Acid-Labile Protecting Groups for the Synthesis of Lipidated Peptides

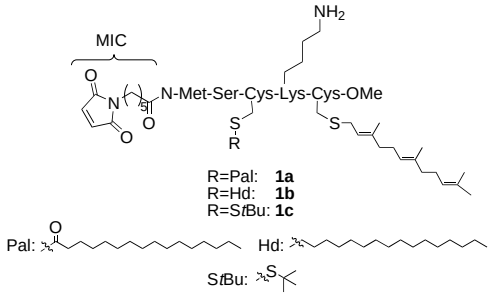
Dieter Kadereit,[a, b] Patrick Deck,[a] Ines Heinemann,[a] and Herbert Waldmann*[a]

Abstract: Lipidated peptides and their neolipoprotein derivatives are efficient tools for the investigation of biological processes in molecular detail. These compounds are often acid- and base-labile, and their synthesis requires the use of a combination of blocking groups that can be removed under very mild conditions. In this article we demonstrate that the Boc urethane and different trityl-type protecting groups can be cleaved selectively under acidic conditions that are mild enough to be compatible with the demands of lipopeptide synthesis. Thus, the Boc group was cleaved with TMS triflate in the presence of lutidine, and the methyltrityl (Mtt) and the methoxytrityl (Mmt) group were removed with 1% TFA in dichloromethane in the presence of triethylsilane as cation scavenger. Removal of the phenylfluorenyl group was achieved with up to 3% TFA in dichloromethane in the presence of triethylsilane at 0°C. These protecting-group techniques were successfully applied in the synthesis of differently lipidated H-Ras peptides.

Keywords: lipids · lipoproteins · peptides · protecting groups · Ras proteins

Introduction

Lipidated proteins embodying palmitic acid thioesters and farnesyl or geranylgeranyl thioethers play important roles in the transduction of extracellular signals across the cell membrane and in numerous intracellular processes like vesicle formation and targeting.[1, 2] Synthetic lipidated proteins carrying different lipid modifications may be employed as invaluable tools for the study of such biological processes in molecular detail. For example, we recently described the synthesis of lipidated Ras proteins based on the combination of the techniques of molecular biology and organic synthesis, that is, the bacterial expression of a truncated non-lipidated Ras protein and its coupling with different lipidated peptides via a maleimido linker.[2] Biophysical and cell-biological investigation of these neolipoproteins then gave insight into the mechanisms by which Ras proteins are specifically targeted to the plasma membrane, a process which is crucial to signalling via Ras. The availability of efficient methods for the synthesis of lipidated peptides is paramount to the success of this method. These peptide conjugates are both base-labile (if they embody palmitic acid thioesters) and acid-sensitive (if they embody prenyl groups, i.e. farnesyl- or geranylgeranyl thioethers) ruling out the use of many established protecting groups. While the introduction of enzyme- and noble-metal-sensitive protecting groups[3] provided efficient solutions to this synthetic problem additional degrees of freedom are urgently required if longer peptides and peptides incorporating amino acids with reactive side-chain functionalities like NH_2 and SH groups have to be constructed.[4] This problem is particularly apparent in the case of our target compound, the maleimido-modified H-Ras C-terminus **1a** (Scheme 1). Apart from the maleimido moiety, the structure carries an acid-labile farnesyl thioether, a base-labile thioester and a nucleophilic NH_2 side chain that requires protection during peptide synthesis or is susceptible to an undesired S → N acyl



Scheme 1. Differently modified target peptides corresponding to the H-Ras C-terminus for the synthesis of protein–peptide conjugates.

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shift. Therefore, for the NH_2 functionality a protecting group was required that is compatible with the demands of lipopeptide synthesis. The obvious Pd^0-catalyzed removal of allyl-type protecting groups, which is one of the most powerful methods for lipopeptide synthesis, cannot be employed in the presence of maleimido groups since under the conditions required for the removal of this blocking group undesired side reactions occur, in particular attack of the alkyl-accepting nucleophile and/or the phosphine ligands on the α,β-unsaturated double bond.

The very pronounced base lability of the palmitic acid thioesters (hydrolysis occurs at $pH > 7$) excludes the use of base-labile protection groups, an assumption that was easily confirmed by some preliminary experiments. Deprotection by oxidation or reduction was considered to be equally problematic owing to the presence of the thioether and the double bonds of the prenyl groups. However, initial orientating experiments revealed that the prenyl thioethers tolerated very low concentrations of acids like TFA. Since the palmitic acid thioesters are stable to acid treatment we investigated the use of different acid-sensitive protecting groups in lipopeptide synthesis. In this paper we report that the Boc urethane and different trityl-type blocking groups can be employed successfully for the synthesis of multifunctional lipopeptides. We demonstrate their use in the construction of maleimido-modified H-Ras peptides and the coupling of these peptide conjugates to a truncated H-Ras protein.

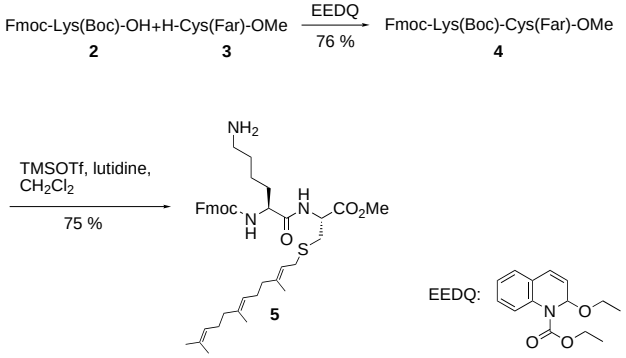
Results and Discussion

From the numerous acid-labile protecting groups available for the one approach, we chose the Boc group since it is one of the standard urethanes widely employed in peptide chemistry for which numerous deprotection conditions are available. For the other approach, trityl-type groups removable under mild conditions were investigated as possible alternatives for side-chain protection, since for this purpose urethane protecting groups are not necessary and since the lability of the trityl core can be fine-tuned by introducing appropriate substituents into the aromatic ring systems.

Initial experiments evaluating the suitability of these protecting groups were carried out with model dipeptides of the type Fmoc-Lys(Xxx)-Cys(Far)-OMe, which were synthesized from an appropriately protected amino acid derivative and *S*-farnesylated cysteine methyl ester.[5]

From the numerous different procedures for Boc cleavage, treatment with $SnCl_4$ in dichloromethane[6] and amine-buffered trimethylsilyl trifluoromethanesulfonate (TMSOTf)[7] were chosen as promising methods. Both had previously been successfully used in the presence of acid-labile functionalities. The former method was used for Boc deprotection in the presence of thioamides,[6] while the latter represents a procedure to remove *tert*-butyloxycarbonyl urethanes in the presence of *tert*-butyl esters or the acid-labile Rink linker.[8] Additionally, this technique offers the advantage that the reactivity can be adjusted by the choice and the relative amount of amine base.

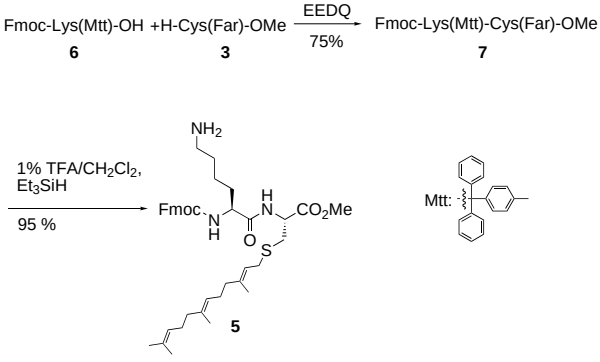
Treatment of dipeptide **4** as test substrate with $SnCl_4$ led to rapid cleavage of the Boc group as well as to side reactions in the farnesyl moiety, which were clearly evident as judged by NMR. In contrast, application of a TMSOTf/lutidine mixture removed the Boc group cleanly without side reactions (Scheme 2). Furthermore, a wide range of amines could be



Scheme 2. Cleavage of a Boc group in the presence of a farnesyl moiety. EEDQ: 1-ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline.

used including volatile amines like $EtMe_2N$. Evaporation of the excess amine and solvent after completion of the reaction avoids an aqueous workup step, which can be a tedious procedure when multiply lipid-modified peptides are involved.

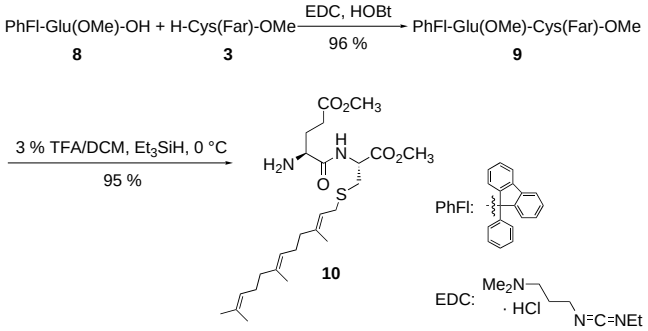
As an alternative for lysine side-chain protection, the 4-methyltrityl group[9] (Mtt) was investigated. This group can be removed by treatment with a mixture of acetic acid, trifluoroethanol, and CH_2Cl_2 (1:2:7) or a solution of 1 % TFA in CH_2Cl_2.[9] Application of the latter protocol resulted in cleavage of the Mtt group in dipeptide **7**. However, as we were concerned about concentrating the TFA during solvent removal, an excess of $EtMe_2N$ was added prior to distillation. After neutralization partial retritylation occurred, indicating that the addition of a cation scavenger is necessary. Subsequently, treatment of dipeptide **7** with the TFA solution in the presence of $EtSiH_3$ gave **5** in excellent yield (Scheme 3).



Scheme 3. Removal of the methyltrityl group in the presence of a farnesyl moiety.

Motivated by this success, we investigated the limitations of TFA-mediated removal of protecting groups in the presence of acid-labile prenyl groups. Test reactions demonstrated that

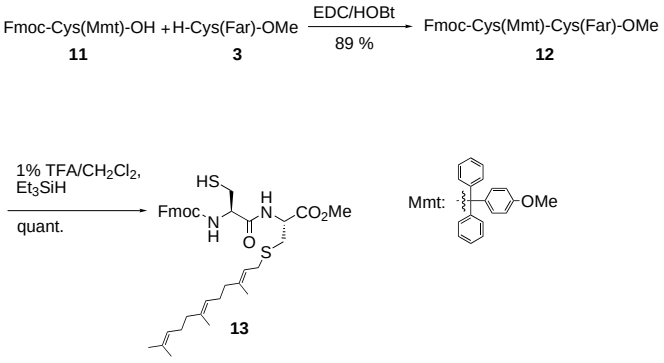
peptides incorporating the farnesyl group can be treated with up to 5 vol% of TFA in CH_2Cl_2 for one hour without side reactions. This was demonstrated by the removal of the phenylfluorenyl group (PhFl) from **9** (Scheme 4). The PhFl



Scheme 4. Removal of the phenylfluorenyl group in the presence of a farnesyl moiety. EDC: 1-ethyl-3(dimethylamino)propylcarbodiimide hydrochloride, HOBt: 1-hydroxybenzotriazole.

group is 6000 times more stable towards hydrolysis than the parent trityl group.[10] Nevertheless, it can be cleaved under mild conditions. Upon treatment of PhFl-protected glutamyl peptide **9** with 3% TFA in methylene chloride in the presence of Et_3SiH as cation scavenger, dipeptide **10** was isolated in 95% yield.

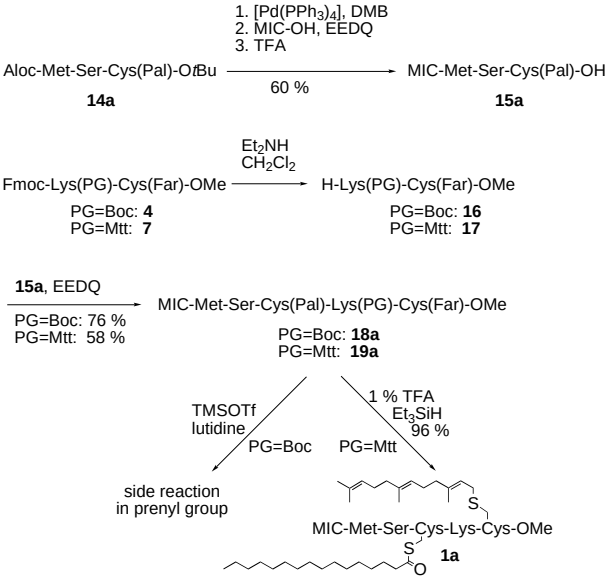
Trityl-type protecting groups with comparable acid lability are known for other functionalities as well. For instance, the *S*-methoxytrityl group (Mmt) represents the equivalent thiol group protection, and can be removed by treatment with 1% TFA in dichloromethane. This was demonstrated for dipeptide **12** as a test substrate. The free thiol **13** was isolated in quantitative yield under similar conditions to those used for *N*-Mtt removal (Scheme 5).



Scheme 5. Removal of the *S*-methoxytrityl group (Mmt) in the presence of a farnesyl moiety.

Having established two different types of acid-labile protecting groups, namely the Boc urethane and a trityl-type blocking function, for the synthesis of prenylated peptides, we tested the Boc and Mtt groups in the synthesis of the palmitoylated, farnesylated, and maleimido-modified target peptide **1a**. For this purpose a block condensation strategy was chosen because it readily gives access to differently protected pentapeptides and differently lipidated analogues thereof.

To start with, the allyloxycarbonyl (Aloc) group in the palmitoylated tripeptide **14a**[11] was replaced by the maleimido linker group in a two-step procedure. Subsequent acid-mediated removal of the *tert*-butyl ester gave fully modified building block **15a** (Scheme 6). The other building blocks, **16**



Scheme 6. Application of Boc and Mtt protecting-group strategies in the synthesis of **1a**. DMB: *N*,*N*′-dimethyl barbituric acid.

and **17**, were accessible by careful removal of the Fmoc group from Boc- and Mtt-protected dipeptides **4** and **7**, respectively. Dipeptides of this type carry the intrinsic risk of diketopiperazine formation by nucleophilic attack of the unmasked lysine α-amino group on the cysteine methyl ester, thereby rendering isolation of the dipeptides **16** and **17** problematic. This side reaction was not observed during Fmoc removal. However, after subsequent block condensation the diketopiperazines were isolated as the major by-products.

As the final step, the protecting groups on the lysine side chain in pentapeptides **18a** and **19a** had to be removed. In the case of the Boc group the conditions that were successfully applied for deprotection of dipeptide **4** permitted undesired side reactions in the prenyl group. Variation of this procedure did not result in an improvement. However, to our delight, treatment of Mtt-protected pentapeptide **19a** with 1% TFA in dichloromethane and Et_3SiH gave target peptide **1a** in 96% isolated yield (Scheme 6). No undesired side reactions were observed at all.

This result prompted us to select the Mtt protecting group for the synthesis of the differently lipid-modified analogous target peptides **1b** and **1c**. These analogues were synthesized by means of the strategy developed for palmitoylated peptide **1a**. The required tripeptide building blocks were synthesized by condensation of dipeptide **20**, which is accessible by *N*-hydroxysuccinimide(HOSu)-mediated coupling of Fmoc-Met-OH with serine,[12] with the already modified cysteine *tert*-butyl ester **21b** or **21c**. Fmoc removal, condensation with maleimidocaproic acid, and subsequent treatment with TFA delivered the tripeptide building blocks **15b** and **15c** (Scheme 7).

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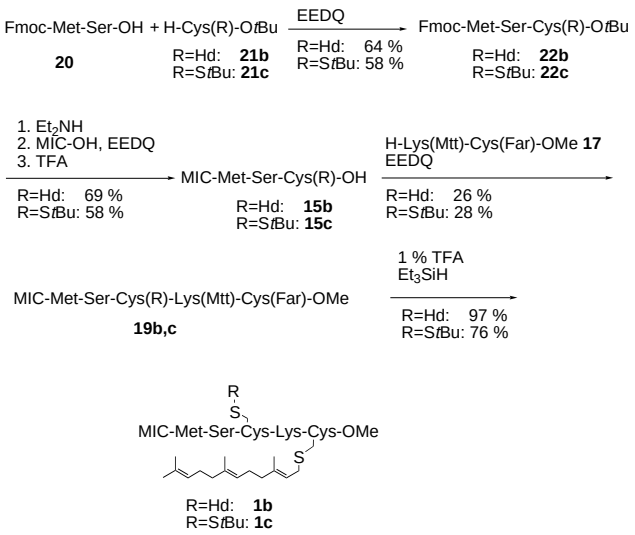

Scheme 7. Synthesis of the analogous peptides **1b** and **1c** by the Mtt strategy.

In the case of both tripeptides **15b** and **15c** the subsequent block condensation with dipeptide **17** gave the desired pentapeptides in only low yield. It proved difficult to improve this result as significant amounts of diketopiperazine were formed as a side product during the condensation reaction, and the separation of diketopiperazine and pentapeptide was problematic. The final deprotection reaction under the conditions described above once more proceeded smoothly and without any undesired side reactions. The target peptides **1b** and **1c** were isolated in 97% and in 76% yield, respectively.

Conclusion

Acid-labile protecting groups can be cleaved in the presence of acid-sensitive prenyl groups under mild conditions. In particular, trityl-based groups, namely the *N*-methyltrityl (Mtt), the *N*-phenylfluorenyl (PhFl), and the *S*-methoxytrityl (Mmt) group, may be applied advantageously in the synthesis of lipidated H-Ras peptides.

Experimental Section

General: ^{1}H and ^{13}C NMR spectra were recorded on Bruker AC-250, Bruker AM-400, and Bruker DRX-500 spectrometers and were based on tetramethylsilane (TMS) as internal standard. EI-MS and FAB-MS were recorded on a Finnigan MAT 90 mass spectrometer with 3-nitrobenzyl alcohol (3-NBA) as a matrix for FAB-MS. Combustion analysis was carried out with a Heraeus CHN-Rapid analyser. Flash chromatography was executed on columns packed with Baker silica gel (30–60 μm). TLC was performed on Kieselgel 60 F_{254} aluminum sheets (Merck Darmstadt, Germany). All reagents were obtained from Fluka, Aldrich, Sigma, or Novabiochem. All solvents were dried and distilled by standard procedures.

***N*$^{(\alpha)}$-Fluorenylmethoxycarbonyl-*N*$^{(\varepsilon)}$-*tert*-butyloxycarbonyl-(L)-lysyl-*S*-farnesyl-(L)-cysteine methyl ester (Fmoc-Lys(Boc)-Cys(Far)-OMe) (4)**: 1-Ethoxycarbonyl-2-ethoxy-1,2-dihydroquinoline (EEDQ; 227 mg, 0.917 mmol) was added to a solution of Fmoc-Lys(Boc)-OH (**2**; 331 mg, 0.705 mmol) and H-Cys(Far)-OMe[5] (**3**; 239 mg, 0.705 mmol) in dichloromethane (10 mL). After 16 h the solvent was evaporated in vacuo and the residue was dissolved in ethyl acetate (50 mL). The solution was extracted twice with 0.5 N hydrochloric acid (50 mL) and saturated $NaHCO_3$ solution (50 mL), dried over $MgSO_4$, and filtered. The solvent was evaporated in vacuo and the product was isolated by flash chromatography (hexane/ethyl acetate 2:1) to yield **4** (425 mg, 0.538 mmol, 76%) as a colorless solid. M.p. 57 °C; $[\alpha]_D^{20} = -8.9$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz, TMS): $\delta = 7.76$ (d, $J = 7.5$ Hz, 2H, Fmoc-CH), 7.60 (d, $J = 7.0$ Hz, 2H, Fmoc-CH), 7.40 (t, $J = 7.4$ Hz, 2H, Fmoc-CH), 7.31 (t, $J = 7.4$ Hz, 2H, Fmoc-CH), 6.71 (b, 1H, NH), 5.50 (b, 1H, NH), 5.15–5.19 (m, 1H, CH=C Far), 5.04–5.12 (m, 2H, CH=C Far), 4.72–4.79 (m, 1H, α-CH Cys), 4.66 (b, 1H, NH), 4.38–4.45 (m, 2H, Fmoc-CH_2), 4.20–4.26 (m, 2H, α-CH Lys, Fmoc-CH), 3.74 (s, 3H, $COOCH_3$), 3.06–3.20 (m, 4H, CH_2 Far, ε-CH_2 Lys), 2.95 (dd, $J = 13.9$ Hz, $J = 4.8$ Hz, 1H, β-CH_{2a} Cys), 2.85 (dd, $J = 13.9$ Hz, $J = 6.3$ Hz, 1H, β-CH_{2b} Cys), 1.93–2.10 (m, 8H, CH_2 Far), 1.80–1.92 (m, 1H, CH_{2a} Lys), 1.38–1.74 (m, 5H, CH_2 Lys), 1.68 (s, 3H, CH_3 Far), 1.62 (s, 6H, CH_3 Far), 1.60 (s, 3H, CH_3 Far), 1.43 (s, 9H, CH_3 Boc); ^{13}C NMR ($CDCl_3$, 100 MHz, TMS): $\delta = 171.7$ (*C*=O), 171.1 (*C*=O), 156.2 (*C*=O), 143.9 (quart, Fmoc), 143.7 (quart, Fmoc), 141.3 (quart, Fmoc), 140.1 (quart, Far), 135.3 (quart, Far), 131.3 (quart, Far), 127.7 (Fmoc-CH), 127.1 (Fmoc-CH), 125.1 (Fmoc-CH), 124.3 (Far-CH), 123.7 (Far-CH), 120.0 (Fmoc-CH), 119.4 (Far-CH), 79.1 (quart, Boc), 67.1 (Fmoc-CH_2), 54.6 (α-CH), 52.6 (COO*C*H_3), 51.8 (α-CH), 47.1 (Fmoc-CH), 39.9 (CH_2), 39.7 (CH_2), 39.6 (CH_2), 33.1 (CH_2), 32.2 (CH_2), 29.8 (CH_2), 29.6 (CH_2), 28.4 (Boc-CH_3), 26.7 (CH_2), 26.4 (CH_2), 25.7 (Far-CH_3), 22.3 (CH_2), 17.7 (Far-CH_3), 16.1 (Far-CH_3), 16.0 (Far-CH_3); MS (FAB, 3-NBA): *m*/*z*: 790.4 $[M+H]^+$, 690.4 $[M - Boc+H]^+$.

***N*$^{(\alpha)}$-Fluorenylmethoxycarbonyl-*N*$^{(\varepsilon)}$-(4-tolyldiphenylmethyl)-(L)-lysyl-*S*-farnesyl-(L)-cysteine methyl ester (Fmoc-Lys(Mtt)-Cys(Far)-OMe) (7)**: EEDQ (142 mg, 0.575 mmol) was added to a solution of Fmoc-Lys(Mtt)-OH[9] (**6**; 276 mg, 0.442 mmol) and H-Cys(Far)-OMe[5] (**3**; 150 mg, 0.442 mmol) in dichloromethane (5 mL). After 20 hours the solvent was evaporated in vacuo and the residue was dissolved in ethyl acetate (30 mL). The solution was extracted twice with 0.5 N hydrochloric acid (20 mL) and saturated $NaHCO_3$ solution (20 mL), dried over $MgSO_4$, and filtered. The solvent was evaporated in vacuo and the product was isolated by flash chromatography (hexane/ethyl acetate 3:1) to yield **6** (312 mg, 0.330 mmol, 75%) as a colorless oil. $[\alpha]_D^{20} = -10.1$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz, TMS): $\delta = 7.74$ (d, $J = 7.1$ Hz, 2H, Fmoc-CH), 7.57 (d, $J = 7.9$ Hz, 2H, Fmoc-CH), 7.45 (d, $J = 7.6$ Hz, 4H, Mtt-CH), 7.23–7.38 (m, 10H, Fmoc-CH, Mtt-CH), 7.15 (t, $J = 7.3$ Hz, 2H, Mtt-CH), 7.06 (d, $J = 8.1$ Hz, 2H, Mtt-CH), 6.59 (b, 1H, NH), 5.29 (d, $J = 7.7$ Hz, 1H, NH), 5.14–5.17 (m, 1H, CH=C Far), 5.05–5.10 (t, $J = 7.7$ Hz, 2H, CH=C Far), 4.72–4.79 (m, 1H, α-CH Cys), 4.37–4.43 (m, 2H, Fmoc-CH_2), 4.16–4.22 (m, 2H, α-CH Lys, Fmoc-CH), 3.69 (s, 3H, $COOCH_3$), 3.15 (dd, $J = 13.1$ Hz, $J = 8.2$ Hz, 1H; CH_{2a} Far), 3.06 (dd, $J = 13.2$ Hz, $J = 7.4$ Hz, 1H, CH_{2b} Far), 2.93 (dd, $J = 13.9$ Hz, $J = 4.8$ Hz, 1H, β-CH_{2a} Cys), 2.85 (dd, $J = 13.9$ Hz, $J = 5.0$ Hz, 1H, β-CH_{2b} Cys), 2.29 (s, 3H, Mtt-CH_3), 1.93–2.12 (m, 10H, CH_2 Far, ε-CH_2 Lys), 1.78–1.88 (m, 1H, CH_{2a} Lys), 1.45–1.74 (m, 3H, CH_2 Lys), 1.67 (s, 3H, CH_3 Far), 1.62 (s, 3H, CH_3 Far), 1.59 (s, 3H, CH_3 Far), 1.58 (s, 3H, CH_3 Far), 1.41–1.49 (m, 2H, CH_2 Lys); ^{13}C NMR ($CDCl_3$, 125 MHz, TMS): $\delta = 171.6$ (*C*=O), 171.0 (*C*=O), 156.0 (*C*=O), 146.4 (quart, Mtt), 143.9 (quart, Fmoc), 143.7 (quart, Fmoc), 143.3 (quart, Mtt), 141.3 (quart, Fmoc), 140.1 (quart, Far), 135.3 (quart, Far), 135.4 (quart, Mtt), 131.3 (quart, Far), 128.6 (Mtt, CH), 128.6 (Mtt, CH), 128.5 (Mtt, CH), 127.7 (Fmoc-CH), 127.1 (Fmoc-CH), 126.1 (Mtt, CH), 125.1 (Fmoc-CH), 124.3 (Far-CH), 123.7 (Far-CH), 120.0 (Fmoc-CH), 119.5 (Far-CH), 70.6 (quart, Mtt), 67.1 (Fmoc-CH_2), 54.8 (α-CH), 52.6 (COO*C*H_3), 51.7 (α-CH), 47.2 (Fmoc-CH), 43.4 (CH_2), 39.7 (CH_2), 39.6 (CH_2), 33.1 (CH_2), 32.9 (CH_2), 30.7 (CH_2), 29.9 (CH_2), 29.7 (CH_2), 26.4 (CH_2), 25.7 (Far-CH_3), 23.2 (CH_2), 20.9 (Mtt-CH_3), 17.7 (Far-CH_3), 16.1 (Far-CH_3), 16.0 (Far-CH_3); MS (FAB, 3-NBA): *m*/*z*: 945.4 $[M+H]^+$, 868.4 $[M - C_6H_5]^+$, 854.4 $[M - C_6H_4CH_3]^+$, 257.1 $[Mtt]^+$.

***N*$^{(\alpha)}$-Fluorenylmethoxycarbonyl-(L)-lysyl-*S*-farnesyl-(L)-cysteine methyl ester (Fmoc-Lys-Cys(Far)-OMe) (5)**: *By Boc deprotection*: TMSOTf (12.0 μL, 66.5 μmol) was added to a solution of Fmoc-Lys(Boc)-Cys(Far)-OMe (**4**; 10.5 mg, 13.3 μmol) and 2,6-lutidine (15.4 μL, 133 μmol) in dry dichloromethane (0.7 mL). After one hour, dichloromethane (20 mL) was added, the solution was extracted twice with 0.5 N hydrochloric acid (20 mL) and saturated $NaHCO_3$ solution (20 mL), dried over $MgSO_4$, and filtered. The solvent was evaporated in vacuo and the product was isolated by flash chromatography to yield **5** (6.9 mg, 9.98 μmol, 75%) as a colorless solid.

By Mtt removal: Et_3SiH (14.1 μL, 88.8 μmol) was added to a solution of Fmoc-Lys(Mtt)-Cys(Far)-OMe (**7**; 8.4 mg, 8.88 μmol) in a 1% solution of TFA in dichloromethane (0.7 mL). Within five minutes the mixture changed from yellow to colorless. After 60 minutes a solution of 1% $EtMe_2N$ in dichloromethane (0.8 mL) was added, the solvent was evaporated in vacuo, and the product was isolated by flash chromatography ($CHCl_3$/MeOH 8:1) to yield **5** (5.8 mg, 8.41 μmol, 95%) as a colorless solid. M.p. 57 °C; $[\alpha]_D^{20} = -4.3$ ($c = 0.3$, $CHCl_3$); ^{1}H NMR ($CDCl_3/CD_3OD$ 6:1, 500 MHz, TMS): $\delta = 7.77$ (d, $J = 7.5$ Hz, 2H, Fmoc-CH), 7.60–7.64 (m, 2H, Fmoc-CH), 7.40 (t, $J = 7.4$ Hz, 2H, Fmoc-CH), 7.30–7.6 (m, 2H, Fmoc-CH), 5.19 (t, $J = 7.5$ Hz, 1H, CH=C Far), 5.08 (t, $J = 6.9$ Hz, 2H, CH=C Far), 4.67 (dd, $J = 7.5$ Hz, $J = 5.0$ Hz, 1H, α-CH Cys), 4.37–4.44 (m, 2H, Fmoc-CH_2), 4.23 (t, $J = 6.8$ Hz, 2H, α-CH Lys, Fmoc-CH), 3.75 (s, 3H, $COOCH_3$), 3.20 (dd, $J = 12.9$ Hz, $J = 8.4$ Hz, 1H, CH_{2a} Far), 3.10 (dd, $J = 12.7$ Hz, $J = 7.3$ Hz, 1H, CH_{2b} Far), 2.90–2.97 (m, 3H, β-CH_{2a} Cys, ε-CH_2 Lys), 2.81 (dd, $J = 13.9$ Hz, $J = 7.6$ Hz, 1H, β-CH_{2b} Cys), 1.93–2.10 (m, 10H, CH_2 Far, CH_2 Lys), 1.78–1.85 (m, 1H, CH_{2a} Lys), 1.55–1.74 (m, 1H, CH_{2b} Lys), 1.68 (s, 3H, CH_3 Far), 1.65 (s, 3H, CH_3 Far), 1.60 (s, 3H, CH_3 Far), 1.59 (s, 3H, CH_3 Far), 1.31–1.42 (m, 2H, CH_2 Lys); ^{13}C NMR ($CDCl_3/CD_3OD$ 6:1, 125 MHz, TMS): $\delta = 172.5$ (*C*=O), 171.5 (*C*=O), 156.8 (*C*=O), 144.0 (quart, Fmoc), 143.8 (quart, Fmoc), 141.4 (quart, Fmoc), 140.2 (quart, Far), 135.5 (quart, Far), 131.5 (quart, Far), 127.9 (Fmoc-CH), 127.2 (Fmoc-CH), 125.2 (Fmoc-CH), 124.4 (Far-CH), 123.8 (Far-CH), 120.0 (Fmoc-CH), 119.6 (Far-CH), 67.2 (Fmoc-CH_2), 54.2 (α-CH), 52.7 ($COOCH_3$), 52.2 (α-CH), 47.2 (Fmoc-CH), 39.8 (CH_2), 39.8 (CH_2), 39.3 (CH_2), 32.7 (CH_2), 31.9 (CH_2), 29.7 (CH_2), 26.8 (CH_2), 26.7 (CH_2), 26.6 (CH_2), 25.7 (Far-CH_3), 22.0 (CH_2), 17.7 (Far-CH_3), 16.1 (Far-CH_3), 16.0 (Far-CH_3); MS (FAB, 3-NBA): *m/z*: 690.5 $[M+H]^+$; HRMS (FAB, 3-NBA) calcd for $C_{40}H_{55}N_3O_5S$ (689.95): 690.3941; found: 690.3954.

***N*-[9-(9-Phenylfluorenyl)]-α-(L)-(γ-methylglutamyl)-*S*-farnesyl-(L)-cysteine methyl ester (PhFl-Glu(OMe)-Cys(Far)-OMe) (9)**: At 0 °C, 1-ethyl-3(dimethylamino)propylcarbodiimide hydrochloride (EDC; 173 mg, 0.884 mmol) was added to a solution of PhFl-Glu(OMe)-OH[13] (**8**, 300 mg, 0.736 mmol), H-Cys(Far)-OMe (**3**, 250 mg, 0.736 mmol) and 1-hydroxybenzotriazole (HOBt; 230 mg, 1.47 mmol) in dichloromethane (40 mL). After 20 h the solution was extracted twice with 0.5 N hydrochloric acid (10 mL) and 10% Na_2CO_3 solution (10 mL), dried over Na_2SO_4, and filtered. The solvent was evaporated in vacuo and the product was isolated by flash chromatography (hexane/ethyl acetate 3:1) to yield **9** (518 mg, 0.7 mmol, 96%) as a colorless oil. $[\alpha]_D^{20} = -8.9$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz, TMS): $\delta = 7.69–7.09$ (m, 14H, NH, PhFl), 5.19 (m, 1H, CH=C Far), 5.10 (m, 2H, CH=C Far), 4.48 (m, 1H, α-CH Cys), 3.75 (s, 3H, OCH_3 Cys), 3.59 (s, 3H, OCH_3 Glu), 3.09 (m, 3H, CH_2 Far, NH), 2.75 (dd, $J = 14$ Hz, $J = 5$ Hz, 1H, β-CH_{2a} Cys), 2.68 (dd, $J = 14$ Hz, $J = 5$ Hz, 1H, β-CH_{2b} Cys), 2.48 (m, 1H, α-CH Glu), 2.40–2.25 (m, 2H, γ-CH_2 Glu), 2.13–1.96 (m, 8H, CH_2 Far), 1.75 (m, 2H, β-CH_2 Glu), 1.68 (s, 3H, CH_3 Far), 1.66 (s, 3H, CH_3 Far), 1.60 (s, 6H, CH_3 Far); ^{13}C NMR ($CDCl_3$, 100 MHz, TMS): $\delta = 174.4$ (*C*=O), 171.7 (*C*=O), 141.1 (quart, PhFl), 140.8 (quart, PhFl), 140.4 (quart, Far), 135.8 (quart, Far), 131.7 (quart, Far), 128.9 (quart, PhFl), 128.4 (CH PhFl), 127.8 (CH PhFl), 127.7 (CH PhFl), 126.9 (CH PhFl), 125.3 (CH PhFl), 124.7 (CH Far), 124.1 (CH Far), 120.4 (CH PhFl), 120.3 (CH PhFl), 119.9 (CH Far), 73.4 (quart, PhFl), 56.6 (α-CH), 52.9 (OCH_3), 52.0 (OCH_3), 51.6 (α-CH), 40.1 (CH_2), 34.1 (CH_2), 30.6 (CH_2), 30.3 (CH_2), 30.1 (CH_2), 27.1 (CH_2), 26.8 (CH_2), 26.1 (CH_2), 18.1 (CH_3 Far), 16.5 (CH_3 Far), 16.4 (CH_3 Far); MS (FAB, 3-NBA): *m/z* calcd for $C_{44}H_{54}N_2O_5S$: 722.9760; found: 722.9719.

α-(L)-(γ-Methylglutamyl)-*S*-farnesyl-(L)-cysteine methyl ester (H-Glu-(OMe)-Cys(Far)-OMe) (10): At 0 °C, Et_3SiH (440 μL, 2.77 mmol) was added to a solution of **9** (200 mg, 0.277 mmol) in a 3% solution of TFA in dichloromethane. Within five minutes the mixture turned from yellow to colorless. After 3 h at 0 °C Et_2MeN (1 mL) was added, the solvent was evaporated in vacuo, and the product was isolated by flash chromatography ($CHCl_3$/MeOH 30:1) to yield **10** (127 mg, 0.263 mmol, 95%) as a yellowish oil. $[\alpha]_D^{20} = -12.7$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 400 MHz, TMS): $\delta =$ 5.21 (m, 1H, CH=C Far), 5.09 (m, 2H, CH=C Far), 4.75 (m, 1H, α-CH Cys), 3.76 (s, 3H, OCH_3 Cys), 3.68 (s, 3H, OCH_3 Glu), 3.48 (m, 1H, NH), 3.24–3.11 (m, 3H, CH_2 Far, α-CH Glu), 2.95 (dd, $J = 14$ Hz, $J = 5$ Hz, 1H, β-CH_{2a} Cys), 2.88 (dd, $J = 14$ Hz, $J = 5$ Hz, 1H, β-CH_{2b} Cys), 2.53–2.46 (m, 2H, γ-CH_2 Glu), 2.15–1.88 (m, 10H, CH_2 Far, β-CH_2 Glu), 1.68 (s, 6H, CH_3 Far), 1.60 (s, 6H, CH_3 Far); ^{13}C NMR ($CDCl_3$, 100 MHz, TMS): $\delta = 174.2$ (*C*=O), 171.7 (*C*=O), 140.4 (quart, Far), 135.8 (quart, Far), 131.7 (quart, Far), 124.7 (CH Far), 124.1 (CH Far), 119.9 (CH Far), 54.8 (α-CH), 52.9 (OCH_3), 52.1 (OCH_3), 51.8 (α-CH), 40.0 (CH_2), 33.8 (CH_2), 30.7 (CH_2), 30.5 (CH_2), 30.2 (CH_2), 27.1 (CH_2), 26.8 (CH_2), 26.1 (CH_2), 18.1 (CH_3 Far), 16.5 (CH_3 Far), 16.4 (CH_3 Far); MS (FAB, 3-NBA): *m/z* calcd for $C_{25}H_{42}N_2O_5S$: 482.6775; found: 482.6736.

***N*-Fluorenylmethoxycarbonyl-(*S*-4-methoxytrityl)-(L)-cysteyl-(*S*-farnesyl)-(L)-cysteine methyl ester (Fmoc-Cys(Mmt)-Cys(Far)-OMe) (12)**: At 0 °C, HOBt (48 mg, 0.35 mmol) and EDC (54 mg, 0.28 mmol) were added to a solution of Fmoc-Cys(Mmt)-OH[14] (145 mg, 2.40 mmol) and **3** (80 mg, 2.4 mmol) in dichloromethane (20 mL). The solution was stirred at room temperature for 15 h. Ethyl acetate was added and the solution was extracted twice with 0.5 N hydrochloric acid (15 mL) and with 0.5 N $NaHCO_3$ solution (15 mL), and once with brine (15 mL), dried over Na_2SO_4, and filtered. The solvent was evaporated in vacuo and the product was isolated by flash chromatography (*n*-hexane/ethyl acetate 2:1) to yield **12** (197 mg, 89%) as a colorless oil. $[\alpha]_D^{20} = -3.2$ ($c = 0.5$); ^{1}H NMR ($CDCl_3$, 500 MHz, TMS): $\delta = 7.74$ (dd, $J = 9.8$ Hz, $J = 7.7$ Hz, 2H, Fmoc-CH), 7.56 (b, 2H, Fmoc-CH), 7.42 (d, $J = 7.6$ Hz, 4H, Mmt-CH), 7.25–7.40 (m, 10H, Fmoc-CH, Mmt-CH), 7.20 (t, $J = 7.3$ Hz, 2H, Mmt-CH), 6.80 (d, $J = 8.9$ Hz, 2H, Mmt-CH), 6.61 (d, $J = 7.8$ Hz, 1H, NH), 5.14 (t, $J = 7.6$ Hz, 1H, CH=C Far), 5.03–5.09 (m, 2H, CH=C Far), 4.35–4.36 (m, 2H, Fmoc-CH_2), 4.21 (t, $J = 6.9$ Hz, 1H, Fmoc-CH), 3.82–3.86 (m, 2H, α-CH Cys), 3.76 (s, 3H, $COOCH_3$), 3.69 (s, 3H, OCH_3), 3.11 (dd, $J = 13.1$ Hz, $J =$ 8.2 Hz, 1H, CH_{2a} Far), 3.05 (dd, $J = 13.0$ Hz, $J = 7.4$ Hz, 1H, CH_{2b} Far), 2.89 (dd, $J = 13.9$ Hz, $J = 5.0$ Hz, 1H, β-CH_{2a} Cys), 2.80 (dd, $J = 13.9$ Hz, $J = 6.0$ Hz, 1H, β-CH_{2b} Cys), 2.72 (dd, $J = 13.3$ Hz, $J = 7.7$ Hz, 1H, β-CH_{2a} Cys), 2.65 (dd, $J = 13.3$ Hz, $J = 5.3$ Hz, 1H, β-CH_{2b} Cys), 1.94–2.06 (m, 8H, CH_2 Far), 1.67 (s, 3H, CH_3 Far), 1.62 (s, 3H, CH_3 Far), 1.59 (s, 3H, CH_3 Far), 1.57 (s, 3H, CH_3 Far); ^{13}C NMR ($CDCl_3$, 125 MHz, TMS): $\delta = 170.7$ (*C*=O), 170.0 (*C*=O), 158.3 (*C*=O), 155.9 (quart, Mmt), 144.7 (quart, Mmt), 143.8 (quart, Fmoc), 143.7 (quart, Fmoc), 141.3 (quart, Fmoc), 140.0 (quart, Far), 136.4 (quart, Mmt), 135.3 (quart, Far), 131.3 (quart, Far), 130.8 (Mmt, CH), 129.5 (Mmt, CH), 128.1 (Mmt, CH), 127.7 (Fmoc-CH), 127.1 (Fmoc-CH), 126.9 (Mmt, CH), 125.1 (Fmoc-CH), 124.3 (Far-CH), 123.7 (Far-CH), 120.0 (Fmoc-CH), 119.5 (Far-CH), 113.4 (Mmt, CH), 67.2 (Fmoc-CH_2), 66.9 (quart, Mmt), 55.2 ($COOCH_3$), 53.9 (OCH_3), 52.5 (α-CH), 52.0 (α-CH), 47.1 (Fmoc-CH), 39.7 (CH_2), 39.6 (CH_2), 33.8 (CH_2), 33.2 (CH_2), 29.9 (CH_2), 26.7 (CH_2), 26.4 (CH_2), 25.7 (CH_3), 17.7 (CH_3), 16.1 (CH_3), 16.0 (CH_3); HRMS (FAB, 3-NBA): *m/z* calcd for $C_{57}H_{64}NaN_2S_2O_6$ ($[M+Na]^+$): 959.4139; found: 959.4104.

***N*-Fluorenylmethoxycarbonyl-(L)-cysteyl-(*S*-farnesyl)-(L)-cysteine methyl ester (Fmoc-Cys-Cys(Far)-OMe) (13)**: At room temperature, 1% trifluoroacetic acid in dichloromethane (1.5 mL) was added to a solution of **12** (30 mg, 32 μmol) in dichloromethane (0.2 mL). Et_3SiH (51 μL, 0.32 mmol) was added to the orange solution. After 30 min toluene (4 mL) was added and the solvents were evaporated in vacuo. The addition of toluene and evaporation was repeated twice. The product was isolated by flash chromatography (*n*-hexane/ethyl acetate 2:1) to yield **13** quantitatively as a colorless solid. M.p. 67–69 °C; $[\alpha]_D^{20} = -8.5$ ($c = 0.7$); ^{1}H NMR ($CDCl_3$, 500 MHz, TMS): $\delta = 7.77$ (d, $J = 7.5$ Hz, 2H, Fmoc-CH), 7.60 (d, $J = 6.7$ Hz, 2H, Fmoc-CH), 7.40 (t, $J = 7.5$ Hz, 2H, Fmoc-CH), 7.31–7.34 (m, 2H, Fmoc-CH), 6.91 (b, 1H, NH), 5.75 (b, 1H, NH), 5.17–5.20 (m, 1H, CH=C Far), 5.07–5.10 (m, 2H, CH=C Far), 4.74–4.78 (m, 1H, α-CH Cys), 4.45–4.47 (m, 3H, α-CH Cys, Fmoc-CH_2), 4.24 (t, $J = 6.8$ Hz, 1H, Fmoc-CH), 3.76 (s, 3H, $COOCH_3$), 3.19 (dd, $J = 13.1$ Hz, $J = 8.2$ Hz, 1H, CH_{2a} Far), 3.06–3.15 (m, 1H, β-CH_{2a} Cys), 3.11 (dd, $J = 13.1$ Hz, $J = 7.4$ Hz, 1H, CH_{2b} Far), 2.99 (dd, $J = 13.9$ Hz, $J = 4.7$ Hz, 1H, β-CH_{2a} Cys), 2.85 (dd, $J =$ 13.4 Hz, $J = 6.3$ Hz, 1H, β-CH_{2b} Cys), 2.69–2.80 (m, 1H, β-CH_{2b} Cys), 1.95–2.11 (m, 8H, CH_2 Far), 1.68 (s, 3H, CH_3 Far), 1.65 (s, 3H, CH_3 Far), 1.60 (s, 3H, CH_3 Far), 1.59 (s, 3H, CH_3 Far); ^{13}C NMR ($CDCl_3$, 125 MHz, TMS): $\delta = 170.9$ (*C*=O), 169.5 (*C*=O), 155.9 (*C*=O), 155.9, 143.8 (quart, Fmoc), 143.6 (quart, Fmoc), 141.4 (quart, Fmoc), 140.3 (quart, Far), 135.4 (quart, Far), 131.3 (quart, Far), 127.8 (Fmoc-CH), 127.1 (Fmoc-CH), 125.0 (Fmoc-CH), 124.3 (Far-CH), 123.7 (Far-CH), 120.1 (Fmoc-CH), 119.3 (Far-CH), 67.3 (Fmoc-CH_2), 55.9 ($COOCH_3$), 52.8 (α-CH), 51.9 (α-CH), 47.2 (Fmoc-CH), 39.7 (CH_2), 39.6 (CH_2), 39.3 (CH_2), 34.3 (CH_2), 33.1 (CH_2), 29.8 (CH_2), 27.2 (CH_2), 26.7 (CH_2), 26.4 (CH_2), 25.7 (CH_3), 17.7 (CH_3), 16.1 (CH_3), 16.0 (CH_3); HRMS (FAB, 3-NBA): *m/z* calcd for $C_{37}H_{49}N_2S_2O_5$: 665.3118; found: 665.3083.

(L)-Methionyl-(L)-seryl-*S*-palmitoyl-(L)-cysteine *tert*-butyl ester (H-Met-Ser-Cys(Pal)-O*t*Bu): $PhSiH_3$ (34 μL, 0.279 mmol) and $[Pd(PPh_3)_4]$ (16 mg,

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0.0139 mmol) were added under an argon atmosphere to a solution of Aloc-Met-Ser-Cys(Pal)-O*t*Bu[11] (**14a**; 100 mg, 0.139 mmol) in THF (1 mL). After two hours the solvent was evaporated in vacuo and H-Met-Ser-Cys(Pal)-O*t*Bu was isolated by size-exclusion chromatography (Sephadex LH-20, $CHCl_3$/MeOH 1:1) as a colorless, waxy solid (50 mg, 0.0789 mmol, 57%). $[\alpha]_D^{20} = -20.9$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR (500 MHz, $CDCl_3$, TMS): $\delta = 8.06$ (d, $J = 7.7$ Hz, 1H, NH), 7.27 (d, $J = 7.3$ Hz, 1H, NH), 4.66–4.70 (m, 1H, α-CH), 4.47–4.50 (m, 1H, α-CH), 4.07 (dd, $J = 11.5$ Hz, $J = 5.3$ Hz, 1H, β-CH_{2a} Ser), 3.60 (dd, $J = 11.5$ Hz, $J = 4.7$ Hz, 1H, β-CH_{2b} Ser), 3.59 (dd, $J = 8.1$ Hz, $J = 4.7$ Hz, 1H, α-CH Met), 3.47 (dd, $J = 14.1$ Hz, $J = 4.1$ Hz, 1H, β-CH_{2a} Cys), 3.27 (dd, $J = 14.1$ Hz, $J = 6.1$ Hz, 1H, β-CH_{2b} Cys), 2.60–2.65 (m, 2H, γ-CH_2 Met), 2.56 (t, $J = 7.6$ Hz, 2H, CH_2 Pal), 2.43 (b, 2H, NH_2), 2.11–2.19 (m, 1H, β-CH_{2a} Met), 2.11 (s, 3H, SCH_3 Met), 1.77–1.84 (m, 1H, β-CH_{2b} Met), 1.58–1.65 (m, 2H, CH_2 Pal), 1.46 (s, 9H, $CO_2C(CH_3)_3$), 1.25 (s, 24H, $CH_3(CH_2)_{12}(CH_2)_2$COS), 0.87 (t, $J = 6.9$ Hz, 3H, ω-CH_3 Pal); ^{13}C NMR (125 MHz, $CDCl_3$, TMS): $\delta = 199.4$, 175.3, 170.7, 168.8 (4 *C*=O), 83.3 ($CO_2C(CH_3)_3$), 62.8 (β-CH_2 Ser), 54.2, 54.0, 53.0 (3 α-CH), 44.1 (β-CH_2 Cys), 33.9, 31.9, 30.6, 30.2, 29.7, 29.7, 29.6, 29.4, 29.4, 29.2, 29.0 (11 CH_2), 27.9 ($CO_2C(CH_3)_3$), 25.6, 22.7 (2 CH_2), 15.3 (SCH_3 Met), 14.1 (CH_3 Pal); MS (FAB, 3-NBA): *m/z*: 634.4 $[M+H]^+$; elemental analysis calcd (%) for $C_{31}H_{59}N_3O_6S_2$: C 58.55, H 8.84, N 5.85; found: C 58.49, H 8.61, N 5.54.

Maleimidocaproyl-(L)-methionyl-(L)-seryl-*S*-palmitoyl-(L)-cysteine *tert*-butyl ester (MIC-Met-Ser-Cys(Pal)-O*t*Bu): EEDQ (29.3 mg, 118 μmol) was added to a solution of MIC-OH (18.3 mg, 86.8 μmol) and H-Met-Ser-Cys(Pal)-O*t*Bu (50 mg, 78.9 μmol) in dichloromethane (1 mL). After 18 hours the solvent was evaporated in vacuo and the product was isolated by flash chromatography (dichloromethane/ethanol 30:1, then 20:1) as a colorless, waxy solid (41.5 mg, 50.2 μmol, 64%). $[\alpha]_D^{20} = -3.6$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz, TMS): $\delta = 7.37-7.39$ (m, 2H, NH), 6.78 (d, $J = 7.9$ Hz, 1H, NH), 6.69 (s, 2H, MIC-CH), 4.63–4.74 (m, 2H, α-CH Cys, α-CH Met), 4.57–4.62 (m, 1H, α-CH Ser), 3.99 (dd, $J = 11.4$ Hz, $J = 4.2$ Hz, 1H, β-CH_{2a} Ser), 3.69 (dd, $J = 11.5$ Hz, $J = 5.6$ Hz, 1H, β-CH_{2b} Ser), 3.49–3.53 (m, 2H, MIC-NCH_2), 3.46 (dd, $J = 14.0$ Hz, $J = 4.4$ Hz, 1H, β-CH_{2a} Cys), 3.32 (dd, $J = 14.0$ Hz, $J = 6.1$ Hz, 1H, β-CH_{2b} Cys), 2.52–2.61 (m, 4H, γ-CH_2 Met, CH_2 Pal), 2.23 (t, $J = 7.5$ Hz, 2H, MIC), 2.08–2.15 (m, 1H, β-CH_{2a} Met), 2.11 (s, 3H, SCH_3 Met), 1.95–2.03 (m, 1H, β-CH_{2b} Met), 1.56–1.69 (m, 6H, CH_2 Pal, CH_2 MIC), 1.46 (s, 9H, $CO_2C(CH_3)_3$), 1.24–1.36 (m, 26H, $CH_3(CH_2)_{12}(CH_2)_2$COS, CH_2 MIC), 0.88 (t, $J = 6.9$ Hz, 3H, ω-CH_3 Pal); ^{13}C NMR ($CDCl_3$, 125 MHz, TMS): $\delta = 199.0$ (*C*=O), 173.2 (*C*=O), 171.9 (*C*=O), 170.9 (2 *C*=O MIC), 170.1 (*C*=O), 168.9 (*C*=O), 134.1 (2 CH MIC), 83.3 ($CO_2C(CH_3)_3$), 62.8 (β-CH_2 Ser), 54.6 (α-CH), 53.1 (α-CH), 52.3 (α-CH), 44.1 (β-CH_2 Cys), 37.6 (CH_2), 36.1 (CH_2), 33.6 (CH_2), 31.9 (CH_2), 30.3 (CH_2), 30.1 (CH_2), 29.7 (CH_2), 29.7 (CH_2), 29.6 (CH_2), 29.4 (CH_2), 29.4 (CH_2), 29.2 (CH_2), 29.0 (CH_2), 28.2 (CH_2), 27.9 ($CO_2C(CH_3)_3$), 26.3 (CH_2), 26.2 (CH_2), 25.6 (CH_2), 25.0 (CH_2), 22.7 (CH_2), 15.3 (SCH_3 Met), 14.1 (CH_3 Pal); MS (FAB, 3-NBA): *m/z*: 849 $[M+Na]^+$, 827 $[M+H]^+$, 771 $[M - t\text{Bu}+H]^+$; HRMS (FAB, 3-NBA): calcd for $C_{41}H_{71}N_4O_9S_2$: 827.4662; found: 827.4622.

Maleimidocaproyl-(L)-methionyl-(L)-seryl-*S*-palmitoyl-(L)-cysteine (MIC-Met-Ser-Cys(Pal)-OH) (15a): TFA (1 mL) was added to a solution of MIC-Met-Ser-Cys(Pal)-O*t*Bu (35.7 mg, 43.2 μmol) in dichloromethane (1 mL). After 3 h toluene (2 mL) was added and the solvents were evaporated in vacuo. The addition of toluene and evaporation was repeated twice to yield **15a** quantitatively as a colorless solid. M.p. 161 °C; $[\alpha]_D^{20} = -8.5$ ($c = 1.1$, $CHCl_3/CH_3OH$ 2:1); ^{1}H NMR ($CDCl_3/CD_3OD$ 6:1, 500 MHz, TMS): $\delta =$ 6.75 (s, 2H, MIC-CH), 4.65 (dd, $J = 7.4$ Hz, $J = 4.6$ Hz, 1H, α-CH Cys), 4.53 (dd, $J = 8.3$ Hz, $J = 5.6$ Hz, 1H, α-CH Met), 4.44 (t, $J = 5.1$ Hz, 1H, α-CH Ser), 3.87 (dd, $J = 11.4$ Hz, $J = 4.8$ Hz, 1H, β-CH_{2a} Ser), 3.73 (dd, $J =$ 11.4 Hz, $J = 5.3$ Hz, 1H, β-CH_{2b} Ser), 3.50–3.54 (m, 3H, MIC-NCH_2, β-CH_{2a} Cys), 3.27 (dd, $J = 14.0$ Hz, $J = 7.5$ Hz, 1H, β-CH_{2b} Cys), 2.52–2.61 (m, 4H, γ-CH_2 Met, CH_2 Pal), 2.25 (t, $J = 7.5$ Hz, 2H, MIC), 2.09–2.17 (m, 1H, β-CH_2 Met), 2.12 (s, 3H, SCH_3 Met), 1.92–1.99 (m, 1H, β-CH_{2a} Met), 1.57–1.70 (m, 6H, CH_2 Pal, 4CH_2 MIC), 1.24–1.36 (m, 26H, $CH_3(CH_2)_{12}(CH_2)_2$COS, CH_2 MIC), 0.88 (t, $J = 6.9$ Hz, 3H, ω-CH_3 Pal); ^{13}C NMR ($CDCl_3/CD_3OD$ 6:1, 125 MHz, TMS): $\delta = 199.9$ (*C*=O), 174.8 (*C*=O), 172.6 (*C*=O), 172.1 (*C*=O), 171.6 (2 *C*=O MIC), 170.8 (*C*=O), 134.5 (2 CH MIC), 62.3 (β-CH_2 Ser), 55.3 (α-CH), 52.9 (α-CH), 52.8 (α-CH), 44.3 (β-CH_2 Cys), 37.9 (CH_2), 36.1 (CH_2), 34.1 (CH_2), 32.2 (CH_2), 31.6 (CH_2), 30.4 (CH_2), 30.3 (CH_2), 30.0 (CH_2), 29.9 (CH_2), 29.9 (CH_2), 29.7 (CH_2), 29.6 (CH_2), 29.5 (CH_2), 29.3 (CH_2), 28.5 (CH_2), 26.6 (CH_2), 26.5 (CH_2), 25.8 (CH_2), 25.4 (CH_2), 22.9 (CH_2), 15.3 (SCH_3 Met), 14.2 (CH_3 Pal); MS (FAB, 3-NBA): *m/z*: 793.4 $[M+Na]^+$, 771.4 $[M+H]^+$; HRMS (FAB, 3-NBA) calcd for $C_{37}H_{63}N_4O_9S_2$: 771.4036; found 771.4068.

Maleimidocaproyl-(L)-methionyl-(L)-seryl-*S*-palmitoyl-(L)-cysteyl-$N^{(\varepsilon)}$-*tert*-butoxycarbonyl-(L)-lysyl-*S*-farnesyl-(L)-cysteine methyl ester (MIC-Met-Ser-Cys(Pal)-Lys(Boc)-Cys(Far)-OMe) (18a): Et_2NH (0.1 mL) was added to a solution of **7** (28.8 mg, 36.5 μmol) in dichloromethane (0.4 mL). After 2 h the solvent was evaporated in vacuo, the residue was dissolved in dichloromethane (2 mL), and **15a** (23.4 mg, 30.4 μmol) and EEDQ (9.8 mg, 36.5 μmol) were added. After 18 h the solvent was evaporated in vacuo and the product was isolated by flash chromatography (dichloromethane/ethanol 30:1, then 20:1) to yield **18a** (30.3 mg, 22.9 μmol, 76%) as a colorless, waxy solid. M.p. 119 °C; $[\alpha]_D^{20} = -24.6$ ($c = 1.3$, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz): $\delta = 7.63$ (b, 1H, NH), 7.45 (b, 1H, NH), 7.29 (b, 1H, NH), 7.15 (b, 1H, NH), 6.98 (d, $J = 7.3$ Hz, 1H, NH), 6.69 (s, 2H, MIC-CH), 5.17–5.27 (m, 1H, CH=C Far), 5.07–5.15 (m, 2H, CH=C Far), 4.74–4.86 (m, 1H, α-CH), 4.63–4.72 (m, 2H, α-CH), 4.44–4.51 (m, 1H, α-CH), 4.05–4.10 (m, 1H, α-CH), 3.95–4.03 (m, 1H, β-CH_{2a} Ser), 3.78–3.88 (m, 1H, β-CH_{2b} Ser), 3.76 (s, 3H, $COOCH_3$), 3.50 (t, $J = 7.2$ Hz, 2H, MIC-NCH_2), 3.05–3.45 (m, 6H, 2 CH_2 Far, 2 ε-CH_2 Lys, β-CH_2 Cys_{Pal}), 2.94–3.04 (m, 1H, β-CH_{2a} Cys_{Far}), 2.85 (m, 1H, β-CH_2 Cys_{Far}), 2.49–2.67 (m, 4H, γ-CH_2 Met, CH_2 Pal), 2.21–2.27 (m, 2H, MIC), 1.88–2.19 (m, 14H, β-CH_2 Met, SCH_3 Met, CH_{2a} Lys, CH_2 Far), 1.35–1.72 (m, 11H, CH_2 Pal, CH_2 MIC, CH_2 Lys), 1.68 (s, 3H, CH_3 Far), 1.62 (s, 6H, CH_3 Far), 1.60 (s, 3H, CH_3 Far), 1.45 (s, 9H, CH_3 Boc), 1.22–1.34 (m, 26H, $CH_3(CH_2)_{12}(CH_2)_2$COS, CH_2 MIC), 0.88 (t, 3H, $J = 6.9$ Hz, ω-CH_3 Pal); ^{13}C NMR ($CDCl_3$, 125 MHz): $\delta = 173.4$ (*C*=O), 171.7 (*C*=O), 170.8 (*C*=O), 169.4 (*C*=O), 167.5 (*C*=O), 166.9 (*C*=O), 156.3 (*C*=O), 140.0 (quart, Far), 135.3 (quart, Far), 134.1 (2 CH MIC), 131.3 (quart, Far), 124.3 (Far-CH), 123.8 (Far-CH), 119.5 (Far-CH), 79.1 (quart, Boc), 64.2 (β-CH_2 Ser), 54.9 (α-CH), 53.8 (α-CH), 53.4 (α-CH), 52.9 (α-CH), 52.6 ($COOCH_3$), 44.1 (CH_2), 40.4 (CH_2), 39.7 (CH_2), 39.6 (CH_2), 37.7 (CH_2), 36.0 (CH_2), 35.5 (CH_2), 32.7 (CH_2), 31.9 (CH_2), 29.7 (CH_2), 29.7 (CH_2), 29.6 (CH_2), 29.5 (CH_2), 29.4 (CH_2), 29.1 (CH_2), 29.0 (CH_2), 28.4 (Boc-CH_3), 26.8 (CH_2), 26.6 (CH_2), 26.4 (CH_2), 25.7 (Far-CH_3), 25.7 (CH_2), 22.6 (CH_2), 17.7 (Far-CH_3), 16.2 (Far-CH_3), 16.0 (Far-CH_3) 15.4 (SCH_3 Met), 14.1 (CH_3 Pal); MS (FAB, 3-NBA): *m/z*: 1342.9 $[M+Na]^+$, 1322.0 $[M+H]^+$, 1220.8 $[M - \text{Boc}+H]^+$; HRMS (FAB, 3-NBA) calcd for $C_{62}H_{106}N_7O_{11}S_3$ ($[M - \text{Boc}+H]^+$): 1220.711; found: 1220.719.

Maleimidocaproyl-(L)-methionyl-(L)-seryl-*S*-palmitoyl-(L)-cysteyl-$N^{(\varepsilon)}$-(4-tolyldiphenylmethyl)-(L)-lysyl-*S*-farnesyl-(L)-cysteine methyl ester (MIC-Met-Ser-Cys(Pal)-Lys(Mtt)-Cys(Far)-OMe) (19a): Et_2NH (0.2 mL) was added to a solution of **7** (50.0 mg, 55.1 μmol) in dichloromethane (0.8 mL). After 4 h the solvent was evaporated in vacuo, the residue was dissolved in dichloromethane (2 mL), and **15a** (32.7 mg, 42.4 μmol) and EEDQ (13.6 mg, 55.1 μmol) were added. After 16 h the solvent was evaporated in vacuo and the product was isolated by flash chromatography (dichloromethane/ethanol 30:1, then 20:1) to yield **19a** (36.5 mg, 24.7 μmol, 58%) as a colorless, waxy solid. M.p. 183 °C; $[\alpha]_D^{20} = -23.8$ ($c = 1.6$, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz, TMS): $\delta = 7.65$ (b, 1H, NH), 7.52 (b, 1H, NH), 7.49 (d, $J = 7.6$ Hz, 4H, Mtt-CH), 7.41 (d, $J = 7.2$ Hz, 2H, Mtt-CH), 7.34–7.38 (m, 6H, Mtt-CH), 7.21 (d, $J = 8.1$ Hz, 2H, Mtt-CH), 6.69 (s, 2H, MIC-CH), 5.16–5.20 (m, 1H, CH=C Far), 5.07–5.11 (m, 2H, CH=C Far), 4.57–4.63 (m, 1H, α-CH), 4.47–4.53 (m, 1H, α-CH), 4.38–4.44 (m, 2H, α-CH), 4.15–4.23 (m, 1H, α-CH), 3.88–3.95 (m, 1H, β-CH_{2a} Ser), 3.75–3.80 (m, 1H, β-CH_{2b} Ser), 3.68 (s, 3H, $COOCH_3$), 3.48 (t, $J = 7.2$ Hz, 2H, MIC-NCH_2), 3.39–3.45 (m, 1H, β-CH_{2a} Cys_{Pal}), 3.13–3.25 (m, 2H, 1 CH_{2a} Far, β-CH_2 Cys_{Pal}), 3.07–3.12 (m, 1H, 1 CH_{2b} Far), 2.81–3.01 (m, 4H, β-CH_2 Cys_{Far}, β-CH_2 Lys), 2.49–2.62 (m, 4H, γ-CH_2 Met, CH_2 Pal), 2.35 (s, 3H, Mtt-CH_3), 2.19–2.28 (m, 2H, MIC), 1.92–2.15 (m, 14H, β-CH_2 Met, SCH_3 Met, CH_{2a} Lys, CH_2 Far), 1.35–1.72 (m, 11H, CH_2 Pal, CH_2 MIC, CH_2 Lys), 1.68 (s, 3H, CH_3 Far), 1.65 (s, 3H, CH_3 Far), 1.60 (s, 6H, CH_3 Far), 1.19–1.38 (m, 26H, $CH_3(CH_2)_{12}(CH_2)_2$COS, CH_2 MIC), 0.88 (t, $J = 6.7$ Hz, 3H, ω-CH_3 Pal); ^{13}C NMR ($CDCl_3$, 125 MHz, TMS): $\delta = 173.3$ (*C*=O), 171.8 (*C*=O), 170.7 (*C*=O), 168.9 (*C*=O), 146.5 (quart, Mtt), 143.2 (quart, Mtt), 140.0 (quart, Far), 135.5 (quart, Mtt), 135.3 (quart, Far), 134.0 (2 CH MIC), 131.3 (quart, Far), 128.6 (Mtt, 3 CH), 127.1 (Mtt, CH), 126.1 (Mtt, CH), 124.3 (Far-CH), 123.8 (Far-CH), 119.5 (Far-CH), 70.6 (quart, Mtt), 54.7 (α-CH), 53.8 (α-CH), 53.2 (α-CH), 52.5 ($COOCH_3$), 44.0 (CH_2), 40.4 (CH_2), 43.6 (CH_2), 39.7 (CH_2), 39.6 (CH_2), 37.7 (CH_2), 36.0 (CH_2), 35.7 (CH_2), 32.8 (CH_2), 31.9 (CH_2), 30.2 (CH_2), 29.7 (CH_2), 29.7 (CH_2), 29.4 (CH_2), 29.1

(CH_2), 28.4 (CH_2), 27.9 (CH_2), 26.8 (CH_2), 26.6 (CH_2), 26.4 (CH_2), 25.7 (Far-CH_3), 25.6 (CH_2), 25.2 (CH_2), 22.7 (CH_2), 20.9 (Mtt-CH_3), 17.7 (Far-CH_3), 16.2 (Far-CH_3), 16.0 (Far-CH_3), 15.3 (SCH_3 Met), 14.1 (CH_3 Pal); MS (FAB, 3-NBA): *m/z*: 1474.5 $[M-H]^+$, 1399.6 $[M-C_6H_5]^+$, 1385.5 $[M-C_6H_4CH_3]^+$, 1220.8 $[M-Mtt+H]^+$, 257.3 $[Mtt]^+$.

Maleimidocaproyl-(L)-methionyl-(L)-seryl-*S*-palmitoyl-(L)-cysteyl-(L)-lysyl-*S*-farnesyl-(L)-cysteine methyl ester (MIC-Met-Ser-Cys(Pal)-Lys-Cys-(Far)-OMe) (1a): Et_3SiH (16.5 μL, 104 μmol) was added to a solution of **19a** (15.3 mg, 10.4 μmol) in a 1% solution of TFA in dichloromethane (0.5 mL). After 70 min a solution of 1% $EtMe_2N$ in dichloromethane (0.6 mL) was added, the solvent was evaporated in vacuo, and the product was isolated by size-exclusion chromatography (Sephadex LH-20, $CHCl_3$/MeOH 1:1) to yield **1a** (12.1 mg, 9.91 μmol, 96%) as a colorless, waxy solid. M.p. 160 °C; $[\alpha]_D^{20} = -24.0$ ($c = 0.6$, $CHCl_3$); ^{1}H NMR ($CDCl_3$/CD_3OD 4:1, 500 MHz, TMS): $\delta = 7.99$ (d, $J = 6.8$ Hz, 1H, NH), 7.89–7.93 (m, 2H, NH), 7.85 (d, $J = 6.7$ Hz, 1H, NH), 7.70 (d, $J = 8.0$ Hz, 1H, NH), 6.73 (s, 2H, MIC-CH), 5.17–5.23 (m, 1H, CH=C Far), 5.05–5.14 (m, 2H, CH=C Far), 4.62–4.67 (m, 1H, α-CH), 4.38–4.50 (m, 3H, α-CH), 4.31–4.36 (m, 1H, α-CH), 3.77–3.81 (m, 2H, β-CH_2 Ser), 3.76 (s, 3H, $COOCH_3$), 3.51 (t, $J = 7.2$ Hz, 2H, 2MIC-NCH_2), 3.35–3.41 (m, 1H, β-CH_{2a} Cys_{Pal}), 3.19–3.28 (m, 2H, CH_{2a} Far, β-CH_{2b} Cys_{Pal}), 3.09–3.16 (m, 1H, CH_{2b} Far), 2.92–3.00 (m, 3H, β-CH_{2a} Cys_{Far}, ε-CH_2 Lys), 2.78–2.84 (m, 1H, β-CH_{2a} Cys_{Far}), 2.51–2.62 (m, 4H, γ-CH_2 Met, CH_2 Pal), 2.24–2.32 (m, 2H, MIC), 1.88–2.14 (m, 14H, β-CH_2 Met, SCH_3 Met, CH_{2a} Lys, CH_2 Far), 1.52–1.76 (m, 9H, CH_2 Pal, CH_2 MIC, CH_2 Lys), 1.68 (s, 6H, CH_3 Far), 1.60 (s, 6H, CH_3 Far), 1.38–1.52 (m, 2H, CH_2 Lys), 1.21–1.38 (m, 26H, $CH_3(CH_2)_{12}(CH_2)_2COS$, CH_2 MIC), 0.88 (t, $J = 6.9$ Hz, 3H, ω-CH_3 Pal); ^{13}C NMR ($CDCl_3$/CD_3OD 4:1, 125 MHz, TMS): $\delta = 200.6$ (*C*=O), 175.2 (*C*=O), 173.0 (*C*=O), 172.0 (*C*=O), 171.4 (*C*=O), 171.3 (*C*=O), 170.8 (*C*=O), 140.3 (quart, Far), 135.5 (quart, Far), 134.3 (2CH MIC), 131.5 (quart, Far), 124.5 (Far-CH), 123.9 (Far-CH), 119.7 (Far-CH), 61.8 (β-CH_2 Ser), 55.8 (α-CH), 54.6 (α-CH), 53.1 (α-CH), 52.8 (α-CH), 52.7 ($COOCH_3$), 52.3 (α-CH), 44.2 (CH_2), 39.8 (CH_2), 39.6 (CH_2), 37.8 (CH_2), 35.9 (CH_2), 32.7 (CH_2), 32.1 (CH_2), 31.0 (CH_2), 30.9 (CH_2), 30.4 (CH_2), 29.8 (CH_2), 29.8 (CH_2), 29.7 (CH_2), 29.6 (CH_2), 29.5 (CH_2), 29.4 (CH_2), 29.2 (CH_2), 28.4 (CH_2), 26.9 (CH_2), 26.6 (CH_2), 26.4 (CH_2), 26.4 (CH_2), 25.7 (Far-CH_3), 25.7 (CH_2), 25.2 (CH_2), 22.8 (CH_2), 22.1 (CH_2), 17.7 (Far-CH_3), 16.2 (Far-CH_3), 16.1 (Far-CH_3), 15.3 (SCH_3 Met), 14.1 (CH_3 Pal); MS (FAB, 3-NBA): *m/z*: 1220.5 $[M+H]^+$.

***N*-Fluorenylmethoxycarbonyl-(L)-methionyl-(L)-serine (Fmoc-Met-Ser-OH) (20)**: DCC (1.00 g, 4.85 mmol) was added at 0 °C to a solution of Fmoc-Met-OH (1.50 g, 4.04 mmol) and *N*-hydroxysuccinimide (0.47 g, 4.04 mmol) in dry THF (5 mL). After 2.5 h the solution was filtered and the solvent was evaporated in vacuo. The residue was dissolved in dioxane (10 mL) and added at 0 °C to a solution of serine (0.64 g, 6.06 mmol) and NaOH (0.24 g, 6.06 mmol) in water (10 mL). After the mixture had spent two days at ambient temperature, water (50 mL) was added and the solution was extracted twice with ethyl acetate (50 mL). By addition of hydrochloric acid the pH of the aqueous layer was adjusted to 3 and the solution was again extracted with ethyl acetate (3 × 50 mL). The combined organic layers were dried over $MgSO_4$ and filtered, and the solvent was evaporated in vacuo to yield **20** (1.52 mg, 3.32 mmol, 82%) as a colorless solid, which was used without further purification. $[\alpha]_D^{20} = +10.6$ ($c = 1.0$, chloroform/methanol 2:1); ^{1}H NMR ($CDCl_3$/CD_3OD 6:1, 500 MHz, TMS): $\delta = 7.75$ (d, $J = 7.5$ Hz, 2H, CH Fmoc), 7.61 (t, $J = 6.5$ Hz, 2H, CH Fmoc), 7.39 (t, $J = 7.4$ Hz, 2H, CH Fmoc), 7.30 (t, $J = 7.5$ Hz, 2H, CH Fmoc), 4.55 (t, $J = 3.5$ Hz, 1H, α-CH Ser), 4.43 (dd, $J = 10.5$ Hz, $J = 7.2$ Hz, 1H, α-CH Met), 4.34–4.39 (m, 2H, CH_2 Fmoc), 4.21 (t, $J = 6.9$ Hz, 1H, CH Fmoc), 3.98 (dd, $J = 11.5$ Hz, $J = 3.9$ Hz, 1H, β-CH_{2a} Ser), 3.86 (dd, $J = 11.5$ Hz, $J = 3.4$ Hz, 1H, β-CH_{2b} Ser), 2.51–2.60 (m, 2H, γ-CH_2 Met), 2.05–2.14 (m, 1H, β-CH_{2a} Met), 2.09 (s, 3H, Met-CH_3), 1.86–1.98 (m, 1H, β-CH_{2b} Met); ^{13}C NMR ($CDCl_3$/CD_3OD 6:1, 125 MHz, TMS): $\delta = 172.5$ (*C*=O), 172.3 (*C*=O), 157.0 (*C*=O), 144.1 (quart, arom, Fmoc), 143.9 (quart, arom, Fmoc), 141.5 (quart, arom, Fmoc), 128.0 (arom, Fmoc-CH), 127.3 (arom, Fmoc-CH), 125.3 (arom, Fmoc-CH), 120.2 (arom, Fmoc-CH), 67.3 (Fmoc-CH_2), 62.3 (β-CH_2 Ser), 55.0 (α-CH), 54.2 (α-CH), 47.3 (Fmoc-CH), 32.1 (γ-CH_2 Met), 30.2 (β-CH_2 Met), 15.2 (SCH_3 Met); MS (FAB, 3-NBA): *m/z*: 481.2 $[M+Na]^+$, 459.2 $[M+H]^+$; HRMS (FAB, 3-NBA) calcd for $C_{23}H_{27}N_2O_6S$: 459.1590; found 459.1564, elemental analysis calcd (%) for $C_{23}H_{26}N_2O_6S$: C 60.25, H 5.72, N 6.11; found:C 60.09, H 5.82, N 5.63.

***S*-Hexadecyl-(L)-cysteine *tert*-butyl ester (H-Cys(Hd)-O*t*Bu) (21)**: Under an argon atmosphere, dithiothreitol (1.4 g, 9.08 mmol) was added to a solution of (H-Cys-O*t*Bu)$_2$[15] (500 mg, 2.27 mmol) and Et_3N (630 μL, 4.54 mmol) in dichloromethane (10 mL). After 100 min, dichloromethane (40 mL) was added and the solution was extracted three times with water (50 mL), dried over $MgSO_4$, and filtered. The solvent was evaporated in vacuo, the residue was dissolved in dry DMF (10 mL), and hexadecyl bromide (2.77 mL, 9.08 mmol) and Et_3N (630 μL, 4.54 mmol) were added under an argon atmosphere. After two days, water (50 mL) was added and the solution was extracted three times with diethyl ether (50 mL). The combined organic layers were dried over $MgSO_4$ and filtered, the solvent was evaporated in vacuo, and the product was isolated by flash chromatography (hexane/ethyl acetate 2:1) to yield **21b** (723 mg, 1.80 mmol, 40%) as a colorless oil. $[\alpha]_D^{20} = -3.5$ ($c = 2.0$, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz, TMS): $\delta = 3.52$ (dd, $J = 7.3$ Hz, $J = 4.7$ Hz, 1H, α-CH Cys), 2.89 (dd, $J = 13.3$ Hz, $J = 4.6$ Hz, 1H, β-CH_{2a} Cys), 2.74 (dd, $J = 13.3$ Hz, $J = 7.3$ Hz, 1H, β-CH_{2b} Cys), 2.54 (t, $J = 7.4$ Hz, 2H, CH_2 Hd), 2.00 (s, 2H, NH_2), 1.53–1.61 (m, 2H, CH_2 Hd), 1.48 (s, 9H, $CO_2C(CH_3)_3$), 1.31–1.39 (m, 2H, CH_2 Hd), 1.25 (s, 24H, $CH_3(CH_2)_{12}(CH_2)_3S$), 0.88 (t, $J = 6.8$ Hz, 3H, ω-CH_3 Hd); ^{13}C NMR ($CDCl_3$, 125 MHz, TMS): $\delta = 173.2$ (*C*=O), 81.6 ($CO_2C(CH_3)_3$), 54.7 (α-CH Cys), 37.4 (CH_2), 32.8 (CH_2), 31.9 (CH_2), 29.7 (CH_2), 29.7 (CH_2), 29.6 (CH_2), 29.5 (CH_2), 29.4 (CH_2), 29.2 (CH_2), 28.8 (CH_2), 28.0 ($CO_2C(CH_3)_3$), 22.7 (CH_2), 14.1 (CH_3 Hd); MS (FAB, 3-NBA): *m/z*: 402.4 $[M+H]^+$, 346.3 $[M-tBu+H]^+$.

***N*-Fluorenylmethoxycarbonyl-(L)-methionyl-(L)-seryl-*S*-hexadecyl-(L)-cysteine *tert*-butyl ester (Fmoc-Met-Ser-Cys(Hd)-O*t*Bu) (22b)**: EEDQ (382 mg, 1.55 mmol) was added to a solution of **20** (600 mg, 1.31 mmol) and H-Cys(Hd)-O*t*Bu (**21b**; 478 mg, 1.19 mmol) in DMF (7 mL). After 3 days, ethyl acetate (50 mL) was added and the solution was extracted twice with 0.5 N hydrochloric acid (50 mL), saturated $NaHCO_3$ solution (50 mL), and brine (50 mL), dried over $MgSO_4$, and filtered. The solvent was evaporated in vacuo and the product was isolated by flash chromatography (hexane/ethyl acetate 1:1) to yield **22b** (639 mg, 0.759 mmol, 64%) as a colorless solid. M.p. 114 °C; $[\alpha]_D^{20} = -12.3$ ($c = 1.5$, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz, TMS): $\delta = 7.76$ (d, $J = 7.5$ Hz, 2H, Fmoc-CH), 7.59 (dd, $J = 7.4$ Hz, $J = 3.8$ Hz, 2H, Fmoc-CH), 7.40 (t, $J = 7.4$ Hz, 2H, Fmoc-CH), 7.32 (t, $J = 7.4$ Hz, 2H, Fmoc-CH), 7.03–7.09 (m, 2H, NH), 5.50 (d, $J = 7.4$ Hz, 1H, NH), 4.61–4.69 (m, 1H, α-CH), 4.48–4.54 (m, 1H, α-CH), 4.38–4.45 (m, 3H, Fmoc-CH_2, α-CH), 4.22 (t, $J = 6.9$ Hz, 1H, Fmoc-CH), 4.05–4.12 (m, 1H, β-CH_{2a} Ser), 3.62–3.71 (m, 1H, β-CH_{2b} Ser), 2.98 (dd, $J = 13.8$ Hz, $J = 4.5$ Hz, 1H, β-CH_{2a} Cys), 2.91 (dd, $J = 13.6$ Hz, $J = 6.1$ Hz, 1H, β-CH_{2b} Cys), 2.52–2.61 (m, 2H, γ-CH_2 Met), 2.50 (t, $J = 7.4$ Hz, 2H, CH_2 Hd), 2.11 (s, 3H, SCH_3 Met), 2.08–2.16 (m, 1H, β-CH_{2a} Met), 1.95–2.03 (m, 1H, β-CH_{2b} Met), 1.49–1.58 (m, 2H, CH_2 Hd), 1.47 (s, 9H, $CO_2C(CH_3)_3$), 1.22–1.38 (m, 26H, $CH_3(CH_2)_{13}(CH_2)_2S$), 0.88 (t, $J = 6.9$ Hz, 3H, ω-CH_3 Hd); ^{13}C NMR ($CDCl_3$, 125 MHz, TMS): $\delta = 171.8$ (*C*=O), 170.2 (*C*=O), 169.7 (*C*=O), 156.2 (*C*=O), 143.8 (quart, Fmoc), 143.7 (quart, Fmoc), 141.3 (quart, Fmoc), 127.7 (Fmoc-CH), 127.1 (Fmoc-CH), 125.1 (Fmoc-CH), 120.0 (Fmoc-CH), 83.1 ($CO_2C(CH_3)_3$), 67.2 (Fmoc-CH_2), 62.9 (β-CH_2 Ser), 54.5 (α-CH), 54.0 (α-CH), 52.6 (α-CH), 47.1 (Fmoc-CH), 34.0 (CH_2), 32.7 (CH_2), 32.0 (CH_2), 31.9 (CH_2), 30.1 (CH_2), 29.7 (CH_2), 29.7 (CH_2), 29.6 (CH_2), 29.6 (CH_2), 29.5 (CH_2), 29.4 (CH_2), 29.3 (CH_2), 28.8 (CH_2), 28.0 ($CO_2C(CH_3)_3$), 22.7 (CH_2), 15.3 (SCH_3 Met), 14.1 (CH_3 Hd); MS (FAB, 3-NBA): *m/z*: 864.5 $[M+Na]^+$, 842.7 $[M+H]^+$, 786.4 $[M-tBu+H]^+$; elemental analysis calcd (%) for $C_{46}H_{71}N_3O_7S_2$: C 65.60, H 8.50, N 4.99; found: C 65.55, H 8.36, N 4.97.

Maleimidocaproyl-(L)-methionyl-(L)-seryl-*S*-hexadecyl-(L)-cysteine *tert*-butyl ester (MIC-Met-Ser-Cys(Hd)-O*t*Bu): Et_2NH (4 mL) was added to a solution of **22b** (203 mg, 0.242 mmol) in dichloromethane (4 mL). After 1.5 h the solvents were evaporated in vacuo, the residue was dissolved in dichloromethane (2 mL), and MIC-OH (56.1 mg, 0.226 mmol) and EEDQ (77.6 mg, 0.314 mmol) were added. After 15 h the solvent was evaporated in vacuo and the residue was dissolved in ethyl acetate (40 mL). The solution was extracted twice with 0.5 N hydrochloric acid (40 mL), saturated $NaHCO_3$ solution (40 mL), and brine (40 mL), dried over $MgSO_4$, and filtered. The solvent was evaporated in vacuo and the product was isolated by flash chromatography (hexane/ethyl acetate 1:3) to yield the product (135 mg, 0.166 mmol, 69%) as a colorless solid. M.p. 88 °C; $[\alpha]_D^{20} = -12.0$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz, TMS): $\delta = 7.12$ (d, $J = 7.7$ Hz, 2H, NH), 6.69 (s, 2H, MIC-CH), 6.40 (d, $J = 7.5$ Hz, 1H, NH), 4.60–4.68 (m, 2H, 2α-CH), 4.49–4.54 (m, 1H, α-CH), 4.54–4.58 (m, 1H, α-CH), 4.06 (dd, $J = 11.4$ Hz, $J = 3.6$ Hz, 1H, β-CH_{2a} Ser), 3.69 (dd, $J = 11.5$ Hz, $J = 5.5$ Hz, 1H, β-CH_{2b} Ser), 3.51 (t, $J = 7.2$ Hz, 2H, MIC-NCH_2), 2.99 (dd, $J =$

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13.7 Hz, $J = 4.7$ Hz, 1H, β-CH_{2a} Cys), 2.91 (dd, $J = 13.6$ Hz, $J = 6.2$ Hz, 1H, β-CH_{2b} Cys), 2.53–2.61 (m, 2H, γ-CH_2 Met), 2.52 (t, $J = 7.4$ Hz, 2H, CH_2 Hd), 2.22 (t, $J = 7.5$ Hz, 2H, MIC), 2.08–2.14 (m, 1H, β-CH_{2a} Met), 2.12 (s, 3H, Met-CH_3), 1.96–2.04 (m, 1H, β-CH_{2b} Met), 1.50–1.70 (m, 6H, CH_2 MIC, CH_2 Hd), 1.48 (s, 9H, $CO_2C(CH_3)_3$), 1.22–1.37 (m, 28H, $CH_3(CH_2)_{13}(CH_2)_2S$, CH_2 MIC), 0.88 (t, $J = 6.9$ Hz, 3H, ω-CH_3 Hd); ^{13}C NMR ($CDCl_3$, 125 MHz, TMS): $\delta = 173.1$ (*C*=O), 171.5 (*C*=O), 170.9 (2*C*=O MIC), 170.1 (*C*=O), 169.7 (*C*=O), 134.1 (2CH MIC), 83.1 ($CO_2C(CH_3)_3$), 62.7 (β-CH_2 Ser), 54.5 (α-CH), 52.5 (α-CH), 52.5 (α-CH), 37.6 (CH_2), 36.2 (CH_2), 34.0 (CH_2), 32.6 (CH_2), 31.9 (CH_2), 31.3 (CH_2), 30.3 (CH_2), 29.7 (CH_2), 29.7 (CH_2), 29.6 (CH_2), 29.5 (CH_2), 29.4 (CH_2), 29.2 (CH_2), 28.8 (CH_2), 28.2 (CH_2), 28.0 ($CO_2C(CH_3)_3$), 26.3 (CH_2), 24.9 (CH_2), 22.7 (CH_2), 15.3 (SCH_3 Met), 14.1 (CH_3 Hd); MS (FAB, 3-NBA): *m/z*: 835.5 $[M+Na]^+$, 813.5 $[M+H]^+$, 757.4 $[M-t\text{Bu}+H]^+$; HRMS (FAB) calcd for $C_{41}H_{72}N_4NaO_8S_2$ ($[M+Na]^+$): 835.4689; found: 835.4727.

Maleimidocaproyl-(L)-methionyl-(L)-seryl-*S*-hexadecyl-(L)-cysteine (MIC-Met-Ser-Cys(Hd)-OH) (15b): TFA (0.25 mL) was added to a solution of MIC-Met-Ser-Cys(Hd)-O*t*Bu (61.0 mg, 75.0 μmol) in dichloromethane (0.25 mL). After 3 h, toluene (4 mL) was added and the solvent was evaporated in vacuo. Addition of toluene and evaporation were repeated twice to yield **15b** in quantitative yield as a colorless solid. M.p. 141 °C; $[\alpha]_D^{20} = -9.6$ ($c = 1.0$, $CHCl_3$/MeOH 2:1); ^{1}H NMR ($CDCl_3/CD_3OD$ 6:1, 500 MHz, TMS): $\delta = 6.72$ (s, 2H, MIC-CH), 4.48–4.70 (m, 3H, 3 α-CH), 3.88–3.93 (m, 1H, β-CH_{2a} Ser), 3.73–3.78 (m, 1H, β-CH_{2b} Ser), 3.52 (t, $J = 7.2$ Hz, 2H, MIC-NCH_2), 3.04 (dd, $J = 13.8$ Hz, $J = 4.4$ Hz, 1H, β-CH_{2a} Cys), 2.95 (dd, $J = 13.9$ Hz, $J = 6.8$ Hz, 1H, β-CH_{2b} Cys), 2.49–2.58 (m, 4H, γ-CH_2 Met, CH_2 Hd), 2.24 (t, $J = 7.3$ Hz, 2H, MIC), 2.06–2.16 (m, 1H, β-CH_{2a} Met), 2.11 (s, 3H, Met-CH_3), 1.90–1.98 (m, 1H, β-CH_{2b} Met), 1.53–1.69 (m, 6H, CH_2 MIC, CH_2 Hd), 1.22–1.41 (m, 28H, $CH_3(CH_2)_{13}(CH_2)_2S$, CH_2 MIC), 0.88 (t, $J = 6.8$ Hz, 3H, ω-CH_3 Hd); ^{13}C NMR ($CDCl_3/CD_3OD$ 6:1, 125 MHz, TMS): $\delta = 174.4$ (*C*=O), 172.7 (*C*=O), 172.4 (*C*=O), 171.3 (2*C*=O MIC), 170.6 (*C*=O), 134.3 (2CH MIC), 62.4 (β-CH_2 Ser), 55.0 (α-CH), 52.6 (α-CH), 52.3 (α-CH), 37.8 (CH_2), 36.0 (CH_2), 33.7 (CH_2), 32.6 (CH_2), 32.0 (CH_2), 31.6 (CH_2), 30.3 (CH_2), 29.8 (CH_2), 29.8 (CH_2), 29.7 (CH_2), 29.7 (CH_2), 29.5 (CH_2), 29.4 (CH_2), 29.0 (CH_2), 28.3 (CH_2), 26.4 (CH_2), 25.1 (CH_2), 22.8 (CH_2), 15.3 (SCH_3 Met), 14.2 (CH_3 Hd); MS (FAB, 3-NBA): *m/z*: 779.5 $[M+Na]^+$, 757.4 $[M+H]^+$; HRMS (FAB) calcd for $C_{37}H_{64}N_4NaO_8S_2$ ($[M+Na]^+$): 779.4063; found: 779.3996.

Maleimidocaproyl-(L)-methionyl-(L)-seryl-*S*-hexadecyl-(L)-cysteyl-$N^{(\varepsilon)}$-(4-tolyldiphenylmethyl)-(L)-lysyl-*S*-farnesyl-(L)-cysteine methyl ester (MIC-Met-Ser-Cys(Hd)-Lys(Mtt)-Cys(Far)-OMe) (19b): Et_2NH (0.4 mL) was added to a solution of **7** (90.0 mg, 95.1 μmol) in dichloromethane (1.6 mL). After 4 h the solvent was evaporated in vacuo, the residue was dissolved in a mixture of dichloromethane (5 mL) and trifluoroethanol (0.1 mL), and **15b** (55.9 mg, 73.8 μmol) and EEDQ (23.7 mg, 95.9 μmol) were added. After 15 h the solvent was evaporated in vacuo and the product was isolated by flash chromatography (dichloromethane/ethanol 30:1) to yield **19b** (28.3 mg, 19.3 μmol, 26%) as a colorless, waxy solid. M.p. 175 °C; $[\alpha]_D^{20} = -21.2$ ($c = 1.0$, $CHCl_3$); ^{1}H NMR ($CDCl_3/CD_3OD$ 6:1, 500 MHz, TMS): $\delta = 7.42$ (d, $J = 7.6$ Hz, 4H, Mtt-CH), 7.22–7.33 (m, 2H, Mtt-CH), 7.24–7.34 (m, 6H, Mtt-CH), 7.13–7.20 (m, 2H, Mtt-CH), 7.05–7.09 (m, 2H, Mtt-CH), 6.71 (s, 1H, MIC-CH), 6.70 (s, 1H, MIC-CH), 5.15–5.23 (m, 1H, CH=C Far), 5.05–5.12 (m, 2H, CH=C Far), 4.34–4.64 (m, 5H, 5 α-CH), 3.88–3.95 (m, 1H, β-CH_{2a} Ser), 3.69–3.78 (m, 1H, β-CH_{2b} Ser), 3.69 (s, 3H, $COOCH_3$), 3.47–3.55 (m, 2H, MIC-NCH_2), 3.21 (dd, $J = 13.1$ Hz, $J = 8.2$ Hz, 1H, CH_{2a} Far), 3.09 (dd, $J = 13.1$ Hz, $J = 7.4$ Hz, 1H, CH_{2b} Far), 2.90–2.98 (m, 4H, β-CH_2 Cys_{Hd}, β-CH_{2a} Cys_{Far}), 2.79 (dd, $J = 13.8$ Hz, $J = 7.8$ Hz, 1H, β-CH_{2b} Cys_{Far}), 2.45–2.58 (m, 6H, γ-CH_2 Met, CH_2 Hd, ε-CH_2 Lys), 2.30 (b, 3H, Mtt-CH_3), 2.18–2.26 (m, 2H, MIC), 1.82–2.15 (m, 14H, β-CH_2 Met, SCH_3 Met, CH_{2a} Lys, CH_2 Far), 1.48–1.69 (m, 11H, CH_2 MIC, CH_2 Lys, CH_2 Hd), 1.68 (s, 3H, CH_3 Far), 1.67 (s, 3H, CH_3 Far), 1.60 (s, 6H, CH_3 Far), 1.19–1.39 (m, 28H, $CH_3(CH_2)_{13}(CH_2)_2S$, CH_2 MIC), 0.88 (t, $J = 6.8$ Hz, 3H, ω-CH_3 Hd); ^{13}C NMR ($CDCl_3/CD_3OD$ 6:1, 125 MHz, TMS): $\delta = 174.3$ (*C*=O), 172.2 (*C*=O), 172.1 (*C*=O), 171.2 (2*C*=O MIC), 170.6 (*C*=O), 176.9 (*C*=O), 169.4 (*C*=O), 146.4 (quart, Mtt), 143.3 (quart, Mtt), 140.1 (quart, Far), 135.8 (quart, Mtt), 135.4 (quart, Far), 134.2 (2CH MIC), 131.4 (quart, Far), 128.7 (Mtt, 3CH), 127.8 (Mtt, CH), 126.2 (Mtt, CH), 124.4 (Far-CH), 123.9 (Far-CH), 119.6 (Far-CH), 70.7 (quart, Mtt), 62.1 (β-CH_2 Ser), 54.9 (α-CH), 53.6 (α-CH), 53.0 (α-CH), 52.8 (α-CH), 52.5 ($COOCH_3$), 52.1 (α-CH), 43.6 (CH_2), 39.8 (CH_2), 37.7 (CH_2), 35.9 (CH_2), 33.9 (CH_2), 33.5 (CH_2), 32.8 (CH_2), 32.6 (CH_2), 32.4 (CH_2), 32.0 (CH_2), 31.3 (CH_2), 30.6 (CH_2), 30.3 (CH_2), 29.8 (CH_2), 29.7 (CH_2), 29.7 (CH_2), 29.4 (CH_2), 29.0 (CH_2), 28.3 (CH_2), 26.8 (CH_2), 26.6 (CH_2), 26.4 (CH_2), 26.3 (CH_2), 25.7 (Far-CH_3), 25.0 (CH_2), 22.8 (CH_2), 20.9 (Mtt-CH_3), 17.7 (Far-CH_3), 16.2 (Far-CH_3), 16.0 (Far-CH_3), 15.4 (SCH_3 Met), 14.1 (CH_3 Hd); MS (FAB, 3-NBA): *m/z*: 1484.6 $[M+Na]^+$, 1462.4 $[M+H]^+$, 1460.4 $[M-H]^+$, 1384.5 $[M-C_6H_5-H]^+$, 1371.5 $[M-C_6H_4CH_3]^+$, 1206.6 $[M-\text{Mtt}+H]^+$, 257.2 $[\text{Mtt}]^+$.

Maleimidocaproyl-(L)-methionyl-(L)-seryl-*S*-hexadecyl-(L)-cysteyl-(L)-lysyl-*S*-farnesyl-(L)-cysteine methyl ester (MIC-Met-Ser-Cys(Hd)-Lys-Cys-(Far)-OMe) (1b): Et_3SiH (16.2 μL, 102 μmol) was added to a solution of **19b** (14.9 mg, 10.2 μmol) in a 1% solution of TFA in dichloromethane (0.8 mL). After 50 min a solution of 1% $EtMe_2N$ in dichloromethane (0.9 mL) was added, the solvent was evaporated in vacuo, and the product was isolated by size-exclusion chromatography (Sephadex LH-20, $CHCl_3$/MeOH 1:1) to yield **1b** (11.9 mg, 9.86 μmol, 97%) as a colorless, waxy solid. M.p. 193 °C; $[\alpha]_D^{20} = -39.0$ ($c = 0.3$, $CHCl_3$/MeOH 2:1); ^{1}H NMR ($CDCl_3/CD_3OD$ 6:1, 500 MHz, TMS): $\delta = 6.74$ (s, 2H, MIC-CH), 5.20 (t, $J = 7.3$ Hz, 1H, CH=C Far), 5.06–5.13 (m, 2H, CH=C Far), 4.63 (dd, $J = 8.2$ Hz, $J = 5.0$ Hz, 1H, α-CH), 4.42–4.48 (m, 3H, 3 α-CH), 4.38 (t, $J = 4.6$ Hz, 1H, α-CH), 3.92 (dd, $J = 11.5$ Hz, $J = 4.4$ Hz, 1H, β-CH_{2a} Ser), 3.79 (dd, $J = 11.5$ Hz, $J = 4.9$ Hz, 1H, β-CH_{2b} Ser), 3.75 (s, 3H, $COOCH_3$), 3.52 (t, $J = 7.2$ Hz, 2H, MIC-NCH_2), 3.24 (dd, $J = 13.1$ Hz, $J = 8.3$ Hz, 1H, CH_{2a} Far), 3.12 (dd, $J = 13.2$ Hz, $J = 7.2$ Hz, 1H, CH_{2b} Far), 3.00 (dd, $J = 13.6$ Hz, $J = 6.5$ Hz, 1H, β-CH_{2a} Cys_{Hd}), 2.93–2.99 (m, 3H, β-CH_{2a} Cys_{Far}, ε-CH Lys), 2.89 (dd, $J = 13.6$ Hz, $J = 7.9$ Hz, 1H, β-CH_{2b} Cys_{Hd}), 2.80 (dd, $J = 13.9$ Hz, $J = 8.2$ Hz, 1H, β-CH_{2b} Cys_{Far}), 2.51–2.61 (m, 4H, γ-CH_2 Met, CH_2 Hd), 2.29 (t, 2H, $J = 7.0$ Hz, MIC), 1.94–2.15 (m, 14H, β-CH_2 Met, SCH_3 Met, CH_{2a} Lys, CH_2 Far), 1.54–1.78 (m, 9H, CH_2 Hd, CH_2 MIC, CH_2 Lys), 1.68 (s, 6H, CH_3 Far), 1.61 (s, 6H, CH_3 Far), 1.41–1.50 (m, 2H, CH_2 Lys), 1.21–1.39 (m, 28H, $CH_3(CH_2)_{13}(CH_2)_2S$, CH_2 MIC), 0.88 (t, $J = 7.0$ Hz, 3H, ω-CH_3 hexadecyl); ^{13}C NMR ($CDCl_3/CD_3OD$ 6:1, 125 MHz, TMS): $\delta = 175.3$ (*C*=O), 173.0 (*C*=O), 172.3 (*C*=O), 171.7 (*C*=O), 171.4 (*C*=O), 171.4 (*C*=O), 140.3 (quart, Far), 135.6 (quart, Far), 134.4 (2CH MIC), 131.5 (quart, Far), 124.5 (Far-CH), 124.0 (Far-CH), 119.8 (Far-CH), 62.0 (β-CH_2 Ser), 55.8 (α-CH), 53.9 (α-CH), 53.3 (α-CH), 52.9 (α-CH), 52.7 ($COOCH_3$), 52.4 (α-CH), 39.9 (CH_2), 39.7 (CH_2), 37.8 (CH_2), 36.0 (CH_2), 33.0 (CH_2), 32.8 (CH_2), 32.5 (CH_2), 32.1 (CH_2), 31.0 (CH_2), 30.7 (CH_2), 30.5 (CH_2), 29.9 (CH_2), 29.9 (CH_2), 29.8 (CH_2), 29.7 (CH_2), 29.6 (CH_2), 29.5 (CH_2), 29.1 (CH_2), 28.4 (CH_2), 26.9 (CH_2), 26.7 (CH_2), 26.5 (CH_2), 26.5 (CH_2), 25.8 (Far-CH_3), 25.3 (CH_2), 22.9 (CH_2), 22.2 (CH_2), 17.8 (Far-CH_3), 16.2 (Far-CH_3), 16.1 (Far-CH_3), 15.3 (SCH_3 Met), 14.2 (CH_3 Hd); MS (FAB, 3-NBA): *m/z*: 1206.5 $[M+H]^+$.

***N*-Fluorenylmethoxycarbonyl-(L)-methionyl-(L)-seryl-*S*-*tert*-butylthio-(L)-cysteine *tert*-butyl ester (Fmoc-Met-Ser-Cys(*t*Bu)-O*t*Bu) (22c)**: EEDQ (488 mg, 1.97 mmol) was added to a solution of Fmoc-Met-Ser-OH (**20**; 835 mg, 1.82 mmol) and H-Cys(*t*Bu)-O*t*Bu (**21c**; 403 mg, 1.82 mmol) in DMF (8 mL). After 15 h ethyl acetate (50 mL) was added and the solution was extracted twice with 0.5N hydrochloric acid (50 mL), saturated $NaHCO_3$ solution (50 mL), and brine (50 mL), dried over $MgSO_4$, and filtered. The solvent was evaporated in vacuo and the product was isolated by flash chromatography (hexane/ethyl acetate 2:1) to yield **22c** (618 mg, 0.875 mmol, 58%) as a colorless solid. M.p. 133 °C; $[\alpha]_D^{20} = -49.8$ ($c = 1.1$, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz, TMS): $\delta = 7.75$ (d, $J = 7.5$ Hz, 2H, Fmoc-CH), 7.58 (dd, $J = 7.4$ Hz, $J = 3.7$ Hz, 2H, Fmoc-CH), 7.39 (t, $J = 7.5$ Hz, 2H, Fmoc-CH), 7.27–7.34 (m, 3H, $J = 7.4$ Hz, 2 Fmoc-CH, 1 NH), 7.24 (d, $J = 6.9$ Hz, 1H, NH), 5.50 (d, $J = 7.7$ Hz, 1H, NH), 4.72–4.79 (m, 1H, α-CH), 4.57–4.63 (m, 1H, α-CH), 4.35–4.47 (m, 3H, α-CH, Fmoc-CH_2), 4.21 (t, $J = 6.9$ Hz, 1H, Fmoc-CH), 4.06 (dd, $J = 11.3$ Hz, $J = 3.0$ Hz, 1H, β-CH_{2a} Ser), 3.70 (dd, $J = 11.0$ Hz, $J = 5.1$ Hz, 1H, β-CH_{2b} Ser), 3.22 (dd, $J = 13.6$ Hz, $J = 4.1$ Hz, 1H, β-CH_{2a} Cys), 3.10 (dd, $J = 13.6$ Hz, $J = 6.1$ Hz, 1H, β-CH_{2b} Cys), 2.52–2.65 (m, 2H, γ-CH_2 Met), 2.07–2.18 (m, 1H, β-CH_{2a} Met), including 2.10 (s, 3H, Met-CH_3), 1.95–2.05 (m, 1H, β-CH_{2b} Met), 1.47 (s, 9H, $CO_2C(CH_3)_3$), 1.30 (s, 9H, $SC(CH_3)_3$); ^{13}C NMR ($CDCl_3$, 125 MHz, TMS): $\delta = 171.6$ (*C*=O), 170.2 (*C*=O), 169.2 (*C*=O), 156.2 (*C*=O), 143.9 (quart, arom, Fmoc), 143.7 (quart, arom, Fmoc), 141.3 (quart, arom, Fmoc), 127.7 (arom, Fmoc-CH), 127.1 (arom, Fmoc-CH), 125.1 (arom, Fmoc-CH), 120.0 (arom, Fmoc-CH), 83.3 ($CO_2C(CH_3)_3$), 67.2 (Fmoc-CH_2), 62.7 (β-CH_2 Ser), 54.3 (α-CH), 54.0 (α-CH), 53.1 (α-CH), 48.4 ($SC(CH_3)_3$), 47.1 (Fmoc-CH), 42.2 (β-CH_2 Cys), 32.0 (CH_2), 30.1 (CH_2), 29.8 ($SC(CH_3)_3$), 27.9 ($CO_2C(CH_3)_3$), 15.3 (SCH_3 Met); MS (FAB, 3-NBA): *m/z*: 737.1 $[M+K]^+$, 728.2 $[M+Na]^+$, 706.3 $[M+H]^+$, 650.2 $[M-t\text{Bu}+H]^+$,

593.1 $[M-2t\text{Bu}+\text{H}]^+$; elemental analysis calcd (%) for $C_{34}H_{47}N_3O_7S_3$: C 57.85, H 6.71, N 5.95; found: C 58.13, H 6.77, N 6.11.

Maleimidocaproyl-(L)-methionyl-(L)-seryl-*S*-*tert*-butylthio-(L)-cysteine *tert*-butyl ester (MIC-Met-Ser-Cys(*t*Bu)-O*t*Bu): Et_2NH (3 mL) was added to a solution of **22c** (100 mg, 0.142 mmol) in dichloromethane (3 mL). After 5 h the solvent was evaporated in vacuo, the residue was dissolved in dichloromethane (2 mL), and MIC-OH (32.9 mg, 0.156 mmol) and EEDQ (45.5 mg, 0.184 mmol) were added. After 18 h dichloromethane (40 mL) was added, the solution was extracted twice with 0.5 N hydrochloric acid (40 mL), saturated $NaHCO_3$ solution (40 mL), and brine (40 mL), dried over $MgSO_4$, and filtered. The solvent was evaporated in vacuo and the product was isolated by flash chromatography (dichloromethane/ethanol 30:1, then 20:1) to yield the product (55.8 mg, 82.4 mmol, 58%) as a colorless solid. M.p. 98 °C; $[\alpha]_D^{20} = -41.7$ ($c = 0.8$, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz, TMS): $\delta = 7.29$ (d, $J = 7.8$ Hz, 1H, NH), 7.23 (d, $J = 7.2$ Hz, 1H, NH), 6.69 (s, 2H, MIC-CH), 6.52 (d, $J = 7.3$ Hz, 1H, NH), 4.72–4.77 (m, 1H, α-CH), 4.62–4.68 (m, 1H, α-CH), 4.54–4.58 (m, 1H, α-CH), 4.06 (dd, $J = 11.5$ Hz, $J = 3.9$ Hz, 1H, β-CH_{2a} Ser), 3.71 (dd, $J = 11.5$ Hz, $J = 5.4$ Hz, 1H, β-CH_{2b} Ser), 3.51 (t, $J = 7.2$ Hz, 2H, MIC-NCH_2), 3.23 (dd, $J = 13.7$ Hz, $J = 4.4$ Hz, 1H, β-CH_{2a} Cys), 3.12 (dd, $J = 13.6$ Hz, $J = 6.2$ Hz, 1H, β-CH_{2b} Cys), 2.53–2.65 (m, 2H, γ-CH_2 Met), 2.23 (t, $J = 7.5$ Hz, 2H, MIC), 2.09–2.18 (m, 1H, β-CH_{2a} Met), 2.12 (s, 3H, Met-CH_3), 1.96–2.05 (m, 1H, β-CH_{2b} Met), 1.64–1.70 (m, 2H, CH_2 MIC), 1.57–1.63 (m, 2H, CH_2 MIC), 1.49 (s, 9H, $CO_2C(CH_3)_3$), 1.28–1.36 (m, 2H, CH_2 MIC), 1.32 (s, 9H, $SC(CH_3)_3$), ^{13}C NMR ($CDCl_3$, 125 MHz, TMS): $\delta = 173.2$ (*C*=O), 171.7 (*C*=O), 170.9 (2 *C*=O MIC), 170.1 (*C*=O), 169.2 (*C*=O), 134.1 (2 CH MIC), 83.3 ($CO_2C(CH_3)_3$), 62.6 (β-CH_2 Ser), 54.5 (α-CH), 53.1 (α-CH), 52.5 (α-CH), 48.4 ($SC(CH_3)_3$), 42.2 (β-CH_2 Cys), 37.6 (CH_2), 36.1 (CH_2), 31.5 (CH_2), 30.3 (CH_2), 29.8 ($SC(CH_3)_3$), 29.7 (CH_2), 28.2 (CH_2), 28.0 ($CO_2C(CH_3)_3$), 26.3 (CH_2), 24.9 (CH_2), 15.3 (SCH_3 Met); MS (FAB, 3-NBA): *m/z*: 699.3 $[M+\text{Na}]^+$, 677.3 $[M+\text{H}]^+$, 621.3 $[M-t\text{Bu}+\text{H}]^+$, 564.2 $[M-2t\text{Bu}]^+$; HRMS (FAB) calcd for $C_{29}H_{49}N_4O_8S_3$: 677.2713; found: 677.2728.

Maleimidocaproyl-(L)-methionyl-(L)-seryl-*S*-*tert*-butylthio-(L)-cysteine (MIC-Met-Ser-Cys(*t*Bu)-OH) (15c): TFA (0.25 mL) was added to a solution of MIC-Met-Ser-Cys(*t*Bu)-O*t*Bu (15.1 mg, 22.3 μmol) in dichloromethane (0.25 mL). After 3 h, toluene (1 mL) was added and the solvent was evaporated in vacuo. Addition of toluene and evaporation were repeated twice to yield **15c** in quantitative yield as a colorless solid. M.p. 123 °C; $[\alpha]_D^{20} = -48.6$ ($c = 0.7$, $CHCl_3$/MeOH 2:1); ^{1}H NMR ($CDCl_3$/CD_3OD 6:1, 500 MHz, TMS): $\delta = 7.78$ (d, $J = 7.6$ Hz, 1H, NH), 7.71 (d, $J = 7.6$ Hz, 1H, NH), 7.45 (d, $J = 7.7$ Hz, 1H, NH), 6.73 (s, 2H, MIC-CH), 4.73–4.79 (m, 1H, α-CH), 4.47–4.55 (m, 2H, 2α-CH), 3.89 (dd, $J = 11.5$ Hz, $J = 4.8$ Hz, 1H, β-CH_{2a} Ser), 3.75 (dd, $J = 11.5$ Hz, $J = 5.3$ Hz, 1H, β-CH_{2b} Ser), 3.52 (t, $J = 7.2$ Hz, 2H, MIC-NCH_2), 3.26 (dd, $J = 13.7$ Hz, $J = 4.4$ Hz, 1H, β-CH_{2a} Cys), 3.11 (dd, $J = 13.7$ Hz, $J = 7.6$ Hz, 1H, β-CH_{2b} Cys), 2.52–2.58 (m, 2H, γ-CH_2 Met), 2.24 (t, $J = 7.5$ Hz, 2H, MIC), 2.07–2.16 (m, 1H, β-CH_{2a} Met), 2.12 (s, 3H, Met-CH_3), 1.90–1.99 (m, 1H, β-CH_{2b} Met), 1.56–1.69 (m, 4H, CH_2 MIC), 1.26–1.35 (m, 2H, CH_2 MIC), 1.33 (s, 9H, $SC(CH_3)_3$); ^{13}C NMR ($CDCl_3$/CD_3OD 6:1, 125 MHz, TMS): $\delta = 174.5$ (*C*=O), 172.4 (*C*=O), 171.3 (2 *C*=O MIC), 170.6 (*C*=O), 134.3 (2 CH MIC), 62.2 (β-CH_2 Ser), 55.0 (α-CH), 52.7 (α-CH), 52.3 (α-CH), 48.3 ($SC(CH_3)_3$), 41.7 (β-CH_2 Cys), 37.8 (CH_2), 36.0 (CH_2), 31.5 (CH_2), 30.3 (CH_2), 29.9 ($SC(CH_3)_3$), 28.4 (CH_2), 26.4 (CH_2), 25.2 (CH_2), 15.3 (SCH_3 Met); MS (FAB, 3-NBA): *m/z*: 659.3 $[M+\text{K}]^+$, 643.2 $[M+\text{Na}]^+$, 621.2 $[M+\text{H}]^+$; HRMS (FAB) calcd for $C_{25}H_{41}N_4O_8S_3$: 621.2087; found: 621.2094.

Maleimidocaproyl-(L)-methionyl-(L)-seryl-*S*-*tert*-butylthio-(L)-cysteyl-$N^{(\varepsilon)}$-(4-tolyldiphenylmethyl)-(L)-lysyl-*S*-farnesyl-(L)-cysteine methyl ester (MIC-Met-Ser-Cys(*t*Bu)-Lys(Mtt)-Cys(Far)-OMe) (19c): Et_2NH (0.4 mL) was added to a solution of **7** (85.6 mg, 90.5 μmol) in dichloromethane (1.6 mL). After 4 h the solvent was evaporated in vacuo, the residue was dissolved in a mixture of dichloromethane (2 mL) and trifluoroethanol (0.1 mL), and **15c** (47.1 mg, 69.6 μmol) and EEDQ (24.2 mg, 90.5 μmol) were added. After 13 h the solvent was evaporated in vacuo and the product was isolated by flash chromatography (dichloromethane/ethanol 30:1) to yield **19c** (25.9 mg, 19.5 μmol, 28%) as a colorless, waxy solid. M.p. 155 °C; $[\alpha]_D^{20} = -47.2$ ($c = 0.5$, $CHCl_3$); ^{1}H NMR ($CDCl_3$, 500 MHz, TMS): $\delta = 7.43$ (d, $J = 7.8$ Hz, 4H, Mtt-CH), 7.30 (d, $J = 8.2$ Hz, 2H, Mtt-CH), 7.01–7.26 (m, 8H, Mtt-CH), 6.68 (s, 1H, MIC-CH), 6.66 (s, 1H, MIC-CH), 5.16–5.23 (m, 1H, CH=C Far), 5.03–5.11 (m, 2H, CH=C Far), 4.40–4.75 (m, 5H, 5 α-CH), 3.91–3.99 (m, 1H, β-CH_{2a} Ser), 3.60–3.80 (m, 4H, β-CH_{2b} Ser, $COOCH_3$), 3.44–3.52 (m, 2H, MIC-NCH_2), 3.05–3.22 (m, 4H, β-CH_2 Cys_{StBu}, CH_2 Far), 2.94 (dd, $J = 14.8$ Hz, $J = 4.9$ Hz, 1H, β-CH_{2a} Cys_{Far}), 2.84 (dd, $J = 14.1$ Hz, $J = 7.2$ Hz, 1H, β-CH_{2b} Cys_{Far}), 2.48–2.62 (m, 4H, γ-CH_2 Met, ε-CH_2 Lys), 2.29 (b, 3H, Mtt-CH_3), 2.19–2.28 (m, 2H, MIC), 1.82–2.15 (m, 14H, β-CH_2 Met, SCH_3 Met, CH_{2a} Lys, CH_2 Far), 1.45–1.72 (m, 9H, CH_2 MIC, CH_2 Lys), 1.67 (s, 3H, CH_3 Far), 1.65 (s, 3H, CH_3 Far), 1.59 (s, 6H, CH_3 Far), 1.24–1.41 (m, 11H, $SC(CH_3)_3$, CH_2 MIC); ^{13}C NMR ($CDCl_3$, 125 MHz, TMS): $\delta = 173.4$ (*C*=O), 171.1 (*C*=O), 170.9 (*C*=O), 170.5 (*C*=O), 146.5 (quart, Mtt), 143.2 (quart, Mtt), 140.1 (quart, Far), 135.5 (quart, Mtt), 135.3 (quart, Far), 134.1 (2 CH MIC), 131.3 (quart, Far), 128.6 (Mtt, 3 CH), 127.8 (Mtt, CH), 126.2 (Mtt, CH), 124.3 (Far-CH), 123.8 (Far-CH), 119.5 (Far-CH), 70.6 (quart, Mtt), 62.3 (β-CH_2 Ser), 54.3 (α-CH), 53.5 (α-CH), 52.8 (α-CH), 52.5 ($COOCH_3$), 51.8 (α-CH), 48.6 (CH_2), 48.4 ($SC(CH_3)_3$), 41.0 (CH_2), 39.7 (CH_2), 37.6 (CH_2), 36.2 (CH_2), 36.1 (CH_2), 32.8 (CH_2), 31.0 (CH_2), 30.9 (CH_2), 30.3 (CH_2), 29.8 ($SC(CH_3)_3$), 29.7 (CH_2), 28.3 (CH_2), 28.2 (CH_2), 26.7 (CH_2), 26.5 (CH_2), 26.4 (CH_2), 26.3 (CH_2), 25.7 (Far-CH_3), 24.9 (CH_2), 21.0 (Mtt-CH_3), 17.7 (Far-CH_3), 16.2 (Far-CH_3), 16.0 (Far-CH_3), 15.4 (SCH_3 Met); MS (FAB, 3-NBA): *m/z*: 1348.5 $[M+\text{Na}]^+$, 1326.5 $[M+\text{H}]^+$, 1248.4 $[M-C_6H_5]^+$, 1334.5 $[M-C_6H_4CH_3]^+$, 1070.5 $[M-\text{Mtt}+\text{H}]^+$, 257.1 $[\text{Mtt}]^+$.

Maleimidocaproyl-(L)-methionyl-(L)-seryl-*S*-*tert*-butylthio-(L)-cysteyl-(L)-lysyl-*S*-farnesyl-(L)-cysteine methyl ester (MIC-Met-Ser-Cys(*t*Bu)-Lys-Cys-(Far)-OMe) (1c): Et_3SiH (11.4 μL, 71.6 μmol) was added to a solution of **19c** (9.5 mg, 7.2 μmol) in a 1% solution of TFA in dichloromethane (0.8 mL). After 60 min a solution of 1% $EtMe_2N$ in dichloromethane (0.9 mL) was added, the solvent was evaporated in vacuo and the product was isolated by size-exclusion chromatography (Sephadex LH-20, $CHCl_3$/MeOH 1:1) to yield **1c** (5.8 mg, 5.4 μmol, 76%) as a colorless, waxy solid. M.p. 155 °C; $[\alpha]_D^{20} = -59.0$ ($c = 0.2$, $CHCl_3$/MeOH 2:1); ^{1}H NMR ($CDCl_3$/CD_3OD 6:1, 500 MHz, TMS): $\delta = 6.74$ (s, 2H, MIC-CH), 5.21 (t, $J = 7.3$ Hz, 1H, CH=C Far), 5.08–5.14 (m, 2H, CH=C Far), 4.63 (dd, $J = 8.1$ Hz, $J = 5.0$ Hz, 1H, α-CH), 4.60 (dd, $J = 8.5$ Hz, $J = 5.7$ Hz, 1H, α-CH), 4.42–4.56 (m, 2H, 2 α-CH), 4.38 (t, $J = 4.7$ Hz, 1H, α-CH), 3.92 (dd, $J = 11.4$ Hz, $J = 4.7$ Hz, 1H, β-CH_{2a} Ser), 3.81 (dd, $J = 11.4$ Hz, $J = 5.0$ Hz, 1H, β-CH_{2b} Ser), 3.76 (s, 3H, $COOCH_3$), 3.52 (t, $J = 7.2$ Hz, 2H, 2 MIC-NCH_2), 3.18–3.26 (m, 2H, CH_{2a} Far, β-CH_{2a} Cys_{StBu}), 3.12 (dd, $J = 13.5$ Hz, $J = 7.5$ Hz, 1H, CH_{2b} Far), 3.08 (dd, $J = 13.6$ Hz, $J = 8.5$ Hz, 1H, β-CH_{2b} Cys_{StBu}), 2.93–2.99 (m, 3H, β-CH_{2a} Cys_{Far}, ε-CH_2 Lys), 2.80 (dd, $J = 13.9$ Hz, $J = 8.2$ Hz, 1H, β-CH_{2b} Cys_{Far}), 2.52–2.61 (m, 2H, γ-CH_2 Met), 2.30 (td, $J = 7.5$ Hz, $J = 2.3$ Hz, 2H, MIC), 1.92–2.16 (m, 14H, β-CH_2 Met, SCH_3 Met, CH_{2a} Lys, CH_2 Far), 1.59–1.79 (m, 7H, CH_2 MIC, CH_2 Lys), 1.68 (s, 6H, CH_3 Far), 1.61 (s, 6H, CH_3 Far), 1.41–1.50 (m, 2H, CH_2 Lys), 1.30–1.39 (m, 11H, $SC(CH_3)_3$, CH_2 MIC); ^{13}C NMR ($CDCl_3$/CD_3OD 6:1, 125 MHz, TMS): $\delta = 175.4$ (*C*=O), 173.1 (*C*=O), 172.2 (*C*=O), 171.5 (*C*=O), 171.4 (*C*=O MIC), 171.3 (*C*=O), 140.3 (quart, Far), 135.6 (quart, Far), 134.4 (2 CH MIC), 131.5 (quart, Far), 124.5 (Far-CH), 124.0 (Far-CH), 119.8 (Far-CH), 61.9 (β-CH_2 Ser), 55.9 (α-CH), 54.0 (α-CH), 53.4 (α-CH), 53.0 (α-CH), 52.7 ($COOCH_3$), 52.4 (α-CH), 40.7 (CH_2), 39.9 (CH_2), 39.7 (CH_2), 37.8 (CH_2), 36.0 (CH_2), 32.7 (CH_2), 31.0 (CH_2), 30.8 (CH_2), 30.5 (CH_2), 30.0 ($SC(CH_3)_3$), 29.7 (CH_2), 28.5 (CH_2), 26.9 (CH_2), 26.7 (CH_2), 26.5 (CH_2), 25.8 (Far-CH_3), 25.3 (CH_2), 22.2 (CH_2), 17.8 (Far-CH_3), 16.2 (Far-CH_3), 16.1 (Far-CH_3), 15.4 (SCH_3 Met); MS (FAB, 3-NBA): *m/z*: 1070.5 $[M+\text{H}]^+$; HRMS (FAB) calcd for $C_{50}H_{83}N_7O_{10}S_4$: 1070.516; found: 1070.519.

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