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Positive Homotropic Allosteric Receptors for Neutral Guests: Annulated Tetrathiafulvalene–Calix[4]pyrroles as Colorimetric Chemosensors for Nitroaromatic Explosives

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Dedicated to Professor Jean-Marie Lehn on the occasion of his 70th birthday

Abstract: The study of positive homotropic allosterism in supramolecular receptors is important for elucidating design strategies that can lead to increased sensitivity in various molecular recognition applications. In this work, the cooperative relationship between tetrathiafulvalene (TTF)-calix[4]pyrroles and several nitroaromatic guests is examined. The design and synthesis of new annulated TTF-calix[4]pyrrole receptors with the goal of rigidifying the system to accommodate better nitroaromatic guests is outlined. These new derivatives, which display significant improvement in terms of binding constants, also display a positive homotropic allosteric relationship, as borne out from the sigmoidal nature of the binding isotherms and analysis by using the Hill equation, Adair equation, and Scatchard plots. The host–guest complexes themselves have been characterized by single-crystal X-ray diffraction analyses and studied by means of UV-spectroscopic titrations. Investigations into the electronic nature of the receptors were made by using cyclic voltammetry; this revealed that the binding efficiency was not strictly related to the redox potential of the receptor. On the other hand, this work serves to illustrate how cooperative effects may be used to enhance the recognition ability of TTF-calix[4]pyrrole receptors. It has led to new allosteric systems that function as rudimentary colorimetric chemosensors for common nitroaromatic-based explosives, and which are effective even in the presence of potentially interfering anions.

Keywords: allosterism · chemosensors · cooperative effects · explosives · supramolecular chemistry · tetrathiafulvalenes

Introduction

The design and construction of new artificial receptors displaying a positive homotropic allosteric response for targeted guests is a well-appreciated challenge for the biomimetic chemist.[1] Allosteric interactions, and the associated cooperative reaction to initial binding events, are key features of many biological processes; they provide, for instance, the tight regulation of oxygen binding by hemoglobin[2] and underlie the exquisite control observed in many enzymatic processes.[3] Among the four distinctive categories of allostery conceivable for synthetic receptors with multiple binding sites for a single substrate, namely 1) negative heterotropic, 2) positive heterotropic, 3) negative homotropic, and 4) positive homotropic, only the latter provides an amplified, positive response to the initial binding of a given chemical species. Unfortunately, simple systems capable of eliciting this type of allosteric response are rare.[4] This is particularly true in the case of neutral substrate recognition, and in the specific case of nitroaromatic explosives detection[5] we are unaware of any examples where the principles of cooperativity have been exploited to enhance the binding response. Here, we report an extremely simple, yet effective, in situ colorimetric sensing material ("chemosensor") for ni-

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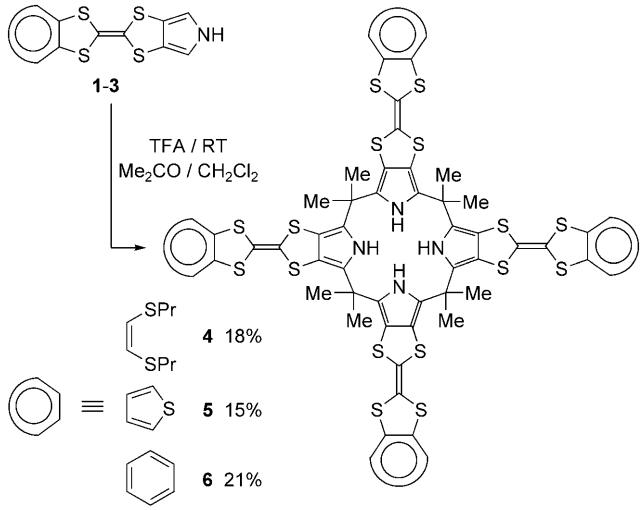
troaromatic explosives that is based on the use of modified tetrathiafulvalene (TTF)-calix[4]pyrrole frameworks.

Prior to this work, we described the synthesis of the TTF-functionalized calix[4]pyrrole **4** (Scheme 1).[6] This first-generation system was found to undergo a fast, easy to visualize color change (from yellow to green) when exposed to 1,3,5-trinitrobenzene (TNB) in organic solvents in the absence of chloride ion. However, receptor **4** is characterized by only modest binding affinities towards TNB and was found to require relatively high concentrations to produce a naked eye detectable colorimetric response (e.g., [TNB] $\approx$ 2 mM and [**4**] = 1 mM) when studied in $CHCl_3$. We reasoned that by modifying the electronics of the system and exploiting cooperative effects, we could overcome this deficiency. As discussed below, large enhancements in the response sensitivity—up to 1000-fold in the most favorable cases—have now been successfully achieved by replacing the TTF subunits originally used to construct **4** with the electronically modified aromatic thiophene and benzene TTF subunits. Specifically, we have found that the resulting annulated TTF-calix[4]pyrroles, receptors **5** and **6** (Scheme 1), display unique positive homotropic allosterism in their binding with the test polynitroaromatic explosives TNB, picric acid (TNP), and 2,4,6-trinitrotoluene (TNT). These cooperative effects are correlated with a corresponding increase in colorimetric sensing ability.

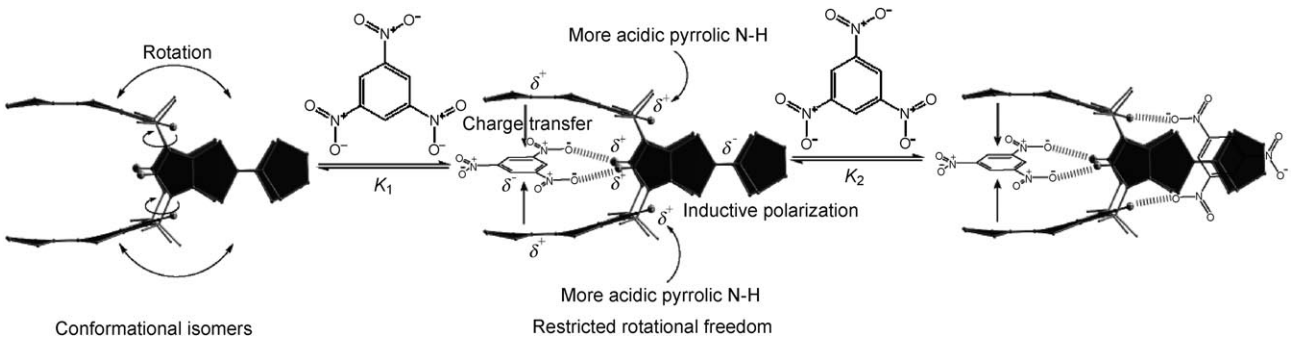


Scheme 1. Synthesis of the TTF-calix[4]pyrrole derivatives **4**, **5**, and **6**.

The design rationale for the present work was the realization that the parent TTF-calix[4]pyrrole **4** could be modified through annulation of an aromatic moiety onto the TTF subunit. Specifically, we thought that this approach could be exploited to enlarge and rigidify the "TTF walls", thus, making this class of electron-rich receptors a better match in terms of size and shape for flat electron-deficient substrates, such as TNB, TNP, and TNT. This annulation was also expected to reduce the flexibility of the system as a whole. This, in turn, was considered likely to enhance the binding of a second substrate in accord with the generalized mechanistic postulate given in Scheme 2. On the other hand, it was also appreciated that annulation with an aromatic moiety would modulate both the nature of the π surface (size and shape), as well as the electronic properties of the system (e.g., redox potentials, donor ability, dipole moment orientation, etc.), in addition to providing flat π surfaces that are extended relative to those present in **4**. Depending on their specific nature, such variations could serve to increase or decrease the propensity to form donor–acceptor complexes. They could also affect the hydrogen bonding donor ability of the pyrrolic NH protons. Thus, one goal of the present study was to gain insight into the interplay of these potentially competing factors with the hope of developing systems that displayed an enhanced guest-dependent colorimetric response for canonical nitroaromatic analytes represented by our test substrates TNB, TNP, and TNT.

Results and Discussion

With the above considerations in mind, the new thieno- and benzo-annulated TTF-calix[4]pyrrole derivatives **5** and **6** were targeted for synthesis. Preliminary DFT calculations (see the Supporting Information) led to the consideration that the requisite precursors, namely **2** and **3**, differ from **1** in terms of their π-electron donating properties (energies of



Scheme 2. Proposed origin of the positive homotropic allosteric effect seen when receptors **4**, **5**, and **6** are titrated with the test nitroaromatic explosives TNB, TNP, and TNT.

their respective HOMOs). Specifically, these systems are predicted to be somewhat less electron-rich. Thus, in the absence of compensating size and shape complementarity effects and the associated benefits of cooperativity as per Scheme 2, it was expected that the receptors derived from these precursors (i.e., **5** and **6**) would be less effective than **4**. As detailed below, this did not prove to be the case.

As shown in Scheme 1, receptors **5** and **6** were prepared in 15% and 21% yield, respectively, from the trifluoroacetic acid (TFA) catalyzed condensation of **2** and **3**, respectively, with an excess of Me_2CO at room temperature in CH_2Cl_2. Under these same conditions, receptor **4** was obtained in 18% yield from **1**. Whereas **1** and **4** are known substances,[6] compounds **2**, **3**, **5**, and **6** have not been reported in the literature.

Although known to exist in several limiting conformations (e.g., 1,2- and 1,3-alternate, partial-cone, cone), in the absence of a strongly bound substrate calix[4]pyrroles typically adopt the so-called 1,3-alternate conformation.[7] It is this conformation that, in the case of **4**, was found to favor TNB binding.[6] It is thus worth noting that all three receptors (i.e., **4**, **5**, and **6**) adopt 1,3-alternate conformations in the solid state. Such a conclusion is established by X-ray structural analyses, which revealed the presence of two bound Me_2CO molecules in the case of **6** and two included MeOH molecules for **5**; in both cases, these solvent molecules are bound through hydrogen-bond interactions and are held within the two cliplike cavities defined by the calix[4]pyrrole framework in this conformation (see Figure 1 and the Supporting Information).[8] While not unexpected given the chemistry of calix[4]pyrroles, such findings were thought to augur well for the use of these systems as receptors for nitroaromatic substrates.

Initial evidence that the new calix[4]pyrrole derivatives **5** and **6** could complex nitroaromatic guests came from a single crystal X-ray diffraction analysis of the 1:2 complex **6**·2TNP. The resulting structure (Figure 1, bottom) revealed that two molecules of TNP are located on opposite sides of the central calix[4]pyrrole core and that both guests are fully "sandwiched" within the two cavities provided by the overall 1,3-alternate conformation. The short distances of 3.2809(74)–3.4900(75) Å between the two imaginary planes defined by the electron-rich benzo-TTF-pyrroles and the electron-deficient TNP guest are fully consistent with strong face-to-face π electron donor–acceptor interactions.[9] For each of the TNP guests, hydrogen-bond interactions are present between the oxygen atoms of the nitro groups in the 2- and 4-positions of the TNP guests and two of the pyrrolic protons provided by the tetra-benzo-TTF-calix[4]pyrrole receptor **6** (the relevant distances range from 2.9215(86)–3.2044(97) Å). X-ray crystallographic studies of the complexes formed between **4** with TNT and TNB[6] also revealed that these latter substrates are likewise fully "sandwiched" within the two cavities defined by the 1,3-alternate conformation of receptor **4** (see the Supporting Information).

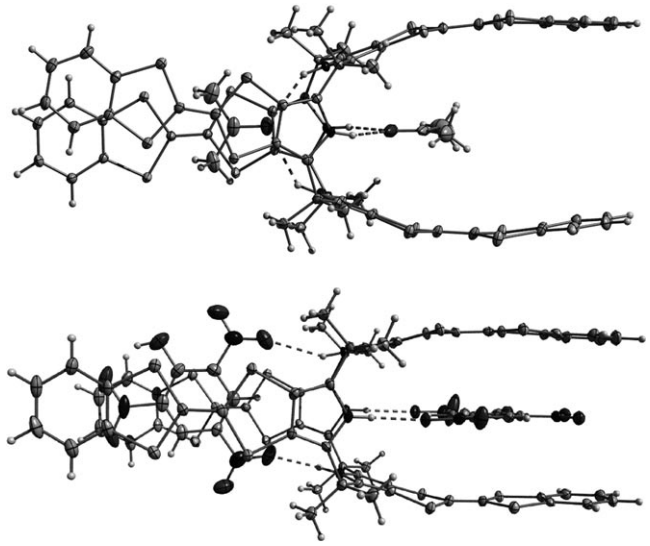


Figure 1. X-ray crystal structure of **6**·2Me_2CO (top) and **6**·2TNP (bottom) shown with the thermal ellipsoids at 30%.[8]

The interaction of the three TTF-calix[4]pyrroles with the test nitroaromatic explosives TNB, TNP, and TNT was further investigated through visible spectrophotometric titration experiments carried out in $CHCl_3$ solution. The guest binding events were easily visualized by following the progressive color change produced as the test nitroaromatic explosives were added to receptors **4**–**6**. Plots of the associated changes in absorption intensity as a function of TNB, TNP, or TNT concentration were then used to construct binding isotherms (see the Supporting Information).

As can be seen from an inspection of Figure 2, the titration isotherms were characterized by a sigmoidal curvature, as would be expected for a cooperative binding process. Such presumed cooperative binding was particular evident in the case of receptor **5**. In all cases, however, the allosteric nature of the interaction was fully analyzed by using the Hill equation,[10] Scatchard plots,[11] and non-linear regressions of the two-site Adair equation.[12] These results provided support for a 1:2 host–guest binding mode. Job plots[13] were also constructed, and these were fully consistent with the proposed 1:2 host–guest stoichiometry (see the Supporting Information).

Linear Hill plots ($\log(Y/(1-Y)) = n\log[\text{explosives}] + \log K_a$ (where Y, n, and K_a are the fractional saturation of host, the Hill coefficient, and the association constant, respectively) corresponding to each individual titration isotherm with a satisfactory correlation coefficient ($R > 0.99$) were then made. From the slopes and the intercepts of these plots, both the association constants (K_a) and the Hill coefficients (n) were obtained. As summarized in Table 1, the values of the Hill coefficients ranged from 1.23 to 1.86 (vs. a mathematical limit of 2.0 for a perfectly cooperative two-site receptor binding two molar equivalents of an identical substrate). Further evidence for the proposed allosterism came from an upward curvature in Scatchard plots, which is a characteristic of positive cooperativity (see the Supporting Information).

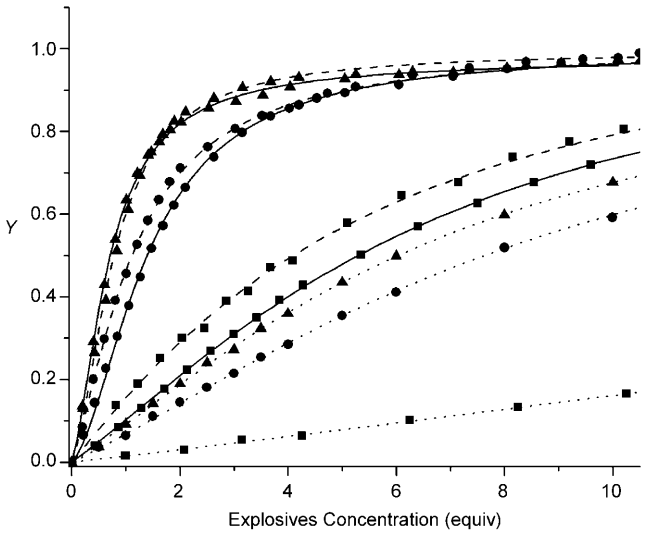


Figure 2. Binding curves obtained from the visible spectroscopic titration of receptors **4** (0.2 mM, •••••), **5** (0.2 mM, -----), and **6** (0.2 mM, ——) with increasing amounts of TNB (▲), TNP (●), and TNT (■) in $CHCl_3$ at room temperature (symbols) and the calculated binding isotherms derived by using the two-site Adair equation.[12] The *y* axis denotes the fractional saturation of receptors **4**, **5** and **6**.

Table 1. Microscopic association constants[a] and cooperativity parameters for receptors **4**, **5**, and **6**, as obtained from fits to the Hill[10] and Adair[12] equations.

	K_a [M^{-2}]	n	K_1 [M^{-1}]	K_2 [M^{-1}]	K_2/K_1
4·2TNB	4.3×10^3	1.27	3.9×10^2	1.4×10^3	3.6
4·2TNP	3.8×10^3	1.30	2.8×10^2	1.2×10^3	4.1
4·2TNT	3.3×10^2	1.23	5.9×10^1	2.0×10^2	3.3
5·2TNB	3.4×10^6	1.70	1.3×10^3	3.1×10^4	24
5·2TNP	3.7×10^6	1.86	6.4×10^2	2.0×10^4	31
5·2TNT	2.3×10^4	1.45	3.2×10^2	2.8×10^3	10
6·2TNB	1.5×10^5	1.34	2.8×10^3	1.7×10^4	6.2
6·2TNP	9.1×10^4	1.34	1.7×10^3	1.1×10^4	6.5
6·2TNT	1.2×10^4	1.31	5.7×10^2	2.6×10^3	4.5

[a] Estimated errors for calculated binding constants are within 12%.

The binding data were also fitted to the two-site Adair equation ($Y=(K'_1[G]+2K'_1K'_2[G]^2/(1+K'_1[G]+2K'_1K'_2[G]^2)$ where K'_1 and K'_2 are macroscopic Adair constants and [G] is the concentration of the explosive in question); this allowed the determination of the individual microscopic binding constants K_1 and K_2 and the ratio K_2/K_1.[4,14] These latter values, included in Table 1, were found to range from 3.3 to 31 (correlation coefficients $R>0.99$). Compared to the theoretical value expected for a non-cooperative system in the case of identical and independent binding sites ($K_2/K_1=0.25$), these high values provide additional support for the proposed positive homotropic allosteric nature of the binding process. Finally, a good agreement was found between the values of the Hill coefficients for each receptor–substrate pair and the corresponding K_2/K_1 ratio (and vice versa) as shown in Table 1.

These findings are rationalized in terms of the effects of aromatic annulation. Specifically, in accord with the mechanism proposed in Scheme 2, we suggest that the origin of the positive cooperative binding seen for the interaction of TNB, TNP, and TNT with **4**, and to a greater extent for **5** and **6**, reflects the fact that the binding of a first equivalent of an electron-deficient guest forces the inherently flexible (and normally conformationally mobile) calix[4]pyrrole receptor to adopt a more rigidified 1,3-alternate conformation. This leads to a loss of rotational freedom and provides a pre-organized framework for the subsequent binding of another equivalent of the guest. It is an enhancement in this rigidity, resulting from the replacement of the freely rotating thiopropyl substituents with rigid aromatic rings whose shape and electronic features match well the targeted substrates, to which we ascribe the greater efficacy of the new receptors **5** and **6** relative to **4**.[15]

Based on the quantitative analyses summarized in Table 1, receptor **5** was found to display the highest degree of positive homotropic allostery across the board, with in general, the sequence **5** > **6** > **4** being observed in terms of the manifest cooperativity. Receptor **5** also displays the highest relative affinity for all three electron-deficient guests, as reflected in the fact that significantly larger K_a values (as derived from the Hill analyses) are obtained for **5** than for **4** or **6**. The K_a ratios are also calculated; this was done by dividing the K_a values of **5** by those of **4** and **6**, respectively. The values found in this way were 790 and 23 (TNB), 973 and 41 (TNP), and 70 and 2 (TNT), as would be expected for a highly cooperative system. In all cases, the greatest affinity was seen for TNP, with this species also acting as the most efficient allosteric effector, as inferred from both the Hill coefficients and the K_2/K_1 ratios (TNP > TNB > TNT).

It is important to appreciate that although the calix[4]pyrrole **5** was found to bind the test substrates TNB, TNP, and TNT with greater affinity than the benzo-fused system **6**, this latter was a considerably more effective receptor than **4**. In fact, in comparison with the first generation system **4**, the new annulated TTF-calix[4]pyrrole derivatives **5** and **6** display significantly enhanced binding constants for all three nitroaromatic analytes (by up to three orders of magnitude). As pointed out in the literature,[16] the strength of the π donor–acceptor (D–A) complexation is governed not only by the electron-donating properties, as gauged by the first redox potential, but also by the overlap integral between the donor and acceptor. In spite of the finding that the first half-wave oxidation potential of receptor **4** obtained by cyclic voltammetry ($E^1_{1/2}=0.29$ V in $CHCl_3$ vs. SCE) is lower than that of either **5** ($E^1_{1/2}=0.37$ V in $CHCl_3$ vs. SCE) or **6** ($E^1_{1/2}=0.30$ V in $CHCl_3$ vs. SCE), significantly enhanced binding of **5** and **6** compared to **4** toward nitroaromatic explosives is seen (see the Supporting Information). This is ascribed to the larger area and more suitable orientation of the receptor π surfaces, as well as the cooperative effects that underlie the overall two-step binding process (see above).

The high affinities displayed by the new receptors **5** and **6** led us to consider that they might act as improved colori-

metric chemosensors ("sensors") for TNB, TNP, or TNT, effective even in the presence of competing anions or in the presence of H_2O. In fact, from the linear region of spectroscopic titration curves at receptor concentrations of 0.2 mM, detection limits in the low sub-ppm levels were estimated for the test nitroaromatic explosives (TNB: 0.44, 0.30, 1.56 $\mu g\,mL^{-1}$; TNP: 0.77, 0.64, 2.68 $\mu g\,mL^{-1}$; TNT: 3.04, 2.72, 15.3 $\mu g\,mL^{-1}$, for receptors **6**, **5**, and **4**, respectively). To test further their utility as possible sensors, 0.1 mM $CHCl_3$ solutions of **4**, **5**, and **6** were mixed with 0.2 mM aqueous solutions of the three test nitroaromatic explosives. As can be seen in Figure 3, these additions result in an immediate

Figure 3. Visual color changes induced by the addition of 2 mL of 0.1 mM solutions of **4**, **5**, and **6** in $CHCl_3$ to 3 mL of 0.2 mM aqueous solutions of the test nitroaromatic explosives TNB, TNP, and TNT in the absence and presence of salts. Here, "salts" refers to a mixture of $NaHCO_3$, K_2CO_3, $MgSO_4$, $CaCl_2$, and NH_4Cl, 2 mM in each experiment. The contents of the vials from left to right are as follows: 1) pure **4**, 2) **4** + TNB, 3) **4** + TNB + salts, 4) **4** + TNT, 5) **4** + TNP, 6) pure **5**, 7) **5** + TNB, 8) **5** + TNB + salts, 9) **5** + TNT, 10) **5** + TNP, 11) pure **6**, 12) **6** + TNB, 13) **6** + TNB + salts, 14) **6** + TNT, 15) **6** + TNP.

change in the color of the $CHCl_3$ solution, with the actual variations depending on the specific choice of receptor and explosives. Presumably, the observed color changes reflect D–A interactions between the occupied HOMOs of TTF-functionalized pyrroles to the empty π^* orbitals of the nitroaromatic compounds. These interactions are expected to be more favorable in the case of receptors that can provide for a better spatial fit as noted above.

Consistent with this conclusion is the finding that the aromatic-fused TTF-calix[4]pyrroles **5** and **6** displayed changes that were significantly greater than those produced by the first generation system **4**. Furthermore, the addition of salts ($NaHCO_3$, K_2CO_3, $MgSO_4$, $CaCl_2$, and NH_4Cl, each at a concentration of 2 mM) to the aqueous phase failed to inhibit the colorimetric response.[17] This stands in marked contrast to what has been reported to be true for other colorimetric nitroaromatic sensors, such as colored reaction-based commercial field test kits,[5a,18] where the color change produced by formation of a Jackson–Meisenheimer anion when nitroaromatic compounds are treated with strong bases, or amine-functionalized Au nanoparticles[19] that rely on D–A interactions between cysteamine and TNT and an aggregation-induced color change. These perceived advantages lead us to suggest that compounds **5** and **6** could play a role as chemosensors for nitroaromatic explosives, particularly when a quick, qualitative response is needed that does not rely on an instrumental response. It is to be noted, however, that the latter methods are much more sensitive than even the best of the new systems reported here, although those do offer potential advantages in terms of ease of use.

Conclusions

In summary, we have successfully demonstrated that the affinity of TTF-calix[4]pyrrole derivatives for nitroaromatic explosives can be significantly enhanced through electronic modulation of the parent TTF-pyrrole and the action of positive allosterism. As far as we are aware, this is the first synthetic artificial receptor that displays biommetic positive homotropic allostrism in the binding process of nitroaromatic explosives. It is also the first that can operate in an aqueous environment free of potential interference from potentially competing ions, such as chloride. While far less sensitive than more complex methods, we believe that these novel positive homotropic allosteric receptors may offer some advantages relative to current chemosensory technologies,[20] many of which still suffer from lack of selectivity and complexity of setup and use. Work is thus underway to test these systems more fully and to increase sensitivity through, for example, incorporating of our new chemosensor materials into various polymer matrixes.

Experimental Section

General methods: All reagents were purchased from Aldrich and used without further purification. ^1H and ^{13}C NMR spectra were recorded at 25 °C with a 500 MHz Varian Innova instrument. High Resolution ESI mass spectrometry was performed using a Varian QF ESI 9.4 Tesla with Internal Calibration.

Compounds **1**,[21] **4**,[6] thieno[3,4-d][1,3]dithiole-2-thione,[22] and benzo[d]-[1,3]dithiole-2-thione[23] were prepared according to literature procedures.

Synthesis of 2: A mixture of 5-tosyl-5*H*-[1,3]dithiolo[4,5-c]pyrrol-2-one (3.11 g, 10 mmol) and thieno[3,4-d][1,3]dithiole-2-thione (3.61 g, 20 mmol) in neat triethylphosphite (150 mL) was stirred for four hours at 140 °C and then cooled to room temperature. Addition of MeOH (200 mL) to the reaction mixture led to precipitation of a yellow solid, which was collected by filtration. The solid obtained in this way was purified by column chromatography (silica gel, CH_2Cl_2/hexane 2:1) to afford the tosyl-protected compound **2a** (2.04 g, 4.5 mmol, 42 %). ^1H NMR (500 MHz, $CDCl_3$): $\delta = 7.71$ (d, $J = 8.29$ Hz, 2H; Ar-H), 7.29 (d, $J = 8.05$ Hz, 2H; Ar-H), 6.92 (s, 2H; thiophene α-H), 6.85 (s, 2H; pyrrolic α-H), 2.39 ppm (s, 3H; tosyl CH_3). To a solution of **2a** (2.04 g, 4.5 mmol) in a mixture of methanol (100 mL) and THF (100 mL) sodium methoxide (30 % in MeOH, 10 equiv) was added. The mixture was heated to 50 °C for 30 min and concentrated under reduced pressure until the volume was 50 mL. The reaction mixture was poured into an aqueous solution of NH_4Cl (200 mL). The resulting yellow precipitate was collected by filtration and washed with water. The yellow solid obtained in this way was purified by column chromatography (silica gel, CH_2Cl_2/hexanes 3:2) to yield **2** as a yellow solid (1.28 g, 4.27 mmol, 95 %). ^1H NMR (500 MHz, $CDCl_3$): $\delta = 8.16$ (brs, 1H; N-H), 6.84 (s, 2H; thiophen α-H), 6.59 ppm (d, $J = 2.44$ Hz, 2H; pyrrolic α-H); ^{13}C NMR (500 MHz, $CDCl_3$): $\delta =$ 136.1, 123.2, 119.9, 119.7, 111.8, 109.7 ppm; HRMS (ESI): m/z: calcd for $C_{10}H_5NS_5$: 298.90200 [M^+]; found: 298.90201.

Synthesis of 3: A mixture of 5-tosyl-5*H*-[1,3]dithiolo[4,5-c]pyrrol-2-one (3.11 g, 10 mmol) and benzo[d][1,3]dithiole-2-thione (3.68 g, 20 mmol) in neat triethylphosphite (150 mL) was stirred for four hours at 140 °C and cooled to room temperature. MeOH (200 mL) was then added to the reaction mixture, which led to precipitation of an orange solid, which was collected by filtration. The resulting solid was purified by column chromatography (silica gel, CH_2Cl_2/hexane 3:2) to afford the tosyl-protected compound **3a** (2.82 g, 6.3 mmol, 63 %). ^1H NMR (500 MHz, $CDCl_3$): $\delta =$ 7.71 (d, $J = 8.42$ Hz, 2H; tosyl), 7.28 (d, $J = 8.12$ Hz, 2H; tosyl), 7.22 (m,

2H; benzene), 7.10 (m, 2H; benzene), 6.91 (s, 2H; pyrrolic α-H), 2.39 ppm (s, 3H; tosyl). To a solution of **3a** (2.82 g, 6.3 mmol) in a mixture of methanol (150 mL) and THF (150 mL) sodium methoxide (30% in MeOH, 10 equiv) was added. The resulting mixture was heated to 50°C for 30 min and concentrated under reduced pressure until the volume was 50 mL. The reaction mixture was poured into an aqueous solution of NH_4Cl (200 mL). The yellow precipitate was filtered and washed with water. The resulting yellow solid was purified by column chromatography (silica gel, CH_2Cl_2/hexanes 2:1) to afford **3** as a yellow solid (1.80 g, 6.13 mmol, 97%). ^{1}H NMR (500 MHz, $CDCl_3$): $\delta = 8.16$ (br s, 1H; N-H), 7.23 (q, $J = 3.41$ Hz, 2H; benzene α-H), 7.09 (q, $J = 3.17$ Hz, 2H; bezene β-H), 6.59 ppm (d, $J = 2.44$ Hz, 2H; pyrrolic α-H); ^{13}C NMR (500 MHz, $CDCl_3$): $\delta = 136.5$, 125.7, 121.8, 120.1, 119.9, 111.3, 109.6 ppm; HRMS (ESI): m/z: calcd for $C_{12}H_7NS_4$: 292.9458 [M^+]; found: 292.94558.

Synthesis of 5 and 6: general procedure for the synthesis of aromatic annulated TTF-calix[4]pyrroles 5 and 6: Compound **2** (1.00 g, 3.34 mmol) for **5**, or compound **3** (1.00 g, 3.4 mmol) for **6** was dissolved in a mixture of acetone (250 mL) and dichloromethane (250 mL) and the solution was degassed with argon for 30 min before trifluroacetic acid (3 mL) was slowly added. The mixture was stirred overnight at room temperature before triethylamine (6 mL) was slowly added. After removal of the solvent by evaporation, a solid yellow residue was obtained, which was washed with water, dried in vacuo, and purified by column chromatography (silica gel, CH_2Cl_2/hexanes 2:1) to give **5** (0.17 g, 0.13 mmol, 15%) or **6** (0.238 g, 0.178 mmol, 21%) as appropriate in the form of yellow solids. For **5**: ^{1}H NMR (500 MHz, $CDCl_3$): $\delta = 7.13$ (br s, 4H; N-H), 6.74 (s, 8H; thiophene α-H), 1.58 ppm (s, 24H; CH_3); ^{13}C NMR (500 MHz, $CDCl_3$): $\delta = 136.0$, 127.0, 123.9, 123.6, 115.3, 111.6, 30.9, 27.6 ppm; HRMS (MALDI): m/z: calcd for $C_{52}H_{36}N_4S_{20}$: 1355.7346 [M^+]; found: 1355.73487. For **6**: ^{1}H NMR (500 MHz, $CDCl_3$): 7.14 (br s, 4H; N-H), 7.09 (m, 8H; benzene C-H), 6.95 (m, 8H; benzene C-H), 1.59 ppm (s, 24H; CH_3); ^{13}C NMR (500 MHz, $CDCl_3$): $\delta = 136.4$, 127.0, 125.5, 121.6, 118.2, 115.7, 112.2, 30.9, 27.6 ppm; HRMS (ESI): m/z: calcd for $C_{60}H_{44}N_4S_{16}$: 1331.9121 [M^+]; found: 1331.90918.

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