_2), 52.6 (CH), 55.5 (CH), 79.3 (Cq), 155.6 (Cq); IR (KBr pellet): $\tilde{\nu} = 3340$, 3050, 2970, 2930, 2870, 1705 (C=O), 1520, 1460, 1390, 1365, 1250, 1170 cm^{-1}; MS (CI): *m/z* (%): 247 (84) $[M+18]^+$, 230 (49) $[M+1]^+$, 191 (52) $[M^+ + 18 - C_4H_8]$, 174 (14) $[M^+ + 1 - C_4H_8]$, 52 (100) $[C_4H_4]^+$; $C_{12}H_{23}NO_3$ (229.2): calcd C 62.85, H 10.11, N 6.11; found C 63.22, H 10.37, N 6.01.

(3*S*,4*R*,5*S*)-4-(*tert*-Butoxycarbonylamino)-3-hydroxy-5-methylheptanonitrile (16): A solution of the oxirane **15** (0.55 g, 2.40 mmol), 2-hydroxy-2-methylpropanonitrile (0.24 g, 2.85 mmol) and triethylamine (0.40 mL, 2.85 mmol) in anhydrous THF (1.0 mL) was refluxed for 16 h, after which it was cooled, diluted with diethyl ether (5 mL) and poured into water (5 mL). The phases were separated and the aqueous phase was extracted with ether. The combined organic phase was dried over anhydrous magnesium sulfate and stripped of solvents under reduced pressure to give 0.61 g of a crude product, which was purified by column chromatography on triethylamine-pretreated silica gel (2.5% v/v) with hexane/ethyl acetate mixtures of increasing polarity as eluents to afford 0.42 g (68% yield) of hydroxynitrile **16**. White solid; m.p. 74–75 °C; $[\alpha]_D^{23} = -1.64$ ($c = 1.79$ in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 0.92$ (t, $^3J(H,H) = 7$ Hz, 3H; CH_3), 0.93 (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.20–1.40 (m, 2H; CH_2), 1.44 (s, 9H; $C(CH_3)_3$), 1.85 (m, 1H; CH), 2.51 (A of ABX system, $^2J(H,H) = 17$ Hz, 1H; CHH'CN), 2.65 (B of ABX system, $^2J(H,H) = 17$ Hz, 1H; CHH'CN), 3.27 (d, $^3J(H,H) = 6.5$ Hz, 1H; OH), 3.65 (m, 1H; CH), 3.90 (m, 1H; CH), 4.48 (brd, 1H; NH); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 11.4$ (CH_3), 13.2 (CH_3), 23.5 (CH_2), 26.8 (CH_2), 28.2 ($3CH_3$), 33.8 (CH), 57.5 (CH), 68.9 (CH), 80.1 (Cq), 118.6 (Cq), 156.5 (Cq); IR (KBr pellet): $\tilde{\nu} = 3340$ (br), 2930, 2870, 2250 (C≡N), 1695 (br, C=O), 1525, 1460, 1395, 1370, 1250, 1170, 1075 cm^{-1}; MS (CI): *m/z* (%): 274 (100) $[M+18]^+$, 257 (51) $[M+1]^+$, 218 (45) $[M^+ + 18 - C_4H_8]$, 201 (15) $[M^+ + 1 - C_4H_8]$; $C_{13}H_{24}N_2O_3$ (256.2): calcd C 60.91, H 9.44, N 10.93; found C 61.04, H 9.62, N 10.82.

(3*S*,4*R*,5*S*)-3-(*tert*-Butyldimethylsilyloxy)-4-(*tert*-butoxycarbonylamino)-5-methyl heptanonitrile (17): A mixture of hydroxynitrile **16** (0.32 g, 1.25 mmol), *tert*-butyldimethylsilyl chloride (0.56 g, 3.74 mmol), imidazole (0.51 g, 7.49 mmol) and dry DMF (320 mL) was stirred at room temperature for 5 h, diluted with diethyl ether (5 mL) and poured into water. The phases were separated; the aqueous one was extracted with diethyl ether and the combined organic extracts were washed with saturated aqueous ammonium chloride and dried over magnesium sulfate. The solvents were eliminated under reduced pressure to give 0.74 g of a crude product, which was purified by column chromatography on triethylamine-pretreated silica gel (2.5% v/v) with hexane/ethyl acetate mixtures of increasing polarity as eluents, affording 0.405 g (87% yield) of the nitrile **17**. White solid; m.p. 86–87 °C; $[\alpha]_D^{23} = +17.1$ ($c = 2.77$ in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 0.11$ (s, 3H; $SiCH_3$), 0.17 (s, 3H; $SiCH_3$), 0.85 (t, $^3J(H,H) = 7$ Hz, 3H; CH_3), 0.91 (s, 9H; $SiC(CH_3)_3$), 0.95 (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.20–1.40 (m, 2H; CH_2), 1.45 (s, 9H; $OC(CH_3)_3$), 1.85 (m, 1H; CH), 2.47 (A of ABX system, $^2J(H,H) = 17$ Hz, 1H; CHH'CN), 2.62 (B of ABX system, $^2J(H,H) = 17$ Hz, 1H; CHH'CN), 3.70 (m, 1H; CH), 3.89 (m, 1H; CH), 4.47 (brd, 1H; NH); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = -4.9$ ($SiCH_3$), −4.3 ($SiCH_3$), 11.7 (CH_3), 13.5 (CH_3), 17.9 (SiCq), 23.9 (CH_2), 25.6 ($SiC(CH_3)_3$), 27.3 (CH_2), 28.3 ($OC(CH_3)_3$), 33.4 (CH), 56.8 (CH), 69.0 (CH), 79.6 (Cq), 117.8 (Cq), 155.8 (Cq); IR (NaCl film): $\tilde{\nu} = 3360$ (br), 2960, 2920, 2860, 2250 (C≡N), 1700 (br, C=O), 1500, 1460, 1390, 1360, 1250, 1160, 1110 cm^{-1}; MS (CI): *m/z* (%): 388 (100) $[M+18]^+$, 371 (25) $[M+1]^+$, 332 (15) $[M^+ + 18 - C_4H_8]$, 315 (5) $[M^+ + 1 - C_4H_8]$; $C_{19}H_{38}N_2O_3Si$ (370.2): calcd C 61.58, H 10.33, N 7.56; found C 61.51, H 10.31, N 7.30.

(3*S*,4*R*,5*S*)-3-(*tert*-Butyldimethylsilyloxy)-4-(*tert*-butoxycarbonylamino)-5-methyl heptanamide (8): To a suspension of the silylated hydroxynitrile **17** (70 mg, 0.19 mmol) in a 6:4 mixture of *tert*-butyl alcohol/water (2.8 mL) were added successively a 1 M NaOH aqueous solution (0.21 mL) and aqueous hydrogen peroxide (31 mL of a 33% w/v solution, 0.30 mmol). The mixture was stirred at room temperature for 66 h, during which the addition of the same quantities of NaOH and H_2O_2 was repeated four times until no more starting material was detected by TLC. After dilution with dichloromethane (5 mL), the reaction mixture was poured into water (2 mL). The phases were separated; the aqueous one is extracted with dichloromethane, and the combined organic extracts were washed successively with 1 M aqueous hydrochloric acid, saturated aqueous sodium bicarbonate solution and brine, and dried over magnesium sulfate. The solvents were eliminated under reduced pressure to give 95 mg of a crude product, which was purified by column chromatography on triethylamine-pretreated silica gel (2.5% v/v) with hexane/ethyl acetate mixtures of increasing polarity as eluents, affording 50 mg (68% yield) of the protected isostatine amide **8**. White solid; m.p. 57–59 °C; $[\alpha]_D^{23} = +9.11$ ($c = 2.80$ in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 0.08$ (s, 6H; $Si(CH_3)_2$), 0.80 (t, $^3J(H,H) = 7$ Hz, 3H; CH_3), 0.87 (s, 9H; $SiC(CH_3)_3$), 0.95 (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.10–1.30 (m, 2H; CH_2), 1.40 (s, 9H; $OC(CH_3)_3$), 1.78 (m, 1H; CH), 2.35 (A of ABX system, $^2J(H,H) = 17$ Hz, 1H; CHH'$CONH_2$), 2.48 (B of ABX system, $^2J(H,H) = 17$ Hz, 1H; CHH'$CONH_2$), 3.64 (m, 1H; CH), 4.07 (m, 1H; CH), 4.51 (brd, 1H; carbamate NH), 5.67 (brs, 1H; CONHH'), 6.13 (brs, 1H; CONHH'); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = -4.3$ ($SiCH_3$), −5.0 ($SiCH_3$), 11.7 (CH_3), 13.5 (CH_3), 17.9 (SiCq), 25.8 ($SiC(CH_3)_3$), 27.2 (CH_2), 28.4 ($OC(CH_3)_3$), 34.0 (CH), 41.4 (CH_2), 57.5 (CH), 69.8 (CH), 79.2 (Cq), 156.3 (Cq), 173.3 (Cq); IR (NaCl film): $\tilde{\nu} = 3360$ (br), 3200 (br), 2980, 2940, 2880, 1680 (br, carbamate C=O), 1615 (amide C=O), 1470, 1400, 1370, 1250, 1090 cm^{-1}; MS (CI): *m/z* (%): 389 (84) $[M+1]^+$, 333 (100) $[M^+ + 1 - C_4H_8]$, 289 (65) $[M^+ + 1 - C_5H_8O_2]$; $C_{19}H_{40}N_2O_4Si$ (388.2): calcd C 58.72, H 10.37, N 7.21; found C 58.54, H 10.37, N 7.01.

(4*R*,5*S*)-3-(*tert*-Butoxycarbonyl)-5-cyanomethyl-2,2-dimethyl-4-[(*S*)-1-methylpropyl]-1,3-oxazolidine (18) [7a]: A mixture of hydroxynitrile **16** (0.19 g, 0.74 mmol), 2-methoxypropene (174 μL, 1.85 mmol), *p*-toluenesulfonic acid monohydrate (28 mg, 0.15 mmol) and dry *N*,*N*-dimethylformamide (3.8 mL) was stirred at room temperature for 24 h, after which it was diluted with diethyl ether (5 mL) and poured into water. The phases were separated, the aqueous one was extracted with diethyl ether and the combined organic extracts were washed successively with aqueous saturated $NaHCO_3$ solution and brine. Drying of the organic phase over anhydrous magnesium sulfate and elimination of the solvents under reduced pressure gave a crude product (0.385 g) that was purified by column chromatography on triethylamine-pretreated silica gel (2.5% v/v) with hexane/diethyl ether mixtures of increasing polarity as eluents. This method furnished 0.12 g (55% yield, 81% based on reacted **16**) of the oxazolidine **18** as a colourless oil, which according to NMR contained less than 5% of the *trans* diastereomer **19**. 60 mg (32%) of the starting hydroxynitrile **16** was also recovered. $[\alpha]_D^{23} = +13.2$ ($c = 1.32$ in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 55 °C, TMS): $\delta = 0.85$ (t, $^3J(H,H) = 7$ Hz, 3H; CH_3), 0.88 (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.14 (m, 2H; CH_2), 1.40 (s, 9H; $OC(CH_3)_3$), 1.46 (m, 1H; CH), 1.50 (brs, 3H; oxazolidine CH_3), 1.51 (brs, 3H; oxazolidine CH_3), 2.54 (A of ABX system, $^2J(H,H) = 16.5$ Hz, 1H; CHH'CN), 2.62 (B of ABX system, $^2J(H,H) = 16.5$ Hz, 1H; CHH'CN), 3.90 (m, 1H; C_4-oxazolidine CH), 4.22 (dt, $^3J(H,H) = 7$ Hz, $^3J'(H,H) = 5.4$ Hz, 1H; C_5-oxazolidine CH); ^{13}C NMR (75 MHz, [D$_6$]DMSO, 60 °C, TMS): $\delta = 11.4$ (CH_3), 15.5 (CH_3), 18.0 (CH_2), 24.2 (oxazolidine CH_3), 26.0 (oxazolidine CH_3), 27.8 ($OC(CH_3)_3$), 27.9 (CH_2), 34.2 (CH), 60.8 (CH), 72.3 (CH), 79.2 (Cq), 92.7 (oxazolidine Cq), 118.2 (Cq), 152.2 (Cq); IR (NaCl film): $\tilde{\nu} = 2980$, 2940, 2880, 2270 (C≡N), 1700 (br, carbamate C=O), 1465, 1390, 1370, 1255, 1180, 1120 cm^{-1}; MS (CI): *m/z* (%): 314 (100) $[M+18]^+$, 297 (74) $[M+1]^+$, 258 (14) $[M^+ + 18 - C_4H_8]$, 241 (8) $[M^+ + 1 - C_4H_8]$, 117 (30) $[M^+ + 18 - C_5H_{11}NO_2]$. When the oxazolidine formation was effected under the more vigorous conditions described by Garner and Park [29] (0.35 mmol of **16**, 10.51 mmol of 2,2-dimethoxypropane added in two portions, cat. *p*-toluenesulfonic acid (1.4% mol), 2 mL benzene, reflux continuous distillation of methanol, 3 h), we obtained 32 mg after chromatographic purification (31% yield) of an 8:1 mixture of **18** and the (4*R*,5*R*) diastereomer **19**, easily identified by the ^{1}H NMR spectrum (300 MHz, $CDCl_3$, 55 °C, TMS) of the mixture: $\delta = 2.53$ (m, 2H; CH_2CN), 4.10 (dt, $^3J(H,H) = 6.6$ Hz, $^3J'(H,H) = 2.4$ Hz, 1H; C_5-oxazolidine CH), among other signals.

(4*S*,5*S*)-3-(*tert*-Butoxycarbonyl)-5-formylmethyl-2,2-dimethyl-4-[(*S*)-1-methylpropyl]-1,3-oxazolidine (20): To a cold (−40 °C) solution of the nitrile **18** (0.11 g, 0.37 mmol) in anhydrous diethyl ether (2.7 mL) was added dropwise 0.66 mL (0.67 mmol) of a 20% solution of DIBAL-H in hexanes. The mixture was stirred at −40 °C for 5 min, at −20 °C for 30 min, treated at this temperature with ethyl acetate (2.7 mL) and with a saturated aqueous solution of sodium potassium tartrate (2.7 mL), allowed to attain room temperature and vigorously stirred for 2 h. The resulting mixture was diluted with diethyl ether (5 mL), the phases were separated, and the aqueous one was extracted with diethyl ether. The combined organic extracts were then washed with brine and dried over anhydrous magnesium sulfate. Evaporation of the solvents afforded 0.11 g (quantitative yield) of the aldehyde **20**, pure enough to be used in subsequent steps. Colourless oil; $[\alpha]_D^{23} = +8.26$ ($c = 2.05$ in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 0.87$ (t, $^3J(H,H) = 7$ Hz, 3H; CH_3), 0.90 (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.20 (m, 2H; CH_2), 1.48 (s, 9H; $OC(CH_3)_3$), 1.51, 1.55* (brs, 3H; oxazolidine CH_3), 1.61 (brs, 3H; oxazolidine CH_3), 1.70 (m, 1H; CH), 2.74 (m, 2H; CH_2CN), 3.85, 4.00* (m, 1H; C_4-oxazolidine CH), 4.56 (m, 1H; C_5-oxazolidine CH), 9.81 (s, 1H; CHO) (signals marked with an asterisk correspond to a rotamer of the *N*-Boc group); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 11.7$, 12.0* (CH_3), 15.4, 15.8* (CH_3), 23.5, 25.2* (oxazolidine CH_3), 26.0, 26.6* (oxazolidine CH_3), 28.3 (CH_2), 28.4 ($OC(CH_3)_3$), 35.2, 35.9* (CH), 43.6 (CH_2), 61.5, 61.8* (CH), 71.5, 71.9* (CH), 80.0 (Cq), 93.4 (oxazolidine Cq), 153.0 (br, Cq), 199.7 (Cq); IR (NaCl film): $\tilde{\nu} = 2970$, 2920, 2850, 2710, 1720 (aldehyde C=O), 1690 (br, carbamate C=O), 1450, 1380, 1360, 1250, 1170, 1080 cm^{-1}; MS (CI): *m/z* (%): 317 (66) $[M+18]^+$, 300 (100) $[M+1]^+$, 261 (7) $[M^+ + 18 - C_4H_8]$, 244 (7) $[M^+ + 1 - C_4H_8]$, 200 (9) $[M^+ + 18 - C_5H_{11}NO_2]$.

(4*R*,5*S*)-3-(*tert*-Butoxycarbonyl)-5-carboxymethyl-2,2-dimethyl-4-[(*S*)-1-methylpropyl]-1,3-oxazolidine (21): To a vigorously stirred mixture of the aldehyde **20** (0.11 g, 0.37 mmol), *tert*-butyl alcohol (2.3 mL) and 5% aqueous NaH_2PO_4 (1.5 mL) at room temperature, a 1 M aqueous solution of $KMnO_4$ (2.2 mL, 2.2 mmol) was added dropwise. The excess permanganate was removed by addition of aqueous Na_2SO_3 solution, and the solution was cooled to 0 °C and acidified (pH 4–5) with precooled (0 °C) 10% aqueous HCl solution until the colloidal precipitate of MnO_2 redissolved. The aqueous phase was thoroughly extracted with ethyl acetate, and the combined organic extracts were washed with brine, dried over anhydrous magnesium sulfate and stripped of solvents under reduced pressure to give 0.11 g (94% yield) of the acid **21**. Colourless oil; $[\alpha]_D^{23} = +8.45$ ($c = 1.32$ in $CHCl_3$); ^{1}H NMR (200 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 0.88$ (t, $^3J(H,H) = 7$ Hz, 3H; CH_3), 0.91 (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.20 (m, 2H; CH_2), 1.48 (s, 9H; $OC(CH_3)_3$), 1.54 (brs, 6H; oxazolidine $C(CH_3)_2$), 1.60 (m, 1H; CH), 2.70 (d, $^3J(H,H) = 7$ Hz, 2H; CH_2COOH), 3.87, 4.02* (m, 1H; C_4-oxazolidine CH), 4.48 (m, 1H; C_5-oxazolidine CH), 8.28 (brs, 1H; COOH) (signals marked with an asterisk correspond to a rotamer of the *N*-Boc group); ^{13}C NMR (50 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 11.9$, 12.0* (CH_3), 15.6, 15.9* (CH_3), 23.4, 24.9* (oxazolidine CH_3), 25.9, 26.5* (oxazolidine CH_3), 28.3 ($OC(CH_3)_3$), 29.6 (CH_2), 34.4 (CH_2), 35.3, 35.7* (CH), 61.4, 61.6* (CH), 72.7, 73.0* (CH), 80.0 (Cq), 92.5 (oxazolidine Cq), 152.7 (Cq), 175.4 (Cq); IR (NaCl film): $\tilde{\nu} = 3200$ (br), 2970, 2930, 2880, 1730 (carboxylic acid C=O), 1690 (br, carbamate C=O), 1450, 1360 (br), 1360, 1250, 1170, 1090 cm^{-1}; MS (CI): *m/z* (%): 333 (100) $[M+18]^+$, 316 (95) $[M+1]^+$, 277 (53) $[M^+ + 18 - C_4H_8]$, 260 (17) $[M^+ + 1 - C_4H_8]$, 216 (93) $[M^+ + 18 - C_5H_{11}NO_2]$.

(4*R*,5*S*)-3-(*tert*-Butoxycarbonyl)-5-carboxymethyl-2,2-dimethyl-4-[(*S*)-1-methylpropyl]-5-methoxycarbonyl-1,3-oxazolidine (22): A solution of the carboxylic acid **21** (0.11 g, 0.35 mmol) in anhydrous diethyl ether (1 mL) was treated with an excess (persistence of the yellow colour) of ethereal diazomethane. Evaporation of the solvents and short-path silica gel filtration gave 0.11 g (95% yield) of the *N*-Boc-*N*,*O*-oxazolidine-protected isostatine methyl ester **22**, whose spectral data were fully coincident with those of an authentic sample [7g]. Colourless oil; $[\alpha]_D^{23} = +11.4$ ($c = 1.90$ in $CHCl_3$); ^{1}H NMR (300 MHz, $CDCl_3$, 25 °C, TMS): $\delta = 0.87$ (t, $^3J(H,H) = 7$ Hz, 3H; CH_3), 0.90 (d, $^3J(H,H) = 7$ Hz, 3H; CH_3), 1.00–1.20 (m, 2H; CH_2), 1.47 (s, 9H; $OC(CH_3)_3$), 1.54 (brs, 3H; oxazolidine CH_3), 1.58 (brs, 3H; oxazolidine CH_3), 1.60–1.70 (m, 1H; CH), 2.65 (m, 2H; CH_2COOCH_3), 3.71 (s, 3H; CH_2COOCH_3), 3.84, 4.00* (m, 1H; C_4-oxazolidine CH), 4.49 (m, 1H; C_5-oxazolidine CH) (the signal marked with an asterisk corresponds to a rotamer of the *N*-Boc group); ^{13}C NMR (75 MHz, $CDCl_3$, 55 °C, TMS): $\delta = 11.9$ (CH_3), 15.8 (CH_3), 24.6, 25.4* (oxazolidine CH_3), 26.4 (oxazolidine CH_3), 28.4 ($OC(CH_3)_3$), 28.6 (CH_2), 34.6 (CH_2), 35.7 (CH), 51.7 (CH_3), 61.8 (CH), 73.2 (CH), 79.8 (Cq), 92.5 (oxazolidine Cq), 152.7 (Cq), 171.1 (Cq); IR (NaCl film): $\tilde{\nu} = 2980$, 2940, 2880, 1745 (ester C=O), 1695 (br, carbamate C=O), 1450, 1435, 1365, 1250, 1170, 1100 cm^{-1}; MS (CI): *m/z* (%): 347 (74) $[M+18]^+$, 330 (100) $[M+1]^+$, 291 (10) $[M^+ + 18 - C_4H_8]$, 274 (1) $[M^+ + 1 - C_4H_8]$, 230 (11) $[M^+ + 18 - C_5H_{11}NO_2]$.

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[28] The diastereomeric excess of **11** was established by comparison with the mixture obtained by treatment of the allyl alcohol **10** (1.87 mmol) with *m*-chloroperbenzoic acid (3.75 mmol) in the presence of sodium carbonate (5.62 mmol) in dichloromethane solution (15 mL) at room temperature for 3.5 h, which gave a 4:6 mixture of **11** and the (2*R*,3*R*,4*S*) diastereomer (78% yield after chromatographic purification).

[29] P. Garner, J. M. Park, *J. Org. Chem.* **1987**, *52*, 2361–2364.