# Anion-induced conformational changes in 2,7-disubstituted indole-based receptors†

**Damjan Makuc,[a] Martina Lenarčič,[a] Gareth W. Bates,[c] Philip A. Gale*[c] and Janez Plavec*[a,b]**

***Received 6th May 2009, Accepted 12th June 2009***
***First published as an Advance Article on the web 9th July 2009***
**DOI: 10.1039/b908947k**

The conformational preorganization and anion-induced conformational changes of indole-based receptors functionalized with an amide group at the 2-position and a variety of amide, urea and thiourea moieties at the 7-position have been studied by the means of NMR spectroscopy. NOE experiments showed that *anti–anti* orientation across C2–C2α and C7–N7α bonds is preferred for receptors **1–4** in acetone solution in the absence of anions. Anion–receptor interactions have been evaluated through $^{1}$H and $^{15}$N chemical shift changes. In 2,7-bis-carboxamido functionalized indoles the interaction with chloride and bromide anions primarily occurs at the indole H1 proton. The introduction of urea and thiourea moieties increases the number of hydrogen bond donor sites which manifests itself in a distribution of halide–receptor interactions among the H1, H7α and H7γ protons. Acetate anions also interact strongly with indole and urea NH donor groups, whereas nitrate anions interact solely with H7α and H7γ urea/thiourea protons. NOE enhancements in the presence of anions revealed that anion–receptor complexes favour the *syn–syn* conformation of the C2 and C7 substituents.

## Introduction

The development of unique anion receptors, sensors and transporters is an area of intense research activity.[1–5] Potential applications in the separation and extraction of anionic species, in the development of new sensing systems and in the design of new compounds that may have potential biological activity have driven the synthesis of a plethora of receptors containing amides and thioamides, pyrroles and indoles, ureas and thioureas, ammonium, guanidinium and imidazolium moieties.[6,7]

Indole is employed by Nature in the sulfate binding protein[8] and in the enzymatic active site of haloalkane dehalogenase[9] to bind anions, however research in the area of indole-based anion receptors[10–20] is still at an early stage when compared to the range of anion receptors based on pyrrole.[21] The recognition and sensing properties of indole can be effectively regulated by appending additional hydrogen bond donors to the indole skeleton. Amides have been widely used as hydrogen bond donor groups to bind anionic species,[22] whilst urea and thiourea moieties have been extensively employed as receptors for Y-shaped oxo-anions through two directional hydrogen bonds.[23,24]

In this study we have analysed the potential conformational preorganization and conformational changes of four previously synthesized bis-amido and mono-amido-mono-urea 2,7-functionalized indoles[25] in the presence of a diverse range of anionic guests using NMR techniques. The indoles have a variety of substituents in the 7-position including secondary amides, urea and thiourea groups (Fig. 1) and a carboxamidophenyl substituent in the 2-position. Crystal structure elucidation of solid-state complexes of these species with anions and solution stability constant determinations in DMSO-$d_6$–0.5% water have been conducted previously.[25] The results in the current study complement crystallographic data in the sense that equivalent conformers are found in the solid and liquid states when complexed with anions. In contrast, the predominant conformer in solution in the absence of anions is different from the conformation established in the crystal.

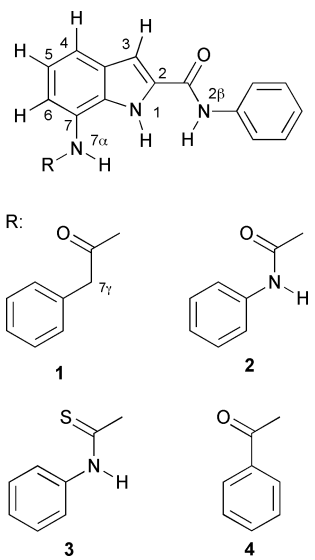


**Fig. 1** Anion receptors **1–4** and atom numbering.

[a]*Slovenian NMR Center, National Institute of Chemistry, Hajdrihova 19, SI-1000 Ljubljana, Slovenia. E-mail: janez.plavec@ki.si; Fax: +386 147 60 300; Tel: +386 147 60 353*
[b]*Faculty of Chemistry and Chemical Technology, University of Ljubljana, SI-1000 Ljubljana, Slovenia*
[c]*School of Chemistry, University of Southampton, Southampton, UK SO17 1BJ. E-mail: philip.gale@soton.ac.uk; Fax: +44 23 8059 6805; Tel: +44 23 8059 3332*

† Electronic supplementary information (ESI) available: $^{1}$H NMR spectra of **1**, **3** and **4** in the absence of anions as well as upon addition of one equivalent of different anions. See DOI: 10.1039/b908947k

**Table 1** Selected $^{1}H$ and $^{15}N$ NMR chemical shifts for **1–4**[a]

| | H1 | H2β | H7α | H7γ | H6 | N1 | N2β | N7α | N7γ |
|---|---|---|---|---|---|---|---|---|---|
| **1** | 10.87 | 9.59 | 9.61 | 3.86 | 7.53 | 136.5 | 128.2 | 131.8 | — |
| **2** | 10.91 | 9.59 | 8.43 | 8.26 | 7.28 | 136.2 | 128.2 | 106.1 | 110.4 |
| **3** | 10.75 | 9.61 | 9.07 | 9.16 | 7.37 | 133.8 | 128.4 | 125.4 | 133.1 |
| **4** | 10.91 | 9.62 | 9.80 | — | 7.64 | 136.6 | 128.3 | 126.4 | — |

[a] Reported chemical shifts (in ppm) correspond to NMR spectra acquired in acetone-$d_6$ at 298 K. The complete set of $^{1}H$ and $^{13}C$ NMR chemical shifts is available in the Experimental section.

## Results

### NMR assignment

NMR spectroscopy has been used to evaluate and correlate structural and conformational properties of anion receptors **1–4** in acetone-$d_6$ to their preorganization for interaction with anions. As a first step, the $^{1}H$, $^{13}C$ and $^{15}N$ resonances of **1–4** have been assigned based on the analysis of 1D proton and carbon spectra as well as $^{13}C$–$^{1}H$ and $^{15}N$–$^{1}H$ correlations in 2D HSQC and HMBC spectra. Selected $^{1}H$ and $^{15}N$ NMR chemical shifts are reported in Table 1, with the full list including $^{13}C$ NMR data available in the Experimental section.

The different physicochemical properties of C7-substituents in 2,7-bisfunctionalized indoles **1–4** are reflected in NMR parameters and in particular in the chemical shielding of their hydrogen bond donors. Receptor **1** with its phenylacetylamido group exhibits H7α and N7α chemical shifts of 9.61 and 131.8 ppm, respectively. The introduction of a urea moiety in **2** manifests itself in the considerable upfield shifts of H7α and N7α to 8.43 and 106.1 ppm, respectively. Additional nitrogen atom N7γ in **2** exhibits a chemical shift of 110.4 ppm with the corresponding H7γ proton resonating at 8.26 ppm. Substitution of the oxygen atom in **2** with sulfur in **3** causes considerable deshielding of H7α, H7γ, N7α and N7γ which is in agreement with the more acidic nature of the thiourea moiety (Table 1). Moderate deshielding of H7α in **4** with respect to **1** has been attributed to its benzoylamido group. On the other hand, N7α in **4** is shielded by 5.4 ppm with regard to **1**. Considerable changes of H7α and N7α chemical shifts in **1–4** correspond to variations in the electron donating/withdrawing nature of N7α-substituents. The minute alterations of chemical shifts of H2β and N2β are in agreement with an invariant C2 substituent. The $^{13}C$ chemical shift changes are insignificant and do not reflect that different groups are attached to the C7 atom (see Experimental section).

### $^{1}H$ NMR chemical shift changes in 1–4 upon addition of anions

The chemical shift values changed upon addition of one equivalent of chloride, bromide, nitrate and acetate ions added as tetrabutylammonium salts to receptors **1–4** (Fig. S1–S3†). The anion–receptor interactions induced a significant change in the chemical shielding of $^{1}H$ and $^{15}N$ NMR resonances and only minor changes in $^{13}C$ resonances. Fig. 2 illustrates $^{1}H$ chemical shift changes in **2** upon interaction with different anions. Significant deshielding of H1, H7α and H7γ protons was observed upon addition of chloride or bromide anions to a solution of **2** (Fig. 2a–c). Interaction of nitrate anions with **2** resulted in relatively smaller downfield shifts of the urea H7α and H7γ protons (Fig. 2d). The

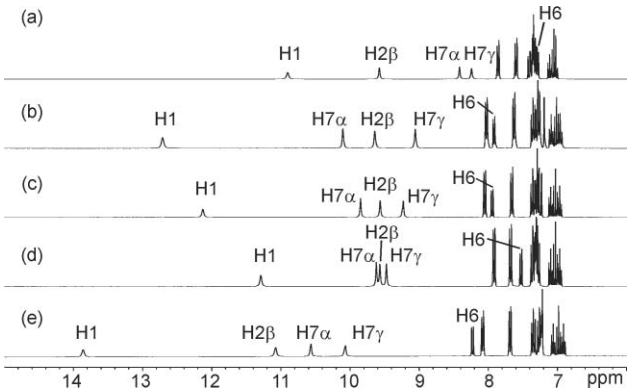


**Fig. 2** $^{1}H$ NMR spectra of **2** in the absence of anions (a) and upon addition of one equivalent of the following anions: chloride (b), bromide (c), nitrate (d) and acetate (e). All spectra were recorded at 298 K.

significant deshielding of all four NH protons of **2** indicated a strong interaction with acetate anions (Fig. 2e).

A comparison of proton $\Delta\delta$ values induced upon addition of one equivalent of the four anions to receptors **1–4** is shown in Fig. 3. The chemical shift of H1 shows the greatest change among all NH protons in **1** upon interaction with chloride (Fig. 3a). Moderate chemical shift changes of H2β and H7α were observed for the **1**·$Cl^-$ complex, while the methylene H7γ shows only a negligible change. The significantly larger $\Delta\delta$ values for both the urea H7α and H7γ protons in **2**·$Cl^-$ should be noted *vs.* **1**·$Cl^-$, whereas the H2β chemical shift change is smaller. However, the $\Delta\delta$ value for indole H1 is the largest $^{1}H$ chemical shift change upon formation of **2**·$Cl^-$. A similar trend was observed for **3**·$Cl^-$, where strong deshielding of H7α and H7γ protons is in accordance with the higher acidity of the thiourea group. Interestingly, a significant chemical shift change of H6 was observed in **3**·$Cl^-$ which is most probably the result of conformational changes and will be discussed later. In contrast to the other three receptors, the smallest $\Delta\delta$ value for H7α and greatly increased deshielding of H1 and H2β protons upon formation of **4**·$Cl^-$ complex can be attributed to the benzoylamido substituent at C7 (Fig. 3a).

The addition of bromide anions resulted in slightly smaller downfield chemical shift changes in all four receptors in comparison to chloride anions (*cf.* Fig. 3a and 3b). As both anions are spherical, the main reason for these differences can be attributed to their different size and basicity. H1 showed significant downfield shifts in **1**·$Br^-$ and **4**·$Br^-$ complexes, whereas **2**·$Br^-$ and **3**·$Br^-$ exhibited considerable deshielding of H1, H7α and H7γ. The latter suggested that all mentioned protons participate in the interaction with bromide anions.

Notably smaller chemical shift changes were observed upon addition of nitrate to **1–4** compared to the other anions studied. Receptors **1** and **4** which lack urea or thiourea moieties show relatively small $\Delta\delta$ values below 0.4 ppm, which leads us to suggest only minor nitrate–receptor interactions are occurring in this case (Fig. 3c). On the other hand, in **2** and **3** deshielding of both H7α and H7γ protons by up to 1.3 ppm was observed upon addition of nitrate anions.

Significant $\Delta\delta$ values of up to 3.3 ppm of NH protons in all four studied receptors occurred upon addition of acetate anions to **1–4** (Fig. 3d) consistent with the formation of strong complexes.[25]

 

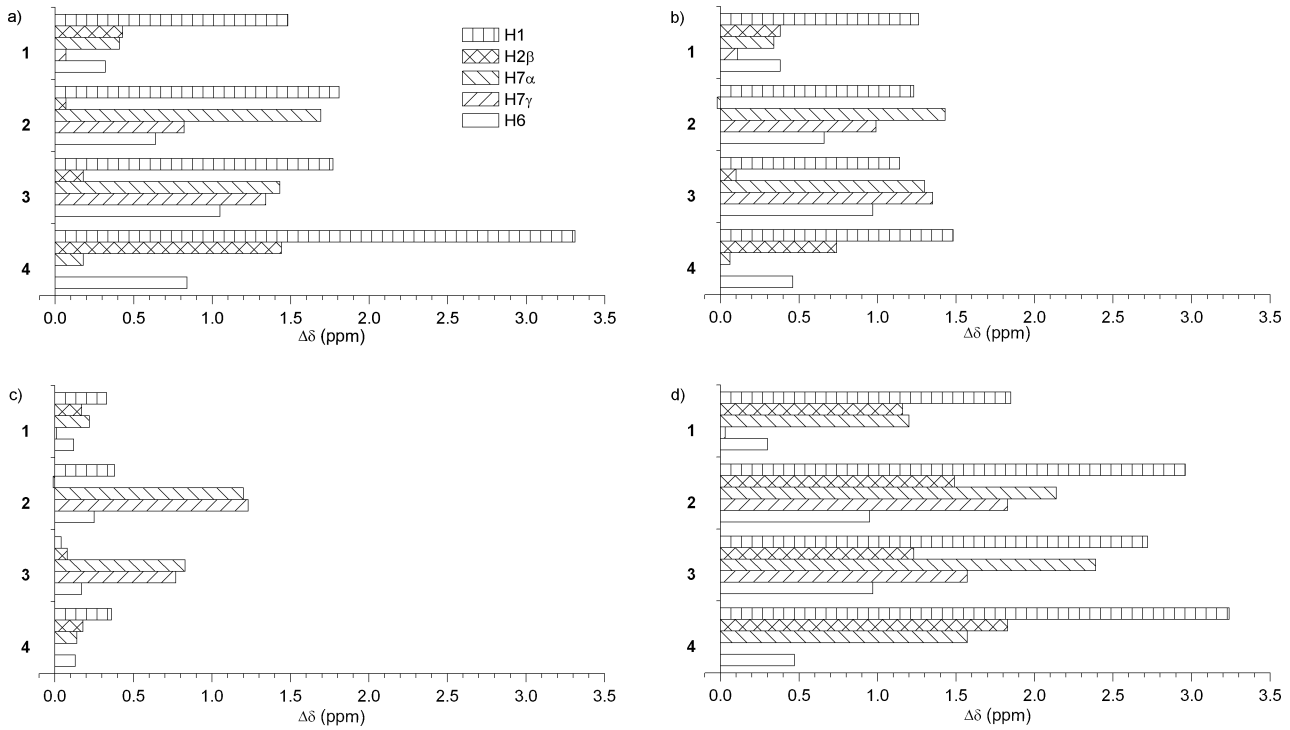


**Fig. 3** $^{1}$H NMR chemical shift changes, $\Delta\delta = \delta$(in the presence of anions) – $\delta$(in the absence of anions), induced by addition of one equivalent of chloride (a), bromide (b), nitrate (c) and acetate (d) anions to receptors **1–4**. Note, there is no H7γ proton in **4**.

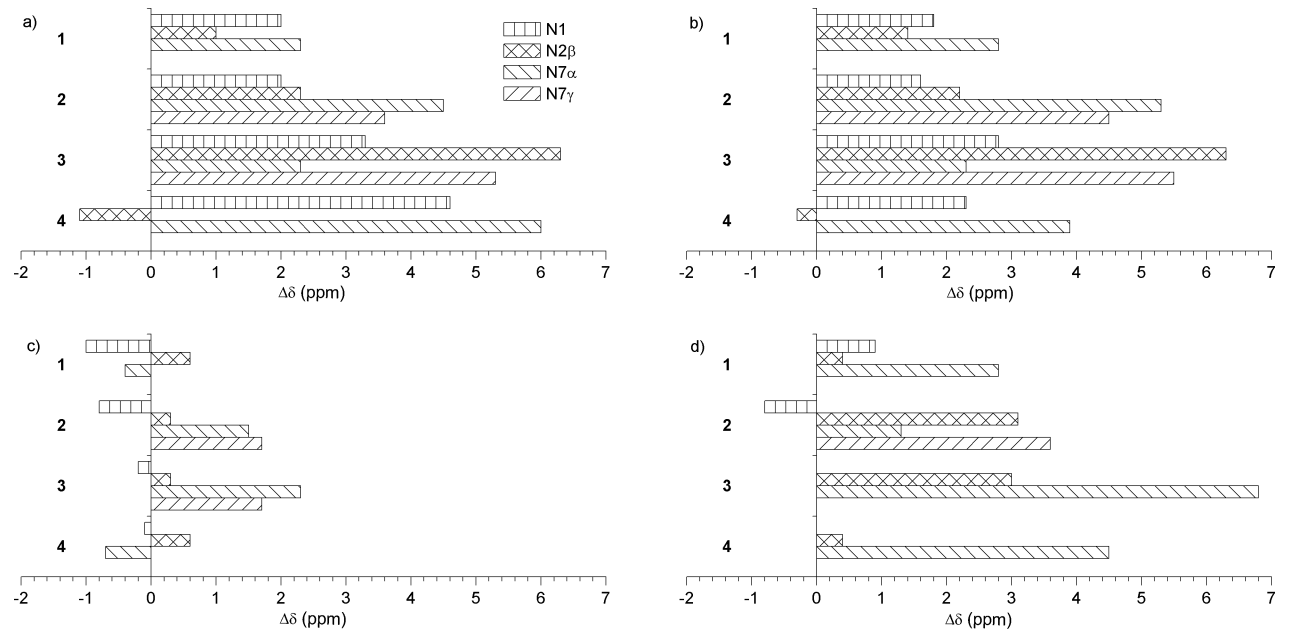


**Fig. 4** $^{15}$N NMR chemical shift changes, $\Delta\delta = \delta$(in the presence of anions) – $\delta$(in the absence of anions), induced by addition of one equivalent of chloride (a), bromide (b), nitrate (c) and acetate (d) anions to receptors **1–4**. Note, $^{15}$N chemical shifts could not be determined for some atoms due to signal broadening after addition of acetate to **3** and **4**. There is no N7γ in **1** and **4**.

### $^{15}$N NMR chemical shift changes induced by anion interactions

Anion–receptor interactions evaluated through $^{1}$H chemical shift changes have been supported by $^{15}$N NMR data (Fig. 4). The largest $\Delta\delta$ values in **1**, **2** and **4** upon addition of chloride and bromide anions were observed for N7α, whereas in **3** chemical shifts of the N2β atom changed the most (Fig. 4a and 4b). N2β and N7γ atoms in **2** and **3** were also deshielded upon interaction with $Cl^-$ and $Br^-$. Interestingly, N1 was deshielded upon addition of one equivalent of chloride and bromide anions, and shielded in the presence of nitrate anions in **1–4** (Fig. 4a–c). The substitution of methylene H7γ in **1** with the NH group in **2** leads to increased deshielding of N2β upon addition of chloride, bromide and acetate anions. The change was magnified even further with $\Delta\delta$ values up

to 6.8 ppm when urea (**2**) was swapped for a thiourea moiety (**3**). A similar increase of N7γ deshielding ($\Delta\delta$ up to 5.5 ppm) in the presence of chloride and bromide was observed in **3** with respect to **2**. The interaction of $Cl^-$ with **4** leads to deshielding of N1 and N7α by up to 4.6 and 6.0 ppm, respectively, whereas N2β is shielded in **4**·$Cl^-$ complex (Fig. 4a). Analogous albeit smaller chemical shift changes were observed in **4**·$Br^-$ complex (Fig. 4b). Addition of nitrate anions results in lower $\Delta\delta$ values. The largest $^{15}N$ chemical shift changes of up to 2.5 ppm were observed for N7α and N7γ atoms in **2** and **3** (Fig. 4c).

### Conformational properties of receptors 1–4 in the absence and in the presence of anions

Conformational equilibria of 2,7-functionalized indole receptors have been studied with the use of 1D difference NOE experiments. As an example, 1D difference NOE spectra of **2** in the absence and upon addition of $Cl^-$ ions are shown in Fig. 5. Four well resolved NH protons have enabled unequivocal quantification of NOE enhancements. The predominant orientation of 2-phenylcarboxamide group in **2** has been established by the strong NOE enhancement of 14.5% at H3 upon saturation of the carboxamide H2β proton (Fig. 5e). In full agreement, the saturation of H2β has resulted in a very weak NOE at H1 (0.5%). The orientation along the C7–C7α bond in **2** has been established through NOE enhancements among H1, H6 and H7α. Relatively strong {H7α}–H6 (7.3%) and weaker {H1}–H7α NOEs (2.9%) have suggested the predominant conformation with H7α pointing away from indole H1 proton (Fig. 5c and 5g). A considerable decrease of {H2β}–H3 and {H7α}–H6 NOE enhancements to 7.5% and 2.7% was observed upon addition of chloride anions, respectively (Fig. 5f and 5h). Furthermore, observation of NOE enhancements among H1–H2β and H1–H7α leads us to suggest conformational changes occur in **2** upon interaction with chloride anions (*cf.* Fig. 5c–d, 5e–f and 5g–h).

The key NOE enhancements for **1–4** in the absence and in the presence of one equivalent of anions are presented in Table 2. Relatively strong {H2β}–H3 NOE (8.5%) in **1** is evidence to support the spatial proximity of H2β and H3 protons. Such an orientation across the C2–C2α bond is in accordance with weak NOEs between H2β and H1 (≤1.0%). The saturation of H7α has resulted in weak NOE enhancement at H1 (≤1.0%), whereas stronger NOE was observed at H6 (3.2%). The saturation of H1 yields a weak overall NOE at H2β and H7α protons (Table 2). Although the H2β and H7α signals overlap in **1**, differences in overall NOE values in the absence and in the presence of anions have turned out to be very informative in the assessment of conformational properties. Addition of one equivalent of chloride to **1** has altered several NOE enhancements. The saturation of H2β and H7α has resulted in an increase of NOE at H1 from ≤1.0% to 6.9%, and a decrease of {H2β}–H3 NOE from 8.5% to 4.9% and {H7α}–H6 NOE from 3.2% to 2.1% (Table 2). Furthermore, the saturation of H1 has caused a major increase of overall NOE at H2β and H7α (13.0%). Addition of bromide to **1** has triggered very similar changes which leads us to suggest an analogous predominant conformation as established for **1**·$Cl^-$ complex

**Table 2** The key NOE enhancements observed for **1–4** in the absence and in the presence of one equivalent of anions

| | Saturated: | H2β | | H1 | | H7α | | H6 |
|---|---|---|---|---|---|---|---|---|
| Receptor | Enhanced: | H3 | H1 | H2β | H7α | H1 | H6 | H7α |
| **1** | without | 8.5 | ≤1[a] | 1.2[a] | | ≤1[a] | 3.2 | 0.6 |
| | chloride | 4.9 | 6.9[a] | 13.0[a] | | 6.9[a] | 2.1 | 0.9 |
| | bromide | 4.1 | 6.7[a] | 11.5[a] | | 6.7[a] | 1.6 | 0.9 |
| | nitrate | 9.9 | 2.5[a] | 5.2[a] | | 3.6[a] | 4.4 | 1.9 |
| | acetate | 4.5 | —[b] | —[b] | | —[b] | 2.5 | 0.1 |
| **2** | without | 14.5 | 0.5 | 0.2 | 2.9 | 5.0 | 7.3 | 1.0 |
| | chloride | 7.5 | 12.0 | 13.6 | 12.9 | 13.8 | 2.7 | 0.9 |
| | bromide | 8.2 | 9.7 | 12.0 | 10.7 | 11.4 | 3.7 | 0.7 |
| | nitrate | 10.0 | 4.7 | 3.0 | 5.2 | 6.3 | 6.8 | 2.9 |
| | acetate | 1.5 | 12.9 | 15.8 | 10.2 | 8.8 | 0.9 | 0 |
| **3** | without | 15.2 | 0 | 0.1 | 1.0 | 1.0[c] | 2.5[c] | —[d] |
| | chloride | 7.1 | 7.2 | 6.4 | 5.7 | 3.6[c] | 1.1[c] | 0.5 |
| | bromide | 6.9 | 5.8 | 5.3 | 4.4 | 3.8[c] | 1.4[c] | 0.2 |
| | nitrate | 15.3 | 1.4 | 1.1 | 0 | 0[c] | 2.8[c] | 0.7 |
| **4** | without | 15.3 | 0 | 0 | 0.9 | 1.5[a] | 6.5 | 0.6[d] |
| | chloride | 1.1 | 8.0 | 8.0 | 5.3 | 5.6 | —[d] | 0.3[d] |
| | bromide | 7.3 | 8.2 | 6.8 | 5.4 | 5.9 | 3.9 | 0.5[d] |
| | nitrate | 13.1 | 2.5[a] | 1.3 | 2.3 | 3.9[a] | 5.9 | 1.0[d] |

[a] H2β and H7α were saturated simultaneously or integrated together due to small $\Delta\delta$ values. [b] Negative NOEs due to proton exchange. [c] Both H7α and H7γ were saturated simultaneously or integrated together due to small $\Delta\delta$ values. [d] $^1H$ signals were overlapped and pair-wise enhancements could not be quantified unequivocally.

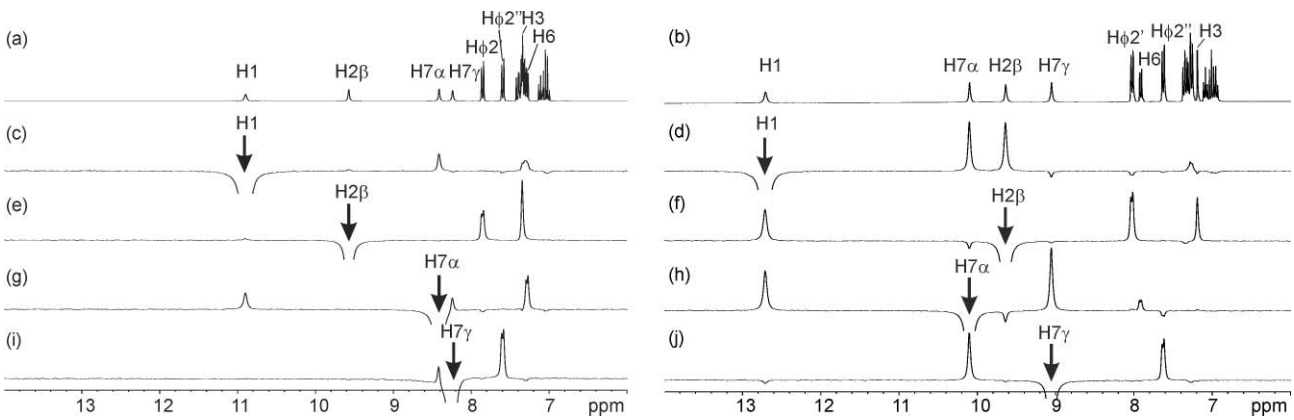


**Fig. 5** $^1H$ NMR spectra of **2** in the absence (a) and in the presence of $Cl^-$ ions (b), and corresponding 1D difference NOE spectra upon saturation of H1 (c, d), H2β (e, f), H7α (g, h) and H7γ protons (i, j). All spectra were recorded at 298 K.

 

(Table 2). In contrast, addition of nitrate anions to **1** has resulted in strong {H2β}–H3 NOE and moderate increase of {H7α}–H6 NOE which indicates negligible conformational changes in **1** upon interaction with $NO_3^-$ (Table 2). The decrease of {H2β}–H3 and {H7α}–H6 NOEs for **1**·$AcO^-$ complex is in agreement with conformational changes across the C2–C2α and C7–N7α bonds. Unfortunately, negative values of other key NOEs due to proton exchange prevented more detailed conclusions regarding conformational changes of **1** upon interaction with acetate anions.

The interaction of bromide anions with **2** resulted in an increase of {H2β}–H1, {H1}–H2β, {H1}–H7α and {H7α}–H1 NOEs (Table 2). Changes of NOE enhancements are analogous to those observed in **2**·$Cl^-$, evidence for comparable conformational changes in **2** upon interaction with bromide anions. Strong {H2β}–H3 and {H7α}–H6 NOEs of 10.0% and 6.8%, respectively, suggest only minor conformational {H2β}–H3 and {H7α}–H6 NOE values of 1.5% and 0.9%, respectively (Table 2). Strong NOEs between H1 and H2β, and likewise between H1 and H7α in **2**·$AcO^-$ complex indicate major conformational changes along C2–C2α and C7–N7α bonds.

Receptor **3** with its 7-thiourea moiety exhibited strong {H2β}–H3 NOE (15.2%) which indicated conformational preorganization along the C2–C2α bond. The predominance of the conformer of **3** with H2β and H3 being spatially close is further supported by negligible NOEs between H1 and H2β protons (<0.1%). The weak NOEs between H1 and H7α (1.0%) and a slightly stronger {H7α}–H6 NOE of 2.5% allude to the predominance of the C7–C7α conformer, where H6 and H7α are spatially closer than H1 and H7α. The addition of chloride and bromide anions results in a decrease of NOEs between H2β and H3 as well as between H7α and H6 (Table 2). Moreover, the increase of H1–H2β and H1–H7α NOEs is consistent with conformational changes in **3** upon addition of chloride and bromide anions. In the case of the **3**·$NO_3^-$ complex, small NOE changes were observed (Table 2). Regrettably, negative NOEs in **3**·$AcO^-$ complex NOEs thwarted a conformational study of this complex.

The conformational preorganization of **4**, where H2β and H7α point away from the indole H1 proton, is supported by strong {H2β}–H3 and {H7α}–H6 NOEs as well as weak NOE enhancements between H1–H2β and H1–H7α. The addition of chloride or bromide anions triggers conformational changes in **4**, which were clearly indicated by decreased {H2β}–H3 and {H7α}–H6 NOEs and increased NOEs between indole H1 and both H2β and H7α protons (Table 2). Insignificant changes of key NOEs in the **4**·$NO_3^-$ complex lead us to suggest that the predominant conformation remained mostly unchanged upon addition of nitrate anions. The saturation of proton signals in **4**·$AcO^-$ complex yielded negative NOEs, which did not allow detailed conformational analysis.

## Discussion

Acquired heteronuclear NMR data on four different anion receptors showed distinct changes as a result of their interactions with anions. Both chemical shift changes and conformational rearrangements can be attributed to structural details of receptors **1–4** as well as the anions' properties including their binding affinities. Examination of chemical shift changes showed a correlation between the nature of the C7 substituent and the magnitude and localization of chloride–receptor interactions. $\Delta\delta$ values in **1** suggested that interactions between the H1 protons and chloride anions are the strongest as deshielding is significantly more pronounced than for the H2β or H7α amide protons. Changing the phenylacetylamido moiety in **1** to a urea group in **2** led to increased deshielding of the H7α and H7γ protons which suggested that the anion–receptor interactions in **2**·$Cl^-$ complex involved the indole and urea NH protons. Our observed differences in proton $\Delta\delta$ values are in agreement with the published stability constants for **1**·$Cl^-$ and **2**·$Cl^-$ complexes.[25] The comparison of structurally related indoles **2** and **3** showed similar chemical shift changes for both receptors upon addition of chloride anions. The largest deshielding of H1 and H2β protons was observed in **4**·$Cl^-$ with respect to corresponding complexes of **1–3**, which suggested the predominant involvement of H1 and H2β donor groups in interaction with chloride anions.

Interaction of bromide anions with receptors **1–4** caused analogous albeit smaller chemical shift changes in comparison to chloride anions, in agreement with bromide's lower basicity. Considerable deshielding of the H7α and H7γ protons was observed upon addition of Y-shaped nitrate anions to **2** and **3** which contain urea and thiourea moieties, respectively. In contrast, only minor $^1H$ chemical shift changes were observed upon addition of $NO_3^-$ anions to **1** and **4** which contain amide groups at C2 and C7. Urea and thiourea moieties were shown to be preferred for interaction with trigonal planar anions due to their suitable shape and capability to form two hydrogen bonds.[23] The largest $^1H$ chemical shift changes were observed upon addition of acetate anions to **1–4**. Major deshielding of H1 protons in **1–4** is consistent with a strong interaction with acetate anions. Additionally, significant $\Delta\delta$ values of H2β and H7α show that acetate anions strongly interact with the other two donor groups in **1** and **4**. In conclusion, acetate and nitrate are both planar oxoanions that are predisposed for bidentate interactions with the urea and thiourea moieties in **2** and **3**. Larger chemical shift changes in **1–4** upon addition of acetate with respect to nitrate anions are consistent with the lower basicity of nitrate.

Substituents attached to the indole C2 and C7 carbons make the resultant anion receptors conformationally flexible. Four conformers with respect to the orientations across the C2–C2α and C7–N7α bonds are expected to be preferred (Fig. 6). A preliminary *ab initio* computational study of C2 and C7 functionalized indoles showed that the *anti–anti* conformer is energetically preferred in the absence of anions, whereas the *syn–syn* conformer is favoured for anion–receptor complexes.

Conformational preorganization of derivatives **1–4** has been assessed by NOE enhancements. 1D difference NOE experiments showed strong H2β–H3 and negligible H2β–H1 NOEs in the absence of anions which suggested the predominance of an *anti* orientation along the C2–C2α bonds in the receptors **1–4**. A relatively strong NOE between H7α–H6 together with a weak NOE between H7α and H1 is evidence that supports the predominance of an *anti* orientation along the C7–N7α bond in the absence of anions. The prevalent *anti–anti* conformers of **1–4** where the substituents' NH groups are pointing away from the indole H1 proton are in agreement with negligible H1–H2β and H1–H7α NOE enhancements. The arrangement of the C2α and C7β carbonyl groups in an *anti–anti* orientation is predisposed to act as an intramolecular hydrogen bond acceptor for the H1 proton.

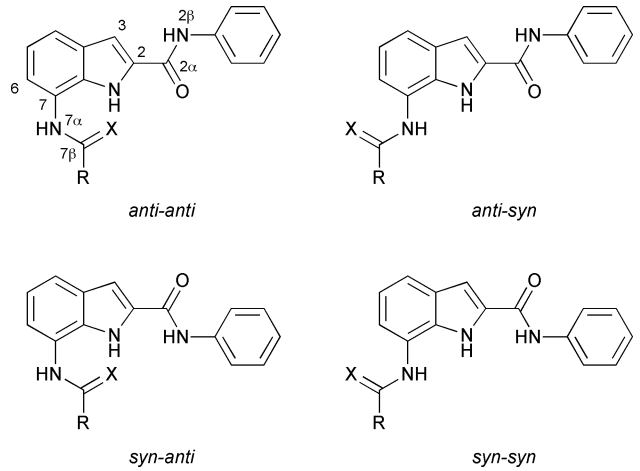


**Fig. 6** Four major conformational families with respect to C2 and C7 substituents in indole receptors. The first notation of individual conformer refers to the orientation along N1–C2–C2α–N2β, while the second refers to the C6–C7–N7α–C7β fragment. R stands for phenyl, benzyl and phenylamine substituents, whereas X symbolizes O or S atom as shown in Fig. 1.

The addition of anions to receptors **1–4** resulted in significant changes of key NOE enhancements. A major decrease of H2β–H3 as well as a minor decrease of H7α–H6 NOEs occurred upon addition of chloride, bromide and acetate anions. Simultaneous increases of NOEs among H1 and H2β as well as H1 and H7α furthermore suggested a conformational conversion of the receptors from *anti–anti* to *syn–syn* upon interaction with anions. Rotations along C2–C2α and C7–C7α bonds are also supported by H6 chemical shift changes. As this proton is not directly involved in anion–receptor interactions its $\Delta\delta$ values are most likely a result of variations of chemical environment upon conformational changes. No conformational changes were observed upon addition of nitrate anions to **1–4** as evidenced by almost unchanged NOE enhancements.

Our NMR data demonstrate that binding of chloride anions occurs primarily at H1 and H2β or H7α (Fig. 7a). Analogous interactions were established for receptors **1–4** upon addition of bromide anions. Proton chemical shift changes upon addition of nitrate anions to **1** and **4** suggest very weak interactions. However, introduction of urea or thiourea groups significantly improves the binding properties of nitrate to **2** and **3**. Negligible NOE changes in nitrate complexes with **1–4** together with corresponding $\Delta\delta$ values indicate that interaction of $NO_3^-$ anions occurs through the urea (**2**) or thiourea (**3**) group without rotation along the C7–C7α bond (Fig. 7b). Fig. 7c illustrates the proposed binding mode of acetate to **2**. Large chemical shift changes hint at strong interactions with indole and urea NH groups whereas interactions with H2β proton are weaker. Rotation from *anti–anti* to *syn–syn* conformer is anticipated in the other three acetate–receptor complexes but could not be confirmed experimentally.

## Experimental

$^1$H, $^{13}$C and $^{15}$N NMR spectra were acquired on a Varian Unity Inova 300 MHz NMR spectrometer. All data were recorded in acetone-$d_6$ at 298 K. Chemical shifts were referenced to the residual solvent signal of acetone-$d_6$ at $\delta$ 2.05 ppm for $^1$H (297.801 MHz) and $\delta$ 29.92 ppm for $^{13}$C (76.190 MHz), whereas $^{15}$N (30.188 MHz) chemical shifts were referenced relative to external benzamide ($\delta$ 103.55 ppm). The saturation delay in the 1D difference NOE experiment was 5.0 s. All anions were added as tetrabutylammonium salts.

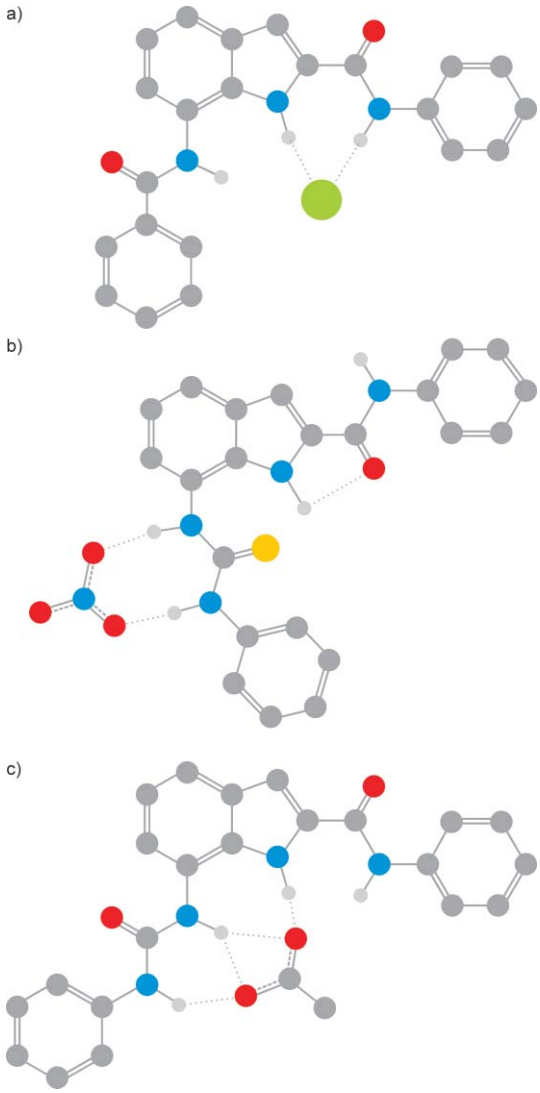


**Fig. 7** Conformations and positions of chloride anion in **4**·$Cl^-$ (a), nitrate anion in **3**·$NO_3^-$ (b) and acetate anion in **2**·$AcO^-$ (c) complexes based on chemical shift changes and NOE enhancements.

### 7-Phenylacetylamino-1*H*-indole-2-carboxylic acid phenylamide 1

$\delta_H$(300 MHz; acetone-$d_6$) 3.86 (H7γ), 7.04 (H5), 7.11 (N2β-*Ph*), 7.26 (C7γ-*Ph*), 7.34 (H3 and C7γ-*Ph*), 7.36 (N2β-*Ph*), 7.45 (H4 and C7γ-*Ph*), 7.53 (H6), 7.85 (N2β-*Ph*), 9.59 (H2β), 9.61 (H7α) and 10.87 (H1); $\delta_C$(75 MHz; acetone-$d_6$) 44.4 (C7γ), 104.5 (C3), 116.2 (C6), 119.2 (C4), 120.8 (Ph), 120.9 (Ph), 121.4 (C5), 124.6 (N2β-*Ph*), 125.5 (C3a), 127.7 (C7γ-*Ph*), 129.3 (Ph), 129.7 (Ph), 130.2 (Ph), 130.8 (C7), 132.5 C2), 136.9 (C7γ-*Ph*), 140.1 (N2β-*Ph*), 160.5 (C2α) and 170.5 (C7β).

### 7-(3-Phenylureido)-1*H*-indole-2-carboxylic acid phenylamide 2

$\delta_H$(300 MHz; acetone-$d_6$) 7.02 (C7γ-*Ph*), 7.05 (H5), 7.11 (N2β-*Ph*), 7.28 (H6), 7.29 (C7γ-*Ph*), 7.34 (H3 and N2β-*Ph*), 7.40 (H4), 7.60

 

(C7γ-*Ph*), 7.86 (N2β-*Ph*), 8.26 (H7γ), 8.43 (H7α), 9.59 (H2β) and 10.91 (H1); $\delta_C$(75 MHz; acetone-$d_6$) 104.6 (C3), 115.6 (C6), 118.3 (C4), 119.8 (C7γ-*Ph*), 120.9 (N2β-*Ph*), 121.6 (C5), 123.3 (C7γ-*Ph*), 124.6 (N2β-*Ph*), 126.0 (C3a), 129.7 (Ph, 2×) 130.0 (C7a), 130.8 (C7), 140.1 (N2β-*Ph*), 140.8 (C7γ-*Ph*).

**7-(3-Phenylthioureido)-1*H*-indole-2-carboxylic acid phenylamide 3**

$\delta_H$(300 MHz; acetone-$d_6$) 7.11 (C7γ-*Ph*), 7.12 (H5), 7.16 (N2β-*Ph*), 7.36 (N2β-*Ph* and C7γ-*Ph*), 7.37 (H6), 7.39 (H3), 7.58 (H4), 7.62 (C7γ-*Ph*), 7.84 (N2β-*Ph*), 9.07 (H7α), 9.16 (H7γ), 9.61 (H2β) and 10.75 (H1); $\delta_C$(75 MHz; acetone-$d_6$) 105.0 (C3), 120.8 (N2β-*Ph*), 120.9 (C4), 121.3 (C5), 121.5 (C6), 124.6 (N2β-*Ph*), 125.3 (C7γ-*Ph*), 126.0 (C7γ-Ph), 129.5 (Ph), 129.7 (Ph), 130.7 (C7), 140.0 (Ph) and 140.4 (Ph).

**7-Benzoylamino-1*H*-indole-2-carboxylic acid phenylamide 4**

$\delta_H$(300 MHz; acetone-$d_6$) 7.11 (C7γ-*Ph*), 7.12 (H5), 7.36 (Ph), 7.39 (H3), 7.5-7.6 (H4 and Ph), 7.64 (H6), 7.85 (N2β-*Ph*), 8.10 (C7γ-*Ph*), 9.62 (H2β), 9.80 (H7α), and 10.91 (H1); $\delta_C$(75 MHz; acetone-$d_6$) 104.7 (C3), 117.8 (C6), 119.7 (C4), 120.8 (Ph), 120.9 (Ph), 121.3 (C5), 124.6 (N2β-*Ph*), 125.3 (C3a), 128.8 (C7γ-*Ph*), 129.4 (C7γ-*Ph*), 129.7 (N2β-*Ph*), 130.9 (C7), 132.6 (C7γ-*Ph*), 136.0 (Ph) and 140.1 (Ph).

## Conclusions

Bis-amido and mono-amido-mono-urea 2,7-functionalized indoles **1–4** were characterized by heteronuclear NMR spectroscopy. NOE based conformational analysis revealed that all four receptors exhibit conformational preorganization in acetone solution, where *anti–anti* conformer is predominant. Such an orientation places C2α and C7β carbonyl groups in the proximity of indole H1 proton which leads to stabilization by intramolecular hydrogen bonds. Anion-induced chemical shift changes demonstrate that binding of halides (chloride, bromide) takes place predominantly at H1 proton. Receptors **2** and **3** with urea and thiourea moieties offer more donor groups and therefore the anions interact with H1, H7α and H7γ protons. Nitrate anions favour interaction with H7α and H7γ urea and thiourea protons, whereas acetate anions interact strongly with all available hydrogen bond donors. Comparison of the NOE enhancements in the absence and in the presence of anions revealed conformational changes of receptors **1–4** induced by complexation of chloride, bromide and acetate anions. Anion–receptor complexes preferably adopt a *syn–syn* conformation where all NH protons are spatially close and involved in interaction with anions. However, no conformational changes were observed upon addition of nitrate anions to **1–4**. Our study demonstrates that the indole ring is an intriguing scaffold for the design of novel anion receptors in order to tune affinities and selectivities for anions.

## Acknowledgements

The authors gratefully acknowledge the Slovenian Research Agency (ARRS, Program No. P1-0242), the EPSRC and COST D31/0021/05 for financial support.

## References

1 *Supramolecular Chemistry of Anions*, ed. A. Bianchi, K. Bowman-James and E. García-España, Wiley-VCH, New York, 1997.
2 J. L. Sessler, P. A. Gale and W. S. Cho, *Anion Receptor Chemistry*, Royal Society of Chemistry, Cambridge, UK, 2006.
3 P. D. Beer and P. A. Gale, *Angew. Chem., Int. Ed.*, 2001, **40**, 486–516.
4 K. Bowman-James, *Acc. Chem. Res.*, 2005, **38**, 671–678.
5 R. Vilar, *Eur. J. Inorg. Chem.*, 2008, 357–367.
6 P. A. Gale, S. E. García-Garrido and J. Garric, *Chem. Soc. Rev.*, 2008, **37**, 151–190.
7 C. Caltagirone and P. A. Gale, *Chem. Soc. Rev.*, 2009, **38**, 520–563.
8 J. W. Pflugrath and F. A. Quiocho, *J. Mol. Biol.*, 1988, **200**, 163–180.
9 K. H. G. Verschueren, F. Seljee, H. J. Rozeboom, K. H. Kalk and B. W. Dijkstra, *Nature*, 1993, **363**, 693–698.
10 P. A. Gale, *Chem. Commun.*, 2008, 4525–4540.
11 C. Caltagirone, P. A. Gale, J. R. Hiscock, S. J. Brooks, M. B. Hursthouse and M. E. Light, *Chem. Commun.*, 2008, 3007–3009.
12 C. Caltagirone, J. R. Hiscock, M. B. Hursthouse, M. E. Light and P. A. Gale, *Chem.–Eur. J.*, 2008, **14**, 10236–10243.
13 J.-m. Suk, M. K. Chae, N.-K. Kim, U.-l. Kim and K.-S. Jeong, *Pure Appl. Chem.*, 2008, **80**, 599–608.
14 G. W. Bates, P. A. Gale and M. E. Light, *Chem. Commun.*, 2007, 2121–2123.
15 F. M. Pfeffer, K. F. Lim and K. J. Sedgwick, *Org. Biomol. Chem.*, 2007, **5**, 1795–1799.
16 J. L. Sessler, D. G. Cho and V. Lynch, *J. Am. Chem. Soc.*, 2006, **128**, 16518–16519.
17 K.-J. Chang, M. K. Chae, C. Lee, J.-Y. Lee and K.-S. Jeong, *Tetrahedron Lett.*, 2006, **47**, 6385–6388.
18 K.-J. Chang, D. Moon, M. S. Lah and K.-S. Jeong, *Angew. Chem., Int. Ed.*, 2005, **44**, 7926–7929.
19 K. J. Chang, B. N. Kang, M. H. Lee and K.-S. Jeong, *J. Am. Chem. Soc.*, 2005, **127**, 12214–12215.
20 S. O. Kang, R. A. Begum and K. Bowman-James, *Angew. Chem., Int. Ed.*, 2006, **45**, 7882–7894.
21 J. L. Sessler, S. Camiolo and P. A. Gale, *Coord. Chem. Rev.*, 2003, **240**, 17–55.
22 C. R. Bondy and S. J. Loeb, *Coord. Chem. Rev.*, 2003, **240**, 77–99.
23 D. A. Jose, A. Singh, A. Das and B. Ganguly, *Tetrahedron Lett.*, 2007, **48**, 3695–3698.
24 D. E. Gomez, L. Fabbrizzi, M. Licchelli and E. Monzani, *Org. Biomol. Chem.*, 2005, **3**, 1495–1500.
25 G. W. Bates, Triyanti, M. E. Light, M. Albrecht and P. A. Gale, *J. Org. Chem.*, 2007, **72**, 8921–8927.