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Linear and Cyclic Amides with a Thiophene Backbone: Ultrasound-Promoted Synthesis and Crystal Structures

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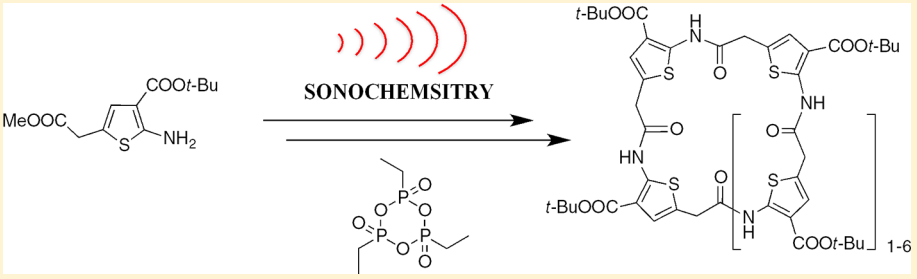
Supporting Information



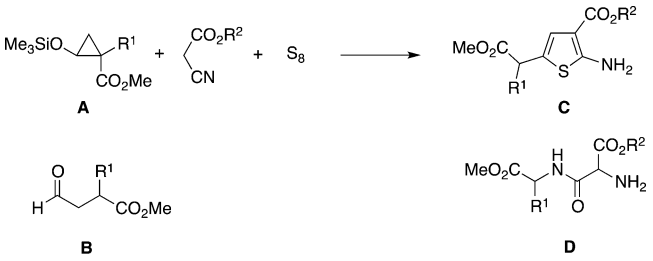
ABSTRACT: A full synthetic study of linear and cyclic thiophene oligoamides has been carried out. The combination of an ultrasonic technique to diminish the intramolecular backfolding of longer oligoamide chains, therefore enhancing the accessibility of the carboxylic acid, and T3P as coupling reagent led to shorter reaction time and higher yields for both linear and cyclic oligoamides. By controlling the degree of dilution, macrocyclic amides with different sizes can selectively be prepared. Different crystal structures of cyclic thiophene oligoamides were also analyzed.

INTRODUCTION

Due to their high stability, amide bonds have been chosen by nature as a linkage for the construction of its basic structures, *e.g.*, proteins and glycoconjugates to amino acids.[1] In the field of peptidomimetics, numerous research groups have dedicated their full focus to the development of methodologies for mimicking the vital processes of nature. The discovery of biological activities of some natural products built up by amide bonds, *e.g.* those extracted from marine plants,[2,3] shows the importance of these investigations. For example, cyclopeptides containing five-membered heterocycles such as the antitumor trunkamide A[4−6] and the cytotoxic ascidiacyclamide[2,7,8] (both from the Lissoclinum class of cyclic peptides) play an undeniably important role in nature, as they are found in many of these marine organisms. The alternating sequence of heterocyclic rings and amino acid units which characterize these structures has led to speculation that the metabolites may have a role to play *in vivo* as host agents for metal transport, and/or that metals may act as templates in their biological assembly from the constitutive amino acids and heterocyclic rings.[9] The importance of this research area has encouraged us to search for optimal methods for the synthesis of oligopeptides containing heterocycles in their backbones.

Within our group, the synthesis of 5-aminothiophenecarboxylic acids via the three-component Gewald reaction[10−15] has been explored[16,17] using 2-siloxycyclopropanecarboxylates **A** as aldehyde equivalents **B** (Scheme 1).[18−35] This process gave direct access to the unnatural amino acids **C** containing a thiophene backbone, which can be considered as isosteres to the natural dipeptides **D**.

Scheme 1. Synthesis of 5-Aminothiophenecarboxylic Acids



The synthetic pathway described above affords new opportunities to prepare a variety of linear and cyclic oligoamides containing thiophene moieties. These oligopeptides can be compared to the better known *meta*-substituted[36,37] (or *para*-substituted)[38−40] phenyl peptidomimetics, but the heterocyclic structure may result in different properties over a wide area of interest, *e.g.*, these oligoamides could act as ligands for metal cations, and the extra coordination might enhance or inhibit some biological activities. The more conformationally rigid structure due to the aryl groups could be an advantage for efficient and flexible synthetic access to both linear and cyclic peptide analogues; in the case of a cyclopeptide, the rigidity could lead to a better fit for the host−guest interaction.

For peptide bond formation, the traditional synthetic methodology (under conventional stirring) involves the

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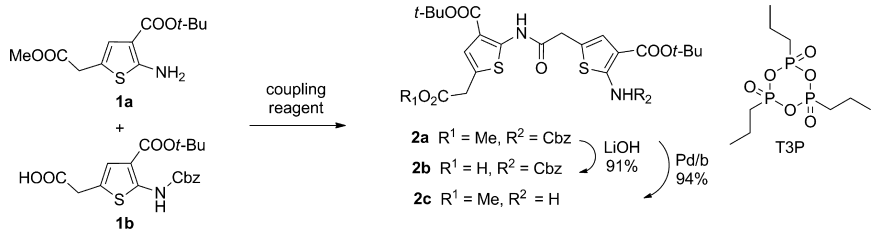
Scheme 2. Synthesis of Linear Thienyl Amides 2a−c with Different Coupling Reagents



reaction of carboxylic acids with amines in the presence of peptide coupling reagents.[41] New techniques such as microwave-assisted peptide couplings have been widely reported in recent decades, both in solution[42−45] and on solid phase.[46,47] But another phenomenon has drawn our attention, namely ultrasonication. Sonochemistry using energy from cavitation bubbles to promote chemical reactions has already been known for more than 70 years.[48] It has regained more attention in recent decades due to the availability of reliable ultrasonic equipment and offers some advantages over conventional procedures, such as shorter reaction times and higher yields. To our knowledge, only a few examples have been reported that employ ultrasound activation in peptide synthesis.[49−53] A second aspect concerns propylphosphonic anhydride T3P, which has gained our interest because it offers several advantages over other traditional coupling reagents, *e.g.*, higher yields, low toxicity, easy workup, and short reaction times.[54−57]

In the previous report, we described the preparation of linear and cyclic thiophene oligoamides with the backbone formed between an aliphatic carboxylic acid derived from **C** and the notoriously unreactive 5-amino group of the thiophene ring, using EDCI·HCl (see footnote in Table 1) as coupling reagent (Scheme 2).[17] The described synthesis gave easy access to linear oligomers, though a clear trend of decreasing yields was observed for higher homologues (from 81% to 61%). In addition, the macrocyclization proved to be inefficient using the described procedure (1−18% yield). In the current work, we report a detailed study of the synthesis of linear and cyclic thiophene oligoamides, employing a variety of coupling reagents under both conventional reaction conditions and ultrasonication. This report discloses that ultrasound activation combined with T3P as the peptide coupling reagent is the best choice for our systems, leading to strong improvements and allowing for the synthesis of compounds previously regarded as "difficult".

RESULTS AND DISCUSSION

To construct thiophene oligoamides via a sequence of peptide couplings, monomers with unprotected amino and carboxylic acid moieties, respectively **1a** and **1b**, were prepared according to the literature procedures.[17] Different coupling reagents were employed to carry out the peptide coupling, affording the dimer **2a** (Scheme 2, Table 1). No desired product **2a** was observed when BOP or TFFH was used in the presence of DMAP (entries 1 and 2). By replacing DMAP with DIPEA, 68% of **2a** could be isolated with TFFH as coupling reagent (entry 3) and 32% with HATU (entry 4). These observations clearly show that not only does the coupling reaction play an important role but the type of amine base also seems to considerably influence the efficiency of the amide bond formation. The best yields were obtained with PyBroP/DIPEA, EDCI·HCl, and T3P/Et_3N (up to 87%, entries 5−7).

Table 1. Reaction Conditions for the Synthesis of Linear Oligoamides

entry	acid	amine	coupling reagent[a]	reaction conditions	product	yield (%)
1	**1b**	**1a**	BOP, DMAP	24 h	**2a**	–
2	**1b**	**1a**	TFFH, DMAP	3 d	**2a**	–
3	**1b**	**1a**	TFFH, DIPEA	3 d	**2a**	68
4	**1b**	**1a**	HATU, DIPEA	24 h	**2a**	32
5	**1b**	**1a**	EDCI·HCl	24 h	**2a**	81
6	**1b**	**1a**	PyBroP, DIPEA	24 h	**2a**	87
7	**1b**	**1a**	T3P, Et_3N	24 h	**2a**	80
8	**2b**	**1a**	EDCI·HCl	24 h	**3a**	69
9	**2b**	**1a**	PyBroP, 2,4,6-collidine	24 h	**3a**	57
10	**2b**	**1a**	T3P, Et_3N	24 h	**3a**	33
11	**2b**	**2c**	EDCI·HCl	2 d	**4a**	61
12	**2b**	**2c**	PyBroP, 2,4,6-collidine	24 h	**4a**	59
13	**2b**	**2c**	T3P, Et_3N	24 h	**4a**	38
14	**2b**	**1a**	T3P, Et_3N	30 min[b]	**3a**	77
15	**2b**	**2c**	T3P, Et_3N	60 min[b]	**4a**	75
16	**3b**	**1a**	T3P, Et_3N	20 min[b]	**4a**	11
17	**3b**	**2c**	T3P, Et_3N	60 min[b]	**5**	38
18	**1b**	**1a**	T3P, Et_3N	5 min[b]	**2a**	98
19	**1b**	**2c**	T3P, Et_3N	20 min[b]	**3a**	quant.
20	**1b**	**3c**	T3P, Et_3N	15 min[b]	**4a**	92
21	**1b**	**4c**	T3P, Et_3N	6 min[b]	**5**	94
22	**1b**	**2c**	EDCI·HCl	2 h[b]	**3a**	68

[a]EDCI·HCl = *N*-ethyl-*N*′-(3-dimethylaminopropyl)carbodiimide hydrochloride, BOP = benzotriazol-1-yloxytris(dimethylamino)-phosphonium hexafluorophosphate, TFFH = *N*,*N*,*N*′,*N*′-tetramethylfluoroformamidinium hexafluorophosphate, DMAP = 4-(dimethylamino)pyridine, DIPEA = *N*,*N*-diisopropylethylamine, HATU = 2-(1*H*-7-azabenzotriazol-1-yl)-1,1,3,3-tetramethyluronium hexafluorophosphate, PyBroP = bromotripyrrolidinophosphonium hexafluorophosphate. [b]Ultrasonic activation.

To investigate the effect of the different coupling reagents on extended chains, the amino and carboxylic acid moieties of dimer **2a** were deprotected selectively either by saponification with lithium hydroxide or by hydrogenolysis employing palladium (black) to obtain **2b** and **2c**, respectively (Scheme 2).[17] These building blocks were then used for the chain elongation. When the synthesis of **3a** was carried out starting from monomer **1a** and dimer **2b**, the yield dropped for all coupling reagents (Scheme 3, Table 1: entries 8−10). This trend continues further with the preparation of the linear tetramer **4a** by peptide coupling, starting from **2b** and **2c** (entries 11−13). When the yields of the di-, tri-, and tetramers prepared by different coupling reagents were compared, one could clearly observe that the yields decreased for higher oligomers, from 87% to 59% for PyBroP (entries 6, 9, 12) and 81% to 61% for EDCI·HCl (entries 5, 8, 11), respectively. This

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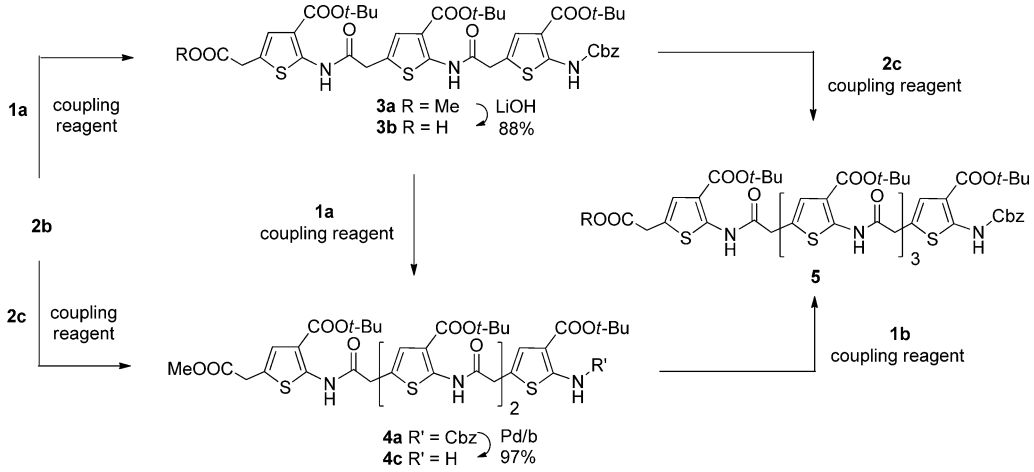
Scheme 3. Synthesis of Trimer 3a, Tetramer 4a, and Pentamer 5

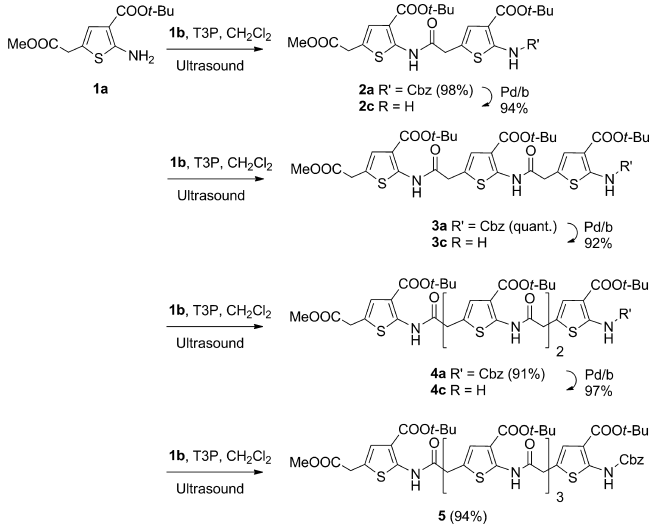


effect was observed more distinctly with the use of T3P where the yield dropped from 80% to 33%, although most of the starting material was recovered. The unreacted precursors did not seem to contribute further to the desired coupling, even when the reaction time was extended or additional reagent T3P was added.

This observation might be a result of the possible conformation of the longer chain compounds. The active carboxylic acid moiety could be shielded by intramolecular folding and thus be less accessible to the coupling reagent, hence resulting in less efficient couplings. To support this hypothesis, ultrasonic activation was applied during the reaction to overcome this proposed intramolecular folding. The decrease in yield for longer amide chains is most apparent in the case of T3P as coupling reagent (Table 1, entries 7, 10, and 13). Therefore, T3P was chosen as the coupling reagent for a further optimization process. Under ultrasonification and using T3P as coupling reagent, the yields of trimer **3a** and tetramer **4a** were enhanced to 77% and 75%, respectively (entries 14 and 15). As expected, the effect of ultrasonic activation is less distinct when the prolongation was extended to higher oligomer chains (tetramer **4a** and pentamer **5**), starting from free carboxylic trimeric precursor **3b**. The yields dropped to 11% and 38%, respectively (entries 16 and 17). Entries 14−17 clarify that the size of the oligomer with the free amino group does not impact the yield much; however, the length of the precursor with the free carboxylic acid does appear to have a significant effect.

These observations seem to confirm our hypothesis that deactivation of the carboxylic group for amide coupling is due to intramolecular backfolding of the longer chain. Therefore, we changed our strategy and carried out the extension of the oligomer chain by successive peptide couplings with only monomer **1b** containing the unprotected carboxylic moiety (Scheme 4). Under the same reaction conditions used previously, the yields were enhanced in all cases up to quantitative (entries 18−21). The reaction times were also reduced from 1−2 days (without sonification) to 5−20 min. Ultrasonication not only appears to unfold the oligomeric chain, leading to a better accessibility of the functional groups, but it also accelerates the coupling reaction itself. The latter can be explained by the high cavitation energy from ultrasonication, which easily overcomes the activation energy barrier. The concentration plays an important role during sonification. The

Scheme 4. Chain Elongation of Oligomeric Amides 3a, 4a, and 5 with Monomer 1b Bearing the Free Carboxyl Group



T3P-mediated peptide coupling occurred much faster (only 5−20 min) when high concentrations were applied while in the case of high dilution, ultrasonification did not speed up the reaction compared to conventional stirring.

Next we questioned whether the improvement of yields was only a result of the sonification effect and the choice of a suitable monomer (free carboxyl *vs* amino groups). To have a better understanding, diamide **3a** was prepared under the optimized procedure above, but with EDCI·HCl as coupling reagent. The reaction resulted in the desired product **3a** with 68% yield (entry 22). Longer reaction times did not lead to the completion of the transformation. Therefore, we can conclude that the procedure using T3P as coupling reagent combined with ultrasonification gives the optimal reaction conditions for the synthesis of thiophene oligoamides. This optimized procedure for amide coupling has been applied to synthesize oligoamides containing more conventional amino amides. The obtained results are very promising but out of the scope of this work and will be reported in the near future.

The high occurrence of macrocyclic oligoamides in nature encouraged our search for optimal cyclization conditions for linear amide chains or starting from single monomers.[58−60] With the tri- and tetramer chains in hand, a range of

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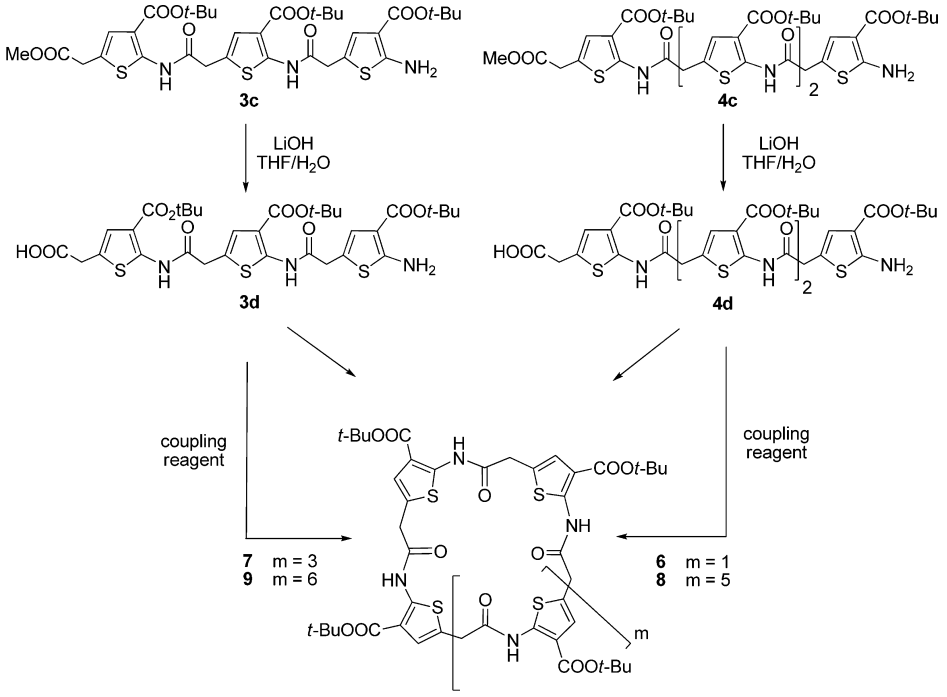
Scheme 5. Synthesis of Macrocyclic Amides 6−9



macrocycles can potentially be prepared by intramolecular cyclization. Similar to the above couplings, the macrocyclizations were subjected to an optimization process with different coupling reagents and reaction conditions. As reported in our preliminary study, EDCI·HCl-mediated macrocyclization of tetramer **4d** at a concentration of 0.004 mol/L gave after three days 18% of the desired cyclic tetramer **6** with traces of cyclic octamer **8** (Scheme 5, Table 2, entry 1).[17] When PyBroP with DIPEA was employed as coupling reagent, no cyclic tetramer **6** was obtained (entry 2), and only in the presence of 2,4,6-collidine could 3% of octamer **8** be isolated (entry 3). The combination of TFFH and DIPEA gave a low yield for **6** (12%), but no formation of **8** was observed (entry 4), while TFFH with 2,4,6-collidine led to the formation of both macrocycles **6** (3%) and **8** (3%, entry 5). With 2,4,6-collidine as base, dimerization seemed to occur faster than the cyclization leading to a higher yield of final cyclic octamer **8**. We then used T3P as coupling reagent with and without ultrasonication. When the mixture of **4d** and T3P was added dropwise to the stirring solution of Et_3N over one day followed by conventional stirring for an additional three days, 31% of cyclic tetramer **6** and 16% of octamer **8** were obtained (entry 6). This protocol created *in situ* a highly diluted solution leading to the expected considerably higher yield of the smaller macrocycle. By ultrasound activation of the entire mixture of precursor, T3P, and Et_3N, the reaction time for both intermolecular and intramolecular amide interaction was decreased, and very much to our pleasure the yields of **6** and **8** were strongly enhanced to 37% and 50%, respectively (entry 7). Starting from **4d**, the combination of the *pseudo* high dilution and ultrasonication influenced the yields to the advantage of the cyclic tetramer **6** (46%) over the octamer **8** (21%). The high dilution and fast reaction activation by ultrasound apparently favored cyclization of the linear oligomers before they could dimerize.

While in the case of tetrapeptide **4d** TFFH-mediated cyclization gave 12% of the cyclic derivative, no cyclic tri-, hexa-, or nonamer was observed during the cyclization of linear trimer **3d** (Scheme 5, Table 2, entry 9). Even in a highly diluted reaction mixture, the EDCI·HCl activation of **3d** did not lead to intramolecular macrocyclization. Only 1% of the cyclic hexamer

Table 2. Reaction Conditions for Macrocyclization

entry	linear oligomer	reagents[a]	concn (mol/L)	time	product (yield)
1[17]	**4d**	EDCI·HCl	0.004	3 d	**6** (18%) **8** (traces)
2	**4d**	PyBroP, DIPEA	0.004	5 d	**6** (0%) **8** (0%)
3	**4d**	PyBroP, 2,4,6-collidine	0.004	3 d	**6** (0%) **8** (3%)
4	**4d**	TFFH, DIPEA	0.004	4 d	**6** (12%) **8** (0%)
5	**4d**	TFFH, 2,4,6-collidine	0.004	3 d	**6** (3%) **8** (3%)
6	**4d**	T3P, Et_3N	≤0.0002[b]	1 d + 3 d	**6** (31%) **8** (16%)
7	**4d**	T3P, Et_3N	0.00025[c]	2 h	**6** (37%) **8** (50%)
8	**4d**	T3P, Et_3N	≤0.0009[d]	2 h + 2 h	**6** (46%) **8** (21%)
9	**3d**	TFFH, DIPEA	0.004	5 d	**7** (0%) **9** (0%)
10[17]	**3d**	EDCI·HCl	0.004	3 d	**7** (1%) **9** (0%)
11	**3d**	T3P, Et_3N	≤0.0004[b]	4 d + 1 d	**7** (27%) **9** (0%)
12	**3d**	T3P, Et_3N	0.0008[d]	2 h + 2 h	**7** (33%) **9** (6%)
13	**3d**	T3P, Et_3N	0.027[c]	1 h (gel)	**7** (26%) **9** (32%)

[a]See footnote in Table 1 for abbreviations. [b]Dropping of activated linear oligomers into a stirring Et_3N solution in CH_2Cl_2 followed by stirring at rt. [c]Sonification of the mixture. [d]Dropwise addition of activated linear oligomers in Et_3N solution in CH_2Cl_2 under ultrasound activation followed by additional sonification.

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7 was obtained (entry 10).[17] The yield of 7 was increased to 27% when the slow addition method was applied in the presence of T3P under conventional stirring (entry 11). For the first time a cyclic nonamer **9** was isolated with 6% yield when we combined the dropwise addition protocol with ultrasonification, and 33% of the hexacyclic peptide 7 was also isolated (entry 12). When the concentration was raised to 0.027 mol/L, gel formation took place after 1 h of sonification. A higher concentration of the reaction mixture led to a more efficient formation of nonamer **9**, resulting in a yield of 32% with 26% of the hexamer 7 (entry 13). These observations confirm the trend as described above. With higher concentration, the reaction occurred faster and was more selective for larger macrocycles.

CRYSTALLOGRAPHIC DATA

In our previous work, the crystal structure of tetrameric amide **6** was reported as a molecule with a central core close to a square (angles of 84.9° and 95.1°), in which the sp^3-hybridized carbon atoms C5−C11−C5i−C11i form the corners.[17] The cavity within the macrocycles was occupied by one hexane molecule, employed as solvent for crystallization.

Two different solvent systems were applied to crystallize hexameric amide 7, namely slow diffusion of hexane into the solution of 7 in CH_2Cl_2 and slow evaporation of 1,2-dichloroethane as solvent. Compared to macrocycle **6**, the crystal structure of the hexameric amide 7 obtained from slow evaporation of 1,2-dichloroethane showed a nearly chairlike structure with a rectangular void with the angles of 87.3° and 93.7° (Figure 1). Taking the sp^3-hybridized carbon atoms C5, C11, C5i, and C11i for its four corners, the cavity had side lengths of 7.4 (C5−C11) and 14.1 Å (C5−C11i). The chair structure was derived from the direction in which the four *tert*-butoxycarbonyl substituents are oriented. In pairs, they are aimed in opposite directions (Figure 1). Interestingly, the cavity of the macrocycle was not occupied by a solvent molecule but by the opposed *tert*-butoxycarbonyl groups connected to the C14 and C14i carbons (Figure 1). Due to sterical hindrance, these ester groups did not lie in the same plane but above and below the rectangle plane with an angle of 12.8° to 18.5°.

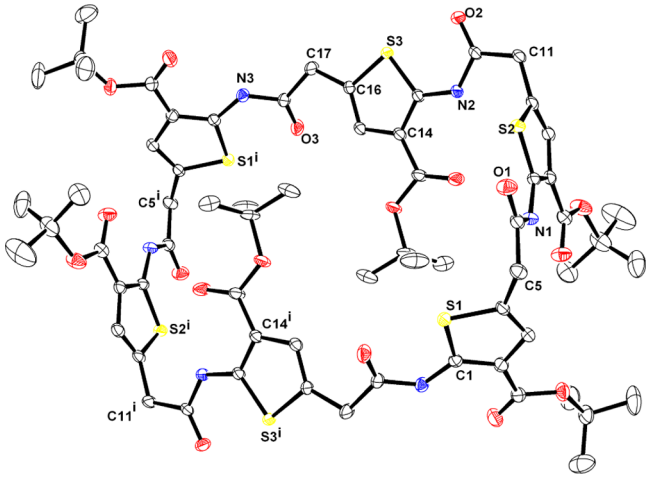

Figure 1. X-ray structure of hexameric amide 7 obtained from slow evaporation of 1,2-dichloroethane (top view).

As mentioned above, two small differences were observed between the two crystal structures of 7 obtained by different solvent systems. With the first solvent system (hexane−dichloromethane), two molecules of CH_2Cl_2 were incorporated within the cavity, whereas 1,2-dichloroethane was too large and only found around the molecule. The distortion of the inner *tert*-butoxycarbonyl group out of the cavity plane created enough space for CH_2Cl_2 to fill the opposite sites of the cavity. As shown in Figure 2, a possible hydrogen bond can be formed between the hydrogens of dichloromethane and the sulfur atoms S3 and S3i of the macrocycle.

In the dichloromethane and 1,2-dichoroethane solvates of compound 7 exist further voids of 560 and 438 $Å^3$, respectively, which are filled by disordered solvent molecules. The electron density of these molecules was taken into account using the program SQEEZE as integrated in the PLATON program package. The positions of these voids in the unit cell are depicted in Figure 3 using ball and stick and space-filling models.

CONCLUSION

An efficient synthetic protocol has been developed for the preparation of linear and cyclic oligoamides containing thiophene moieties. Via a sequence of peptide couplings with monomeric amino acids containing a free carboxylic acid moiety, linear peptide chains were obtained with very high to quantitative yields. The combination of T3P as coupling reagent and ultrasonification proved to be the optimal reaction conditions for a fast and high-yielding amide coupling. This optimized procedure was applied to prepare cyclic amides. The yields of the macrocycles with different sizes were selectively raised by varying the concentration of the reaction mixture. As expected, high dilution led to good yields of smaller macrocycles, *e.g.*, cyclic tetra- and hexamers, while increased concentrations selectively gave higher quantities of cyclic octa- and nonamers. Crystal structures of macrocyclic hexamer 7 revealed a chairlike solid-state structure with the central *tert*-butoxycarbonyl groups filling the rectangular cavity.

EXPERIMENTAL SECTION

General. ^1H and ^{13}C NMR spectra were recorded on commercial 250, 400, and 700 MHz instruments in $CDCl_3$ solution, and chemical shifts (δ) are reported in parts per million (ppm) referenced to tetramethylsilane or the internal (NMR) solvent signals. Detailed NMR peak assignments were obtained by analysis of DEPT, HSQC, and COSY NMR spectra. The high resolution mass spectra were obtained with an ESI-TOF spectrometer. Silica gel (0.040−0.063 mm) was used for column chromatography. Melting points are not corrected. X-ray crystallography: Single crystals for the X-ray diffraction experiment were selected using a microscope and mounted on the top of a glass fiber. Crystallographic data were collected using a diffractometer using Mo K_α radiation (λ = 0.71073 Å, graphite monochromator) at 133 K. Ultrasound bath with 35 kHz, 100% power, was used for syntheses.

The compounds **1a**, **1b**, **2b**, **2c**, **3c**, **3d**, **4c**, and **4d** have been prepared according to the literature procedure.[16,17]

General Procedure for T3P-Mediated Peptide Couplings under Ultrasonication (Scheme 4, Table 1; entries 18−21). To a solution of amine (1 equiv) in CH_2Cl_2 were added carboxylic acid **1b** (1.2 equiv), T3P (2 equiv), and triethylamine (2 equiv). The mixture was activated in the ultrasound bath at rt (5−20 min) under Ar atmosphere, and the completion was followed by TLC. The crude solution was washed with water (three times) and brine. After drying of the organic layer with Na_2SO_4, the solvent was evaporated under reduced pressure. Pure oligomers were obtained after purification by column chromatography (silica gel).

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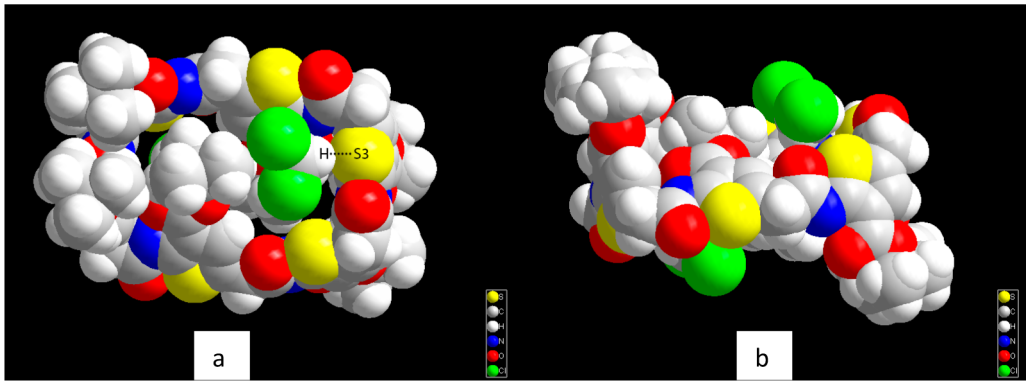

Figure 2. Top view (a) and side view (b) of macrocycle **7** containing two CH_2Cl_2 molecules in its cavity.

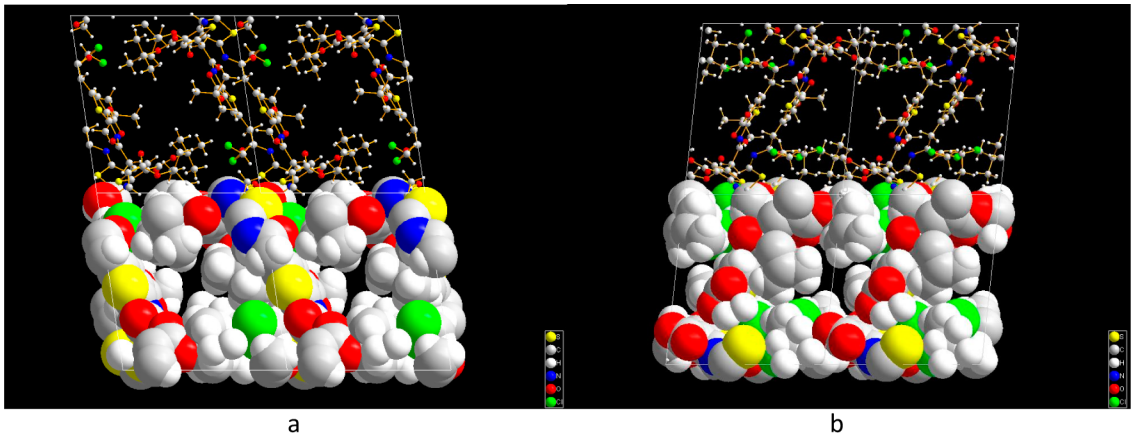

Figure 3. Packing of macrocycle **7** crystallized from CH_2Cl_2/hexane (a) and 1,2-dichloroethane (b).

Dimer 2a. According to general procedure, 60 mg of **1a** (0.22 mmol) and 103 mg of **1b** (0.26 mmol) in CH_2Cl_2 (5 mL) afforded after 5 min sonification 143 mg of dimer **2a** (143 mg, 98%); eluent 15% EtOAc in hexane; spectroscopic and physical properties agree with previously published data.[17]

Trimer 3a. According to general procedure, 100 mg of **2c** (0.20 mmol) and 92 mg of **1b** (0.24 mmol) in CH_2Cl_2 (5 mL) afforded after 20 min sonification quantitative yield of trimer **3a** (173 mg); eluent 30% EtOAc in hexane; spectroscopic and physical properties agree with previously published data.[17]

Tetramer 4a. According to general procedure, 107 mg of **3c** (0.14 mmol) and 92 mg of **1b** (0.17 mmol) in CH_2Cl_2 (5 mL) afforded after 15 min sonification tetramer **4a** (147 mg, 92%); eluent 10% EtOAc in CH_2Cl_2; spectroscopic and physical properties agree with previously published data.[17]

Pentamer 5. According to general procedure, 100 mg of **4c** (0.10 mmol) and 48 mg of **1b** (0.12 mmol) in CH_2Cl_2 (5 mL) afforded after 6 min sonification pentamer **5** (128 mg, 94%) as colorless solid; mp 190−192 °C; eluent 10% EtOAc in CH_2Cl_2; HRMS (ESI+) calcd for $C_{64}H_{75}N_5O_{18}S_5K$ 1400.3342; found 1400.3316; ^{1}H NMR (400 MHz, $CDCl_3$, 25 °C, TMS): δ = 11.26 (s, 1H, NH); 11.23 (s, 2H, NH); 11.19 (s, 1H, NH); 10.34 (s, 1H, NH); 7.42−7.34 (m, 5H, Ph); 7.08−7.07 (m, 4H, thioph); 6.94 (s, 1H, thioph); 5.24 (s, 2H, CH_2Ph); 3.84 (s, 8H, CH_2); 3.71 (s, 3H, OMe); 3.68 (s, 2H, CH_2COO); 1.54, 1.52, 1.51 (3s, 9H, 27H, 9H, *t*-Bu); ^{13}C NMR (101 MHz, $CDCl_3$, 25 °C, TMS): δ = 170.8 (CO_2Me); 166.64, 166.61, 166.57 (CONH); 164.88, 164.86, 164.76 (CO_2*t*-Bu); 153.0 (Cbz); 149.9, 147.6, 147.3 (SCN); 135.5 (Ph); 128.8, 128.7, 128.6 (Ph); 125.3, 125.0, 124.5 (CH_{thioph}); 124.4, 124.33, 124.31 ($SCCH_2$); 123.7 (CH_{thioph}); 123.1 ($SCCH_2$); 114.97, 114.96, 114.5, 113.5 (CCO_2*t*-Bu); 81.89, 81.87, 81.82, 81.7 (*t*-Bu); 68.1 (CH_2Ph); 52.5 (OMe); 37.1, 37.0, 35.0 (CH_2); 28.42, 28.37 (*t*-Bu).

General Saponification Procedure of Methyl Esters. To a solution of the ester (1 equiv) in THF/H_2O (3:2) was added LiOH (3−4 equiv). The mixture was stirred at rt, and the completion was followed by TLC. After acidification by HCl (1 M) to pH 2, EtOAc was added, and the solution was washed with water and brine and dried with Na_2SO_4. The solvent was evaporated under reduced pressure. Purification by column chromatography (silica gel, 9:1 CH_2Cl_2:MeOH) afforded pure carboxylic acid as a colorless solid.

Deprotected Trimer 3b. According to general saponification procedure, 400 mg of **3a** (0.45 mmol) and 33 mg of LiOH (1.38 mmol) in a mixture of THF/H_2O (16 mL) afforded carboxylic acid **3b** (345 mg, 88%) as colorless solid; mp 164−166 °C; HRMS (ESI+) calcd for $C_{41}H_{47}N_3O_{12}S_3Na$ 892.2220; found 892.2194; ^{1}H NMR (400 MHz, $CDCl_3$, 25 °C, TMS): δ = 11.26 (s, 1H, NH); 11.20 (s, 1H, NH); 10.34 (s, 1H, NH); 7.41−7.33 (m, 5H, Ph); 7.08 (s, 1H, thioph); 7.07 (s, 1H, thioph); 6.96 (s, 1H, thioph); 5.23 (s, 2H, CH_2Ph); 3.85 (s, 4H, CH_2); 3.73 (s, 2H, CH_2COO); 1.542, 1.515, 1.511 (3s, each 9H, *t*-Bu); ^{13}C NMR (101 MHz, $CDCl_3$, 25 °C, TMS): δ = 166.73, 166.70 (CONH); 164.9, 164.8 (CO_2*t*-Bu); 153.0 (Cbz); 149.9, 147.6, 147.4 (SCN); 135.5, 128.7, 128.66, 128.62 (Ph), 125.3, 124.4, 124.2 (thiophene), 124.6, 124.0, 123.1 ($SCCH_2$); 115.0, 114.6, 113.5 (CCO_2*t*-Bu); 81.9 (*t*-Bu); 68.1 (CH_2Ph); 37.05, 37.04, 37.00 (CH_2); 28.41, 28.37, 28.35 (*t*-Bu).

Procedure for Macrocyclizations (Scheme 5, Table 2). *Cyclic Octamer 6.* To an activated solution of Et_3N (20 μL, 0.14 mmol) in CH_2Cl_2 (70 mL) in an ultrasound bath was added dropwise, over 2 h, a mixture of linear tetramer **4d** (69 mg, 0.071 mmol) and T3P (45 mg, 0.14 mmol). The solution was sonificated for an additional 2 h. The crude solution was washed with water and brine. After drying the organic layer with Na_2SO_4, the solvent was evaporated under reduced pressure. Pure macrocycle **6** (31 mg, 46%) was obtained as a yellow solid after purification by column chromatography (silica gel, eluent 1% MeOH in CH_2Cl_2). The spectroscopic and physical properties of

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cyclic tetramer **6** agree with previously published data.[17] A 14 mg amount of **8** (21%) was obtained during this experiment.

*Cyclic Octamer **8***. A solution of tetramer **4d** (50 mg, 0.051 mmol), T3P (32 mg, 0.10 mmol), and Et_3N (14 μL) in CH_2Cl_2 (200 mL) was activated in the ultrasound bath at rt for 2 h. The crude solution was washed with water and brine. After drying the organic layer with Na_2SO_4, the solvent was evaporated under reduced pressure. Pure macrocycle **8** (25 mg, 50%) was obtained as a yellow solid after purification by column chromatography (silica gel, eluent 4:1 EtOAc/hexane). Mp 190−195 °C; HRMS (ESI+) calcd for $C_{88}H_{104}N_8O_{24}S_8Na$ 1935.4821; found 1935.4806; ^1H NMR (250 MHz, $CDCl_3$, 25 °C, TMS): δ = 11.22 (s, 8H, NH); 7.05 (s, 8H, thioph); 3.83 (s, 16H, CH_2), 1.47 (s, 72H, *t*-Bu); ^{13}C NMR (176 MHz, $CDCl_3$, 25 °C, TMS): δ = 166.6, 164.9 (CON); 147.7 (SCN); 124.7 ($SCCH_2$); 124.3 (CH_{thioph}); 115.0, 110.14 (CCO_2t-Bu); 82.0 (*t*-Bu); 42.0 (CH_2); 28.4 (*t*-Bu). A 18 mg amount of **6** (37%) was obtained during this experiment.

*Cyclic Hexamer **7***. To an activated solution of Et_3N (19 μL, 0.135 mmol) in CH_2Cl_2 (70 mL) in an ultrasound bath was added dropwise, over 2 h, a mixture of linear tetramer **3d** (50 mg, 0.068 mmol) and T3P (43 mg, 0.135 mmol). The solution was sonificated for additional 2 h. The crude solution was washed with water and brine. After drying the organic layer with Na_2SO_4, the solvent was evaporated under reduced pressure. Pure macrocycle **7** (16 mg, 33%) was obtained as a yellow solid after purification by column chromatography (silica gel, eluent 1% MeOH in CH_2Cl_2). The spectroscopic and physical properties of cyclic tetramer **7** agree with previously published data.[17] A 3 mg amount of **9** (6%) was obtained during this experiment. Two different solvent systems were employed to obtain crystals from **7**, namely by slow diffusion of hexane into the solution of **7** in CH_2Cl_2 and by slow evaporation of 1,2-dichloroethane as solvent.

*Cyclic Nonamer **9***. A solution of tetramer **3d** (100 mg, 0.136 mmol), T3P (87 mg, 0.273 mmol), and Et_3N (38 μL, 0.273 mmol) in CH_2Cl_2 (5 mL) was activated in the ultrasound bath at rt for 1 h. The crude solution was washed with water and brine. After drying the organic layer with Na_2SO_4, the solvent was evaporated under reduced pressure. Pure macrocycle **9** (31 mg, 32%) was obtained as an orange-red solid after purification by column chromatography (silica gel, eluent 4:1 EtOAc/hexane). Mp 135−137 °C; HRMS (ESI^+) calcd for $C_{99}H_{117}N_9O_{27}S_9Na$ 2175.5477; found 2175.5482; ^1H NMR (250 MHz, $CDCl_3$, 25 °C, TMS): δ = 11.22 (s, 9H, NH); 7.06 (s, 9H, thioph); 3.83 (s, 18H, CH_2); 1.49 (s, 81H, *t*-Bu); ^{13}C NMR (176 MHz, $CDCl_3$, 25 °C, TMS): δ = 166.6, 164.9 (CON); 147.7 (SCN); 124.7 ($SCCH_2$); 124.3 (CH_{thioph}); 115.0, 110.14 (CCO_2t-Bu); 82.0 (*t*-Bu); 37.0 (CH_2); 28.4 (*t*-Bu). A 25 mg amount of **7** (26%) was obtained during this experiment.

■ ASSOCIATED CONTENT

Ⓢ Supporting Information

Crystallographic data of macrocycle **7** and ^1H and ^{13}C NMR spectra of new compounds **3b**, **5**, **8**, and **9**. This material is available free of charge via the Internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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