

Scheme 1

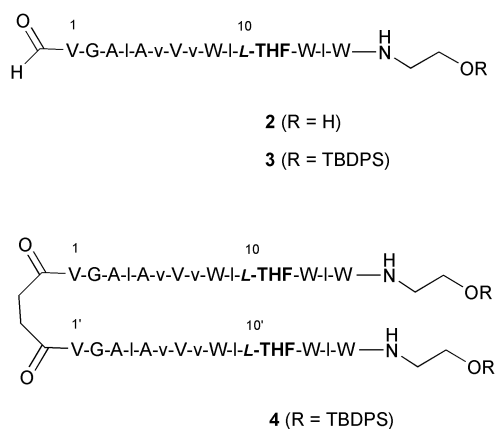


Fig. 1

chelation control). Epoxidation of the terminal double bond and a subsequent acid-mediated intramolecular epoxide opening gave, after chromatographic separation, the *D*-*cis*-THF alcohol (2*R*,5*S*)-**12** and the corresponding *trans*-THF alcohol together with the minor stereoisomers from the cuprate addition. A one-step oxidation of (2*R*,5*S*)-**12** led to the desired *N*-Boc-protected *D*-*cis*-THF amino acid **13** (Scheme 2). The synthesis of the target peptides was accomplished by fragment coupling in solution. Although protocols for the solid-phase synthesis of gA are available,<sup>19</sup> we chose the fragment-coupling strategy<sup>20</sup> for the following reasons: higher purity, larger quantities, modular approach with predictable diversity.

The retrosynthetic analysis of the target peptides is summarized in Scheme 3. All four monomers **2**, **3**, **5**, and **6** share the common tetramer **14**. Thus, disconnection between *D*-Leu4 and Ala5 leads to the decamer **15** containing the *L*-THF amino acid

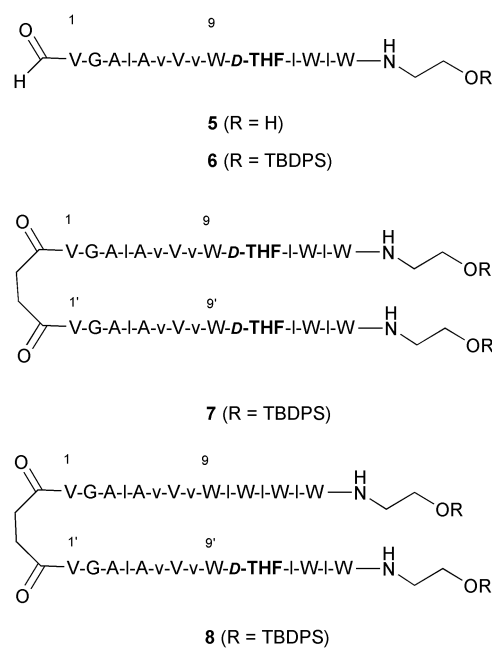
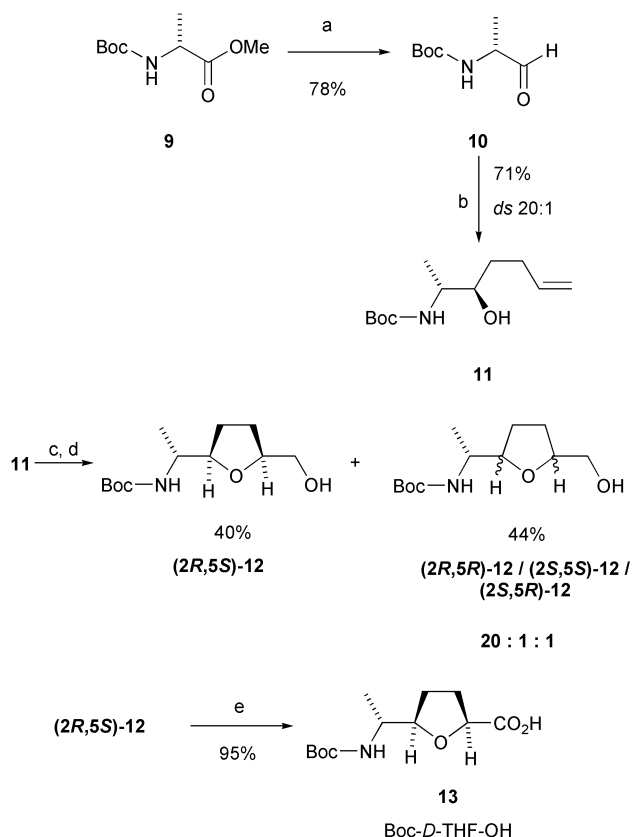


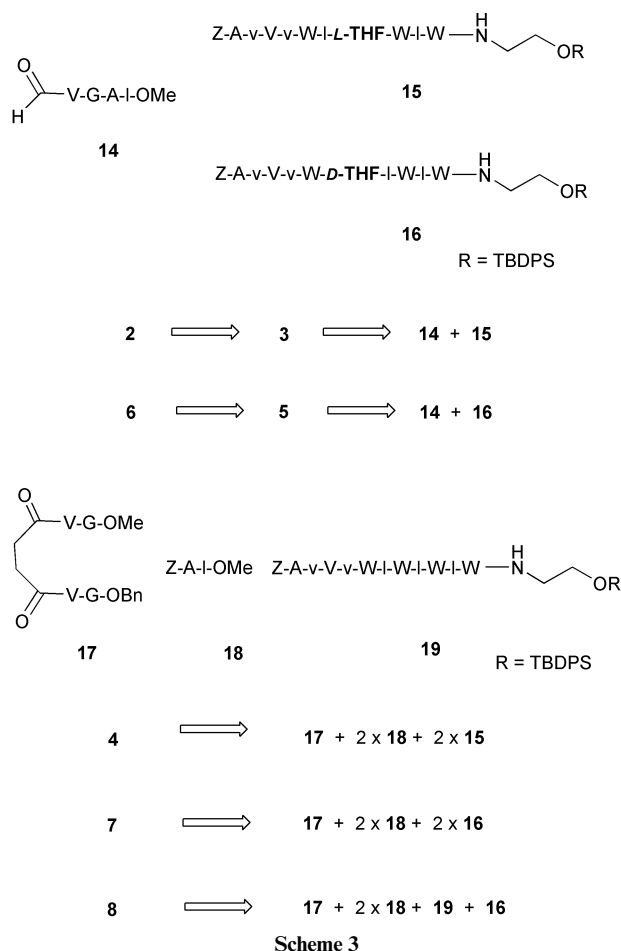
Fig. 2



**Scheme 2** (a) DIBAL-H (2.0 eq.), CH<sub>2</sub>Cl<sub>2</sub>-*n*-hexane 30 : 1, -60 °C; (b) (i) lithiation of 1-bromo-3-butene (4.5 eq.): *t*-BuLi (9.9 eq.), Et<sub>2</sub>O-*n*-hexane 1 : 1, -78 °C; (ii) cuprate-addition: 1-lithium-3-butene (4.5 eq.), CuBr·SMe<sub>2</sub> (2.2 eq.), Et<sub>2</sub>O-*n*-hexane 2 : 1, -40 °C → -80 °C; (c) MCPBA (2.0 eq.), CH<sub>2</sub>Cl<sub>2</sub>, 0 °C → rt; (d) CSA (0.04 eq.), CH<sub>2</sub>Cl<sub>2</sub>, rt; (e) iodobenzene diacetate (2.2 eq.), 2,2,6,6-tetramethylpiperidin-1-yl oxyl free radical (0.05 eq.), CH<sub>2</sub>Cl<sub>2</sub>, rt.

**1** and the decamer **16** which includes the *D*-THF amino acid **ent-1**.

The covalently linked dimers **4**, **7**, and **8** were assembled from the two building blocks **17** and **18**, as well as the decamer **15**, the decamer **16** and the undecamer **19**. The use of the tetramer clamp **17** results in final Gly-Ala coupling steps, which in our hands proved to be more efficient than the previously reported

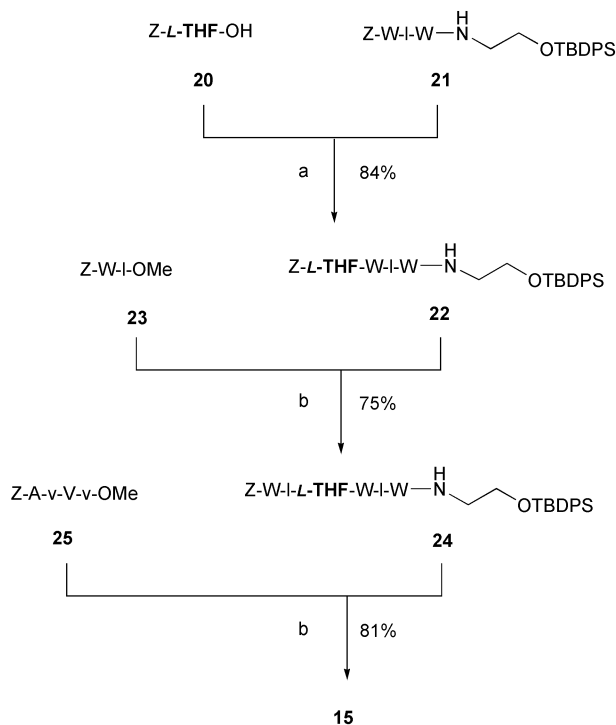


succinyl–Val or tartrate–Val couplings for the synthesis of covalently linked gA dimers.<sup>13,20</sup> The lipophilic TBDPS-group at the aminoethanol terminus increases the solubility in organic solvents and eases chromatographic purification. Z/OMe protections were used throughout the synthesis due to their mild deprotection conditions and the possibility of leaving the Trp side chain unprotected.

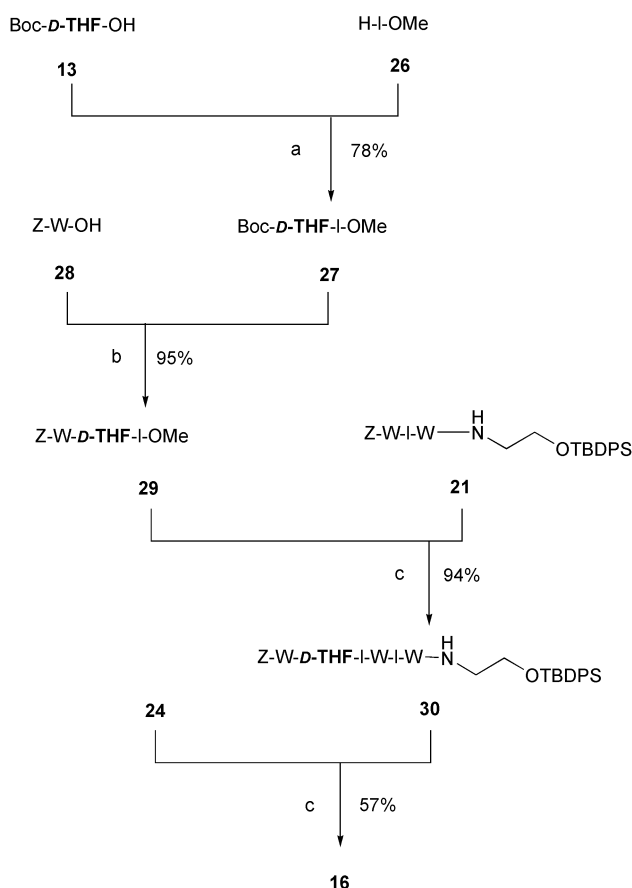
The synthesis of the decamer **15** containing the L-THF **1** utilizes HBTU–HOBt couplings in CH<sub>2</sub>Cl<sub>2</sub> or CH<sub>2</sub>Cl<sub>2</sub>–DMF to assemble the peptide sequence (Scheme 4). Reaction of Z-L-THF–OH **20** with the ethanolamine-tagged tripeptide **21**<sup>20</sup> resulted in the tetramer **22**. A detailed discussion of the proper choice of deprotection conditions (H<sub>2</sub>, Pd/C and LiOH–THF–MeOH) is given in our earlier work on minigramicidins.<sup>20</sup> Coupling of **22** with the dipeptide **23**<sup>20</sup> gave the hexamer **24**, which was further elaborated using the tetramer **25**<sup>20</sup> to yield the desired decamer **15**.

The decamer **16** containing the D-THF **ent-1** was synthesized starting with the coupling of Boc-D-THF–OH **13** and H-D-Leu–OMe **26** to the dimer **27** (Scheme 5). Addition of Z-Trp–OH **28** led to **29**. The latter was coupled with the ethanolamine-tagged tripeptide **21** to produce the hexamer **30**. The conjugation of **24** and **30** completed the assembly of the decamer **16**.

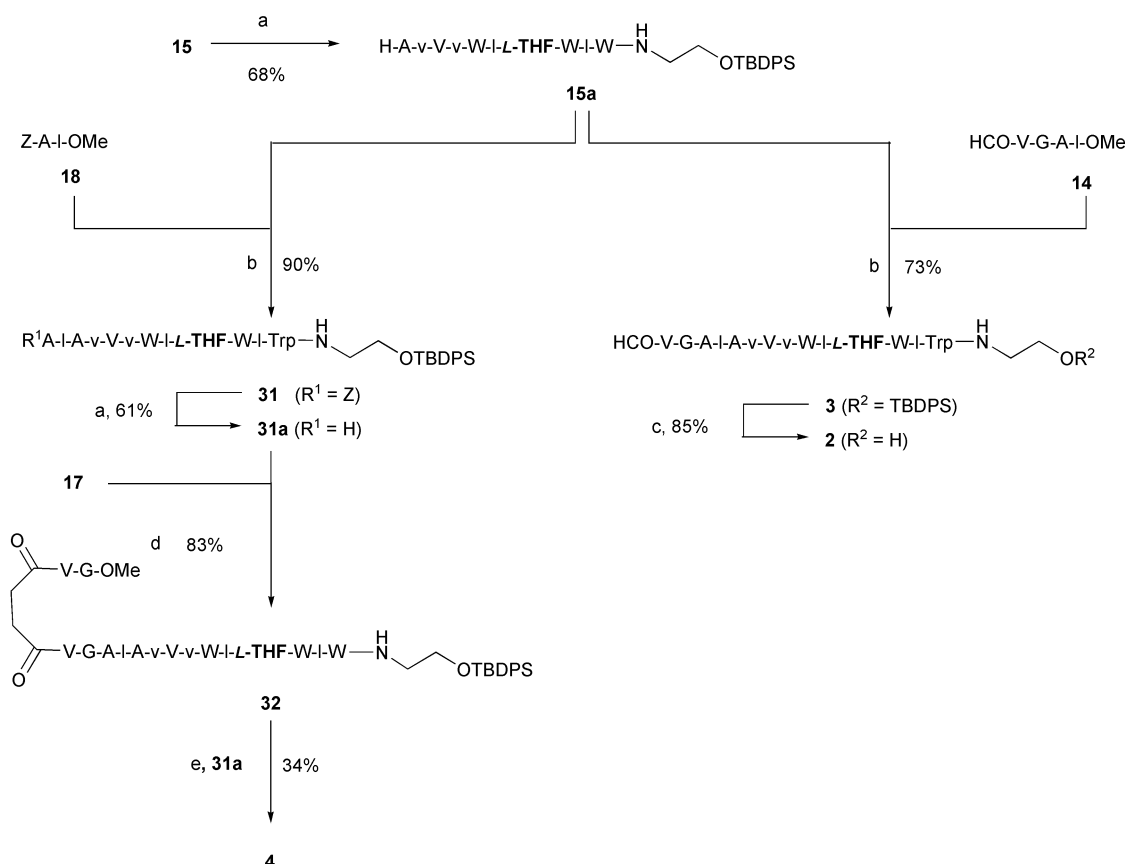
To proceed towards the L-THF–gA hybrids **2**, **3**, and **4** (Scheme 6), the decamer **15** was *N*-deprotected by hydrogenolysis and the resulting amine **15a** was coupled with the *N*-formyl tetramer **14** to yield compound **3** in 73% yield. Cleavage of the TBDPS-group in **3** with 5% HF in CH<sub>3</sub>CN gave **2**. Treatment of the methyl ester **18** with LiOH resulted in the corresponding carboxylic acid, which was coupled with the amine **15a** to give the dodecamer **31**. After hydrogenolytic cleavage of the Z-group the amine **31a** was obtained and subsequently conjugated with the tetrameric clamp **17** to produce compound **32**. Hydrolysis of the methyl ester in **32** and



**Scheme 4** (a) Z-group cleavage: Pd/C, H<sub>2</sub> (1 bar), MeOH, rt; coupling: EDC (1.5 eq.), HOBt (3.0 eq.), (*i*-Pr)<sub>2</sub>EtN (3.0 eq.), CH<sub>2</sub>Cl<sub>2</sub>, 0 °C→rt; (b) methyl ester deprotection: 2.5 eq LiOH, THF–H<sub>2</sub>O 3 : 1, 0 °C; Z-group cleavage: Pd/C, H<sub>2</sub> (1 bar), MeOH, 40 °C; coupling: HBTU (1.5 eq.), HOBt (3.0 eq.), (*i*-Pr)<sub>2</sub>EtN (3.0 eq.), CH<sub>2</sub>Cl<sub>2</sub>–DMF 5 : 1, –15 °C.



**Scheme 5** (a) EDC (1.5 eq.), HOBt (3.0 eq.), (*i*-Pr)<sub>2</sub>EtN (3.0 eq.), CH<sub>2</sub>Cl<sub>2</sub>, 0 °C→rt; (b) Boc-group cleavage: TFA, CH<sub>2</sub>Cl<sub>2</sub>, rt; coupling: EDC (1.5 eq.), HOBt (3.0 eq.), (*i*-Pr)<sub>2</sub>EtN (3.0 eq.), CH<sub>2</sub>Cl<sub>2</sub>, 0 °C→rt; (c) methyl ester deprotection: 2.5 eq LiOH, THF–H<sub>2</sub>O 3 : 1, 0 °C; Z-group cleavage: Pd/C, H<sub>2</sub> (1 bar), MeOH, 40 °C; coupling: HBTU (1.5 eq.), HOBt (3.0 eq.), (*i*-Pr)<sub>2</sub>EtN (3.0 eq.), CH<sub>2</sub>Cl<sub>2</sub>–DMF 5 : 1, –15 °C.



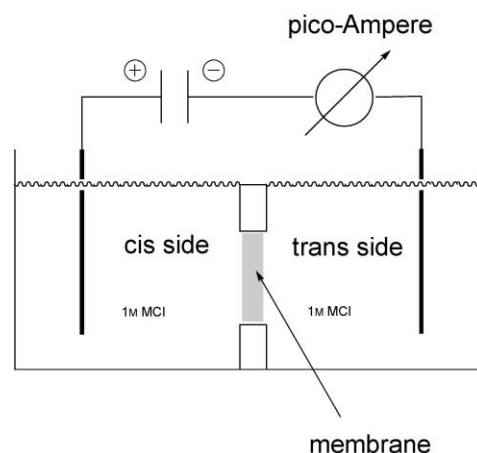
**Scheme 6** (a) Pd/C, H<sub>2</sub> (1 bar), EtOH, 50 °C; (b) methyl ester deprotection: 2.5 eq LiOH, THF–H<sub>2</sub>O 3 : 1, 0 °C; coupling: HBTU (1.5 eq.), HOBT (3.0 eq.), (*i*-Pr)<sub>2</sub>EtN (3.0 eq.), CH<sub>2</sub>Cl<sub>2</sub>–DMF 5 : 1, –15 °C; (c) HF (5% in CH<sub>3</sub>CN), CH<sub>2</sub>Cl<sub>2</sub>–DMF 10 : 1, rt; (d) benzyl ester deprotection: Pd/C, H<sub>2</sub> (1 bar), MeOH, rt; coupling: HBTU (1.5 eq.), HOBT (3.0 eq.), (*i*-Pr)<sub>2</sub>EtN (3.0 eq.), CH<sub>2</sub>Cl<sub>2</sub>–DMF 5 : 1, –15 °C; (e) methyl ester deprotection: 2.5 eq LiOH, THF–H<sub>2</sub>O 3 : 1, 0 °C; coupling: HATU (1.5 eq.), HOAt (3.0 eq.), (*i*-Pr)<sub>2</sub>EtN (3.0 eq.), CH<sub>2</sub>Cl<sub>2</sub>–DMF 5 : 1, –15 °C.

coupling of the resulting carboxylic acid with the amine **31a** led to the symmetrical bis-L-THF channel hybrid **4**.

The decamer **16** was converted into the free amine **16a** and coupled with the carboxylic acid prepared from the tetrameric methyl ester **14** (Scheme 7). The desired monomeric D-THF–gA hybrid **6** could be obtained in 87% yield. Deprotection of the silyl ether led to compound **5**. In order to get to the linked dimers containing the D-THF amino acid **ent-1**, the methyl ester **18** was transformed into the corresponding carboxylic acid and coupled with the amine **16a** to yield the dodecamer **33** and its Z-deprotected form **33a**, respectively. The benzyl ester **17** could be converted into the free acid and coupled with the amine **33a** to produce compound **34**. Hydrolysis of the methyl ester in **34** and subsequently coupling with a second molecule of **33a** gave the symmetrically bis-D-THF channel hybrid **7**. After deprotection of the methyl ester in **35**,<sup>20</sup> the coupling of the resulting carboxylic acid with the amine **33a** produced the unsymmetrical mono-D-THF channel hybrid **8**.

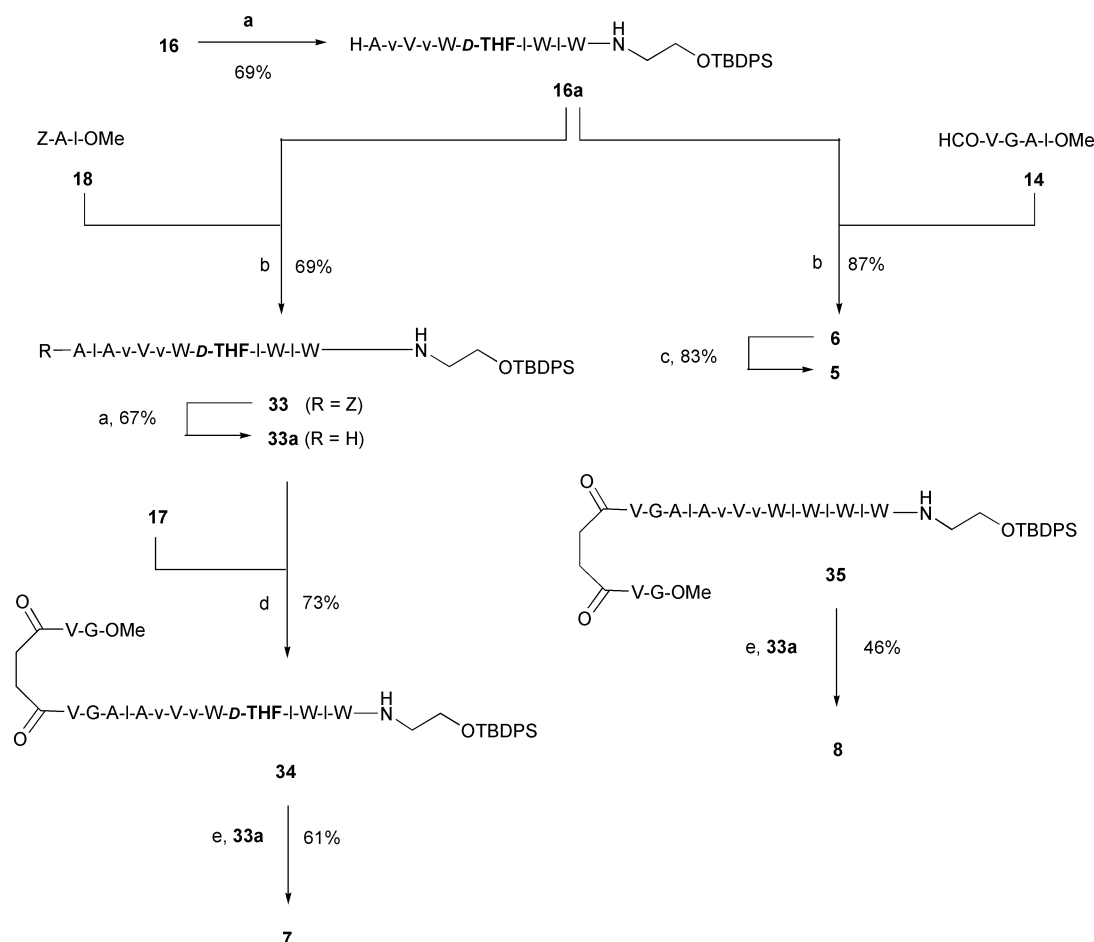
All gA-hybrid channels **2–8** were purified by RP-HPLC and characterized by <sup>1</sup>H-NMR and MS (see experimental section and electronic supplementary information (ESI)†). The characterisation of ion channels by single-channel currents is a single-molecule technique and has to fulfil high-purity criteria in order to avoid misinterpretation of channel-active impurities. The advanced synthetic intermediates of Schemes 6 and 7 were all tested routinely for ion channel activity. HPLC-separation was additionally monitored by single-channel current analysis.

For the functional analysis of the ion channels, planar phospholipid bilayers (black-lipid membranes, Fig. 3)<sup>1</sup> were used. If not stated otherwise, soybean lecithin was the phospholipid of choice. The channel formers were added from a methanol stock solution to the *cis* and/or the *trans* side of the measurement chamber. Details are given in the experimental section.

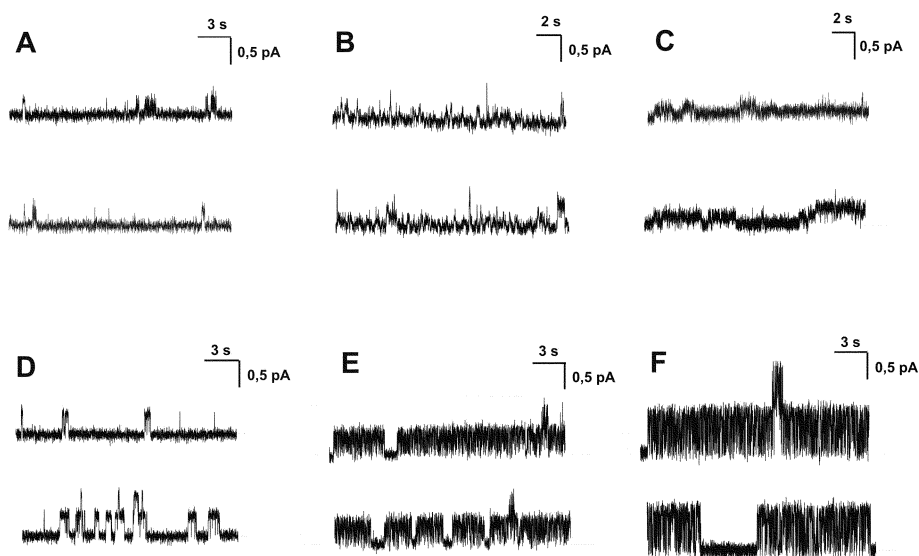


**Fig. 3** Experimental setup for single-channel measurements in planar phospholipid bilayers.

The replacement of Trp11-D-Leu12 of gA with the L-THF amino acid **1** resulted in a considerable change in the ion channel properties. Selected traces are shown in the upper part of Fig. 4. For the monomeric hybrids **2** and **3** it was necessary to add the compounds in high concentrations from the *cis* and the *trans* side to see any channels at all. Conductivities were reduced to 10% compared with gA. The observation of single channel currents was restricted to Cs<sup>+</sup>. For compounds **2** and **3** multiple noisy events were found. The covalently linked dimer **4** showed better resolved channel signals at lower concentrations (Fig. 4c). Here, it was sufficient to add the compound from the *cis* side only. The channel conductivity of **4** was still low and allowed the detection of Cs<sup>+</sup> currents only. Thus, the use of the L-THF-amino acid **1** in positions 11 and 12 of gA reduces the transport efficiency of the gA channel strongly.



**Scheme 7** (a) Pd/C, H<sub>2</sub> (1 bar), EtOH, 50 °C; (b) methyl ester deprotection: 2.5 eq LiOH, THF–H<sub>2</sub>O 3 : 1, 0 °C; coupling: HBTU (1.5 eq.), HOBT (3.0 eq.), (*i*-Pr)<sub>2</sub>EtN (3.0 eq.), CH<sub>2</sub>Cl<sub>2</sub>–DMF 5 : 1, –15 °C; (c) HF (5% in CH<sub>3</sub>CN), CH<sub>2</sub>Cl<sub>2</sub>–DMF 10 : 1, rt; (d) Pd/C, H<sub>2</sub> (1 bar), EtOH, 50 °C; coupling: HBTU (1.5 eq.), HOBT (3.0 eq.), (*i*-Pr)<sub>2</sub>EtN (3.0 eq.), CH<sub>2</sub>Cl<sub>2</sub>–DMF 5 : 1, –15 °C; (e) methyl ester deprotection: 2.5 eq LiOH, THF–H<sub>2</sub>O 3 : 1, 0 °C; coupling: HATU (1.5 eq.), HOAt (3.0 eq.), (*i*-Pr)<sub>2</sub>EtN (3.0 eq.), CH<sub>2</sub>Cl<sub>2</sub>–DMF 5 : 1, –15 °C.



**Fig. 4** Channel traces of THF–gA–hybrids. The upper part shows the results from the replacement of positions 11 and 12 by the L–THF amino acid **1** (A: **2**, B: **3**, C: **4**); the lower part shows the results from the replacement of positions 10 and 11 by the D–THF amino acid **ent-1** (D: **5**, E: **6**, F: **7**) in asolectin (1 M CsCl, +170 mV).

The effect of the replacement of D–Leu10–Trp11 with the D–THF amino acid **ent-1** was examined next. The monomeric D–THF hybrids **5** and **6** as well as the symmetric, dimeric D–THF–gA hybrid **7** show single channel behavior for Cs<sup>+</sup>, Rb<sup>+</sup>, K<sup>+</sup> and Na<sup>+</sup> ions (Table 1). The conductivities were much lower

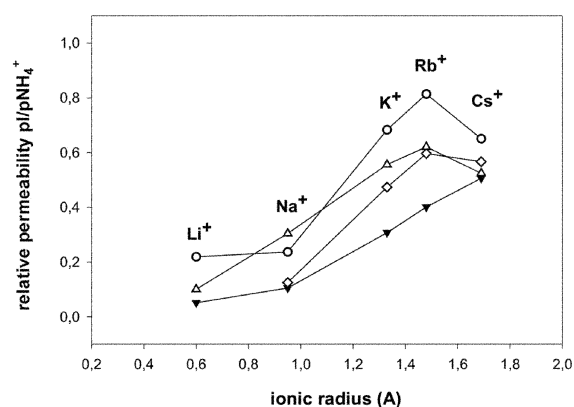
for **5**, **6**, and **7** than for gA but the channels were uniform and well defined.

Representative traces are shown in the lower part of Fig. 4. The conductivity series Cs<sup>+</sup> > Rb<sup>+</sup> > K<sup>+</sup> > Na<sup>+</sup> is similar to that for gA (see electronic supplementary information (ESI)†).



**Table 1** Single-channel conductances of compounds **7**, **8** and succ(gA-TBDPS)<sub>2</sub> for different cations

Compound	Conductance/pS		
	1 M CsCl	1 M KCl	1 M NaCl
<b>8</b>	5.7	2.2	0.6
<b>7</b>	7.0	1.8	0.6
Succ(gA-TBDPS) <sub>2</sub>	19.1	14.0	7.7

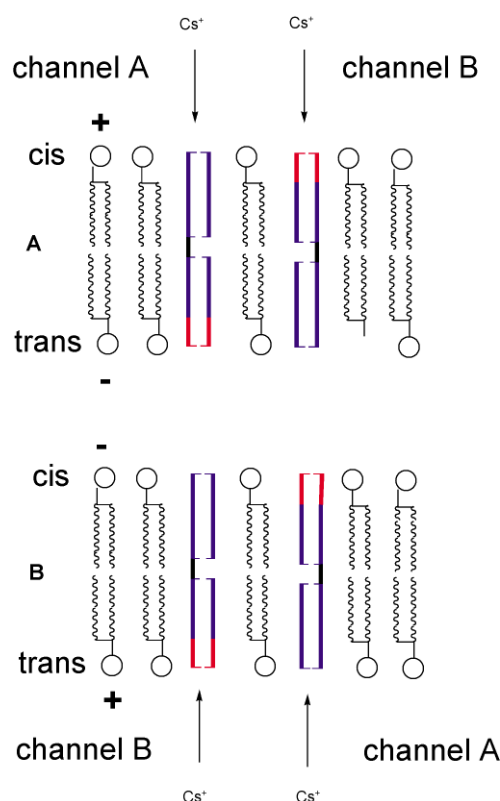
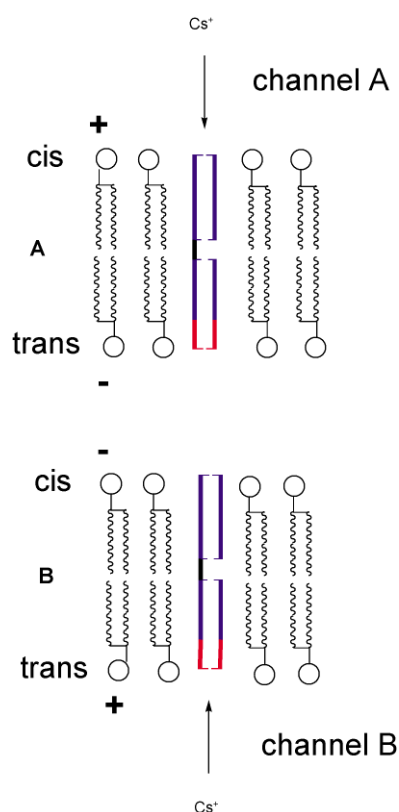
**Fig. 5** Relative permeabilities of compounds **5**, **6**, **7** and gA for monovalent cations in relation to the ionic radius of the cation; —○— **6**; —△— **7**; —◆— **5**; —▼— gA.

The permeability ratios relative to NH<sub>4</sub><sup>+</sup> reveal a tendency from an Eisenman I towards an Eisenman III selectivity with Rb<sup>+</sup> > Cs<sup>+</sup> (see Fig. 5). This behavior is most pronounced for compound **6** and indicates the presence of the donor ether oxygen as a new binding site in the channel.

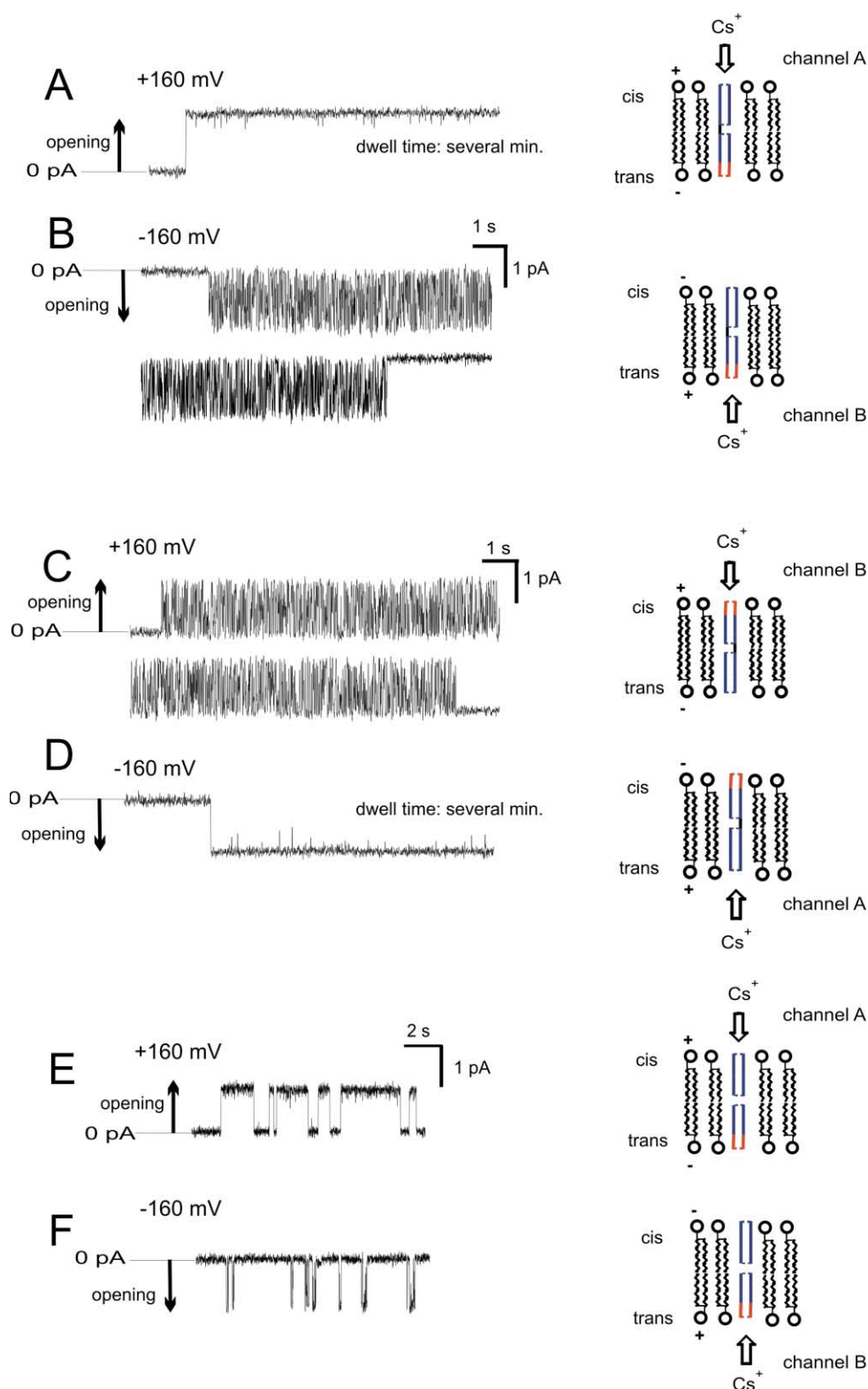
The single channel events for **6** and **7** display dynamic behavior in the millisecond range (openings and closings: 10–20 ms). This occurs with the monomeric compound **6** and the dimeric compound **7**. Therefore, in the case of **6**, the dimer formation and dissociation cannot be the reason for this type of channel opening and closing. Its cause is the presence of the THF amino **ent-1**, which leads to a type of conformational gating.

The asymmetric compound **8** in its extended  $\beta$ -helical conformation can be schematically described as a cylinder-type structure with a gramicidin (grami) end and a THF end. In Figs. 6–8 the THF end is depicted in red and the grami end in blue. If compound **8** is added from the *cis* side to the membrane there are two modes of insertion possible: a *THF-end-first* mode and a *grami-end-first* mode. If the insertion occurs randomly, two types of channel, A and B, should be possible. Let us assume that both channels are different in functionality, so that both are distinguishable by their current traces. In channel type A the cations enter the channel through the grami end and leave it through the THF end. In channel B the cations enter the channel through the THF end and leave it through the grami end. The entrance and the exit site are determined by the membrane potential. Independent of the membrane potential one expects the occurrence of both channel types, A and B, if the insertion occurs randomly (Fig. 6). If the insertion occurs selectively, one type of channel either A or B is possible (Fig. 7). A positive potential at the *cis* side should lead to the observation of channel type A (Fig. 7A). After change of the potential the channel type B should be observed (Fig. 7B).

The addition of compound **8** from the *cis* side to a diphytanoyl phosphatidyl choline (DPhPC) membrane with 1 M CsCl and a membrane potential of +160 mV at the *cis* side gave rise to only one type of channel with a very long dwell time (type 1, Fig. 8A). Change of the potential to –160 mV gave rise to a second type of channel (type 2, Fig. 8B) with a long overall dwell time but with the same short closings and openings that

**Fig. 6** Two channel types A and B resulting from random insertion of **8** into a membrane.**Fig. 7** One channel type either A or B resulting from selective insertion of **8** into a membrane.

were observed for compounds **6** and **7**. Switching the potential back to +160 mV gave rise to the first channel type. This behavior indicates a selective insertion of compound **8** into the membrane. A control experiment to support this hypothesis was done by the addition of compound **8** from the *trans* side. This time, at a membrane potential of +160 mV, the type 2



**Fig. 8** Selective insertion of compound **8** into the phospholipid bilayer. A and B: addition of **8** from the *cis* side; C and D: addition of **8** from the *trans* side, E and F: addition of gA from the *cis* side and **5** from the *trans* side. Current traces were recorded with 1 M CsCl and DPhPC membranes.

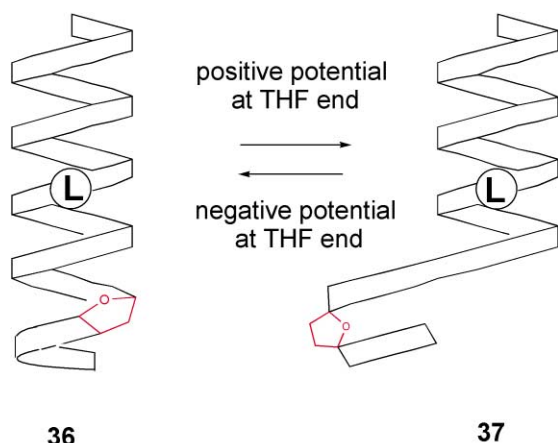
channel (Fig. 8C) and at  $-160$  mV the type 1 channel (Fig. 8D) were observed.

Having proven that a selective insertion had occurred, the question of a *THF-first* or a *grami-first* insertion was addressed next. Towards this end, the monomer compound **5** was added to the *trans* side and gA to the *cis* side. By this token, a predictable *trans* orientation of the THF end and a *cis* orientation of the grami end were accomplished. Mixed **5**–gA channels were formed that showed single channel behavior as depicted in Figs. 7 E and F. Because of the lack of covalent succinyl linker,

the dwell times were shorter, but the characteristics in the channel dynamics observed in Figs. 7A–D were found again. If the cations enter the channel from the grami end, stable grami-type channels are formed (Fig. 8E). If the cations enter from the THF end, channels with ms-dynamics are formed (Fig. 8F). It has to be noted, that after 30 min of experiments no transversal mixing was noticeable. The TBDPS-deprotected compounds **5** and gA gave reproducible mixed channels, while their protected counterparts **6** and gA-TBDPS caused trans-bilayer motion (flip/flop) problems due to their higher

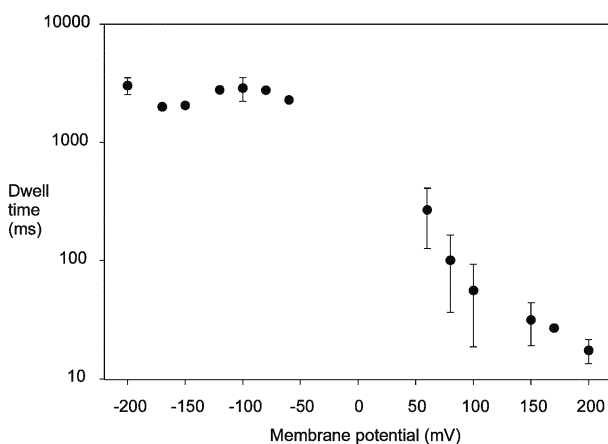
lipophilicity resulting in the gA-TBDPS dimer as the major detectable species.

The asymmetric orientation leads to a potential-dependent function, which can be compared with the voltage gating of biological channels. The ms-dynamics (short closings and openings) occur if a positive potential is applied at the THF end. A type of potential-driven conformational switching (**36**→**37**) around the THF building block in the gA sequence may cause this behavior. As depicted in Fig. 9 the positive potential next to the THF end may lead a switch into a partial  $\beta$ -turn,<sup>15</sup> channel-inactive form of **37**.



**Fig. 9** Schematic representation of a potential-driven conformational switch from an active conformation **36** into an inactive conformation **37**.

The voltage dependence of the gating dynamics for compound **8** was studied over a broader potential range between +200 mV and –200 mV (Fig. 10). Due to the low conductivities in the range between +50 mV and –50 mV no data could be collected in this region. The data show that, for negative potentials, the long lived channel of type A dominates. In the positive range of channel type B a decrease in the dwell time with an increase of the potential is observed.



**Fig. 10** Voltage dependence of the dwell time for **8** in DPhPC and 1 M CsCl.

No significant difference between positive and negative potential was observed for the conductivity of **8**. There is a rectifying effect on the channel-gating dynamics, but not on the channel conductivities. Single-channel currents for  $\text{Cs}^+$ ,  $\text{K}^+$  and  $\text{Na}^+$  are summarized in Table 1. Succ(gA-TBDPS)<sub>2</sub><sup>20</sup> was used as a reference compound. The conductivities of **7** and **8** are of comparable size and smaller than for succ(gA-TBDPS)<sub>2</sub>, with an increased selectivity for  $\text{Cs}^+$  versus  $\text{K}^+$  and  $\text{Na}^+$ .

For a detailed understanding of the potential-dependent function of **8** it will be necessary to perform a detailed

structural NMR-analysis of compound **8** in a membrane-like environment with different electrical potentials. This is a challenging problem which hopefully can be solved. Data from other examples of potential-dependent channel transport by Matile<sup>4f</sup> and Kobuke<sup>4h</sup> will be useful for comparison. The structural reasons for the selective insertion of **8** into the membrane also remain to be clarified.

## Conclusion

In summary, it was found that THF amino acids are valuable building blocks to modulate the ion channel activity of gA. The choice of the THF amino acid and the positions of replacement are crucial. The replacement of positions 11 and 12 by the L-THF amino acid **1** leads to compounds **2**, **3**, and **4** with a drastically reduced channel performance. In contrast, replacement of positions 10 and 11 by the D-THF amino acid **ent-1** gave rise to compounds **5–8** with new and interesting channel properties. The ion selectivity shifts from Eisenman I towards Eisenman III selectivity and the channels display ms-dynamics. Most remarkable is the asymmetric compound **8**, which inserts selectively into a DPhPC membrane and displays voltage-dependent gating dynamics.

## Experimental

### Analytical techniques

**Single channel measurement.** Planar lipid membranes were prepared by painting a solution of soybean lecithin (40%, Avanti Polar Lipids, Alabaster, AL) or diphyanoylphosphatidylcholin (DPhPC, Avanti Polar Lipids, Alabaster, AL) in *n*-decane (25 mg ml<sup>–1</sup>) over the aperture of a polystyrene cuvette with an inner diameter of 0.15 mm. All experiments were performed at ambient temperature. The used electrolyte solutions at a concentration of 1 M each were unbuffered. Probes of monomolecular channels, dissolved in methanol, were added to the *trans* or *cis* side (containing the measuring electrode) of the cuvette. Probes of bimolecular channels, dissolved in methanol, were added to both *trans* and *cis* sides of the cuvette. Current detection and recording was performed with a patch-clamp amplifier Axopatch 200B, a Digidata A/D converter and pClamp6 software (Axon Instruments, Foster City, MA). The acquisition frequency was 5 kHz. The data were filtered with a digital filter at 50 Hz for further analysis.

**NMR-spectroscopy.** NMR spectroscopy was carried out on Bruker AMX300, DPX600, AMX500 and AX400 instruments. All resonances are referenced to residual solvent signals ( $\text{CDCl}_3$ : 7.25 ppm,  $\text{DMSO}-d_6$ : 2.49 ppm).

**HPLC.** HPLC was carried out on a Rainin Dynamax HPLC-System, DA-Detector. Column: Jupiter C5 250 × 4 mm. Method 1 (M1): 65%→85% B in 25 min, A:  $\text{H}_2\text{O}$ ; B: acetonitrile–isopropanol 2 : 1; 0.7 ml min<sup>–1</sup>, 30 °C. Method 2 (M2): 60%→100% B in 20 min, A:  $\text{H}_2\text{O}$  + 0.1% TFA; B: acetonitrile–isopropanol 2 : 1 + 0.1% TFA; 0.7 ml min<sup>–1</sup>, 30 °C. Method 3 (M3) 55%→65% B in 20 min, A:  $\text{H}_2\text{O}$ ; B: acetonitrile–isopropanol 2 : 1; 0.7 ml min<sup>–1</sup>, 30 °C. Method 4 (M4): 70%→100% B in 25 min, A:  $\text{H}_2\text{O}$  + 0.1% TFA; B: acetonitrile–isopropanol 2 : 1 + 0.1% TFA; 0.7 ml min<sup>–1</sup>, 30 °C.

### General procedures

The term “aqueous work-up” refers to an extraction of the organic layer with 1 M  $\text{NaHSO}_4$ , sat.  $\text{NaHCO}_3$  and brine consecutively (a third of the volume of the organic layer each), followed by drying over  $\text{MgSO}_4$  and evaporation. Thin layer chromatography (TLC) and flash column chromatography (FCC) were performed on silica gel (Merck Si60, 40–60  $\mu\text{m}$ ). Columns for FCC were slurry-packed. TLC-spots were visual-



ised by UV-light and by staining with 2% molybdophosphoric acid in EtOH or 2% *p*-methoxy-benzaldehyde in EtOH followed by 20% H<sub>2</sub>SO<sub>4</sub> and heating. Optical rotations are given in 10<sup>-1</sup> deg cm<sup>2</sup> g<sup>-1</sup>.

**Methyl ester deprotection (GP 1).** The methyl ester was dissolved in THF (0.05 M) and cooled to 0 °C. 2.5 eq. LiOH·H<sub>2</sub>O was added as an aqueous solution (1 : 3 v/v H<sub>2</sub>O–THF) and stirred until complete conversion. The mixture was then neutralised with a 2 M HCl solution, THF was evaporated and the residue was partitioned between EtOAc (0.01 M) and 1 M NaHSO<sub>4</sub>. The aqueous layer was extracted with EtOAc, the collected organic fractions were washed with brine and dried over MgSO<sub>4</sub>. After evaporation the residue was dried to constant weight (0.01 Torr). The crude carboxylic acid proved to be sufficiently pure (TLC, NMR, HPLC).

**Z-Group cleavage and benzyl ester deprotection (GP 2 A+B).** The Z-protected compound (or benzyl ester) was dissolved in the indicated solvent (0.02 M) and 10 mol% (2 mol% for benzyl ester deprotection) Pd from Pd/C (5%, Degussa E101 NO/W) was added. The reaction mixture was then degassed, purged with H<sub>2</sub> and hydrogenated under 1 bar at the given temperature. (A): the mixture was vigorously stirred until complete conversion (TLC). The catalyst was then filtered off on Celite® 512 (2 cm). After evaporation and coevaporation with MeOH–toluene the residue was dried to constant weight (0.01 Torr). The crude amine obtained was sufficiently pure. (B): After stirring for 1.5 h the reaction was stopped and the catalyst filtered off on Celite® 512 (2 cm). The fractions were collected, the mixture was reduced to the initial volume and hydrogenated a second time under the same conditions for 2.5 h. After filtration on Celite®, evaporation and coevaporation with MeOH–toluene the residue was dried to constant weight (0.01 Torr). In several cases the crude amines had to be further purified by FCC.

**EDC–HOBt-mediated coupling (GP 3).** The materials were dissolved in CH<sub>2</sub>Cl<sub>2</sub> (indicated concentration) and cooled to 0 °C. EDC, HOBt, and diisopropylethylamine (1.5 eq, 3.0 eq and 3.0 eq respectively, relative to the carboxylic acid) were then added consecutively. After 30 min at 0 °C the reaction mixture was stirred at room temperature until conversion was complete.

**HBTU–HOBt-mediated coupling (GP 4).** The materials were dissolved in CH<sub>2</sub>Cl<sub>2</sub>–DMF 5 : 1 (indicated concentration) and cooled to –15 °C. HBTU, HOBt, and diisopropylethylamine (1.5 eq, 3.0 eq and 3.0 eq respectively, relative to the carboxylic acid) were then added consecutively. After 2 h at –15 °C the reaction mixture was stirred at room temperature until conversion was complete.

**HATU–HOAt-mediated coupling (GP 5).** The materials were dissolved in CH<sub>2</sub>Cl<sub>2</sub>–DMF 5 : 1 (indicated concentration) and cooled to –15 °C. HATU, HOAt, and diisopropylethylamine (1.5 eq, 3.0 eq and 3.0 eq respectively, relative to the carboxylic acid) were then added consecutively. After 2 h at –15 °C the reaction mixture was stirred at room temperature for 22 h.

**Work-up of preparative-HPLC acidic fractions (GP 6).** The collected acidic HPLC-fractions were neutralised with NaHCO<sub>3</sub> (pH paper) and reduced (50%) by evaporation. The obtained mixture was then diluted with CHCl<sub>3</sub> (same volume) and after phase separation, the aqueous layer was extracted four times with CHCl<sub>3</sub>–*i*-PrOH 5 : 1. The collected organic fractions were dried over Na<sub>2</sub>SO<sub>4</sub> and after evaporation the obtained pure compound was dried to constant weight.

#### Experimental procedures and compound characterisation

**(2R)-2-*N*-*t*-Butoxycarbonyl-aminopropanal 10.** 29.5 ml of DIBAL-H (1 M in *n*-hexane, 29.5 mmol) was added dropwise at

–60 °C to a solution of *N*-Boc-D-alanine methyl ester **9** in CH<sub>2</sub>Cl<sub>2</sub> (0.1 M, 150 ml). After 1.5 h the reaction was quenched with *i*-PrOH (15 ml), warmed to room temperature and the resulting mixture was stirred for 3 h with K-Na-tartrate (30 ml, 1 M). The layers were then separated and the aqueous phase was extracted with CH<sub>2</sub>Cl<sub>2</sub> (3 × 20 ml). The collected organic layers were washed with brine and dried over MgSO<sub>4</sub>. After evaporation and FCC (220 g, CH<sub>2</sub>Cl<sub>2</sub>–acetone 25 : 1) 2.0 g (11.6 mmol, 78%) of pure aldehyde **10** was obtained. *R*<sub>f</sub> = 0.68 (CH<sub>2</sub>Cl<sub>2</sub>–acetone 20 : 1); *v*<sub>max</sub>(CHCl<sub>3</sub>)/cm<sup>-1</sup> 3687, 3620, 3441, 3013, 2979, 1734, 1707, 1501, 1451, 1393, 1369, 1236, 1166, 1048, 878, 668; *δ*<sub>H</sub>(75 MHz; CDCl<sub>3</sub>; Me<sub>4</sub>Si) 1.27 (3 H, d, *J* 7.5, Ala 3-H<sub>3</sub>), 1.39 (9H, s, 3 × *t*-Bu-H<sub>3</sub>), 4.09–4.24 (1H, m, 2-H), 4.94–5.11 (1H, m, NH), 9.50 (1H, s, CHO); *δ*<sub>C</sub> (300 MHz; CDCl<sub>3</sub>; Me<sub>4</sub>Si) 14.8 (3-CH<sub>3</sub>), 28.3 (*t*-Bu-CH<sub>3</sub>), 65.4 (2-CH), 199.7 (1-CHO); [*α*]<sub>D</sub> = –33.1, [*α*]<sub>365</sub> = –174.0, (*c* = 1.06, CHCl<sub>3</sub>, 25 °C).

**(2R)-2-*N*-*t*-Butoxycarbonylamino-6-hepten-3-ol 11.** 22 ml of a 1.7 M solution of *t*-BuLi in *n*-hexane were added at –30 °C to 30 ml dry Et<sub>2</sub>O. The mixture was then cooled to –78 °C and 1-bromo-3-butene (1.7 ml, 17.1 mmol) was added dropwise under continuous stirring. In a second flask CuBr·SMe<sub>2</sub> (1.77 g, 8.6 mmol) was dried under vacuum (0.01 Torr) and successively suspended in 50 ml dry Et<sub>2</sub>O. After cooling to –78 °C, the metallation product 1-lithium-3-butene (52 ml Et<sub>2</sub>O–*n*-hexane 1 : 1 mixture) was transferred into the suspension *via* cannula within 1 min. The mixture was warmed to –40 °C and after 5 min a colour change from yellow to dark green indicated the formation of the cuprate. After cooling to –80 °C a solution of the aldehyde **10** (654 mg, 3.8 mmol) in Et<sub>2</sub>O (20 ml) was added dropwise and the resulting mixture was stirred for 1.5 h. The reaction was quenched at room temperature by addition of 50 ml of half sat. NH<sub>4</sub>Cl. After phase separation the organic layer was extracted with aqueous NH<sub>3</sub> (3 × 50 ml), washed with sat. NH<sub>4</sub>Cl (2 × 30 ml), brine and dried over anhydrous MgSO<sub>4</sub>. Evaporation followed by FCC (100 g, CH<sub>2</sub>Cl<sub>2</sub>–acetone 20 : 1) gave 614 mg (2.7 mmol, 71%) of the chromatographically inseparable diastereomeric mixture (**11 syn/anti** 20 : 1, estimated by <sup>1</sup>H NMR upon integration of the C-1 CH<sub>3</sub> signals) as a colourless oil. *R*<sub>f</sub> = 0.32 (CH<sub>2</sub>Cl<sub>2</sub>–acetone 20 : 1); *v*<sub>max</sub>(CHCl<sub>3</sub>)/cm<sup>-1</sup> 3442, 3016, 2980, 2935, 1702, 1502, 1453, 1393, 1368, 1236, 1166, 917; *m/z* (EI) 229.1678 (M<sup>+</sup>. C<sub>12</sub>H<sub>23</sub>NO<sub>3</sub> requires 229.1678).

**(2S,5R,1′R)-2-(1′-*t*-Butoxycarbonylamino-ethyl)-5-hydroxy-methyl-tetrahydrofuran (2S,5R)-12.** The diastereomeric alcohol mixture **11** 614 mg (2.70 mmol) was dissolved in 40 ml CH<sub>2</sub>Cl<sub>2</sub> and cooled to 0 °C. A solution of MCPBA (*m*-chloroperbenzoic acid) in CH<sub>2</sub>Cl<sub>2</sub> (20 ml, 0.26 M) was then added dropwise and the reaction was stirred overnight at room temperature. After dilution with CH<sub>2</sub>Cl<sub>2</sub> (20 ml) the mixture was washed with sat. Na<sub>2</sub>SO<sub>3</sub> (2 × 20 ml), brine (2 × 15 ml) and dried over anhydrous Na<sub>2</sub>SO<sub>4</sub>. The clear solution obtained was reduced to 30 ml and stirred for 3 h with 24 mg (0.10 mmol) of CSA (camphorsulfonic acid). CH<sub>2</sub>Cl<sub>2</sub> (30 ml) was then added and the mixture was washed with sat. NaHCO<sub>3</sub>, brine and dried over MgSO<sub>4</sub>. After evaporation and FCC (90 g MTBE–PE 3 : 1) 250 mg of the *cis*-THF-alcohol **2S,5R-12** (1.02 mmol, 40% over two steps) were obtained as a colourless oil as well as 275 mg of the inseparable diastereomeric mixture (**2S,5S**)-**12**, (**2R,5R**)-**12** and (**2R,2S**)-**12** (20 : 1 : 1 respectively). *R*<sub>f</sub> = 0.38 (MTBE–PE 3 : 1); *v*<sub>max</sub>(CHCl<sub>3</sub>)/cm<sup>-1</sup> 3439, 3009, 2980, 2877, 1699, 1510, 1455, 1368, 1336, 1238, 1165, 1100, 1076, 884; *δ*<sub>H</sub> (300 MHz; CDCl<sub>3</sub>; Me<sub>4</sub>Si) 1.08 (3H, d, *J* 6.7, 2′-H<sub>3</sub>), 1.42 (9H, s, 3 × *t*-Bu-H<sub>3</sub>), 1.67–1.73 (1H, m, 3-HH), 1.82–2.01 (3 H, m, 4-H<sub>2</sub> and 3-HH), 2.15 (1H, s, OH), 3.45 (1H, dd, *J* 10.6 and 7.0, CHOH), 3.65–3.73 (2H, m, CHOH and 2-H), 3.99–4.03 (1H, m, 5-H), 4.62 (1H, d, *J* 8.4, NH); *δ*<sub>C</sub> (75 MHz; CDCl<sub>3</sub>; Me<sub>4</sub>Si) 18.1 (2′-CH<sub>3</sub>), 26.3 (3-CH<sub>2</sub>), 28.3 (*t*-Bu-CH<sub>3</sub>), 28.8 (4-CH<sub>2</sub>), 50.3 (1′-CH), 64.0 (CH<sub>2</sub>OH), 79.4 (*t*-Bu-C(CH<sub>3</sub>)<sub>3</sub>),

80.3 (5-CH), 83.4 (2-CH), 156.6 (C=O, Boc);  $[a]_D = +3.0$ ,  $[a]_{436} = +3.9$  ( $c = 1.10$ ,  $\text{CHCl}_3$ , 25 °C);  $m/z$  (FAB) 246.1711 ( $M + H^+$ ,  $\text{C}_{12}\text{H}_{24}\text{NO}_4$  requires 246.1705).

**(5*S*,2*R*,1'*R*)-5-(1'-*t*-Butoxycarbonylamino-ethyl)-tetrahydrofuran-2-carboxylic acid (Boc-D-THF-OH, **13**).** 280 mg (1.13 mmol) of the alcohol **2S,5*R*-12** were dissolved in 15 ml  $\text{CH}_2\text{Cl}_2$ . Two drops of  $\text{H}_2\text{O}$ , 800 mg (2.48 mmol) of iodobenzene diacetate (IBDA) and 9.0 mg (0.06 mmol) of TEMPO were added and the mixture was vigorously stirred for 3 h at room temperature. After dilution with 40 ml  $\text{CH}_2\text{Cl}_2$  and phase separation, the organic layer was washed with sat.  $\text{Na}_2\text{S}_2\text{O}_3$  (4 × 15 ml) until reduction of the residual IBDA was complete (KI/starch paper). The organic layer was then extracted with sat.  $\text{NaHCO}_3$  (4 × 10 ml) and after phase separation the collected aqueous fractions were acidified with 2 M HCl to pH = 3 (pH paper) and extracted with  $\text{CH}_2\text{Cl}_2$  (4 × 30 ml). The organic layer was washed with brine and dried with  $\text{MgSO}_4$ . After evaporation and drying to constant weight (0.01 Torr) 278 mg (1.07 mmol, 95%) of the pure carboxylic acid **13** was obtained.  $R_f = 0.31$  ( $\text{CHCl}_3$ -MeOH- $\text{HCO}_2\text{H}$  200 : 10 : 1);  $\nu_{\text{max}}(\text{CHCl}_3)/\text{cm}^{-1}$  3442, 3100, 2975, 2932, 2877, 1748, 1691, 1600, 1510, 1430, 1395, 1380, 1343, 1244, 1164, 1086, 840, 770;  $\delta_{\text{H}}$  (300 MHz;  $\text{CDCl}_3$ ;  $\text{Me}_4\text{Si}$ ) 1.18 (3H, d,  $J$  6.8, 2'- $\text{H}_3$ ), 1.44 (9H, s, 3 × *t*-Bu- $\text{H}_3$ ), 1.66 (1H, dddd,  $J$  12.3, 10.4, 9.1 and 8.1, 3- $\text{HH}$ ), 1.93 (1H, dddd,  $J$  12.5, 7.8, 6.2 and 3.3, 3- $\text{HH}$ ), 2.09 (1H, dddd,  $J$  12.7, 8.1, 4.0 and 3.3, 4- $\text{HH}$ ), 2.30 (1H, dddd,  $J$  12.7, 10.4, 9.0 and 7.8, 4- $\text{HH}$ ), 3.65 (1H, dq,  $J$  6.8 and 3.2, 1'-H), 3.98 (1H, ddd,  $J$  9.1, 6.2 and 3.3, 5-H), 4.42 (1H, dd,  $J$  9.0 and 4.0, 2-H);  $\delta_{\text{C}}$  (75 MHz;  $\text{CDCl}_3$ ;  $\text{Me}_4\text{Si}$ ) 19.5 (2'-CH), 25.5 (4- $\text{CH}_2$ ), 28.6 (*t*-Bu- $\text{CH}_3$ ), 85.8 (5-CH), 158.9 (C=O, Boc).  $[a]_D = +14.0$ ,  $[a]_{365} = +43.8$  ( $c = 1.80$  in EtOH, 25 °C);  $m/z$  (FAB) 260.1511 ( $M + H^+$ ,  $\text{C}_{12}\text{H}_{22}\text{NO}_5$  requires 260.1498).

**Z-L-THF-Trp-Leu-Trp-2-(*t*-butyldiphenylsilyloxy)-ethylamide **22**.** 1.70 g (1.85 mmol) tripeptide **21** were *N*-deprotected according to GP 2 (MeOH, rt, 2 h;  $R_f = 0.38$  in  $\text{CHCl}_3$ -MeOH- $\text{NH}_3$ (aq.) 100 : 10 : 1). The crude product and 390 mg (1.33 mmol) of carboxylic acid **20** were then combined and coupled according to GP 3 (0.03 M, 3 h). After aqueous work-up, FCC (120 g, 3% MeOH in  $\text{CHCl}_3$ ) gave 1.19 g (1.12 mmol, 84%) of ethylamide **22** as a colourless foam.  $R_f = 0.49$  ( $\text{CHCl}_3$ -MeOH- $\text{HCO}_2\text{H}$  100 : 10 : 5);  $\delta_{\text{H}}$  (300 MHz;  $\text{CDCl}_3$ ;  $\text{Me}_4\text{Si}$ ) 0.60 (3H, d,  $J$  6.0, Leu  $\delta$ - $\text{H}_3$ ), 0.64 (3H, d,  $J$  6.4, Leu  $\delta$ - $\text{H}_3$ ), 0.97 (12H, s, 2'- $\text{H}_3$  and 3 × *t*-Bu- $\text{H}_3$ ), 1.08–1.19 (2H, m, Leu  $\beta$ - $\text{H}_2$ ), 1.20–1.40 (2H, m, Leu  $\gamma$ -H and THF 3- or 4- $\text{CHH}$ ), 1.58–1.77 (1H, m, THF 3- or 4- $\text{CHH}$ ), 1.83–2.00 (2H, m, THF 3- or 4- $\text{CHH}$ ), 2.90 (1H, dd,  $J$  13.9 and 10.2, Trp  $\beta$ - $\text{HH}$ ), 3.05–3.21 (3H, m, 1 × Trp  $\beta$ - $\text{HH}$  and 2 × Trp  $\beta$ - $\text{HH}$ ), 3.28 (2H, t,  $J$  7.4, 1- $\text{H}_2$ ), 3.60 (2H, t,  $J$  6.4, 2- $\text{H}_2$ ), 3.62–3.75 (2H, m, THF 1'-CH and THF 5-H), 4.09–4.27 (2H, m, THF 2-H and Leu  $\alpha$ -H), 4.37–4.57 (2H, m, 2 × Trp  $\alpha$ -H), 4.94 (1H, d,  $J$  12.4, Z- $\text{HH}$ ), 5.05 (1H, d,  $J$  12.4, Z- $\text{HH}$ ), 6.87–6.95 (2H, m, 2 × ar. CH), 6.98–7.08 (3H, m, 3 × ar. CH), 7.12 (1H, s, ar. CH), 7.22–7.34 (7H, m, 7 × ar. CH), 7.36–7.45 (6H, m, 6 × ar. CH), 7.48–7.69 (7H, m, THF NH and 6 × ar. CH), 7.93–8.05 (2H, m, Leu NH and AE NH), 8.15 (1H, d,  $J$  8.7, Trp NH), 8.35 (1H, d,  $J$  7.2, Trp NH), 10.78 (1H, s, indole NH), 10.80 (1H, s, indole NH);  $[a]_D = +38.3$ ,  $[a]_{365} = +134.2$  ( $c = 1.2$ ,  $\text{CHCl}_3$ , 20 °C); HPLC: 12.8 min (M1);  $m/z$  (FAB) 1060.5379 ( $M + H^+$ ,  $\text{C}_{61}\text{H}_{74}\text{N}_7\text{O}_8\text{Si}$  requires 1060.5368).

**Z-Trp-D-Leu-L-THF-Trp-D-Leu-Trp-2-(*t*-butyldiphenylsilyloxy)-ethylamide **24**.** 627 mg (1.35 mmol) of dipeptide **23** were C-deprotected according to GP 1 and 1.06 g (1.00 mmol) of peptide **22** were *N*-deprotected according to GP 2 (MeOH, 40 °C, 3 h,  $R_f = 0.26$  in  $\text{CHCl}_3$ -MeOH- $\text{NH}_3$ (aq.) 100 : 10 : 1). The crude products were combined and coupled according to GP 4 (0.02 M, 1 h). After aqueous work-up, FCC (123 g 3%

MeOH in  $\text{CHCl}_3 \rightarrow 5\%$  MeOH in  $\text{CHCl}_3 + 0.5\%$   $\text{HCO}_2\text{H}$ ) gave 1.02 g (0.75 mmol, 75%) of peptide **24** as a colourless foam.  $R_f = 0.49$  ( $\text{CHCl}_3$ -MeOH- $\text{NH}_3$ (aq.) 100 : 10 : 1);  $\delta_{\text{H}}$  (400 MHz;  $\text{DMSO}-d_6$ ;  $\text{Me}_4\text{Si}$ ) (ppm): 0.63 (3H, d,  $J$  6.3, Leu  $\delta$ - $\text{H}_3$ ), 0.67 (d,  $J$  6.6, 3H, Leu  $\delta$ - $\text{H}_3$ ), 0.80 (d,  $J$  5.9, 3H, Leu  $\delta$ - $\text{H}_3$ ), 0.83 (3H, d,  $J$  5.9, Leu  $\delta$ - $\text{H}_3$ ), 0.94–1.03 (12H, m, THF C-2'- $\text{H}_3$  and 3 × *t*-Bu- $\text{H}_3$ ), 1.03–1.12 (1H, m, Leu  $\gamma$ -H), 1.13–1.22 (2H, m, Leu  $\beta$ - $\text{H}_2$ ), 1.24–1.36 (1H, m, THF 4- $\text{HH}$ ), 1.39–1.54 (3H, m, Leu  $\beta$ - $\text{H}_2$  and Leu  $\gamma$ -H), 1.64–1.74 (1H, m, THF 3- $\text{HH}$ ), 1.85–1.94 (2H, m, THF 4- $\text{HH}$  and THF 3- $\text{HH}$ ), 2.65 (1H, dd,  $J$  8.8 and 14.0, Trp  $\beta$ - $\text{HH}$ ), 2.91 (1H, dd,  $J$  10.2 and 14.0, Trp  $\beta$ - $\text{HH}$ ), 3.05–3.21 (4H, m, Trp  $\beta$ - $\text{HH}$  and 3 × Trp  $\beta$ - $\text{HH}$ ), 3.21–3.28 (2H, m, AE 2- $\text{H}_2$ ), 3.61 (2H, t,  $J$  6.6, AE 1- $\text{H}_2$ ), 3.67–3.75 (1H, m, THF 5-H), 3.87 (1H, dt,  $J$  2.3 and 7.0, THF 1'-H), 4.12 (1H, t,  $J$  5.5, THF 2-H), 4.19–4.27 (1H, m, Leu  $\alpha$ -H), 4.37–4.53 (4H, m, Leu  $\alpha$ -H and 3 × Trp  $\alpha$ -H), 4.94 (1H, d,  $J$  12.4, Z- $\text{HH}$ ), 5.05 (1H, d,  $J$  12.4, Z- $\text{HH}$ ), 6.89–6.97 (3H, m, 3 × ar. CH), 6.99–7.06 (3H, m, 3 × ar. CH), 7.08 (1H, s, ar. CH), 7.14 (2H, d,  $J$  15.6, 2 × ar. CH), 7.26–7.33 (8H, m, 8 × ar. CH), 7.38–7.48 (7H, m, Trp NH and 6 × ar. CH), 7.53–7.65 (7H, m, 7 × ar. CH), 7.98 (1H, t,  $J$  5.8, AE NH), 8.06 (1H, d,  $J$  7.9, Leu NH), 8.09–8.15 (2H, m, Leu NH and Trp NH), 8.18 (1H, d,  $J$  8.6, THF NH), 8.38 (1H, d,  $J$  7.3, Trp NH), 10.73 (1H, s, indole NH), 10.79 (2H, s, indole NH);  $[a]_D = +19.5$ ,  $[a]_{365} = +90.8$  ( $c = 1.3$ ,  $\text{CHCl}_3$ , T = 20 °C); HPLC: 14.4 min (M1);  $m/z$  (ESI) 1381.8 ( $M + \text{Na}^+$ ,  $\text{C}_{78}\text{H}_{94}\text{N}_{10}\text{NaO}_{10}\text{Si}$  requires 1381.7).

**Z-Ala-D-Val-Val-D-Val-Trp-D-Leu-L-THF-Trp-D-Leu-Trp-2-(*t*-butyldiphenylsilyloxy)-ethylamide **15**.** 480 mg (0.9 mmol) of tetrapeptide **25** were C-deprotected according to GP 2 and 800 mg (0.60 mmol) of peptide **24** were *N*-deprotected according to GP 1 (MeOH, 40 °C, 3 h;  $R_f = 0.12$  in  $\text{CHCl}_3$ -MeOH- $\text{HCO}_2\text{H}$  100 : 10 : 1). The crude products were combined and coupled according to GP 4 (0.01 M, 2 h). After aqueous work-up, FCC (120 g, 3% MeOH in  $\text{CHCl}_3$ ) gave 828 mg (0.48 mmol 81%) of product **15** as a colourless foam.  $R_f = 0.55$  ( $\text{CHCl}_3$ -MeOH- $\text{HCO}_2\text{H}$  100 : 10 : 1);  $\delta_{\text{H}}$  (500 MHz;  $\text{DMSO}-d_6$ ;  $\text{Me}_4\text{Si}$ ) 0.56–0.63 (9H, m, 2 × Leu  $\delta$ - $\text{H}_3$  and Val  $\gamma$ - $\text{H}_3$ ), 0.64–0.67 (6H, m, Leu  $\delta$ - $\text{H}_3$  and Val  $\gamma$ - $\text{H}_3$ ), 0.69 (3H, d,  $J$  6.2, Leu  $\delta$ - $\text{H}_3$ ), 0.77–0.87 (12H, m, 4 × Val  $\gamma$ - $\text{H}_3$ ), 0.99 (9H, s, 3 × *t*-Bu- $\text{H}_3$ ), 0.95–1.05 (4H, m, THF 2'- $\text{H}_3$  and Leu  $\gamma$ -H), 1.20 (3H, d,  $J$  6.9, Ala  $\beta$ - $\text{H}_3$ ), 1.08–1.23 (3H, m, Leu  $\beta$ - $\text{H}_2$  and Leu  $\gamma$ -H), 1.28–1.39 (2H, m, THF 4- $\text{HH}$  and THF 3- $\text{HH}$ ), 1.40–1.49 (2H, m, Leu  $\beta$ - $\text{H}_2$ ), 1.66–1.74 (1H, m, THF 4- $\text{HH}$ ), 1.84–1.94 (2H, m, Val  $\beta$ -H and THF 3- $\text{HH}$ ), 1.96–2.06 (2H, m, 2 × Val  $\beta$ -H), 2.89–3.01 (2H, m, 2 × Trp  $\beta$ - $\text{HH}$ ), 3.22–3.08 (4H, m, Trp  $\beta$ - $\text{HH}$  and 3 × Trp  $\beta$ - $\text{HH}$ ), 3.23–3.31 (2H, m, AE 2- $\text{H}_2$ ), 3.61 (2H, t,  $J$  6.4, AE 1- $\text{H}_2$ ), 3.68–3.75 (1H, m, THF 5-H), 3.85–3.93 (1H, m, THF 1'-H), 4.09–4.19 (2H, m, THF 2-H and Ala  $\alpha$ -H), 4.20–4.28 (3H, m, Val  $\alpha$ -H and 2 × Leu  $\alpha$ -H), 4.29–4.36 (2H, m, 2 × Val  $\alpha$ -H), 4.41–4.47 (1H, m, Trp  $\alpha$ -H), 4.51 (1H, dd,  $J$  7.3 and 14.2, Trp  $\alpha$ -H), 4.57 (1H, dd,  $J$  7.7 and 14.1, Trp  $\alpha$ -H), 4.98 (1H, d,  $J$  13.0, Z- $\text{HH}$ ), 5.01 (1H, d,  $J$  13.0, Z- $\text{HH}$ ), 6.89–6.97 (3H, m, 3 × ar. CH), 6.99–7.06 (3H, m, 3 × ar. CH), 7.08 (1H, s, ar. CH), 7.15 (2H, d,  $J$  7.1, 2 × ar. CH), 7.26–7.37 (8H, m, 8 × ar. CH), 7.38–7.48 (7H, m, 6 × ar. CH and Ala NH), 7.53–7.60 (3H, m, 3 × ar. CH), 7.62 (4H, d,  $J$  6.6, 4 × ar. CH), 7.83 (1H, d,  $J$  9.9, NH), 7.86 (1H, d,  $J$  9.2, NH), 7.90 (1H, d,  $J$  8.7, THF NH), 7.92 (1H, d,  $J$  8.7, NH), 7.99 (1H, t,  $J$  4.8, AE NH), 8.02 (1H, d,  $J$  8.3, NH), 8.10–8.17 (2H, m, 2 × Trp NH), 8.23 (1H, d,  $J$  8.3, NH), 8.46 (1H, d,  $J$  7.1, Trp NH), 10.74 (1H, s, indole NH), 10.79 (2H, s, indole NH); HPLC: 16.6 min (M1);  $m/z$  (ESI) 1728.6 ( $M + H^+$ ,  $\text{C}_{96}\text{H}_{127}\text{N}_{14}\text{O}_{14}\text{Si}$  requires 1727.9).

**H-Ala-D-Val-Val-D-Val-Trp-D-Leu-L-THF-Trp-D-Leu-Trp-2-(*t*-butyldiphenylsilyloxy)-ethylamide **15a**.** 400 mg (231  $\mu\text{mol}$ ) of peptide **15** were *N*-deprotected according to GP 2A (EtOH, 50 °C). After filtration, evaporation, FCC (20 g,  $\text{CHCl}_3$ -MeOH- $\text{NH}_3$  100 : 10 : 1) of the crude product gave 250 mg (157  $\mu\text{mol}$ , 68%) of amine **15a**.  $R_f = 0.16$  ( $\text{CHCl}_3$ -MeOH- $\text{HCO}_2\text{H}$

100 : 10 : 1). HPLC: 11.8 min (M2).  $m/z$  (ESI) 1615.6 (M + Na<sup>+</sup>). C<sub>88</sub>H<sub>120</sub>N<sub>14</sub>NaO<sub>12</sub>Si requires 1615.9).

**HCO-Val-Gly-Ala-D-Leu-Ala-D-Val-Val-D-Val-Trp-D-Leu-L-THF-Trp-D-Leu-Trp-2-(*t*-butyldiphenylsilyloxy)-ethylamide 3.** 55.0 mg (137  $\mu$ mol) of tetrapeptide **14** were C-deprotected according to GP 1 ( $R_f$  = 0.26 in CHCl<sub>3</sub>-MeOH-HCO<sub>2</sub>H 100 : 10 : 1) and then coupled with 100 mg (62.7  $\mu$ mol) of amine **15a** according to GP 4 (6.0 mm, 3 h). After aqueous work-up and FCC (12 g, CHCl<sub>3</sub>-MeOH-HCO<sub>2</sub>H 200 : 10 : 1) 90.0 mg (46.0  $\mu$ mol, 73%) of peptide **3** were obtained as a colourless foam. Prior to single-channel measurement, purification of 10.0 mg of compound **3** by preparative HPLC (250 mm Rainin C 8, 60→99% B in 20 min, A: H<sub>2</sub>O; B: acetonitrile-isopropanol 2 : 1; 10 ml min<sup>-1</sup>) afforded 8.0 mg (80%) of purest product.  $R_f$  = 0.52 (CHCl<sub>3</sub>-MeOH-HCO<sub>2</sub>H 100 : 10 : 1);  $\delta_H$  (400 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.55–0.63 (9H, m, 3 × Leu  $\delta$ -H<sub>3</sub>), 0.64–0.71 (9H, m, 3 × Leu  $\delta$ -H<sub>3</sub>), 0.77–0.88 (24H, m, 8 × Val  $\gamma$ -H<sub>3</sub>), 0.95–1.05 (13H, m, THF 2'-H<sub>3</sub>, 3 × *t*-Bu-H<sub>3</sub> and Leu  $\gamma$ -H), 1.19 (3H, d, *J* 4.3, Ala  $\beta$ -H<sub>3</sub>), 1.21 (3H, d, *J* 4.3, Ala  $\beta$ -H<sub>3</sub>), 1.06–1.25 (4H, m, Leu  $\gamma$ -H, Leu  $\beta$ -H<sub>2</sub> and THF 4-*HH*), 1.27–1.37 (2H, m, THF 3-*HH* and Leu  $\beta$ -*HH*), 1.39–1.49 (3H, m, Leu  $\beta$ -*HH* and Leu  $\beta$ -H<sub>2</sub>), 1.50–1.60 (1H, m, Val  $\beta$ -H), 1.65–1.75 (1H, m, THF 4-*HH*), 1.83–2.07 (5H, m, THF 3-*HH*, Leu  $\gamma$ -H and 3 × Val  $\beta$ -H), 2.86–3.02 (2H, m, Trp  $\beta$ -H<sub>2</sub>), 3.07–3.20 (4H, m, 2 × Trp  $\beta$ -H<sub>2</sub>), 3.21–3.31 (2H, m, AE 1-H<sub>2</sub>), 3.60 (2H, t, *J* 6.4, AE 1-H<sub>2</sub>), 3.67–3.76 (3H, m, Gly  $\alpha$ -H<sub>2</sub> and THF 5-H), 3.89 (1H, dd, *J* 14.7 and 7.4, THF 1'-H), 4.12 (1H, t, *J* 5.8, Val  $\alpha$ -H), 4.17–4.34 (9H, m, 3 × Val  $\alpha$ -H, 3 × Leu  $\alpha$ -H, 2 × Ala  $\alpha$ -H and THF 2-H), 4.40–4.58 (3H, m, 3 × Trp  $\alpha$ -H), 6.88–6.96 (3H, m, 3 × ar. CH), 6.98–7.05 (3H, m, 3 × ar. CH), 7.06 (1H, d, *J* 1.9, ar. CH), 7.12 (1H, d, *J* 1.9, ar. CH), 7.14 (1H, d, *J* 1.9, ar. CH), 7.26–7.32 (3H, m, 3 × ar. CH), 7.36–7.47 (6H, m, 6 × ar. CH), 7.51–7.64 (7H, m, 7 × ar. CH), 7.85 (1H, d, *J* 8.6, NH), 7.88–8.02 (7H, m, 7 × NH), 8.04 (1H, s, CHO), 8.09–8.26 (5H, m, 5 × NH), 8.31 (1H, t, *J* 5.6, AE NH), 8.46 (1H, d, *J* 7.0, Trp NH), 10.71 (1H, s, indole NH), 10.77 (2H, s, indole NH); HPLC: 14.3 min (M1);  $m/z$  (ESI) 1962.7 (M + H<sup>+</sup>). C<sub>105</sub>H<sub>149</sub>N<sub>18</sub>O<sub>17</sub>Si requires 1962.1).

**<sup>11,12</sup>[L-THF]-ga 2.** 20.0 mg (10.2  $\mu$ mol) of peptide **3** were dissolved in 20 ml CH<sub>2</sub>Cl<sub>2</sub>-DMF 10 : 1. Under stirring at rt 1.8 ml of a 5% solution of HF in CH<sub>3</sub>CN were then added dropwise. After 45 min the reaction mixture was diluted with 20 ml CH<sub>2</sub>Cl<sub>2</sub>, carefully washed with sat. NaHCO<sub>3</sub> (2 × 10 ml) and brine (1 × 10 ml) and dried over Na<sub>2</sub>SO<sub>4</sub>. After evaporation, 20.0 mg of the crude peptide **2** were isolated and purified by preparative HPLC (250 mm Rainin C 8, 60%→99% B in 20 min, A: H<sub>2</sub>O; B: acetonitrile-isopropanol 2 : 1; 10 ml min<sup>-1</sup>). Pure product **2** (15.0 mg 8.70  $\mu$ mol, 85%) was isolated as a colourless solid.  $R_f$  = 0.40 (CHCl<sub>3</sub>-MeOH-HCO<sub>2</sub>H 100 : 10 : 1);  $\delta_H$  (400 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.56–0.64 (9H, m, 3 × Leu  $\delta$ -H<sub>3</sub>), 0.65–0.73 (9H, m, 3 × Leu  $\delta$ -H<sub>3</sub>), 0.77–0.88 (24H, m, 8 × Val  $\gamma$ -H<sub>3</sub>), 0.99 (3H, d, *J* 6.6, THF 2'-H<sub>3</sub>), 0.95–1.08 (2H, m, 2 × Leu  $\gamma$ -H), 1.11–1.28 (5H, m, Leu  $\gamma$ -H and 2 × Leu  $\beta$ -H<sub>2</sub>), 1.19 (3H, d, *J* 7.6, Ala  $\beta$ -H<sub>3</sub>), 1.21 (3H, d, *J* 7.6, Ala  $\beta$ -H<sub>3</sub>), 1.28–1.41 (3H, m, Leu  $\beta$ -H<sub>2</sub> and THF 4-*HH*), 1.43–1.51 (1H, m, THF 3-*HH*), 1.52–1.61 (1H, m, Val  $\beta$ -H), 1.69–1.79 (1H, m, THF 4-*HH*), 1.86–2.08 (5H, m, THF 3-*HH*, Leu  $\gamma$ -H and 3 × Val  $\beta$ -H), 2.89 (1H, dd, *J* 14.5 and 9.9, Trp  $\beta$ -*HH*), 2.98 (1H, dd, *J* 14.0 and 8.4, Trp  $\beta$ -*HH*), 3.06–3.24 (6H, m, Trp  $\beta$ -*HH*, 3 × Trp  $\beta$ -*HH* and AE 2-H<sub>2</sub>), 3.35–3.43 (1H, m, AE 1-*HH*), 3.66–3.80 (3H, m, Gly  $\alpha$ -H<sub>2</sub> and THF 5-H), 3.90 (1H, dd, *J* 15.4 and 7.7, THF 1'-H), 4.13–4.33 (10H, m, 4 × Val  $\alpha$ -H, 3 × Leu  $\alpha$ -H, 2 × Ala  $\alpha$ -H and THF 2-H), 4.43 (1H, dt, *J* 4.9 and 9.1, Trp  $\alpha$ -H), 4.51 (2H, dd, *J* 9.1 and 4.9, 2 × Trp  $\alpha$ -H), 4.67–4.73 (1H, m, AE 1-*HH*), 6.88–6.97 (3H, m, 3 × ar. CH), 6.98–7.05 (3H, m, 3 × ar. CH), 7.08 (1H, d, *J* 1.9, ar. CH), 7.09 (1H, d, *J* 1.9, ar. CH), 7.16 (1H, d, *J* 1.9, ar. CH), 7.25–7.34 (3H, m, 3 × ar. CH), 7.52–7.62 (3H, m, 3 × ar. CH), 7.89 (1H, t, *J* 5.6, AE NH), 7.93–8.12 (8H, m, 7 × NH and CHO), 8.17 (1H, d, *J* 8.6, Trp

NH), 8.22–8.31 (3H, m, 3 × NH), 8.35 (1H, t, *J* 5.8, Gly NH), 8.47 (1H, d, *J* 7.6, Trp NH), 10.73 (1H, s, indole NH), 10.79 (2H, s, indole NH); HPLC: 10.9 min (M3);  $m/z$  (ESI) 1745.9 (M + Na<sup>+</sup>). C<sub>89</sub>H<sub>130</sub>N<sub>18</sub>NaO<sub>17</sub> requires 1746.0).

**Z-Ala-D-Leu-Ala-D-Val-Val-D-Val-Trp-D-Leu-L-THF-Trp-D-Leu-Trp-2-(*t*-butyldiphenylsilyloxy)-ethylamide 31.** 43.0 mg (123  $\mu$ mol) of dipeptide **18** were C-deprotected according to GP 1 and coupled with 60.0 mg (37.7  $\mu$ mol) of amine **15a** according to GP 4 (6.0 mm, 2.5 h). After aqueous work-up and FCC (12 g, 3% MeOH in CHCl<sub>3</sub>→4% MeOH in CHCl<sub>3</sub> + 0.4% HCO<sub>2</sub>H) 65.0 mg (34.0  $\mu$ mol, 90%) of peptide **31** were obtained as a colourless foam.  $R_f$  = 0.52 (CHCl<sub>3</sub>-MeOH-HCO<sub>2</sub>H 100 : 10 : 1);  $\delta_H$  (500 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.57–0.63 (9H, m, 3 × Leu  $\delta$ -H<sub>3</sub>), 0.65 (6H, d, *J* 6.4, 2 × Leu  $\delta$ -H<sub>3</sub>), 0.70 (3H, d, *J* 6.6, Leu  $\delta$ -H<sub>3</sub>), 0.76–0.86 (18H, m, 6 × Val  $\gamma$ -H<sub>3</sub>), 0.95–1.01 (13H, m, 3 × *t*-Bu-H<sub>3</sub>, THF 2'-H<sub>3</sub> and Leu  $\gamma$ -H), 1.08–1.16 (3H, m, Leu  $\gamma$ -H and Leu  $\beta$ -H<sub>2</sub>), 1.17–1.23 (7H, m, 2 × Ala  $\beta$ -H<sub>3</sub> and THF 3-*HH*), 1.28–1.35 (2H, m, THF 4-*HH* and Leu  $\gamma$ -H), 1.42–1.49 (4H, m, Leu  $\beta$ -H<sub>2</sub>), 1.51–1.59 (1H, m, Val  $\beta$ -H), 1.66–1.75 (1H, m, THF 4-*HH*), 1.84–1.94 (2H, m, Leu  $\gamma$ -H, and THF 3-*HH*), 1.95–2.05 (2H, m, Val  $\beta$ -H), 2.88–3.00 (2H, m, 2 × Trp  $\beta$ -*HH*), 3.07–3.21 (4H, m, Trp  $\beta$ -*HH* and 3 × Trp  $\beta$ -*HH*), 3.23–3.33 (2H, m, AE 2-H<sub>2</sub>), 3.61 (2H, t, *J* 6.2, AE 1-H<sub>2</sub>), 3.71 (1H, dd, *J* 15.1 and 7.1, THF 5-H), 3.89 (1H, dd, *J* 14.4 and 7.5, THF 1'-H), 4.07 (1H, t, *J* 7.2, Ala  $\alpha$ -H), 4.14 (1H, t, *J* 5.6, THF 2-H), 4.19–4.28 (4H, m, Val  $\alpha$ -H and 3 × Leu  $\alpha$ -H), 4.29–4.35 (3H, m, Ala  $\alpha$ -H and 2 × Val  $\alpha$ -H), 4.41–4.47 (1H, m, Trp  $\alpha$ -H), 4.45 (1H, dd, *J* 14.7 and 7.1, Trp  $\alpha$ -H), 4.57 (1H, dd, *J* 14.3 and 7.0, Trp  $\alpha$ -H), 4.99 (1H, d, *J* 12.4, Z-*HH*), 5.02 (1H, d, *J* 12.1, Z-*HH*), 6.88–6.96 (3H, m, 3 × ar. CH), 6.98–7.05 (3H, m, 3 × ar. CH), 7.07 (1H, d, *J* 1.8, ar. CH), 7.13 (1H, d, *J* 1.7, ar. CH), 7.16 (1H, d, *J* 1.8, ar. CH), 7.26–7.36 (8H, m, 8 × ar. CH), 7.39–7.47 (7H, m, 6 × ar. CH and Ala NH), 7.53–7.60 (3H, m, 3 × ar. CH), 7.61–7.65 (4H, m, 4 × ar. CH), 7.77 (1H, d, *J* 8.7, NH), 7.86 (1H, d, *J* 9.9, NH), 7.88–7.93 (2H, m, NH and THF NH), 7.96 (1H, d, *J* 7.1, NH), 7.99 (1H, t, *J* 6.0, AE NH), 8.02 (1H, d, *J* 8.5, NH), 8.09–8.17 (3H, m, NH and 2 × Trp NH), 8.23 (1H, d, *J* 8.3, NH), 8.46 (1H, d, *J* 7.1, Trp NH), 10.73 (1H, s, indole NH), 10.79 (2H, s, indole NH); HPLC: 10.7 min (M4);  $m/z$  (ESI) 1934.3 (M + Na<sup>+</sup>). C<sub>105</sub>H<sub>142</sub>N<sub>16</sub>NaO<sub>16</sub>Si requires 1934.0).

**H-Ala-D-Leu-Ala-D-Val-Val-D-Val-Trp-D-Leu-L-THF-Trp-D-Leu-Trp-2-(*t*-butyldiphenylsilyloxy)-ethylamide 31a.** 124 mg (64.9  $\mu$ mol) of peptide **31** were N-deprotected according to GP 2A (EtOH, 50 °C). After filtration and evaporation FCC (15 g, CHCl<sub>3</sub>-MeOH-NH<sub>3</sub>(conc.) 100 : 10 : 1) gave 70.0 mg (39.4  $\mu$ mol, 61%) of amine **31a**.  $R_f$  = 0.28 (CHCl<sub>3</sub>-MeOH-NH<sub>3</sub>(conc.) 100 : 10 : 1); HPLC: 7.2 min (M4).  $m/z$  (ESI) 1799.4 (M + Na<sup>+</sup>). C<sub>97</sub>H<sub>136</sub>N<sub>16</sub>NaO<sub>14</sub>Si requires 1800.0).

**Succ-1-(1'-*t*-butyldiphenylsilyloxy)-<sup>11,12</sup>[L-THF]-gramicidin A)-4-(Val-Gly-OMe)-diamide 32.** 30.0 mg (56.2  $\mu$ mol) of benzyl ester **17** were C-deprotected according to GP 2A. The crude product was coupled with amine **31a** (30.0 mg, 17.0  $\mu$ mol) according to GP 4 (3.0 mm, 3 h). After aqueous work-up FCC (5 g, 3% MeOH in CHCl<sub>3</sub>→4% MeOH in CHCl<sub>3</sub> + 0.4% HCO<sub>2</sub>H) gave 31.0 mg (14.1  $\mu$ mol, 83%) of pure diamide **32** as a colourless solid.  $R_f$  = 0.42 (CHCl<sub>3</sub>-MeOH-HCO<sub>2</sub>H 100 : 10 : 1);  $\delta_H$  (600 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.52–0.71 (18H, m, 6 × Leu  $\delta$ -H<sub>3</sub>), 0.74–0.89 (30H, m, 10 × Val  $\gamma$ -H<sub>3</sub>), 0.91–1.02 (13H, m, 3 × *t*-Bu-H<sub>3</sub>, Leu  $\gamma$ -H and THF 2'-H<sub>3</sub>), 1.03–1.13 (3H, m, Leu  $\gamma$ -H and Leu  $\beta$ -H<sub>2</sub>), 1.18 (3H, d, *J* 6.8, Ala  $\beta$ -H<sub>3</sub>), 1.19 (3H, d, *J* 6.8, Ala  $\beta$ -H<sub>3</sub>), 1.16–1.23 (1H, m, THF 3-*HH*), 1.25–1.35 (2H, m, THF 4-*HH* and Leu  $\gamma$ -H), 1.41–1.51 (4H, m, 2 × Leu  $\beta$ -H<sub>2</sub>), 1.52–1.59 (1H, m, Val  $\beta$ -H), 1.64–1.74 (1H, m, THF 4-*HH*), 1.82–2.07 (5H, m, 4 × Val  $\beta$ -H and THF 3-*HH*), 2.36–2.45 (4H, m, succ 2-H<sub>2</sub> and 3-H<sub>2</sub>), 2.85–3.03 (2H, m, 2 × Trp  $\beta$ -*HH*), 3.04–3.21 (4H, m, Trp  $\beta$ -*HH* and 3 × Trp  $\beta$ -*HH*), 3.22–3.28



(2H, m, AE 2-H<sub>2</sub>), 3.60 (3H, s, OCH<sub>3</sub>), 3.55–3.63 (3H, m, THF 5-H and AE 1-H<sub>2</sub>), 3.63–3.74 (4H, m, 2 × Gly α-H<sub>2</sub>), 3.85–3.93 (1H, m, THF 1'-H), 4.03–4.33 (11H, m, 3 × Leu α-H, 5 × Val α-H, THF 2-H and 2 × Ala α-H), 4.37–4.60 (3H, m, 3 × Trp α-H), 6.87–6.95 (3H, m, ar. 3 × CH), 6.97–7.03 (3H, m, 3 × ar. CH), 7.05 (1H, d, *J* 1.9, ar. CH), 7.11 (1H, d, *J* 1.9, ar. CH), 7.14 (1H, d, *J* 1.5, ar. CH), 7.24–7.32 (3H, m, 3 × ar. CH), 7.35–7.45 (6H, m, 6 × ar. CH), 7.49–7.57 (3H, m, 3 × ar. CH), 7.58–7.64 (4H, m, 4 × ar. CH), 7.78 (1H, d, *J* 8.7, NH), 7.83–7.93 (6H, m, 6 × NH), 7.94–8.05 (4H, m, 4 × NH), 8.06–8.15 (2H, m, 2 × NH), 8.17–8.28 (2H, m, 2 × NH), 8.37 (1H, t, *J* 5.8, AE NH), 8.45 (1H, d, *J* 5.3, Trp NH), 10.71 (1H, s, indole NH), 10.76 (2H, s, indole NH); HPLC: 14.7 min (M4); *m/z* (ESI) 2226.0 (M + Na<sup>+</sup>. C<sub>116</sub>H<sub>166</sub>N<sub>20</sub>NaO<sub>21</sub>Si requires 2226.2).

**Succ-(<sup>11,12</sup>[L-THF]-gA-TBDPS)<sub>2</sub> 4.** 15.0 mg (6.8 μmol) of methyl ester **32** were C-deprotected according to GP 1 (*R*<sub>f</sub> = 0.20 in CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H 100 : 10 : 1; HPLC: 7.2 min M4). 12.0 mg of the crude carboxylic acid were combined with 12.0 mg (6.8 μmol) of amine **31a** and coupled according to GP 5 (1.0 mm, 12 h). After aqueous work-up 24.0 mg of crude diamide **4** were obtained and purified by preparative HPLC (250 mm Rainin C 4, 60%→100% B in 25 min, A: H<sub>2</sub>O + 0.1% TFA; B: acetonitrile–isopropanol 2 : 1 + 0.1% TFA; 14 ml min<sup>-1</sup>). Treatment of the collected fractions according to GP 6 gave 9.0 mg (2.3 μmol, 34%) of pure diamide **4** as a colourless solid. δ<sub>H</sub> (500 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.55–0.73 (30H, m, 10 × Leu δ-H<sub>3</sub>), 0.75–0.91 (54H, m, 16 × Val γ-H<sub>3</sub> and 2 × Leu δ-H<sub>3</sub>), 0.92–1.05 (26H, m, 6 × *t*-Bu-H<sub>3</sub>, 2 × THF 2'-H<sub>3</sub> and 2 × Leu γ-H), 1.05–1.39 (20 H, m, 4 × Ala β-H<sub>3</sub> and 4 × Leu β-H<sub>2</sub>), 1.40–1.59 (6H, m, 2 × Leu γ-H, 2 × Val β-H and 2 × THF 3- or 4-HH), 1.61–1.78 (4H, m, 2 × Leu β-H<sub>2</sub>), 1.85–2.08 (12H, m, 2 × THF 3- or 4-HH, 2 × THF 3- or 4-HH, 6 × Val β-H and 2 × Leu γ-H), 2.26–2.32 (2H, m, 2 × THF 3- or 4-HH), 2.36–2.45 (4H, m, succ 2-H<sub>2</sub> and 3-H<sub>2</sub>), 2.88–3.05 (6H, m, 6 × Trp β-HH), 3.08–3.29 (6H, m, 6 × Trp β-HH), 3.42 (4H, t, *J* 5.3, 2 × AE 2-H<sub>2</sub>), 3.56–3.63 (6H, m, 2 × AE 1-H<sub>2</sub> and 2 × Gly αCHH), 3.66–3.76 (2H, m, 2 × Leu α-H), 3.86–3.95 (4H, m, 2 × THF 1'-H and 2 × Val α-H), 4.05–4.15 (6H, m, 2 × Val α-H, 2 × Gly α-HH and 2 × Leu α-H), 4.18–4.34 (12H, m, 4 × Val α-H, 4 × Ala α-H, 2 × Leu α-H and 2 × THF 5-H), 4.39–4.56 (6H, m, 6 × Trp α-H), 4.56–4.62 (2H, m, 2 × THF 2-H), 6.89–6.97 (6H, m, 6 × ar. CH), 6.99–7.05 (6H, m, 6 × ar. CH), 7.07 (2H, s, 2 × ar. CH), 7.15 (4H, d, *J* 12.8, 4 × ar. CH), 7.30 (6H, t, *J* 7.7, 6 × ar. CH), 7.38–7.48 (12H, m, 12 × ar. CH), 7.52–7.66 (14H, m, 14 × ar. CH), 7.90 (16H, m, 16 × NH), 8.15–8.21 (6H, m, 6 × NH), 8.24–8.36 (6H, m, 6 × NH), 8.49–8.56 (2H, m, 2 × NH), 10.72 (2H, s, indole NH), 10.80 (4H, s, indole NH); HPLC: 18.6 min (M4); *m/z* (ESI) 1999.9 (M + 2Na<sup>+</sup>. C<sub>212</sub>H<sub>298</sub>N<sub>36</sub>Na<sub>2</sub>O<sub>34</sub>Si<sub>2</sub> requires 1997.1).

**Boc-D-THF-D-Leu-OMe 27.** 289 mg (1.15 mmol) of carboxylic acid **5** were combined with 616 mg (3.40 mmol) of D-Leucine hydrochloride and coupled according to GP 3 (0.04 M 2 h). After aqueous work-up FCC (35g, 1% MeOH in CHCl<sub>3</sub>) gave 346 mg (0.90 mmol, 78%) of pure methyl ester **27** as a colourless foam. *R*<sub>f</sub> = 0.57 (CHCl<sub>3</sub>–MeOH 20 : 1); δ<sub>H</sub> (300 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.84 (3H, d, *J* 6.4, Leu δ-H<sub>3</sub>), 0.91 (3H, d, *J* 6.4, Leu δ-H<sub>3</sub>), 0.99 (3H, d, *J* 5.5, THF 2'-H<sub>3</sub>), 1.29–1.39 (1H, m, Leu β-HH), 1.40 (9H, s, 3 × *t*-Bu-H<sub>3</sub>), 1.54 (1H, ddd, *J* 13.2, 9.9 and 3.6, THF 4-HH), 1.61–1.71 (1H, m, Leu γ-H), 1.83–1.92 (2H, m, Leu β-HH and THF 3-HH), 2.00–2.09 (1H, m, THF 3-HH), 2.11–2.18 (1H, m, THF 4-HH), 3.64 (3H, s, OCH<sub>3</sub>), 3.65–3.71 (2H, m, THF 1'-H and Leu α-H), 4.34 (1H, d, *J* 8.2, THF 5-H), 4.39 (1H, ddd, *J* 11.5, 8.4 and 3.5 THF 2-H), 7.06 (1H, d, *J* 8.0, THF NH), 8.68 (1H, d, *J* 8.6, Leu NH); [α]<sub>D</sub> = –9.3, [α]<sub>365</sub> = –42.1 (*c* = 1.0, CHCl<sub>3</sub>, 20 °C); HPLC: 7.9 min (M1); *m/z* (FAB) 387.2491 (M + H<sup>+</sup>. C<sub>19</sub>H<sub>35</sub>N<sub>2</sub>O<sub>6</sub> requires 387.2495).

**Z-Trp-D-THF-D-Leu-OMe 29.** 216 mg (0.56 mmol) of methyl ester **27** were dissolved in a solution of CF<sub>3</sub>CO<sub>2</sub>H in CH<sub>2</sub>Cl<sub>2</sub> (20 ml, 20%) and stirred 20 min. at room temperature. The reaction mixture was then diluted with CH<sub>2</sub>Cl<sub>2</sub> (30 ml), washed carefully with sat. NaHCO<sub>3</sub> (10 ml) and brine (20 ml) and dried over MgSO<sub>4</sub>. After evaporation, 160 mg of crude amine (*R*<sub>f</sub> = 0.28 in CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H 100 : 10 : 1) were obtained and directly coupled with 379 mg (1.12 mmol) Z-Trp-OH according to GP 3 (0.02 M, 2 h). After aqueous work-up and FCC (7 g, 3% MeOH in CHCl<sub>3</sub>) 322 mg (0.53 mmol, 95%) of pure peptide **29** were obtained as a colourless foam. *R*<sub>f</sub> = 0.50 (CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H 100 : 10 : 1); δ<sub>H</sub> (500 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.80 (3H, d, *J* 6.4, Leu δ-H<sub>3</sub>), 0.87 (3H, d, *J* 6.4, Leu δ-H<sub>3</sub>), 0.98 (3H, d, *J* 6.9, THF 2'-H<sub>3</sub>), 1.38–1.48 (1H, m, THF 4-HH), 1.52 (1H, ddd, *J* 13.3, 9.6 and 3.9, THF 3-HH), 1.57–1.67 (1H, m, Leu γ-H), 1.86–1.95 (2H, m, THF 3-HH and Leu β-HH), 2.04–2.13 (2H, m, THF 4-HH and Leu β-HH), 2.96 (1H, dd, *J* 13.9 and 8.8, Trp β-HH), 3.31 (1H, dd, *J* 14.4 and 4.8, Trp β-HH), 3.60 (3H, s, OCH<sub>3</sub>), 3.76–3.83 (1H, m, Leu α-H), 3.91–3.99 (1H, m, THF C-1'-H), 4.25–4.32 (1H, m, THF 5-H), 4.32–4.41 (2H, m, Trp α-H and THF 2-H), 4.97 (2H, s, Z-H<sub>2</sub>), 6.97 (1H, t, *J* 7.5, ar. CH), 7.01–7.09 (2H, m, 2 × ar. CH), 7.10–7.21 (1H, m, ar. CH), 7.22–7.27 (2H, m, 2 × ar. CH), 7.27–7.37 (4H, m, 4 × ar. CH), 7.61 (1H, d, *J* 8.0, THF NH), 8.16 (1H, d, *J* 8.5, Trp NH), 8.68 (1H, d, *J* 8.0, Leu NH), 10.84 (1H, s, indole NH); [α]<sub>D</sub> = –35.8, [α]<sub>365</sub> = –133.2 (*c* = 1.0, CHCl<sub>3</sub>, 20 °C); HPLC: 6.6 min (M1). *m/z* (FAB) 607.3139 (M + H<sup>+</sup>. C<sub>33</sub>H<sub>43</sub>N<sub>4</sub>O<sub>7</sub> requires 607.3132).

**Z-Trp-D-THF-D-Leu-Trp-D-Leu-Trp-2-(*t*-butyldiphenylsilyloxy)-ethylamide 30.** 150 mg (0.25 mmol) of peptide **29** were C-deprotected according to GP 1 (*R*<sub>f</sub> = 0.28 in CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H), as well as 1.20 g (1.30 mmol) of tripeptide **21** were N-deprotected according to GP 2A (MeOH, rt, 2 h; *R*<sub>f</sub> = 0.38 in CHCl<sub>3</sub>–MeOH–NH<sub>3</sub>(aq.) 100 : 10 : 1). The crude carboxylic acid and a portion (1/4) of the crude amine were collected and coupled according to GP 4 (8.0 mm, 3 h). After aqueous work-up and FCC (40 g, 3% MeOH in CHCl<sub>3</sub>) 319 mg (23.5 mmol, 94%) of peptide **30** were isolated as a colourless foam. *R*<sub>f</sub> = 0.65 (CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H 200 : 10 : 1); δ<sub>H</sub> (400 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.59 (3H, d, *J* 6.6, Leu δ-H<sub>3</sub>), 0.65 (3H, d, *J* 6.3, Leu δ-H<sub>3</sub>), 0.66 (3H, d, *J* 6.6, Leu δ-H<sub>3</sub>), 0.70 (3H, d, *J* 6.3, Leu δ-H<sub>3</sub>), 0.84 (3H, d, *J* 6.6, THF 2'-H<sub>3</sub>), 0.97 (9H, s, 3 × *t*-Bu-H<sub>3</sub>), 0.96–1.03 (1H, m, Leu γ-H), 1.11–1.21 (3H, m, THF 4-HH and Leu β-H<sub>2</sub>), 1.22–1.36 (2H, m, THF 3HH and Leu γ-H), 1.48–1.58 (1H, m, THF 3-HH), 1.68–1.78 (1H, m, THF 4-HH), 1.88–2.05 (2H, m, Leu β-H<sub>2</sub>), 2.83–2.99 (3H, m, 3 × Trp β-HH), 3.05–3.20 (3H, m, 3 × Trp β-HH), 3.20–3.29 (2H, m, AE 2-H<sub>2</sub>), 3.62 (2H, t, *J* 6.6, AE 1-H<sub>2</sub>), 3.65 (1H, dd, *J* 14.9 and 8.9, THF 5-H), 3.81 (1H, sextet, *J* 7.0, THF 1'-H), 4.15–4.31 (4H, m, 2 × Leu α-H, Trp α-H and THF 2-H), 4.47 (1H, dt, *J* 4.4 and 9.1, Trp α-H), 4.53 (1H, dt, *J* 5.6 and 8.5, Trp α-H), 4.92 (1H, d, *J* 13.5, Z-HH), 4.96 (1H, d, *J* 13.2, Z-HH), 6.89–6.96 (3H, m, 3 × ar. CH), 6.98–7.09 (6H, m, 5 × ar. CH and Trp NH), 7.11–7.14 (1H, m, ar. CH), 7.19–7.24 (2H, m, 2 × ar. CH), 7.25–7.34 (6H, m, 6 × ar. CH), 7.37–7.47 (6H, m, 6 × ar. CH), 7.51–7.60 (3H, m, 3 × ar. CH), 7.59–7.65 (4H, m, 4 × ar. CH), 7.83 (1H, d, *J* 7.6, Leu NH), 8.02, (1H, d, *J* 8.6, THF NH), 8.04 (1H, t, *J* 5.5, AE NH), 8.13 (1H, d, *J* 7.9, Trp NH), 8.20 (1H, d, *J* 8.3, Trp NH), 8.24 (1H, d, *J* 7.6, Leu NH), 10.73 (1H, d, *J* 1.9, indole NH), 10.76–10.83 (2H, m, indole NH); [α]<sub>D</sub> = –19.8, [α]<sub>365</sub> = –70.8 (*c* = 1.1, CHCl<sub>3</sub>, 20 °C); HPLC: 13.2 (M1); *m/z* (ESI) 1381.9 (M + Na<sup>+</sup>. C<sub>78</sub>H<sub>94</sub>N<sub>10</sub>NaO<sub>10</sub>Si requires 1381.7).

**Z-Ala-D-Val-Val-D-Leu-Trp-D-THF-D-Leu-Trp-2-(*t*-butyldiphenylsilyloxy)-ethylamide 16.** 319 mg (0.23 mmol) of peptide **30** were N-deprotected according to GP 2A (EtOH, 50 °C, 2h; *R*<sub>f</sub> = 0.18 in CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H 200 : 10 : 1) and 369 mg (0.70 mmol) of tetrapeptide **24** were C-deprotected according to GP 1. The crude amine and a portion (1/4) of the crude saponification product were then collected and coupled

according to GP 4 (3.0 mm, 3 h). After aqueous work-up, FCC (20 g, 3% MeOH in CHCl<sub>3</sub>→5% MeOH in CHCl<sub>3</sub> + 0.5% HCO<sub>2</sub>H) gave 231 mg (0.13 mmol, 57%) of pure ethylamide **16** as a colourless foam.  $R_f$  = 0.28 (CHCl<sub>3</sub>–MeOH 20 : 1);  $\delta_H$  (400 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.38 (3H, d,  $J$  6.6, Leu  $\delta$ -H<sub>3</sub>), 0.55 (3H, d,  $J$  6.6, Leu  $\delta$ -H<sub>3</sub>), 0.57 (3H, d,  $J$  6.6, Leu  $\delta$ -H<sub>3</sub>), 0.64 (6H, d,  $J$  5.9, Leu  $\delta$ -H<sub>3</sub> and Val  $\gamma$ -H<sub>3</sub>), 0.72 (3H, d,  $J$  6.3, Val  $\gamma$ -H<sub>3</sub>), 0.74–0.83 (12H, m, 4 × Val  $\gamma$ -H<sub>3</sub>), 0.86 (3H, d,  $J$  6.9, THF 2'-H<sub>3</sub>), 0.98 (9H, s, 3 × *t*-Bu-H<sub>3</sub>), 0.95–1.05 (1H, m, Leu  $\gamma$ -H), 1.09–1.21 (3H, m, THF 4-*HH* and Leu  $\beta$ -H<sub>2</sub>), 1.19 (3H, d,  $J$  7.3, Ala  $\beta$ -H<sub>3</sub>), 1.22–1.35 (2H, m, THF 4-*HH*, and Val  $\beta$ -H), 1.45–1.55 (1H, m, THF 3-*HH*), 1.63 (1H, sextet,  $J$  7.2, Leu  $\gamma$ -H), 1.69–1.79 (1H, m, THF 4-*HH*), 1.90–2.11 (4H, m, 2 × Val  $\beta$ -H and Leu  $\beta$ -H<sub>2</sub>), 2.79 (1H, dd,  $J$  14.2 and 10.1, Trp  $\beta$ -*HH*), 2.84–2.95 (2H, m, 2 × Trp  $\beta$ -*HH*), 3.13–3.24 (2H, m, 2 × Trp  $\beta$ -*HH*), 3.25–3.31 (3H, m, AE 2-H<sub>2</sub> and Trp  $\beta$ -*HH*), 3.64 (2H, t,  $J$  6.4, AE 1-H<sub>2</sub>), 3.64–3.74 (1H, m, THF 5-H), 3.82 (1H, t,  $J$  7.1, Leu  $\alpha$ -H), 3.92 (1H, sextet,  $J$  8.3, THF 1'-H), 4.13 (1H, dt,  $J$  14.5 and 7.3, Ala  $\alpha$ -H), 4.17–4.27 (3H, m, Leu  $\alpha$ -H, Val  $\alpha$ -H and THF 2-H), 4.28–4.36 (2H, m, 2 × Val  $\alpha$ -H), 4.38–4.51 (2H, m, 2 × Trp  $\alpha$ -H), 4.52–4.61 (1H, m, Trp  $\alpha$ -H), 4.98 (1H, d,  $J$  12.9, Z-*HH*), 5.02 (1H, d,  $J$  12.9, Z-*HH*), 7.26–7.35 (11H, m, Ala NH and 10 × ar. CH), 7.36–7.46 (10H, m, 10 × ar. CH), 7.47–7.58 (5H, m, 5 × ar. CH), 7.59–7.64 (6H, m, THF NH and 5 × ar. CH), 7.66 (1H, d,  $J$  8.9, Val NH), 7.68 (1H, d,  $J$  7.3, NH), 7.82 (1H, d,  $J$  6.9, Leu NH), 7.92 (1H, d,  $J$  8.9, Val NH), 8.05 (1H, t,  $J$  5.8, AE NH), 8.13 (1H, d,  $J$  6.9, NH), 8.24 (1H, d,  $J$  8.3, Trp NH), 8.26–8.30 (2H, m, Trp NH), 10.71–10.76 (2H, m, indole NH), 10.80 (1H, d,  $J$  1.3, indole NH); HPLC: 10.9 min (250 × 4 mm Jupiter C 5, 70→100% B in 25 min, A: H<sub>2</sub>O; B: acetonitrile–isopropanol 2 : 1; 0.7 ml min<sup>-1</sup>);  $m/z$  (ESI) 1749.9 (M + Na<sup>+</sup>. C<sub>96</sub>H<sub>126</sub>N<sub>14</sub>O<sub>14</sub>Si requires 1749.9).

**H-Ala-D-Val-Val-D-Val-Trp-D-THF-D-Leu-Trp-D-Leu-Trp-2-(*t*-butyldiphenylsilyloxy)-ethylamide 16a.** 210 mg (120  $\mu$ mol) of peptide **16** were *N*-deprotected according to GP 2A (EtOH, 50 °C). After FCC (8 g; CHCl<sub>3</sub>–MeOH 15 : 1→CHCl<sub>3</sub>–MeOH–NH<sub>3</sub>(conc.) 100 : 10 : 1) 131 mg (0.08 mmol, 69%) of the pure amine **16a** were obtained.  $R_f$  = 0.09 in CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H 100 : 10 : 1; HPLC: 7.4 min (M4).  $m/z$  (ESI) 1616.1 (M + Na<sup>+</sup>. C<sub>88</sub>H<sub>120</sub>N<sub>14</sub>NaO<sub>12</sub>Si requires 1615.9).

**CHO-Val-Gly-Ala-D-Leu-Ala-D-Val-Val-D-Val-Trp-D-THF-D-Leu-Trp-D-Leu-Trp-2-(*t*-butyldiphenylsilyloxy)-ethylamide 6.** 217 mg (0.54 mmol) of tetrapeptide **14** were *C*-deprotected according to GP 1 ( $R_f$  = 0.21 in CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H 100 : 10 : 1) and a portion (1/4) of the crude saponification product was coupled with amine **16a** (121 mg, 76.0  $\mu$ mol) according to GP 4 (4.0 mm, 3 h). After aqueous work-up, FCC (14 g, CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H 150 : 10 : 1) gave 130 mg (66.3  $\mu$ mol, 87%) of the pure peptide **6** as a colourless solid. Prior to single-channel measurements, purification of 10.0 mg of compound **6** by preparative HPLC (250 mm Rainin C 8, 75→90% B in 25 min, A: H<sub>2</sub>O; B: acetonitrile–isopropanol 2 : 1; 10 ml min<sup>-1</sup>) afforded 9.0 mg (90%) of pure product.  $R_f$  = 0.36 (CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H 100 : 10 : 1);  $\delta_H$  (500 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.21 (3H, d,  $J$  6.4, Leu  $\delta$ -H<sub>3</sub>), 0.35 (3H, d,  $J$  6.7, Leu  $\delta$ -H<sub>3</sub>), 0.38 (3H, d,  $J$  6.4, Leu  $\delta$ -H<sub>3</sub>), 0.45 (6H, d,  $J$  5.7, 2 × Leu  $\delta$ -H<sub>3</sub>), 0.50–0.73 (30H, m, Leu  $\delta$ -H<sub>3</sub>, THF 2'-H<sub>3</sub> and 8 × Val  $\gamma$ -H<sub>3</sub>), 0.74–0.87 (11H, m, 3 × *t*-Bu-H<sub>3</sub> and 2 × Leu  $\gamma$ -H), 0.88–1.17 (11H, m, 2 × Ala  $\beta$ -H<sub>3</sub>, 2 × Leu  $\beta$ -H<sub>2</sub> and Leu  $\gamma$ -H), 1.19–1.49 (4H, m, THF 4-*HH*, Val  $\beta$ -H and Leu  $\beta$ -H<sub>2</sub>), 1.50–1.63 (1H, m, THF 3-*HH*), 1.70–1.91 (5H, m, THF 3-*HH*, THF 4-*HH* and 3 × Val  $\beta$ -H), 2.53–2.78 (3H, m, 3 × Trp  $\beta$ -*HH*), 2.91–3.13 (5H, m, 3 × Trp  $\beta$ -*HH* and AE 2-H<sub>2</sub>), 3.45 (2H, t,  $J$  6.2, AE 1-H<sub>2</sub>), 3.49–3.60 (3H, m, Gly  $\alpha$ -H<sub>2</sub> and Leu  $\alpha$ -H), 3.61–3.80 (2H, m, Leu  $\alpha$ -H and THF 1'-H), 3.95–4.17 (9H, m, 4 × Val  $\alpha$ -H, THF 2-H and 5-H, Leu  $\alpha$ -H and 2 × Ala  $\alpha$ -H), 4.18–4.33 (2H, m, 2 × Trp  $\alpha$ -H), 4.34–4.43 (1H, m, Trp  $\alpha$ -H), 6.67–6.78 (3H, m, 3 × ar. CH), 6.79–7.00 (6H, m, 6 × ar. CH), 7.01–7.14 (3H, m, 3 × ar. CH),

7.15–7.28 (6H, m, 6 × ar. CH), 7.29–7.48 (9H, m, 7 × ar. CH, THF NH and NH), 7.51 (1H, d,  $J$  5.8 Leu NH), 7.60–7.98 (8H, m, 8 × NH), 8.00–8.19 (5H, m, 3 × Trp NH, Gly NH and AE NH), 10.55 (2H, s, indole NH), 10.62 (1H, s, indole NH); HPLC: 7.6 min (250 × 4 mm Jupiter C 5, 65→75% B in 25 min, A: H<sub>2</sub>O; B: acetonitrile–isopropanol 2 : 1; 0.7 ml min<sup>-1</sup>);  $m/z$  (ESI): 1984.0 (M + Na<sup>+</sup>. C<sub>105</sub>H<sub>148</sub>N<sub>18</sub>NaO<sub>17</sub>Si requires 1984.1).

<sup>10,11</sup>[D-THF]-gA **5**. 30.0 mg (15.3  $\mu$ mol) of peptide **6** were dissolved in 27 ml CH<sub>2</sub>Cl<sub>2</sub>. A solution of 5% HF in CH<sub>3</sub>CN (2.7 ml) was then added dropwise and the mixture was stirred for 45 min at room temperature. After dilution with 30 ml CH<sub>2</sub>Cl<sub>2</sub> the mixture was carefully washed with sat. NaHCO<sub>3</sub> (2 × 10 ml) and brine, and dried over MgSO<sub>4</sub>. Evaporation gave 33.0 mg of the crude peptide **5**, which was purified by preparative HPLC (250 mm Rainin C 8, 60→75% B in 25 min, A: H<sub>2</sub>O; B: acetonitrile–isopropanol 2 : 1; 10 ml min<sup>-1</sup>). 22.0 mg (12.7  $\mu$ mol, 83%) of pure product were obtained as a colourless solid.  $R_f$  = 0.16 (CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H 100 : 10 : 1);  $\delta_H$  (500 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.33 (3H, d,  $J$  6.8, Leu  $\delta$ -H<sub>3</sub>), 0.47 (3H, d,  $J$  6.4, Leu  $\delta$ -H<sub>3</sub>), 0.51 (3H, d,  $J$  6.8, Leu  $\delta$ -H<sub>3</sub>), 0.59 (6H, d,  $J$  6.8, 2 × Leu  $\delta$ -H<sub>3</sub>), 0.63–0.87 (30H, m, 8 × Val  $\gamma$ -H<sub>3</sub>, THF 2'-H<sub>3</sub> and Leu  $\delta$ -H<sub>3</sub>), 0.88–0.97 (2H, m, 2 × Leu  $\gamma$ -H), 1.04–1.29 (13H, m, 2 × Ala  $\beta$ -H<sub>3</sub>, 3 × Leu  $\beta$ -H<sub>2</sub>, and Leu  $\gamma$ -H), 1.35–1.62 (5H, m, Leu  $\beta$ -H<sub>2</sub>, Leu  $\gamma$ -H, Val  $\beta$ -H and THF 4-*HH*), 1.65–1.77 (1H, m, THF 3-*HH*), 1.83–2.03 (5H, m, THF 4-*HH*, THF 3-*HH* and 3 × Val  $\beta$ -H), 2.64–2.90 (3H, m, 3 × Trp  $\beta$ -*HH*), 3.05–3.25 (5H, m, 3 × Trp  $\beta$ -*HH* and AE 1-H<sub>2</sub>), 3.33 (1H, d,  $J$  6.0, AE 2-*HH*), 3.37 (1H, d,  $J$  6.4, AE 2-*HH*), 3.60–3.73 (3H, m, Gly  $\alpha$ -H<sub>2</sub> and Leu  $\alpha$ -H), 3.74–3.94 (2H, m, Leu  $\alpha$ -H and THF 1'-H), 4.05–4.30 (9H, m, 4 × Val  $\alpha$ -H, 2 × Ala  $\alpha$ -H, Leu  $\alpha$ -H, THF 2-H and 5-H), 4.31–4.44 (2H, m, 2 × Trp  $\alpha$ -H), 4.46–4.57 (1H, m, Trp  $\alpha$ -H), 4.63 (1H, t,  $J$  5.7, OH), 6.82–6.92 (3H, m, 3 × ar. CH), 6.93–7.00 (3H, m, 3 × ar. CH), 7.01–7.08 (3H, m, 3 × ar. CH), 7.23 (3H, d,  $J$  7.9, 3 × ar. CH), 7.47 (3H, dd,  $J$  19.2 and 7.9, 3 × ar. CH), 7.53–7.62 (2H, m, 1H, NH and Leu NH), 7.68 (1H, d,  $J$  7.9, Leu NH), 7.75–7.91 (4H, m, THF NH, AE NH and 2 × NH), 7.94 (1H, d,  $J$  6.8, NH), 8.06 (2H, d,  $J$  7.9, NH), 8.13–8.31 (5H, m, 3 × Trp NH, Gly NH and NH), 10.68 (1H, d,  $J$  0.8, indole NH), 10.70 (1H, d,  $J$  1.9, indole NH), 10.75 (1H, d,  $J$  1.9, indole NH); HPLC: 6.8 min (250 × 4 mm Jupiter C 5, 65→75% B in 25 min, A: H<sub>2</sub>O; B: acetonitrile–isopropanol 2 : 1; 0.7 ml min<sup>-1</sup>);  $m/z$  (ESI) 1745.9 (M + Na<sup>+</sup>. C<sub>89</sub>H<sub>130</sub>N<sub>18</sub>NaO<sub>17</sub> requires 1746.0).

**Z-Ala-D-Leu-Ala-D-Val-Val-D-Val-Trp-D-THF-D-Leu-Trp-D-Leu-Trp-2-(*t*-butyldiphenylsilyloxy)-ethylamide 33.** 210 mg (0.60 mmol) of dipeptide **18** were *C*-deprotected according to GP 1. A portion (1/2) of the crude saponification product was coupled with amine **16a** (220 mg 0.14 mmol) according to GP 4 (3.0 mm, 3 h) and after aqueous work-up FCC (20g, 4% MeOH in CHCl<sub>3</sub> + 0.4% HCO<sub>2</sub>H) gave 215 mg (0.11 mmol, 79%) of pure peptide **33** as a colourless solid.  $R_f$  = 0.49 (CHCl<sub>3</sub>–MeOH–NH<sub>3</sub>(conc.) 100 : 10 : 1);  $\delta_H$  (500 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.40 (3H, d,  $J$  6.6, Leu  $\delta$ -H<sub>3</sub>), 0.54 (3H, d,  $J$  6.4, Leu  $\delta$ -H<sub>3</sub>), 0.58 (3H, d,  $J$  6.4, Leu  $\delta$ -H<sub>3</sub>), 0.65 (6H, d,  $J$  5.5, 2 × Leu  $\delta$ -H<sub>3</sub>), 0.70–0.82 (18H, m, Leu  $\delta$ -H<sub>3</sub> and 5 × Val  $\gamma$ -H<sub>3</sub>), 0.84 (3H, d,  $J$  6.9, Val  $\gamma$ -H<sub>3</sub>), 0.89 (3H, d,  $J$  6.4, THF 2'-H<sub>3</sub>), 0.98 (9H, s, 3 × *t*-Bu-H<sub>3</sub>), 0.95–1.05 (1H, m, Leu  $\gamma$ -H), 1.19 (6H, d,  $J$  7.1, 2 × Ala  $\beta$ -H<sub>3</sub>), 1.09–1.22 (3H, m, THF 3-*HH* and Leu  $\beta$ -H<sub>2</sub>), 1.23–1.36 (2H, m, THF 4-*HH* and Leu  $\gamma$ -H), 1.42–1.58 (4H, m, Val  $\beta$ -H, THF 3-*HH* and Leu  $\beta$ -H<sub>2</sub>), 1.59–1.68 (1H, m, Leu  $\gamma$ -H), 1.71–1.80 (1H, m, THF 4-*HH*), 1.92–2.06 (4H, m, 2 × Val  $\beta$ -H and Leu  $\beta$ -H<sub>2</sub>), 2.80 (1H, dd,  $J$  13.9 and 10.8, Trp  $\beta$ -*HH*), 2.85–2.95 (2H, m, 2 × Trp  $\beta$ -*HH*), 3.14–3.24 (3H, m, 3 × Trp  $\beta$ -*HH*), 3.25–3.20 (2H, m, AE 2-H<sub>2</sub>), 3.65 (2H, t,  $J$  6.5, AE 1-H<sub>2</sub>), 3.73 (1H, dd,  $J$  14.8 and 7.5 THF 5-H), 3.84 (1H, t,  $J$  7.2, Leu  $\alpha$ -H), 3.93 (1H, dd,  $J$  14.4 and 6.9, THF 1'-H), 4.02–4.09 (1H, m, Ala  $\alpha$ -H), 4.17–4.33 (7H, m, 2 × Leu  $\alpha$ -H, Ala  $\alpha$ -H, THF 2-H and 3H and Val  $\alpha$ -H), 4.41–4.51 (2H, m, 2 × Trp  $\alpha$ -H), 4.54–4.60



(1H, m, Trp  $\alpha$ -H), 4.99 (1H, d,  $J$  12.6, Z-*HH*), 5.02 (1H, d,  $J$  12.1, Z-*HH*), 6.90–6.97 (3H, m, 3  $\times$  ar. CH), 6.99–7.05 (3H, m, 3  $\times$  ar. CH), 7.08 (2H, s, 2  $\times$  ar. CH), 7.13 (1H, s, ar. CH), 7.27–7.37 (8H, m, 8  $\times$  ar. CH), 7.38–7.47 (7H, m, Ala NH and 6  $\times$  ar. CH), 7.51 (1H, d,  $J$  7.6, ar. CH), 7.56 (2H, t,  $J$  7.5, 2  $\times$  ar. CH), 7.56–7.60 (1H, m, NH), 7.61–7.67 (5H, m, 4  $\times$  ar. CH and THF NH), 7.71 (1H, d,  $J$  7.3, NH), 7.85 (1H, d,  $J$  6.9, Leu NH), 7.91 (1H, d,  $J$  9.2, NH), 7.98 (1H, d,  $J$  7.3, NH), 8.08 (1H, t,  $J$  5.8, AE NH), 8.11–8.18 (2H, m, NH), 8.25–8.36 (3H, m, Trp NH), 10.73–10.78 (2H, m, indole NH), 10.82 (1H, s, indole NH); HPLC: 11.8 min (M4);  $m/z$  (ESI): 1933.8 (M + Na<sup>+</sup>. C<sub>105</sub>H<sub>142</sub>N<sub>16</sub>NaO<sub>16</sub>Si requires 1934.0).

**H-Ala-D-Leu-Ala-D-Val-Val-D-Val-Trp-D-THF-D-Leu-Trp-D-Leu-Trp-2-(*t*-butyldiphenylsilyloxy)-ethylamide 33a.** 200 mg (104  $\mu$ mol) of peptide **33** were *N*-deprotected according to GP 2A (EtOH, 50 °C). After filtration and evaporation FCC (25 g, CHCl<sub>3</sub>–MeOH–NH<sub>3</sub>(conc.) 100 : 10 : 1) gave 125 mg (70.0  $\mu$ mol, 67%) of amine **33a**.  $R_f$  = 0.15 (CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H 100 : 10 : 1). HPLC: 9.9 min (250  $\times$  4 mm Dynamax C8, 60 $\rightarrow$ 100% B in 25 min, A: H<sub>2</sub>O + 0.1% TFA; B: acetonitrile–isopropanol 2 : 1 + 0.1% TFA; 0.7 ml min<sup>–1</sup>).  $m/z$  (ESI): 1799.7 (M + Na<sup>+</sup>. C<sub>97</sub>H<sub>136</sub>N<sub>16</sub>NaO<sub>14</sub>Si requires 1800.0).

**Succ-1-(1'-*t*-butyldiphenylsilyloxy)-<sup>10,11</sup>[D-THF]-gramicidin A)-4-(Val-Gly-OMe)-diamide 34.** 23.5 mg (44.0  $\mu$ mol) of benzyl ester **17** were *C*-deprotected according to GP 2A. The crude carboxylic acid and 40.0 mg (22.5  $\mu$ mol) of amine **33a** were then collected and coupled according to GP 4 (1.0 mM, 3 h). After aqueous work-up FCC (4 g, 4% MeOH in CHCl<sub>3</sub>–6% MeOH in CHCl<sub>3</sub> + 0.6% HCO<sub>2</sub>H) gave 35.0 mg (16.0 mmol, 73%) of pure diamide **34** as a colourless solid.  $R_f$  = 0.35 (CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H 100 : 10 : 1);  $\delta_H$  (500 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.40 (3H, d,  $J$  6.6, Leu  $\delta$ -H<sub>3</sub>), 0.55 (3H, d,  $J$  6.4, Leu  $\delta$ -H<sub>3</sub>), 0.58 (3H, d,  $J$  6.4, Leu  $\delta$ -H<sub>3</sub>), 0.65 (6H, d,  $J$  6.2, 2  $\times$  Leu  $\delta$ -H<sub>3</sub>), 0.72 (3H, d,  $J$  6.6, Leu  $\delta$ -H<sub>3</sub>), 0.74–0.82 (15H, m, 5  $\times$  Val  $\gamma$ -H<sub>3</sub>), 0.83–0.93 (18H, m, 5  $\times$  Val  $\gamma$ -H<sub>3</sub> and THF 2'-H<sub>3</sub>), 0.98 (9H, s, 3  $\times$  *t*-Bu-H<sub>3</sub>), 0.99–1.04 (1H, m, Leu  $\gamma$ -H), 1.08–1.34 (13H, m, 2  $\times$  Ala  $\beta$ -H<sub>3</sub>, THF 3-*HH* and 4-*HH*, 2  $\times$  Leu  $\beta$ -H<sub>2</sub> and Leu  $\gamma$ -H), 1.41–1.60 (4H, m, Leu  $\beta$ -H<sub>2</sub>, THF 3-*HH* and Val  $\beta$ -H), 1.61–1.69 (1H, m, Leu  $\gamma$ -H), 1.71–1.81 (1H, m, THF 4-*HH*), 1.91–2.07 (4H, m, 4  $\times$  Val  $\beta$ -H), 2.36–2.47 (4H, m, succ 3-H<sub>2</sub> and 4-H<sub>2</sub>), 2.75–2.83 (1H, m, Trp  $\beta$ -*HH*), 2.85–2.96 (2H, m, 2  $\times$  Trp  $\beta$ -*HH*), 3.13–3.30 (5H, m, 3  $\times$  Trp  $\beta$ -*HH* and AE 2-H<sub>2</sub>), 3.62 (3H, s, OCH<sub>3</sub>), 3.65 (2H, t,  $J$  6.5, AE 1-H<sub>2</sub>), 3.67–3.89 (6H, m, 2  $\times$  Gly  $\alpha$ -H<sub>2</sub>, THF 5-H and Leu  $\alpha$ CH), 3.90–3.96 (1H, m, THF 1'-H), 4.06–4.34 (10H, m, 5  $\times$  Val  $\alpha$ -H, 2  $\times$  Leu  $\alpha$ -H, 2  $\times$  Ala  $\alpha$ -H, and THF 2-H), 4.40–4.50 (2H, m, 2  $\times$  Trp  $\alpha$ -H), 4.54–4.61 (1H, m, Trp  $\alpha$ -H), 6.90–6.97 (3H, m, 3  $\times$  ar. CH), 6.99–7.05 (3H, m, 3  $\times$  ar. CH), 7.08 (2H, s, 2  $\times$  ar. CH), 7.13 (1H, d,  $J$  1.4, ar. CH), 7.28–7.32 (3H, m, 3  $\times$  ar. CH), 7.38–7.47 (6H, m, 6  $\times$  ar. CH), 7.51 (1H, d,  $J$  7.6, ar. CH), 7.56 (2H, t,  $J$  7.1, ar. CH), 7.59–7.64 (5H, m, 4  $\times$  CH and NH), 7.65 (1H, d,  $J$  8.5, THF NH), 7.71 (1H, d,  $J$  6.7, NH), 7.83–7.93 (4H, m, Leu NH and 3  $\times$  NH), 7.96 (1H, d,  $J$  8.9, NH), 8.05–8.11 (2H, m, 2  $\times$  NH), 8.12–8.17 (1H, m, NH), 8.20–8.37 (4H, m, 4  $\times$  NH), 8.39 (1H, t,  $J$  5.84, Gly NH), 10.75 (1H, s, indole NH), 10.77 (1H, s, indole NH), 10.82 (1H, s, indole NH). HPLC: 9.8 min (M4);  $m/z$  (ESI): 2226.4 (M + Na<sup>+</sup>. C<sub>116</sub>H<sub>166</sub>N<sub>20</sub>NaO<sub>21</sub>Si requires 2226.2).

**Succ-(<sup>10,11</sup>[D-THF]-gA-TBDPS)<sub>2</sub> 7.** 39.0 mg (17.7  $\mu$ mol) of methyl ester **34** were *C*-deprotected according to GP 1 ( $R_f$  = 0.20 in CHCl<sub>3</sub>–MeOH–HCO<sub>2</sub>H 100 : 10 : 1; HPLC: 9.8 min, 250  $\times$  4 mm Jupiter C 5, 70 $\rightarrow$ 100% B in 25 min, A: H<sub>2</sub>O + 0.1% TFA; B: acetonitrile–isopropanol 2 : 1 + 0.1% TFA; 0.7 ml min<sup>–1</sup>). The crude carboxylic acid and 40.0 mg (22.5  $\mu$ mol) of amine **33a** were collected and coupled according to GP 5 (1.0 mM, 12 h). After aqueous work-up 64.0 mg of crude diamide **7** were isolated and purified directly by preparative HPLC (250 mm Rainin C 4, 60 $\rightarrow$ 100% B in 25 min, A: H<sub>2</sub>O + 0.1% TFA;

B: acetonitrile–isopropanol 2 : 1 + 0.1% TFA; 14 ml min<sup>–1</sup>). Treatment of the collected fractions according to GP 6 gave 36.0 mg (9.1  $\mu$ mol, 61%) of pure diamide **7** as a colourless solid.  $\delta_H$  (500 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.40 (6H, d,  $J$  6.6, 2  $\times$  Leu  $\delta$ -H<sub>3</sub>), 0.54 (6H, d,  $J$  6.4, 2  $\times$  Leu  $\delta$ -H<sub>3</sub>), 0.58 (6H, d,  $J$  6.4, 2  $\times$  Leu  $\delta$ -H<sub>3</sub>), 0.65 (12H, d,  $J$  6.0, 4  $\times$  Leu  $\delta$ -H<sub>3</sub>), 0.72 (6H, d,  $J$  6.19, 2  $\times$  Leu  $\delta$ -H<sub>3</sub>), 0.74–0.82 (30H, m, 10  $\times$  Val  $\gamma$ -H<sub>3</sub>), 0.83–0.87 (18H, m, 6  $\times$  Val  $\gamma$ -H<sub>3</sub>), 0.89 (6H, d,  $J$  6.6, 2  $\times$  THF 2'-H<sub>3</sub>), 0.98 (18H, s, 6  $\times$  *t*-Bu-H<sub>3</sub>), 0.99–1.05 (2H, m, 2  $\times$  Leu  $\gamma$ -H), 1.09–1.38 (26H, m, 4  $\times$  Ala  $\beta$ -H<sub>3</sub>, 4  $\times$  Leu  $\beta$ -H<sub>2</sub>, 2  $\times$  THF 4-*HH*, 2  $\times$  THF 3-*HH* and 2  $\times$  Leu  $\gamma$ -H), 1.39–1.59 (8H, m, 2  $\times$  Leu  $\beta$ -H<sub>2</sub>, 2  $\times$  THF 3-*HH* and 2  $\times$  Val  $\beta$ -H), 1.61–1.68 (2H, m, 2  $\times$  Leu  $\gamma$ -H), 1.72–1.80 (2H, m, 2  $\times$  THF 4-*HH*), 1.90–2.06 (6H, m, 6  $\times$  Val  $\beta$ -H), 2.36–2.47 (4H, m, succ 2-H<sub>2</sub> and 3-H<sub>2</sub>), 2.76–2.84 (2H, m, 2  $\times$  Trp  $\beta$ -*HH*), 2.85–2.95 (4H, m, 4  $\times$  Trp  $\beta$ -*HH*), 3.14–3.31 (6H, m, 6  $\times$  Trp  $\beta$ -*HH*), 3.52 (2H, s, 2  $\times$  Gly  $\alpha$ -*HH*), 3.61–3.68 (6H, m, 2  $\times$  AE 2-H<sub>2</sub> and 2  $\times$  Gly  $\alpha$ -*HH*), 3.69–3.78 (4H, m, 2  $\times$  THF 5-H and 2  $\times$  Leu  $\alpha$ -H), 3.81–3.88 (2H, m, 2  $\times$  Leu  $\alpha$ -H), 3.89–3.97 (2H, m, 2  $\times$  THF 1'-H), 4.07–4.13 (2H, m, 2  $\times$  Val  $\alpha$ -H), 4.15–4.33 (14H, m, 6  $\times$  Val  $\alpha$ -H, 2  $\times$  THF 2-H, 4  $\times$  Ala  $\alpha$ -H and 2  $\times$  Leu  $\alpha$ -H), 4.39–4.51 (4H, m, 4  $\times$  Trp  $\alpha$ -H), 4.54–4.61 (2H, m, 2  $\times$  Trp  $\alpha$ -H), 6.90–6.97 (6H, m, 6  $\times$  ar. CH), 6.99–7.06 (6H, m, 6  $\times$  ar. CH), 7.08 (4H, s, 4  $\times$  ar. CH), 7.13 (2H, s, 2  $\times$  ar. CH), 7.30 (6H, d,  $J$  8.0, 6  $\times$  ar. CH), 7.37–7.47 (12H, m, 12  $\times$  ar. CH), 7.51 (2H, d,  $J$  7.8, 2  $\times$  ar. CH), 7.56 (4H, t,  $J$  6.5, 4  $\times$  ar. CH), 7.59–7.67 (12H, m, 8  $\times$  ar. CH, NH and THF NH), 7.68–7.73 (2H, m, 2  $\times$  NH), 7.82–7.94 (8H, m, 6  $\times$  NH and 2  $\times$  Leu NH), 8.00–8.11 (6H, m, 4  $\times$  NH and 2  $\times$  AE NH), 8.12–8.18 (2H, m, 2  $\times$  NH), 8.21–8.39 (8H, m, 6  $\times$  Trp NH and 2  $\times$  Gly NH), 10.73–10.79 (4H, m, 4  $\times$  indole NH), 10.82 (2H, s, 2  $\times$  indole NH); HPLC: 18.1 min (M4);  $m/z$  (ESI): 1998.0 (M + 2Na<sup>+</sup>. C<sub>212</sub>H<sub>298</sub>N<sub>36</sub>Na<sub>2</sub>O<sub>34</sub>Si requires 1997.1).

**Succ-1-(<sup>10,11</sup>[D-THF]-gA-TBDPS)-4-(gA-TBDPS) 8.** 70.0 mg (29.6  $\mu$ mol) of methyl ester **35** were *C*-deprotected according to GP 1. The crude carboxylic acid and 40.0 mg (22.5  $\mu$ mol) of amine **33a** were collected and coupled according to GP 5 (1.0 mM, 12 h). After aqueous work-up 81.0 mg of crude diamide **8** were obtained and purified by preparative HPLC (250 mm Rainin C 4, 80 $\rightarrow$ 90% B in 10 min, then 90 $\rightarrow$ 100% B in 15 min. A: H<sub>2</sub>O + 0.1% TFA; B: acetonitrile–isopropanol 2 : 1 + 0.1% TFA; 14 ml min<sup>–1</sup>). Treatment of the collected fractions according to GP 6 gave 43.0 mg (10.5  $\mu$ mol, 46%) of pure diamide **8** as a colourless solid.  $\delta_H$  (500 MHz; DMSO-*d*<sub>6</sub>; Me<sub>4</sub>Si) 0.40 (3H, d,  $J$  6.4, Leu  $\delta$ -H<sub>3</sub>), 0.49 (6H, d,  $J$  6.4, 2  $\times$  Leu  $\delta$ -H<sub>3</sub>), 0.51–0.62 (15H, m, 5  $\times$  Leu  $\delta$ -H<sub>3</sub>), 0.61–0.68 (9H, m, 3  $\times$  Leu  $\delta$ -H<sub>3</sub>), 0.72 (3H, d,  $J$  6.2, Leu  $\delta$ -H<sub>3</sub>), 0.74–0.91 (54H, m, 2  $\times$  Leu  $\delta$ -H<sub>3</sub> and 16  $\times$  Val  $\gamma$ -H<sub>3</sub>), 0.92–0.95 (3H, m, THF 2'-H<sub>3</sub>), 0.97 (9H, s, 3  $\times$  *t*-Bu-H<sub>3</sub>), 0.98 (9H, s, 3  $\times$  *t*-Bu H<sub>3</sub>), 1.09–1.32 (26H, m, 4  $\times$  Ala  $\beta$ -H<sub>3</sub>, 5  $\times$  Leu  $\beta$ -H<sub>2</sub> and 4  $\times$  Leu  $\gamma$ -H), 1.42–1.59 (8H, m, 2  $\times$  Leu  $\beta$ -H<sub>2</sub>, 2  $\times$  Val  $\beta$ -H, THF 4-*HH* and THF 3-*HH*), 1.61–1.67 (1H, m, Leu  $\gamma$ -H), 1.74–1.82 (1H, m, Leu  $\gamma$ -H), 1.90–2.05 (7H, m, 6  $\times$  Val  $\beta$ -H and Leu  $\gamma$ -H), 2.19 (1H, t,  $J$  7.3, THF 3-*HH*), 2.25–2.32 (1H, m, THF 4-*HH*), 2.36–2.47 (4H, m, succ 2-H<sub>2</sub> and 3-H<sub>2</sub>), 2.74–2.95 (6H, m, 3  $\times$  Trp  $\beta$ -H<sub>2</sub>), 3.06–3.29 (8H, m, 3  $\times$  Trp  $\beta$ -H<sub>2</sub> and AE 2-*HH*), 3.35–3.43 (2H, m, AE 2-*HH*), 3.46–3.50 (2H, m, Trp  $\beta$ -H<sub>2</sub>), 3.58–3.68 (6H, m, 2  $\times$  C-2 H<sub>2</sub> and Gly  $\alpha$ -H<sub>2</sub>), 3.69–3.75 (2H, m, 2  $\times$  Leu  $\alpha$ -H), 3.82–3.88 (1H, m, Leu  $\alpha$ -H), 3.90–3.95 (2H, m, THF 1'-H and Leu  $\alpha$ -H), 4.05–4.16 (5H, m, 2  $\times$  Val  $\alpha$ -H, Gly  $\alpha$ -H<sub>2</sub> and Leu  $\alpha$ -H), 4.17–4.34 (13H, m, 4  $\times$  Ala  $\alpha$ -H, Leu  $\alpha$ -H, THF 5-H and 2-H and 6  $\times$  Val  $\alpha$ -H), 4.41–4.61 (7H, m, 7  $\times$  Trp  $\alpha$ -H), 6.88–6.97 (6H, m, 6  $\times$  ar. CH), 6.99–7.05 (6H, m, 6  $\times$  ar. CH), 7.08 (6H, s, 6  $\times$  ar. CH), 7.13 (1H, s, ar. CH), 7.25–7.32 (7H, m, 7  $\times$  ar. CH), 7.36–7.47 (12H, 12  $\times$  ar. CH), 7.49–7.58 (7H, m, 7  $\times$  ar. CH), 7.59–7.63 (12H, m, 10  $\times$  ar. CH and 2  $\times$  NH), 7.64–7.67 (1H, m, THF NH), 7.68–7.82 (3H, m, NH), 7.85 (1H, d,  $J$  5.7, Leu NH), 7.87–7.96 (7H, m, 7  $\times$  NH), 7.98 (1H, d,  $J$  5.7, Leu NH), 8.01–8.18 (8H, m, 7  $\times$  NH and Trp NH), 8.21–8.31 (6H, m, Gly NH and 5  $\times$  Trp NH), 8.33 (1H, d,  $J$  6.4, Trp NH), 10.71

(1H, s, indole NH), 10.76 (5H, s, 5 × indole NH), 10.82 (1H, s, indole NH). HPLC: 10.6 min (10 × 250 mm Rainin C 4, 80→90% B in 10 min, then 90→100% B in 15 min. A: H<sub>2</sub>O + 0.1% TFA; B: acetonitrile-isopropanol 2 : 1 + 0.1% TFA; 3.3 ml min<sup>-1</sup>); *m/z* (ESI) 2077.4 (M + 2Na<sup>+</sup>. C<sub>222</sub>H<sub>308</sub>N<sub>38</sub>Na<sub>2</sub>O<sub>34</sub>Si<sub>2</sub> requires 2076.2).

## Abbreviations

### Amino acid residues

For the schemes the common one letter code was used, where capital letters denote L- and small letters D-configuration. V = L-Val, G = Gly, A = L-Ala, I = D-Leu, v = D-Val, W = L-Trp.

**Z**: carboxy-benzyl; **TBDPS**: *t*-butyl-diphenyl-silyl; **HOBt**: 1-hydroxy-benzotriazole monohydrate; **EDC**: *N*-ethyl-*N'*-(3-dimethylamino-propyl)-carbodiimide hydrochloride; **HBTU**: *O*-(benzotriazol-1'-yl)-1,1,3,3-tetramethyl-uronium hexafluorophosphate; **DIEA**: di-*iso*-propyl-ethylamine; **HATU**: *O*-(7'-aza-benzotriazol-1'-yl)-1,1,3,3-tetramethyl-uronium hexafluorophosphate; **HOAt**: 1-hydroxy-7-aza-benzotriazole; **FCC**: flash column chromatography; **TFE**: trifluoroethanol; **DMSO**: dimethylsulfoxide; **DMF**: dimethylformamide; **DIBAL-H**: diisobutyl aluminium hydride; **MCPBA**: *m*-chloro-perbenzoic acid.

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