Design and Synthesis of Novel Thiazole-Containing Cross-Linked Polyamides Related to the Antiviral Antibiotic Distamycin

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A family of naturally occurring oligopeptides includes netropsin, distamycin, anthelvencin, kikumycin B, amidinomycin, and norformycin. Netropsin (**I**) and distamycin (**II**) express their biological activities by targeting specific sequences of chemical functionalities in the minor groove of DNA. Both netropsin and distamycin can be regarded as polyamide chains in which each α-carbon has been replaced by a five-membered pyrrole ring. The repeat distance in such an augmented polyamide chain is almost the same as the distance from one base pair to the next along the floor of a minor groove within *β*-DNA. In this paper we report the synthesis of **16**−**21** cross-linked polyamides containing a thiazole heterocyclic ring bearing the active functionalites NH_2, NHCHO, or H. **16** and **17** were synthesized by DCC and HOBt catalyzed reaction of **5** with **14** and **15**, while the formylation products **18** and **19** were obtained by coupling the formylated 4-methyl-thiazolated acid **6** with **14** and **15**. The deaminated compounds **20** and **21** were obtained by the coupling of 5-trichloroacetyl-4-methylthiazole **7** synthesized from 4-methylthiazole. All the six cross-linked polyamides **16**−**21** were tested for their DNA gyrase inhibition. The studies have shown these polyamides have better sequence recognition and a greater percentage of inhibition than the corresponding monomers. The compound **17** shows complete inhibition of gyrase at 0.5 *μ*M concentration as compared to the naturally occurring distamycin at 1.0 *μ*M.

Introduction

The current interest in the control of gene expression[1] has driven synthetic chemists to attempt to develop DNA sequence specific agents. Conceptually there are a number of approaches[2−4] to this problem, e.g., using *β*-oligonucleotides or their backbone-modified counterparts,[5−8] which take advantage of inherent Watson−Crick base pairing to target single-stranded nucleic acid sequences, or with hybrid probes incorporating an intercalator.[9−11] Another approach is to take advantage of the ability of certain oligonucleotides to form triplex structures and thereby target double-stranded nucleic acid sequences.[12] A complementary approach is to develop sequence specific probes based on natural DNA minor-groove-binding agents. Groove-binding agents have several advantages over intercalators for this purpose in that, unlike the latter, groove binders in general cause minimal structural distortion of the DNA and correspondingly less disturbance of the information inherent in the DNA sequences.[13] An understanding of drug−DNA interactions at the molecular level is crucial in facilitating the design of new drugs and probes that can recognize specific DNA sequences.

A family of naturally occurring oligopeptides includes netropsin (**I**),[14] distamycin (**II**),[15,16] anthelvencin (**III**),[17] kikumycin B (**IV**),[18] amidinomycin (**V**),[19] and norformycin (**VI**)[20] (Figure 1). Netropsin (**I**) and distamycin (**II**) express their biological activities by targeting specific AATT sequences in the minor groove of DNA. Both netropsin and distamycin can be regarded as polyamides chains in which each α-carbon has been replaced by a five-membered pyrrole ring. The repeat distance in such

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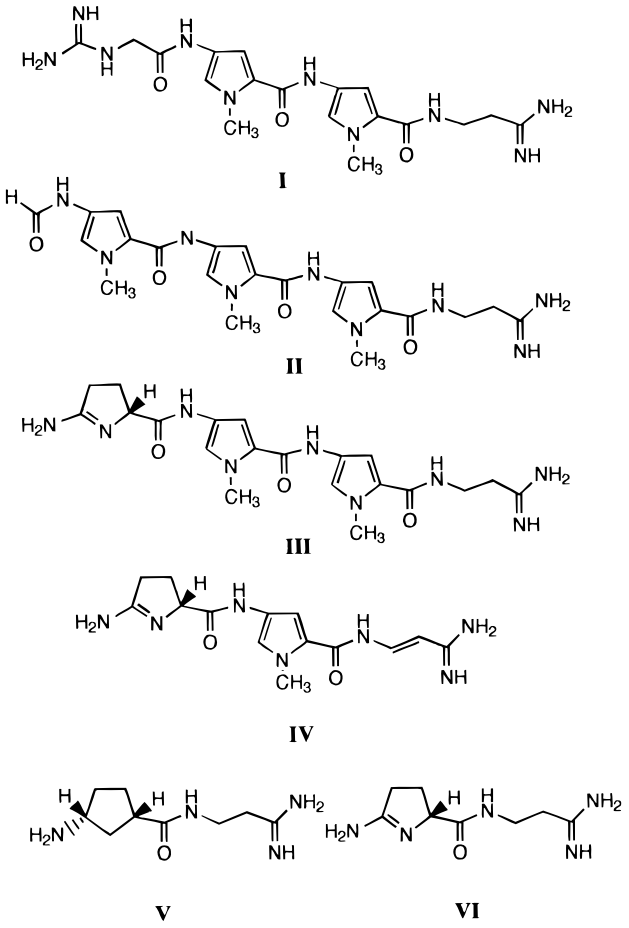

Figure 1.

an augmented polyamide chain is almost the same as the distance from one base pair to the next along the floor of a minor groove within β-DNA.[21] Lown and co-workers[22] have synthesized lexitropsins, i.e., longer chain analogues of netropsin that retain pyrrole groups at those positions where an AT base pair was to be read, but substituted imidazole or an equivalent heterocyclic ring at sites where GC base pair reading was desired. The imidazole provides room for a guanine 2-amine group and provides an acceptor for a new hydrogen bond. These changes incorporated in the parent natural product showed promising results in the recognition of GC regions,[23−25] but it was still not possible, at that stage in the development, to discriminate between end-for-end reversals (such as AT for TA) of either an AT or a GC base pair. However, these minor groove binders have the potential of reading only half the information in the minor groove. NMR studies of Pelton and Wemmer[26] have shown that, at sufficently high drug-to-DNA ratios, two distamycin molecules are simultaneously located in the minor groove in a highly overlapped antiparallel side-by-side manner with the N → C direction of each peptide strand parallel to the 5′ → 3′ direction of its adjacent DNA strands. This observation provided new scope for the information-reading capacity of polyamides. Each binding molecule interacts specifically with only one DNA strand, which constitutes a unique strand-specific information-reading pattern. In a 2:1 mode each polyamide ring contacts a single base, instead of the center of a base pair, so true base specificity is conceptually possible. Accordingly the differentiation of sequences related by GC or AT transversion can be accomplished, a problem not resolved in the original lexitropsin proposal.

The challenge for design has been to discover linkages that favor the proper alignment of the two strands in a 2:1 binding mode. The polyamides can be linked in two ways, as a single polyamide with a flexible central linkage that allows binding as a hairpin[27] or with a lateral bridge or staple that connects the centers of the two side-by-side molecules.[28,29] Hairpin linkage has the advantage of facile linear synthesis, easily allowing synthesis of different subunit sequences on the two side-by-side strands. The course of synthesis with the stapled linkages makes this more difficult. On the other hand, stapled or cross-linked polyamides may minimize the possibility of inappropriate binding modes because they are constrained to bind in the 2:1 mode, whereas hairpin-linked molecules have the possibility of binding in the 2:1 mode or in an extended 1:1 mode. Given a suitable linker, the bidentate-binding mode than complements the hairpin motif owing to its much larger binding strength.[30]

Encouraged by these results we have systematically explored several structural factors in the ligands that might be expected to contribute to the processes of molecular recognition. One legitimate aspect of molecular design in the context of polyamides is the introduction of heterocyclic moieties capable of specific DNA recognition by hydrogen bond acceptance and donation.

The structural consideration we examine in the present paper is the complementary one of site avoidance. In addition, we address the question whether this property can be incorporated into the design of agents capable of recognizing and binding to unique sequences. Accordingly we report the synthesis of novel thiazole-containing cross-linked polyamides in such a way that the sulfur of the thiazole is oriented toward the floor of the minor groove. The base-recognizing properties of the thiazole moiety are employed in the side chain of the glycopeptide antitumor antibiotic bleomycin,[31] which is thought to determine the sequence-recognizing properties of the latter.[32] Thiazole-containing polyamides have shown high preference for AT base pairs.[33,34]

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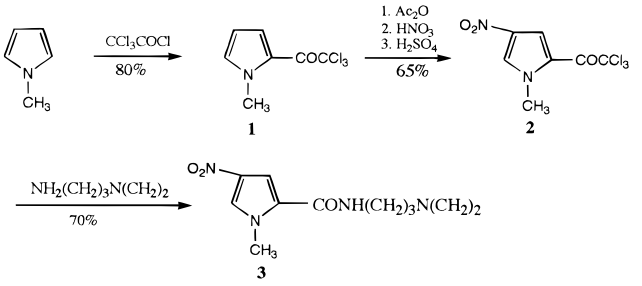
Scheme 1



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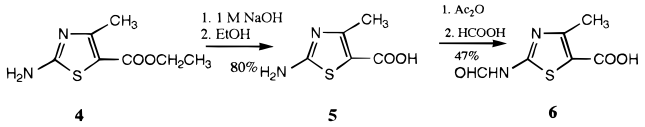
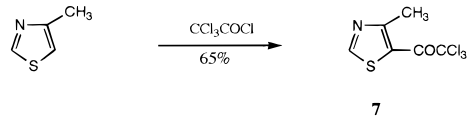


Scheme 3



Results and Discussion

Only monocationic drugs can form 2:1 side-by-side complexes, and these molecules are staggered so as to place their cationic termini far from one another. Replacing a pyrrole ring of distamycin with an imidazole ring allows the formation of a hydrogen bond between G(2)−NH_2 in the minor groove and the N3 of the imidazole moiety.[35] Earlier studies[36] conducted in our group identified that the pentamethylene linker is the shortest linker to permit a bidentate-binding interaction and the heptamethylene is a much better cross linker than either the 5- or 6-carbon linker. On the basis of these studies, we have designed the thiazolated cross- linked polyamides in such a way that the central cross linking is between the 1-H of the two imidazole moieties in order to recognize the GC-containing base sequences. The linker length was selected to be five and seven carbons. The cationic terminius was selected for the 1-methylpyrrole moiety as dimethylaminopropylamido directed outward to the 3′ end of its neighboring DNA strand. The thiazole groups with the appropriate active functionalities (NH_2, NHCHO, or H) were introduced on the other side of this cross-linked bis(imidazole) dipyrrole moiety.

The alkane linkers C5 and C7 were introduced by the alkylation of imidazole with 1,5 dibromopentane and 1,7 dibromoheptane, respectively, and this process was successfully achieved by dropwise addition of the alkyl halides to the potassium salt of imidazole generated in situ from potassium and imidazole in refluxing tetrahydrofuran. The crude product was fractionally distilled to provide *N*,*N*′-bis(imidazolyl) pentane **8** and *N*,*N*′-bis-(imidazolyl) heptane **9**, respectively. The addition of alkyl halides to the reaction mixture just after the addition of the potassium metal to the solution of imidazole helped to shorten the time necessary for the metal to dissolve completely from several hours to one. This procedure avoided the use of a large volume of the solvent required to allow efficient stirring of the suspension. *N*,*N*′-bis-(imidazole)s **8** and **9** were then trichloroacetylated to afford **10** and **11**, respectively, and the same methodology[37] was used for the introduction of the trichloroacetyl group in *N*-methylpyrrole to give **1** (Scheme 1). The intermediates **10** and **11** were then nitrated with fuming nitric acid in acetic anhydride to give **12** and **13**, respectively. Similarly, **1** was nitrated to afford **2**, which was condensed with 3-dimethylaminopropylamine to give **3**. The coupling of the dinitro derivatives **12** and **13** with a amine, freshly prepared from **3**, went smoothly in 62% yield to give **14** and **15** (Scheme 4), respectively.

Having completed the common four-ring core intermediates **14** or **15**, it remained to introduce the thiazole moieties bearing the active functionalites on the other side of **14** and **15**. The required key starting material ethyl-2-amino-4-methylthiazole-5-carboxylate **4** was prepared following the reported procedure[38] (Scheme 2). Compound **4** was then converted into the corresponding acid **5** and was coupled with the freshly prepared amine of **14** or **15**. This coupling was catalyzed by DCC and HOBt to give the final products **16** and **17**, respectively. To introduce the *N*-formyl group into these aminothiazolated compounds **16** and **17**, we first tried to protect the amino group with Boc and then to treat this with excess of formic acid (which can remove the Boc and introduce the formyl group in case of pyrrole and imidazole). However, this procedure proved unsuccessful in thiazolated cases. Then we tried to treat **16** or **17** with formic acid in acetic anhydride to obtain direct formylation, but this method also failed. Finally, we successfully converted **5** into the *N*-formyl derivative **6** by treating it with formic acid and acetic anhydride before coupling it with the amine of **14** or **15**. This procedure worked satisfactorily to afford compounds **18** and **19**, respectively. To obtain a deaminated final product, we tried the reported[39] deamination procedure on compounds **16** and **17** and also on **5**, but,while this method failed to deaminate these particular compounds, these methods worked well in the case of aromatic amine deamination. Accordingly we synthesized 4-methyl-5-trichloroacetylthiazole **7** from 4-methylthiazole (Scheme 3) and coupled it with the freshly prepared amines of **14** or **15**, which afforded **20** and **21**, respectively, in good yields.

These thiazolated cross-linked oligopeptides **16**−**21** were examined for their DNA-binding characterstics and their comparative in vitro studies on the influence on DNA gyrase. Compounds were investigated for their influence on the DNA supercoiling and cleavage reaction catalyzed by gyrase from *S. noursei*. Inhibition of the supercoiling reaction of DNA gyrase by these cross-linked polyamides was comparable with that of the naturally occurring distamycin. All the six compounds have shown the beginning of inhibition at 0.1 μM, and the action of gyrase is completely inhibited at 1−2 μM. In contrast, distamycin dimer and the monomer polyamide concentrations are higher than 1 and 10 μM, respectively. Compound **17** has shown complete inhibition of the enzyme at 0.5 μM concentration, which is better than

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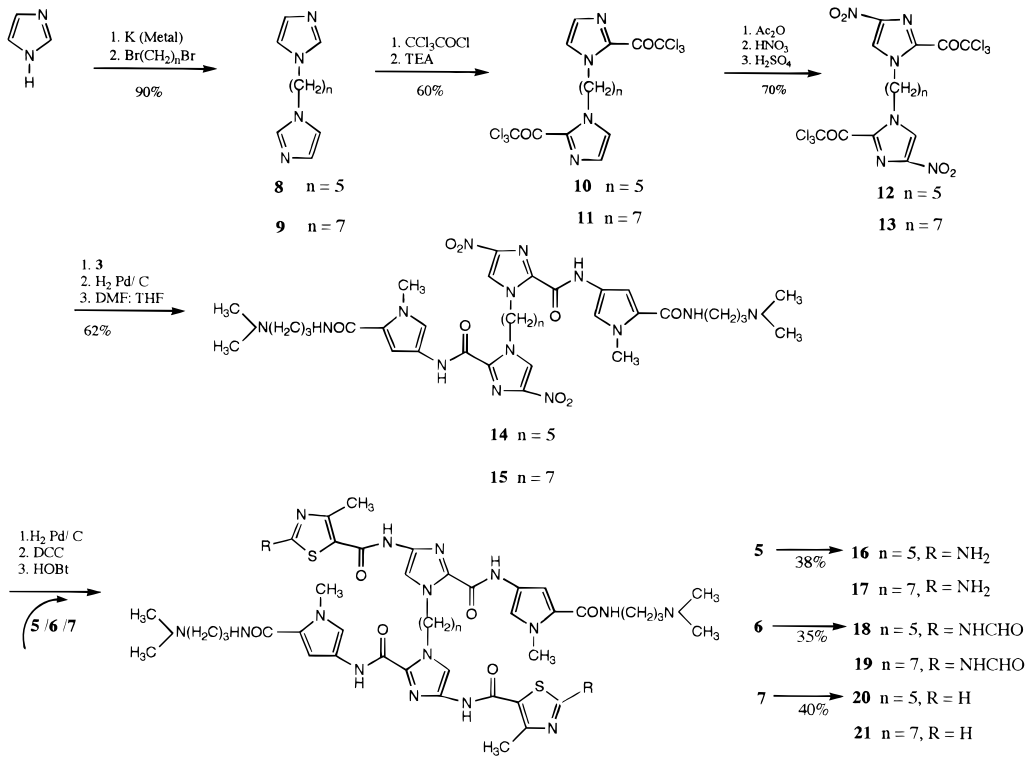
Scheme 4



that of the naturally occurring distamycin. The cross-linked dimers have both a higher sequence recognition capacity and inhibitory gyrase potency than do the corresponding monomers.[40]

Experimental Section

All chemicals used were of reagent grade. The reactions were carried out in anhydrous solvents. Anhydrous *N*,*N*-dimethylformamide (DMF), methanol (MeOH), *N*,*N*-diisopropylamine (DIEA), triethylamine (TEA), 1,3-dicyclohexylcarbodiimide (DCC), 1-hydroxybenzotriazole (HOBt), and 4-methylthiazole were purchased from Aldrich Chemical Co. and were used without any purification. Anhydrous tetrahydrofuran (THF) was distilled freshly over sodium/benzophenone at the time of reaction. The reactions were monitered by analytical thin-layer chromatography (TLC) using silica gel (60F-254 mesh: Merck) coated aluminum-backed plates. ^{1}H NMR spectra were recorded on a 300 MHz spectrometer, and the chemical shifts are reported in δ ppm with respect to tetramethylsilane as an internal standard. The spliting of resonance peaks are indicated as singlet (s), doublet (d), triplet (t), quartet (q), and multiplet (m). The values for the coupling constants (J) are expressed in hertz. Mass spectra and high-resolution mass spectra (HRMS) were done by electrospray. Melting points were determined on an electrothermal melting point apparatus and are uncorrected.

Compounds **1**−**5** were synthesized as reported in the literature.[37,38]

2-(Formylamino)-4-methylthiazole-5-carboxylic acid (6). To a solution of **5** (632 mg, 4 mmol) in 98% formic acid (8 mL) was added dropwise acetic anhydride (2.7 mL), and the reaction mixture was stirred for 18 h at room temperature. The solvent was evaporated under reduced pressure, and water (30 mL) was added to the resulting residue. The resulting mixture was extracted with 1:1 THF:ethyl acetate (3 × 30 mL). The combined organic extract was washed successively with 2% HCl (2 × 20 mL) and brine (2 × 20 mL) and then dried (Na_2SO_4). Evaporation of the solvent gave **6**, 350 mg (47% yield), mp 220 °C; ^{1}H NMR (DMSO-d_6) δ 2.50 (s, 3 H), 8.52 (s, 1H), 12.53 (br s,1H, exchanged with D_2O); MS (*m*/*z* rel. intensity) $C_6H_6N_2O_3S$ 186.046, found 186.046.

4-Methyl-5-trichloroacetylthiazole (7). 4-Methylthiazole (1.00 g, 10.00 mmol) was dissolved in dry dichloromethane (100 mL), and the solution was cooled to 0 °C. To this mixture trichloroacetyl chloride (2.04 g, 10.00 mmol) in dichloromethane was added dropwise over a period of 30 min under nitrogen. After complete addition the reaction mixture was brought to room temperature and stirred for 18 h. The solvent was removed under reduced pressure, and the crude product was purified by silica gel column chromatography, eluting by 3:1 hexane:ethyl acetate to give compound **7** as a colorless oil, yield 65%; ^{1}H NMR (DMSO-d_6) δ 2.65 (s, 3 H), 7.45 (s, 1H); MS (*m*/*z* rel. intensity) 244.905 (M^+), 126.001(100).

1,1′-(1,5-Pentamethylene)bis(imidazole) (8). To a solution of imidazole (5.00 g, 73 mmol) in anhydrous tetrahydrofuran (50 mL) was introduced pieces of potassium metal (2.90 g, 73 mmol) under nitrogen atmosphere. 1,5-Dibromopentane (7.5 g, 35 mmol) was added dropwise just after the addition of potassium metal over a period of 30 min, and the mixture was heated to reflux for 12 h. The reaction mixture was cooled to room temperature and filtered. The white solid residue was dissolved in water, and the solution was extracted with ether (3 × 100 mL). The combined organic fractions were concentrated in vacuo to give **8** as a colorless oil. Yield 90%; ^{1}H NMR ($CDCl_3$) δ 1.45 (m, 2 H), 1.95 (q, 4 H), 3.95 (t, J = 7.1 Hz, 4 H), 6.82 (s, 2 H), 7.10 (s, 2 H), 7.45 (s, 2 H); MS (*m*/*z* rel. intensity) 204.136 (M^+), 82.053 (100).

1,1′-(1,7-Heptamethylene)bis(imidazole) (9). The title compound was synthesized in a similar way as compound **8** using 1,7-dibromoheptane affording **9**, as a colorless oil, yield 93%; ^{1}H NMR ($CDCl_3$) δ 1.25 (m, 6 H), 1.70 (q, 4 H), 3.88 (t, J = 7.1 Hz, 4 H), 6.84 (s, 2 H), 7.00 (s, 2 H), 7.40 (s, 2 H); MS (*m*/*z* rel. intensity) 232.168 (M^+), 82.053 (100).

1,1′-(1,5-Pentamethylene)-2,2′-bis(trichloroacetyl)bis(imidazole) (10). Compound **8** (3.89 g, 1.94 mmol) was dissolved in anhydrous dichloromethane (150 mL), and trichloroacetyl chloride (7.80 g, 38.20 mmol) was added to this mixture dropwise over a period of 1 h under nitrogen at room

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temperature. Rapid evolution of gas was observed, and the resulting solution was stirred for 24 h at room temperature. The mixture was cooled to 0 °C, dry triethylamine (9.6 mL, 68.9 mmol) was added to this stirred mixture over a period of 30 min, and then the solution was concentrated in vacuo and the residue purified by silica gel column chromatography eluting with dichloromethane to give **10** as a viscous brown oil. Yield 68%; ^{1}H NMR ($CDCl_3$) δ 1.53 (m, 2 H), 2.10 (q, 4 H), 4.40 (t, J = 7.1 Hz, 4 H), 7.20 (s, 2 H), 7.38 (s, 2 H); MS (*m*/*z* rel. intensity) 495.918 (M^+), 377.013 (85), 55.050 (100).

1,1′-(1,7-Heptamethylene)-2,2′-bis(trichloroacetyl)bis-(imidazole) (11). Obtained in a similar procedure as a viscous dark yellow oil. Yield 67%; ^{1}H NMR ($CDCl_3$) δ 1.35 (m, 6 H), 1.80 (m, 4 H), 4.40 (t, J = 7.1 Hz, 4 H), 7.18 (s, 2 H), 7.38 (s, 2 H); MS (*m*/*z* rel intensity) 523.950 (M^+), 405.045 (85), 55.050 (100).

1,1′-(1,5-Pentamethylene)-2,2′-bis(trichloroacetyl)-4,4′-dinitrobis(imidazole) (12). A suspension of **10** (2.00 g, 4.04 mmol) in acetic anhydride (9.00 mL) and dichloromethane(35 mL) was cooled to −78 °C, and fuming nitric acid (0.65 mL) was added to it dropwise over a period of 30 min. This procedure was followed by the addition of concentrated sulfuric acid (0.4 mL). The brown mixture was brought to room temperature gradually and stirred for 3 h. The final reaction mixture was diluted with dichloromethane, neutralized with saturated sodium bicarbonate solution, extracted with dichloromethane (3 × 50 mL), dried over Na_2SO_4, and then filtered and concentrated to a brown foam. The impure product was purified by silica gel column chromatography eluting with ethyl acetate−hexane (1:2) to give **12** as pale yellow crystals. Yield 70%; mp 132 °C; ^{1}H NMR ($CDCl_3$) δ 1.52 (m, 2 H), 2.10 (q, 4 H), 4.45 (t, J = 7.1 Hz, 4 H), 8.01 (s, 2 H); MS (*m*/*z* rel. intensity) 585.913 (M^+), 465.411 (13.8), 55.050 (100).

1,1′-(1,7-Heptamethylene)-2,2′-bis(trichloroacetyl)-4,4′-dinitrobis(imidazole) (13). The title compound was obtained by a similar procedure as pale yellow crystals. Yield 70%; mp 120 °C; ^{1}H NMR ($CDCl_3$) δ 1.44 (m, 6 H), 1.92 (m, 4 H), 4.50 (t, J = 7.1 Hz, 4 H), 7.98 (s, 2 H); MS (*m*/*z* rel. intensity) 613.520 (M^+), 493.018 (13.8), 55.050 (100).

1,1′-(1,5-Pentamethylene)-2,2′bis[*N*-[(dimethylamino)-propyl]1-methyl-4-pyrrolyl]4,4′-dinitrobis(imidazole)-2,2′-carboxamide (14). Palladium charcol (10%, 430 mg) was added to *N*-[(dimethylamino)propyl]1-methyl-4-nitropyrrole-carboxamide **3** (869 mg, 3.40 mmol) in anhydrous DMF:MeOH (1:1, 10 mL). After being degassed with argon, the mixture was hydrogenated in a Parr shaker at 50 psi for 2 h. The catalyst was removed by filtration and washed thoroughly with methanol, and the combined filtrates were evaporated in vacuo. The residue was dried under high vacuum to remove traces of residual solvent. To this residue dry DMF was reintroduced, and the solution was cooled to 0 °C. A solution of **12** (1.00 g, 1.70 mmol) in DMF:THF (10 mL, 1:1 v/v) was added to the above solution under stirring over a duration of 30 min. The reaction mixture was brought to room temperature, and stirring was continued for 18 h. A TLC examination at this time showed that the reaction was complete. The solvent were evaporated in vacuo, and the impure product was purified by silica gel column chromatography. Eluting with CH_2Cl_2:MeOH:NH_4OH (80:20:1, v/v/v) gave the pure product **14** as pale yellow crystals. Yield 62%; mp 120 °C; 1H NMR (DMSO-d_6) δ 1.28 (m, 2 H), 1.57 (q, 2 H), 1.85 (q, 4 H), 2.15 (s, 12 H), 2.25 (t, J = 7.0 Hz, 4 H), 3.20 (q, J = 6.2 Hz, 4 H), 3.89 (s, 6 H), 4.41 (t, J = 7.2 Hz, 4 H). 6.95 (d, J = 1.7 Hz, 2 H), 7.29 (d, J = 1.7 Hz, 2 H), 8.15 (t, J = 5.5 Hz, 2 H), 8.65 (s, 2 H), 10.62 (s, 2 H); HRMS calcd for $C_{35}H_{51}N_{14}O_8$ 795.401, found 795.402.

1,1′-(1,7-Heptamethylene)-2,2′bis[*N*-[(dimethylamino)-propyl]1-methyl-4-pyrrolyl]4,4′-dinitrobis(imidazole)-2,2′-carboxamide (15). This compound was synthesized from **13** in a similar way as described for **14** as pale yellow crystals **15**, yield 62%; mp 101 °C; 1H NMR (DMSO-d_6) δ 1.30 (m, 6 H), 1.62 (q, 2 H), 1.90 (m, 4 H), 2.16 (s, 12 H), 2.28 (t, J = 7.0 Hz, 4 H), 3.18 (q, J = 6.8 Hz,4 H), 3.80 (s, 6 H), 4.48 (t, J = 7.0 Hz, 4 H). 7.00 (d, J = 1.6 Hz, 2 H), 7.25 (d, J = 1.6 Hz, 2 H), 8.15 (t, J = 6.0 Hz, 2 H), 8.68 (s, 2 H), 10.68 (s, 2 H); HRMS calcd for $C_{37}H_{55}N_{14}O_8$ 823.432, found 823.432.

1,1′-(1,5-Pentamethylene)bis[*N*-[5-[[[(3,3-dimethyl-amino)propyl]carbonyl]-1-methyl-4-pyrrol-3-yl]-4-[[[2-amino-4-methylthiazol-5-yl]carbonyl]amino]-imidazole]-2-carboxamide (16). Palladium charcoal (10%, 500 mg) was added to a solution of **14** (1.00 g, 1.25 mmol) in anhydrous DMF:MeOH (1:1 v/v; 20 mL). Degassing of the solution was done by argon, and then the mixture was hydrogenated in a Parr shaker for 2 h at 50 psi. The catalyst was then removed by filtration, and the filtrate was washed several times with methanol. The combined filtrates were evaporated under high vacuum to remove traces of the solvent. The residual reduced product was redissolved in anhydrous DMF (10 mL), and compound **5** (396.00 mg, 2.51 mmol) and HOBt (474.91 mg, 3.51 mmol) were added to it with constant stirring. A solution of DCC (723.04 mg, 3.51 mmol) in anhydrous DMF (3 mL) was added slowly to this stirred mixture, and the stirring was continued at 25 °C for an additional 18 h. The reaction mixture was filtered, and the solvent was removed in vacuo. The impure product was purified on silica gel column chromatography, eluting with CH_2Cl_2:MeOH:NH_4OH (80:20:4,v/v/v) to give compound **16** as a white solid.Yield 39%; mp 154−156 °C; ^{1}H NMR (DMSO-d_6) δ 1.29 (m, 2 H), 1.60 (q, 2 H), 1.85 (q, 4 H), 2.15 (s, 12 H), 2.26 (t, J = 7.0 Hz, 4 H),2.40 (s, 6 H), 3.20 (q, J = 6.2 Hz, 4 H), 3.88 (s, 6 H), 4.41 (t, J = 7.2 Hz, 4 H), 7.00 (d, J = 1.7 Hz, 2 H), 7.27 (d, J = 1.7 Hz, 2 H), 8.15 (t, J = 5.5 Hz, 2 H), 8.60 (s, 2 H), 9.60 (s, 2 H), 10.64 (s, 2 H); HRMS calcd for $C_{45}H_{63}N_{18}O_6S_2$ 1015.461, found 1015.462.

1,1′-(1,7-Heptamethylene)bis[*N*-[5-[[[(3,3-dimethyl-amino)propyl]carbonyl]-1-methyl-4-pyrrol-3-yl]-4-[[[2-amino-4-methylthiazol-5-yl]carbonyl]amino]-imidazole]-2-carboxamide (17). The title compound was prepared in a similar way as a white solid, yield 38%; mp 150−153 °C; ^{1}H NMR (DMSO-d_6) δ 1.26 (m, 6 H), 1.60 (q, J = 7.0 Hz, 4 H), 1.75 (m, 4 H), 2.15 (s, 12 H), 2.25 (t, J = 7.0 Hz, 4 H),2.40 (s, 6 H),3.20 (t, J = 6.2 Hz, 4 H), 3.80 (s, 6 H), 4.40 (t, J = 7.2 Hz, 4 H). 6.90 (d, J = 1.6 Hz, 2 H), 7.20 (d, J = 1.6 Hz, 2 H),7.46 (s, 4 H), 7.50 (s, 2 H), 8.10 (t, J = 6.0 Hz, 2 H), 9.64 (s, 2 H), 10.06 (s, 2 H); HRMS calcd for $C_{47}H_{67}N_{18}O_6S_2$ 1043.493, found 1043.491.

1,1′-(1,5-Pentamethylene)bis[*N*-[5-[[[(3,3-dimethyl-amino)propyl]carbonyl]-1-methyl-4-pyrrol-3-yl]-4-[[[2-formylamino-4-methylthiazol-5-yl]carbonyl]amino]-imidazole]-2-carboxamide (18). Compound **14** (1.00 g, 1.25 mmol) was reduced by palladium charcoal (500 mg) in a similar way as described for **16**. The reduced product was redissolved in anhydrous DMF (10 mL), and **6** (465.00 mg, 2.50 mmol), and HOBt (472.50 mg, 3.50 mmol) were added to it at room temperature under nitrogen. A solution of DCC (721.00 mg, 3.50 mmol) in anhydrous DMF (3 mL) was added slowly to this stirred mixture, and stirring was continued at 25 °C for an additional 18 h. The reaction mixture was filtered, and the solvent was removed in vacuo. The impure product was purified on silica gel column chromatography, eluting with CH_2Cl_2:MeOH:NH_4OH (80:20:3,v/v/v) to give compound **18** as a yellow-colored powder; yield 35%; mp 165−167 °C; ^{1}H NMR (DMSO-d_6) δ 1.30 (m, 2 H), 1.85 (q, 4 H), 2.20 (s, 12 H), 2.25 (t, J = 7.0 Hz, 4 H),2.45 (s, 6 H),3.20 (q, J = 6.2 Hz, 4 H), 3.88 (s, 6 H), 4.45 (t, J = 7.2 Hz, 4 H). 6.95 (d, J = 1.7 Hz, 2 H), 7.25 (d, J = 1.7 Hz, 2 H), 7.59 (s, 2 H, exchanged with D_2O), 8.15 (t, J = 5.5 Hz, 2 H), 8.55 (s, 2 H), 10.05 (s, 2 H, exchanged with D_2O), 10.42 (s, 2 H, exchanged with D_2O); HRMS calcd for $C_{47}H_{63}N_{18}O_8S_2$ 1071.451, found 1071.451.

1,1′-(1,7-Heptamethylene)bis[*N*-[5-[[[(3,3-dimethyl-amino)propyl]carbonyl]-1-methyl-4-pyrrol-3-yl]-4-[[[2-formylamino-4-methylthiazol-5-yl]carbonyl]amino]-imidazole]-2-carboxamide (19). The title compound was obtained in a similar way as a yellow powder, yield 35%; mp 174−176 °C; ^{1}H NMR (DMSO-d_6) δ 1.25 (m, 6 H), 1.65 (q, 4 H), 1.80 (m, 4 H), 2.20 (s, 12 H), 2.30 (t, J = 7.0 Hz, 4 H), 2.45 (s, 6 H),3.20 (t, J = 6.2 Hz, 4 H), 3.85 (s, 6 H), 4.45 (t, J = 7.0 Hz, 4 H). 6.95 (d, J = 1.6 Hz, 2 H), 7.25 (d, J = 1.6 Hz, 2 H), 7.60 (s, 2 H, exchanged with D_2O) 8.15 (t, J = 6.0 Hz, 2 H),

8.56 (s, 2 H), 10.15 (s, 2 H, exchanged with D_2O), 10.30 (s, 2H, exchanged with D_2O); HRMS calcd for $C_{49}H_{67}N_{18}O_8S_2$ 1099.483, found 1099.484.

1,1′-(1,5-Pentamethylene)bis[*N*-[5-[[[(3,3-dimethylamino)propyl]carbonyl]-1-methyl-4-pyrrol-3-yl]-4-[[[4-methylthiazol-5-yl]carbonyl]amino]imidazole]-2-carboxamide (20). Compound **14** (1.20 g, 1.00 mmol) was reduced by palladium charcoal (10%, 600 mg) in a similar way as described for **16**. The reduced product was redissolved in anhydrous DMF (10 mL); **7** (490.80 mg, 2.00 mmol) in DMF: THF (1:1,v/v,20 mL) was added to the above reduced product dropwise at room temperature under nitrogen over a period of 30 min, and the stirring was continued at 25 °C for additional 18 h. The solvent was removed under reduced pressure. The impure product was purified on silica gel column chromatography, eluting with CH_2Cl_2:MeOH:NH_4OH (80:20:4,v/v/v) to give compound **20** as yellow powder, yield 42%; mp 150−152 °C; ^{1}H NMR (DMSO-d_6) δ 1.30 (m, 2 H), 1.85 (q, 4 H), 2.20 (s, 12 H), 2.25 (t, J = 7.0 Hz 4 H), 2.45 (s, 6 H), 3.20 (q, J = 6.2 Hz, 4 H), 3.80 (s, 6 H), 4.45 (t, J = 7.2 Hz, 4 H). 6.95 (d, J = 1.7 Hz, 2 H), 7.26 (d, J = 1.7 Hz, 2 H), 7.65 (s, 2 H), 7.75 (s, 2 H) 10.10 (s, 2 H, exchanged with D_2O), 10.30 (s, 2 H, exchanged with D_2O); HRMS calcd for $C_{45}H_{61}N_{16}O_6S_2$ 985.440, found 985.440.

1,1′-(1,7-Heptamethylene)bis[*N*-[5-[[[(3,3-dimethylamino)propyl]carbonyl]-1-methyl-4-pyrrol-3-yl]-4-[[[4-methylthiazol-5-yl]carbonyl]amino]imidazole]-2-carboxamide (21). The title compound was obtained in a similar way as a yellow powder, yield 40%; mp 163−165 °C; ^{1}H NMR (DMSO-d_6) δ 1.39 (m, 6 H), 1.65 (q, 4 H), 1.75 (m, 4 H), 2.20 (s, 12 H), 2.35 (t, J = 7.0 Hz, 4 H), 2.55 (s, 6 H), 3.19 (t, J = 6.2 Hz, 4 H), 3.79 (s, 6 H), 4.25 (t, J = 7.0 Hz, 4 H). 6.95 (d, J = 1.6 Hz, 2 H), 7.25 (d, J = 1.6 Hz, 2 H), 7.61 (s, 2 H,), 7.72 (s, 2 H), 8.15 (t, J = 6.0 Hz, 2 H), 10.21 (bs, 2 H, exchanged with D_2O), 10.30 (s, 2H, exchanged with D_2O); HRMS calcd for $C_{47}H_{65}N_{16}O_6S_2$ 1013.471, found 1013.471.

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Supporting Information Available: ^{1}H NMR and HRMS spectra of compounds **15**, **17**, **19**, and **21**. This material is available free of charge via the Internet at http://pubs.acs.org.

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