Self-Folding Molecules: A Well Defined, Stable Loop Formed by a Carboxylate−Guanidinium Zwitterion in DMSO

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Two zwitterions **1a**,**b** have been synthesized, in which a carboxylate group is attached via a flexible alkyl chain of different length (butylene and ethylene, respectively) to a guanidiniocarbonyl pyrrole cation moiety. For **1b**, no signs for either an intra- or intermolecular association between these two groups in polar solution (DMSO) could be found. In contrast to this, the ^{1}H NMR spectrum of **1a** shows clear evidence for a strong interaction between the carboxylate and the guanidiniocarbonyl pyrrole cation. According to variable-temperature and concentration-dependent NMR studies, this interaction stems from an intramolecular complexation. It was shown by ROESY and H/D-solvent exchange experiments that **1a**, even in DMSO, folds into a well-defined intramolecular loop conformation held together by multiple weak interactions.

Introduction

The molecular properties and thus the function of a given molecule depend not only on its chemical constitution but also on its specific conformation. The various conformations accessible for a large and flexible molecule are often associated with different molecular properties. However, the basic principles that control such folding processes are still poorly understood.[1,2] Therefore, the design of molecules that fold into a predictable, specific conformation can help learn more about these self-organization processes and design molecules with new, tailored functions and properties (e.g., designer proteins, new catalysts, and drug candidates). However, a lot of systems reported so far[3] either are rather rigid and therefore structurally biased toward a particular conformation[3e,4] or adopt their specific conformation only in organic solvents of low polarity (e.g., chloroform),[5] because their folding is mainly determined by hydrogen bonds,[6] which are easily disrupted when the polarity of the solvent increases.[3b,7] Here, the synthesis and structural characterization of a conformationally flexible zwitterion **1a** is reported. Without any built-in structural bias, this zwitterion folds, even in a highly polar solvent such as DMSO, into a well-defined loop conformation due to strong intramolecular interactions between two self-complementary groups.

It was shown recently that guanidiniocarbonyl pyrroles are excellent receptors for carboxylates even in aqueous solvents with association constants in the order of $K \approx 10^3$ mol^{-1} in 40% water−DMSO (v/v).[8,9] By linking a pyrrole carboxylate moiety to such a guanidiniocarbonyl pyrrole receptor unit through a flexible spacer, molecules capable self-organizing into higher ordered structures are obtained. Depending on the length and flexibility of the spacer, either monomolecular self-folding molecules or supramolecular oligomers and polymers should result (Scheme 1).[1,10,11]

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(1) (a) Lehn, J.-M. *Supramolecular Chemistry*; *Concepts and Perspectives*; VCH: Weinheim, 1995. (b) Whitesides, G. M.; Mathias, J. P.; Seto, C. T. *Science* **1991**, *254*, 1312−1319. (c) Philp, D.; Stoddart, J. F. *Angew. Chem., Int. Ed. Engl.* **1999**, *38*, 1154−1196.

(2) Gellman, S. H. *Chem. Rev.* **1997**, *97*, 1231−1232.

(3) For selected reviews and articles on this area see: (a) Nowick, J. S.; Tsai, J. H.; Bui, Q.-C. D.; Maitra, S. *J. Am. Chem. Soc.* **1999**, *121*, 8409−8410. (b) Nowick, J. S. *Acc. Chem. Res.* **1999**, *32*, 287−296. (c) Gellman, S. H. *Acc. Chem. Res.* **1998**, *31*, 173−180. (d) Nowick, J. S.; Smith, E. M.; Pairish, M. *Chem. Soc. Rev.* **1996**, 401−415. (e) Schneider, J. P.; Kelly, J. W. *Chem. Rev.* **1995**, *95*, 2169−2187.

(4) (a) Feigel, M. *J. Am. Chem. Soc.* **1986**, *108*, 181−182. (b) Wagner, G.; Feigel, M. *Tetrahedron* **1993**, *49*, 10831−10842. (c) Brandmeier, V.; Sauer, W. H. B.; Feigel, M. *Helv. Chim. Acta* **1994**, *77*, 70−85.

(5) Recently, short artificial peptides have been shown to form stable helices and sheets in aqueous solvents, e.g.: (a) Apella, D. H.; Barchi, J. J., Jr.; Durell, S. R.; Gellman, S. H. *J. Am. Chem. Soc.* **1999**, *121*, 2309−2310. (b) Seebach, D.; Abele, S.; Gademann, K.; Jaun, B. *Angew. Chem., Int. Ed. Engl.* **1999**, *38*, 1595−1597. (c) See also: Blanco, F.; Ramirez-Alvarado, M.; Serrano, L. *Curr. Opin. Struct. Biol.* **1998**, *8*, 107−111.

(6) Another approach is persued by Hoffmann et al., who use gauche interactions within 2,4-dimethyl pentane units to induce specific conformations in acyclic hydrocarbons in unpolar solvents: Schopfer, U.; Stahl, M.; Brandl, T.; Hoffmann, R. W. *Angew. Chem., Int. Ed. Engl.* **1997**, *36*, 1745−1747 and references therein.

(7) For solvent effects on the strengths of H-bonds in complexes, see, e.g.: (a) Horvath, P.; Gergely, A.; Noszal, B. *J. Chem. Soc., Perkin Trans. 1* **1996**, 1419−1422. (b) Kelly, T. R.; Kim, M. H. *J. Am. Chem. Soc.* **1994**, *116*, 7072−7080. (c) Ariga, K.; Anslyn, E. V. *J. Org. Chem.* **1992**, *57*, 417−419.

(8) (a) Schmuck, C. *Chem. Commun.* **1999**, 843−844. (b) Schmuck, C. *Chem. Eur. J.* **2000**, *4*, 709−718.

(9) Selected examples of other synthetic receptors that function in polar solvents: (a) Hossain, A.; Schneider, H.-J. *J. Am. Chem. Soc.* **1998**, *120*, 11208−11209. (b) Davies, M.; Bonnat, M.; Gullier, F.; Kilburn, J. D.; Bradley, M. *J. Org. Chem.* **1998**, *63*, 8696−8703. (c) Berger, M.; Schmidtchen, F. P. *Angew. Chem., Int. Ed. Engl.* **1998**, *37*, 2694−2696. (d) Jagessar, R. C.; Shang, M.; Scheldt, W. R.; Burns, D. H. *J. Am. Chem. Soc.* **1998**, *120*, 11684−11692. (e) Niikura, K.; Metzger, A.; Anslyn, E. V. *J. Am. Chem. Soc.* **1998**, *120*, 8533−8534. (f) Peczuh, M. W.; Hamilton, A. D.; Sanchez-Qesada, J.; deMendoza, J.; Haak, T.; Giralt, E. *J. Am. Chem. Soc.* **1997**, *119*, 9327−9328. (g) Buhlmann, P.; Nishizawa, S.; Xiao, K. P.; Umewaza, Y. *Tetrahedron* **1997**, *53*, 1647−1654. (h) Berger, M.; Schmidtchen, F. P. *J. Am. Chem. Soc.* **1996**, *118*, 8947−8948. (i) LaBrenz, S. R.; Kelly, J. W. *J. Am. Chem. Soc.* **1995**, *117*, 1655−1656. (j) Albert, J. S.; Goodman, M. S.; Hamilton, A. D. *J. Am. Chem. Soc.* **1995**, *117*, 1143−1144. (k) Kato, Y.; Conn, M. M.; Rebek, J. Jr. *J. Am. Chem. Soc.* **1994**, *116*, 3279−3284. (l) Schiessl, P.; Schmidtchen, F. P. *Tetrahedron Lett.* **1993**, 2449−2452. (m) Fan, E.; Anslyn, E. V. *J. Am. Chem. Soc.* **1993**, *115*, 10042−10055.

(10) For a sampling of hydrogen-bonded self-assembled dimers in nonpolar solvents, see, e.g.: (a) Kang, J.; Rebek, J., Jr. *Nature* **1997**, *385*, 50−52. (b) Kang, J.; Rebek, J., Jr. *Nature* **1996**, *382*, 239−241. (c) Meissner, R. S.; de Mendoza, J.; Rebek, J., Jr. *Science* **1995**, *270*, 1485−1488. (d) For a comprehensive review article, see: Conn, M. M.; Rebek, J., Jr. *Chem. Rev.* **1997**, *97*, 1647−1668.

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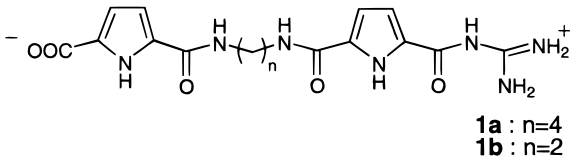
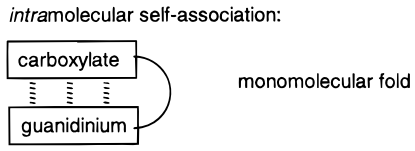
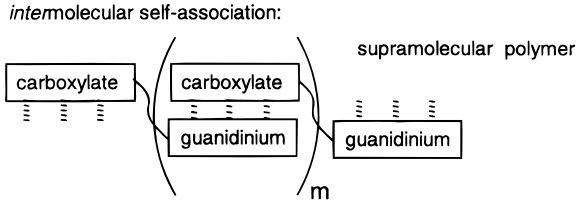
Scheme 1. Schematic Representation of Possible Self-Assembled Structures for Zwitterions Such as 1







Results and Discussion

Synthesis. The C4-linked zwitterion **1a** ($n = 4$) along with the C2-linked control compound **1b** ($n = 2$) was synthesized according to Scheme 2. The acid **2**[12] is reacted with the mono tBoc-protected diamines **3**[13] in methylene chloride with DCC as the coupling reagent and catalytic amounts of DMAP. After deprotection with TFA, the amine **5** is reacted with the acyl chloride **6**,[8] prepared from the corresponding acid by reaction with oxalyl chloride in the presence of catalytic amounts of DMF in methylene chloride. The resulting methyl ester **7** can then be hydrolyzed with LiOH in aqueous THF to give the desired zwitterions **1**. Alternatively, the diamide **10** can be prepared from the acyl chloride **8** and the diamines **9**.[14] Guanidinylation[15] of compound **10** with guanidinium chloride in sodium methoxide gives the same mono ester **7** as before. However, the yields following this second route are lower (especially the guanidinylation step is low yielding), and the workup and purification of the products is more difficult so that the first synthetic alternative, though being slightly longer, is preferred.

Structural Characterization. According to molecular modeling calculations, zwitterion **1a** with its C4 linker should be capable of folding intramolecularly into a loop conformation, whereas in zwitterion **1b** the C2 linker is much too short to allow such a self-folding. And indeed, the ^{1}H NMR spectra of these two compounds in DMSO are remarkably different. Zwitterion **1b** shows the "normal" spectrum expected for a simple guanidiniocarbonyl pyrrole (Figure 1; all peaks were clearly assigned with the help of 2D NMR experiments): A broad signal for the two CH_2 groups at $\delta = 3.4$, multiplets for the four pyrrole CHs around $\delta = 6.7$, a broad signal for the four guanidinium protons $NH^{f,g}$ at $\delta = 7.3$, two signals for the amide NHs at $\delta = 8.4$ (NH^b) and 8.6 (NH^c), respectively, and signals for the two pyrrole protons NH^a and NH^d and the guanidinium amide NH^e around $\delta = 11.7$. These shifts are identical with similar, more simple guanidiniocarbonyl pyrroles, which lack any possible complementary binding group.[8,16] It is well-known that any complexation between a guanidiniocarbonyl group and a carboxylate group causes significant complexation-induced shift changes in the ^{1}H NMR spectrum.[8,16,17] Therefore, the absence of any such shift changes in the spectrum of **1b** clearly shows that at least under these experimental conditions (millimolar solution in DMSO) no complexation, neither inter- nor intramolecular of the carboxylate and the guanidiniocarbonyl pyrrole, occurs.

In contrast to this, the spectrum of zwitterion **1a** is completely different: The signal for the four guanidiniocarbonyl pyrrole protons splits into two signals (two protons each), at $\delta = 8.0$ (NH^g) and 9.9 (NH^f), respectively. The two amide NHs give signals at $\delta = 8.6$ (NH^b) and 9.5 (NH^c). One of the two pyrrole NHs is shifted downfield by 1.4 ppm relative to **1b** and resonates at $\delta = 13.2$ (NH^d), the other resonating at $\delta = 11.8$ (NH^a). And perhaps most strikingly, the guanidinio amide NH^e gives a signal at $\delta = 14.8$ that corresponds to a downfield shift of more than 3 ppm (relative to **1b**). This ^{1}H NMR pattern is exactly the same as already known from intermolecular complexes between simple carboxylates and guanidiniocarbonyl pyrrole receptors.[8,19] In these complexes, the guanidinium cation forms an ion pair with the carboxylate that is simultaneously hydrogen bonded by the pyrrole NH and the various amide NHs. The two guanidinium protons H^g, which do not form hydrogen bonds to the carboxylate, resonate more or less at the same field as in **1b**, whereas the other two guanidinium protons H^f, the amide NH^e and the pyrrole NH^d, all of which take part in the complexation process, give signals at much lower field relative to the corresponding signals in uncomplexed guanidiniocarbonyl pyrrole compounds (e.g., the spectrum of **1b**), indicating strong hydrogen bonding interactions. This clearly shows that even in DMSO complexation of the carboxylate by the guanidiniocarbonyl pyrrole moiety takes place.[8]

In principle, such a complexation can occur inter- or intramolecularly. However, the principal binding interactions between the carboxylate and the guanidiniocarbonyl

(11) For selected examples of the formation of supramolecular oligomers in nonpolar solvents, see, e.g.: (a) Yamaguchi, N.; Gibson, H. W. *Angew. Chem., Int. Ed. Engl.* **1999**, *38*, 143−147. (b) Ashton, P. R.; Parson, I. W.; Raymo, F. M.; Stoddart, J. F.; White, A. J. P.; Williams, D. J.; Wolf, R. *Angew. Chem., Int. Ed. Engl.* **1998**, *37*, 1913−1916. (c) Yamaguchi, N.; Nagvekar, D. S.; Gibson, H. W. *Angew. Chem., Int. Ed. Engl.* **1998**, *37*, 2361−2364.

(12) Barker, P.; Gendler, P.; Rapoport, H. *J. Org. Chem.* **1978**, *43*, 4849−4853 and references therein.

(13) Pons, J.-F.; Fauchère, J.-L.; Lamaty, F.; Molla, A.; Lazaro, R. *Eur. J. Org. Chem.* **1998**, *3*, 853−859.

(14) Shama, S. A.; Tran, T. L. *J. Chem. Educ.* **1978**, *55*, 816.

(15) Bicking, J. B.; Robb, C. M.; Kwong, S. F.; Cragoe, E. J. *J. Med. Chem.* **1967**, *10*, 598−602.

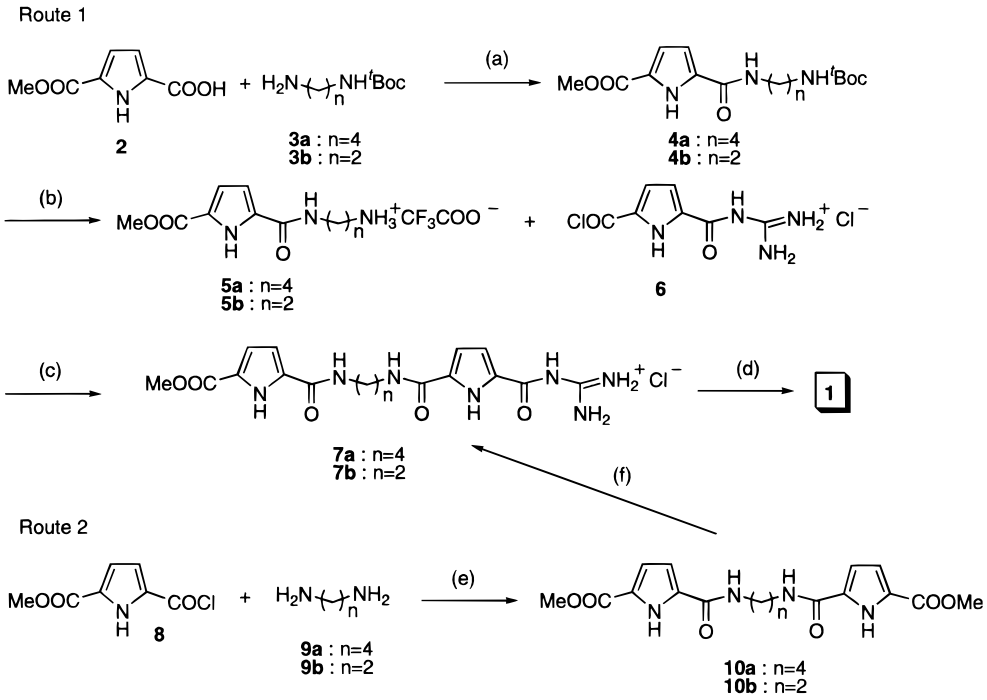
(16) Dixon, R. D.; Geib, S. J.; Hamilton, A. D. *J. Am. Chem. Soc.* **1992**, *114*, 365−366.

(17) Recent review articles on the binding of carboxylates by artificial receptors including guanidinium-based systems: (a) Schmidtchen, F. P.; Berger, M. *Chem. Rev.* **1997**, *97*, 1609−1646. (b) Schmidtchen, F. P. *Artificial Anion Hosts: Concepts for Structure and Guest Binding* in *Supramolecular Chemistry of Anions*; Bianchi, A., Bowman-James, K., García-España, E., Eds.; Wiley-VCH: New York, 1997; pp 79−146. (c) Atwood, J. L.; Steed, J. W. Structural and Topological Aspects of Anion Coordination. In *Supramolecular Chemistry of Anions*; Bianchi, A., Bowman-James, K., García-España, E., Eds.; Wiley-VCH: New York, 1997; pp 147−216. (d) Seel, C.; Galán, A.; de Mendoza, J. *Top. Curr. Chem.* **1995**, *175*, 101−132.

(18) The ^{1}H NMR spectrum of a saturated solution of **1b** (ca. 50 mM) in DMSO at 20 °C shows two but still overlapping signals for the guanidinium NHs (in contrast to more dilute solutions which only show one signal, see Figure 1). This might be a first hint to the formation of oligomers in such highly concentrated solutions.

(19) Schmuck, C. *Eur. J. Org. Chem.* **1999**, 2397−2403.

Scheme 2. Synthesis of Zwitterions 1a and 1b[a]



[a] Key: (a) DCC, DMAP (cat.), THF/CH_2Cl_2 (1:1), 0 °C then rt, 70−80%; (b) TFA, CH_2Cl_2, 0 °C, quant; (c) NEt_3, THF, 0 °C, 12 h, rt, 30−45%; (d) LiOH, THF/water (4:1), rt; (e) C_6H_6, reflux, 24 h, 35−45%; (f) guanidinium chloride, MeONa, MeOH, reflux, 24 h, 18−25%.

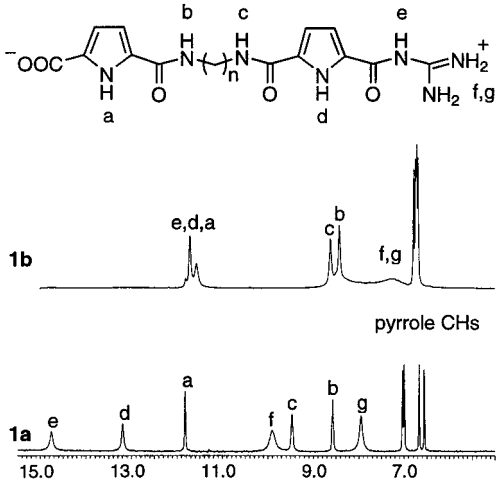


Figure 1. Downfield part of the ^{1}H NMR spectra (300 MHz, DMSO-d_6, 1 mM, 298 K) of zwitterions **1a** and **1b** clearly showing the complexation of the carboxylate group by the guanidiniocarbonyl pyrrole moiety in the case of **1a** in contrast to **1b**.

pyrrole group are the same in **1a** and **1b**, but only in **1a** and not in **1b** an interaction between the carboxylate and the guanidiniocarbonyl pyrrole group can be seen. If the above-described features in the NMR spectrum of **1a** were due to intermolecular oligomerization, both **1a** and **1b** should show similar spectra, which is not the case. Any intermolecular complexation is possible in both compounds in exactly the same manner as far as the binding interactions are concerned and should even be more favorable in **1b** due to its less flexible nature. Therefore, the complexation-induced shift changes in the NMR spectrum of **1a** cannot be caused by dimerization or oligomerization but rather indicate that **1a** forms a specifically folded monomer, in which the carboxylate

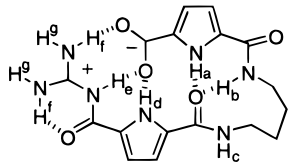


Figure 2. Proposed structure for zwitterion **1a** that folds into a loop stabilized by multiple intramolecular weak interactions (shown here in form of dashed lines).

group on the one end of the molecule interacts with the guanidiniocarbonyl pyrrole moiety on the other.

Furthermore, such an intramolecular self-folding is concentration independent, whereas with increasing dilution the formation of intermolecular self-associated oligomers or polymers becomes less and less favorable. As the ^{1}H NMR splitting pattern and shifts observed in the spectrum of **1a** do not change in the concentration range from 100 mM to as low as 0.01 mM oligomer formation can most likely be ruled out. The C2-linked zwitterion **1b** cannot self-fold intramolecularly due to geometric reasons and could only oligomerize intermolecularly.[18] Accordingly, in contrast to **1a** the zwitterion **1b** is much less soluble. Whereas **1a** can be recrystallized from water and is readily soluble in methanol, **1b** is nearly completely insoluble in these solvents. Even in DMSO **1b** is only very poorly soluble and immediately precipitates from the solution upon addition of chloroform, whereas **1a** readily forms millimolar solutions even in 10% DMSO−$CHCl_3$.

Therefore, **1b** probably forms oligomers in the solid state that dissociate in polar solutions, whereas **1a** most likely folds monomolecularly into a loop conformation (Figure 2). In this conformation, the carboxylate is bound by the guanidiniocarbonyl pyrrole unit in the same way as already known from intermolecular complexes between carboxylates and guanidiniocarbonyl pyrrole re-

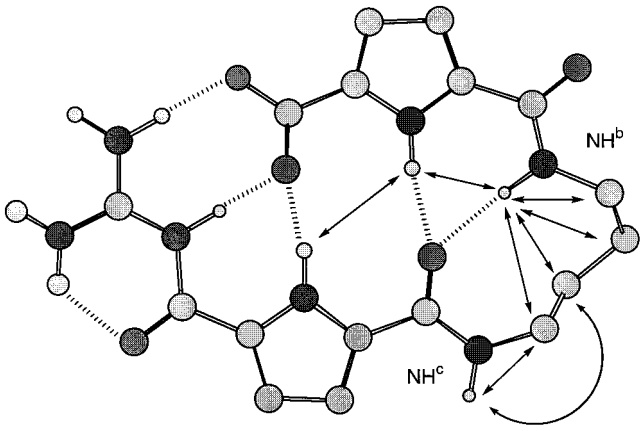


Figure 3. Key NOEs (arrows) detected by ROESY studies (500 MHz, DMSO-d_6, rt) of zwitterion **1a** (dashed lines indicate intramolecular hydrogen bonds; hydrogens attached to carbon atoms are omitted for clarity).

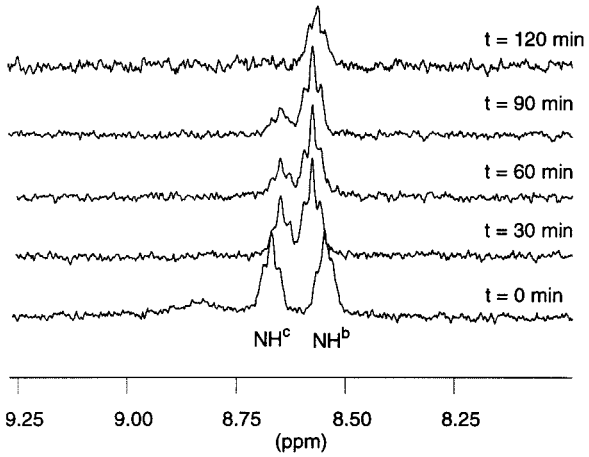


Figure 4. ^{1}H NMR H/D-exchange experiment of **1a** (D_2O added to a 1 mM solution of **1a** in 10% DMSO−$CDCl_3$ (v/v) at room temperature, 300 MHz).

ceptors,[8] consistent with the observed shifts for the various NH protons in the ^{1}H NMR spectrum as described above. In addition to these binding interactions between the carboxylate and the guanidiniocarbonyl moiety, the pyrrole NH^a and the amide NH^b of the first pyrrole moiety can form hydrogen bonds to the carbonyl oxygen next to the guanidiniocarbonyl pyrrole further stabilizing this conformation (Figure 2).

ROESY experiments also indicate that **1a** folds into a well-defined loop. In this conformation, amide NH^b points inward and is involved in hydrogen bonding to the opposite carbonyl group, whereas amide NH^c points outward toward the solvent. Therefore, as expected, amide NH^b shows NOE signals to all four CH_2 groups of the linker and the neighboring pyrrole NH^a (Figure 3). Amide NH^c, however, only exhibits NOEs with the two nearest CH_2 groups and no NOE with its neighboring pyrrole NH^d. Even a weak NOE signal between the two pyrrole NHs a and d is observed, which indicates that both pyrrole units, though being located at the opposite ends of a rather long and flexible alkyl chain, obviously are quite close to each other. Therefore, **1a** in contrast to **1b** does not exist in a linear extended conformation or a random coil. In that case, both amide NHs should give similar NOEs to the methylene groups in the linker and no NOE between the pyrrole NHs would be expected. All observed NOEs, however, are in full agreement with the proposed intramolecular loop structure for **1a**.

The difference between the two amide protons b and c, one forming intramolecular hydrogen bonds (NH^b) the other being only exposed to the solvent (NH^c), can also be proved by variable-temperature and H/D-solvent exchange NMR studies. Protons that form stable intramolecular hydrogen bonds show a much smaller shift change upon increase in temperature and also exchange much more slowly with the solvent than protons that are only exposed to the solvent.[3d] And indeed the temperature gradient for the shift change of amide NH^c is more than twice as large as that of amide proton NH^b. In DMSO, the signal for amide NH^c shifts from $\delta = 9.4$ at 30 °C to 8.0 at 170 °C, whereas the signal for amide NH^b shifts from $\delta = 8.5$ only to 7.9. Furthermore, upon addition of D_2O to a 1 mM solution of **1a** in 10% DMSO-d_6 in $CDCl_3$ at room temperature, amide proton NH^c has completely exchanged after 2 h and its signal has disappeared while amide NH^b is still detectable even after 2 days (see Figure 4). These observed differences in solvent exchange rate and temperature gradient clearly reflect a strinkingly different chemical environment to which the two amide NHs b and c are exposed. This rules out any extended or random coil-like structure, but again is in total agreement with the self-folded loop structure proposed for **1a**.

To probe the stability of this intramolecular complexation in **1a**, NMR studies were done at 140 °C to see whether dissociation could be seen. Opening up of this folded conformation should lead to a random coil structure with an NMR spectrum similar to that of the nonassociated zwitterion **1b**. However, though the signals become increasingly broad and difficult to detect at higher temperatures, the spectrum of **1a** does not change its appearance accordingly. Hence, it seems as if the loop is conformationally completely stable in DMSO even at elevated temperatures. This is remarkable as very often small molecules adopt their specific conformation only in rather unpolar solvents and are completely unfolded in polar solvents.[3] From earlier NMR titration studies, we already know that the intermolecular complexation of pyrrole carboxylate (NMe_4^+ salt) with [5-(methoxycarbonyl)-1*H*-pyrrole-2-carbonyl]guanidinium (picrate salt) in DMSO has an association constant of $K \approx 10^6$ mol^{-1}, and even in 40% water−DMSO (v/v) it still has $K \approx 5 \times 10^3$ mol^{-1}.[19] As the intramolecular association in **1a** should entropically be more favorable, it is expected that the association constant is even $K \geq 10^6$ mol^{-1}. Therefore, it is not surprising that, under the conditions studied here, no unfolding of the molecule could be seen.

Theoretical calculations also support the high stability of this loop. Molecular dynamics calculations were performed using the Amber* force field and the GB/SA water solvation treatment as implemented in Macromodel 6.5.[20] Molecule **1a** was allowed to thermally equilibrate at 300 K for 5 ps and was then heated to 500 K within 20 ps. After another equilibration period of 100 ps at this temperature, the molecule was slowly cooled to 300 K again (over 50 ps) and equilibrated for another 10 ps. This procedure enables the molecule to overcome any possible conformational energy barriers on its hypersurface before optimizing the final structure. The resulting conforma-

(20) Mohamadi, F.; Richards, N. G. J.; Guida, W. C.; Liskamp, R.; Lipton, M.; Caufiled, C.; Chang, G.; Hendrickson, T.; Still, W. C. *J. Comput. Chem.* **1990**, *11*, 440−467.

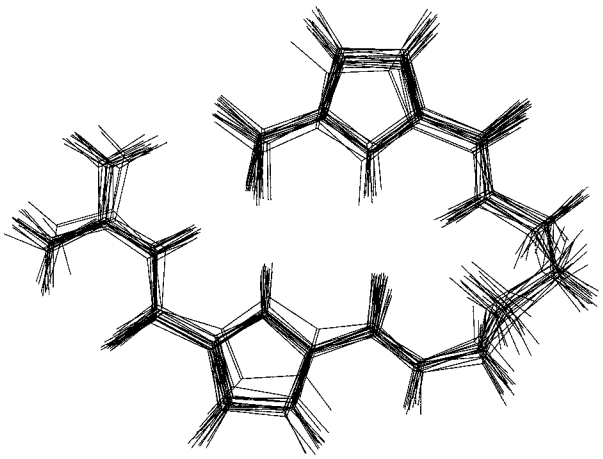


Figure 5. Superposition of 15 structures of **1a**, sampled from a Molecular Dynamics calculation over a time period of 100 ps at 300 K.

tion, which was the same as the energy minimum found by a conventional Monte Carlo simulation, corresponds to the loop already deduced from the NMR studies and shown in Figure 2. As a constant temperature MD calculation on this energy minimum conformation shows (100 ps simulation at 300 K; Figure 5), the structure is well defined and rather stable. At least during the time period of this simulation, only the inner methylene groups of the linker show some flexibility, but none of the bonding interactions is lost, nicely illustrating the high thermal stability of this conformation.

Conclusions

In conclusion, a novel self-folding motif based on multiple weak interactions within a zwitterion, in principle expected to be rather flexible, has been identified and structurally characterized. It could be shown here by NMR studies with H/D solvent exchange and ROESY experiments that zwitterion **1a**, in contrast to the very similar **1b**, folds into a well-defined loop, which is conformationally completely stable even in DMSO. This folding motif can be useful for the design of supramolecular polymers by varying the linker length and its flexibility.

Experimentals:

General Remarks. Solvents were dried and distilled under argon before use. All other reagents were used as obtained from either Aldrich or Fluka. All experiments were run in oven-dried glassware under argon unless otherwise stated. ^{1}H and ^{13}C NMR shifts are reported relative to the deuterated solvents. Peak assignments are based on DEPT, 2D NMR studies and comparison with literature data.[8,16]

General Procedure for the Preparation of tBoc-Protected Amides 4. To a suspension of acid **2** (37 mmol) and amine **3**[13] (37 mmol) in THF−CH_2Cl_2 (300 mL, 1:1 mixture, v/v) were added DCC (8.4 g, 41 mmol) and DMAP (0.25 g, 2 mmol) at 0 °C, and the resulting mixture was stirred at room temperature for 24 h. After filtration and removal of the solvent under reduced pressure, the crude product was purified by flash chromatography on silica gel (eluent: ethyl acetate/hexane, 2:1, with 0.1% NEt_3). **4a** (9.9 g, 79%): mp 108 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 1.41 (s, 9 H, tBu), 1.45 (m, 4 H, CH_2), 2.92 (m, 2 H, CH_2), 3.28 (m, 2 H, CH_2), 3.78 (s, 3 H, CH_3), 6.70 (m, 2 H, pyrrole CH), 6.81 (t, 2 H, amide NH), 8.25 (t, 1 H, amide NH), 12.08 (s, 1 H, pyrrole NH); ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 26.63, 27.26 (both CH_2), 28.46 (tBu), 38.65, 39.75 (both CH_2), 51.64 (CH_3), 112.50 (CH), 115.22 (CH), 124.18, 131.43, 138.99, 159.33, 160.72 (all quat C). Anal. Calcd for $C_{16}H_{25}N_3O_5$ (339.18): C, 56.61;, H, 7.43; N, 12.39. Found: C, 56.46; H, 7.24; N, 12.24. **4b** (8.2 g, 71%): mp 148 °C; ^{1}H NMR (300 MHz, DMSO-d_6) δ 1.32 (s, 9 H, tBu), 3.02 (m, 2 H, CH_2), 3.25 (m, 2 H, CH_2), 3.78 (s, 3 H, CH_3), 6.68 (m, 2 H, pyrrole CH), 6.91 (t, 2 H, amide NH), 8.32 (t, 1 H, amide NH), 12.07 (s, 1 H, pyrrole NH); ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 28.39 (tBu), 39.09, 39.45 (both CH_2), 51.63 (CH_3), 112.48 (CH), 115.21 (CH), 124.22, 131.35, 155.85, 159.62, 160.70 (all quat C). Anal. Calcd for $C_{14}H_{21}N_3O_5$ (311.15): C, 53.99; H, 6.80; N, 13.50. Found: C, 53.94; H, 6.85; N, 13.49.

General Procedure for the Preparation of Methyl Esters 7. The tBoc-protected amide **4** (30 mmol) was dissolved in CH_2Cl_2 (100 mL) and the solution cooled to 0 °C. Over a period of 3 h, TFA (10 mL) was added, and the mixture was stirred at room temperature for 24 h. After removal of the solvent under reduced pressure, the residue was extracted with acetone and the resulting white precipitate of the crude amine salt was filtered off and dried over phosphorus pentoxide. The amine **5** (4.4 mmol) was then reacted without further purification with acyl chloride **6**[19] (4 mmol) in the presence of NEt_3 (1.82 g, 18 mmol) in THF (30 mL), following a procedure already described.[8] After removal of the solvent under reduced pressure, the residue was purified by chromatography on alumina (eluent: CH_2Cl_2/MeOH, 4:1, with 0.1% NEt_3). The yields were generally in the range from 30 to 45%. **7a**: ^{1}H NMR (300 MHz, DMSO-d_6) δ 1.51 (m, 4 H, CH_2), 3.21 (m, 4 H, CH_2), 3.81 (s, 3 H, CH_3), 6.71 (m, 2 H, pyrrole CH), 6.83 (m, 1 H, pyrrole CH), 7.49 (m, 1 H, pyrrole NH), 8.29 (t, 1 H, amide NH), 8.45 (t, 1 H, amide NH), 8.52 (brs, 4 H, guanidinium NH_2), 11.81 (s, 1 H, amide NH), 12.09, 12.35 (both, s, 1 H, pyrrole NH); ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 26.74, 26.77, 38.55, 38.63 (all CH_2), 51.63 (CH_3), 112.53, 115.22, 116. 09, 118.42 (all CH), 124.19, 125.49, 131.39, 133.12, 155.0.58, 159.18, 159.38, 159.80, 160.71 (all quat C). **7b**: ^{1}H NMR (300 MHz, DMSO-d_6) δ 3.32 (m, 4 H, CH_2), 3.75 (s, 3 H, CH_3), 6.72 (m, 2 H, pyrrole CH), 6.85 (m, 1 H, pyrrole CH), 7.41 (m, 1 H, pyrrole CH), 8.30 (brs, 4 H, guanidinium NH_2), 8.50, (t, 1 H, amide NH), 8.68 (t, 1 H, amide NH), 11.81 (brs, 1 H, pyrrole NH), 12.12 (s, amide NH), 12.31 (s, 1 H, pyrrole NH); ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 38.68, 39.45 (both CH_2), 51.67 (CH_3), 112.65 (CH), 115.28 (CH), 124.35, 131.28, 159.55, 159.74, 160.71 (all quat C).

General Procedure for the Preparation of the Zwitterions 1. The methyl ester **7** (0.5 mmol) was stirred with LiOH monohydrate (63 mg, 1.5 mmol) in THF/water (4 mL, 4:1, v/v) overnight at room temperature. According to TLC, the conversion was quantitative. After evaporation of the solvent, the residue was taken up in water and the pH adjusted to pH = 6 with hydrochloric acid. The resulting white precipitate was filtered off, washed with water, and in the case of **1a** recrystallized from methanol. **1a**: mp 300 °C dec; ^{1}H NMR (300 MHz, DMSO-d_6) δ 1.51 (m, 2 H, CH_2), 1.62 (m, 2 H, CH_2), 3.28 (m, 2 H, CH_2), 3.51 (m, 2 H, CH_2), 6.51, 6.63, 7.00, 7.03 (all, m, 1 H, pyrrole CH), 7.95 (brs, 2 H, guanidinium NH_2), 8.61 (t, 1 H, amide NH), 9.53 (t, 1 H, amide NH), 9.98 (brs, 2 H, guanidinium NH_2), 11.80 (s, 1 H, pyrrole NH), 13.21 (s, 1 H, pyrrole NH), 14.82 (s, 1 H, amide NH); ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 24.08, 25.15, 35.15, 36.61 (all CH_2), 111.65, 112.34, 1113.76, 117.93 (all CH), 127.36, 128.43, 130.04, 130.75, 156.94, 159.30, 161.29, 165.95 (all quat C). **1b**: mp 300 °C dec; ^{1}H NMR (300 MHz, DMSO-d_6) δ 3.35 (m, 2 H, CH_2), 3.48 (m, 2 H, CH_2), 6.72 (m, 4 H, pyrrole CH), 7.32 (brs, 4 H, guanidinium NH_2), 8.41 (t, 1 H, amide NH), 8.62 (t, 1 H, amide NH), 11.70 (s, 1 H, pyrrole NH), 11.89 (s, 1 H, pyrrole NH), 12.02 (s, 1 H, amide NH); ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 38.89, 38.92 (all CH_2), 111.96, 112.5, 113.68, 114.14 (all CH), 127.37, 129.12, 129.92, 131.63, 160.04, 160.34, 160.57, 163.16 (all quat C).

General Procedure for the Preparation of Diamides 10. Acyl chloride **8** (20 mmol) was prepared from the corresponding acid[12] (3.4 g, 20 mmol) by reaction with oxalyl chloride (5.1 g, 40 mmol) in the presence of DMF (three drops) in CH_2Cl_2 (50 mL) for 2 h. After removal of the solvent under

reduced pressure, the crude acyl chloride was taken up in benzene (50 mL) and the diamine **9** (10 mmol) was added. The reaction mixture was refluxed for 48 h. After the HCl evolution ceased, the solvent was evaporated and the residue either recrystallized from methanol (**10a**) or purified by flash chromatography on silica gel (eluent: CH_2Cl_2/MeOH, 9:1, with 0.1% NEt_3) (**10b**). **10a** (1.67 g, 43%): mp 190 °C dec; ^{1}H NMR (300 MHz, DMSO-d_6) δ = 1.51 (m, 4 H, CH_2), 3.38 (m, 4 H, CH_2), 3.81 (s, 6 H, CH_3), 6.72 (m, 4 H, pyrrole CH), 8.38 (t, 2 H, amide NH), 12.08 (s, 2 H, pyrrole NH); ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 26.83 (CH_2), 38.60 (CH_2), 51.69 (CH_3), 112.55 (CH), 115.27 (CH), 124.25, 131.43, 159.41, 160.75 (all quat C). Anal. Calcd for $C_{18}H_{22}N_4O_6$ (390.40): C, 55.38; H, 5.68; N, 14.35. Found: C, 55.10; H, 5.88; N, 14.20. **10b** (1.31 g, 36%): mp 240 °C dec; ^{1}H NMR (300 MHz, DMSO-d_6) δ 3.37 (m, 4 H, CH_2), 3.77 (s, 6 CH_3), 6.78 (m, 4 H, pyrrole CH), 8.45 (t, 2 H, amide NH), 12.11 (s, 2 H, pyrrole NH); ^{13}C NMR (75.5 MHz, DMSO-d_6) δ 38.75 (CH_2), 51.17 (CH_3), 112.61 (CH), 115.28 (CH), 124.34 (quat C), 131.28, 159.72, 160.70 (all quat C). Anal. Calcd for $C_{16}H_{18}N_4O_6 \cdot {}^1/_2H_2O$ (371.13): C, 51.73; H, 5.16; N, 15.09. Found: C, 51.75; H, 4.81; N, 14.92.

General Procedure for the Preparation of Methyl Esters 7 by Guanidinylation of Diamide 10. Compound **10** (4 mmol) was dissolved in a solution of sodium methoxide (prepared by dissolving sodium (176 mg, 8 mmol) in 20 mL of methanol). Guanidinium chloride (764 mg, 8 mmol) was added, and the reaction mixture was refluxed for 12 h. After removal of the solvent under reduced pressure, the residue was purified by chromatography on alumina (eluent: CH_2Cl_2/MeOH, 4:1, with 0.1% NEt_3) in the same way as already described above. The yields were in the range of 18−25%.

Molecular Modeling. All calculations described in this paper were performed on an SGI O_2 workstation using the software package Macromodel 6.5.[20] Monte Carlo conformational searches were performed with at least 1000 steps until the energy minimum structure had been found several times. Molecular dynamics calculations were carried out as described in the text. The Amber* force field and the GB/SA water solvation model implemented in Macromodel were used in all studies.

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Supporting Information Available: ^{1}H and ^{13}C NMR and DEPT spectra for compounds **1a**,**b**, **4a**,**b**, **7a**,**b**, and **10a**,**b** and the ROESY spectrum of **1a**. This material is available free of charge via the Internet at http://pubs.acs.org.

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