SYNTHESIS OF NOVEL THIAZOLE-CONTAINING BIS -1-CHLOROMETHYL- 5-HYDROXY-1,2-DIHYDRO-3*H*-BENZ[*e*]INDOLE (*seco*-CBI)-POLYAMIDE CONJUGATES AS ANTICANCER AGENTS

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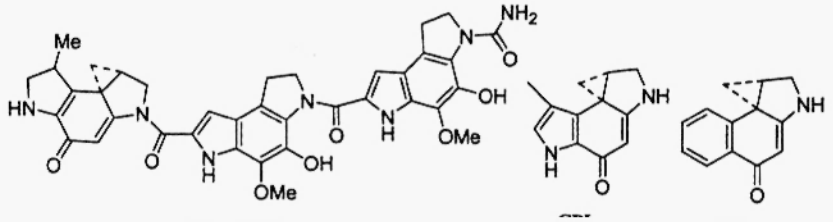
Abstract: The synthesis of novel thiazole-containing bis-1-chloromethyl-5-hydroxy-1,2-dihydro-3*H*-benz[*e*]indole (*seco*-CBI)-polyamide conjugates is described.

Introduction

Certain non-peptide low molecular weight ligands exhibit intermolecular recognition by interacting with double stranded DNA via three different modes: intercalation and minor or major groove binding. Some of these processes can be further classified into covalent bonding, non-covalent binding or electrostatic attraction. Some non-protein molecules that are known to bind to double-stranded DNA include the naturally found antitumor antibiotic CC-1065. The interaction of relatively small molecules of this type with DNA may lead to useful applications in gene control as well as anticancer, antiviral and antibacterial agents.

Extensive research has been done on the highly potent CC-1065[1-6] and its analogues derived from[7-8] 1,2,8,8a-tetrahydro-7-methylcyclopropa[*c*]pyrrolo[3,2-*e*]indole-4-one (CPI) and 1,2,9,9a - tetrahydrocyclopropa[*c*]benz[*e*]indole-4-one (CBI). We have to linked CPI[9, 10] and CBI[11] with polyamides and found that some optimized CPI-polyamide conjugates exhibit up to 10000 times higher potency than CC-1065 against KB human cancer cells.[10] Studies also have shown that some synthetic compounds, which contain two CPI moieties linked from two possible positions by a flexible methylene chain of variable length, are significantly more potent than CC-1065 both in *vitro* and in *vivo.*[12] We also reported the synthesis and biological evaluation of *seco*-CBI dimers against nine types of cancer cells. Certain examples showed significant activity against CCRT-CEM, HL-60 (TB), MOLT-4, leukemia, CNS cancer, melanoma, and prostate cancer cell lines with Gl 50 values < 0.01μm.[13]



CC - 1065 CPI CBI

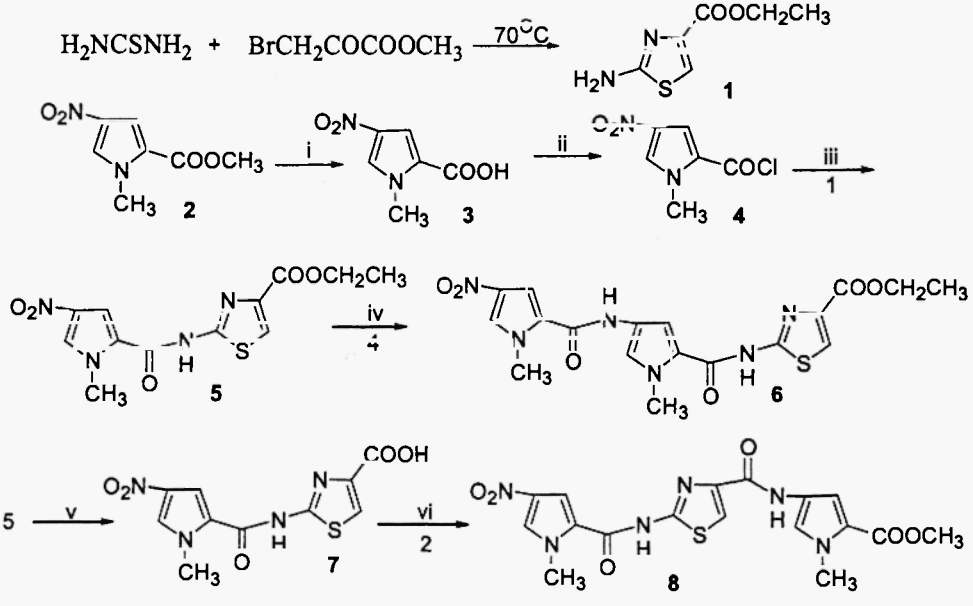
In our previous studies, we reported the synthesis of bis-1-chloromethyl-5-hydroxy-1,2-dihydro-3*H*-benz[*e*]indole (*seco*-CBI) pyrrole and imidazole polyamide conjugates[14,15] which contain two

racemic CBI moieties linked from two different positions to pyrrole and imidazole polyamides by a methylene chain of variable length. The structural consideration we are examining in the present paper is the complementary one that a given heterocyclic moiety can actually exclude binding at a particular base site. Accordingly, we report the synthesis of novel thiazole-containing bis *seco*-CBI polyamide conjugates. In their complexes with DNA the sulfur of the thiazole is oriented toward the floor of the minor groove thereby preventing binding at a G-C site. The base-recognizing properties of the thiazole moiety are employed in the side chain of the glycopeptide antitumor antibiotic bleomycin,[16] which is thought to determine the sequence-recognizing properties of the latter.[17] Thiazole-containing polyamides have shown high preference for AT base pairs and very good cytotoxicity against different type of cancer cells. We herein describe the synthesis of bis-1-chloromethyl-5-hydroxy-1,2-dihydro-3*H*-benz[*e*]indole)-thiazole containing polyamide conjugates which contain two racemic CBI moieties linked from two different positions with thiazole contaning polyamide by a flexible methylene chain of variable length.

Results and discussion

The compounds **1-8** were made according to the routes described in Scheme 1. Pyruvic acid was brominated and the product was condensed with thiourea to afford hydrobromide salt of 2-aminothiazole-4-carboxylic acid, which was then esterified and neutralized to provide compound **1**. Condensation of the acid chloride of 1-methyl-4-nitropyrrole-2-carboxylic acid with **1** proceeded normally to give **5** in 70% yield. The nitro group of compound **5** was reduced to the amine group and the amine derivative was then coupled with the acid chloride of 1-methyl-4-nitropyrrole-2-carboxylic acid to afford compound **6** in 60% yield. Hydrolysis of compound **5** gave its corresponding carboxylic acid **7** which was then coupled with the reduced compound of **2** to give compound **8** in 70% yield.

SCHEME 1

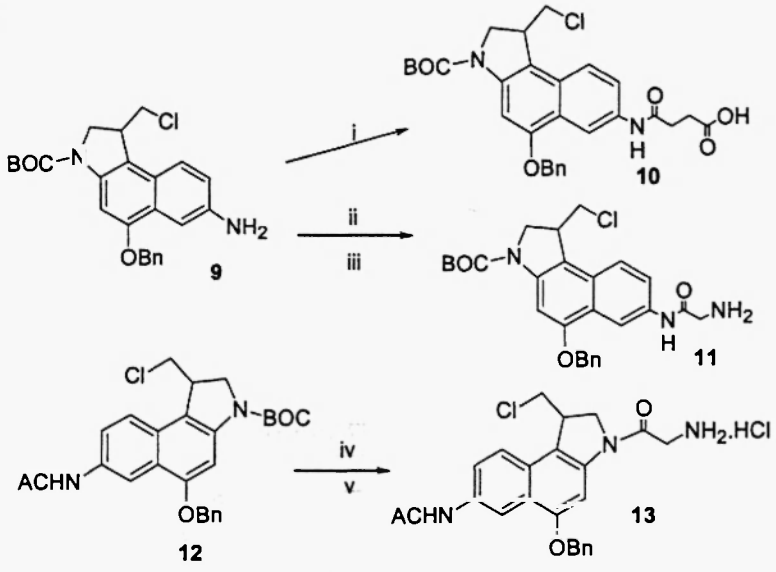


(i) 1N NaOH, THF:MeOH 1:1 (ii) $SOCl_2$, 60 °C (iii) **1**, THF, Et_3N, RT (iv) H_2 Pd / C, MeOH, **4**, DMF, RT (v) 1N NaOH, THF:MeOH 1:1, 60 °C (vi) **2**, EDCI, HOBT, DMF, RT

The *seco*-CBI compounds **10, 11** and **13**(Scheme 2) were prepared by the earlier reported procedure in good yield.[13-15] Condensation of the *seco*-CBI acid **10** with the amine moiety of the thiazole containing polyamides **5, 6, 8,** using EDCI and HOBt as the coupling agents, in dry DMF afforded the corresponding coupled *seco*-CBI thiazole containing polyamide methyl esters **14-16** in 80% yield. Hydrolysis of **14-16** with 0.5 N NaOH produced the corresponding *seco*-CBI polyamide acid compounds **17-19** in 70% yield (Scheme 3). The corresponding amino compounds were then prepared by hydrogenation of the respective nitro polyamides **5, 6, 8**. The *seco*-CBI polyamide acids **17-19** were then coupled with the *seco*-CBI **11,** or **13** containing a more nucleophilic primary amine group, under EDCI, HOBt coupling conditions in dry DMF to afford the corresponding coupled bis *seco*-CBI thiazole containing polyamides **20-22** and **26-28** in 60% yield. Treatment of **20-22** and **26-28** with ammonium formate in the presence of Pd-C for about 2 h provided the final C7-C7 and C7-N3 bis *seco*-CBI thiazole containing polyamide dimers **23-25** and **29-31** in fair yield (Schemes 4 and 5).

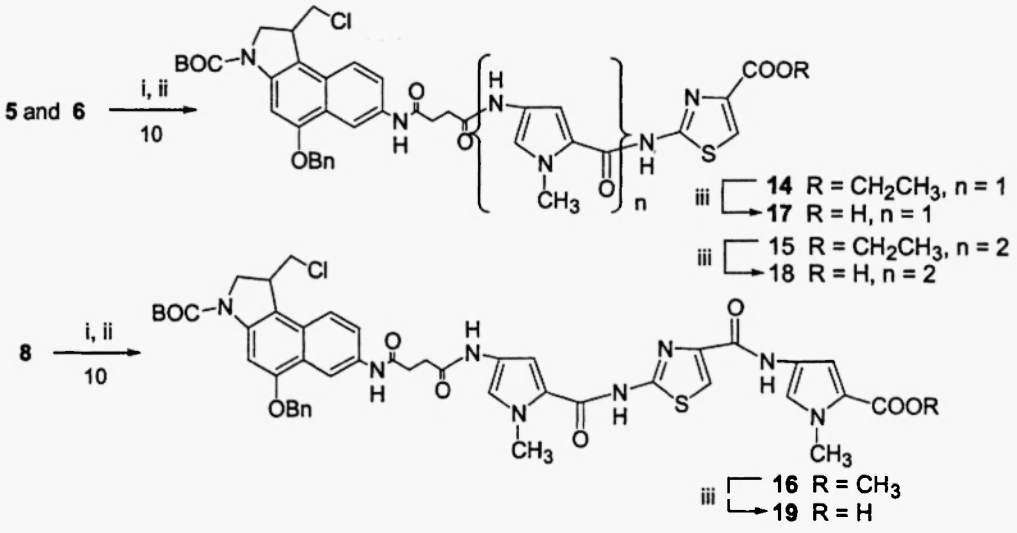
In summary, we have described the first synthesis of the thiazole-containing bis -1-chloromethyl-5-hydroxy-1,2-dihydro-3*H*-benz[*e*]indole (*seco*-CBI)-polyamide conjugates **23-25** and **29-31** (bis *seco*-CBI-thiazole containing polyamide dimers). Results on the DNA sequence preferences and biological evaluation will be reported in due course.

SCHEME 2



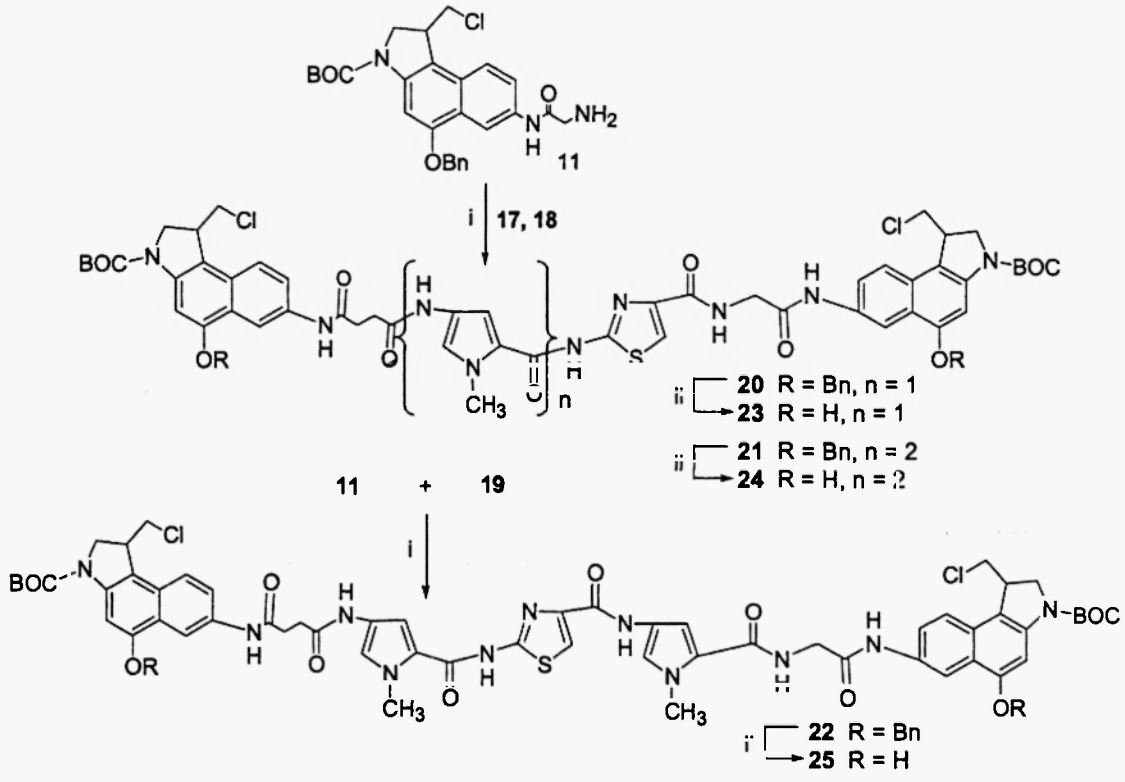
(i) Succinic anhydride, Et $_3$ N, THF, RT. (ii) N-FmOC glycine, EDCI, HOBt, DMF, RT (iii)TBAF, THF, RT. (iv) 4M HCl in dioxane, RT, 2h. N-BOC glycine, EDCI, HOBt,$NaHCO_3$, DMF, RT(v) 4M HCl in dioxane, RT, 2h

SCHEME 3



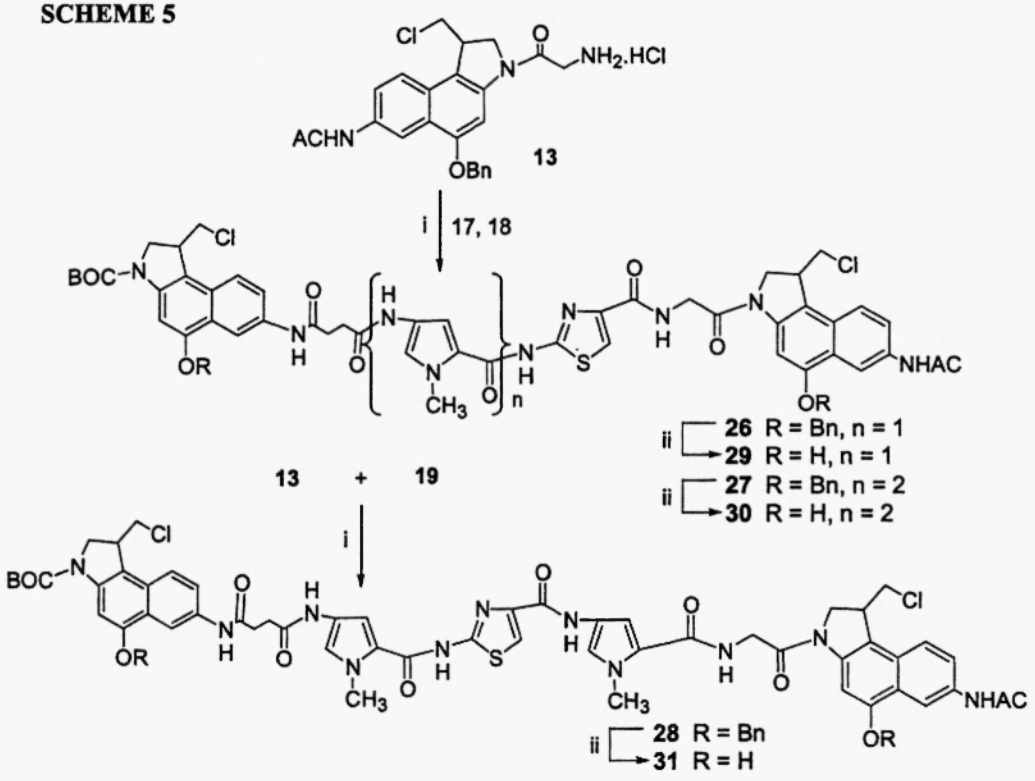
(i) H2, Pd/C, MeOH or DMF, RT. (ii) **10**, EDCI, HOBt, DMF, RT.(iii) 1 N NaOH, THF/MeOH (1:1), RT.

SCHEME 4



(i) EDCI, HOBT, DMF, RT, 12h. (ii) 10% $HCOONH_4$, 10% Pd/C, THF RT.

SCHEME 5



(i) EDCI, HOBT, DMF, RT, 12h. (ii) 10% $HCOONH_4$, 10% Pd/C, THF RT.

Experimental

General. The ^{1}H-NMR and IR spectra were recorded at 300 MHz in DMSO-d_6 and in nujol, respectively.

Ethyl 2-Aminothiazole-4-carboxylate (1). Synthesis of this compound was essentially based on the reported method;[18] mp 171-4 ^{0}C; IR ν 3450, 1690, 1620, 1540, 1460cm^{-1}; ^{1}H-NMR δ1.1(t, 3H, CH_3), 4.1 (q, 2H, CH_2), 7.13 (br s, 2H, NH_2), 7.35 (s, 1H, Ar-H) ; MS, m/z calcd for $C_6H_8N_2O_2S$ 172.0306, found 172.0306.

2-[(1-Methyl-4-nitro-1*H*-pyrrole-2-carbonyl)amino]thiazole-4-carboxylic acid ethyl ester (5). 1-Methyl-4-nitropyrrole-2-carboxylic acid (1.0g, 5.88 mmol) was refluxed in 20 ml of thionyl chloride in for 2 h. The mixture was concentrated under reduced pressure, and the residue was treated with toluene and concentrated again to remove traces of thionyl chloride. The resulting acid chloride was used without further purification. This acid chloride was dissolved in 10 ml of dry THF and the solution was added to a mixture of ethyl 2-aminothiazole-4-carboxylate **1** (1.01g, 5.87 mmol) and 0.5 ml of triethylamine in 10 ml of dry THF at 0 ^{0}C. The mixture was allowed to attain room temperature and stirred for 10 h. The THF was evaporated in vacuo, and water was added to precipitate a solid. This solid was collected, washed with acid, alkali, and water, and dried in vacuo; yield 1.5g (70%); mp 215-8 ^{0}C; IR 3250, 1720, 1660, 1540, 1510, 1460cm^{-1}; ^{1}H-NMR δ 1.26 (t, 3H, CH_3), 3.96 (s, 3H, CH_3), 4.28 (q, 2H, CH_2), 8.10 (s, 1H, Ar-H), 8.02 and 8.3 (2d, 2H, Ar-H) 10.58 (br s, 1H, NH); MS, *m/z* calcd for $C_{12}H_{12}N_4O_5S$ 324.0528, found 324.0528.

2-({1-Methyl-4-[(1-methyl-4-nitro-1*H*-pyrrole-2-carbonyl)amino]-1*H*-pyrrole-2-carbonyl}amino)thiazole-4-carboxylic acid ethyl ester (6).

A solution of compound **5** (1.0g, 3.08 mmol) in methanol (50ml) was hydrogenated over 10 % Pd/C at room temperature for 2.5 h. After completion of the reaction the catalyst was removed by filtration, and the filtrate was concentrated to give its the amino compound. This amine was used immediately in the following step.

To the solution of the above amino compound and triethylamine (0.5ml) in THF was added acid chloride of 1-methyl-4-nitropyrrole-2-carboxylic acid (0.53g, 3.12 mmol) in THF (20ml) dropwise at 0 ^{0}C. The mixture was allowed to attain room temperature and stirred for 12 h. The solvent was evaporated to dryness under reduced pressure and the resulting solid was purified by flash column chromatogarphy using DCM MeOH (9:1) to give **6** in 70 % yield; ^{1}H-NMR δ 1.26 (t, 3H, CH_3), 3.96 (s, 3H, CH_3), 3.98 (s, 3H, CH_3), 4.28 (q, 2H, CH_2), 7.75 (d, J = 2.2 Hz, 1H, Py-H), 7.95 (d, J = 2.2 Hz, 1H, Py-H), 8.10 (s, 1H, Thio-H), 8.02 and 8.3 (2d, J = 2.2 Hz, 2H, Py-H), 10.45 (s, 1H, NH), 10.58 (s, 1H, NH); MS, *m/z* calcd for $C_{18}H_{18}N_6O_6S$ 446.05, found 446.0528.

2-[(1-Methyl-4-nitro-1*H*-pyrrole-2-carbonyl)-amino]thiazole-4-carboxylic acid (7).

A suspension of 1.0g (3.08-m mole) of compound **5** in 10ml of 2 M NaOH in 1:1 water-methanol was stirred for 2 h. Or until the solid dissolved. Insoluble solids, if any, were removed by filtration, and the methanol was evaporated in vacuo. The remaining water solution was cooled to 5 ^{0}C and acidified with cold 6 N HCl. The solid **7** was collected, washed with water, and dried in vacuo, 700mg (70% yield); mp 313-5^{0}C; IR ν 3600, 1850, 1680, 1570, 1540, 1500, 1460cm^{-1}; ^{1}H-NMR δ 3.99 (s, 3H, CH_3), 8.03 (s, 1H, Ar-H), 8.01 and 8.31 (2d, 2H, Ar-H), 10.52 (br s, 1H, NH); MS, *m/z* calcd for $C_{10}H_8N_4O_5S$ 296.0215, found 296.0219.

1-Methyl-4-({2-[(1-methyl-4-nitro-1*H*-pyrrole-2-carbonyl)amino]thiazole-4-carbonyl}-amino)-1*H*-pyrrole-2-carboxylic acid methyl ester (8).

A solution of methyl 1-methyl-4-nitropyrrole-2-carboxylate (0.45g, 2.44 mmol) in MeOH (50.0 ml) was hydrogenated over 10% Pd-C at 50 psi for 2 h and then filtered. The filtrate was concentrated to dryness under reduced pressure (at RT) to afford the corresponding amino compound. Due to the sensitivity of the amine to oxidation, it was used for the next reaction immediately. It was dissolved in dry DMF (30 ml) and a mixture of the acid **7** (0.718 g, 2.42 mmol), HOBT (0.327 g, 2.41 mmol), and EDCI (1.16 g, 6.05 mmol) in DMF (15 ml) was added. This mixture was stirred at RT for 12 h and the solvent was removed under reduced pressure to afford a dark oil which was purified by flash chromatography on silica gel (methanol/dichloromethane, 1:9 to 2:8) to afford coupled polyamide **8** as a white solid in 65% yield. ^{1}H-NMR δ 3.86 (s, 3H, CH_3), 3.96 (s, 3H, CH_3), 3.98 (s, 3H, CH_3), 7.35 (d, J= 2.2Hz, 1H, Py-H), 7.56 (d, J = 2.2 Hz, 1H, Py-H), 8.10 (s, 1H, Thio-H), 8.02 and 8.3 (2d, J= 2.2 Hz, 2H, Py-H), 10.42 (s, 1H, NH), 10.56 (s, 1H, NH); MS, *m/z* calcd for $C_{17}H_{16}N_6O_6S$ 432.05, found 432.049.

General procedure for the synthesis of compounds 14-16.

A solution of the nitropolyamides **5, 6 or 8** in MeOH or DMF was hydrogenated over 10% Pd/C at 50 psi pressure for two hours and the catalyst was removed by filtration through a Celite pad. The filtrate was concentrated to dryness under reduced pressure (at RT) to afford the corresponding amine. Owing to the sensitivity of the amine to oxidation, it was used for the next reaction immediately. It was dissolved in dry DMF and a mixture of the acid **10**, hydroxybenzotriazole and EDCI in DMF was added. This mixture was stirred at RT for 12 h and after completion of the reaction the solvent was removed under reduced pressure to afford a dark oil which was purified by flash column chromatography on silica gel by using methanol-dichloromethane as eluent to afford the *seco*-CBI polyamide esters **14-16** as white solids in 60% yield.

5-Benzyloxy-1-chloromethyl-7-{3-[5-(4-ethoxycarbonylthiazol-2-ylcarbamoyl)-1-methyl-1*H*-pyrrol-3-ylcarbamoyl]propionylamino}-1,2-dihydrobenz[*e*]indole-3-carboxylic acid *tert*-butyl ester (14). ^{1}H-NMR δ 1.41 (s, 3H, CH_3), 1.52 (s, 9H, Boc-H), 2.56-2.70 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.90-4.10 (m, 5H, -CH, CH_2Cl, CH_2N), 4.10 (q, 2H, -CH_2CH_3), 5.24 (s, 2H, -

$OCH_2C_6H_5$), 7.20 (d, 1H, J = 1.8 Hz, Py-H), 7.50 (d, 1H, J = 1.8 Hz, Py-H), 7.52-7.90 (m, 8H, Ar-H), 8.0 (s, 1H, Thia-H), 8.40 (s, 1H, C6-H), 10.0 (s, 1H), 10.20 (s, 1H), 10.32 (s, 1H); ES–MS m/z calcd for $C_{41}H_{43}N_6O_8ClSNa$ 836.50, found 836.50 ($M+Na^+$).

5-Benzyloxy-1-chloromethyl-7-(3-{5-[5-(4-ethoxycarbonyl-thiazol-2-ylcarbamoyl)-1-methyl-1*H*-pyrrol-3-ylcarbamoyl]-1-methyl-1*H*-pyrrol-3-ylcarbamoyl}-propionylamino)-1,2-dihydrobenz[*e*]indole-3-carboxylic acid *tert*-butyl ester (15): ^{1}H-NMR δ 1.39 (s, 3H, CH_3), 1.50 (s, 9H, Boc-H), 2.56-2.70 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.85 (s, 3H, -NCH_3), 3.90-4.10 (m, 5H, -CH, CH_2Cl, CH_2N), 4.10 (q, 2H, -CH_2CH_3), 5.26 (s, 2H, -$OCH_2C_6H_5$), 6.96 (d, 1H, J = 1.8 Hz, Py-H), 7.05 (d, 1H, J = 1.8 Hz, Py-H), 7.18 (d, 1H, J = 1.8 Hz, Py-H), 7.45 (d, 1H, J = 1.8 Hz, Py-H), 7.50-7.90 (m, 8H, Ar-H), 8.10 (s, 1H, Thia-H), 8.45 (s, 1H, C6-H), 10.10 (s, 1H), 10.12 (s, 1H), 10.20 (s, 1H), 10.30 (s, 1H); ES–MS, m/z calcd for $C_{47}H_{49}N_8O_9ClSNa$ 959.40, found 959.401 ($M+Na^+$).

5-Benzyloxy-1-chloromethyl-7-(3-{5-[4-(5-methoxycarbonyl-1-methyl-1*H*-pyrrol-3-ylcarbamoyl)thiazol-2-ylcarbamoyl]-1-methyl-1*H*-pyrrol-3-ylcarbamoyl}-propionylamino)-1,2-dihydrobenz[*e*]indole-3-carboxylic acid *tert*-butyl ester (16): ^{1}H-NMR δ 1.52 (s, 9H, Boc-H), 2.56-2.70 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.85 (s, 3H, -NCH_3), 3.89 (s, 3H, -OCH_3), 3.92-4.10 (m, 5H, -CH, CH_2Cl, CH_2N), 5.25 (s, 2H, -$OCH_2C_6H_5$), 6.96 (d, 1H, J = 1.8 Hz, Py-H), 7.05 (d, 1H, J = 1.8 Hz, Py-H), 7.20 (d, 1H, J = 1.8Hz, Py-H), 7.41 (d, 1H, J = 1.8 Hz, Py-H), 7.45-7.90 (m, 8H, Ar-H), 8.20 (s, 1H, Thia-H), 8.45 (s, 1H, C6-H), 9.95 (s, 1H), 10.12 (s, 1H), 10.20 (s, 1H), 10.35 (s, 1H); ES–MS, m/z calcd for $C_{46}H_{47}N_8O_9ClSNa$ 945.40, found 945.30 ($M+Na^+$).

General procedure for the synthesis of compounds 17-19.

A mixture of *seco*-CBI polyamide ester **14, 15 or 16** in methanol and 10 ml of 0.5N NaOH was placed in a flask, then the mixture was stirred at room temperature until the ester completely disappeared as shown by TLC. The mixture was cooled in ice with stirring and neutralized with 0.5 N HCl slowly to pH 2. The mixture was extracted with ethyl acetate and THF (1:1) three times and dried over sodium sulfate, and the solvent was removed under reduced pressure. The residue was purified by column chromatography using MeOH / dichloromethane as eluent solvent to afford the *seco*-CBI polyamide acids **17-19** in 60 % yield.

5-Benzyloxy-7-{3-[5-(4-carboxythiazol-2-ylcarbamoyl)-1-methyl-1*H*-pyrrol-3-ylcarbamoyl]propionylamino}-1-chloromethyl-1,2-dihydrobenz[*e*]indole-3-carboxylic acid *tert*-butyl ester (17): ^{1}H-NMR δ 1.52 (s, 9H, Boc-H), 2.56-2.70 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.90-4.10 (m, 5H, -CH, CH_2Cl, CH_2N), 5.24 (s, 2H, -$OCH_2C_6H_5$), 7.18 (d, 1H, J = 1.8 Hz, Py-H), 7.45 (d, 1H, J = 1.8 Hz, Py-H), 7.52-7.90 (m, 8H, Ar-H), 8.0 (s, 1H, Thia-H), 8.40 (s, 1H, C6-H), 10.0 (s, 1H), 10.20 (s, 1H), 10.32 (s, 1H), 12.52 (s, 1H, -COOH); ES–MS, m/z calcd for $C_{39}H_{39}N_6O_8ClSNa$ 808.50, found 808.50 ($M+Na^+$).

5-Benzyloxy-7-(3-{5-[5-(4-carboxythiazol-2-ylcarbamoyl)-1-methyl-1*H*-pyrrol-3-ylcarbamoyl]-1-methyl-1H-pyrrol-3-ylcarbamoyl}propionylamino)-1-chloromethyl-1,2-dihydrobenz[*e*]indole-3-carboxylic acid *tert*-butyl ester (18): ^{1}H-NMR δ 1.50 (s, 9H, Boc-H), 2.56-2.70 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.85 (s, 3H, -NCH_3), 3.90-4.10 (m, 5H, -CH, CH_2Cl, CH_2N), 5.25 (s, 2H, -$OCH_2C_6H_5$), 6.98 (d, 1H, J = 1.8 Hz, Py-H), 7.10 (d, 1H, J = 1.8 Hz, Py-H), 7.20 (d, 1H, J = 1.8 Hz, Py-H), 7.49 (d, 1H, J = 1.8 Hz, Py-H), 7.52-7.90 (m, 8H, Ar-H), 8.10 (s, 1H, Thia-H), 8.49 (s, 1H, C6-H), 10.10 (s, 1H), 10.12 (s, 1H), 10.20 (s, 1H), 10.30 (s, 1H), 12.50 (s, 1H, -COOH); ES–MS, m/z calcd for $C_{45}H_{45}N_8O_9ClSNa$ 931.42, found 931.42 ($M+Na^+$).

5-Benzyloxy-7-(3-{5-[4-(5-carboxy-1-methyl-1*H*-pyrrol-3-ylcarbamoyl)thiazol-2-ylcarbamoyl]-1-methyl-1*H*-pyrrol-3-ylcarbamoyl}propionylamino)-1-chloromethyl-1,2-dihydrobenz[*e*]indole-3-carboxylic acid *tert*-butyl ester (19): ^{1}H-NMR δ 1.52 (s, 9H, Boc-H), 2.56-2.70 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.85 (s, 3H, -NCH_3), 3.92-4.10 (m, 5H, -CH,

CH_2Cl, CH_2N), 5.24 (s, 2H, -OC**H**$_2C_6H_5$), 6.96 (d, 1H, J = 1.8 Hz, Py-H), 7.15 (d, 1H, J = 1.8 Hz, Py-H), 7.20 (d, 1H, J = 1.8 Hz, Py-H), 7.45 (d, 1H, J = 1.8 Hz, Py-H), 7.49-7.90 (m, 8H, Ar-H), 8.10 (s, 1H, Thia-H), 8.45 (s, 1H, C6-H), 9.95 (s, 1H), 10.12 (s, 1H), 10.20 (s, 1H), 10.35 (s, 1H) 12.56 (s, 1H, -COOH); ES–MS, m/z calcd for $C_{45}H_{45}N_8O_9ClSNa$ 931.42, found 931.32 ($M+Na^+$).

General procedure for the synthesis of compounds 20-22 and 26-28.

To a solution of *seco*-CBI polyamide acids **17-19** in dry DMF (20ml) were added EDCI (2.5 mol), HOBt (1.0 mol), and *seco*-CBI amine **11** (1.1 mol) for compounds **20-22** or *seco*-CBI amine **13** (1.1 mol) and $NaHCO_3$ (3.0 mol) for compounds **26-28** under a nitrogen atmosphere and the mixture was stirred for 12h. When TLC indicated the absence of starting material, DMF was removed under reduced pressure. The dark residue was purified by column chromatography on silica gel using MeOH / dichloromethane as eluent to afford the coupled conjugates **20-22** and **26-28** as white solids in 70 % yield.

Compound 20. ^{1}H-NMR δ 1.52 (s, 18H, 2×Boc-H), 2.56-2.70 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.89-4.10 (m, 12H, Cl, 2-H, 2×CH_2Cl, 2×CH_2N, NHC**H**$_2$), 5.24 (s, 2H, -OC**H**$_2C_6H_5$), 5.25 (s, 2H, -OC**H**$_2C_6H_5$), 6.96 (d, 1H, J = 1.8 Hz, Py-H), 7.18 (d, 1H, J = 1.8 Hz, Py-H), 7.59-7.75 (m, 16H, Ar-H), 8.12 (s, 1H, Thia-H), 8.32 (m, 1H, N**H**CH_2), 8.40 (s, 2H, 2×C6-H), 9.91 (s, 1H), 10.09 (s, 1H), 10.12 (s, 1H), 10.30 (s, 1H); ES–MS, m/z calcd for $C_{66}H_{67}N_9O_{11}Cl_2SNa$ 1286.40, found 1286.40 ($M+Na^+$).

Compound 21. ^{1}H-NMR δ 1.56 (s, 18H, 2×Boc-H), 2.56-2.70 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.85 (s, 3H, -NCH_3), 3.90-4.10 (m, 12H, Cl, 2-H, 2×CH_2Cl, 2×CH_2N, NHC**H**$_2$), 5.23 (s, 2H, -OC**H**$_2C_6H_5$), 5.25 (s, 2H, -OC**H**$_2C_6H_5$), 6.96 (d, 1H, J = 1.8 Hz, Py-H), 7.18 (d, 1H, J = 1.8 Hz, Py-H), 7.24 (d, 1H, J = 1.8 Hz, Py-H), 7.26 (d, 1H, J = 1.8 Hz, Py-H), 7.59-7.85 (m, 16H, Ar-H), 8.10 (s, 1H, Thia-H), 8.35 (m, 1H, N**H**CH_2), 8.40 (s, 2H, 2×C6-H), 9.91 (s, 1H), 9.95 (s, 1H), 10.09 (s, 1H), 10.12 (s, 1H), 10.30 (s, 1H); ES–MS, m/z calcd for $C_{72}H_{73}N_{11}O_{12}Cl_2SNa$ 1408.40, found 1408.40 ($M+Na^+$).

Compound 22. ^{1}H-NMR δ 1.54 (s, 18H, 2×Boc-H), 2.56-2.70 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.85 (s, 3H, -NCH_3), 3.90-4.10 (m, 12H, Cl, 2-H, 2×CH_2Cl, 2×CH_2N, NHC**H**$_2$), 5.24 (s, 2H, -OC**H**$_2C_6H_5$), 5.26 (s, 2H, -OC**H**$_2C_6H_5$), 6.88 (d, 1H, J = 1.8 Hz, Py-H), 7.05 (d, 1H, J = 1.8 Hz, Py-H), 7.24 (d, 1H, J = 1.8 Hz, Py-H), 7.26 (d, 1H, J = 1.8 Hz, Py-H), 7.60-7.85 (m, 16H, Ar-H), 8.15 (s, 1H, Thia-H), 8.35 (m, 1H, N**H**CH_2), 8.40 (s, 2H, 2×C6-H), 9.91 (s, 1H), 9.95 (s, 1H), 10.09 (s, 1H), 10.12 (s, 1H), 10.32 (s, 1H); ES–MS, m/z calcd for $C_{72}H_{73}N_{11}O_{12}Cl_2SNa$ 1408.40, found 1408.40 ($M+Na^+$).

Compound 26. ^{1}H-NMR δ 1.52 (s, 9H, Boc-H), 2.04 (s, 3H, CH_3CON), 2.58-2.70 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.90-4.40 (m, 12H, Cl, 2-H, 2×CH_2Cl, 2×CH_2N, NHC**H**$_2$), 5.24 (s, 2H, -OC**H**$_2C_6H_5$), 5.25 (s, 2H, -OC**H**$_2C_6H_5$), 6.96 (d, 1H, J = 1.5 Hz, Py-H), 7.16 (d, 1H, J = 1.5 Hz, Py-H), 7.60-7.79 (m, 16H, Ar-H), 8.10 (s, 1H, Thia-H), 8.21-8.30 (m, 1H, N**H**CH_2), 8.35-8.42 (m, 2H, 2×C6-H), 10.03 (s, 1H), 10.07 (s, 1H), 10.14 (s, 1H), 10.32 (s, 1H); HR-ESMS, m/z calcd for $C_{63}H_{61}N_9O_{10}Cl_2SNa$ 1228.30, found 1228.30 ($M+Na^+$).

Compound 27. ^{1}H-NMR δ 1.54 (s, 9H, Boc-H), 2.04 (s, 3H, CH_3CON), 2.58-2.70 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.85 (s, 3H, -NCH_3), 3.90-4.35 (m, 12H, Cl, 2-H, 2×CH_2Cl, 2×CH_2N, NHC**H**$_2$), 5.22 (s, 2H, -OC**H**$_2C_6H_5$), 5.24 (s, 2H, -OC**H**$_2C_6H_5$), 6.96 (d, 1H, J = 1.5 Hz, Py-H), 7.16 (d, 1H, J = 1.5 Hz, Py-H), 7.23 (d, 1H, J = 1.5 Hz, Py-H), 7.26 (d, 1H, J = 1.5 Hz, Py-H), 7.61-7.85 (m, 16H, Ar-H), 8.15 (s, 1H, Thia-H), 8.25-8.32 (m, 1H, N**H**CH_2), 8.35-8.42 (m, 2H, 2×C6-H), 10.02 (s, 1H), 10.05 (s, 1H), 10.09 (s, 1H), 10.18 (s, 1H), 10.30 (s, 1H); HR-ESMS, m/z calcd for $C_{69}H_{67}N_{11}O_{11}Cl_2SNa$ 1350.39 found 1350.39 ($M+Na^+$).

Compound 28. ^{1}H-NMR δ 1.52 (s, 9H, Boc-H), 2.04 (s, 3H, CH_3CON), 2.58-2.70 (m, 4H, 2×CH_2CO-), 3.83 (s, 3H, -NCH_3), 3.85 (s, 3H, -NCH_3), 3.89-4.40 (m, 12H, Cl, 2-H, 2×CH_2Cl,

2×CH_2N, NHC**H**$_2$), 5.25 (s,2H, -OC**H**$_2C_6H_5$), 5.26 (s, 2H, -OC**H**$_2C_6H_5$), 6.98 (d, 1H, J = 1.5 Hz, Py-H), 7.18 (d, 1H, J = 1.5 Hz, Py-H), 7.24 (d, 1H, J = 1.5 Hz, Py-H), 7.27 (d, 1H, J = 1.5 Hz, Py-H), 7.59-7.85 (m, 16H, Ar-H), 8.10 (s, 1H, Thia-H), 8.20-8.30 (m, 1H, N**H**CH_2), 8.35-8.42 (m, 2H, 2×C6-H), 10.00 (s, 1H), 10.08 (s, 1H), 10.15 (s, 1H), 10.22 (s, 1H)10.32 (s, 1H); HR-ESMS, m/z calcd for $C_{69}H_{67}N_{11}O_{11}Cl_2SNa$ 1350.30 found 1350.30 ($M+Na^+$).

General procedure for the synthesis of compounds 23-25 and 29-31.

To a solution of compounds **20-22** and **26-28** in THF or DMF was added Black-Pd-C under argon. The mixture was cooled to 0^0C and 10% aqueous ammonium formate was added. The mixture was stirred at 23 ^{0}C until the reaction was complete (TLC). The mixture was then filtered through a pad of Celite, and concentrated in vacuo. The crude product was purified by flash column chromatography using MeOH / dichloromethane as eluent to afford the thiazole-containing bis-*seco*-CBI-polyamide conjugate compounds **23-25** and **29-31** as white solids in 70 % yield.

Compound 23. ^{1}H-NMR δ 1.52 (s, 18H, 2×Boc-H), 2.50-2.71 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.90-4.10 (m, 12H, Cl, 2-H, 2×CH_2Cl, 2×CH_2N, NHC**H**$_2$), 6.96 (d, 1H, J = 1.8 Hz, Py-H), 7.18 (d, 1H, J = 1.8 Hz, Py-H), 7.59-7.75 (m, 6H, 2×C4-H, C7-H, C8-H), 8.12 (s, 1H, Thia-H), 8.32 (m, 1H, N**H**CH_2), 8.40 (s, 2H, 2×C6-H), 9.91 (s, 1H), 9.95 (s, 1H), 10.09 (s, 1H), 10.12 (s, 1H), 10.30 (s, 1H), 10.32 (s, 1H); ES–MS, m/z calcd for $C_{52}H_{55}N_9O_{11}Cl_2SNa$ 1106.40, found 1106.40 ($M+Na^+$).

Compound 24. ^{1}H-NMR δ 1.56 (s, 18H, 2×Boc-H), 2.56-2.70 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.85 (s, 3H, -NCH_3), 3.89-4.20 (m, 12H, Cl, 2-H, 2×CH_2Cl, 2×CH_2N, NHC**H**$_2$), 6.96 (d, 1H, J = 1.8 Hz, Py-H), 7.05 (d, 1H, J = 1.8 Hz, Py-H), 7.24 (d, 1H, J = 1.8 Hz, Py-H), 7.26 (d, 1H, J = 1.8 Hz, Py-H), 7.55-7.85 (m, 6H, 2×C4-H, C7-H, C8-H), 8.18 (s, 1H, Thia-H), 8.32 (m, 1H, N**H**CH_2), 8.43 (s, 2H, 2×C6-H), 9.89 (s, 1H), 9.91 (s, 1H), 9.95 (s, 1H), 10.09 (s, 1H), 10.12 (s, 1H), 10.30 (s, 1H), 10.32 (s, 1H); ES–MS, m/z calcd for $C_{58}H_{61}N_{11}O_{12}Cl_2SNa$ 1228.40, found 1228.40 ($M+Na^+$).

Compound 25. ^{1}H-NMR δ 1.54 (s, 18H, 2×Boc-H), 2.56-2.72 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.85 (s, 3H, -NCH_3), 3.90-4.15 (m, 12H, Cl, 2-H, 2×CH_2Cl, 2×CH_2N, NHC**H**$_2$), 6.96 (d, 1H, J = 1.8 Hz, Py-H), 7.05 (d, 1H, J = 1.8 Hz, Py-H), 7.24 (d, 1H, J = 1.8 Hz, Py-H), 7.26 (d, 1H, J = 1.8 Hz, Py-H), 7.55-7.85 (m, 6H, 2×C4-H, C7-H, C8-H), 8.18 (s, 1H, Thia-H), 8.32 (m, 1H, N**H**CH_2), 8.43 (s, 2H, 2×C6-H), 9.89 (s, 1H), 9.91 (s, 1H), 9.95 (s, 1H), 10.09 (s, 1H), 10.12 (s, 1H), 10.30 (s, 1H), 10.32 (s, 1H); ES–MS, m/z calcd for $C_{58}H_{61}N_{11}O_{12}Cl_2SNa$ 1228.40, found 1228.40 ($M+Na^+$).

Compound 29. ^{1}H-NMR δ 1.52 (s, 9H, Boc-H), 2.04 (s, 3H, CH_3CON), 2.58-2.70 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.90-4.40 (m, 12H, Cl, 2-H, 2×CH_2Cl, 2×CH_2N, NHC**H**$_2$), 6.96 (d, 1H, J = 1.5 Hz, Py-H), 7.16 (d, 1H, J = 1.5 Hz, Py-H), 7.60-7.79 (m, 5H, 2×C8, C9-H, C4-H), 7.90 (d, 1H, C4-H), 8.18 (s, 1H, Thia-H), 8.20-8.30 (m, 1H, N**H**CH_2), 8.35-8.42 (m, 2H, 2×C6-H), 9.91 (s, 1H), 9.94 (s, 1H), 10.05 (s, 1H), 10.07 (s, 1H), 10.14 (s, 1H), 10.32 (s, 1H); HR-ESMS, m/z calcd for $C_{49}H_{49}N_9O_{10}Cl_2SNa$ 1048.30, found 1048.30 ($M+Na^+$).

Compound 30. ^{1}H-NMR δ 1.54 (s, 9H, Boc-H), 2.04 (s, 3H, CH_3CON), 2.60-2.72 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.85 (s, 3H, -NCH_3), 3.91-4.40 (m, 12H, Cl, 2-H, 2×CH_2Cl, 2×CH_2N, NHC**H**$_2$), 6.96 (d, 1H, J = 1.5 Hz, Py-H), 7.16 (d, 1H, J = 1.5 Hz, Py-H), 7.23 (d, 1H, J = 1.5 Hz, Py-H), 7.26 (d, 1H, J = 1.5 Hz, Py-H), 7.60-7.79 (m, 5H, 2×C8, C9-H, C4-H), 7.90 (d, 1H, C4-H), 8.10 (s, 1H, Thia-H), 8.20-8.30 (m, 1H, N**H**CH_2), 8.35-8.42 (m, 2H, 2×C6-H), 9.91 (s, 1H), 9.94 (s, 1H), 10.03 (s, 1H), 10.05 (s, 1H), 10.07 (s, 1H), 10.14 (s, 1H), 10.32 (s, 1H); HR-ESMS, m/z calcd for $C_{55}H_{55}N_{11}O_{11}Cl_2SNa$ 1170.30, found 1170.30 ($M+Na^+$).

Compound 31. ^{1}H-NMR δ 1.56 (s, 9H, Boc-H), 2.04 (s, 3H, CH_3CON), 2.60-2.72 (m, 4H, 2×CH_2CO-), 3.82 (s, 3H, -NCH_3), 3.85 (s, 3H, -NCH_3), 3.91-4.40 (m, 12H, Cl, 2-H, 2×CH_2Cl, 2×CH_2N, NH**C**H_2), 6.96 (d, 1H, J = 1.5 Hz, Py-H), 7.16 (d, 1H, J = 1.5 Hz, Py-H), 7.23 (d, 1H, J = 1.5 Hz, Py-H), 7.26 (d, 1H, J = 1.5 Hz, Py-H), 7.59-7.70 (m, 5H, 2×C8, C9-H, C4-H), 7.90 (d, 1H, C4-H), 8.10 (s, 1H, Thia-H), 8.20-8.30 (m, 1H, N**H**CH_2), 8.35-8.42 (m, 2H, 2×C6-H), 9.91 (s, 1H), 9.94 (s, 1H), 10.03 (s, 1H), 10.05 (s, 1H), 10.07 (s, 1H), 10.14 (s, 1H), 10.32 (s, 1H); HR-ESMS, m/z calcd for $C_{55}H_{55}N_{11}O_{11}Cl_2SNa$ 1170.30 found 1170.30 ($M+Na^+$).

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