Total Synthesis of Amiclenomycin, an Inhibitor of Biotin Biosynthesis

Stéphane Mann, Sophie Carillon†, Olivier Breyne, and Andrée Marquet*[a]

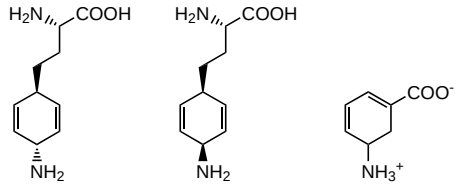
We dedicate this paper to Sophie Carillon, who recently died in a car accident. She performed a great part of this work.

Abstract: We describe the first synthesis of amiclenomycin, a natural product that has been found to inhibit biotin biosynthesis and, as a consequence, to exhibit antibiotic properties. Structure **1**, with a *trans* relationship between the ring substituents, had previously been proposed for amiclenomycin on the basis of its ^{1}H NMR spectrum. We have prepared the *trans* and *cis* isomers **1** and **2** by unequivocal routes and we conclude that the natural product is in fact the *cis* isomer **2**. The properly substituted cyclohexadienyl rings were constructed first. A cycloaddition reaction between 1,2-di(phenylsulfonyl)ethylene and the *N*-allyloxycarbonyl diene **13**, followed by reductive elimination of the phenylsulfinyl groups, gave the *cis* isomer **15**. To obtain the *trans* isomer, the *O*-trimethylsilyl diene was used to give the *cis* hydroxylated Diels–Alder adduct **33**, which was transformed into the corresponding *trans* amino derivative by means of a Mitsunobu reaction. The L-α-amino acid functionality was introduced by means of a Strecker reaction on the aldehydes **16** and **42**, followed by enzymatic hydrolysis with immobilised pronase.

Keywords: amiclenomycin · aminocyclohexadiene · antibiotics · enzymatic resolution · Strecker reaction

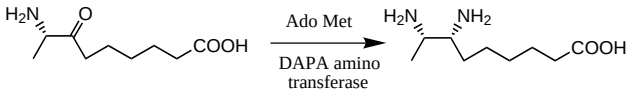
Introduction

Amiclenomycin (**2**) has been isolated from cultures of different *Streptomyces* strains either as the free amino acid[1] or as a component of di- and tripeptides,[2] all of which show antibiotic properties, with a specificity against mycobacteria.[1, 2b,c]



The antimutagenic effects of the dipeptide *N*-methylvalyl-amiclenomycin have also been described.[3] These antibiotic properties were reversed by biotin and some of its precursors, and it has been established that the target of amiclenomycin is 7,8-diaminopelargonic acid (DAPA) aminotransferase[4] (Scheme 1).

The mechanism of inhibition has been investigated,[5] and kinetic studies showed that amiclenomycin was probably recognised at the substrate binding site. Inactivation of the enzyme was observed after preincubation, but this was found to be reversible, the activity being recovered after dialysis. This may correspond to tight binding or, more probably, to the reversible formation of a covalent adduct. The analogy with the inhibition of γ-amino-butyric acid (GABA) aminotransferase by gabaculine (**3**)[6] should be explored.



Scheme 1. Action of DAPA aminotransferase, blocked by amiclenomycin.

In connection with our general interest in inhibitors of biotin biosynthesis with potential herbicidal properties,[7] we decided to revisit the mechanism of action of amiclenomycin.

Unfortunately, the natural product was no longer available, and all our attempts to isolate it from cultures of *S. Lavandulae*—with a strain kindly supplied by Dr. Okami—failed, maybe because of mutations in the strain. Thus, we decided to undertake the total synthesis of amiclenomycin.

A *trans* geometry between the two substituents on the cyclohexadienyl ring in **1** had been tentatively proposed by Okami et al,[1] on the basis of the ^5J coupling constant (7.5 Hz) between the two allylic hydrogens,[1] and this later became accepted.[2b] The other isomer had, of course, not been available, and in order to establish the stereochemistry of the natural product with more certainty, we decided to synthesise both isomers by unequivocal routes.

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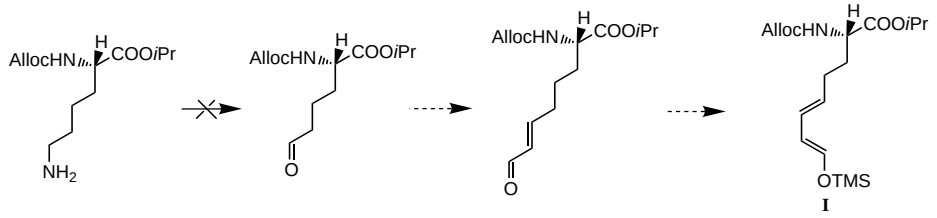
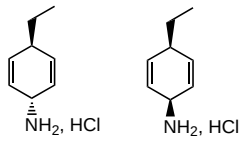
We chose a Diels–Alder strategy to construct the cyclohexadiene ring, with the use of *trans*-1,2-bis(phenylsulfonyl)-ethylene, an acetylene dienophile equivalent described by De Lucchi.[8] We expected that the mild conditions used to regenerate the double bond, reductive elimination of the phenylsulfinyl groups at room temperature with sodium amalgam in methanol buffered with potassium dihydrogenphosphate, would preserve the 2,5-cyclohexadiene. Initial attempts to prepare the oxygenated diene partner **I** of the Diels–Alder reaction from L-lysine according to Scheme 2 were not successful. We thus had to construct the cyclohexadiene part first, and then introduce the amino acid functionality.



I

Scheme 2. Unsuccessful attempts to prepare Diels—Alder diene **I**.

However, due to the expected sensitivity of the 1-amino-2,5-cyclohexadiene ring to conjugation and/or aromatisation, we first examined the synthetic routes to this moiety on a simple model before the construction of the amino acid side-chain. The synthesis of the two isomers **VII** and **VIII** and the assignment of their stereochemistry have been described in another paper.[9] In this work we report the preparation of the two isomers **1** and **2**.



VII **VIII**

Model compounds

The ^5J values observed in the *trans* and *cis* compounds **1** and **2** are 5.6 and 8.1 Hz, respectively; in good agreement with the corresponding values, 5.6 and 8.2 Hz, found in the model compounds **VII** and **VIII**.[9] The reported value for natural amiclenomycin was 7.5 Hz, very close to that of the *cis* compound, and we concluded that the published stereochemistry was very probably incorrect. Comparison of chemical shift values supported this conclusion (Table 1). It would be interesting to study inhibition of DAPA aminotransferase, the crystal structure of which has recently been published,[10] by these two isomers. We intend to examine whether one or both isomers cocrystallise with the protein, and we hope to obtain the three-dimensional structure of the complex(es).

Table 1. NMR chemical shift differences of hydrogen and carbon atoms in amiclenomycin (Acm) present in natural peptides,[2b] and in compounds **1** and **2**. The spectra were recorded in D_2O at 400 MHz (^{1}H NMR) and 100 MHz (^{13}C NMR), except for **1** and **2**, which were recorded at 500 MHz (^{1}H NMR) and 125 MHz (^{13}C NMR).

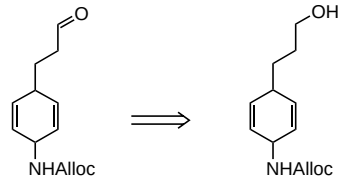
Compounds	$\Delta\delta$ H-1′–H-4′	$\Delta\delta$ C-1′–C-4′	$\Delta\delta$ C-3–C-4
MeIle-Acm	1.47	10.3	0.9
Ile-Acm	1.49	10.4	1.1
MeVal-Acm	1.48	10.4	1.5
MeIle-Acm-Gln	1.47	10.4	1.7
Ile-Acm-Gln	1.47	10.3	1.6
Val-Acm-Gln	1.47	10.3	1.2
1	1.37	10.15	2.39
2	1.46	10.46	1.45

Results and Discussion

Our first targets were precursors of the cyclohexadiene ring with a functionalised side-chain suitable for the introduction of the amino acid moiety.

A number of methods for the (enantioselective) synthesis of amino acids are now available.[11] We first considered the alkylation of glycine derivatives, for which many chiral versions exist.[11] Preliminary experiments were carried out with the anion of imine **II**[12] and the bromo derivatives **III** or **IV**. No reaction occurred at low temperature, whilst at 0 °C **III** and **IV** decomposed into aromatic products **V** and **VI** (Scheme 3).

We thus turned to application of the Strecker reaction to aldehydes **16** and **42**, and prepared their precursors **15** and **41**,

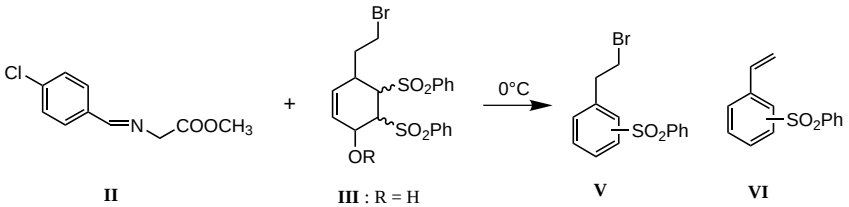


16 : *cis* isomer
42 : *trans* isomer

15 : *cis* isomer
41 : *trans* isomer

which contain hydroxypropyl side-chains, by the same Diels–Alder cycloaddition-based strategy as used for the preparation of the *trans* and *cis* models **VII** and **VIII**.[9]

Synthesis of the *cis* isomer 2: The diene **13** was synthesised from butane-1,4-diol as shown in Scheme 4. This symmetric



II **III** : R = H **V** **VI**

IV : R = THP

Scheme 3. Tentative attempts to alkylate the glycine Schiff base **II**.

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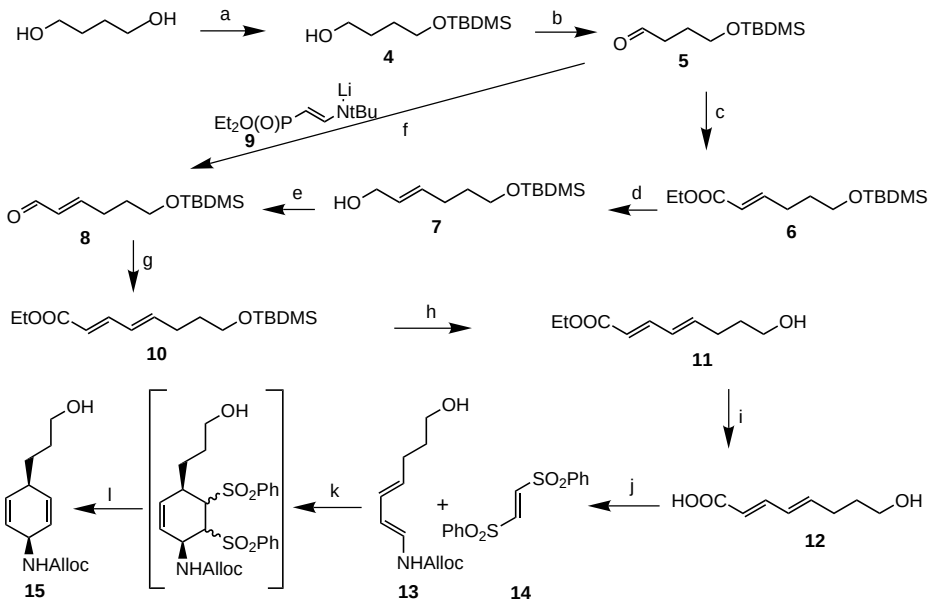


Scheme 4. a) NaH, TBDMSCl, THF, 97%. b) $(COCl)_2$, DMSO, CH_2Cl_2, $-60\,^{\circ}C$; NEt_3, $-20\,^{\circ}C$, 94%. c) NaH, $(EtO)_2P(O)CH_2CO_2Et$, THF, $-20\,^{\circ}C$, 80%. d) DiBAH, CH_2Cl_2, $-60\,^{\circ}C$, 84%. e) $(COCl)_2$, DMSO, CH_2Cl_2, $-60\,^{\circ}C$; NEt_3, $-20\,^{\circ}C$, 94%. f) LDA, *t*BuN=CH–CH_3, $(EtO)_2P(O)Cl$, THF, $-78\,^{\circ}C$; H_3O^+, 67%. g) NaH, $(EtO)_2P(O)CH_2CO_2Et$, THF, $-20\,^{\circ}C$, 70%. h) THF/H_2O/CH_3CO_2H (1:1:3), 71%. i) NaOH, MeOH, 60 °C, 83%. j) $(PhO)_2P(O)N_3$, allyl alcohol, NEt_3, reflux, 70%. k) **14**, *ortho*-xylene, reflux. l) Na(Hg), MeOH, KH_2PO_4, 37%.

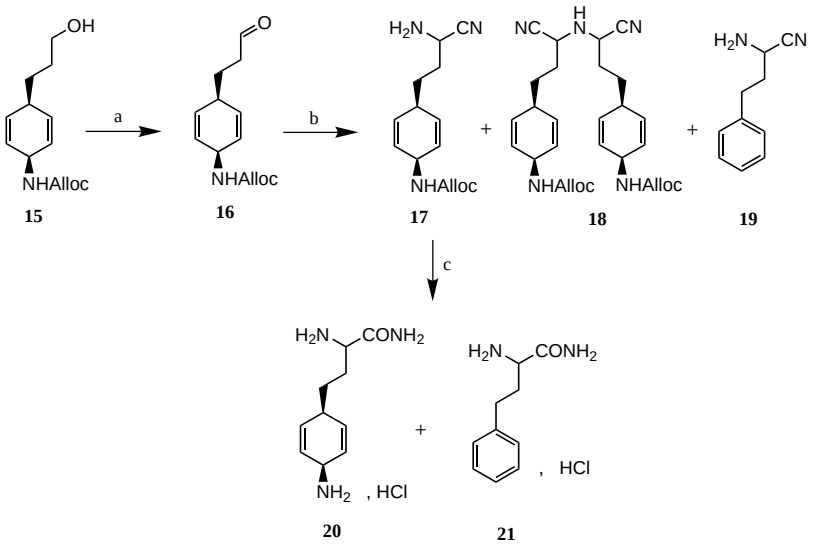
diol was monosilylated in 90% yield with *tert*-butyldimethylsilyl (TBDMS) chloride according to a method described by McDougal et al.,[13] who explained the selectivity of the reaction by the insolubility of the monosodium salt in THF. Alcohol **4** was then oxidised to aldehyde **5** by a Swern reaction and converted into the α,β-unsaturated aldehyde **8**. A Wittig reaction carried out with triethyl phosphonoacetate enabled aldehyde **5** to be transformed into the conjugated ester **6**, which was then reduced by diisobutyl aluminium hydride (DiBAH) to the allylic alcohol **7**. This was then oxidised to give **8**. This reaction could be performed more rapidly and with an equivalent yield by means of the formylolefination reaction developed by Meyers.[14] This conversion proceeds by condensation of lithium enaminophosphonate **9** with aldehyde **5** at $-78\,^{\circ}C$, followed by hydrolysis of the intermediate imine. Compound **9** was prepared in situ from acetaldehyde *tert*-butylimine,[15] by treatment with lithium diisopropylamide (LDA) and diethylchlorophosphate.

Aldehyde **8** was then transformed into the (1*E*,3*E*) conjugated ester **10** by means of a Wittig reaction with the triethyl phosphonoacetate anion. Deprotection of the alcohol moiety in compound **10** under acidic conditions afforded **11**. Saponification of the ester was achieved by using an aqueous sodium hydroxide solution in methanol under reflux. The *N*-protected (1*E*,3*E*) diene **13** was obtained from **12** by means of a Curtius reaction[16] under the same conditions as described for the preparation of the corresponding diene in the model series.

The Diels–Alder reaction between **13** and *trans*-1,2-di-(phenylsulfonyl)ethylene (**14**)[8] was performed in refluxing *ortho*-xylene to afford, after reductive elimination of the phenylsulfinyl groups from the mixture, cyclohexadiene **15**.

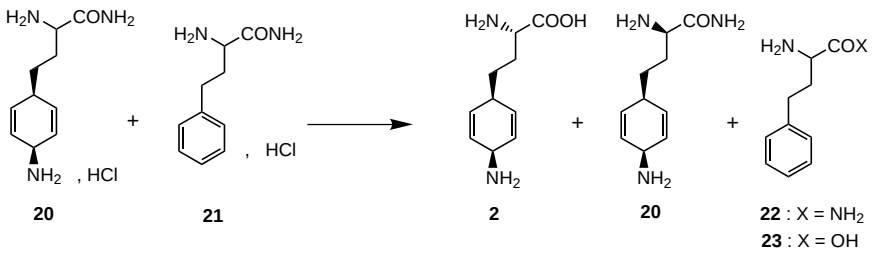
To introduce the amino acid functionality by a Strecker reaction[17] (Scheme 5), alcohol **15** was oxidised to **16** by the Swern method. This aldehyde was treated with trimethylsilyl cyanide in the presence of a catalytic amount of ZnI_2 and then with a saturated solution of ammonia in methanol to give amino nitrile **17**. The dicondensation product **18** and/or the aromatic compound **19** were isolated in some experiments. We found that the quality of the ammonia and the reaction temperature were important parameters. To minimise the formation of **18** and **19**, the reaction has to be performed with very pure ammonia and at a temperature not exceeding 50 °C for 15 min.

Cleavage of the allyloxycarbonyl group of **17** was carried out with $Pd(PPh_3)_4$ in the presence of phenylsilane.[18] During extraction of the resulting amine with an acidic aqueous solution at 0 °C followed by lyophilisation, hydration of the nitrile group took place, and the amide **20** was obtained as a mixture containing 22% of the aromatic compound **21**.

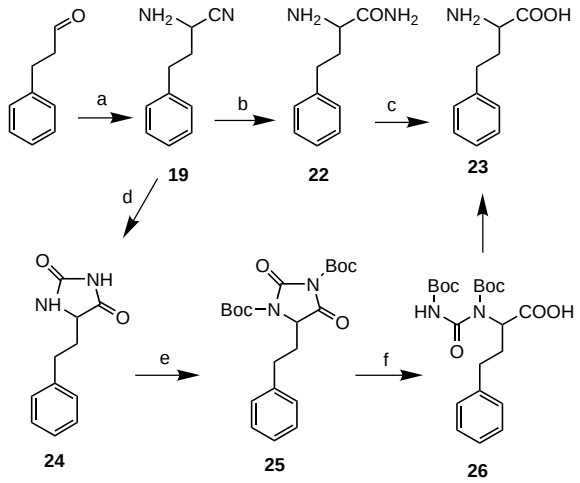


Scheme 5. a) $(COCl)_2$, DMSO, CH_2Cl_2, $-60\,^{\circ}C$; NEt_3, $-20\,^{\circ}C$, 95%. b) TMS–CN, ZnI_2, CH_2Cl_2, RT; NH_3, MeOH, 74%. c) $Pd(PPh_3)_4$, $PhSiH_3$, CH_2Cl_2, RT,; HCl, 60%.

The last step was the conversion of the amino acid amide **20** into the amino acid **2**. To conserve supplies of amide **20**, the reaction conditions were first examined on the aromatic analogue, homophenylalanine amide (**22**). This was prepared from 3-phenylpropanal by a Strecker reaction to provide **19**, which was then treated with a methanolic solution of $NaOH/H_2O_2$ (Scheme 6). Hydrolysis of **22** with reasonable concentrations (0.7 M) of basic reagents (NaOH, LiOH, Na_2O_2[19]) was very slow (over 24 h at room temperature) and therefore not compatible with the amino-cyclohexadienyl moiety.



Scheme 7. Conversion of D,L-amino acid amide **20** into L-amino acid **2** and D-amino acid amide **20** with the aid of immobilised pronase in a phosphate buffer (pH 9.6) at 37 °C.

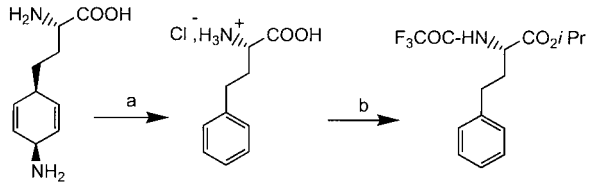


Scheme 6. a) TMS–CN, ZnI_2, CH_2Cl_2, RT; NH_3, MeOH, 73%. b) H_2O_2, NaOH, MeOH, RT, 76%. c) NaOH or LiOH or Na_2O_2, H_2O, RT. d) $(NH_4)_2CO_3$, H_2O, MeOH, 50 °C, 62%. e) Boc_2O, DMAP, THF, RT, 65%. f) LiOH, THF/H_2O (5:1), RT, 92%.

Another method, described as milder, is the hydrolysis of *N*-protected hydantoins by lithium hydroxide at room temperature.[20] Hydantoin **24** was prepared by condensation of ammonium carbonate with amino nitrile **19**. Protection of the nitrogen atoms of **24** with *tert*-butyloxycarbonyl groups afforded **25**. However, hydrolysis of the latter with lithium hydroxide gave compound **26** and homophenylalanine could be identified only after a few days (Scheme 6).

The alternative route of enzymatic hydrolysis was then considered. An enantioselective method for the conversion of α-amino nitriles into L-α-amino acids under moderately basic conditions (pH 10) has been described by Taillades et al.,[21] who used immobilised pronase on a poly(*N*-acryloylpiperidin-4-one) (80%) cross-linked with (1,4-bisacryloylpiperazine) (20%). They showed that the hydration of D,L-amino nitriles to D,L-amino acid amides is efficiently catalysed in phosphate or borate buffers by ketonic sites in the polymer matrix. Pronase, with its amidase activity, then effects the enantioselective transformation of the intermediate D,L-amino acid amides into L-amino acids and D-amino acid amides. Under these conditions, the D,L-mixture **20** (containing 27% of aromatic compound **21**) was converted into L-amino acid **2** and D-amino acid amide **20** (Scheme 7). Compounds **2** (28%) and **20** (29%) and the aromatic compounds **22** and **23** (25%) were separated on a Dowex column with ammonia solution as eluant.

In order to measure the enantiomeric excesses of acid **2** and amide **20**, these compounds were converted by acidic treatment into the corresponding aromatic amino acid **23**, which was derivatised into **27** by classical methods (Scheme 8) and submitted to gas chromatography on a chiral column. The retention times were compared with those of commercial racemic homophenylalanine, also derivatised into **27**. The enantiomeric excesses of **2** and **20** were 74% and 80%, respectively.



Scheme 8. a) 3 N HCl, reflux. b) 2 N *i*PrOH/HCl, 100 °C; $(CF_3CO)_2O$, CH_2Cl_2, RT. The same procedure was used for the derivatisation of **20**.

Synthesis of the *trans* isomer 1: The diene **31** was synthesised from pentane-1,5-diol as shown in Scheme 9. The diol was successively monosilylated, oxidised and converted into α,β-unsaturated aldehyde **30** by the methods described for the synthesis of **8** (Scheme 4).

Dienes **31** and **32** were obtained by the Danishefsky method.[22] Aldehyde **30**, activated by $ZnCl_2$, was treated with bromotrimethylsilane in the presence of triethylamine to afford a 35:65 mixture of **31** (1*E*,3*E*) and **32** (1*E*,3*Z*). We have shown in the case of the model compounds that the percentage of the desired (*E*,*E*) isomer could be improved by isomerisation with iodine but that the yield of the subsequent Diels–Alder reaction is lower and that the overall yield is identical with or without isomerisation.[9] Thus, the cycloaddition here was performed with the initial 35:65 mixture of dienes **31** and **32** and with *trans*-1,2-bis(phenylsulfonyl)ethylene (**14**) as the dienophile.[8]

The protective groups in dienes **31** and **32** were chosen in order to subsequently selectively hydrolyse the secondary hydroxyl group of the Diels–Alder adduct, but no selective cleavage was observed under various conditions. With a dilute solution of HCl in methanol (0.01 eq.) both silyl ethers were removed. With oxalic or acetic acid (0.001 to 0.01 eq.), the monoprotected compounds **34** was observed, but always in a mixture with the diol **33**. We thus had to isolate the

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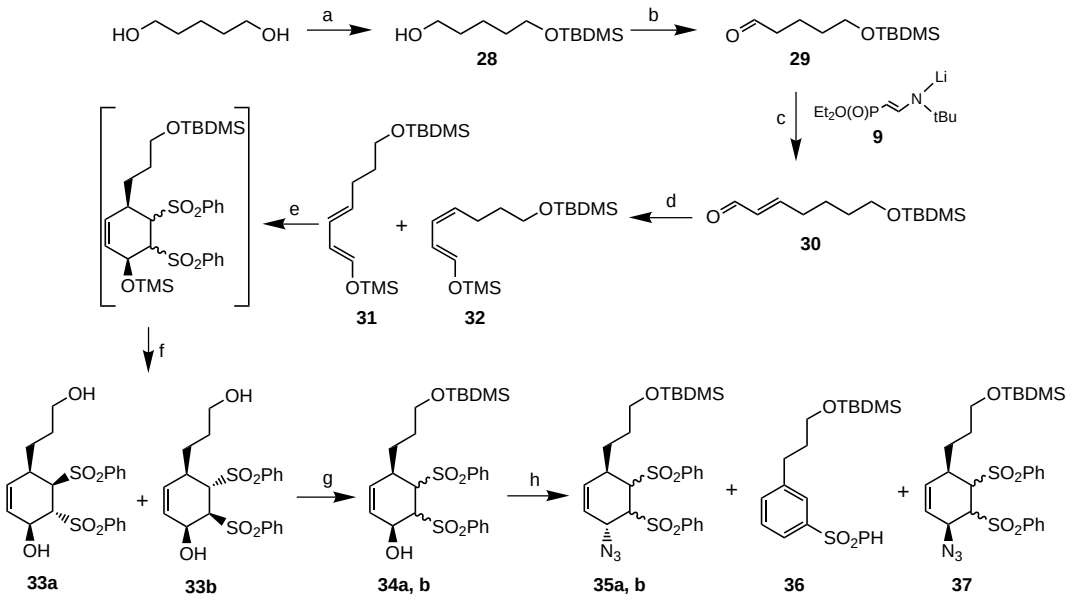


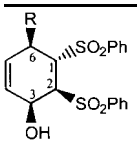
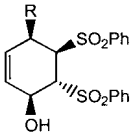
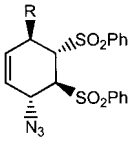
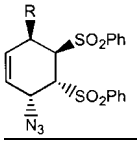
Scheme 9. a) NaH, TBDMSCl, THF, RT, 89%. b) $(COCl)_2$, DMSO, CH_2Cl_2, $-60\,^{\circ}C$; NEt_3, $-20\,^{\circ}C$, 90%. c) LDA, *t*BuN=CH-CH_3, $(EtO)_2P(O)Cl$, THF, $-78\,^{\circ}C$; H_3O^+, RT, 56%. d) TMSBr, $ZnCl_2$, NEt_3, toluene, reflux, 75%. e) **14**, *ortho*-xylene, $120\,^{\circ}C$. f) MeOH, H^+, RT, 66%. g) TBDMSCl, DMAP, CH_2Cl_2, NEt_3, 83%. h) PPh_3, DIAD, $(PhO)_2P(O)N_3$, CH_2Cl_2, $0\,^{\circ}C$, **35a,b**: 52%.

deprotected compound **33** and then reprotect the primary alcohol. From the similarity of the NMR spectra of the Diels–Alder adducts **33** to those of the model series (Table 2), we were able to conclude that **33** was a 80:20 to 65:35 mixture of **33a** and **33b**. The reprotection of the primary alcohol was achieved by treatment of the mixture of **33a** and **33b** with *tert*-butyldimethylsilyl chloride in the presence of triethylamine and a catalytic amount of *N*,*N*-dimethylaminopyridine (DMAP).[23] Alcohols **34** were transformed into azides **35a** and **35b** by a Mitsunobu reaction, with diphenylphosphoryl azide.[24] As in the case of the model compounds, the relative proportions of **35a** and **35b** (75:25 to 60:40) did not exactly match those of **34a** and **34b**. The epimeric azide **37** was also formed in 5–10% yield, as in the model series. The aromatic compound **36** was similarly isolated in a reproducible yield of 30%.

Amine **38**, obtained by reduction of azides **35a** and **35b** under hydrogen (5 bars) in the presence of Lindlar catalyst,[25] was then protected by an allyloxycarbonyl group to furnish **39a** and **39b**. Deprotection of the alcohol by acidic methanolysis, followed by desulfonylation with sodium amalgam gave the cyclohexadiene **41** (Scheme 10).

The last steps of this synthesis were achieved as in the case of the *cis* isomer (Scheme 11). Alcohol **41** was oxidised to **42**,

Table 2. Comparison of chemical shifts in compounds **33a**, **33b**, **35a**, **35b** and models[9] (solvent: $CDCl_3$, ^1H NMR: 400 MHz, ^{13}C NMR: 100 MHz).

Compounds	R	H-1	H-2	H-3	H-6	C-1	C-2	C-3	C-6
					δ				
	Et	4.15	4.37	4.88	2.58	61.59	61.59	65.15	33.60
	$(CH_2)_3OH$	4.10	4.22	4.81	2.61	61.55	61.50	64.58	31.40
	Et	4.16	3.96	4.55	2.96	58.94	66.34	61.73	37.04
	$(CH_2)_3OH$	4.10	3.83	4.52	2.87	58.96	66.46	61.45	34.82
	Et	4.14	4.21	4.42	2.70	57.89	60.10	50.91	33.78
	$(CH_2)_3OTBDMS$	4.13	4.20	4.46	2.82	58.50	61.30	51.21	32.10
	Et	4.67	4.46	4.88	3.02	62.40	60.39	54.99	37.07
	$(CH_2)_3OTBDMS$	4.66	4.43	4.88	3.12	62.53	60.45	54.95	35.03

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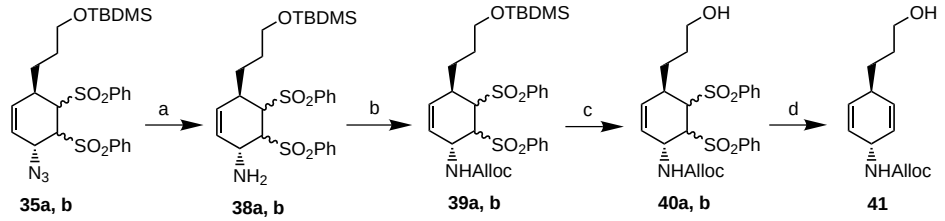
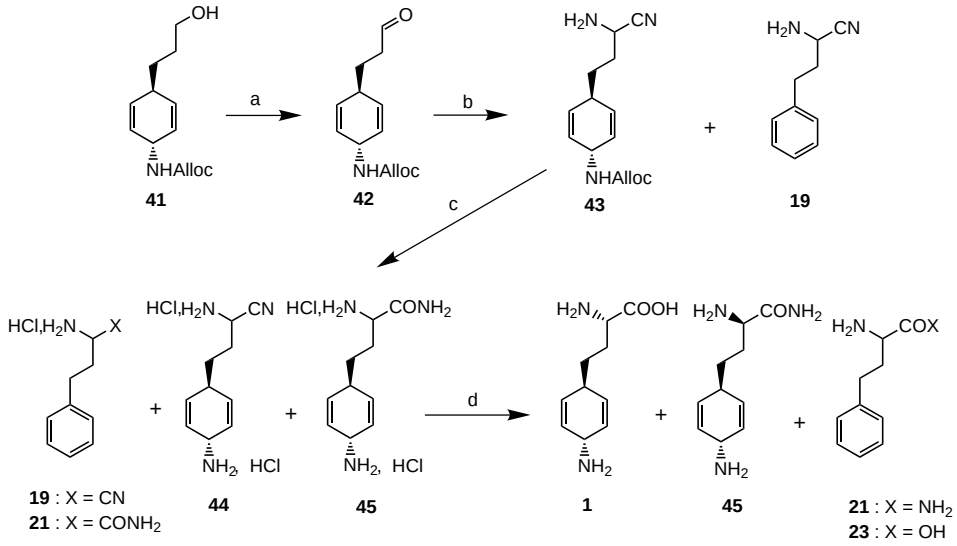


Scheme 10. a) H_2 (5 bars), Pd(5%)/$CaCO_3$-Pb(3.5%), THF/*i*PrOH (1:1), RT. b) $ClCO_2All$, EtOH, $NaHCO_3$, sonication, RT, 76% from **35**. c) MeOH, H^+, RT, 86%. d) Na(Hg), MeOH, KH_2PO_4, RT, 74%.

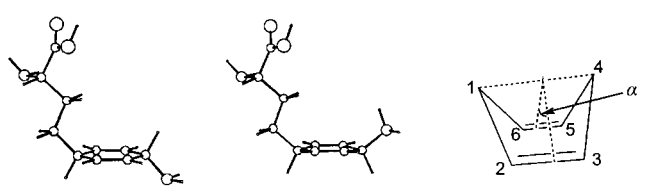


Scheme 11. a) $(COCl)_2$, DMSO, CH_2Cl_2, $-60\,^\circ C$; NEt_3, $-20\,^\circ C$, quantitative yield. b) TMS–CN, ZnI_2, CH_2Cl_2, RT; NH_3, MeOH, 51%. c) $Pd(PPh_3)_4$, $PhSiH_3$, CH_2Cl_2, RT; HCl, 85%. d) immobilised pronase, phosphate buffer, pH 9.6, 37 °C, **1**: 20%, **45**: 20%.

which was converted into amino nitrile **43** accompanied by the aromatic compound **19**. Deprotection of the amine followed by a rapid extraction at 0 °C with 1 N hydrochloric acid gave a mixture of amino nitrile **44** and amino acid amide **45** (in a ratio of 80:20 to 30:70), and their aromatic derivatives **19** and **21** (13 to 23%). This crude mixture, when treated with immobilised pronase, afforded the L-amino acid **1** (20%) and the D-amino acid amide **45** (20%). The enantiomeric excesses of these products were 94% and 88%, respectively (determined by chiral gas chromatography, as described above for **2** and **20**).

Configuration of amiclenomycin: The ^5J(H-1′,H-4′) coupling constants measured after irradiation of H-3′ in the *trans* and *cis* isomers—5.6 and 8.1 Hz, respectively—are in good agreement with the values found in the model compounds **VII** and **VIII** (5.6 and 8.2 Hz). The observed value in the natural product was 7.5 Hz, close to the value found in the *cis* compound. The chemical shift differences reported in Table 1 for the amiclenomycin-containing peptides are also more consistent with those of isomer **2**. Thus, we conclude that the stereochemistry originally attributed to the natural product should be corrected.

As discussed for **VII** and **VIII**,[9] the values of the coupling constants are consistent with a planar ring.[26] AM1 semiempirical calculations on the most stable conformations of **1** and **2** indeed indicated that the ring was almost planar ($\alpha_{cis} = 175.5^\circ$ and $\alpha_{trans} = 178.7^\circ$) (Figure 1). The calculated distance between the two homoallylic hydrogens (4.05 Å in the *cis* compound and 4.75 Å in the *trans*) explains why no NOEs were observed.



1 $\alpha = 178.7°$ **2** $\alpha = 175.5°$

Figure 1. Modeling of compounds **1** and **2** by AM1 semiempirical methods. The dihedral angle α is defined by the intersection of the (C1, C2, C3, C4) and the (C1, C6, C5, C4) planes.

Experimental Section

General procedures: Solvents were dried by distillation under argon over CaH_2 (CH_2Cl_2, toluene, *ortho*-xylene, NEt_3), Mg (MeOH) or Na/benzophenone (THF, Et_2O). All other commercially available reagents were used without further purification. Column chromatography was performed with flash silica (Merck 230, 0.040–0.063 mm). ^1H NMR (400 MHz) and ^{13}C NMR (100 MHz) were recorded on a Bruker ARX400 at room temperature for $CDCl_3$ solutions unless otherwise stated (some spectra were recorded on a Bruker AC200 or a DMX500). All chemical shifts are reported as δ values (ppm) relative to $CDCl_3$ (or CD_3OD): $\delta = 7.28$ (3.34) and $\delta = 77.16$ (49.86) for ^1H NMR and ^{13}C NMR spectra, respectively. IR spectra were recorded on a Perkin–Elmer 1420 instrument. CI mass spectra were obtained with a NERMAG R30–10 apparatus. High-resolution mass spectra were recorded on a JEOL MS700BE (CH_4). MS-MS spectra were recorded on a Micromass Quattro1 instrument with ESI source. Melting points were measured with a Kofler bank and are uncorrected. Elemental analyses were performed by the Service Régional de Microanalyse (SIAR-Jussieu).

Determination of the ^5J coupling constant values: The ^5J coupling constants between H-1′ and H-4′ in compounds **1**, **2**, **20** and **45** were measured from the 4′ proton signal after irradiation of the 3′ protons. The experiments were carried out on a Bruker ARX400 apparatus at 400 MHz in D_2O. For **2** and **20**, ^5J was 8.1 Hz and for **1** and **45**, 5.6 Hz.

4-*tert*-Butyldimethylsilyloxybutan-1-ol (4): Butane-1,4-diol (9.0 g, 100 mmol) was added to a vigorously stirred dispersion of NaH in mineral oil (80%, 3.15 g) in dry THF (200 mL). After being stirred for 1 h, the mixture was cooled in an ice bath, and *tert*-butyldimethylsilyl chloride

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(16.5 g, 109 mmol) was added over 15 min. Stirring was continued at RT for another hour, after which the reaction mixture was diluted with diethyl ether (500 mL) and washed with saturated Na_2CO_3 solution (2×100 mL), followed by brine (2×100 mL). The organic layer was dried over $MgSO_4$ and concentrated to afford a yellow oil that was distilled (b.p. 57 °C, 0.07 mbar). Monosilylated diol **4** was obtained as a colourless oil. Yield: 19.7 g, 97 %; ^{1}H NMR: $\delta = 0.06$ (s, 6H; $SiCH_3$), 0.90 (s, 9H; *t*Bu), 1.64 (m, 4H; H-2, H-3), 3.63–3.66 (m, 4H; H-1, H-4); ^{13}C NMR: $\delta = -5.31$ ($SiCH_3$), 18.39 ($C(CH_3)_3$), 25.99 ($C(CH_3)_3$), 29.95, 30.28 (C-2, C-3), 62.81, 63.44 (C-1, C-4); elemental analysis calcd for $C_{10}H_{24}O_2Si$: C 58.83, H 11.76; found C 58.64, H 11.96.

4-*tert*-Butyldimethylsilyloxybutanal (5): A stirred solution of oxalyl dichloride (8.8 mL, 100 mmol) in anhydrous dichloromethane (200 mL) was cooled to −60 °C, and anhydrous dimethyl sulfoxide (14.3 mL, 201 mmol) was added dropwise. After 10 min, alcohol **4** (17.1 g, 84 mmol) was added, and stirring was maintained for 30 min, during which the solution was allowed to warm to −20 °C. Addition of triethylamine (54 mL, 390 mmol) then produced a white precipitate and, after an additional 20 min stirring at this temperature, the mixture was diluted with diethyl ether (200 mL) and washed with a saturated NH_4Cl solution (100 mL). The organic layer was diluted with cyclohexane (1 L), washed with a saturated Na_2CO_3 solution (100 mL) followed by brine (3×150 mL) and dried over $MgSO_4$. Solvents were removed to deliver the desired aldehyde **5** smoothly. The crude product was used without purification for the next step. Yield: 15.9 g, 94 %; ^{1}H NMR (200 MHz): $\delta = 0.01$ (s, 6H; $SiCH_3$), 0.85 (s, 9H; *t*Bu), 1.83 (tt, $J = 7.1$, 6.5 Hz, 2H; H-3), 2.48 (td, $J = 7.1$, 1.6 Hz, 2H; H-2), 3.62 (t, $J = 6.5$ Hz, 2H; H-4), 9.76 (t, $J = 1.6$ Hz, 1H; H-1); ^{13}C NMR (50 MHz): $\delta = -5.37$ ($SiCH_3$), 18.34 ($C(CH_3)_3$), 25.53 (C-3), 25.95 ($C(CH_3)_3$), 40.84 (C-2), 62.12 (C-4), 202.70 (C-1).

Ethyl (2*E*)-6-*tert*-butyldimethylsilyloxyhex-2-enoate (6): Triethyl phosphonoacetate (23.5 mL, 117 mmol) was added dropwise, with vigorous stirring, to a dispersion of NaH in mineral oil (80 %, 3.52 g, 117 mmol) in dry THF (100 mL) cooled to −20 °C. After the reaction mixture had been stirred at this temperature for 30 min, aldehyde **5** (15.8 g, 78 mmol) was added, and the mixture was kept for 20 min at −20 °C and then for 30 min at RT. The solution was diluted in diethyl ether (500 mL), and the mixture was washed with a saturated NH_4Cl solution. The organic layer was washed with a saturated Na_2CO_3 solution (2×100 mL) followed by brine (2×100 mL) and dried over $MgSO_4$. After concentration and flash chromatography (cyclohexane/ethyl acetate 98:2), **6** was obtained as a colourless oil. Yield: 17 g, 80 %; ^{1}H NMR (200 MHz): $\delta = 0.00$ (s, 6H; $SiCH_3$), 0.84 (s, 9H; *t*Bu), 1.23 (t, $J = 7.1$ Hz, 3H; OCH_2CH_3), 1.62 (tt, $J = 6.8$, 6.8 Hz, 2H; H-5), 2.23 (tdd, $J = 7.3$, 7.3, 1.5 Hz, 2H; H-4), 3.58 (t, $J = 6.1$ Hz, 2H; H-6), 4.13 (q, $J = 7.1$ Hz, 2H; OCH_2CH_3), 5.78 (td, $J = 15.6$, 1.5 Hz, 1H; H-2), 6.94 (td, $J = 15.7$, 6.9 Hz, 1H; H-3); ^{13}C NMR: $\delta = -5.34$ ($SiCH_3$), 14.28 (OCH_2CH_3), 18.38 ($C(CH_3)_3$), 25.92 ($C(CH_3)_3$), 28.67 (C-4), 31.12 (C-5), 60.09 (OCH_2CH_3), 62.14 (C-6), 121.48 (C-2), 148.92 (C-3), 166.64 (C-1); elemental analysis calcd for $C_{14}H_{28}O_3Si$: C 61.78, H 10.29; found C 61.77, H 10.23.

(2*E*)-6-*tert*-Butyldimethylsilyloxyhex-2-en-1-ol (7): Diisobutylaluminium hydride (1M in hexane, 129 mL) was added at −60 °C to a solution of ester **6** (16 g, 59 mmol) in dry dichloromethane. After the mixture had been stirred for 20 min at this temperature, the reaction was quenched with methanol (50 mL). Aluminium salts were slowly hydrolysed with a hydrochloric acid solution (1M). After dilution with diethyl ether (1 L) and water (200 mL), the aqueous layer was extracted with diethyl ether (2×300 mL). The combined organic layers were dried over $MgSO_4$ and concentrated to give the desired alcohol **7** as a colourless oil after chromatography (cyclohexane/ethyl acetate 8:2). Yield: 11.34 g, 84 %; ^{1}H NMR (200 MHz): $\delta = 0.00$ (s, 6H; $SiCH_3$), 0.84 (s, 9H; *t*Bu), 1.55 (tt, $J = 6.9$, 6.9 Hz, 2H; H-5), 2.05 (td, $J = 6.9$, 6.9 Hz, 2H; H-4), 2.23 (m, 1H; OH), 3.56 (t, $J = 6.4$ Hz, 2H; H-6), 4.01 (d, $J = 3.5$ Hz, 2H; H-1), 5.50–5.72 (m, 2H; H-2, H-3); ^{13}C NMR: $\delta = -5.28$ ($SiCH_3$), 18.32 ($C(CH_3)_3$), 25.95 ($C(CH_3)_3$), 28.51 (C-4), 32.21 (C-5), 62.52 (C-6), 63.51 (C-1), 129.29–132.53 (C-2, C-3); elemental analysis calcd for $C_{12}H_{26}O_2Si$: C 62.62, H 11.30; found C 62.57, H 11.44.

(2*E*)-6-*tert*-Butyldimethylsilyloxyhex-2-enal (8):

*Preparation from **7***: Compound **8** was prepared as described for **5**, from **7** (11.3 g, 49 mmol), oxalyl dichloride (5.2 mL, 59 mmol), dimethyl sulfoxide (8.4 mL, 118 mmol) and triethylamine (33 mL, 237 mmol). Yield: 10.5 g, 94 %.

*Preparation from **5***: *Acetaldehyde tert-butylimine*: Acetaldehyde (20 mL, 360 mmol) was introduced dropwise at 0 °C into a flask containing *tert*-butylamine (38 mL, 360 mmol). The mixture was stirred at this temperature for 3 h, then solid KOH (800 mg, 14 mmol) was added. After 12 h at 4 °C, the aqueous layer was removed, and the organic layer was distilled (bp 66–67 °C) to furnish the desired imine as a colourless oil. Yield: 23 g, 66 %; ^{1}H NMR (200 MHz): $\delta = 1.11$ (s, 9H; *t*Bu), 1.90 (d, 3H; $J = 4.8$ Hz, CH_3), 7.62 (q, 1H; $J = 4.8$ Hz, *CH*=); ^{13}C NMR (50MHz): $\delta = 22.77$ (CH_3), 29.61 ($C(CH_3)_3$), 56.66 ($C(CH_3)_3$), 155.00 (CH=).

Acetaldehyde *tert*-butylimine (3 g, 30 mmol) was added at −78 °C to a solution of lithium diisopropylamide (30 mL, 2M in heptane) in anhydrous THF (60 mL). The mixture was stirred at this temperature for 30 min, and diethyl chlorophosphate (4.4 mL, 30 mmol) was added. The mixture was then allowed to warm to −10 °C over 1 h, was stirred for 2 h at this temperature and then for 5 min at −78 °C. Aldehyde **5** was added, and the reaction mixture was allowed to warm to RT over 2 h. The reaction was quenched by addition of a saturated oxalic acid solution (50 mL), and the mixture was diluted with toluene (50 mL). The organic layer was extracted with diethyl ether (3×50 mL) and concentrated to furnish an orange oil, which was flash chromatographed (cyclohexane/ethyl acetate 49:1–41:9). α,β-Unsaturated aldehyde **8** was obtained as an orange oil. Yield: 3.06 g, 67 %; ^{1}H NMR (200 MHz): $\delta = 0.00$ (s, 6H; $SiCH_3$), 0.85 (s, 9H; *t*Bu), 1.68 (tt, $J = 6.7$, 6.7 Hz, 2H; H-5), 2.38 (tdd, $J = 7.2$, 7.2, 1.4 Hz, 2H; H-4), 3.61 (t, $J = 6.0$ Hz, 2H; H-6), 6.08 (tdd, $J = 15.6$, 7.9, 1.5 Hz, 1H; H-2), 6.85 (td, $J = 15.6$, 6.7 Hz, 1H; H-3), 9.46 (d, $J = 7.9$ Hz, 1H; H-1); ^{13}C NMR (50 MHz): $\delta = -5.34$ ($SiCH_3$), 18.28 ($C(CH_3)_3$), 25.92 ($C(CH_3)_3$), 29.37 (C-4), 30.89 (C-5), 62.04 (C-6), 133.04 (C-2), 158.76 (C-3), 194.05 (C-1).

Ethyl (2*E*,4*E*)-8-*tert*-butyldimethylsilyloxyocta-2,4-dienoate (10): Compound **10** was prepared as described for compound **6**, from aldehyde **8** (3.06 g, 13.4 mmol), NaH (80 %, 0.7 g) and triethyl phosphonoacetate (4.7 mL, 23 mmol). It was obtained after chromatography (cyclohexane/ethyl acetate 49:1) as a yellow oil. Yield: 2.82 g, 70 %; ^{1}H NMR: $\delta = 0.04$ (s, 6H; $SiCH_3$), 0.89 (s, 9H; *t*Bu), 1.29 (t, $J = 7.1$ Hz, 3H; CH_2CH_3), 1.64 (tt, $J = 6.8$, 6.8 Hz, 2H; H-7), 2.24 (td, $J = 6.9$, 6.9 Hz, 2H; H-6), 3.61 (t, $J = 6.1$ Hz, 2H; H-8), 4.19 (q, $J = 7.1$ Hz, 2H; CH_2CH_3), 5.78 (d, $J = 15.3$ Hz, 1H; H-2), 6.09–6.22 (m, 2H; H-4, H-5), 7.25 (dd, $J = 15.2$, 9.7 Hz, 1H; H-3); ^{13}C NMR: $\delta = -5.30$ ($Si{-}CH_3$), 14.33 (CH_2CH_3), 18.34 ($C(CH_3)_3$), 25.96 ($C(CH_3)_3$), 29.38 (C-6), 31.76 (C-7), 60.16 (CH_2CH_3), 62.24 (C-8), 119.36 (C-2), 128.66, 144.01 (C-4, C-5), 144.95 (C-3), 167.27 (C-1); elemental analysis calcd for $C_{16}H_{30}O_3Si$: C 64.44, H 10.06; found C 64.45, H 10.17.

Ethyl (2*E*,4*E*)-8-hydroxyocta-2,4-dienoate (11): Protected alcohol **10** (5 g, 16.8 mmol) dissolved in a THF/water/acetic acid (1:1:3; 75 mL) was stirred for 4 h at RT. The solvents were evaporated, and the crude product was purified by chromatography (cyclohexane/ethyl acetate 9:1–1:1) to give **11** as a colourless oil. Yield: 2.21 g, 71 %; ^{1}H NMR (200 MHz): $\delta = 1.20$ (t, $J = 7.1$ Hz, 3H; CH_3), 1.61 (tt, $J = 7.0$, 7.0 Hz, 2H; H-7), 2.18 (td, $J = 6.8$, 6.8 Hz, 2H; H-6), 2.65 (m, 1H; OH), 3.55 (t, $J = 6.3$ Hz, 2H; H-8), 4.10 (q, $J = 7.1$ Hz, 2H; CH_2CH_3), 5.70 (d, $J = 15.4$ Hz, 1H; H-2), 5.97–6.20 (m, 2H; H-4, H-5), 7.17 (dd, $J = 15.3$, 9.9 Hz, 1H; H-3); ^{13}C NMR (50 MHz) : 14.19 (CH_3), 29.22 (C-6), 31.47 (C-7), 60.21 (CH_2CH_3), 61.72 (C-8), 119.31 (C-2), 128.65, 143.74 (C-4, C-5), 144.90 (C-3), 167.37 (C-1); MS: *m/z*: 185 $[M+H]^+$, 202 $[M+NH_4]^+$.

(2*E*,4*E*)-8-Hydroxyocta-2,4-dienoic acid (12): A sodium hydroxide solution (0.8M, 60 mL) was added to a solution of ester **11** (2.21 g, 12 mmol) in methanol (30 mL), and the mixture was stirred for 50 min at 60 °C. Neutralisation with hydrochloric acid (12N) and evaporation of the solvents gave a white solid; this was then flash chromatographed (dichloromethane/ethanol/acetic acid 9:1:0.1). Acid **12** was obtained as a white solid. Yield: 1.56 g, 83 %; ^{1}H NMR (200 MHz, CD_3OD): $\delta = 1.64$ (tt, $J = 6.9$, 6.9 Hz, 2H; H-7), 2.24 (td, $J = 6.8$, 6.8 Hz, 2H; H-6), 3.56 (t, $J = 6.5$ Hz, 2H; H-8), 5.78 (d, $J = 15.3$ Hz, 1H; H-2), 6.08–6.32 (m, 2H; H-4, H-5), 7.24 (dd, $J = 15.3$, 9.8 Hz, 1H; H-3); ^{13}C NMR (50 MHz, CD_3OD) : 30.20 (C-6), 32.44 (C-7), 62.10 (C-8), 120.47 (C-2), 129.84, 145.18 (C-4, C-5), 146.84 (C-3), 170.79 (C-1); elemental analysis calcd for $C_8H_{12}O_3$: C 61.57, H 7.69; found C 61.50, H 7.73.

Allyl (1*E*,3*E*)-7-hydroxy-hepta-1,3-dienyl-carbamate (13): Diphenylphosphoryl azide (5.2 mL, 24 mmol) and triethylamine (3.3 mL, 24 mmol) were

added to a solution of acid **12** (2.66 mg, 17 mmol) in allyl alcohol (80 mL). The mixture was stirred under reflux for 2.5 h, and after cooling to RT, the solvents were evaporated. The resulting oil was purified by flash chromatography (cyclohexane/ethyl acetate 8:2 – 4:6) to afford the desired diene **13** as a yellow oil. Yield: 2.51 g, 70 %; ^{1}H NMR: $\delta = 1.62$ (tt, $J = 7.0$, 7.0 Hz, 2H; H-6), 1.98 (m, 1H; OH), 2.12 (td, $J = 7.0$, 7.0 Hz, 2H; H-5), 3.61 (t, $J = 6.4$ Hz, 2H; H-7), 4.58 (d, $J = 5.4$ Hz, 2H; COOCH_2), 5.20 (d, $J = 10.4$ Hz, 1H; =CH_2), 5.28 (d, $J = 17.3$ Hz, 1H; =CH_2), 5.47 (td, $J = 14.7$, 7.3 Hz, 1H; H-4), 5.64 (dd, $J = 10.8$, 13.7 Hz, 1H; H-2), 5.83 – 5.90 (m, 1H; CH=CH_2), 5.95 (dd, $J = 15.0$, 10.6 Hz, 1H; H-3), 6.59 (dd, $J = 13.8$, 10.6 Hz, 1H; H-1), 6.84 (d, $J = 10.6$ Hz, 1H; NH); ^{13}C NMR: $\delta = 29.02$ (C-5), 32.33 (C-6), 62.30 (C-7), 66.08 (COOCH_2), 111.72 (C-2), 118.25 (=CH_2), 124.94 (C-1), 127.96 (C-3), 130.15 (C-4), 132.28 (CH=CH_2), 153.44 (C=O).

3.5 % Sodium amalgam:[27] Clean sodium (27 g) was placed in a 500 mL round-bottomed flask, fitted with a dropping funnel containing 750 g of mercury in the central sockets. The air was displaced by dry nitrogen between two side sockets. Mercury was added slowly enough to control the temperature of the exothermic reaction. After cooling the mixture to RT, a solid was obtained and powdered.

Allyl [4*c*-(3-hydroxypropyl)cyclohexa-2,5-dien-1*r*-yl]-carbamate (15): A solution of diene **13** (880 mg, 4.2 mmol) in anhydrous *ortho*-xylene (5 mL) was added to *trans*-1,2-bis(phenylsulfonyl)ethylene (**14**; 1.28 g, 4.2 mmol). The mixture was heated at reflux overnight. After evaporation of the solvent, the residual oil was diluted in anhydrous methanol (150 mL) buffered with KH_2PO_4 (17.2 g, 126 mmol). The solution was vigorously stirred with 3.5 % sodium amalgam (2.15 g, 3.04 mmol) under argon for 3 h at RT. The salts and mercury were then removed by filtration and washed with dichloromethane (100 mL). The organic layer was concentrated and purified by flash chromatography (cyclohexane/ethyl acetate 9:1 – 4:6) to furnish the cyclohexa-1,4-diene **15**. Yield: 363 mg, 37 %; ^{1}H NMR: $\delta =$ 1.46 – 1.58 (m, 4H; H-1′, H-2′), 2.06 (m, 1H; OH), 2.70 – 2.72 (m, 1H; H-4), 3.59 (t, $J = 6.0$ Hz, 2H; H-3′), 4.53 (d, $J = 5.5$ Hz, 2H; COOCH_2), 4.68 (m, 1H; H-1), 4.79 (d, $J = 8.8$ Hz, 1H; NH), 5.18 (d, $J = 10.5$ Hz, 1H; =CH_2), 5.27 (dd, $J = 17.2$, 1.2 Hz, 1H; =CH_2), 5.67 – 5.77 (m, 4H; H-2, H-3, H-5, H-6), 5.80 – 5.93 (m, 1H; CH=CH_2); ^{13}C NMR (50 MHz): $\delta = 29.18$, 30.70 (C-1′, C-2′), 34.74 (C-4), 44.86 (C-1), 62.75 (C-3′), 65.58 (COOCH_2), 117.72 (=CH_2), 125.63 (C-2, C-6), 131.47 (C-3, C-5), 132.82 (CH=CH_2), 155.62 (C=O); MS: *m/z*: 238 $[M+H]^+$, 255 $[M+NH_4]^+$.

Allyl [4*c*-(3-oxopropyl)-cyclohexa-2,5-dien-1*r*-yl]-carbamate (16): Alcohol **15** (418 mg, 1.8 mmol) was converted into the aldehyde **16** by means of a Swern reaction as described for **5**. Yield: 393 mg, 95 %; ^{1}H NMR (200 MHz) : 1.81 (td, $J = 6.8$, 6.8 Hz, 2H; H-1′), 2.37 (td, $J = 7.5$, 1.6 Hz, 2H; H-2′), 2.80 – 2.83 (m, 1H; H-4), 4.53 (d, $J = 5.6$ Hz, 2H; COOCH_2), 4.58 – 4.72 (m, 1H; H-1), 4.85 (d, $J = 9.0$ Hz, 1H; NH), 5.17 (ddt, $J = 10.4$, 1.3, 1.3 Hz, 1H; =CH_2), 5.27 (ddt, $J = 17.2$, 1.5, 1.5 Hz, 1H; =CH_2), 5.64 – 5.78 (m, 4H; H-2, H-3, H-5, H-6), 5.79 – 5.98 (m, 1H; CH=CH_2), 9.70 (t, $J =$ 1.7 Hz, 1H; H-3′); ^{13}C NMR (50 MHz): $\delta = 25.88$ (C-1′), 34.07 (C-4), 40.26 (C-2′), 44.69 (C-1), 65.60 (COOCH_2), 117.60 (=CH_2), 126.80 (C-2, C-6), 130.35 (C-3, C-5), 132.83 (CH=CH_2), 155.58 (N–C=O), 202.54 (C-3′); MS: *m/z*: 236 $[M+H]^+$, 253 $[M+NH_4]^+$.

Allyl [4*c*-(3-Amino-3-cyano-propyl)-cyclohexa-2,5-dien-1*r*-yl]-carbamate (17): Aldehyde **16** (423 mg, 1.8 mmol) and trimethylsilyl cyanide (300 μL, 2.25 mmol) in the presence of a catalytic amount of ZnI_2 were stirred for 15 min at RT. A saturated solution of ammonia in methanol (2 mL) at – 40 °C was added, and the mixture was heated at 50 °C for 15 min. After solvent removal and chromatography (cyclohexane/ethyl acetate 9:1 – 0:1), amino nitrile **17** was obtained as a yellow powder. Yield: 346 mg, 74 %; ^{1}H NMR: $\delta = 1.60 – 1.68$ (m, 4H; H-1′, H-2′), 1.71 (m, 2H; NH_2), 2.76 – 2.77 (m, 1H; H-4), 3.62 (t, $J = 6.3$ Hz, 1H; H-3′), 4.51 (d, $J = 5.4$ Hz, 2H; COOCH_2), 4.65 – 4.69 (m, 1H; H-1), 4.88 (d, $J = 9.0$ Hz, 1H; NH), 5.16 (d, $J = 10.4$ Hz, 1H; =CH_2), 5.25 (dd, $J = 17.2$, 1.4 Hz, 1H; =CH_2), 5.68 – 5.74 (m, 4H; H-2, H-3, H-5, H-6), 5.81 – 5.91 (m, 1H; CH=CH_2); ^{13}C NMR (50 MHz): $\delta = 29.86$, 31.59 (C-1′, C-2′), 34.13 (C-4), 43.38 (C-3′), 44.65 (C-1), 65.49 (COOCH_2), 117.63 (=CH_2), 121.94 (CN), 126.45 (C-2, C-6), 130.38 (C-3, C-5), 132.74 (CH=CH_2), 155.48 (C=O); MS: *m/z*: 262 $[M+H]^+$, 279 $[M+NH_4]^+$.

The double condensation by-product **18** and the aromatic compound **19** may be isolated in sizeable yields if the ammonia concentration in the methanolic solution is insufficient or if the ammonia is not sufficiently pure. A single δ value is found for each proton in compound **18**, except for H-3′ for which two signals in a 55:45 ratio are observed. This product is probably a mixture of *meso* and (*RR*,*SS*) compounds. **18**: ^{1}H NMR: $\delta = 1.58 – 1.78$ (m, 8H; H-1′, H-2′), 2.81 (m, 2H; H-4), 3.59 – 3.63 (m, 1H; H-3′$_a$), 3.75 – 3.76 (m, 1H; H-3′$_b$), 4.53 (d, $J = 5.5$ Hz, 4H; COOCH_2), 4.68 – 4.70 (m, 2H; H-1), 4.89 – 4.95 (m, 2H; NH), 5.17 (d, $J = 10.5$ Hz, 2H; =CH_2), 5.26 (dd, $J = 17.2$, 1.2 Hz, 2H; =CH_2), 5.68 – 5.76 (m, 8H; H-2, H-3, H-5, H-6), 5.83 – 5.92 (m, 2H; CH=CH_2); ^{13}C NMR (50MHz): $\delta = 29.64$, 29.90 (C-1′, C-2′), 34.07 (C-4), 44.70 (C-1), 48.66 (C-3′$_a$), 49.23 (C-3′$_b$), 65.56 (COOCH_2), 117.65 (=CH_2), 118.91 (CN), 119.25 (CN), 126.78 (C-2, C-6), 130.17 (C-3, C-5), 132.74 (CH=CH_2), 155.56 (C=O); MS (CI): *m/z*: 506 $[M+H]^+$, 479 $[M+H – HCN]^+$. **19**: ^{1}H NMR: $\delta = 1.65$ (m, 2H; NH_2), 2.08 (dt, $J =$ 7.5, 7.5 Hz, 2H; H-3), 2.79 – 2.94 (m, 2H; H-4), 3.63 (t, $J = 7.2$ Hz, 1H; H-2), 7.22 – 7.35 (m, 5H; Ph); ^{13}C NMR: $\delta = 31.60$ (C-4), 36.83 (C-3), 42.62 (C-2), 122.07 (CN), 126.52 – 139.81 (Ph); MS: *m/z*: 161 $[M+H]^+$, 178 $[M+NH_4]^+$, 134 $[M+H – HCN]^+$.

2-Amino-4-(4*c*-aminocyclohexa-2,5-dien-1*r*-yl)-butanamide hydrochloride (20): Phenylsilane (110 μL, 0.9 mmol) and a solution of $Pd(Ph_3)_4$ (10 mg, 0.009 mmol) in dry dichloromethane (1 mL) were added under argon to a solution of aminonitrile **17** (116 mg, 0.4 mmol) in dry dichloromethane (1 mL). The mixture was stirred for 30 min at RT and extracted with a hydrochloric acid solution (1N) at 0 °C (5 × 1 mL). The combined aqueous layers were lyophilised to afford a mixture of **20** (60 %) and **21** (22 %) as a yellow oil. Yield: 82 mg. **20**: ^{1}H NMR (200 MHz, CD_3OD): $\delta = 1.72$ (dt, $J =$ 5.6, 5.6 Hz, 2H; H-3), 1.95 (td, $J = 5.6$, 5.6 Hz, 2H; H-4), 2.90 (m, 1H; H-1′), 3.98 (m, 1H; H-2), 4.33 (m, 1H; H-4′), 5.87 – 5.91 (m, 2H; H-3′, H-5′), 6.10 – 6.15 (m, 2H; H-2′, H-6′); ^{13}C NMR (50 MHz, CD_3OD): $\delta = 30.02$ (C-3), 29.35 (C-4), 36.02 (C-1′), 54.16 (C-2), 46.10 (C-4′), 122.49 (C-3′, C-5′), 135.71 (C-2′, C-6′), 172.21 (C-1); MS: *m/z* (%): 196 $[M+H]^+$, 179 (30)$[Ph(CH_2)_2CH(NH_2)CONH_2+H]^+$, 134 (100) $[Ph(CH_2)_2CH=NH_2]^+$, 117 (7) $[PhCH_2CH=CH]^+$, 91 (35) $[PhCH_2]^+$; MS-MS: *m/z* (%) 179 (100), 134 (75) $[Ph(CH_2)_2CH=NH_2]^+$, 117 (10) $[PhCH_2CH=CH]^+$, 91 (60) $[PhCH_2]^+$.

2-Amino-4-phenyl-butanenitrile (19): A solution of 3-phenylpropanal (5.07 g, 37.8 mmol) and trimethylsilyl cyanide (6 mL, 46.6 mmol) in the presence of a catalytic amount of ZnI_2 was stirred at RT for 15 min. Then, a saturated solution of ammonia in methanol (38 mL) was added, and stirring was maintained for 3 h at 40 °C. The crude product was concentrated and chromatographed (cyclohexane/ethyl acetate 9:1 – 1:9) to give amino nitrile **19** as a yellow oil. Yield: 4.39 g, 73 %; ^{1}H and ^{13}C NMR: see above. MS: *m/z*: 161 $[M+H]^+$, 178 $[M+NH_4]^+$, 134 $[M+H – HCN]^+$.

2-Amino-4-phenyl-butanamide (22): A sodium hydroxide solution (1N, 27 mL) and a hydrogen peroxide solution (35 %, 5 mL) were added to a solution of nitrile **19** (2.1 g, 13.15 mmol) in methanol (14 mL). After 1 h of stirring at RT, the mixture was extracted with dichloromethane, and the organic layer was dried over $MgSO_4$ to give **22** as yellow crystals. Yield: 1.78 g (76 %); m.p. 89 °C; ^{1}H NMR: $\delta = 1.57$ (m, 2H; NH_2), 1.80 – 1.90 (m, 1H; H-3), 2.16 – 2.25 (m, 1H; H-3), 2.70 – 2.83 (m, 2H; H-4), 3.40 (m, 1H; H-2), 5.84 (m, 1H; $CONH_2$), 7.07 (m, 1H; $CONH_2$), 7.19 – 7.32 (m, 5H; Ph); ^{13}C NMR: $\delta = 32.19$ (C-4), 36.71 (C-3), 54.79 (C-2), 126.21 – 141.21 (Ph), 178.16 (C-1); elemental analysis calcd for $C_{10}H_{14}ON_2$: C 67.39, H 7.91, N 15.72; found C 67.36, H 7.81, N 15.69.

5-Phenethyl-imidazolidine-2,4-dione (24): Amino nitrile **19** (2.62 g, 14.15 mmol) and ammonium carbonate (15.6 g, 198 mmol) in a methanol/water 1:1 solution were stirred for 6 h at 50 °C. After concentration and recrystallisation from water, hydantoin **24** was obtained as white crystals. Yield: 1.78 g, 62 %; m.p. 166 °C; ^{1}H NMR (CD_3OD): $\delta = 1.91 – 2.00$ (m, 1H; CH_2CH_2Ph), 2.06 – 2.15 (m, 1H; CH_2CH_2Ph), 2.69 – 2.76 (m, 2H; CH_2Ph), 4.08 (dd, $J = 7.2$, 4.2 Hz, 1H; N-CH), 7.17 – 7.31 (m, 5H; Ph); ^{13}C NMR (CD_3OD): $\delta = 28.41$ (CH_2Ph), 31.30 (CH_2CH_2Ph), 55.76 (CH–N), 124.69, 126.93, 127.01, 139.46 (Ph), 157.43, 175.55 (C=O); elemental analysis calcd for $C_{11}H_{12}O_2N_2$: C 64.69, H 5.92, N 13.72; found C 65.10, H 5.85, N 13.68.

***N*,*N*′-Bis-*tert*-butyloxycarbonyl-5-phenethyl-imidazolidine-2,4-dione (25)**: Di-*tert*-butyldicarbonate (806 mg, 3.67 mmol) was added to a solution of hydantoin **24** (250 mg, 1.23 mmol) in THF (20 mL) in the presence of a catalytic amount of DMAP, and the mixture was stirred under argon at RT for 45 min. The crude product was concentrated and filtered on silica gel (cyclohexane/ethyl acetate 7:3) to give a yellow oil. Recrystallisation from pentane afforded **25** as white crystals. Yield: 323 mg, 65 %; m.p. 97 °C; ^{1}H NMR: $\delta = 1.53$ (s, 9H; *t*Bu), 1.56 (s, 9H; *t*Bu), 2.34 – 2.41 (m, 2H; CH_2Ph), 2.63 – 2.67 (m, 1H; CH_2CH_2Ph), 2.71 – 2.76 (m, 1H; CH_2CH_2Ph),

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4.46 (dd, $J = 6.6$, 3.3 Hz, 1H; N-CH), 7.16–7.29 (m, 5H; Ph); ^{13}C NMR (50 MHz): $\delta = 27.76$, 27.99 (C(CH_3)$_3$), 29.70, 31.60 (CH_2CH_2Ph), 58.51 (N–CH), 84.83–86.53 ($C(CH_3)_3$), 126.50–139.70 (Ph), 145.10, 147.60, 148.20, 167.30 (C=O); elemental analysis calcd for $C_{21}H_{28}O_6N_2$: C 62.36, H 6.98, N 6.93; found C 62.30, H 7.02, N 7.03.

2-(1,3-di-*tert*-butyloxycarbonyl)urea-4-phenyl-butanoic acid (26): A solution of *N*-protected hydantoin **25** (0.49 mmol, 197 mg) in THF (10 mL) and water (2 mL) was stirred in the presence of lithium hydroxide monohydrate (41 mg, 0.97 mmol) for 2 h at RT. After concentration, dilution in water (3 mL) and acidification with hydrochloric acid (1N, pH 6), the solution was filtered to afford **26** as a white powder. Yield: 189 mg, 92%; m.p. 152 °C; ^{1}H NMR: $\delta = 1.48$ (s, 9H; *t*Bu), 1.52 (s, 9H; *t*Bu), 2.17–2.22 (m, 1H; CH_2CH_2Ph), 2.74–2.78 (m, 1H; CH_2CH_2Ph), 2.51–2.62 (m, 2H; CH_2Ph), 5.40–5.50 (m, 1H; N-CH), 7.18–7.30 (m, 5H; Ph), 10.28 (m, 1H; NH), 10.97 (m, 1H; COOH); ^{13}C NMR: $\delta = 28.24$, 28.43 (C(CH_3)$_3$), 31.65 (CH_2CH_2Ph), 32.90 (CH_2Ph), 56.00 (CH–N), 82.59–86.41 ($C(CH_3)_3$), 126.15, 128.36, 128.57, 141.30 (Ph), 150.16, 151.18, 154.03 (C=O), 175.79 (COOH); MS: *m/z*: 423 $[M+H]^+$, 440 $[M+NH_4]^+$.

Synthesis of the polyacrylamide resin:

1-acryloylpiperidin-4-one: 4-Piperidone monohydrate hydrochloride (15 g, 98 mmol) was added to $NaHCO_3$ (24.7 g, 290 mmol) in water (75 mL) cooled to 0 °C. Acryloyl chloride (9.4 mL, 120 mmol) was added dropwise at this temperature, with efficient mechanical stirring, over 30 min. The solution was kept at 0 °C for 1 h and further stirred for 1 h 15 min at RT. Excess $NaHCO_3$ was removed by filtration, and acryloylpiperidone was extracted with chloroform (5×100 mL). The organic layer was washed with 2M hydrochloric acid (2×20 mL) and dried over $MgSO_4$. After concentration, pure 1-acryloylpiperidin-4-one was obtained. Yield: 13.28 g, 65%; ^{1}H NMR: $\delta = 2.37$ (t, $J = 6.3$ Hz, 4H; NCH_2CH_2), 3.75–3.80 (m, 4H; NCH_2CH_2), 5.63 (dd, $J = 10.5$, 1.8 Hz, 1H; =CH_2), 6.20 (dd, $J = 16.8$, 1.8 Hz, 1H; =CH_2), 6.54 (dd, $J = 16.8$, 10.5 Hz, 1H; CH=CH_2); ^{13}C NMR: $\delta =$ 41.42, 41.47, 41.65, 44.06 (NCH_2CH_2), 127.38 (CH=CH_2), 129.26 (=CH_2), 166.05 (N–C=O), 206.95 (C=O).

1,4-Bisacryloyl-piperazine: Acryloyl chloride (8.1 mL, 100 mmol) was added dropwise over 30 min to a stirred mixture of piperazine (4.3 g, 50 mmol) and triethylamine (14 mL, 100 mmol) in dry dichloromethane (350 mL) cooled to 0 °C. After 45 min of stirring at this temperature, the mixture was allowed to warm to RT over 1 h. The organic layer was washed with water (2×100 mL) and with hydrochloric acid (0.5M, 2×40 mL). After concentration, 1,4-bisacryloyl-piperazine was crystallised from methanol/diethyl ether (15:85), to give of white crystals. Yield: 5.55 g, 57%; m.p. 95–96 °C; ^{1}H NMR: $\delta = 3.60$ (s, 4H; NCH_2), 3.71 (s, 4H; NCH_2), 5.75 (dd, $J = 10.7$, 1.5 Hz, 2H; =CH_2), 6.33 (dd, $J = 16.7$, 1.5 Hz, 2H; =CH_2), 6.57 (dd, $J = 10.7$, 16.6 Hz, 2H; CH=CH_2); ^{13}C NMR: $\delta = 42.32$, 45.78 (NCH_2), 127.36 (CH=CH_2), 129.15 (=CH_2), 165.95 (C=O).

Immobilisation of pronase on poly(N-acryloylpiperidin-4-one): A solution of 1-acryloylpiperidin-4-one (5 g, 24 mmol), 1,4-bisacryloyl-piperazine (1 g, 5 mmol) and pronase (100 mg) in water (6 mL) was deoxygenated by bubbling argon through it for 1 h. *N,N,N′,N′*-tetramethyl ethylene diamine (40 mL) and a deoxygenated ammonium persulfate solution (0,175M, 1 mL) were added quickly. The flask was transferred to an ice bath and left there for 1 h, and then at RT for 1 h. The solid was recovered, crushed, and washed with water. The immobilised pronase can be stored at 4 °C.

(2R)-2-Amino-4-(4c-amino-cyclohexa-2,5-dien-1r-yl)-butanoic acid (2): Immobilised pronase (2 g) and (D,L)-amino acid amide **20** (in the presence of aromatic compound **21**) (94 mg, 0.35 mmol) were added to a phosphate buffer (0.1M, 20 mL, pH 9.6). After 7.5 h stirring at 37 °C, the mixture was filtered, and the resin was washed with water. The combined aqueous layers were acidified to pH 6 with acetic acid and then lyophilised and purified on a Dowex-$SO_3^-NH_4^+$ column (H_2O/NH_4OH (0.2M) 1:0–0:1) to give L-amino acid **2** (20 mg, 28%) contaminated with **21** (27%) and D-amino acid amide **20** (21 mg, 29%). A pure sample of **2** could be obtained after additional DOWEX chromatography; the enantiomeric excess was determined on this sample. ^{1}H NMR (500 MHz, D_2O): $\delta = 1.56–1.62$ (m, 1H; H-4), 1.65–1.71 (m, 1H; H-4), 1.84–1.91 (m, 2H; H-3), 2.96 (m, 1H; H-1′), 3.75 (t, $J = 6.0$ Hz, 1H; H-2), 4.41 (m, 1H; H-4′), 5.87 (d, $J = 10.4$ Hz, 2H; H-3′, H-5′), 6.13 (d, $J = 10.4$ Hz, 2H; H-2′, H-6′); ^{13}C NMR (125 MHz, D_2O): $\delta = 27.83$, 29.29 (C-3, C-4), 35.10 (C-1′), 55.50 (C-2), 45.56 (C-4′), 121.52 (C-3′, C-5′), 135.75 (C-2′, C-6′), 175.32 (C-1); MS-MS: *m/z* (%): 197 $[M+H]^+$, 180 (30) $[Ph(CH_2)_2CH(NH_2)CO_2H+H]^+$, 134 (60) $[Ph(CH_2)_2CH=NH_2]^+$, 117 (18) $[PhCH_2CH=CH]^+$, 91 (100) $[PhCH_2]^+$.

Derivatisation into compound 27: A solution of amino acid **2** in hydrochloric acid (3N, 1 mL) was heated at reflux for 2.5 h. After evaporation of solvent, a solution of 2N isopropanol in HCl was added, and the mixture was heated at 100 °C for 30 min under argon. The solvent was evaporated under draught, and CH_2Cl_2 (1 mL) followed by trifluoroacetic anhydride (10 drops) were added. After 30 min at RT, the solvent was evaporated, and **27** was injected onto a CHIRASYL-VAL 0.20 μ G.C. column (length 50 m, internal diameter 0.32 mm). The same procedure was used for amino acid amide **20** and for the racemic homophenylalanine reference.

5-*tert*-Butyldimethylsilyloxypentan-1-ol (28): Pentane-1,5-diol (27.1 g, 0.27 mol) was transformed into the colourless oil **28** as described for the preparation of **4**. Yield: 50.4 g, 89%; b.p. 122 °C (0.1 mbar); ^{1}H NMR: $\delta =$ 0.00 (s, 6H; $SiCH_3$), 0.85 (s, 9H; *t*Bu), 1.31–1.38 (m, 2H; H-3), 1.46–1.56 (m, 4H; H-2, H-4), 2.33 (s, 1H; OH), 3.54–3.58 (m, 4H; H-1, H-5); ^{13}C NMR: $\delta = -5.28$ ($SiCH_3$), 18.38 ($C(CH_3)_3$), 22.04 (C-3), 25.98 (C(CH_3)$_3$), 32.45–32.51 (C-2, C-4), 62.66–63.19 (C-1, C-5); elemental analysis calcd for $C_{11}H_{26}O_2Si$: C 60.56, H 11.92; found C 60.51, H 12.07.

5-*Tert*-butyldimethylsilyloxypentanal (29): Alcohol **28** (21.8 g, 100 mmol) was transformed into the yellow oil **29** as described for the preparation of **5**. Yield: 19.5 g, 90%; ^{1}H NMR (200 MHz): $\delta = -0.02$ (s, 6H; $SiCH_3$), 0.82 (s, 9H; *t*Bu), 1.40–1.57 (m, 2H; H-4), 1.57–1.68 (m, 2H; H-3), 2.39 (td, $J = 7.1$, 1.7 Hz, 2H; H-2), 3.56 (t, $J = 6.0$ Hz, 2H; H-5), 9.70 (t, $J = 1.8$ Hz, 1H; H-1); ^{13}C NMR (50 MHz): $\delta = -5.37$ ($SiCH_3$), 18.34 ($C(CH_3)_3$), 18.58 (C-3), 25.92 (C(CH_3)$_3$), 32.07 (C-4), 43.59 (C-2), 62.55 (C-5), 202.48 (C-1).

(2*E*)-7-*tert*-Butyldimethylsilyloxyhept-2-enal (30): α,β-Unsaturated aldehyde **30** was obtained as an orange oil from aldehyde **29** in one step as described for the preparation of **8**. Yield: 12.1 g, 56%; ^{1}H NMR (200 MHz): $\delta = 0.00$ (s, 6H; $SiCH_3$), 0.84 (s, 9H; *t*Bu), 1.49–1.58 (m, 4H; H-5, H-6), 2.27–2.33 (m, 2H; H-4), 3.58 (t, $J = 5.8$ Hz, 2H; H-7), 6.07 (tdd, $J = 15.5$, 7.9, 1.3 Hz, 1H; H-2), 6.81 (td, $J = 15.6$, 6.7 Hz, 1H; H-3), 9.45 (d, $J = 7.9$ Hz, 1H; H-1); ^{13}C NMR (50 MHz): $\delta = -5.34$ ($SiCH_3$), 18.31 ($C(CH_3)_3$), 24.22, 32.13 (C-5, C-6), 25.92 (C(CH_3)$_3$), 32.46 (C-4), 62.58 (C-7), 133.01 (C-2), 158.75 (C-3), 194.05 (C-1).

(1*E*,3*E*)-7-*tert*-Butyldimethylsilyloxy-1-trimethylsilyloxyhepta-1,3-diene (31) and (1*E*,3*Z*)-7-*tert*-butyldimethylsilyloxy-1-trimethylsilyloxyhepta-1,3-diene (32): A solution of α,β-unsaturated aldehyde **30** (4.53 g, 18.7 mmol) in toluene (21 mL) was added to a stirred suspension of $ZnCl_2$ (200 mg) in triethylamine (8.6 mL, 62.7 mmol). Trimethylsilyl bromide (5.4 mL, 41.1 mmol) was added dropwise with stirring, and the mixture was heated under reflux overnight. After having cooled to RT, the solution was filtered on a Celite pad, concentrated, diluted with cyclohexane (400 mL) and cooled for 1 h at 4 °C. After concentration, another filtration through a Celite pad afforded a crude oil, which was distilled to give a colourless oil as a mixture of the (1*E*,3*E*) and (1*E*,3*Z*) isomers **31** and **32** in 35:65 ratio. Yield: 75%; b.p. 110 °C (0.5 mbar). **31**: ^{1}H NMR: $\delta = 0.03$ (s, 6H; $Si(CH_3)_2$), 0.19 (s, 9H; $Si(CH_3)_3$), 0.88 (s, 9H; *t*Bu), 1.57 (tt, $J = 7.0$, 7.0 Hz, 2H; H-6), 2.06–2.16 (m, 2H; H-5), 3.59 (t, $J = 6.5$ Hz, 2H; H-7), 5.44 (dt, $J = 14.7$, 7.2 Hz, 1H; H-4), 5.66 (dd, $J = 11.3$, 11.3 Hz, 1H; H-2), 5.83–5.97 (m, 1H; H-3), 6.43 (d, $J = 11.8$ Hz, 1H; H-1); ^{13}C NMR: $\delta = -5.20$ ($Si(CH_3)_2$), -0.39 ($Si(CH_3)_3$), 18.42 ($C(CH_3)_3$), 26.06 (C(CH_3)$_3$), 29.15 (C-5), 32.73 (C-6), 62.67 (C-7), 113.94 (C-2), 126.22 (C-3), 129.11 (C-4), 142.29 (C-1). **32**: ^{1}H NMR: $\delta = 0.03$ (s, 6H; $Si(CH_3)_2$), 0.20 (s, 9H; $Si(CH_3)_3$), 0.88 (s, 9H; *t*Bu), 1.57 (tt, $J = 7.0$, 7.0 Hz, 2H; H-6), 2.06–2.16 (m, 2H; H-5), 3.59 (t, $J = 6.4$ Hz, 2H; H-7), 5.19 (dt, $J = 10.4$, 7.5 Hz, 1H; H-4), 5.83–5.97 (m, 2H; H-2, H-3), 6.49 (d, $J = 11.4$ Hz, 1H; H-1); ^{13}C NMR: $\delta = -5.20$ ($Si(CH_3)_2$), -0.39 ($Si(CH_3)_3$), 18.42 (C(CH_3)$_3$), 24.08 (C-5), 26.06 (C(CH_3)$_3$), 32.94 (C-6), 62.67 (C-7), 109.73 (C-2), 124.66 (C-3), 126.99 (C-4), 144.01 (C-1).

3*c*-Hydroxy-6*c*-(3-hydroxypropyl)-1*r*,2*t*-bis(phenylsulfonyl)-cyclohex-4-ene (33a) and 3*t*-hydroxy-6*t*-(3-hydroxypropyl)-1*r*,2*t*-bis(phenylsulfonyl)-cyclohex-4-ene (33b): *Trans*-1,2-bis(phenylsulfonyl)ethylene (**14**; 2.12 g, 6.88 mmol) was added to the above 65:35 mixture of the dienes **32** and **31** (6.17 g) in anhydrous *ortho*-xylene (7 mL). The mixture was stirred at 120 °C until complete disappearance of sulfone (24 h). After concentration under high vacuum, the crude product was dissolved in methanol (7 mL) with one drop of 12N hydrochloric acid and stirred for 30 min. Concentration followed by purification of the resulting oil by chromatography (cyclohexane/ethyl acetate 95:5–0:1) afforded a white foam as a mixture of

two diastereoisomers **33a** and **33b** in a ratio of 80:20 to 65:35. Yield: 1.98 g, 66%. **33a**: ^{1}H NMR: $\delta = 1.42-1.51$ (m, 1H; H-2′), 1.53–1.64 (m, 1H; H-2′), 1.73–1.81 (m, 1H; H-1′), 1.83–1.91 (m, 1H; H-1′), 2.45 (m, 1H; OH-3′), 2.87 (m, 1H; H-6), 3.49 (t, $J = 6.3$ Hz, 2H; H-3′), 3.60 (d, $J = 9.7$ Hz, 1H; OH-3), 3.83 (m, 1H; H-2), 4.10 (m, 1H; H-1), 4.52 (m, 1H; H-3), 5.74–5.88 (m, 2H; H-4, H-5), 7.33–7.84 (m, 10H; Ph); ^{13}C NMR: $\delta = 28.16$ (C-1′), 30.61 (C-2′), 34.82 (C-6), 58.96 (C-1), 61.45 (C-3), 61.80 (C-3′), 66.46 (C-2), 127.41–129.70 (C-4, C-5), 127.83–141.71 (Ph). **33b**: ^{1}H NMR: $\delta = 1.22-1.26$ (m, 1H; H-2′), 1.31–1.39 (m, 1H; H-2′), 1.54–1.64 (m, 2H; H-1′), 2.45 (m, 1H; OH-3′), 2.61 (m, 1H; H-6), 3.36 (t, $J = 6.3$ Hz, 2H; H-3′), 3.54 (m, 1H; OH-3), 4.10 (m, 1H; H-1), 4.22 (m, 1H; H-2), 4.81 (m, 1H; H-3), 5.64 (d, $J = 10.5$ Hz, 1H; H-4), 5.74–5.88 (m, 1H; H-5), 7.33–7.84 (m, 10H; Ph); ^{13}C NMR: $\delta = 29.60$ (C-1′), 30.86 (C-2′), 31.40 (C-6), 61.50 (C-2), 61.55 (C-1), 61.70 (C-3′), 64.58 (C-3), 127.83–141.71 (Ph), 128.16 (C-4), 128.76 (C-5); **33a** + **33b**: elemental analysis calcd for $C_{21}H_{24}O_6S_2$: C 57.40, H 5.80; found C 57.78, H 5.54.

6*c*-(3-*tert*-Butyldimethylsilyloxypropyl)-3*c*-hydroxy-1*r*,2*t*-bis(phenylsulfonyl)cyclohex-4-ene (34a) and 6*t*-(3-*tert*-butyldimethylsilyloxypropyl)-3*t*-hydroxy-1*r*,2*t*-bis(phenylsulfonyl)cyclohex-4-ene (34b): Anhydrous triethylamine (140 μL, 1.01 mmol) and *tert*-butyldimethylsilyl chloride (280 mg, 1.84 mmol) were added to a stirred solution of diols **33a** and **33b** (400 mg, 0.92 mmol) in anhydrous dichloromethane (10 mL) in the presence of a catalytic amount of DMAP (0.2 eq). After 5.5 h of stirring, the mixture was concentrated and purified by chromatography (cyclohexane/ethyl acetate 95:5–5:5), to afford a yellow oil as a mixture of the two diastereoisomers **34a** and **34b** in a ratio of 80:20 to 65:35. Yield: 83%. **34a**: ^{1}H NMR: $\delta = 0.06$ (s, 6H; $SiCH_3$), 0.90 (s, 9H; *t*Bu), 1.43–1.58 (m, 1H; H-2′), 1.64–1.70 (m, 1H; H-2′), 1.81–1.88 (m, 2H; H-1′), 2.94 (m, 1H; H-6), 3.41 (d, $J = 10.2$ Hz, 1H; OH), 3.56–3.63 (m, 2H; H-3′), 3.97 (s, 1H; H-2), 4.16 (d, $J = 5.4$ Hz, 1H; H-1), 4.55 (m, 1H; H-3), 5.88–5.94 (m, 2H; H-4, H-5), 7.54–5.77 (m, 10H; Ph); ^{13}C NMR: $\delta = -5.17$ ($SiCH_3$), 18.51 ($C(CH_3)_3$), 26.08 ($C(CH_3)_3$), 28.46 (C-1′), 31.31 (C-2′), 35.21 (C-6), 59.45 (C-1), 61.90 (C-3), 62.55 (C-3′), 66.45 (C-2), 127.85–141.70 (C-4, C-5, Ph); **34b**: ^{1}H NMR: $\delta = 0.01$ (s, 6H; $SiCH_3$), 0.87 (s, 9H; *t*Bu), 1.23–1.26 (m, 2H; H-2′), 1.55–1.60 (m, 2H; H-1′), 2.65 (m, 1H; H-6), 3.46 (s, 1H; OH), 3.45 (t, $J = 6.2$ Hz, 2H; H-3′), 4.10 (m, 1H; H-1), 4.36 (m, 1H; H-2), 4.88 (m, 1H; H-3), 5.71–5.75 (m, 1H; H-4), 5.83–5.91 (m, 1H; H-5), 7.54–7.77 (m, 10H; Ph); ^{13}C NMR: $\delta = -5.17$ ($SiCH_3$), 18.51 ($C(CH_3)_3$), 26.08 ($C(CH_3)_3$), 30.38 (C-2′), 31.31 (C-1′), 32.00 (C-6), 61.50–61.55 (C-1, C-2), 62.65 (C-3′), 65.11 (C-3), 127.85–141.70 (C-4, C-5, Ph); **34a** + **34b**: elemental analysis calcd (%) for $C_{27}H_{38}O_6S_2Si$: C 58.77, H 7.24; found C 58.88, H 6.95.

3*t*-Azido-6*c*-(3-*tert*-butyldimethylsilyloxypropyl)-1*r*,2*t*-di(phenylsulfonyl)-cyclohex-4-ene (35a) and 3*c*-azido-6*t*-(3-*tert*-butyldimethylsilyloxypropyl)-1*r*,2*t*-di(phenylsulfonyl)cyclohex-4-ene (35b): A mixture of alcohols **34a** and **34b** (420 mg, 0.76 mmol) and triphenylphosphine (280 mg, 1.06 mmol) in anhydrous dichloromethane (2 mL) was stirred at 0 °C. After 10 min, diisopropylazodicarboxylate (210 μL, 1.07 mmol) and diphenylphosphoryl azide (230 μL, 1.06 mmol) were added. The solution was kept at 0 °C for 1.5 h. Purification of the crude product by chromatography (cyclohexane/ethyl acetate 95:5–7:3) afforded the desired azide as a yellow oil as a 80:20 to 60:40 mixture of the two diastereoisomers **35a** and **35b**. Yield: 52%. **35a**: ^{1}H NMR: $\delta = 0.01$ (s, 6H; $SiCH_3$), 0.88 (s, 9H; *t*Bu), 1.42–1.49 (m, 2H; H-2′), 1.70–1.75 (m, 2H; H-1′), 3.12 (m, 1H; H-6), 3.40–3.46 (m, 2H; H-3′), 4.43 (m, 1H; H-2), 4.66 (m, 1H; H-1), 4.88 (m, 1H; H-3), 5.81 (d, $J =$ 10.4 Hz, 1H; H-5), 5.98 (d, $J = 10.4$ Hz, 1H; H-4), 7.58–7.97 (m, 10H; Ph); ^{13}C NMR: $\delta = -5.24$ ($SiCH_3$), 18.38 ($C(CH_3)_3$), 26.05 ($C(CH_3)_3$), 29.80, 30.67 (C-1′, C-2′), 35.03 (C-6), 54.95 (C-3), 60.45 (C-2), 62.21 (C-3′), 62.53 (C-1), 122.16 (C-5), 128.23–141.47 (C-4, Ph); **35b**: ^{1}H NMR: $\delta = 0.00$ (s, 6H; $SiCH_3$), 0.87 (s, 9H; *t*Bu), 1.42–1.49 (m, 2H; H-2′), 1.54–1.70 (m, 2H; H-1′), 2.82 (m, 1H; H-6), 3.40–3.46 (m, 2H; H-3′), 4.13 (s, 1H; H-1), 4.20 (s, 1H; H-2), 4.46 (s, 1H; H-3), 5.86 (d, $J = 10.5$ Hz, 1H; H-4), 6.15 (d, $J =$ 10.5 Hz, 1H; H-5), 7.58–7.97 (m, 10H; Ph); ^{13}C NMR: $\delta = -5.24$ ($SiCH_3$), 18.38 ($C(CH_3)_3$), 26.05 ($C(CH_3)_3$), 26.95, 29.75 (C-1′, C-2′), 32.10 (C-6), 51.21 (C-3), 58.50 (C-1), 61.30 (C-2), 62.21 (C-3′), 120.88 (C-4), 128.23–141.47 (Ph), 133.41 (C-5); **35a** + **35b**: IR ($CDCl_3$): $\tilde{\nu} = 2100$ cm^{-1} (N_3); MS: *m/z*: 576 $[M+H]^+$, 593 $[M+NH_4]^+$.

3-*tert*-Butyldimethylsilyloxypropyl-1-(phenylsulfonyl)benzene (36): This compound was also isolated by chromatography as a yellow oil in a yield of 30%. ^{1}H NMR: $\delta = 0.05$ (s, 6H; $SiCH_3$), 0.31 (s, 9H; *t*Bu), 1.81–1.87 (m, 2H; CH_2CH_2O), 2.75 (t, $J = 7.7$ Hz, 2H; CH_2Ph), 3.62 (t, $J = 6.1$ Hz, 2H; CH_2O), 7.41 (m, 2H; H-4), 7.41–7.45 (m, 1H; H-5), 7.77 (ddd, $J = 6.1$, 2.3, 2.3 Hz, 1H; H-6), 7.79 (m, 1H; H-2), 7.96 (dd, $J = 8.3$, 1.1 Hz, 2H; H-2′), 7.50–7.60 (m, 3H; H-3′, H-4′); ^{13}C NMR (50 MHz): $\delta = -5.22$ ($SiCH_3$), 18.80 ($C(CH_3)_3$), 26.04 ($C(CH_3)_3$), 29.95 (CH_2CH_2O), 32.04 (CH_2Ph), 61.96 (CH_2O), 125.23–144.20 (Ph).

Compound 37: This compound was also isolated by chromatography, in a yield of 10%, and was characterised by its NMR spectra. ^{1}H NMR: $\delta = 0.06$ (s, 6H; $SiCH_3$), 0.91 (s, 9H; *t*Bu), 1.33–1.36 (m, 2H; H-2′), 1.74–1.83 (m, 1H; H-1′), 1.93–1.98 (m, 1H; H-1′), 2.28–2.33 (m, 1H; H-6), 3.44–3.50 (m, 2H; H-3′), 4.12 (d, $J = 4.1$ Hz, 1H; H-2), 4.28 (s, 1H; H-1), 4.31 (m, 1H; H-3), 5.84 (m, 1H; H-4), 6.28 (m, 1H; H-5), 7.55–7.77 (m, 10H; Ph); ^{13}C NMR: $\delta = -4.89$ ($SiCH_3$), 18.70 ($C(CH_3)_3$), 25.30 (C-1′), 26.34 ($C(CH_3)_3$), 30.32 (C-2′), 38.99 (C-6), 59.49 (C-2), 59.64–60.64 (C-1, C-3), 62.72 (C-3′), 118.94 (C-4), 128.40–139.98 (Ph), 135.84 (C-5); IR ($CDCl_3$): $\tilde{\nu} = 2100$ cm^{-1} (N_3); HRMS: *m/z*: found 576.2027 $[M+H]^+$; $C_{27}H_{38}O_5N_3S_2Si$ calcd 576.2022.

Allyl [6*c*-(3-*tert*-butyldimethylsilyloxypropyl)-1*r*,2*t*-bis(phenylsulfonyl)-cyclohex-4-en-3*t*-yl]carbamate (39a) and allyl [6*t*-(3-*tert*-butyldimethylsilyloxypropyl)-1*r*,2*t*-bis(phenylsulfonyl)-cyclohex-4-en-3*c*-yl]carbamate (39b): A solution of azides **35a** and **35b** (70:30, 335 mg, 0.58 mmol) in THF/isopropyl alcohol (1:1, 14 mL) was introduced into a reactor with a Lindlar catalyst (300 mg). After the mixture had been stirred overnight under hydrogen (5 bars), the catalyst was removed by centrifugation and washed with dichloromethane (2×20 mL). After evaporation of the combined layers, a yellow oil containing the two diastereoisomers **38a** and **38b** was obtained. This crude product was directly used in the following step without purification.

The oil was dissolved in absolute ethanol (14 mL) buffered with $NaHCO_3$ (489 mg, 5.82 mmol), and allyloxycarbonyl chloride (111 μL, 1.05 mmol) was added. The solution was sonicated at RT for 1.5 h and then concentrated. Purification by chromatography (cyclohexane/ethyl acetate 9:1–7:3) afforded the two diastereoisomers **39a** and **39b** in a 65:35 ratio as a yellow oil. Yield: 281 mg, 76% over two steps. **39a**: ^{1}H NMR: $\delta = 0.00$ (s, 6H; $SiCH_3$), 0.84 (s, 9H; *t*Bu), 1.47–1.56 (m, 1H; H-2′), 1.59–1.65 (m, 1H; H-2′), 1.75–1.81 (m, 1H; H-1′), 1.86–1.92 (m, 1H; H-1′), 3.10 (m, 1H; H-6), 3.51–3.57 (m, 2H; H-3′), 3.78–3.83 (ddd, J = 13.2, 5.6, 1.5 Hz, 1H; $COOCH_2$), 4.15–4.19 (m, 2H; $COOCH_2$, H-2), 4.39 (m, 1H; H-1), 5.06 (m, 1H; H-3), 5.07 (m, 1H; NH), 5.12 (dd, $J = 10.2$, 1.5 Hz, 1H; $=CH_2$), 5.14 (dd, $J = 17.3$, 1.5 Hz, 1H; $=CH_2$), 5.54 (d, $J = 10.2$ Hz, 1H; H-4), 5.63–5.71 (m, 1H; $CH=CH_2$), 5.85 (d, $J = 10.7$ Hz, 1H; H-5), 7.35–7.96 (m, 10H; Ph); ^{13}C NMR: $\delta = -4.90$ ($SiCH_3$), 18.73 ($C(CH_3)_3$), 26.38 ($C(CH_3)_3$), 28.55 (C-1′), 31.33 (C-2′), 35.26 (C-6), 46.40 (C-3), 60.64 (C-2), 62.13 (C-1), 62.93 (C-3′), 65.98 ($COOCH_2$), 118.23 ($=CH_2$), 125.04 (C-4), 127.68–141.59 (Ph), 131.24 (C-5), 132.62 ($CH=CH_2$), 154.92 (C=O). **39b**: ^{1}H NMR: $\delta = 0.00$ (s, 6H; $SiCH_3$), 0.86 (s, 9H; *t*Bu), 1.29–1.36 (m, 2H; H-2′), 1.49–1.59 (m, 1H; H-1′), 1.62–1.70 (mm, 1H; H-1′), 2.64 (m, 1H; H-6), 3.39–3.42 (s, 2H; H-3′), 4.10–4.24 (s, 2H; H-1, H-2), 4.40 (m, 1H; $COOCH_2$), 4.69 (m, 1H; H-3), 5.15 (dd, $J = 10.7$, 1.6 Hz, 1H; $=CH_2$), 5.22 (d, $J = 17.3$ Hz, 1H; $=CH_2$), 5.69 (d, $J = 9.7$ Hz, 1H; NH), 5.76–5.86 (m, 1H; $CH=CH_2$), 5.80–5.84 (m, 1H; H-4), 5.94 (dd, $J = 10.2$, 3.6 Hz, 1H; H-5), 7.58–7.94 (m, 10H; Ph); ^{13}C NMR: $\delta = -4.92$ ($SiCH_3$), 18.65 ($C(CH_3)_3$), 26.31 ($C(CH_3)_3$), 29.92, 30.33 (C-1′, C-2′), 31.53 (C-6), 41.50 (C-3), 58.08, 60.49 (C-1, C-2), 62.41 (C-3′), 65.72 ($COOCH_2$), 117.63 ($=CH_2$), 122.63 (C-4), 128.91–137.90 (Ph), 130.39 (C-5), 132.71 ($CH=CH_2$), 155.12 (C=O); **39a** + **39b**: HRMS: *m/z*: found 634.2335 $[M+H]^+$; $C_{31}H_{43}O_7NS_2Si$ calcd 634.2328.

Allyl [6*c*-(3-hydroxypropyl)-1*r*,2*t*-di(phenylsulfonyl)cyclohex-4-en-3*t*-yl]-carbamate (40a) and allyl [6*t*-(3-hydroxypropyl)-1*r*,2*t*-di(phenylsulfonyl)-cyclohex-4-en-3*c*-yl]carbamate (40b): Silylated alcohols **39a** and **39b** (280 mg, 0.44 mmol) solubilised in methanol (5 mL) in the presence of hydrochloric acid (12 N, 1 μL), were stirred for 15 min. After concentration, the mixture was purified by flash chromatography (cyclohexane/ethyl acetate 9:1–3:7) to give a white foam consisting of a mixture of the two diastereoisomers **40a** and **40b**. Yield: 197 mg, 86%. **40a**: ^{1}H NMR: $\delta = 1.54$ (s, 1H; OH), 1.70–1.80 (m, 2H; H-2′), 1.90–1.96 (m, 1H; H-1′), 2.08–2.18 (m, 1H; H-1′), 3.20 (m, 1H; H-6), 3.67 (m, 2H; H-3′), 3.88 (dd, $J = 13.2$, 5.6 Hz, 1H; $COOCH_2$), 4.12–4.46 (m, 2H; H-1, H-2), 4.23 (dd, $J = 13.2$, 5.6 Hz, 2H; $COOCH_2$), 5.12 (m, 1H; H-3), 5.12 (m, 1H; NH), 5.18–5.23 (m, 2H; $=CH_2$), 5.63 (d, $J = 10.6$ Hz, 1H; H-4), 5.68–5.75 (m, 1H; $CH=CH_2$), 5.91 (d, $J = 10.6$ Hz, 1H; H-5), 7.40–8.04 (m, 10H; Ph); ^{13}C NMR: $\delta = 28.43$, 30.86 (C-1′, C-2′), 35.06 (C-6), 46.06 (C-3), 60.38, 61.57 (C-1, C-2), 62.06 (C-3′), 65.70 ($COOCH_2$), 117.93 ($=CH_2$), 124.90 (C-4), 127.33–141.16 (C-5, Ph), 132.61 ($CH=CH_2$), 154.67 (C=O). **40b**: ^{1}H NMR:

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$\delta = 1.10-1.20$ (m, 2H; H-2′), 1.40–1.50 (m, 1H; H-1′), 1.60 (s, 1H; OH), 1.65–1.75 (m, 1H; H-1′), 2.69 (m, 1H; H-6), 3.47 (t, $J = 5.8$ Hz, 2H; H-3′), 4.09–4.29 (m, 2H; H-1, H-2), 4.10 (m, 2H; $COOCH_2$), 4.71 (m, 1H; H-3), 5.16 (dd, $J = 10.6$, 1.1 Hz, 1H; $=CH_2$), 5.23 (d, $J = 17.1$ Hz, 1H; $=CH_2$), 5.69 (d, $J = 9.9$ Hz, 1H; NH), 5.78–5.88 (m, 1H; $CH=CH_2$), 5.84 (m, 1H; H-4), 5.95 (dd, $J = 10.6$, 3.0 Hz, 1H; H-5), 7.59–7.97 (m, 10H; Ph); ^{13}C NMR: $\delta =$ 29.58, 30.23 (C-1′, C-2′), 31.34 (C-6), 41.50 (C-3), 58.08, 60.50 (C-1, C-2), 62.44 (C-3′), 65.70 ($COOCH_2$), 117.67 ($=CH_2$), 122.80 (C-4), 128.91–141.60 (Ph), 130.85 (C-5), 132.67 ($CH=CH_2$), 154.81 (C=O); **40a** + **40b**: elemental analysis calcd for $C_{25}H_{29}O_7NS_2$: C 57.78, H 5.62, N 2.69; found C 57.51, H 5.85, N 2.62.

Allyl [4*t*-(3-hydroxypropyl)-cyclohexa-2,5-dien-1*r*-yl]carbamate (41): A solution of disulfones **40a** and **40b** (253 mg, 0.49 mmol) in dry methanol (11 mL) buffered with KH_2PO_4 (2.56 g, 18 mmol) was vigorously stirred with 3.5% sodium amalgam (2.56 g, 3.9 mmol) under argon atmosphere for 45 min at RT. The salts and mercury were removed by filtration and washed with dichloromethane (10 mL), and the filtrate was concentrated and chromatographed (cyclohexane/ethyl acetate 5:5–4:6). After recrystallisation from ethyl acetate, **41** was obtained as white needles. Yield: 152 mg, 74%; m.p. 91 °C; ^{1}H NMR: $\delta = 1.45-1.47$ (m, 4H; H-1′, H-2′), 2.02 (s, 1H; OH), 2.72 (m, 1H; H-4), 3.60 (t, $J = 5.8$ Hz, 2H; H-3′), 4.56 (d, $J = 5.6$ Hz, 2H; $COOCH_2$), 4.67 (m, 1H; H-1), 4.88 (d, $J = 9.2$ Hz, 1H; NH), 5.23 (d, $J = 10.9$ Hz, 1H; $=CH_2$), 5.32 (d, $J = 17.2$ Hz, 1H; $=CH_2$), 5.78 (d, $J =$ 10.2 Hz, 2H; H-2, H-6), 5.85 (d, $J = 10.2$, 2.4 Hz, 2H; H-3, H-5), 5.91–5.98 (m, 1H; $CH=CH_2$); ^{13}C NMR: $\delta = 29.25$, 30.09 (C-1′, C-2′), 35.29 (C-4), 44.90 (C-1), 63.36 (C-3′), 65.95 ($COOCH_2$), 118.04 ($=CH_2$), 126.27 (C-2, C-6), 131.74 (C-3, C-5), 133.25 ($CH=CH_2$), 155.70 (C=O); elemental analysis calcd for $C_{13}H_{19}O_3N$: C 65.80, H 8.07, N 5.90; found C 65.77, H 8.05, N 5.91.

Allyl [4*t*-(3-oxopropyl)-cyclohexa-2,5-dien-1*r*-yl]-carbamate (42): Aldehyde **42** was prepared, as described for **5**, from alcohol **41** (140 mg, 0.59 mmol), oxalyl dichloride (65 μL, 0.71 mmol) and DMSO (100 μL, 1.42 mmol) in dichloromethane (2 mL); quantitative yield. ^{1}H NMR: $\delta =$ 1.74 (m, 2H; H-1′), 2.33 (m, 2H; H-2′), 2.77 (m, 1H; H-4), 4.50 (d, 2H; $COOCH_2$), 4.62 (m, 1H; H-1), 4.96 (d, $J = 8.6$ Hz, 1H; NH), 5.14 (d, $J =$ 10.5 Hz, 1H; $=CH_2$), 5.24 (d, $J = 17.3$ Hz, 1H; $=CH_2$), 5.68–5.76 (m, 4H; H-2, H-3, H-5, H-6), 5.85–5.95 (m, 1H; $CH=CH_2$), 9.69 (t, $J = 1.4$ Hz, 1H; H-3′); ^{13}C NMR: 26.93 (C-1′), 34.22 (C-4), 39.73 (C-2′), 44.51 (C-1), 65.57 ($COOCH_2$), 117.63 ($=CH_2$), 127.02–130.33 (C-2, C-3, C-5, C-6), 132.94 ($CH=CH_2$), 155.56 (N–C=O), 202.22 (C-3′).

Allyl [4*t*-(3-amino-3-cyano-propyl)cyclohexa-2,5-dien-1*r*-yl]-carbamate (43): Amino acid amide **43** was prepared as described for **17**, from **42** (140 mg, 0.59 mmol) and trimethylsilyl cyanide (120 μL, 0.89 mmol) in the presence of a catalytic amount of ZnI_2 and a saturated methanolic ammonia solution (2.5 mL). Chromatography (cyclohexane/ethyl acetate 9:1–0:1) gave 43 as a yellow powder. Yield: 74 mg, 51%; ^{1}H NMR: $\delta = 1.60-1.70$ (m, 4H; H-1′, H-2′), 1.80 (m, 2H; NH_2), 2.79–2.81 (m, 1H; H-4), 3.63 (t, $J = 7.2$ Hz, 1H; H-3′), 4.54 (d, $J = 5.5$ Hz, 2H; $COOCH_2$), 4.65 (m, 1H; H-1), 4.93 (d, $J = 9.2$ Hz, 1H; NH), 5.19 (d, $J = 10.3$ Hz, 1H; $=CH_2$), 5.27 (d, $J = 17.2$ Hz, 1H; $=CH_2$), 5.77 (m, 4H; H-2, H-3, H-5, H-6), 5.84–5.94 (m, 1H; $CH=CH_2$); ^{13}C NMR: $\delta = 30.62$, 31.31 (C-1′, C-2′), 34.47 (C-4), 43.56 (C-3′), 43.56 ($COOCH_2$), 65.68 (C-1), 117.79 ($=CH_2$), 122.00 (CN), 126.89, 130.51 (C-2, C-3, C-5, C-6), 132.92 ($CH=CH_2$), 155.57 (C=O).

Aromatic compound **19** was also isolated as a yellow oil in a yield of 40%.

2-Amino-4-(4*t*-amino-cyclohexa-2,5-dien-1*r*-yl)-butanenitrile hydrochloride (44) and 2-Amino-4-(4*t*-amino-cyclohexa-2,5-dien-1*r*-yl)-butanamide hydrochloride (45): Amino nitrile **44** and amide **45** were prepared as described for **17**, from **43** (35 mg, 0.13 mmol), phenylsilane (60 μL, 0.50 mmol) and a solution of $Pd(PPh_3)_4$ (10 mg, 0.009 mmol) in dry dichloromethane (1 mL). Yield: 31 mg (85%) of the mixture of amine hydrochlorides **44** and **45**. **44**: ^{1}H NMR (CD_3OD): $\delta = 1.77-1.82$ (m, 2H; H-4′), 1.92–1.99 (m, 2H; H-3), 3.09 (m, 1H; H-1′), 4.36 (m, 1H; H-4′), 4.57 (dd, $J = 8.7$, 5.9 Hz, 1H; H-2), 5.97–6.00 (m, 2H; H-3′, H-5′), 6.17–6.21 (m, 2H; H-2′, H-6′); ^{13}C NMR (50 MHz, CD_3OD): $\delta = 28.35$ (C-3), 30.70 (C-4), 35.79 (C-1′), 42.71 (C-4′), 45.68 (C-2), 116.75 (CN), 123.33 (C-3′, C-5′), 135.78 (C-2′, C-6′). **45**: ^{1}H NMR (CD_3OD): $\delta = 1.63-1.70$ (m, 2H; H-4), 1.80–1.89 (m, 2H; H-3), 3.02 (m, 1H; H-1′), 3.92 (t, $J = 6.2$ Hz, 1H; H-2), 4.32 (m, 1H; H-4′), 5.93 (d, $J = 10.4$ Hz, 1H; H-3′, H-5′), 6.18 (d, $J =$ 10.4 Hz, 1H; H-2′, H-6′); ^{13}C NMR (50 MHz, CD_3OD) : 28.70 (C-3), 30.37 (C-4), 35.79 (C-1′), 46.10 (C-4′), 54.16 (C-2), 122.95 (C-3′, C-5′), 136.61 (C-2′, C-6′), 172.20 (C-1).

Aromatic compounds **19** and **21** were also identified, in a yield of 15%.

(2*R*)-2-Amino-4-(4*t*-amino-cyclohexa-2,5-dien-1*r*-yl)butanoic acid (1): Amino acid **1** was prepared as described for **2**, from a mixture of **44** and **45** (40 mg, 0.149 mmol) and pronase (1 g). Yield: 20%; ^{1}H NMR (500 MHz, D_2O): $\delta = 1.46-1.52$ (m, 1H; H-4), 1.55–1.61 (m, 1H; H-4), 1.74–1.81 (m, 2H; H-3), 2.91 (m, 1H; H-1′), 3.62 (t, $J = 5.9$ Hz, 1H; H-2), 4.28 (m, 1H; H-4′), 5.80 (d, $J = 10.4$ Hz, 2H; H-3′, H-5′), 6.01 (d, $J =$ 10.4 Hz, 2H; H-2′, H-6′); ^{13}C NMR (125 MHz, D_2O): $\delta = 27.10$, 29.49 (C-3, C-4), 35.10 (C-1′), 45.25 (C-4′), 55.50 (C-2), 121.79 (C-3′, C-5′), 135.80 (C-2′, C-6′), 175.32 (C=O).

The D-amino acid amide **45** was isolated in a yield of 20%. MS of **45**: no molecular peak was observed when an ESI or APCI source was used, but a peak at *m/z* 179 was identified as corresponding to the elimination of NH_3. MS-MS: *m/z* (%) 179: 134 (90) $[Ph(CH_2)_2CH=NH_2]^+$, 117 (10) $[PhCH_2CH=CH]^+$, 91 (45) $[PhCH_2]^+$, identical to those observed for the *cis* isomer. Compound **1** shows the same behavior in the MS.

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