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Transmembrane anion transport mediated by halogen bonding and hydrogen bonding triazole anionophores

Laura E. Bickerton, Alistair J. Sterling, Paul D. Beer, Fernanda Duarte*, Matthew J. Langton*

Transmembrane ion transport by synthetic anionophores is typically achieved using polar hydrogen bonding anion receptors. Here we show that readily accessible halogen and hydrogen bonding 1,2,3-triazole derivatives can efficiently mediate anion transport across lipid bilayer membranes with unusual anti-Hofmeister selectivity. Importantly, the results demonstrate that the iodo-triazole systems exhibit the highest reported activity to date for halogen bonding anionophores, and enhanced transport efficiency relative to the hydrogen bonding analogues. In contrast, the analogous fluoro-triazole systems, which are unable to form intermolecular interactions with anions, are inactive. The halogen bonding anionophores also exhibit a remarkable intrinsic chloride over hydroxide selectivity, which is usually observed only in more complex anionophore designs, in contrast to the readily accessible acyclic systems reported here. This highlights the potential of iodo-triazoles as synthetically accessible and versatile motifs for developing more efficient anion transport systems. Computational studies provide further insight into the nature of the anion-triazole intermolecular interactions, examining the origins of the observed transport activity and selectivity of the systems, and revealing the role of enhanced charge delocalisation in the halogen bonding anion complexes.

Introduction

The development of supramolecular anionophores for transmembrane ion transport is driven by their potential utility as tools for studying ion transport processes and as therapeutics for diseases arising from mis-regulation of protein ion channels.1–4 Significant effort has been devoted to designing mobile carrier systems with high anion transport activity in vesicles (particularly for chloride), and more recently, in cells.5–10 As with naturally occurring ion transporters, anion selectivity is crucial, and depends on the delicate balance between transporter anion binding selectivity and anion desolvation. Selectivity for chloride over proton transport (or the functionally equivalent hydroxide) is particularly necessary for applications in which dissipation of transmembrane pH gradients must be avoided. The naturally occurring anionophore, prodigiosin,11 and its synthetic analogues,12 are known to uncouple H+/ATPases or neutralise organelles by facilitated H+/Cl− symport, and for this reason are promising candidates for anti-cancer agents. Conversely, anionophores designed for treating diseases arising from mis-regulated chloride channels, including cystic fibrosis and Bartter syndrome, must have high Cl− > H+/OH− selectivity to avoid toxicity arising from disrupting transmembrane pH gradients. However, designing transporters with such selectivity remains a significant challenge. Polar NH hydrogen bond (HB) donors (such as ureas or squaramides) which are typically used in supramolecular anionophores exhibit no intrinsic Cl− > H+/OH− selectivity.10 Improved chloride selectivity has been achieved by designing anionophores which encapsulate the anion, such as tripod or cholapod-based receptors, but in general, more acidic NH HB-donors that are required for anion binding and efficient transport lead to decreased Cl− > H+/OH− selectivity.10 C–H hydrogen bonding or C–I halogen bonding (XB)13–15 interactions have recently emerged as potential alternatives to the classical N–H or O–H HB interactions in particular can exhibit superior anion binding affinity in competitive polar organic17 or aqueous media18–23 to hydrogen bonding,18–23 pointing to its potential utility in transmembrane anion transport systems. However, to date only a handful of XB-mediated anion transport systems have been reported, exploiting iodo-perfluoro alkane and arene derivatives for anion recognition and transport,24–28 and examples of transporters mediating anion transport solely through C–H HB interactions are also rare.16,29–31 The potential power of XB for anion transport was exemplified by Matile and co-workers who reported that gaseous iodo trifluoromethane facilitates anion transport when bubbled through the vesicle solution.25

Herein we show that simple acyclic XB or HB triazoles (Figure 1) can efficiently mediate anion transport across lipid bilayer membranes with an unusual anti-Hofmeister selectivity. Importantly, we demonstrate unprecedented activity for XB...
anionophores with up to two orders of magnitude improvement over the previously reported iodo-perfluorobenzene systems, and reveal their remarkable intrinsic C=O-H+ selectivity. Iodo- and proto-triazoles are versatile motifs readily accessible via Cu-catalysed azide-alkyne click chemistry, and these results demonstrate their potential for applications in synthetic anion transport systems.

Results and discussion

Synthesis and anion recognition properties

Acyclic 5-iodo/proto/fluoro-1,4-disubstituted-1,2,3-triazoles, of the general form shown in Figure 1, were prepared as minimalistic scaffolds with which to explore the intrinsic activity and selectivity of the hydrophobic C-I XB- and C-H HB-selectivity. The family of compounds spans a range of iodo-triazole derivatives, and their proto-triazole analogues, with varied electron-withdrawing/donating substituents on the aryl group and alkyl chain lengths. The library of 5-iodo-1,4-disubstituted-1,2,3-triazoles (compounds 1b-9b) were prepared by Cu-catalysed azide-alkyne cycloaddition of the respective alkyl azide and iodo-aryl-alkyne, in the presence of tris(1-benzyl-1H-1,2,3-triazol-4-yl)methyl]amine (TBTA) (Scheme 1). The analogous proto-triazoles (compounds 1a-9a) were prepared from the corresponding alkyne. As control compounds, the fluoro-triazole analogues of 1 and 9, namely 1c and 9c, which are unable to form HB or XB interactions, were also prepared from the corresponding iodo-triazole by heating with potassium fluoride. Full synthetic procedures and characterisation are available in the ESI.

The anion recognition capability of representative proto-triazole 1a and iodo-triazole 1b were first investigated by $^1$H NMR binding titrations with Bu$_4$N$^+$ X$^-$ in d$_6$-acetone, monitoring the binding induced chemical shift perturbations of the proto-triazole H and adjacent aryl protons. For each anion, the data could be fitted to a 1:1 binding isotherm (see ESI) and the 1:1 stoichiometric association constants determined using the Bindfit program (Table 1).$^{33,34}$ The data revealed an overall selectivity trend of Cl$^->$Br$^->$I$^-$ for both 1a and 1b, and approximately one order of magnitude enhancement of halide binding affinity for the XB iodo-triazole receptor 1b compared to HB proto-triazole 1a. For both compounds, no measurable binding of nitrate was observed under these conditions.

Transmembrane anion transport activity

The ability of anionophores 1–9 a/b to mediateOH$^+/Cl^-$ transmembrane anion transport was first determined in 1-palmitoyl-2-oleoyl-sn-glycero-3-phosphocholine large unilamellar vesicles (POPC LUVs), loaded with 8-hydroxypyrene-1,3,6-trisulfonate (HPTS) and buffered to pH 7.0 in NaCl solution. HPTS is a pH-sensitive fluorophore, which allows for ratiometric determination of the internal pH of the LUVs. A pH gradient was applied across the membrane by addition of a base pulse, followed by addition of the carrier as a DMSO solution (<0.5% v/v). The ability of the anionophore to dissipate the pH gradient by transmembrane OH$^-$/Cl$^-$ exchange was determined by recording the change in the HPTS emission, $I_{rel}(\lambda_{em} = 510 \text{ nm})$, with time following excitation at $\lambda_{ex}$=405/465 nm (Figure 2a). At the end of each experiment, excess detergent (Triton X-100) was added to lyse the vesicles for calibration of the emission intensity. The HPTS assay was used to determine the concentration dependence of the activity of each anion carrier. Figure 2a shows representative data for the activity of compound 1b at a range of concentrations. The fractional activities (y, the relative intensity at 288 s, immediately prior to lysis) were plotted as a function of concentration (Figure 2b) and fitted to the Hill equation (Equation 1). Original data for all other compounds is available in the ESI.

| Table 1. Binding affinities for triazole derivatives 1a and 1b. |
|---|---|---|
| Anion | $K_a / M^{-1}$ 1a | $K_a / M^{-1}$ 1b |
| Cl$^-$ | 17 ± 1 | 197 ± 14 |
| Br$^-$ | 17 ± 7 | 110 ± 35 |
| I$^-$ | < 3 | 38 ± 9 |
| NO$_3^-$ | > | > |

*Anions added as the TBA salt in d$_6$-acetone, data fitted to a 1:1 binding model using Bindfit. Perturbations too small to obtain binding constant. Errors at the 95% confident limit.
The Hill equation here is used to describe the dependence of the fractional activities \( y \) on the \( n^{th} \) power of the anionophore concentration \( x \), and facilitates comparison of the relative activities of transporters through an effective concentration value \( (EC_{50}) \) required to reach 50% activity in the given assay (Equation 1). Compatibility with the Hill equation reveals endergonic self-assembly of the active supramolecule, and Hill coefficients \( n > 1 \) are indicative of the stoichiometry of the unstable supramolecular assembly that exists in the presence of an excess of uncomplexed anionophores.\(^{35}\)

**Table 2. Characteristics of the HB and XB anion transporters.**

<table>
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<th></th>
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<td>EC(_{50}) / (\mu M)</td>
<td>2.0 (0.1)</td>
<td>1.3 (0.1)</td>
<td>0.9 (0.1)</td>
<td>1.1 (0.1)</td>
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<td>(n^p)</td>
<td>2.4 (0.3)</td>
<td>2.1 (0.4)</td>
<td>3.7 (0.8)</td>
<td>2.6 (0.4)</td>
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<td>cloP(_{clog})</td>
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<td>6.1</td>
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<td>EC(_{50}) / (\mu M)</td>
<td>6.2 (0.2)</td>
<td>3.3 (0.2)</td>
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<tr>
<td>(n^p)</td>
<td>5.6 (0.5)</td>
<td>2.0 (0.2)</td>
<td>(\cdots)</td>
<td>(\cdots)</td>
<td>(\cdots)</td>
<td>(\cdots)</td>
</tr>
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<tr>
<td>EC(_{50}) / (\mu M)</td>
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<td>1.8 (0.2)</td>
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<td>3.6 (0.3)</td>
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<tr>
<td>(n^p)</td>
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<td>3.1 (0.7)</td>
<td>3.6 (0.7)</td>
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<td>7.2 (1.2)</td>
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<td>cloP(_{clog})</td>
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<td>4.8</td>
<td>5.2</td>
<td>5.6</td>
<td>6.0</td>
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\(^*\)Effective concentration to reach 50% of maximal activity in the HPTS assay, determined from Hill analysis of the relative fluorescence intensity at \( t = 288 \) s immediately prior to vesicle lysis. Experiments conducted in LUVs of POPC (mean diameter 200 nm) loaded with 1 mM HPTS, NaCl (100 mM) and buffered at pH 7.0 with 10 mM HEPES. For compounds of low activity, estimated lower bounds for the EC\(_{50}\) value are given. \(^\dagger\)Hill coefficient. \(^\circ\)Calculated log \( P \). \(^\ddagger\)Insufficient activity for Hill analysis.

**Figure 2.** Anion transport by XB transport 1b in the HPTS assay. (a) Change in ratiometric emission \((\lambda_{ex} = 510 \text{ nm}; \lambda_{em} = 405 \text{ nm}, \lambda_{em} = 460 \text{ nm})\) upon addition of 1b in DMSO (0.05 – 50 \( \mu M \)) to POPC vesicles containing 1 mM HPTS, 100 mM internal and external NaCl, buffered with 10 mM HEPES at pH 7.0. A \( \Delta pH \) gradient is generated by addition of a NaOH base pulse (0.5 M), followed by 1b at \( t = 0 \). Vesicle lysis by Triton X-100 calibrates the assay. (b) Dependence on fractional activities \( y \), the relative intensity at \( t = 288 \) s just prior to lysis, on concentration of 1b (black squares), and fit to the Hill equation (black line).

**Equation 1.**

\[
y = y_0 + (y_{max} - y_0) \cdot \frac{x^n}{EC_{50} + x^n}
\]

EC\(_{50}\) values, hill coefficients and calculated partition coefficients (cloP) values for the proto- and iodo-triazoles are shown in Table 2. cloP \( P \) values were calculated using the VCCLab software.\(^{36}\)

In general, the electron withdrawing 3,5-bis(trifluoromethyl)benzene and 4-nitrobenzene substituents led to excellent activity despite the simplicity of the anionophores (compounds 1–3, and 7–9, respectively). The electron deficient halogen bonding transporters 1b and 9b are approximately 3x more active than the most active XB anionophore reported to date, iodoperfluorohexane (EC\(_{50} = 3.1 \mu M\))\(^{25}\), and notably over two orders of magnitude more active than the archetypal XB donor iodoperfluorobenzene under comparable assay conditions (EC\(_{50} = 260 \mu M\))\(^{25}\). Electron donating 4-butylbenzene substituents (compounds 4–6) conversely decreased activity, consistent with the expected decreased XB and HB donor strength. The correlation between activity and log \( P \) is complex, because transport efficiency is also strongly dependent on other factors including anion binding affinity and molecular size, which are not easily decoupled.\(^{37}\)

Nevertheless, the observed trend of maximum activity of compounds with log \( P \) of 4–6, and reduced activity observed for compounds with higher and lower lipophilicities, is broadly consistent with previous reports on the role of lipophilicity on HB-mediated anion carriers.\(^{38}\)

For compounds in which both XB and HB derivatives fall within these optimum parameters (compounds 1, 4, 7 and 8), a general enhancement of activity of around 2x, and up to 8x, is observed for XB systems over their HB analogues. The most lipophilic transporters (3, 5, 6) are
methyl-truncated analogues
mechanism
To confirm the role of the C–I XB and C–H HB interactions in the anion transport process, we investigated the anion transport capability of fluoro-triazole derivative 1c, which is unable to donate XB or HB interactions through the triazole motif. Inactivity of 1c in the HPTS assay confirmed the role of the XB and HB intermolecular interactions in the transport processes (Figure 4a). Similar behaviour was observed for nitro-phenyl fluoro-triazole derivative 9c.

Inactivity of any of the transport systems 1-9 reported here in the carboxyfluorescein dye leakage assay rules out nonspecific leakage by these systems (ESI). Replacement of the zwitterionic POPC lipids in the HPTS assay with anionic egg yolk phosphatidylglycerol (EYPG) lipids led to a significant decrease in observed activity, consistent with the requirement for formation of an anionic complex in the rate limiting transport process. Transport activity at 25 °C with XB transporter 1b and dipalmitylophosphatidylcholine (DPPC) lipids, under otherwise identical conditions, was negligible (Figure 4b). The lipid gel phase inhibits translation of mobile carriers through the membrane which are otherwise mobile in a fluid lipid phase. The gel to fluid phase transition for DPPC lipids is 41°C, and repeating the assay above this temperature (45°C) restored anion transport activity. The observed temperature dependence is indicative of a mobile carrier mechanism, and rules out formation of a membrane-spanning supramolecular channel structure whose activity would be independent of the lipid phase.

Evidence for XB/HB-anion interactions and mobile carrier mechanism
Calculation of the electrostatic potential (ESP) surface for methyl-truncated analogues of 1a/b, 4a/b and 7a/b (denoted 1a’/b’, 4a’/b’ and 7a’/b’ respectively) reveals the characteristic iodine-centred sigma-hole associated with halogen bonding donors, and the accumulation of δ+ surrounding the triazole protons active in hydrogen bonding (Figure 3). Comparison of the Hammett σ values for each substituent demonstrate that the more electron-withdrawing substituents result in a more positive value for the ESP maximum, leading to a stronger XB-/HB-anion intermolecular interaction. This correlates with the observed greater activity of 1a/b and 7a/b over 4a/b (Table 2).

Inactivity of any of the transport systems 1-9 reported here in the carboxyfluorescein dye leakage assay rules out nonspecific leakage by these systems (ESI). Replacement of the zwitterionic POPC lipids in the HPTS assay with anionic egg yolk phosphatidylglycerol (EYPG) lipids led to a significant decrease in observed activity, consistent with the requirement for formation of an anionic complex in the rate limiting transport process. Transport activity at 25 °C with XB transporter 1b and dipalmitylophosphatidylcholine (DPPC) lipids, under otherwise identical conditions, was negligible (Figure 4b). The lipid gel phase inhibits translation of mobile carriers through the membrane which are otherwise mobile in a fluid lipid phase. The gel to fluid phase transition for DPPC lipids is 41°C, and repeating the assay above this temperature (45°C) restored anion transport activity. The observed temperature dependence is indicative of a mobile carrier mechanism, and rules out formation of a membrane-spanning supramolecular channel structure whose activity would be independent of the lipid phase.

Anion selectivity
Dissipation of the transmembrane pH gradient measured in the HPTS assay by the carrier species can in principle occur through either cation (H+/M+) or anion (OH-/A-) antiport (exchange), or H+/A- symport (co-transport) mechanisms. The activities of HB transporter 1a and XB transporter 1b were not affected by isosmolar replacement of the external Na+ cation with Li+, K+, Rb+, Cs+ (see ESI), indicative of selective anion transport (OH-/A- antiport or H+/A- symport) rather than H+/M+ cation antiport. Further evidence to support the cation-independent transport mechanism was provided by conducting analogous experiments in the presence of sodium gluconate, a large hydrophilic anion (ESI). The absence of detectable transport indicates that, as expected, neither OH-/gluconate antiport or H+/gluconate symport mechanisms are active because of the insurmountable dehydration penalty of gluconate, and also that the alternative H+/Na+ cation antiport process is negligible.

OH-/A- antiport and H+/A- symport mechanisms are functionally equivalent and cannot be distinguished through these transport assays. However, the low basicity of triazole derivatives (pKb ~ 0-1) suggests that H+/A- symport (achieved via triazole protonation and XB-/HB-mediated anion recognition) is unlikely to contribute to any significant extent to the ion transport process at neutral pH. This is consistent with
The activity of previously reported XB iodofluoroalkene/arene transporters in the same assay, which do not possess any basic atoms and are therefore most likely operate through an anion antiport mechanism.\textsuperscript{25} For simplicity, we refer to the transport process from here on in as OH\(^{-}/A^{-}\) antiport.

To examine the relative rates of Cl\(^{-}\) vs OH\(^{-}\) transport, we repeated the HPTS assay with the addition of carbonyl cyanide-\(p\)-trifluoromethoxyphenylhydrazone (FCCP), a weak acid protonophore at a low concentration (0.25 \(\mu\)M) insufficient to cause activity alone. FCCP transports protons via transmembrane shuttling of both protonated and deprotonated forms of the molecule. Enhancement of activity by FCCP is indicative of a rate limiting electrogenic OH\(^{-}\) transport process, because FCCP decouples the anionophore-mediated OH\(^{-}/\)Cl\(^{-}\) antiport process (Figure 5a) by facilitating rapid electrogenic H\(^{+}\) transport in an overall coupled Cl\(^{-}\)/H\(^{+}\) symport process (Figure 5b). In this scenario, the observed rate of pH gradient dissipation reports on the rate-limiting electrogenic Cl\(^{-}\) transport process. As such, if the activity of a given carrier is invariant to FCCP addition, Cl\(^{-}\) transport must therefore be rate limiting in the antiport mechanism (i.e. slower than electrogenic OH\(^{-}\) transport). The EC\(_{50}\) values for compounds 1a and 1b with FCCP are shown in Table 3. The activity of HB anionophore is invariant to FCCP addition, demonstrating rapid OH\(^{-}\) transport and rate limiting Cl\(^{-}\) transport by 1a. In contrast, a four-fold increase in activity for XB transporter 1b is observed in the presence of FCCP, revealing rate limiting OH\(^{-}\) transport in its absence. The ratio of EC\(_{50}\) values in the absence and presence of FCCP (Table 3, column 4) therefore reports on the relative Cl\(^{-}\) > OH\(^{-}\) selectivity of the system (for the given conditions and ion transport gradient), and reveals the unusual Cl\(^{-}\) > OH\(^{-}\) selectivity of the XB transporter 1b. Notably, transporter 1b in the presence of FCCP achieved nanomolar activity (EC\(_{50}\) = 300 nM) in the HPTS assay, which to the best of our knowledge is the highest activity reported to date for an XB anion transporter under comparable experimental conditions. This highlights the potential of iodo-triazoles as synthetically accessible and versatile motifs for developing more efficient anion transport systems.

In the presence of FCCP, the observed transport kinetics report on the rate limiting A\(^{-}\) transport for a given anionophore. This allows the relative selectivity of both XB and HB transporters for each anion to be investigated, enabling direct comparison of the relative rates of electrogenic A\(^{-}\) transport (Figure 6). HB transporter 1a exhibits an overall selectivity profile of Cl\(^{-}\) > Br\(^{-}\) > I\(^{-}\) > NO\(_3\)\(^{-}\) (Figure 6, black data). This selectivity is remarkable because in general hydrogen bonding anion transporters exhibit Hofmeister selectivity, whereby more weakly hydrated anions are selectively transported across the membrane due to ease of desolvation.\textsuperscript{44-47} The analogous XB anionophore 1b exhibits a selectivity profile of NO\(_3\)\(^{-}\) > Cl\(^{-}\) > Br\(^{-}\) > I\(^{-}\). Comparison of the relative activities for both transporters in the absence of FCCP allows placement of OH\(^{-}\) into the overall selectivity trends, revealing OH\(^{-}\) > Cl\(^{-}\) > Br\(^{-}\) > I\(^{-}\) > NO\(_3\)\(^{-}\) behaviour for 1a and NO\(_3\)\(^{-}\) > Cl\(^{-}\) > Br\(^{-}\) > OH\(^{-}\) > I\(^{-}\) behaviour for 1b. Similar selectivity trends for analogous compounds 9a and 9b were also observed in these assays (see ESI).

Overall these transport experiments reveal (i) enhanced activity of halogen bonding anionophores vs hydrogen bonding analogues across all anions, (ii) unusual Cl\(^{-}\) > OH\(^{-}\) selectivity for the halogen bonding anionophores and (iii) anti-Hofmeister bias for halide transport, demonstrating the dominant role of ion binding to the anionophore and correlating with NMR determined anion binding affinities (see Table 1).\textsuperscript{45}

---

### Table 3. FCCP dependence of transporters 1a and 1b.

<table>
<thead>
<tr>
<th>Compound</th>
<th>EC(_{50})/ (\mu)M(^{a})</th>
<th>EC(_{50})CCP/ (\mu)M(^{b})</th>
<th>EC(_{50})</th>
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<tr>
<td>1a</td>
<td>2.0 ± 0.1</td>
<td>2.2 ± 0.5</td>
<td>0.9</td>
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<tr>
<td>1b</td>
<td>1.3 ± 0.1</td>
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\(^{a}\) EC\(_{50}\) in the absence of FCCP. \(^{b}\) EC\(_{50}\) in the presence of 0.25 \(\mu\)M FCCP.
Computational studies into anionophore-anion interactions

Computational studies were used to probe the binding energies and structures of the anionophore-anion complexes. Calculation of the relative binding enthalpies for both 1:1 and 2:1 model anionophore-anion complexes in CHCl₃ implicit solvent as a membrane-mimetic environment reveals that ∆Hbind(2:1) > 2 × ∆Hbind(1:1) (Figure 7a), and that 2:1 binding is enhanced by π-stacking between the arenes (see ESI). Bidentate binding was found to be preferred in the model 1:1 HB system 1a′ involving hydrogen bonds from both the alkyl C(sp³)–H₁ and triazole C(sp²)–H₂ (Figure 7b). This finding is in line with the result of the ESP calculations in Figure 3. Monodentate XB interactions are observed with 1b′. Interestingly, our results indicate that in the 2:1 complex, four HB C–H⋯anion interactions per anion exists for 1a′ (Figure 7c), while only one XB C–I⋯anion interaction is present for 1b′ (Figure 7c).

Additionally, bond order and second-order perturbation theory analysis, which allows the quantification of bond energies (E⁽²⁾) indicates that addition of the second transporter ligand weakens the total XB interactions compared with the 1:1 complex (E⁽²⁾XB = 40.6 vs 30.7 kcal mol⁻¹). However, this is partially compensated by dispersion interactions between the two transporter ligands (E⁽¹⁾₁⁻₂ = 16.0 kcal mol⁻¹) and a C–H interaction (10.1 kcal mol⁻¹), which leads to an overall larger stabilisation energy (E⁽²⁾total = 56.8 kcal mol⁻¹).

Analysis of the partial charges on each anion in the 1:1 and 2:1 binding modes suggest a correlation between the rate of transport and the ability of the transport to stabilise the anion charge (Figure 8). For example, for Cl⁻, Br⁻ and I⁻, the negative charge is more effectively delocalised over the XB anionophore than the HB analogue. This is consistent with previous results from Donor K-edge X-ray Absorption Spectroscopy experiments which revealed significant covalency in the XB interaction between related iodo-triazole XB donors and chloride anions and a C–H interaction (10.1 kcal mol⁻¹), which leads to an overall larger stabilisation energy (E⁽²⁾total = 56.8 kcal mol⁻¹).

Figure 7. Characterisation of 1:1 and 2:1 binding of (a) 1a′ and (b) 1b′ to chloride. Enthalpies, entropies and free energies of binding (kcal mol⁻¹) were calculated at the [SMD(CHCl₃)-DLPNO-CCSD(T)/def2-TZVP (ma-def2-TZVP on Cl, I)] level of theory. Mayer bond orders (dashed lines), their associated HB/XB bond energies (E⁽²⁾HB/XB), and interaction energy between ligands (E⁽¹⁾₁⁻₂) in kcal mol⁻¹ were calculated at the [NBO/SMD(CHCl₃)-ωB97X-D3/def2-SVP (ma-def2-SVP on Cl, I)] level of theory. Free energies were calculated using a quasi-RRHO approximation and corrected for a 1 M standard state at 298.15 K.

Figure 8. Partial charges on each anion for 1:1 anionophore-anion complexes of 1a′ and 1b′, calculated using natural population analysis (NPA) at the [NBO/SMD(CHCl₃)-ωB97X-D3/def2-SVP (ma-def2-SVP on anion)] level of theory.
(comparable to that in transition metal chloride complexes), in contrast to proto-triazole analogues with negligible charge transfer contribution to the HB interaction. Improved charge delocalisation over the anion-triazole complex is expected to decrease the barrier to transport across the hydrophobic membrane (Figure 8), and this is consistent with the experimentally observed selectivity trend of the XB anionophores enhancing rates of anion transport over the analogous HB system. A different behaviour is observed for nitrate, where the observed efficient nitrate transport does not correlate with the increased calculated partial charge. We suggest that this behaviour is due to the greater number of thermally-accessible conformations for the binding of the trigonal nitrate anion, each of them having a different charge distribution. In this case, the most stable conformer in solution is likely not the most active for membrane transport (see ESI).

Conclusions

We have shown that acyclic halogen bonding iodo-triazoles can efficiently transport anions across lipid bilayer membranes, with enhanced activity in comparison to hydrogen bonding proto-triazole analogues. Despite the relative simplicity of the design, the iodo-triazole systems are the most active halogen bonding anionophores reported to date. Anion transport experiments reveal an unusual anti-Hofmeister selectivities, and a remarkable intrinsic Cl\(^{-}\) > OH\(^{-}\) selectivity for the halogen bonding transport systems. Experimental and computational studies provide further insight into the binding modes of the anion-anionophore complexes. Calculations demonstrate that the strength of the non-covalent interactions are sufficient to overcome the entropic cost of 2:1 complex formation, and reveal the remarkable ability of the halogen bonding anionophores to delocalise the anion charge over the complex, which correlates with the enhanced anion transport capability observed by experiment. This work demonstrates the utility of the synthetically accessible and highly versatile XB and HB triazole motif for anionophore design. The observed Cl\(^{-}\) > OH\(^{-}\) selectivity of the iodotriazole derivatives also suggest that such motifs may provide the starting point for designing novel ionophores with enhanced selectivity for potential future application in channelopathy therapeutics.

Conflicts of interest

There are no conflicts to declare

Acknowledgements

A.J.S. thanks the EPSRC Centre for Doctoral Training in Synthesis for Biology and Medicine for a studentship (EP/L015838/1), generously supported by AstraZeneca, Diamond Light Source, Defence Science and Technology Laboratory, Evotec, GlaxoSmithKline, Janssen, Novartis, Pfizer, Syngenta, Takeda, UCB and Vertex. A.J.S. also thanks the Oxford-Radcliffe Scholarship for a studentship. A.J.S. and F.D. thank the EPSRC Tier-2 National HPC Facility Service (http://www.cirrus.ac.uk), and the EPSRC Centre for Doctoral Training in Theory and Modelling in Chemical Sciences (EP/L015722/1) for providing access to the Dirac cluster at Oxford. M.J.L. acknowledges funding from the Royal Society. F.D. and M.J.L. thank the John Fell Oxford University Press Research Fund for financial support. M.J.L. is a Royal Society University Research Fellow.

Notes and references

§ Truncated alkyl chains to methyl were used for computational studies. This does not affect the calculated ESP (see ESI for details).

§§ Detailed comparison between the measured association constants in solution and transport kinetics in membranes should be avoided, given that the observed rate of transport is also dependent on many other factors including desolvation, anionophore-ion complex mobility in the membrane, and competing binding interactions of the transporter with the phosphate head groups. 57

§§§ Discussed here are the lowest energy conformers obtained from the sampling procedure described in the ESI.


Halogen and hydrogen bonding 1,2,3-triazole derivatives efficiently mediate anion transport across lipid bilayer membranes with unusual anion selectivity profiles