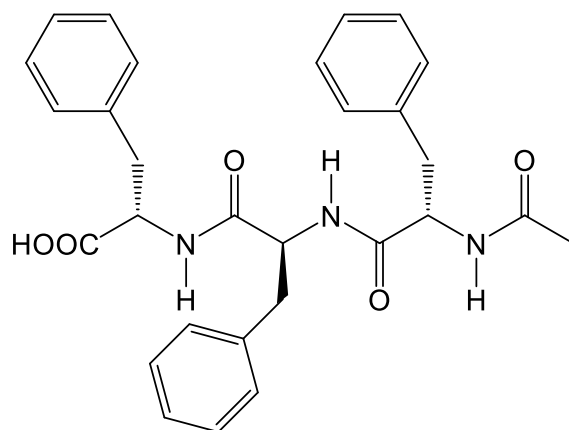


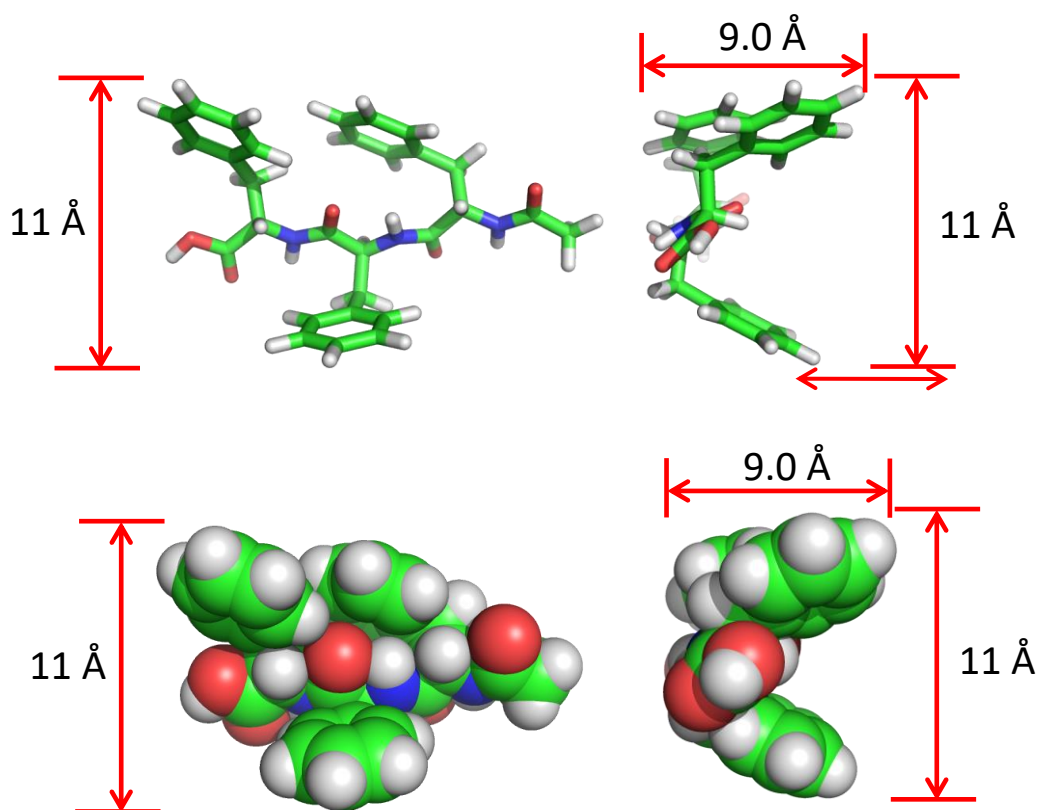
1.	General Remarks.....	S2
2.	Figure S1. Dimensionality of the Tripehnylalaine Side Chains.....	S3
3.	Synthetic Scheme .....	S4
4.	Experimental Procedures .....	S6
5.	MALDI-TOF Data for Polymers .....	S11
6.	Molecular Dynamics Simulations .....	S14
7.	Ion Transport Activity using HPTS Assay.....	S21
8.	Cation Transport under High Salt Gradient Environment .....	S28
9.	Chloride Transport Using the SPQ Assay .....	S30
10.	FCCP-Coupled HPTS Assay .....	S31
11.	The CF Dye Leakage Assay .....	S32
12.	Anion Transport Activity by 1a-Bn .....	S33
13.	Single Channel Current Measurement .....	S34
14.	Determination of Anion Selectivity .....	S35
15.	Determination of Initial Rate Constants .....	S42
16.	Effect of Heavy Iodine Atom .....	S43
17.	$^1\text{H}/^{13}\text{C}$ NMR Spectra .....	S44
18.	References.....	S50

## General Remarks

All the chemicals were purchased from commercial sources and were used as received unless stated otherwise. Solvents were dried by standard methods prior to use or purchased as dry. All reactions were carried out under the nitrogen atmosphere. Thin layer chromatography (TLC) was carried out with silica gel coated plates and column chromatography was performed over silica gel (230-400 mesh) obtained from commercial suppliers. Egg yolk phosphatidylcholine (EYPC) lipid was purchased from Avanti Polar Lipids as a solution dissolved in chloroform (25 mg/mL). All buffer solutions were prepared from MilliQ water and pH of the buffer solutions were measured using Sartorius pH meter (PB-10). HEPES buffer, monobasic sodium phosphate salt, dibasic sodium phosphate salt, HPTS dye, SPQ dye, Triton X-100, NaOH and all inorganic salts of molecular biology grade were purchased from Sigma. Gel-permeation chromatography was performed on a column of Sephadex LH-20 gel (25 × 300 mm,  $V_0 = 25$  mL). Large unilamellar vesicles (LUV) were prepared from EYPC lipid by using mini extruder, equipped with a polycarbonate membrane either of 100 nm or 200 nm pore size, obtained from Avanti Polar Lipids. Fluorescence was recorded using fluorescence spectrophotometer (Hitachi, Model F-7100, Japan). The  $^1\text{H}$  and  $^{13}\text{C}$  spectra were recorded on Bruker ACF-400 spectrometer using either residual solvent signals as an internal reference or from internal tetramethylsilane on the  $\delta$  scale relative to dimethylsulphoxide- $d_6$  ( $\delta$  2.50 ppm), Methanol- $d_4$  ( $\delta$  3.31 ppm) for  $^1\text{H}$  NMR. The chemical shifts ( $\delta$ ) are reported in ppm. All fluorescence data were processed by Origin 8.5 and reaction schemes were prepared by ChemDraw Professional 15.0.

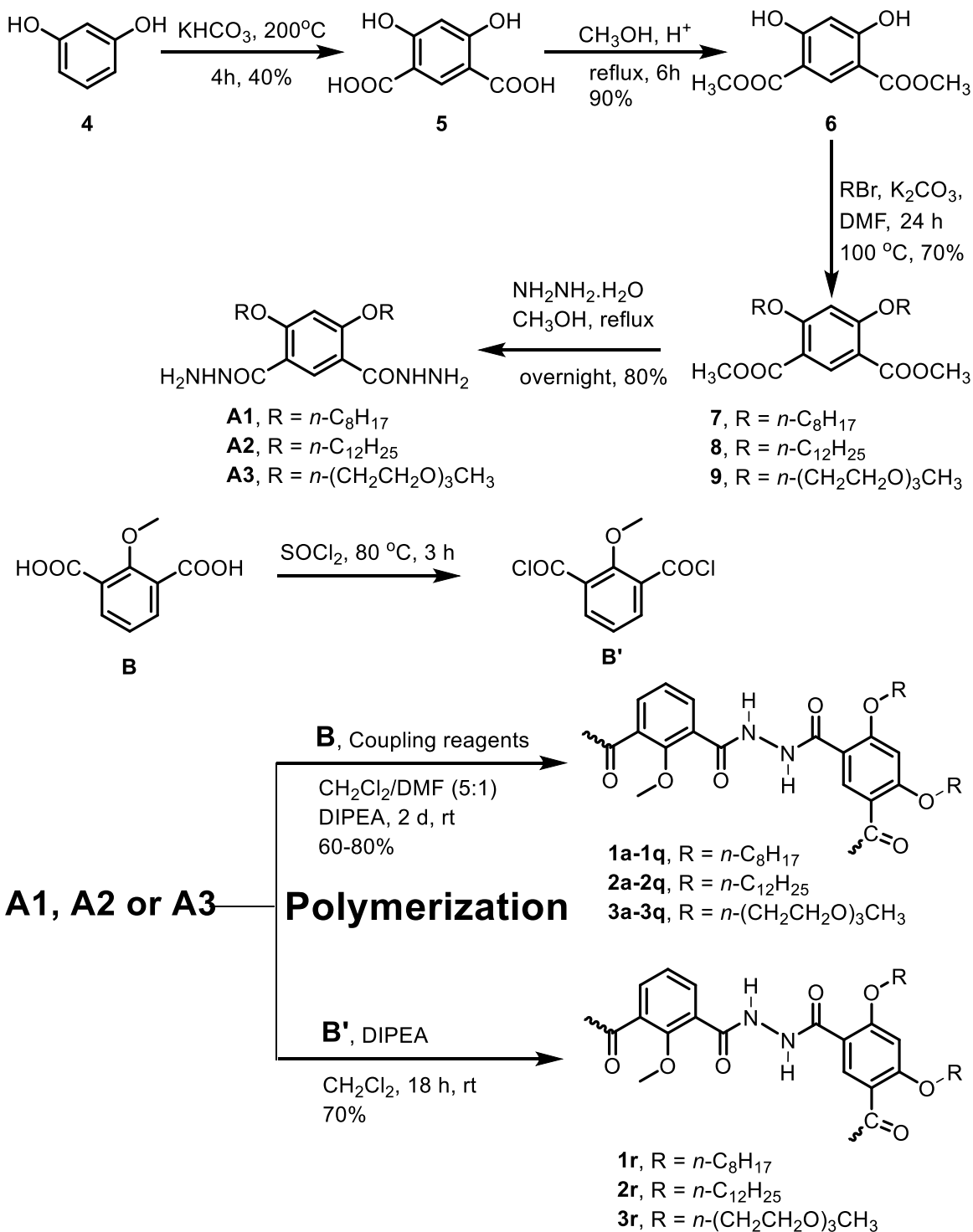


Triphenylalanine-based side chain

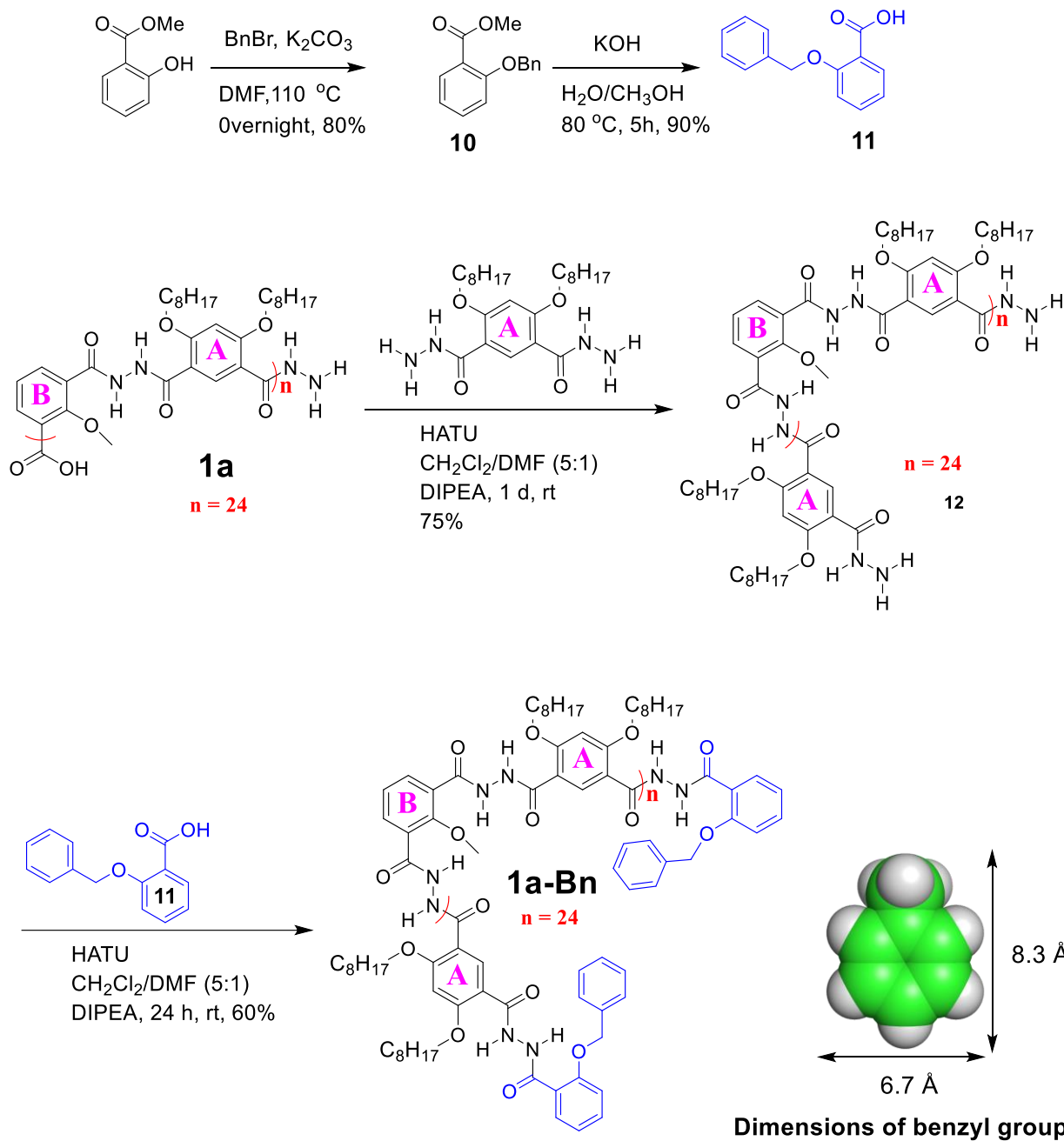


**Figure S1.** Dimensionality of the triphenylalanine-based side chains used by Li and Hou for constructing oligomeric and polymeric ion transporters for preferential transports of cations, rather than anions (See *J. Am. Chem. Soc.* **2014**, *136*, 13078).

## Synthetic Scheme



Scheme S1. Syntheses of polymers 1-3.



**Scheme S2.** Synthesis of **1a-Bn** modified with benzyl groups at its two helical ends. Having a dimension of 6.7 Å x 8.3 Å and pointing toward the interior, the introduced benzyl groups should partially block the cavity of about 6.5 Å across in polymer channel **1a** and diminish its ion transport activities.

## Experimental Procedures

**Preparation of monomeric building blocks A1-A3:** Compound **6** was prepared from compound **5** which was synthesized from resorcinol **4** following a reported protocol.<sup>[1]</sup> Compounds **A1**,<sup>[2]</sup> **A2**,<sup>[3]</sup> and **A3**<sup>[3]</sup> were prepared according to reported protocols.

**General protocol for synthesis of polymers using amide coupling agents:** In a 20 mL reaction vial, amine compound (**A1** - **A3**, 0.118 mmol) and 2-methoxyisophthalic acid **B** (0.118 mmol) were taken. Coupling reagents (0.354 mmol) were added to each reaction vial. This was followed by adding freshly distilled CH<sub>2</sub>Cl<sub>2</sub> (5 mL) and 1 mL DMF (dimethylformamide) in presence of nitrogen atmosphere. 100 μL DIEA (N,N-Diisopropylethylamine) was then added in the reaction mixture, and the solution was stirred for 48 hours at room temperature. After completion of reaction, solvent was evaporated to remove CH<sub>2</sub>Cl<sub>2</sub> and DMF. The obtained residue was washed with 10 mL MeOH/H<sub>2</sub>O (1:1), and off-white precipitate was observed. The obtained precipitate was filtered and washed with 10 mL H<sub>2</sub>O and 10 mL MeOH respectively to obtain polymers as off-white solid powder with yields of 60 - 80%. Molecular weights of all these polymers were determined by Gel Permeation Chromatography (GPC, **Table S1**).

**Table S1.** GPC data for all polymers classified as **1a-1r** with C<sub>8</sub>H<sub>17</sub> side chains, **2a-2r** with C<sub>12</sub>H<sub>25</sub> side chains and **3a-3r** with (C<sub>2</sub>H<sub>4</sub>O)<sub>3</sub>CH<sub>3</sub> side chains and synthesized from repeating units **A** and **B** using 18 different coupling conditions. PDI refers to the polydispersity index values.

Entry	Coupling Reagents	Polymers with C <sub>8</sub> H <sub>17</sub> side chain ( <b>1</b> )		Polymers with C <sub>12</sub> H <sub>25</sub> side chain ( <b>2</b> )		Polymers with (C <sub>2</sub> H <sub>4</sub> O) <sub>3</sub> CH <sub>3</sub> side chain ( <b>3</b> )	
		M <sup>n</sup> (KDa)	PDI	M <sup>n</sup> (KDa)	PDI	M <sup>n</sup> (KDa)	PDI
<b>a</b>	HATU	18.2	1.18	39	1.04	7.8	1.07
<b>b</b>	HBTU	27.3	1.16	3.5	1.21	6.5	1.21
<b>c</b>	BOP reagent	19.4	1.26	55.4	1.69	11.3	1.32
<b>d</b>	PyBroP	10.8	2.42	43.3	1.80	3.4	1.43
<b>e</b>	DEPBT	42.1	2.12	3.1	1.18	5.5	1.05
<b>f</b>	HCTU	6.7	1.41	51	1.94	10.1	1.04
<b>g</b>	DMTMM	71.1	2.01	5.2	1.64	4.6	1.89
<b>h</b>	COMU	-	-	33	1.19	10.2	1.06
<b>i</b>	EDC.HCl	7.8	1.54	4.8	1.07	3.8	1.49
<b>j</b>	TATU	6.8	1.57	7.1	1.07	8.5	1.12
<b>k</b>	TBTU	21.9	1.12	6.8	1.16	9.5	1.05
<b>l</b>	TCTU	6.8	1.33	6.9	1.08	9.8	1.05
<b>m</b>	TOTU	15.5	2.40	6.9	1.07	10.8	1.03
<b>n</b>	TSTU	6.2	1.18	4.4	1.19	9.3	1.55
<b>o</b>	TFFH	5.8	1.34	8.0	1.05	8.7	1.07
<b>p</b>	TDBTU	4.7	1.49	3.1	1.32	5.9	1.40
<b>q</b>	PyOxime	6.9	1.12	2.4	1.45	10.7	1.05
<b>r</b>	Acid Chloride	6.7	1.31	28	1.62	13.2	1.15

**General protocol for synthesis of polymers with acid chloride:** In a 25 mL round bottomed flask, 2-methoxyisophthalic acid **B** (0.118 mmol) was taken and  $\text{SOCl}_2$  (4 mL) was added. This reaction was refluxed for 3 hours. After completion of reaction,  $\text{SOCl}_2$  was evaporated from the solvent to obtain compound **B'**, which was further treated with amine compound (**A1 - A3**, 0.118 mmol). Freshly distilled dry  $\text{CH}_2\text{Cl}_2$  (6 mL) and 100  $\mu\text{L}$  DIEA (N,N-Diisopropylethylamine) were added at 0 °C, and the solution was stirred overnight at room temperature. Solvent was then evaporated and the residue was washed with 10 mL MeOH to remove unreacted starting materials to give polymers **1r**, **2r**, and **3r** as off-white solid powder with ~ 70% yield. Molecular weights of all these polymers were determined by gel permeation chromatography (GPC) (**Table S1**).

**Synthesis of compound 11:** Potassium carbonate (1.36 g, 9.85 mmol, 1.5 equiv.) was added to a solution of methyl salicylate (1.0 g, 6.57 mmol) in dry Acetone (20 mL) and followed by benzyl bromide (1.04 mL, 8.54 mmol, 1.3 equiv.) was added to the reaction mixture and stirred at 55 °C for overnight. After completion of reaction, solvent was evaporated and obtained reaction mixture was washed with water (3 x 50 mL) and extracted with dichloromethane (3 x 50 mL). The organic layer was further washed with brine (100 mL) and dried over  $\text{Na}_2\text{SO}_4$ . The crude product **10**<sup>[4]</sup> was dissolved in  $\text{H}_2\text{O}/\text{CH}_3\text{OH}$  (1:4) and KOH (0.5 g, 8.66 mmol) was added at rt. The reaction mixture was refluxed for 5h. After completion of reaction, solvent was evaporated to remove methanol and pH was adjusted to 2-3 with 1M HCl. The reaction mixture was then extracted with EtOAc (3 x 30 mL) and dried over  $\text{Na}_2\text{SO}_4$  and concentrated under reduced pressure. The crude product was purified by column chromatography (2%  $\text{CH}_3\text{OH}$  in dichloromethane) to yield desired product **11** as white, solid with 90% yield.<sup>[5]</sup>  $^1\text{H}$  NMR: (400 MHz, DMSO)  $\delta$  12.69 (s, 1H), 7.65 (dd,  $J = 7.6, 1.8$  Hz, 1H), 7.53 – 7.44 (m, 3H), 7.42 – 7.36 (m, 2H), 7.35 – 7.28 (m, 1H), 7.19 (d,  $J = 8.4$  Hz, 1H), 7.00 (td,  $J = 7.5, 0.9$  Hz, 1H), 5.20 (s, 2H).

**Synthesis of 1a-Bn:** Polymer **1a** (0.018 mmol) and 4,6-bis(octyloxy)isophthalohydrazide (**A1**, R =  $\text{C}_8\text{H}_{17}$ , 0.06 mmol) were taken in a 20 mL round bottomed flask. HATU (0.07 mmol) was added in the same reaction vial. Freshly distilled  $\text{CH}_2\text{Cl}_2$  (2.5 mL) and 0.5 mL DMF



(dimethylformamide) was added to the reaction vial in presence of nitrogen atmosphere. This was followed by adding 100  $\mu$ L DIEA (N,N-Diisopropylethylamine) in the reaction mixture and stirred for 48 hours at room temperature. After completion of reaction, solvent was evaporated to remove  $\text{CH}_2\text{Cl}_2$  and DMF. The obtained residue was washed with 5 mL MeOH/ $\text{H}_2\text{O}$  (1:1), and off-white precipitate was observed. The obtained precipitate was filtered and washed with 5 mL  $\text{H}_2\text{O}$  and 5 mL MeOH respectively to obtain amine-modified polymer **12** as off-white solid powder with yields of 75%.

Synthesis of channel **1a-Bn**: Amine Polymer **12** (0.018 mmol) and compound **11** (0.06 mmol) were taken in a 20 mL round bottomed flask. HATU (0.07 mmol) was added in the same reaction vial. Freshly distilled  $\text{CH}_2\text{Cl}_2$  (2.5 mL) and 0.5 mL DMF (dimethylformamide) was added to the reaction vial in presence of nitrogen atmosphere. This was followed by adding 100  $\mu$ L DIEA (N,N-Diisopropylethylamine) in the reaction mixture and stirred for 48 hours at room temperature. After completion of reaction, solvent was evaporated to remove  $\text{CH}_2\text{Cl}_2$  and DMF. The obtained residue was washed with 5 mL MeOH/ $\text{H}_2\text{O}$  (1:1), and off-white precipitate was observed. The obtained precipitate was filtered and washed with 5 mL  $\text{H}_2\text{O}$  and 5 mL MeOH respectively to obtain compound as off-white solid powder with 60% yields. Molecular weight was determined by GPC (**Table S2**).

**Table S2.** GPC data for polymers **1a** and **1a-Bn**. PDI refers to the polydispersity index values.

Polymer	Coupling Reagents	Polymers with $\text{C}_8\text{H}_{17}$ side chain ( <b>1</b> )	
		$M^n$ (KDa)	PDI
<b>1a</b>	HATU	18.2	1.18
<b>1a-Bn</b>	HATU	16.8	1.19

### **MALDI-TOF Distribution Patterns of the Polymers**

**MALDI-TOF mass spectra** were acquired with Bruker Ultraflex extreme (Bruker Daltonik GmbH, Germany) equipped with Bruker smartbeam II 355 nm nitrogen laser with an accelerating voltage of 25 kV in the linear configuration. Mass spectra were measured by using the positive mode of mass spectroscopy. The matrix used in the experiment was trans-2-[3-(4-tert-Butylphenyl)-2-methyl-2-propenylidene]malononitrile (DCTB) from TCI and used directly without further purification. The solid matrix was dissolved at 10 mg/mL in chloroform. A volume of 10  $\mu$ L matrix solution was then spotted onto the MALDI sample plate and followed by addition of 5  $\mu$ L of the chloroform solution of polymers (1 mg/mL). Sample was air-dried at room temperature. The dried plate was then inserted into MALDI instrument. Selection of the laser used for ionization was performed directly through the software and required no adjustments to the individual lasers. Polymers **1a**, **1c**, **2a**, **2c** and **3c** were subjected for MALDI-TOF experiment and instrument can detect up to 10,000 Da. All polymers displayed characteristic mass pattern of repeating unit in the MALDI spectra. The interior of the helical channel is highly electron rich and therefore can bind with multiple metal ions such as Na<sup>+</sup> or K<sup>+</sup> ions to give rise to doubly or multiply charged species and all these peaks can be observed in the MALDI spectra.

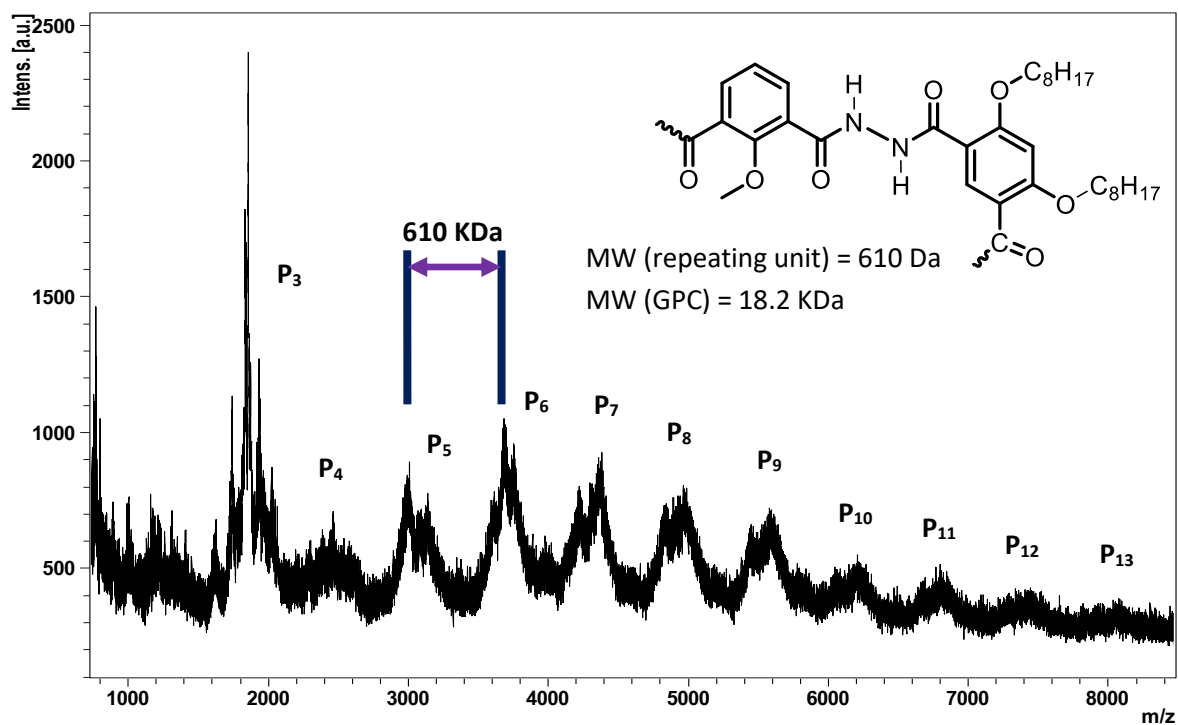


Figure S2. MALDI-TOF pattern of polymer 1a.

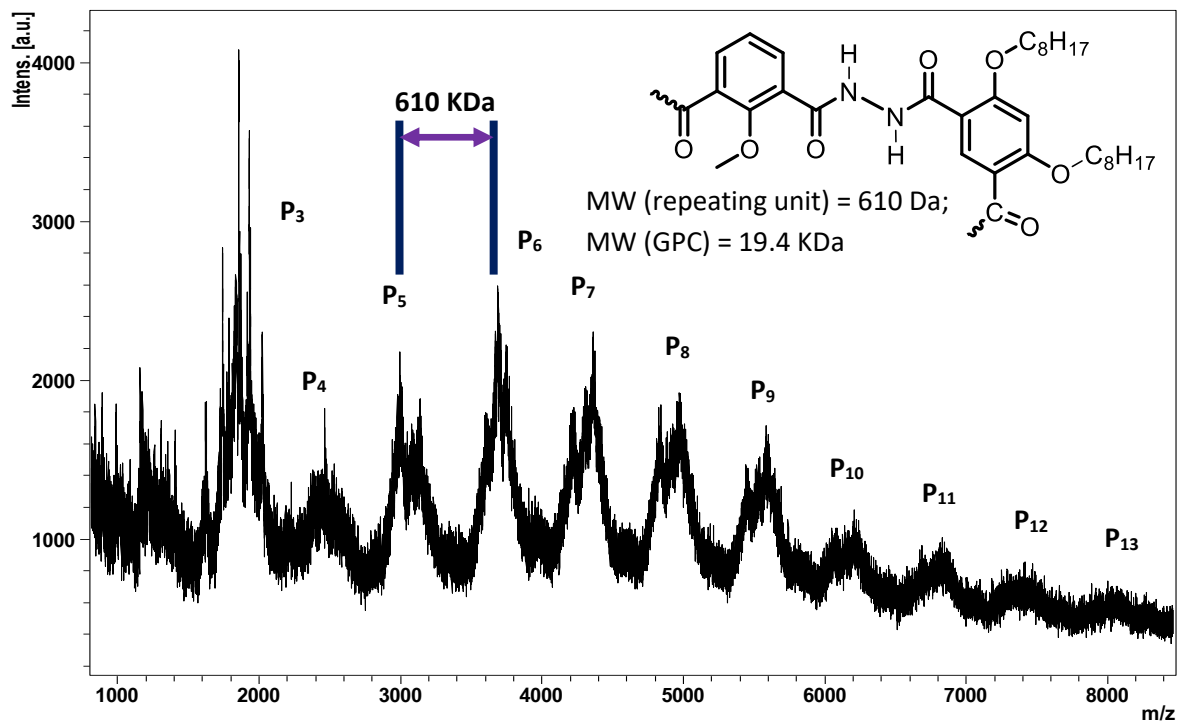


Figure S3. MALDI-TOF pattern of polymer 1c.

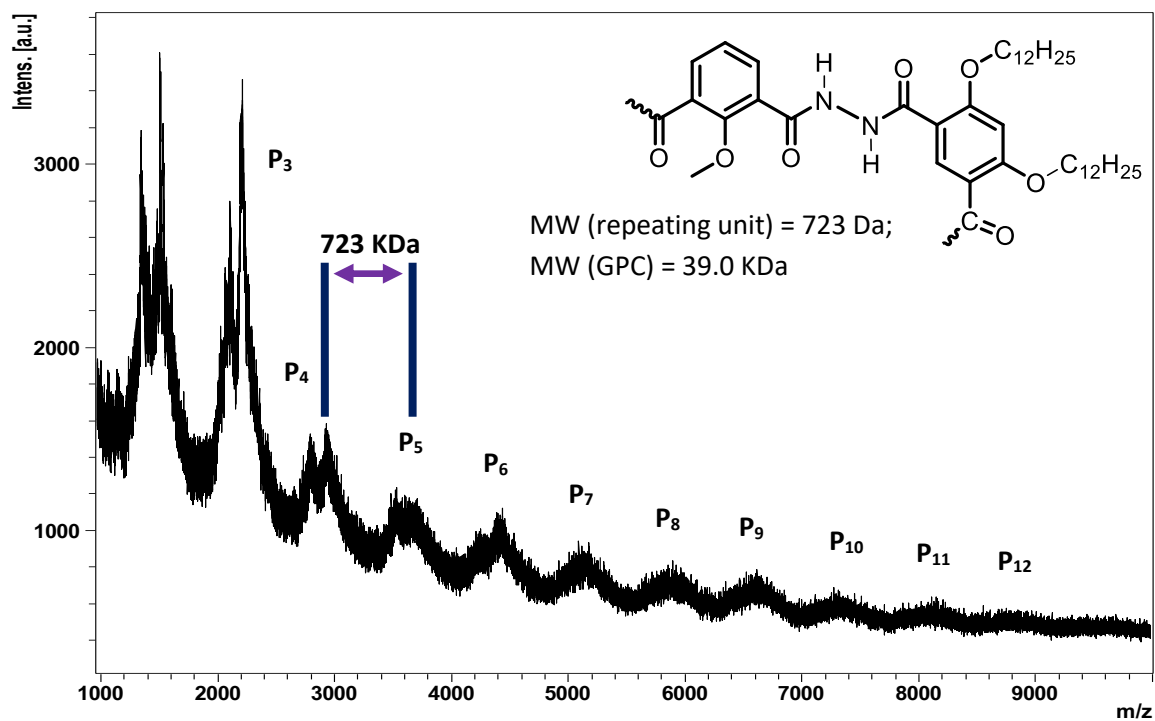


Figure S4. MALDI-TOF pattern of polymer **2a**.

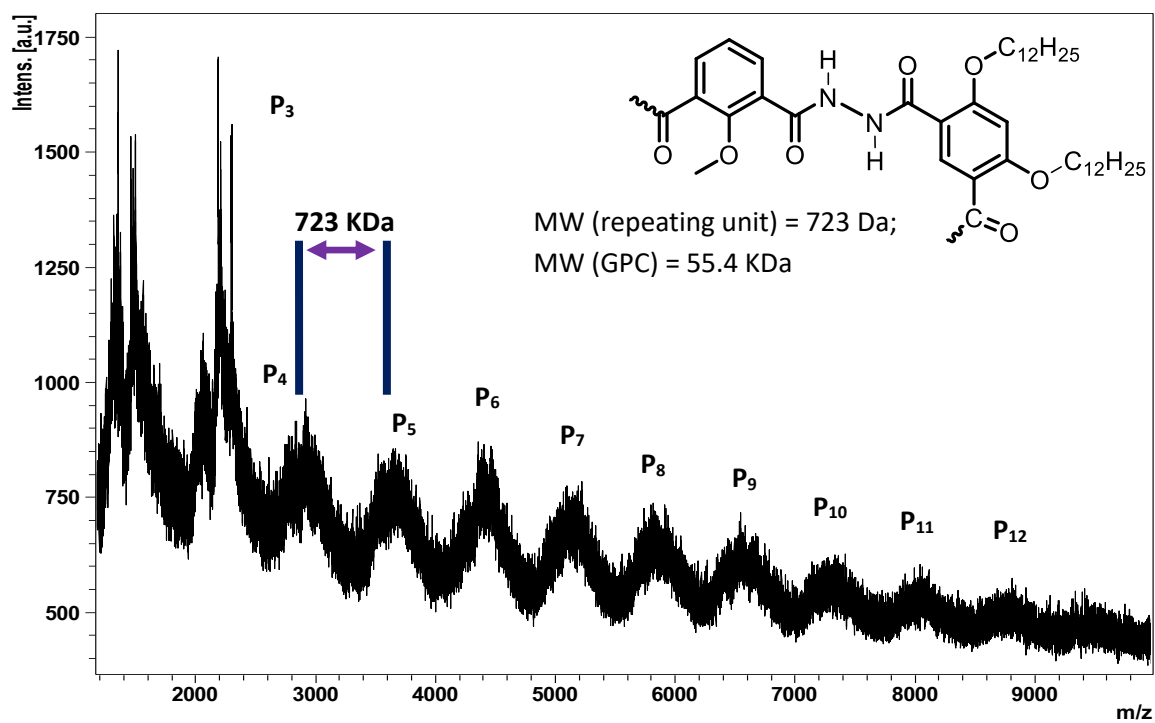


Figure S5. MALDI-TOF pattern of polymer **2c**.

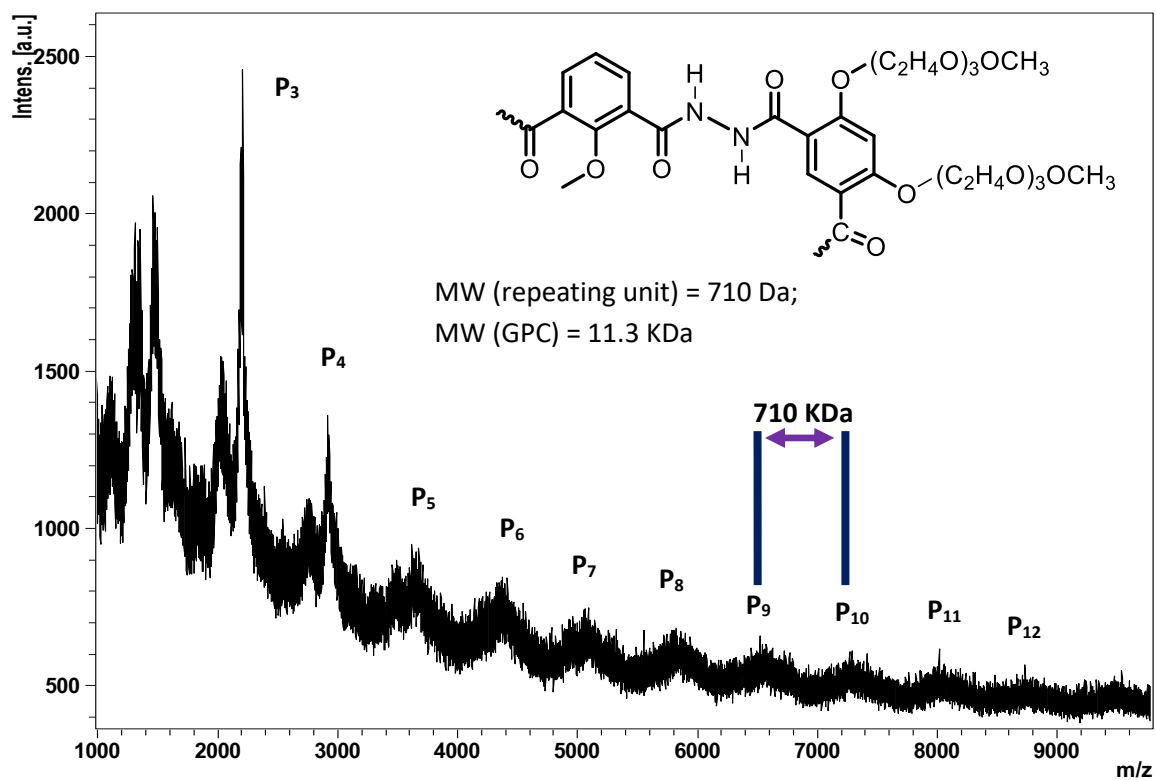


Figure S6. MALDI-TOF pattern of polymer 3c.

## Molecular Dynamics Simulations

To elucidate the microscopic mechanism of ion permeation through the **HP24** channels, we have performed atomistic molecular dynamics (MD) simulations. We constructed an all-atom model of **HP24** channel using psfgen plugin of VMD<sup>[6]</sup>, Supplementary Fig. S1a, Movie SM1 and SM2. To characterize its structural dynamics and ion selectivity in a lipid bilayer membrane, we embedded the channel into a patch of pre-equilibrated 1-palmitoyl-2-oleoyl-sn-glycero-3-phosphatidylcholine (POPC) membrane. We created several systems by solvating the membrane-embedded **HP24** channel with aqueous solutions of KCl, KI, NaCl and NaI at 0.6 M or 1M ionic concentration. Figure S8b shows a typical configuration of a fully assembled all-atom simulation system. All MD simulations were performed using the MD program NAMD2<sup>[7]</sup>, periodic boundary conditions and particle mesh Ewald (PME) method to calculate the long range electrostatics<sup>[8]</sup>. The Nose-Hoover Langevin piston<sup>[9]</sup> and Langevin thermostat<sup>[10]</sup> were used to maintain the constant pressure and temperature in the system. CHARMM36 force field parameters<sup>[11]</sup> described the bonded and non-bonded interactions of among, lipid bilayer membranes, water and ions. We used the standard CHARMM ion parameters by Roux and coworkers<sup>[12]</sup> for the K<sup>+</sup>, Na<sup>+</sup> and Cl<sup>-</sup> ions. Whereas the non-bonded parameters for the I<sup>-</sup> ion were obtained from the recent work of Li et al<sup>[13]</sup>. A 8-10-12 Å cutoff scheme was used to calculate van der Waals and short range electrostatics forces. All simulations were performed using a 2 fs time step to integrate the equation of motion. SETTLE algorithm<sup>[14]</sup> was applied to keep water molecules rigid whereas RATTLE algorithm<sup>[15]</sup> constrained all other covalent bonds involving hydrogen atoms. The coordinates of the system were saved at an interval of 19.2 ps. The analysis and post processing the simulation trajectories were performed using VMD<sup>[6]</sup> and CPPTRAJ<sup>[16]</sup>.

The initial PDB structure of the **HP24** channel was created using Gaussview to contain 24 units of the building blocks **A** and **B** shown in Figure 1. The topology and force field parameters for one unit of the **HP24** channel was obtained using the CHARMM general force fields (CGenFF) webserver.<sup>[17]</sup> The protein structure file (psf) of the **HP24** molecule was constructed using the psfgen tool of VMD. The channel was then embedded into a 9 × 9 nm<sup>2</sup> patch of pre-equilibrated POPC lipid bilayer membrane. The lipid patch was generated using the CHARMM-GUI

membrane builder<sup>[18]</sup> and pre-equilibrated for approximately 400 ns. Lipid molecules that overlapped with the channel were removed. The system was then solvated with water<sup>[19]</sup> using the Solvate plugin of VMD. Sodium and chloride ions were added to 0.6 M concentration using the Autoionize plugin of VMD. The final assembled system measured 9 x 9 x 9 nm<sup>3</sup> and contained approximately 80,000 atoms.

Following the assembly, the system underwent 1200 steps of energy minimization using the conjugate gradient method to remove steric clashes. After energy minimization, the system was subjected to a 100 ns equilibration at a constant number of atoms (N), pressure (P = 1 bar) and temperature (T = 300 K), the NPT ensemble, with harmonic restraints applied to all non-hydrogen atoms of the **HP24** channel that surrounded the transmembrane pore. The restraints were applied relative to the initial coordinates of the atoms, the spring constants were 1 kcal mol<sup>-1</sup> Å<sup>-2</sup>. After 100 ns, the harmonic restraints were removed, and the system was equilibrated free of any restraints for ~550 ns.

Four other systems, containing 1M KCl, KI, NaCl or NaI solution, were built using the configuration of the lipids and the **HP24** channel obtained at the end of the 550 ns equilibration. After energy minimization and a 100 ns NPT equilibration, the systems were simulated in the constant volume (NVT) ensemble for about 100 ns under external electric field applied to produce a +1 or -1 V transmembrane bias. In these simulations, the center of mass of the 24 oxygen atoms of the channel was restrained using zero velocity SMD module in NAMD, maintaining the center of the channel at Z = 0. The ionic current across the membrane was computed using the following formula as described previously

$$I(t) = \frac{1}{\Delta t L_z} \sum_{i=1}^N q_i [(z_i(t + \Delta t) - z_i(t))],$$

where  $\Delta t$  is the time interval between two consecutive frames of the MD trajectory,  $L_z$  is length of the system along Z axis,  $N$  is the number of ions present in the system,  $q_i$  is the charge of the respective ion,  $z_i$  is z coordinate of the respective ion at that time instant if

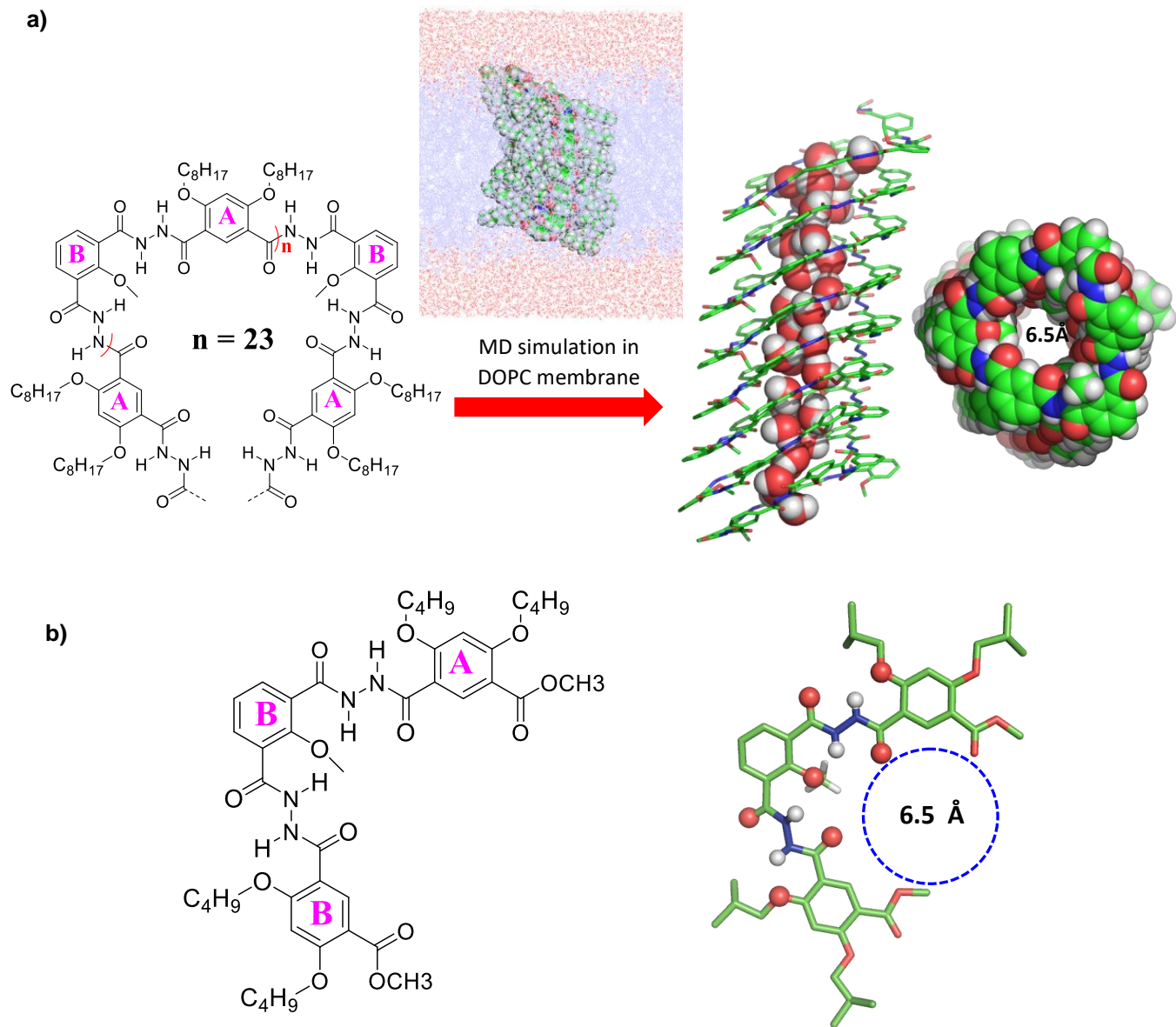
$$\text{abs}(z_i) \leq L_z/2; z_i = z_i$$

else if  $z_i > L_z/2$ ;  $z_i = L_z/2$

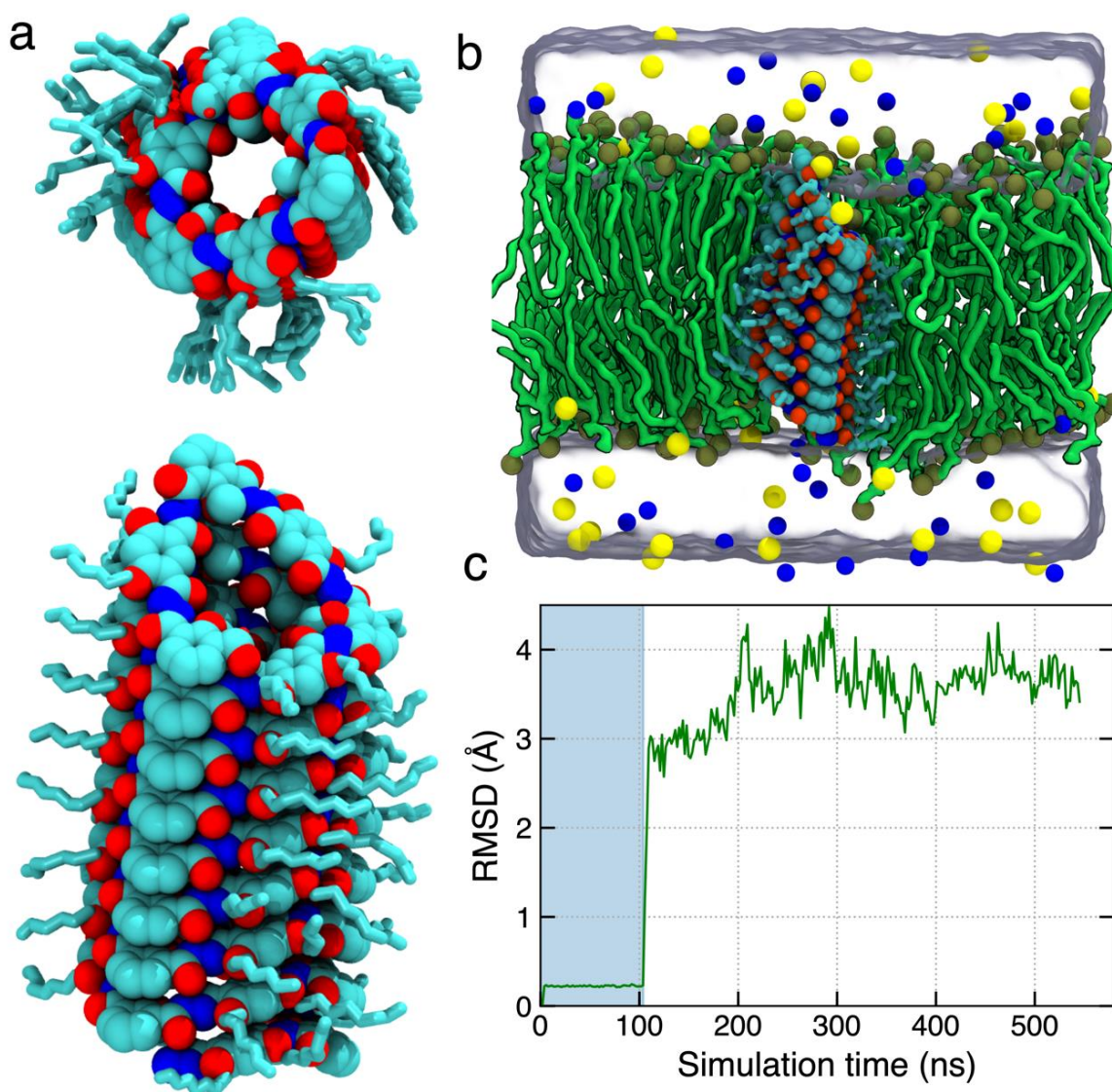
else if  $z_i < -L_z/2$ ;  $z_i = -L_z/2$

To calculate the free energy profile of an ion moving through the central pore of the **HP24** channel, we employed the umbrella sampling technique<sup>[20]</sup> implemented within the *colvar* module of NAMD. In order to ensure the uniform sampling of the reaction space, we performed replica exchange MD simulations<sup>[21]</sup>. Starting from the 100 ns equilibrated systems with 1M molar concentration of different electrolyte solution, we created 69 copies of each of the system by quickly pulling a single ion along the axis of the channel in a 1.4 ns steered molecular dynamics run. We choose the snapshot of the system where the ion falls in the respective sampling window of the 1Å bin along the Z axis and harmonically restrained it to the center of the window. The spring constant of the harmonic potential was 2.5 kcal/mol/ Å<sup>2</sup>. Each replica was simulated for 100 ns and the replicas were allowed to exchange with a probability given by the Metropolis algorithm. Finally, we used WHAM<sup>[22]</sup> program to subtract the contribution from the confining harmonic potential and extracted the PMF of ion along the axis of the channel.

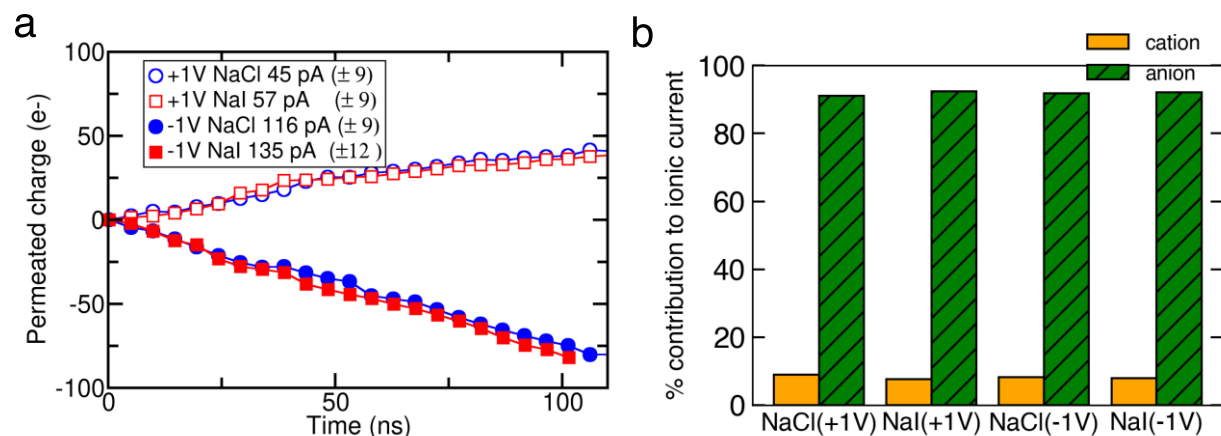




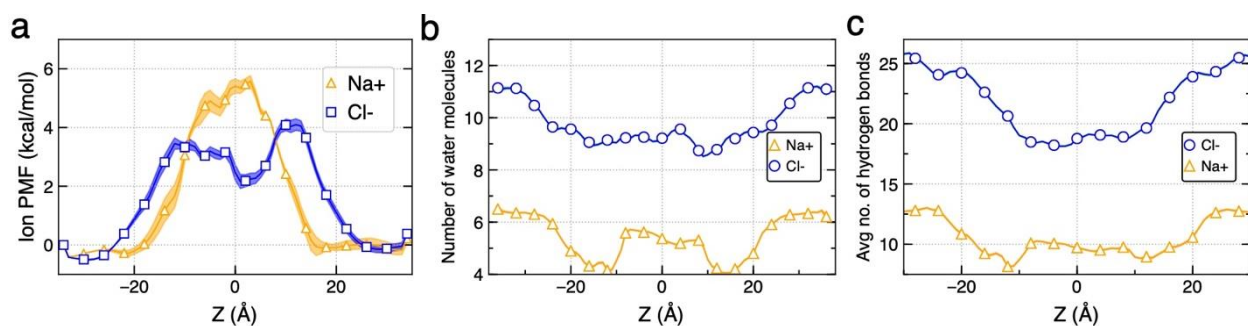
**Figure S7.** a) Molecular dynamics-simulated structure of polyhydrazides containing 24 **A**s and 24 **B**s in POPC membrane. b) The crystal structure of a trimer molecule, containing 2 **A**s and 1 **B**, which shows that about 5.6 repeating units (e.g., 2.8 **A**s and 2.8 **B**s) are required to form one helical turn of 3.4 Å in height and about 6.5 Å in diameter.



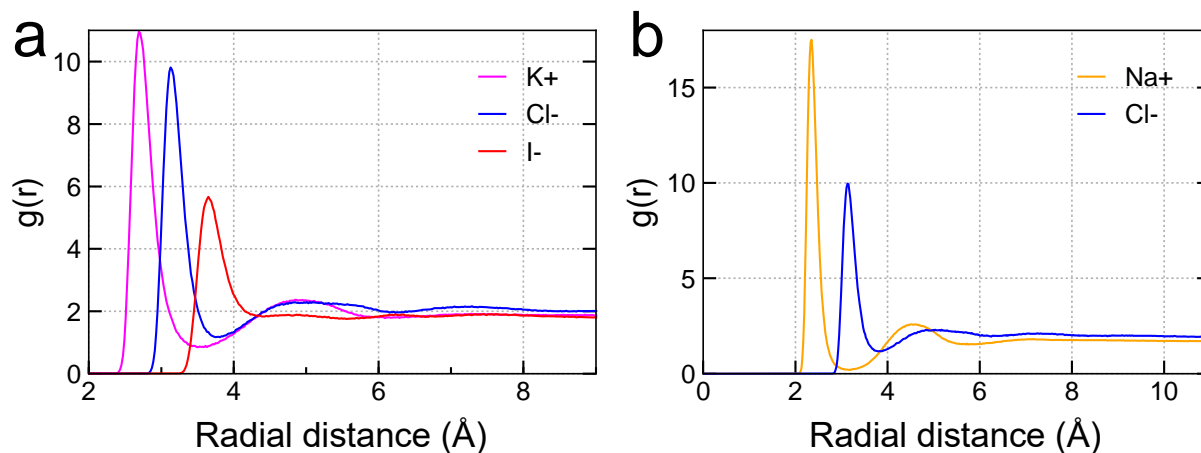
**Figure S8:** All-atom MD simulation of channel **HP24**, which contains 24 **As** and 24 **Bs**, in a lipid bilayer membrane with explicit water and ions. **(a)** Top and side views of all atom model of **HP24**; carbon (cyan), oxygen (red) and nitrogen (blue) atoms of the core of the channel are shown using sphere whereas the side chains of channel are shown in cyan color licorice representation. **(b)** The cut-away view of the channel embedded in the lipid bilayer membrane and solvated in a box of water and ions. The phosphorus atom of the lipid headgroup is shown using blue spheres whereas the tail of the lipid molecules is shown using thick green lines. Na<sup>+</sup> (yellow) and Cl<sup>-</sup> (cyan) ions are shown using spheres and water is represented as transparent white background. **(c)** The RMSD of the channel as a function of simulation time, the shaded blue region shows the period where the non-hydrogen atoms of the channel (represented as spheres in the figS1a) were restrained using harmonic potential.



**Figure S9:** Simulated ionic currents through the **HP24** channel for 1M NaCl and 1M NaI systems. **(a)** The total permeated charge (integrated ionic current) as a function of the simulation time in the units of the elementary charge,  $e$ , the charge of a proton. The slope of each line gives the average current. The inset specified the average current values with the error bars for each system. **(b)** Percentage of total current carried by the cation and anion species in the simulations of the NaCl and NaI systems



**Figure S10:** (a) The free energy profile of the  $\text{Na}^+$  and  $\text{Cl}^-$  ions translocating through the **HP24** channel. (b) Average number of the water molecules in the first solvation shell of ion as a function of the Z-coordinate of the ion. (c) The average number of hydrogen bonds formed by the water molecules in the first solvation shell of the  $\text{Na}^+$  and  $\text{Cl}^-$  ions as a function of their Z-coordinate.



**Figure S11:** The radial distribution function (RDF) of the various ions obtained from the equilibrium MD simulation trajectories. Based on the RDF, we define the radius of the first solvation shell for Na<sup>+</sup>, K<sup>+</sup>, Cl<sup>-</sup> and I<sup>-</sup> as 3.2 Å, 3.5 Å, 4.0 Å 4.35 Å respectively.

**Table S3:** Transmembrane ionic current measurements for **HP24** channel obtained using all-atom MD simulation at several electrolyte conditions and +/-1V. The average current value and its standard error was calculated by splitting the current versus time trace into 15 ns blocks and considering average values within each block as independent measurements.

Electrolyte (1M)	Applied Bias (V)	I <sub>total</sub> (pA)	I <sub>cation</sub> (%)	I <sub>anion</sub> (%)
NaCl	1	45.2 (±9.0)	8.9	91.1
	-1	-115.8 (±9.0)	7.6	92.4
NaI	1	57.1 (±9.0)	8.2	91.8
	-1	-135.0 (±12.1)	7.9	92.1
KCl	1	22.5 (±6.5)	21.9	78.1
	-1	-119.1 (±10.0)	6.8	93.2
KI	1	77.3 (±9.0)	16.7	83.3
	-1	-137.2 (±16.2)	21.0	79

## **Ion Transport Study Using the HPTS Assay**

**Buffer and stock solution preparation:** HEPES buffer (10 mM) was prepared by dissolving solid HEPES in MilliQ water, then NaCl (100 mM) was added and followed by pH was adjusted to 7.0 by adding NaOH solution dropwisely. Stock solutions of all compounds were prepared in HPLC grade DMF.

**Preparation of HPTS-containing LUVs for the HPTS assay:** Egg yolk L- $\alpha$ -phosphatidylcholine (EYPC, 1 ml, 25 mg/mL in CHCl<sub>3</sub>, Avanti Polar Lipids, USA) was taken in a 2 mL microtube and CHCl<sub>3</sub> was evaporated by purging N<sub>2</sub> slowly. After drying the resulting film under high vacuum overnight at room temperature, the film was hydrated with 4-(2-hydroxyethyl)-1-piperazine-ethane sulfonic acid (HEPES) buffer solution (1 mL, 10 mM HEPES, 100 mM NaCl, pH = 7.0) containing a pH sensitive dye 8-hydroxypyrene-1,3,6-trisulfonic acid (HPTS, 0.5 mM) at room temperature for 1 hour (with occasional vortexing after every 15 minutes) to give a milky suspension. The mixture was then subjected to 10 freeze-thaw cycles (freezing in liquid N<sub>2</sub> for 1 minute and heating at 55 °C in water bath for 2.5 minutes). The vesicle suspension was extruded through polycarbonate membrane (0.1  $\mu$ m) to produce a homogeneous suspension of large unilamellar vesicles (LUVs) of about 100 nm in diameter with HPTS encapsulated inside. The extraventricular HPTS dye was removed by using size exclusion chromatography (stationary phase: Sephadex G-50, GE Healthcare, USA, mobile phase: HEPES buffer with 100 mM NaCl) and diluted with the mobile phase to yield 3.2 mL of 10 mM lipid stock solution.

**The HPTS assay for ion transport activity study:** 30  $\mu$ L of HPTS-containing LUVs was added to 1970  $\mu$ L of HEPES buffer (10 mM HEPES, 100 mM NaCl, pH = 8.0) in a clean fluorescence cuvette to generate pH gradient across lipid bilayer. This cuvette was placed on the fluorescence instrument (at  $t = 0$  s) equipped with magnetic stirrer. Fluorescence emission intensity of HPTS dye,  $F_t$  was monitored at  $\lambda_{em} = 510$  nm ( $\lambda_{ex} = 450$  nm) with time. All compounds (dissolved in DMF) were added at  $t = 70$  s and recorded simultaneously for 300 seconds using fluorescence spectrophotometer (Hitachi, Model F-7100, Japan). Finally at  $t = 370$  s, 20  $\mu$ L of 20% Triton X-

100 was added to destroy all vesicles which resulted in destruction of pH gradient to achieve the maximum change in fluorescence emission intensity of HPTS dye.

The time axis was normalized according to Equation S1:

$$t = t - 70 \quad (\text{Equation S1})$$

Fluorescence intensities ( $F_t$ ) were normalized to fractional emission intensity  $I_F$  using Equation S2, whereas  $F_0$  = Fluorescence intensity just before the compound addition (at  $t = 0$  s),  $F_\infty$  = Fluorescence intensity at saturation after complete leakage (at  $t = 300$  s), and  $F_t$  = Fluorescence intensity at time  $t$ .

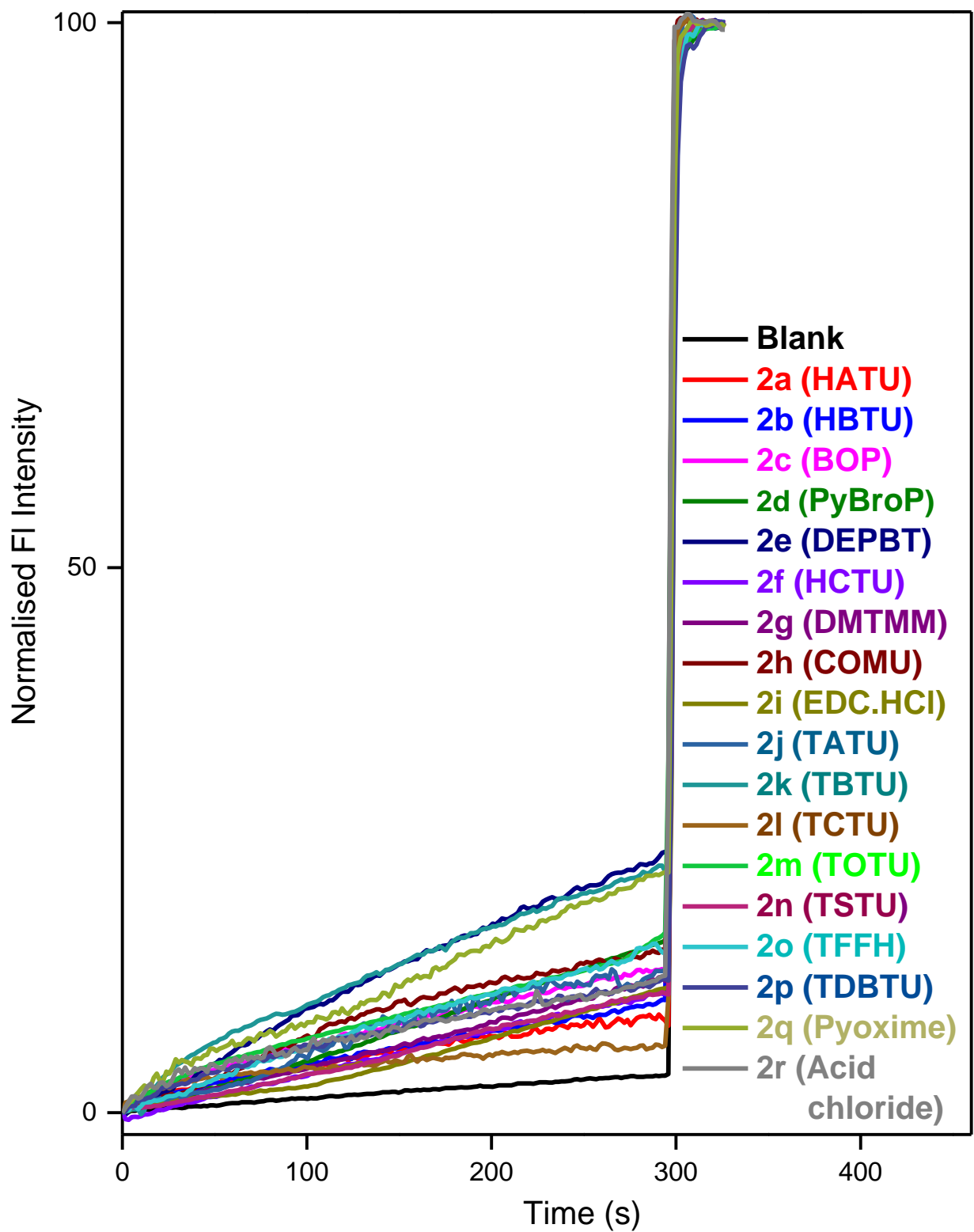
$$F_\infty = I_F = [(F_t - F_0)/(F_\infty - F_0)] \times 100 \quad (\text{Equation S2})$$

The concentration profile data were analysed by Hill Equation (Equation S2) to get the Effective concentration at half maximal activity ( $EC_{50}$ ) and Hill Coefficient ( $n$ ),

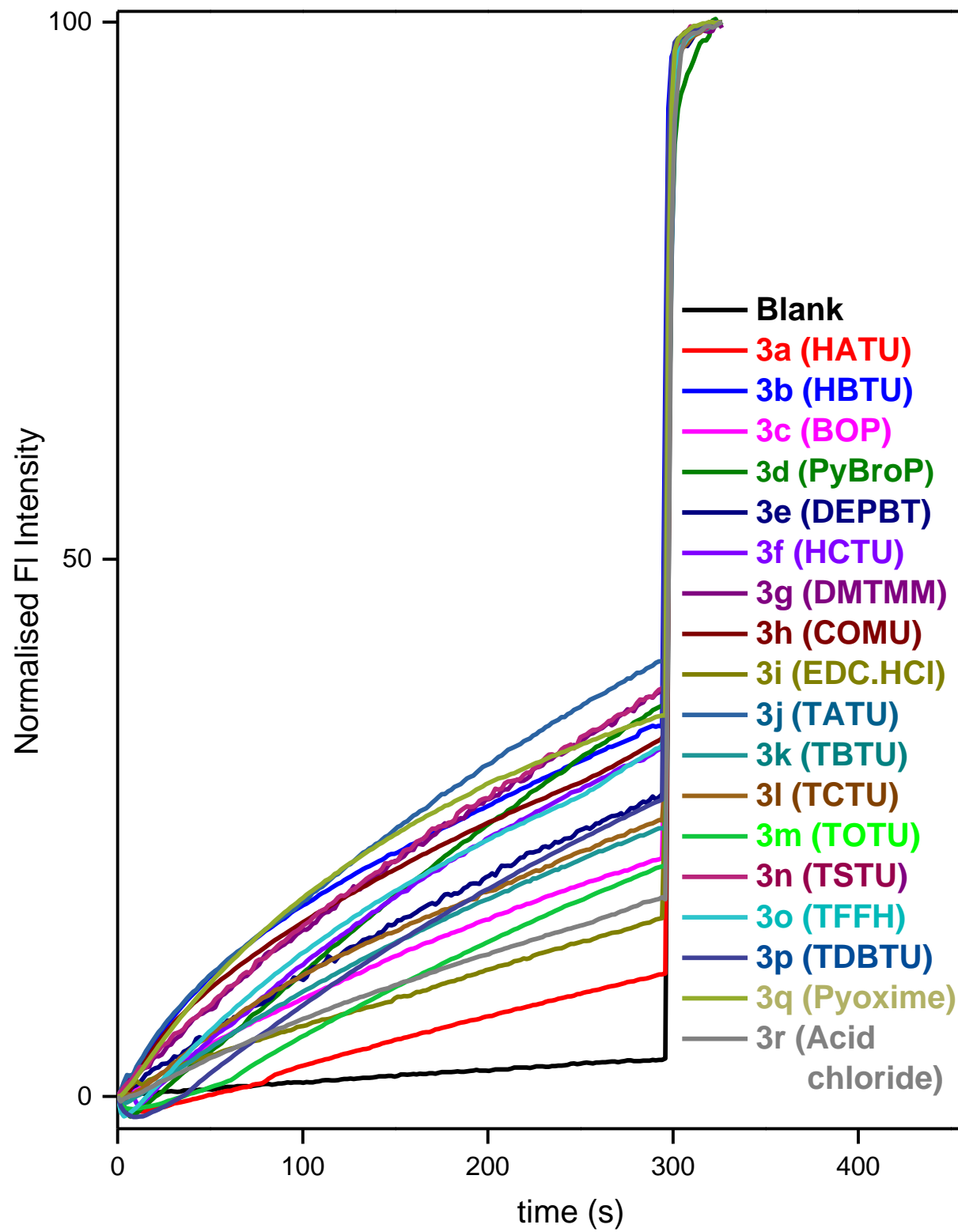
$$Y = I / (1 + (EC_{50}/[c])^n) \quad (\text{Equation S3})$$

Whereas  $Y$  = Fluorescence intensity and  $c$  = Concentration of channel forming molecule.

Take polymers **1** as the example. Ion transport activity of all polymer channels (**1a** - **1r** with R = C<sub>8</sub>H<sub>17</sub>) at 1  $\mu$ M across lipid bilayer were compared at  $t = 370$  s. Intermediate polymers, having molecular weight 15 - 30 KDa, show higher transport ability than those with very short (< 10 KDa) or longer backbones (> 30 KDa), indicating that an optimum length of polymer is crucial to span through membrane and form active structure (**Figure 2b**). The most active channel **1a** was further examined at varied concentrations. The saturation of ion transport activity of **1a** was found at 1  $\mu$ M, and these transport curves were plotted in a dose dependent manner. Hill equation was used to obtain  $EC_{50}$ , *i.e.* effective concentration to reach 50% maximum activity, and Hill coefficient (**Figure S14**).

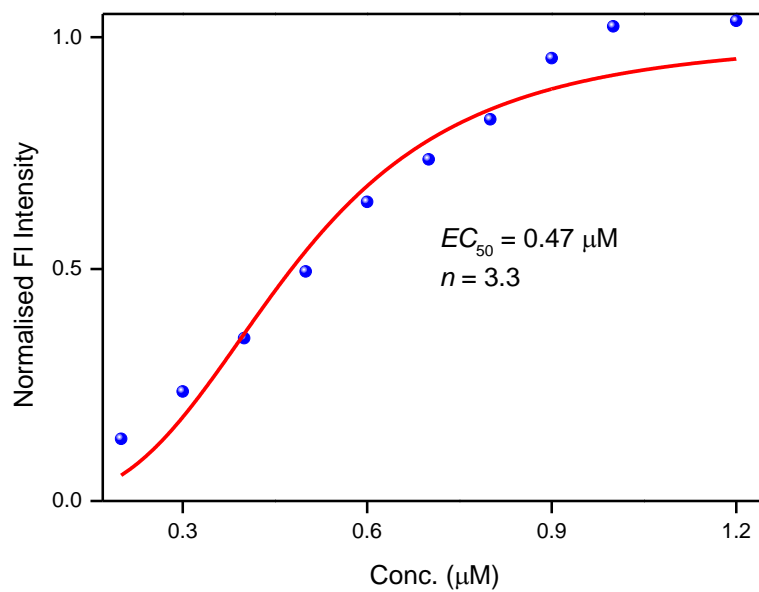


**Figure S12.** Ion transport activities of channels (2a - 2r, 1  $\mu$ M) using the HPTS assay.



**Figure S13.** Ion transport activities of channels (3a - 3r, 1  $\mu$ M) using the HPTS assay.





**Figure S14.** Determination of  $EC_{50}$  value via Hill analysis for channel **1a** with a MW of 18.2 KDa using the HPTS assay.

## Proton Transport Using the HPTS Assay

Egg yolk L- $\alpha$ -phosphatidylcholine (EYPC, 1 ml, 25 mg/mL in CHCl<sub>3</sub>, Avanti Polar Lipids, USA) was taken in a 2 mL microtube and CHCl<sub>3</sub> was evaporated by purging N<sub>2</sub> slowly. After drying the resulting film under high vacuum overnight at room temperature, the film was hydrated with 4-(2-hydroxyethyl)-1-piperazine-ethane sulfonic acid (HEPES) buffer solution (1 mL, 10 mM HEPES, 100 mM Na<sub>2</sub>SO<sub>4</sub>, pH = 7.0) containing a pH sensitive dye 8-hydroxypyrene-1,3,6-trisulfonic acid (HPTS, 0.5 mM) at room temperature for 1 hour (with occasional vortexing after every 15 minutes) to give a milky suspension. The mixture was then subjected to 10 freeze-thaw cycles (freezing in liquid N<sub>2</sub> for 1 minute and heating at 55 °C in water bath for 2.5 minutes). The vesicle suspension was extruded through polycarbonate membrane (0.1  $\mu$ m) to produce a homogeneous suspension of LUVs of about 100 nm in diameter with HPTS encapsulated inside. The extravesicular HPTS dye was removed by using size exclusion chromatography (stationary phase: Sephadex G-50, GE Healthcare, USA, mobile phase: HEPES buffer with 100 mM Na<sub>2</sub>SO<sub>4</sub>, pH = 7.0) and diluted with the mobile phase to yield 3.2 mL of 10 mM lipid stock solution.

The HPTS-containing LUV suspension (30  $\mu$ L, 10 mM in 10 mM HEPES buffer containing 100 mM Na<sub>2</sub>SO<sub>4</sub> at pH = 7.0) was added to a HEPES buffer solution (1.7 mL, 10 mM HEPES, 100 mM H<sub>2</sub>SO<sub>4</sub> at pH = 8.0) to create a pH gradient for ion transport study. A solution of compound in DMF (1  $\mu$ M for polymer **1a** and FCCP, and 2  $\mu$ M for GA) was then injected into the suspension under gentle stirring. Upon the addition of channels, the emission of HPTS was immediately monitored at 510 nm with excitations at 460 nm for 300 seconds using fluorescence spectrophotometer (Hitachi, Model F-7100, Japan) after which time an aqueous solution of Triton X-100 (20  $\mu$ L, 20% v/v) was immediately added to achieve the maximum change in dye fluorescence emission. The final transport trace was normalized based on equation S2.

### **Cation Selectivity Using the HPTS Assay**

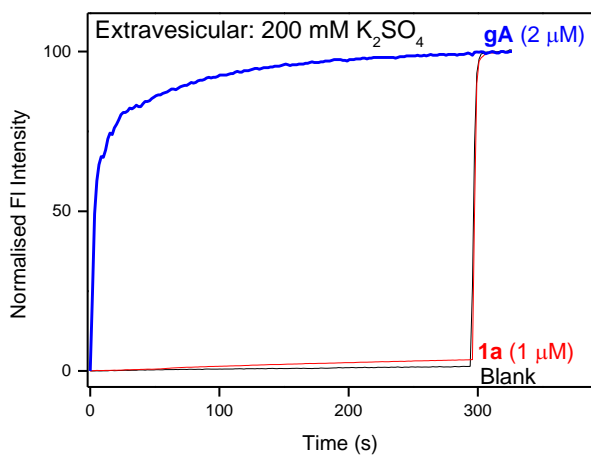
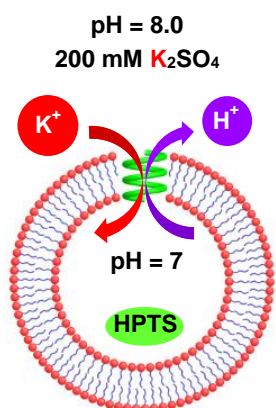
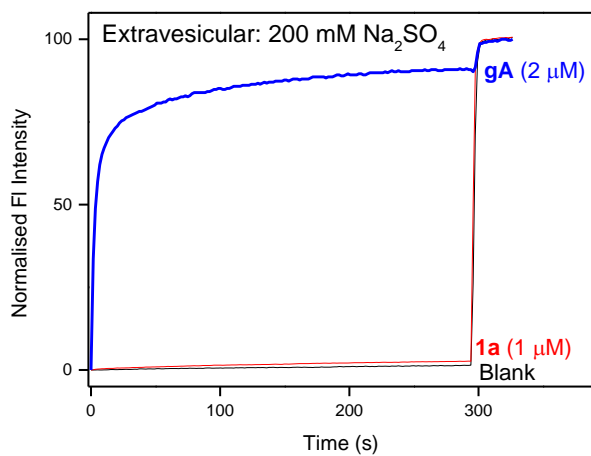
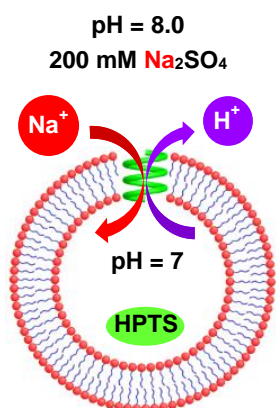
The HPTS-containing LUV suspension (30  $\mu\text{L}$ , 10 mM in 10 mM HEPES buffer containing 100 mM  $\text{Na}_2\text{SO}_4$  at pH = 7.0) was added to a HEPES buffer solution (1.7 mL, 10 mM HEPES, 100 mM  $\text{M}_2\text{SO}_4$  at pH = 8.0, where  $\text{M}^+ = \text{Li}^+, \text{Na}^+, \text{K}^+$  and  $\text{Cs}^+$ ) to create a pH gradient for ion transport study. A solution of polymer **1a** (0.5  $\mu\text{M}$ ) in DMF was then injected into the suspension under gentle stirring. Upon the addition of channels, the emission of HPTS was immediately monitored at 510 nm with excitations at 460 nm for 300 seconds using fluorescence spectrophotometer (Hitachi, Model F-7100, Japan) after which time an aqueous solution of Triton X-100 (20  $\mu\text{L}$ , 20% v/v) was immediately added to achieve the maximum change in dye fluorescence emission. The final transport trace was normalized based on equation S2.

## Cation Transport Using the HPTS Assay under High Salt Gradient Environment

Egg yolk L- $\alpha$ -phosphatidylcholine (EYPC, 1 mL, 25 mg/mL in  $\text{CHCl}_3$ , Avanti Polar Lipids, USA) was taken in a 2 mL microtube and  $\text{CHCl}_3$  was evaporated by purging  $\text{N}_2$  slowly. After drying the resulting film under high vacuum overnight at room temperature, the film was hydrated with 4-(2-hydroxyethyl)-1-piperazine-ethane sulfonic acid (HEPES) buffer solution (1 mL, 10 mM HEPES, pH = 7.0) containing a pH sensitive dye 8-hydroxy-ypyrene-1,3,6-trisulfonic acid (HPTS, 0.5 mM) at room temperature for 1 hour (with occasional vortexing after every 15 minutes) to give a milky suspension. The mixture was then subjected to 10 freeze-thaw cycles (freezing in liquid  $\text{N}_2$  for 1 minute and heating at 55 °C in water bath for 2.5 minutes). The vesicle suspension was extruded through polycarbonate membrane (0.1  $\mu\text{m}$ ) to produce a homogeneous suspension of LUVs of about 100 nm in diameter with HPTS encapsulated inside. The extravesicular HPTS dye was removed by using size exclusion chromatography (stationary phase: Sephadex G-50, GE Healthcare, USA, mobile phase: HEPES buffer, pH = 7.0) and diluted with the mobile phase to yield 3.2 mL of 10 mM lipid stock solution.

The HPTS-containing LUV suspension (30  $\mu\text{L}$ , 10 mM in 10 mM HEPES buffer without any salt at pH = 7.0) was added to a HEPES buffer solution (1.7 mL, 10 mM HEPES, containing 200 mM  $\text{Na}_2\text{SO}_4$  at pH = 8.0) to create salt and pH gradient for ion transport study. A solution of compound (polymer **1a** (1  $\mu\text{M}$ ), and GA (2  $\mu\text{M}$ ) in DMF) was then injected into the suspension under gentle stirring. Upon the addition of channels, the emission of HPTS was immediately monitored at 510 nm with excitations at 460 nm for 300 seconds using fluorescence spectrophotometer (Hitachi, Model F-7100, Japan) after which time an aqueous solution of Triton X-100 (20  $\mu\text{L}$ , 20% v/v) was immediately added to achieve the maximum change in dye fluorescence emission. The final transport trace was normalized based on equation S2.

Based on the data presented in Figure S15, it is clear that polymer **1a** (1  $\mu\text{M}$ ) is unable to transport any cation ( $\text{Na}^+$  or  $\text{K}^+$ ) under high salt gradient environment (200 mM  $\text{Na}_2\text{SO}_4/\text{K}_2\text{SO}_4$ ) whereas gramicidin A (gA, (2  $\mu\text{M}$ ) displays significant  $\text{Na}^+$  and  $\text{K}^+$  transport ability under same assay condition.



**Figure S15.** Schematic representation of LUV-based cation selectivity assay under high salt gradient (200 mM Na<sub>2</sub>SO<sub>4</sub> or K<sub>2</sub>SO<sub>4</sub>) for channel **1a** (1 μM) with comparison to those of gramicidin A (gA, 2 μM) under the same assay condition.

## Chloride Transport Using the SPQ Assay

Egg yolk L- $\alpha$ -phosphatidylcholine (EYPC, 1 ml, 25 mg/mL in  $\text{CHCl}_3$ , Avanti Polar Lipids, USA) were taken in a 2 mL microtube and  $\text{CHCl}_3$  was removed under reduced pressure at room temperature. After drying the resulting film under high vacuum overnight at room temperature, the film was hydrated  $\text{NaNO}_3$  solution (1 mL, 225 mM) containing a  $\text{Cl}^-$ -sensitive dye 6-methoxy-N-(3-sulfopropyl)quinolinium (SPQ) (1 mM) dye at room temperature for 1 hour (with occasional vortexing after every 15 minutes) to give a milky suspension. The mixture was then subjected to 10 freeze-thaw cycles: freezing in liquid  $\text{N}_2$  for 60 seconds and heating at 55 °C for 2.5 minutes. The vesicle suspension was extruded through polycarbonate membrane (0.1  $\mu\text{m}$ ) to produce a homogeneous suspension of LUVs with SPQ encapsulated inside. The extravesicular SPQ dye was removed by using size exclusion chromatography (stationary phase: Sephadex G-50, GE Healthcare, USA, mobile phase: 225 mM  $\text{NaNO}_3$  solution) and diluted with the mobile phase to yield 3.2 mL of 10 mM lipid stock solution. pH of the solution containing 225 mM  $\text{NaCl}$  was measured and found to be 6.2.

The SPQ-containing LUV suspension (30  $\mu\text{L}$ , 10 mM in 225 mM  $\text{NaNO}_3$ ) was added to a  $\text{NaCl}$  solution (1.7 mL, 225 mM) to create an extravesicular chloride gradient. A solution of channel molecule in DMF at different concentrations was then injected into the suspension under gentle stirring. Upon the addition of channels, the emission of SPQ was immediately monitored at 430 nm with excitations at 360 nm for 300 seconds using fluorescence spectrophotometer (Hitachi, Model F-7100, Japan) after which time an aqueous solution of Triton X-100 (20  $\mu\text{L}$ , 20% v/v) was immediately added to completely destruct the chloride gradient. The final transport trace was obtained by normalizing the fluorescence intensity using the following Equation S4,

$$I_F = [(F_t - F_0)/(F_\infty - F_0)] \quad (\text{Equation S4})$$

Where  $F_0$  = Fluorescence intensity just before the compound addition (at  $t = 0$  s),  $F_t$  = Fluorescence intensity at time  $t$ , and  $F_\infty$  = Fluorescence intensity at saturation after complete leakage (at  $t = 300$  s).

### **FCCP-Coupled HPTS Assay**

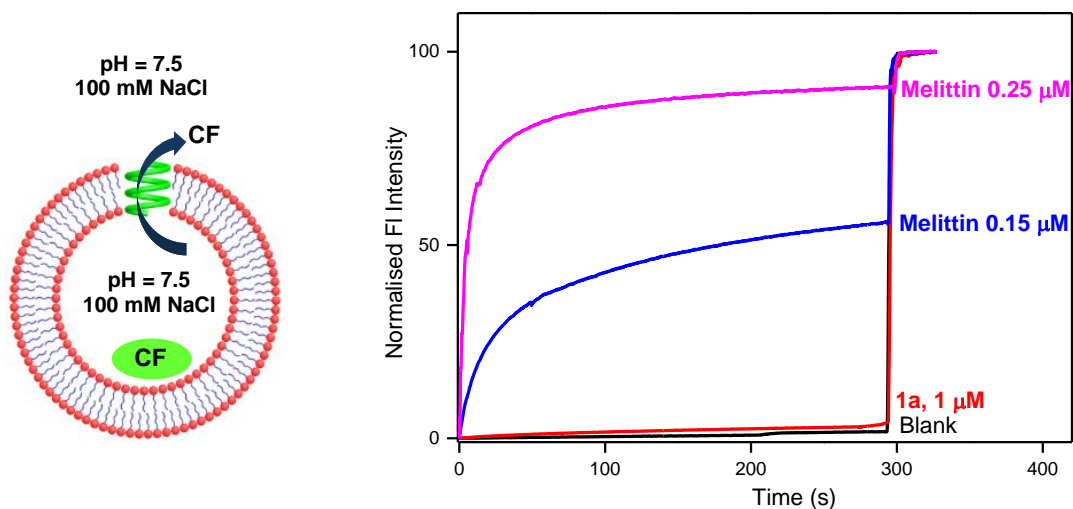
The HPTS-containing LUV suspension (30  $\mu\text{L}$ , 10 mM in 10 mM HEPES buffer containing 100 mM NaCl at pH = 7.0) was added to a HEPES buffer solution (1.7 mL, 10 mM HEPES, 67 mM  $\text{Na}_2\text{SO}_4$ ) to create a pH gradient for ion transport study. A solution of carbonyl cyanide-4-(trifluoromethoxy)phenylhydrazone (FCCP) (1  $\mu\text{M}$ , whenever necessary) and channel molecule **1a** (0.5  $\mu\text{M}$ ) in DMF was then injected into the suspension under gentle stirring at  $t = 40$  s and 70 s, respectively. Upon the addition of channels, the emission of HPTS was immediately monitored at 510 nm with excitations at 460 nm recorded for 300 seconds using fluorescence spectrophotometer (Hitachi, Model F-7100, Japan) after which time an aqueous solution of Triton X-100 (20  $\mu\text{L}$ , 20% v/v) was immediately added to achieve the maximum change in dye fluorescence emission. The final transport trace was obtained and normalized based on the saturation fluorescence intensity after addition of triton according to Equation S1.

## The CF Dye Leakage Assay

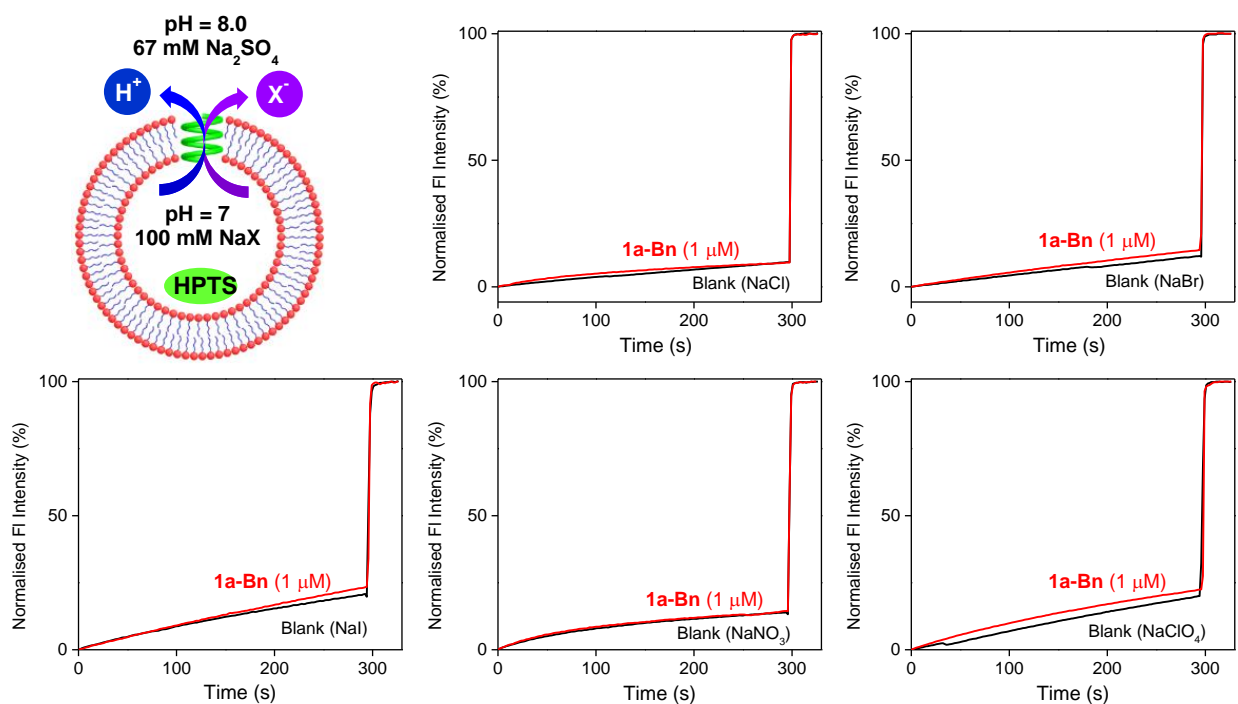
A thin lipid film was prepared in 2 mL microtube by evaporating 1 mL solution EYPC by N<sub>2</sub> flow in (25 mg/mL) and dried in vacuum for overnight. After that lipid film was hydrated with 1 mL buffer (10 mM HEPES, 100 mM NaCl, 50 mM CF, pH 7.5) for 1 h with occasional vortexing for 5 times (after every 15 minutes) and then subjected to 10 freeze-thaw cycle (freezing in liquid N<sub>2</sub> for 60 seconds and heating at 55 °C for 2.5 minutes). The vesicle solution was extruded through a polycarbonate membrane with polycarbonate membrane (0.1 μm) to produce a homogeneous suspension of LUVs with CF encapsulated inside. The extravesicular CF dye was removed by using size exclusion chromatography (stationary phase: Sephadex G-50, GE Healthcare, USA, mobile phase: HEPES buffer with 100 mM NaCl, pH 7.5) and diluted with the mobile phase to yield 3.2 mL of 10 mM lipid stock solution.

In a clean and dry fluorescence cuvette 30 μL of above lipid solution and 1970 μL of 10 mM HEPES buffer (100 mM NaCl, pH 7.5) was taken and kept in slowly stirring condition by a magnetic stirrer equipped with the fluorescence instrument (at  $t = 0$  s). The time course of CF fluorescence emission intensity,  $F_t$  was observed at  $\lambda_{em} = 517$  nm ( $\lambda_{ex} = 492$  nm). Melittin and Compound **1a** was added at  $t = 70$  s and finally at  $t = 370$  s, 20 μL of 20% Triton X- was added to lyse all vesicles for 100% chloride influx. Melittin is a well-known pore forming channel which can cause leakage of carboxyfluorescein through it. Fluorescence intensities ( $F_t$ ) were normalized to fractional emission intensity  $I_F$  according to Equation S2. The data obtained from this experiment was compared with Melittin and it is confirmed polymer **1a** does defect lipid membrane and does not form large pore ( $< 12\text{\AA}$ ).





**Figure S16.** Schematic representation of CF dye leakage assay and fluorescence intensity changes in self-quenching CF dye ( $\lambda_{\text{ex}} = 492 \text{ nm}$ ,  $\lambda_{\text{em}} = 517 \text{ nm}$ ) after additions of polymer **1a** at  $1 \mu\text{M}$  as well as melittin at different concentrations ( $0.15$ , and  $0.2 \mu\text{M}$ ). Inside LUV:  $10 \text{ mM HEPES}$ ,  $500 \text{ mM CF}$ ,  $\text{pH} = 7.5$ ; outside LUV:  $10 \text{ mM HEPES}$ ,  $\text{pH} = 7.5$ . CF dye refers to 5(6)-Carboxyfluorescein with smaller dimensions being  $10 \text{ \AA} \times 10 \text{ \AA}$ .



**Figure S17.** Undetectable anion transport activity by **1a-Bn** at  $1 \mu\text{M}$ . This suggests that membrane defects are not responsible for the observed anion transport by **1a**.

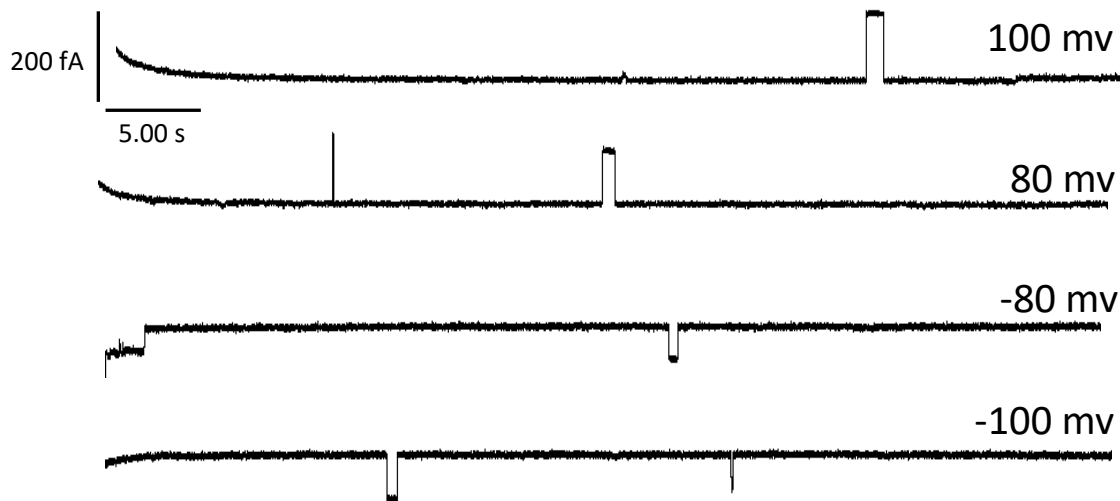
**Single channel current measurement in planar lipid bilayers** (Warner Instruments, Hamden, CT).

The chloroform solution of 1,2-diphytanoyl-*sn*-glycero-3-phosphocholine (diPhyPC, 10 mg/ml, 20 uL) was mixed with polymer channel **1a** at a molar ratio of 20000:1 (diPhyPC:**1a**). Nitrogen gas was used to remove chloroform to form a thin film, which was dissolved in *n*-decane (8 uL). 0.2 uL of this *n*-decane solution was injected into the aperture (diameter = 200 um) of the Delrin®cup (Warner Instruments, Hamden, CT) with the *n*-decane removed by using nitrogen gas. In a typical experiment for conductance measurement, both the chamber (*cis* side) and Delrin cup (*trans* side) were filled with an aqueous KCl solution (1.0 M, 1.0 mL). The obtained current-voltage (I-V) curve was then analysed by FitMaster (HEKA) with a digital filter at 100 Hz and fitted using a linear equation of  $y = a + b \cdot x$  where slope  $b$  is the conductance value ( $\gamma_{K^+}$ ) in the unit of nS.

For selectivity determination, unsymmetric baths (*cis* = 0.5 M KCl, *trans* = 1.0 M KCl) were used. Ag-AgCl electrodes were inserted into the two solutions with the *cis* chamber grounded. Planar lipid bilayer was formed by painting 0.3 uL of the lipid-containing *n*-decane solution around the *n*-decane-pretreated aperture. Successful formation of planar lipid bilayers can be established with a capacitance value ranging from 80-120 pF. These single channel currents were then measured using a Warner BC-535D bilayer clamp amplifier, collected by PatchMaster (HEKA) with a sample interval at 5 kHz and filtered with an 8-pole Bessel filter at 1 kHz (HEKA). The data were analysed by FitMaster(HEKA) with a digital filter at 100 Hz. After following the above procedure to obtain I-V curves, the selectivity of the channels for  $K^+$  over  $M^+$ , defined as the permeability ratio of two ions (e.g.,  $P_{K^+ \text{ trans}}/P_{M^+ \text{ cis}}$ ), can be obtained by fitting the I-V curve using the following simplified Goldman–Hodgkin–Katz equation:

$$\varepsilon_{\text{rev}} = RT/F \times \ln( P_{K^+ \text{ trans}} / P_{M^+ \text{ cis}} )$$

where  $\varepsilon_{\text{rev}}$  is the reversal membrane potential;  $R$  is the universal gas constant ( $8.314 \text{ J.K}^{-1}.\text{mol}^{-1}$ );  $T$  the temperature in Kelvin (298 K);  $F$  is the Faraday's constant ( $96485 \text{ C.mol}^{-1}$ );  $P$  is the permeability of the channel for ions



**Figure S18.** Single channel current traces at various voltages recorded in unsymmetric baths (*cis* = 0.5 M KCl and *trans* = 1.0 M KCl). Fitting the linear current-voltage (I-V curve) using the equation ( $\epsilon_{rev} = RT/F * \ln\{(P_{K^+}[K^+]_{trans} + P_{Cl^-}[Cl^-]_{cis})/(P_{K^+}[K^+]_{cis} + P_{Cl^-}[Cl^-]_{trans})\}$ ) yields reversal membrane potential ( $\epsilon_{rev} = -21.2$  mV) from which selectivity ratio of  $Cl^-/K^+$  was calculated to be 13.

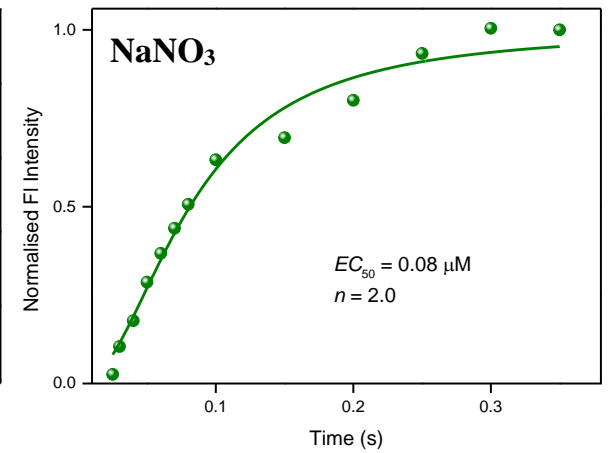
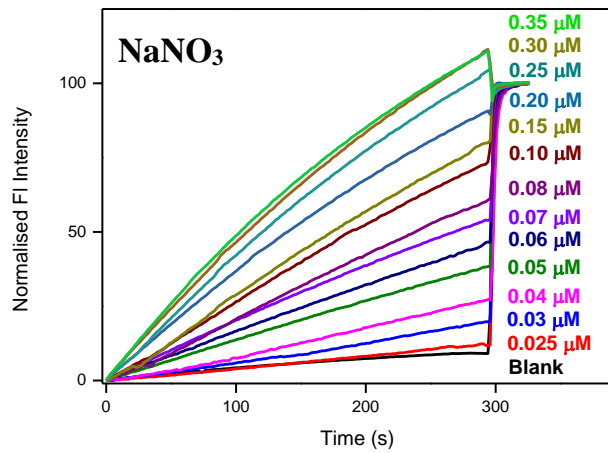
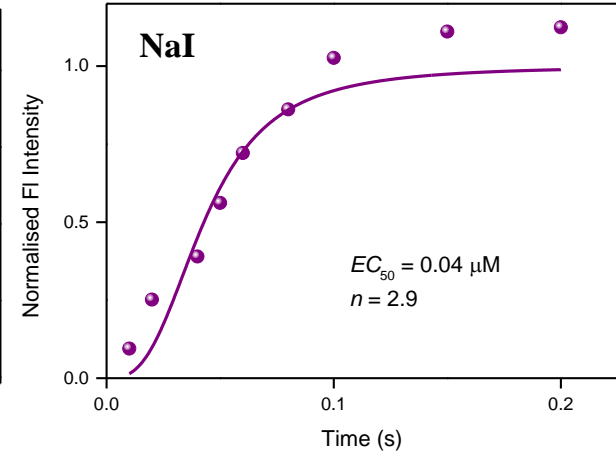
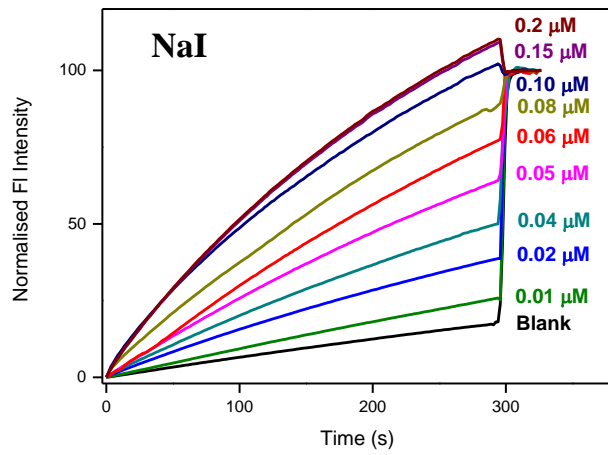
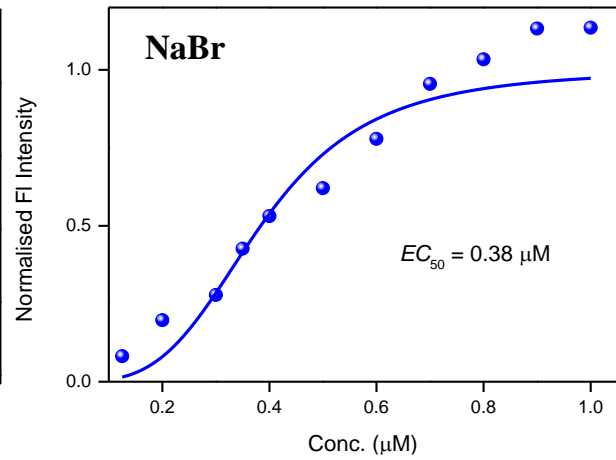
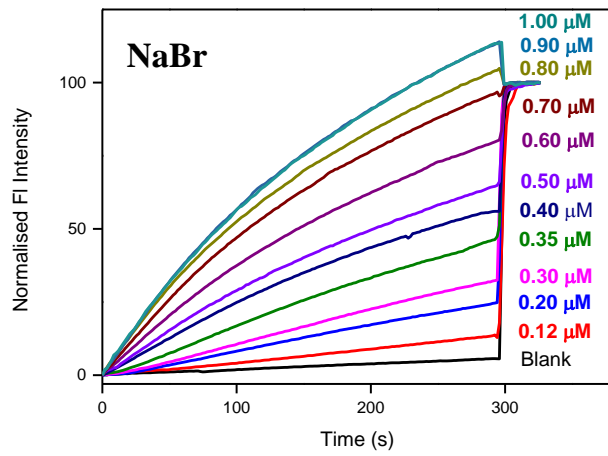
### Anion Selectivity Using the HPTS Assay

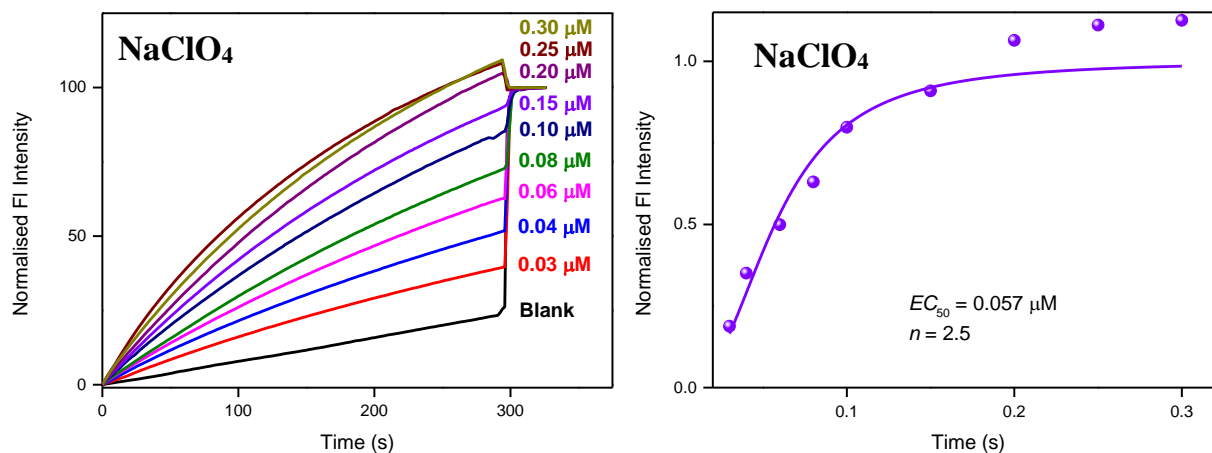
Egg yolk L- $\alpha$ -phosphatidylcholine (EYPC, 1 mL, 25 mg/mL in  $CHCl_3$ , Avanti Polar Lipids, USA) was taken in a 2 mL microtube and  $CHCl_3$  was evaporated by purging  $N_2$  slowly. After drying the resulting film under high vacuum overnight at room temperature, the film was hydrated with 4-(2-hydroxyethyl)-1-piperazine-ethane sulfonic acid (HEPES) buffer solution (1 mL, 10 mM HEPES, 100 mM NaX where  $X^- = Cl^-, Br^-, I^-, NO_3^-, ClO_4^-$  and  $SO_4^{2-}$  pH = 7.0) containing a pH sensitive dye 8-hydroxy-pyrene-1,3,6-trisulfonic acid (HPTS, 0.5 mM) at room temperature for 1 hour (with occasional vortexing after every 15 minutes) to give a milky suspension. The mixture was then subjected to 10 freeze-thaw cycles (freezing in liquid  $N_2$  for 1 minute and heating at 55 °C in water bath for 2.5 minutes). The vesicle suspension was extruded through polycarbonate membrane (0.1  $\mu m$ ) to produce a homogeneous suspension of LUVs of about 100 nm in diameter with HPTS encapsulated inside. The extravesicular HPTS dye was removed by using size exclusion chromatography (stationary phase: Sephadex G-50, GE Healthcare, USA, mobile phase: HEPES buffer with 100 mM NaX where  $X^- = Cl^-, Br^-, I^-, NO_3^-$  and  $ClO_4^-$ , pH = 7.0) and diluted with the mobile phase to yield 3.2 mL of 10 mM lipid stock solution.

The HPTS-containing LUV suspension (30  $\mu\text{L}$ , 10 mM in 10 mM HEPES buffer containing 100 mM NaX where  $\text{X}^- = \text{Cl}^-, \text{Br}^-, \text{I}^-, \text{NO}_3^-, \text{ClO}_4^-$  and  $\text{SO}_4^{2-}$  at pH = 7.0) was added to a HEPES buffer solution (1.7 mL, 10 mM HEPES, 67 mM  $\text{Na}_2\text{SO}_4$ , pH= 8.0) to create a pH gradient for ion transport study. A solution of polymer **1a** (0.15  $\mu\text{M}$ ) in DMF was then injected into the suspension under gentle stirring. Upon the addition of channels, the emission of HPTS was immediately monitored at 510 nm with excitations at 460 for 300 seconds using fluorescence spectrophotometer (Hitachi, Model F-7100, Japan) after which time an aqueous solution of Triton X-100 (20  $\mu\text{L}$ , 20% v/v) was immediately added to achieve the maximum change in dye fluorescence emission. The final transport trace was normalized based on equation S2.

Concentration dependent assay was carried out with all 6 different LUVs containing NaX (where  $\text{X}^- = \text{Cl}^-, \text{Br}^-, \text{I}^-, \text{NO}_3^-, \text{ClO}_4^-$  and  $\text{SO}_4^{2-}$ ). From this concentration dependent plot,  $EC_{50}$  was calculated for each anion salt and compared.

## EC<sub>50</sub> Determination using the HPTS assays

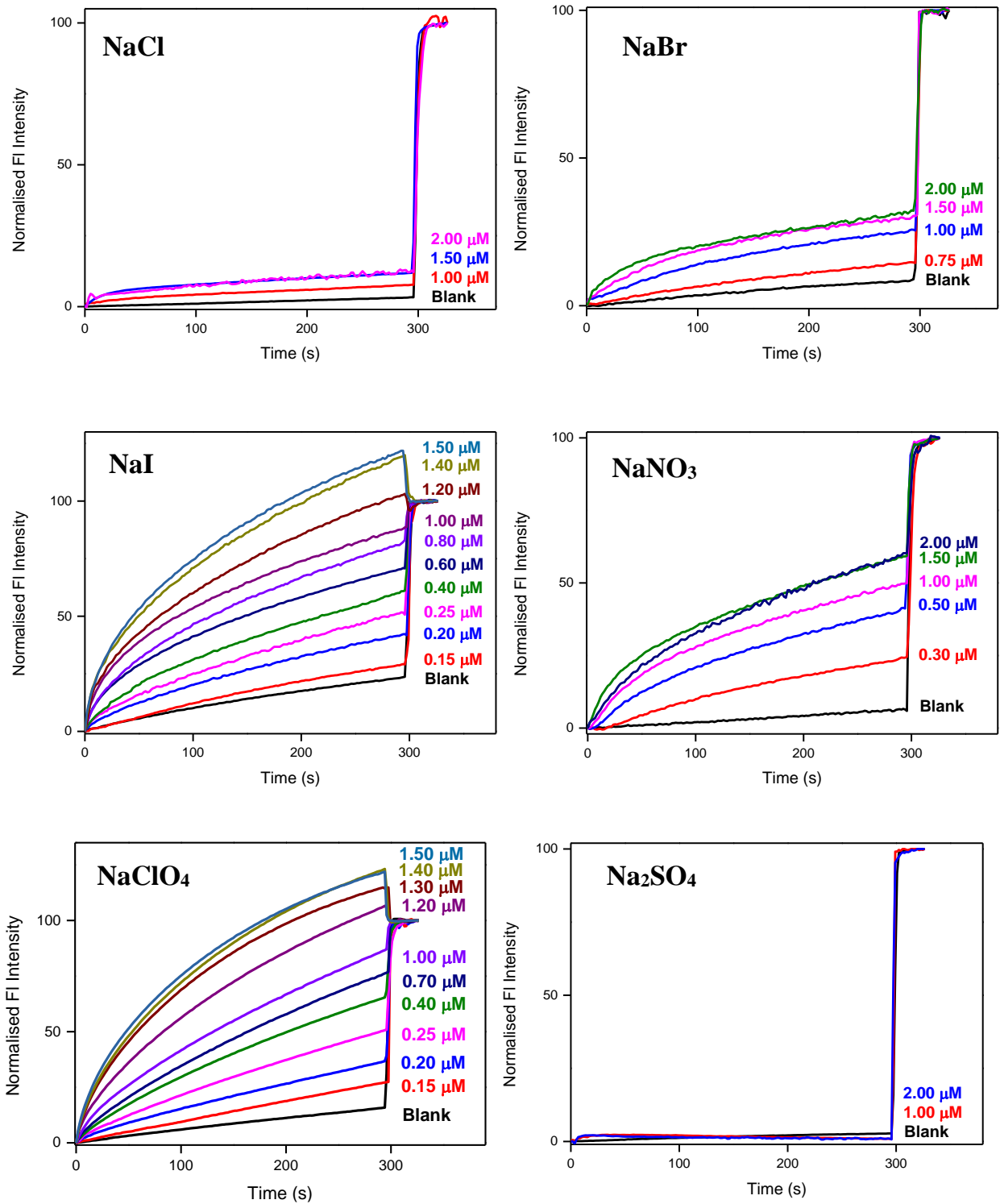




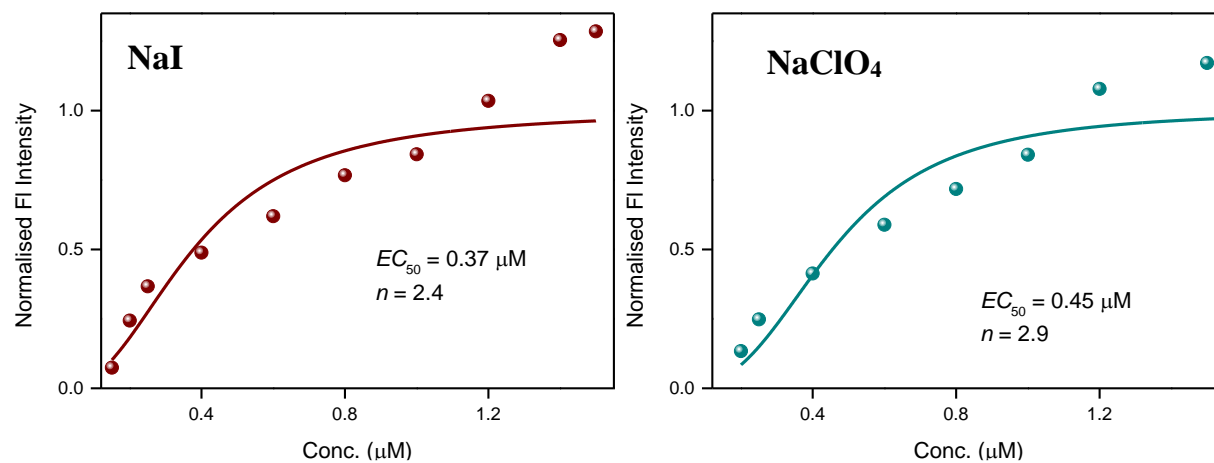
**Figure S19.** Determination of  $EC_{50}$  values from concentration-dependent plots with various anions using the HPTS assay for polymer **1a**.

### Anion Selectivity Using the Cholesterol-Containing HPTS Assay

Egg yolk L- $\alpha$ -phosphatidylcholine (EYPC, 1 mL, 25 mg/mL in  $\text{CHCl}_3$ , Avanti Polar Lipids, USA) and cholesterol (6.3 mg, 0.016 mM) was taken in a 2 mL microtube and  $\text{CHCl}_3$  was evaporated by purging  $\text{N}_2$  slowly. After drying the resulting film under high vacuum overnight at room temperature, the film was hydrated with 4-(2-hydroxyethyl)-1-piperazine-ethane sulfonic acid (HEPES) buffer solution (1 mL, 10 mM HEPES, 100 mM NaX, where  $\text{X}^- = \text{Cl}^-$ ,  $\text{Br}^-$ ,  $\text{I}^-$ ,  $\text{NO}_3^-$ ,  $\text{ClO}_4^-$  and  $\text{SO}_4^{2-}$ , pH = 7.0) containing a pH sensitive dye 8-hydroxy-pyrene-1,3,6-trisulfonic acid (HPTS, 0.5 mM) at room temperature for 1 hour (with occasional vortexing after every 15 minutes) to give a milky suspension. The mixture was then subjected to 10 freeze-thaw cycles (freezing in liquid  $\text{N}_2$  for 1 minute and heating at 55 °C in water bath for 2.5 minutes). The vesicle suspension was extruded through polycarbonate membrane (0.1  $\mu\text{m}$ ) to produce a homogeneous suspension of LUVs of about 100 nm in diameter with HPTS encapsulated inside. The extraventricular HPTS dye was removed by using size exclusion chromatography (stationary phase: Sephadex G-50, GE Healthcare, USA, mobile phase: HEPES buffer with 100 mM NaX, where  $\text{X}^- = \text{Cl}^-$ ,  $\text{Br}^-$ ,  $\text{I}^-$ ,  $\text{NO}_3^-$  and  $\text{ClO}_4^-$ , pH = 7.0) and diluted with the mobile phase to yield 3.2 mL of 10 mM lipid stock solution.



**Figure S20.** Concentration-dependent ion transport curves with various anions using cholesterol-containing HPTS assay for polymer **1a**.



**Figure S21.** Determination of  $EC_{50}$  values from concentration-dependent plots for  $\text{I}^-$  and  $\text{ClO}_4^-$  from Figure S20 for polymer **1a**.



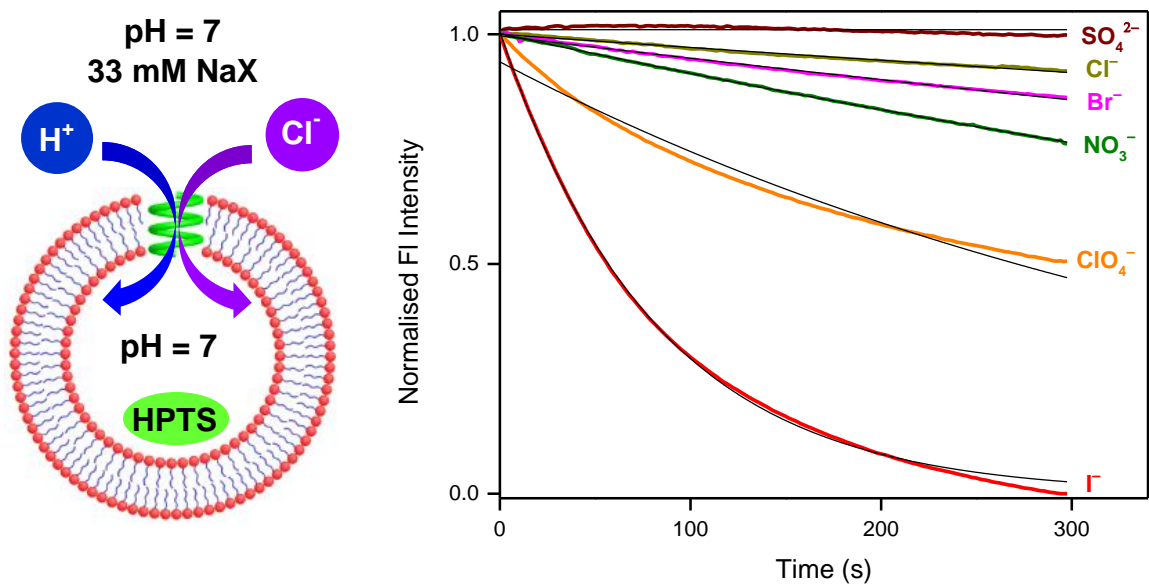
### Anion Selectivity Using the HPTS assay without salt<sup>[23]</sup>

Egg yolk L- $\alpha$ -phosphatidylcholine (EYPC, 1 ml, 25 mg/mL in CHCl<sub>3</sub>, Avanti Polar Lipids, USA) was taken in a 2 mL microtube and CHCl<sub>3</sub> was evaporated by purging N<sub>2</sub> slowly. After drying the resulting film under high vacuum overnight at room temperature, the film was hydrated with 4-(2-hydroxyethyl)-1-piperazine-ethane sulfonic acid (HEPES) buffer solution (1 mL, 10 mM HEPES, pH = 7.0) containing a pH sensitive dye 8-hydroxy-ypyrene-1,3,6-trisulfonic acid (HPTS, 0.5 mM) at room temperature for 1 hour (with occasional vortexing after every 15 minutes) to give a milky suspension. The mixture was then subjected to 10 freeze-thaw cycles (freezing in liquid N<sub>2</sub> for 1 minute and heating at 55 °C in water bath for 2.5 minutes). The vesicle suspension was extruded through polycarbonate membrane (0.1  $\mu$ m) to produce a homogeneous suspension of LUVs of about 100 nm in diameter with HPTS encapsulated inside. The extraventricular HPTS dye was removed by using size exclusion chromatography (stationary phase: Sephadex G-50, GE Healthcare, USA, mobile phase: HEPES buffer of pH = 7.0) and diluted with the mobile phase to yield 3.2 mL of 10 mM lipid stock solution.

The HPTS-containing LUV suspension (30  $\mu$ L, 10 mM in 10 mM HEPES buffer at pH = 7.0) was added to a HEPES buffer solution (1.7 mL, 10 mM HEPES, pH= 7.0). At  $t = 40$  s 33 mM NaX (where X<sup>-</sup> = Cl<sup>-</sup>, Br<sup>-</sup>, I<sup>-</sup>, NO<sub>3</sub><sup>-</sup>, ClO<sub>4</sub><sup>-</sup> and SO<sub>4</sub><sup>2-</sup>) was added in the cuvette. Stock solution of polymer **1a** (0.15  $\mu$ M) in DMF was then injected into the suspension at  $t = 70$  s under gentle stirring. Upon the addition of channels, the emission of HPTS was immediately monitored at 510 nm with excitations at 460 for 300 seconds using fluorescence spectrophotometer (Hitachi, Model F-7100, Japan) after which time an aqueous solution of Triton X-100 (20  $\mu$ L, 20% v/v) was immediately added to achieve the maximum change in dye fluorescence emission. The final transport trace was generated after subtraction of their own respective backgrounds over the whole duration of 300 s, with further normalization using the maximum reading obtained after addition of triton (Figure S22). Therefore, the fluorescence quenching effect of each anion towards HPTS dye as well as their own intrinsic membrane permeability have been nullified. Rate constant values were obtained by fitting transport curves to a single exponential decay equation:

$$I_F = A_1 e^{(-k_1 t)}$$

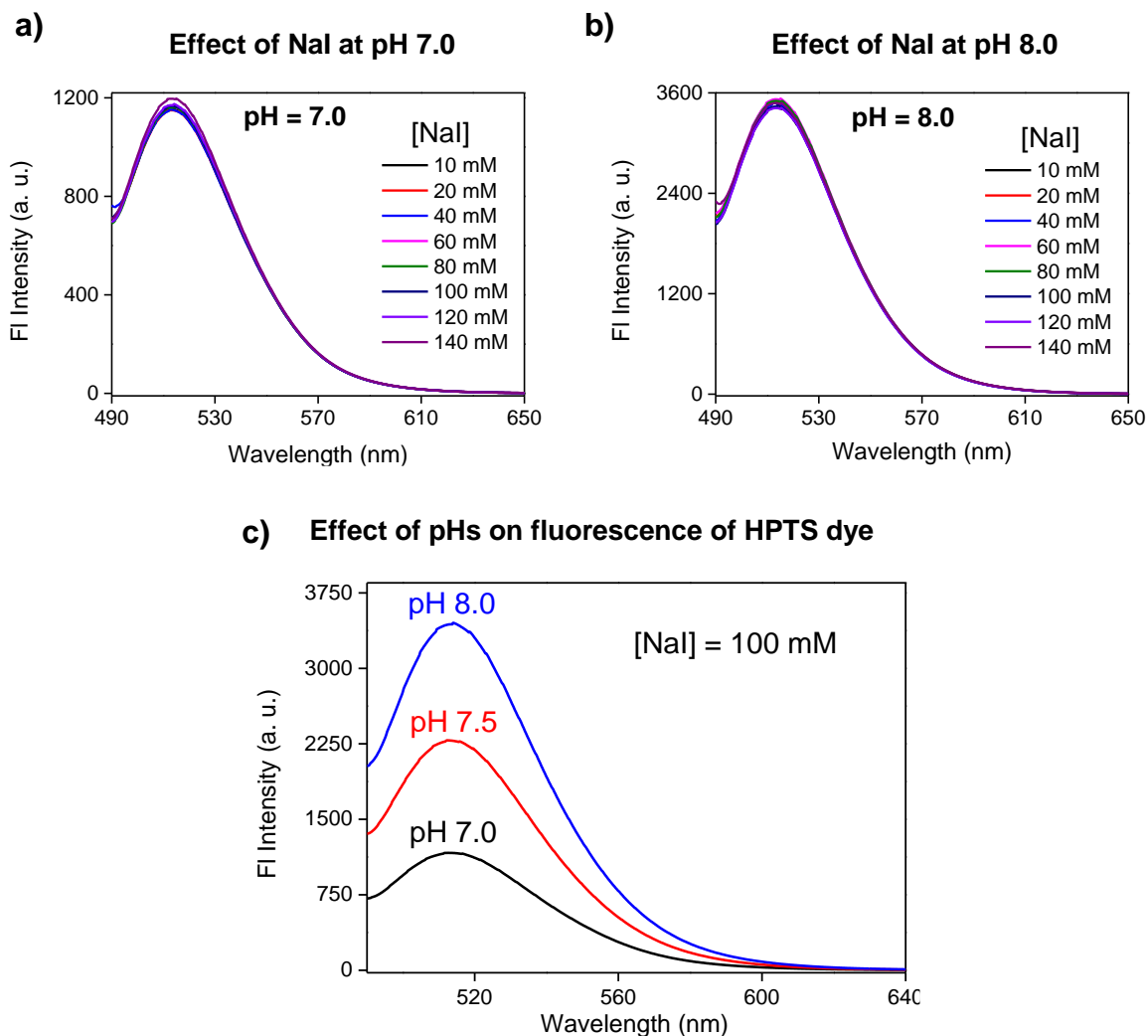
## Determination of initial rate constants



Anion	$k_1$ (rate constant, s <sup>-1</sup> )
SO <sub>4</sub> <sup>2-</sup>	-8.0E <sup>-86</sup>
Cl <sup>-</sup>	2.9E <sup>-4</sup>
Br <sup>-</sup>	5.04E <sup>-4</sup>
I <sup>-</sup>	0.01229
NO <sub>3</sub> <sup>-</sup>	9.04E <sup>-4</sup>
ClO <sub>4</sub> <sup>-</sup>	0.00233

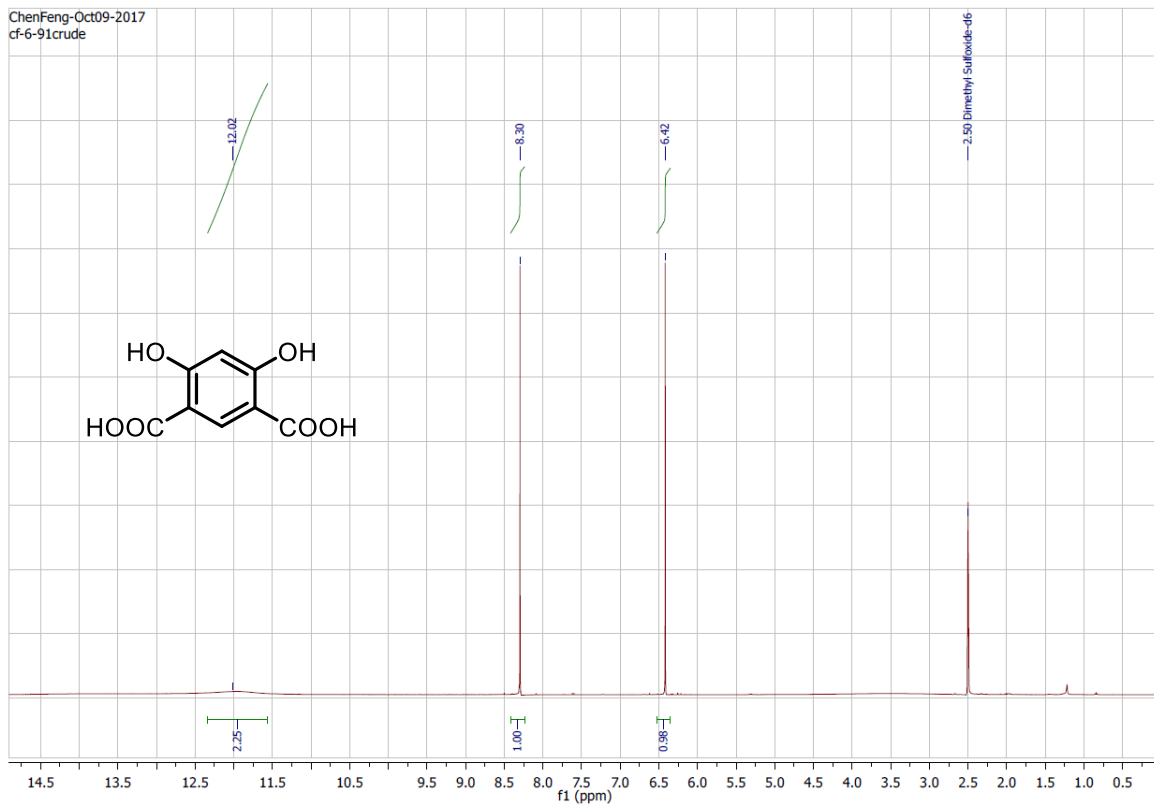
**Figure S22.** Fitted plots (thin black lines) to calculate initial rate constant ( $k_1$ ) values of anion transports by channel **1a** at 0.15  $\mu$ M by the HPTS assay using by Kim.

## Effect of heavy iodine atom on the fluorescence intensity of the HPTS dye

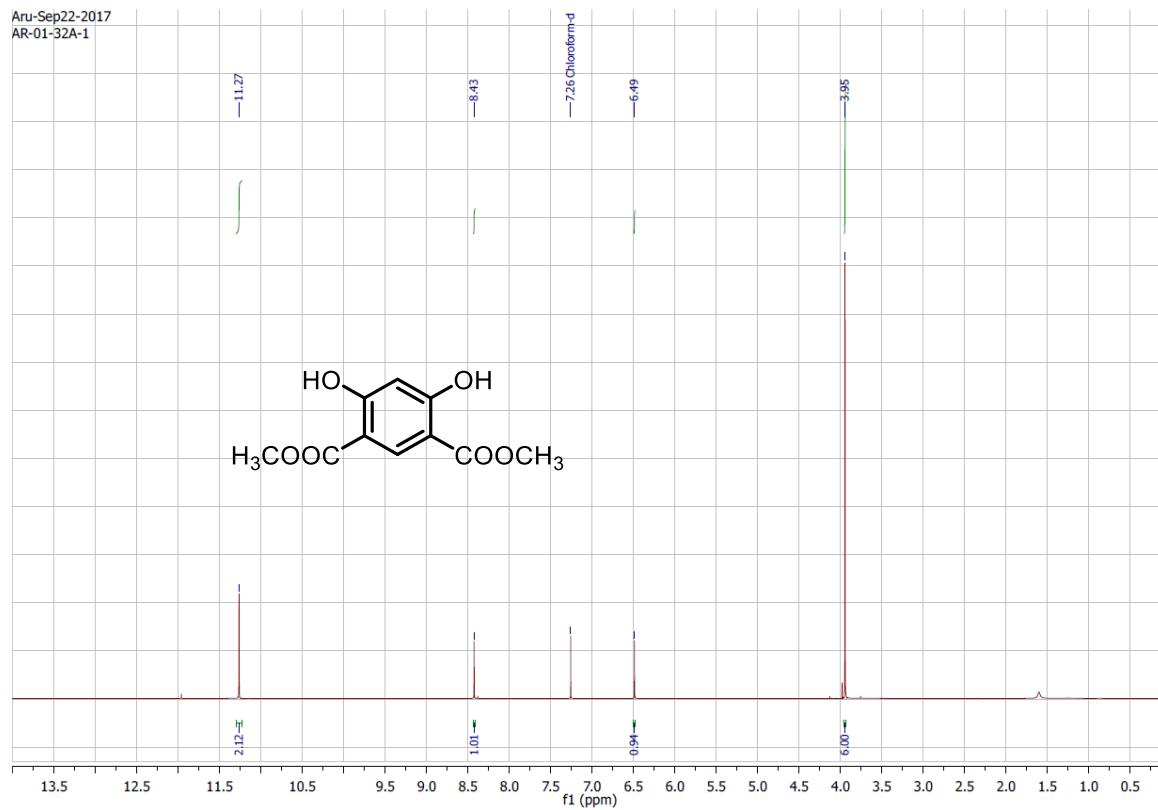


**Figure S23.** Effects of NaI on the fluorescence intensity of the HPTS dye at various concentrations and pHs. These data confirm that heavy iodine atom has no quenching effect on the fluorescence intensity of HPTS dye, and thus all the fluorescence changes observed are a result of efficient transport of iodide anions.

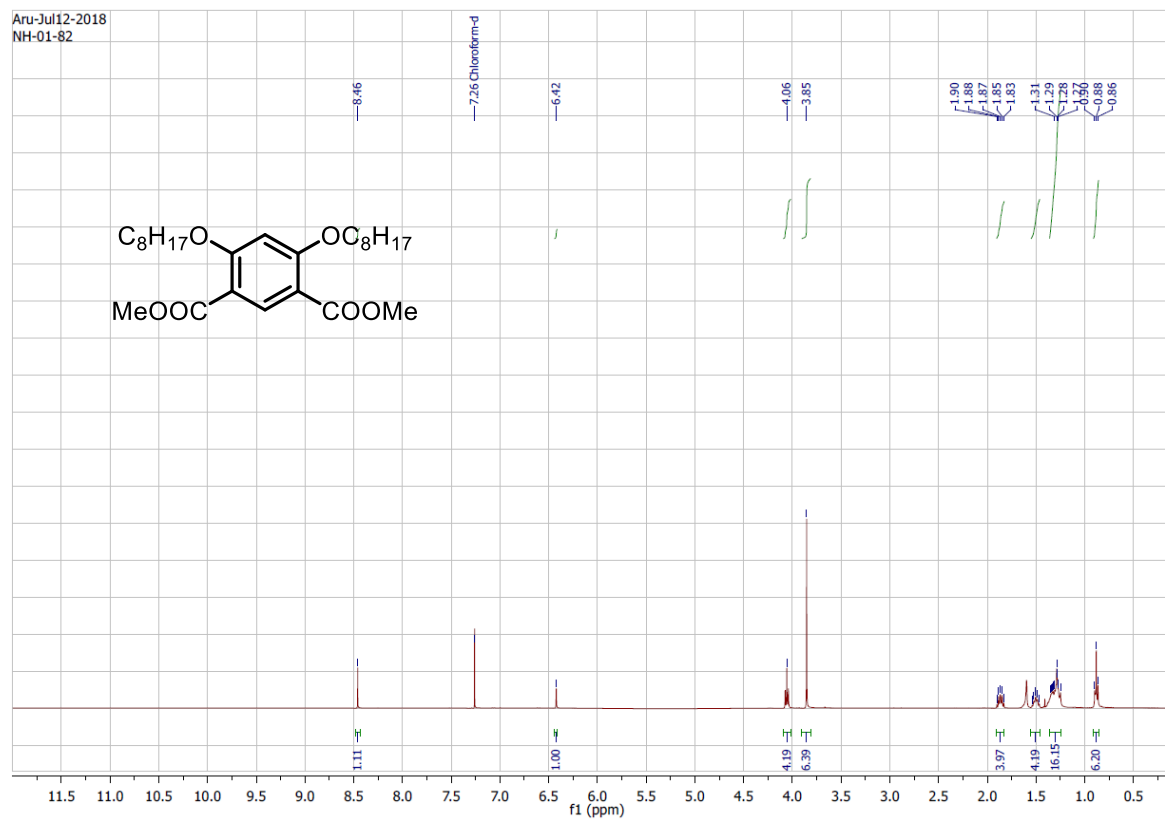
# <sup>1</sup>H NMR Spectra



Aru-Sep22-2017  
AR-01-32A-1

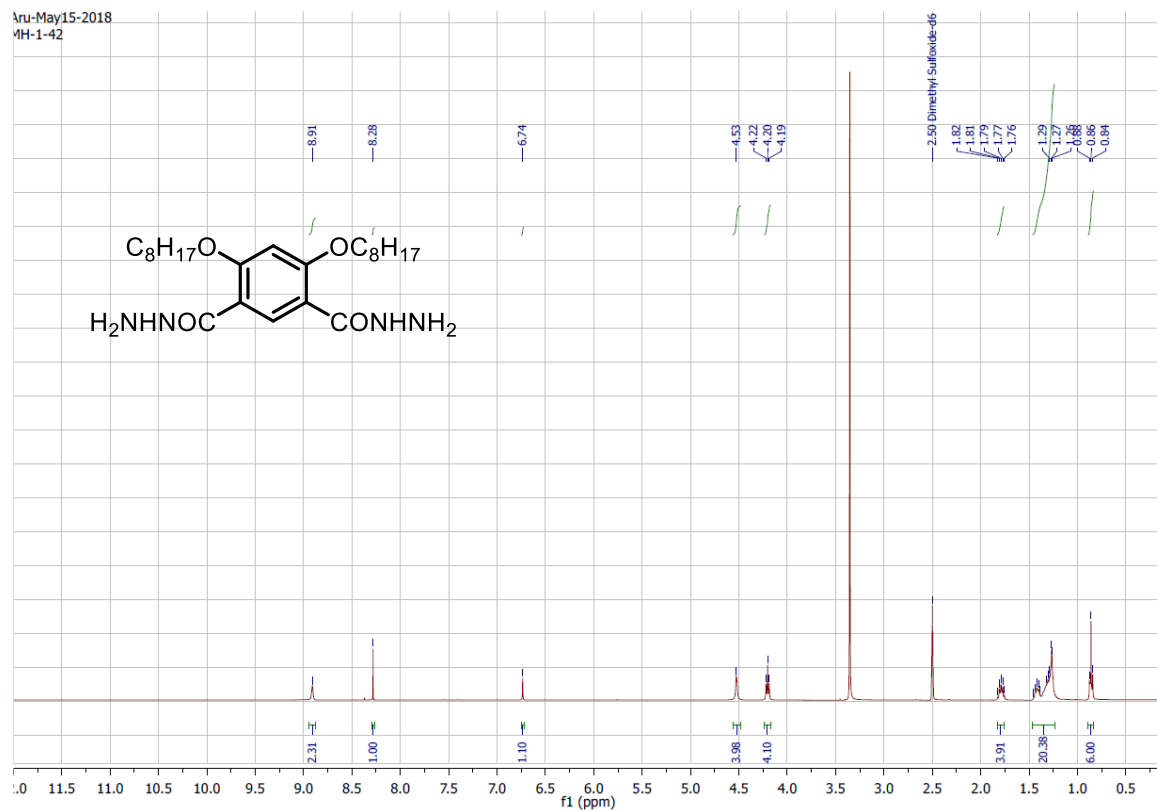


Aru-Jul12-2018  
NH-01-82

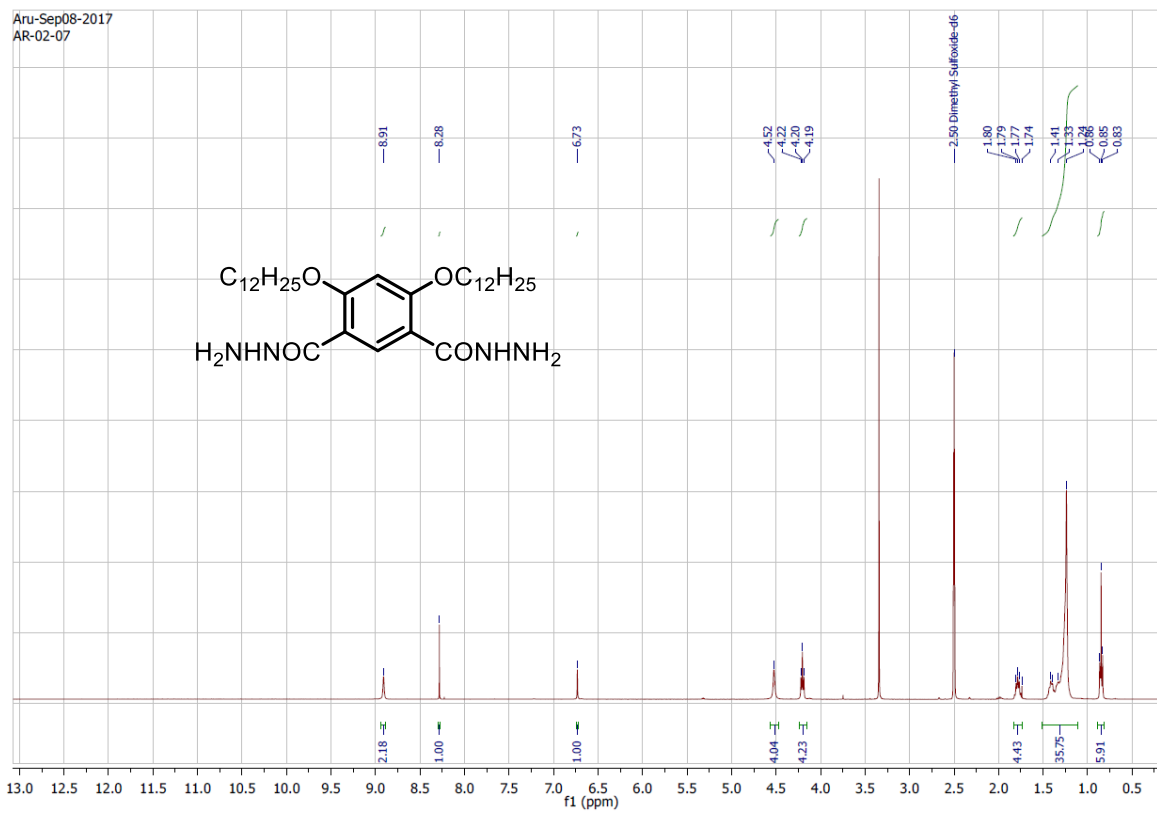




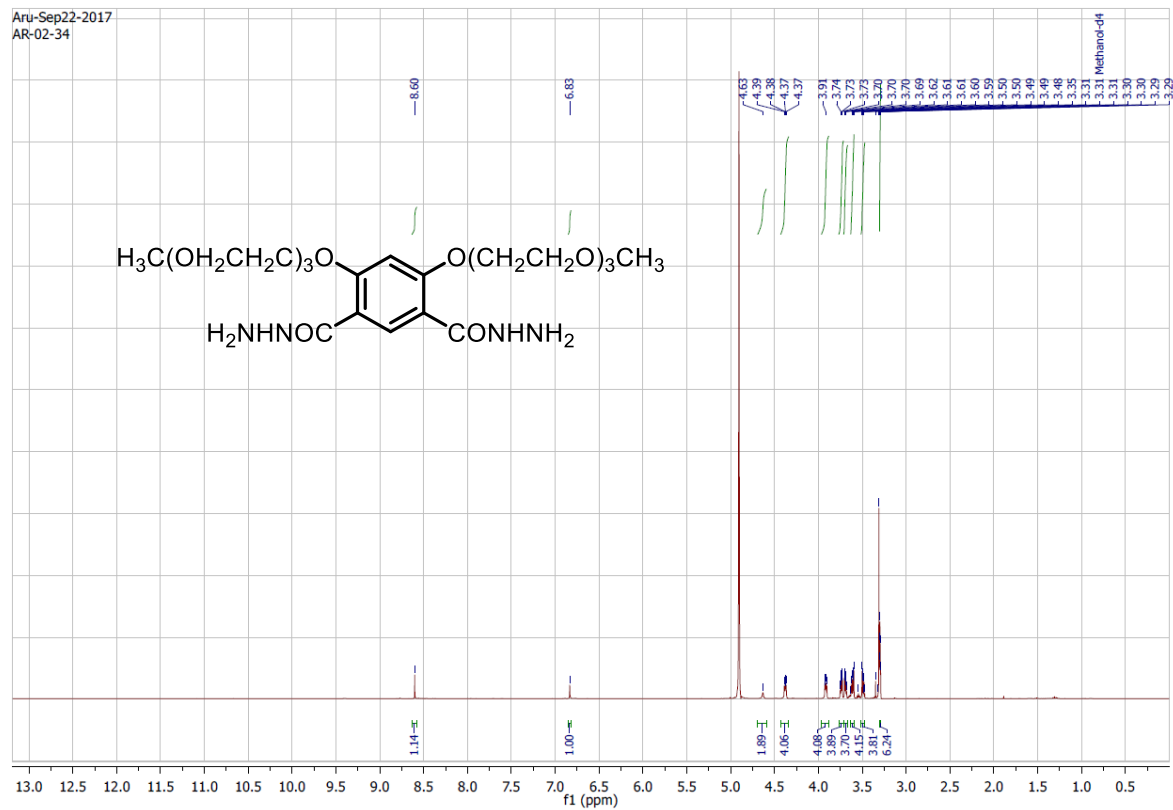
Aru-May15-2018  
MH-1-42



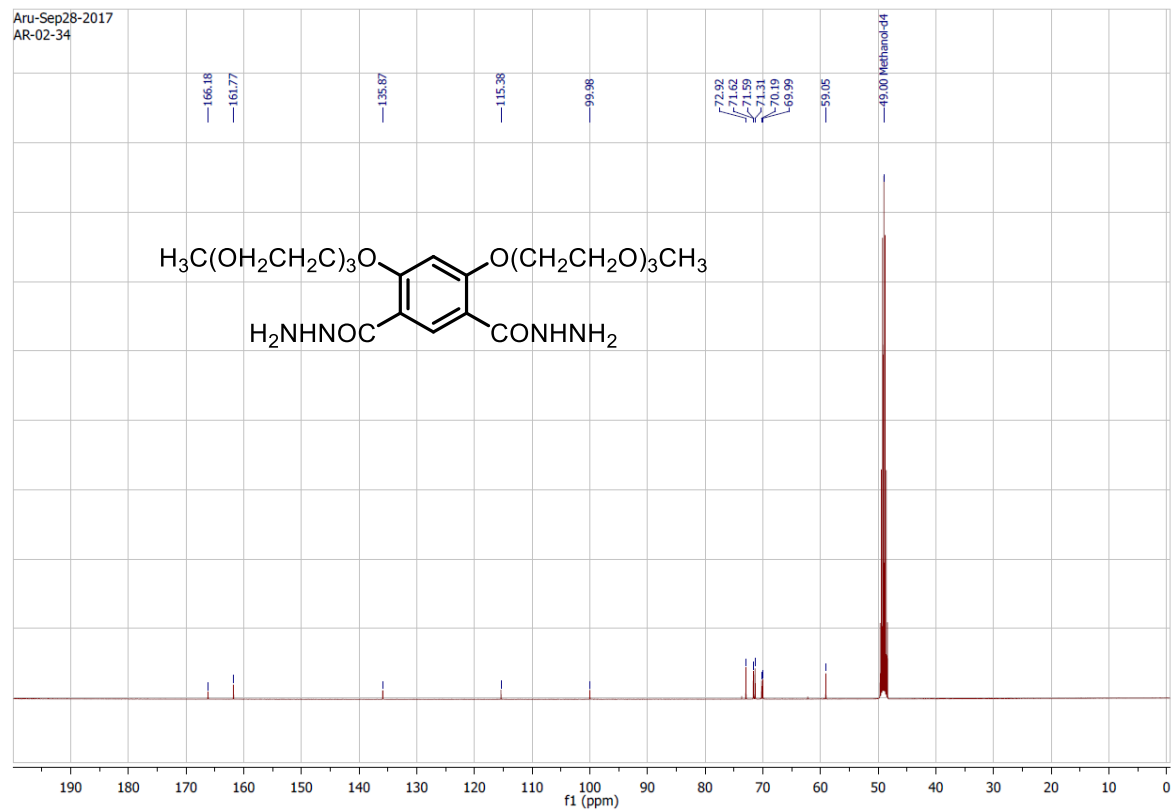
Aru-Sep08-2017  
AR-02-07



Aru-Sep22-2017  
AR-02-34

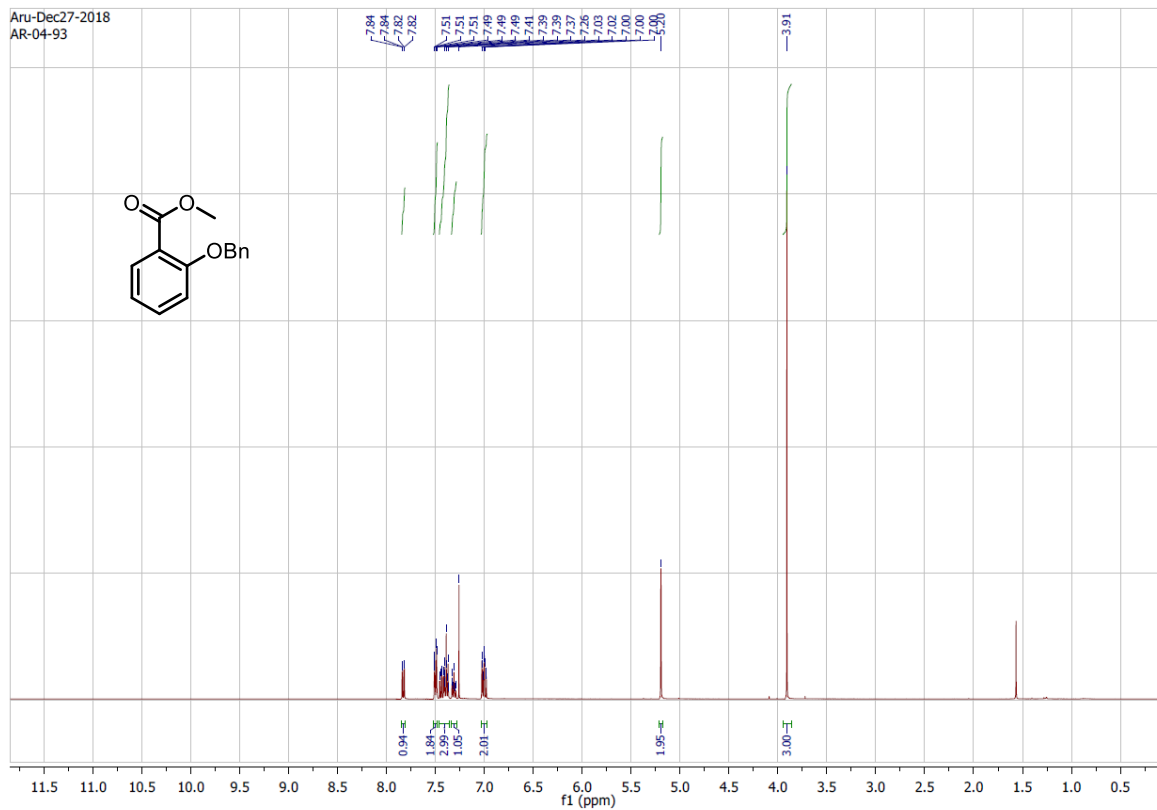


Aru-Sep28-2017  
AR-02-34

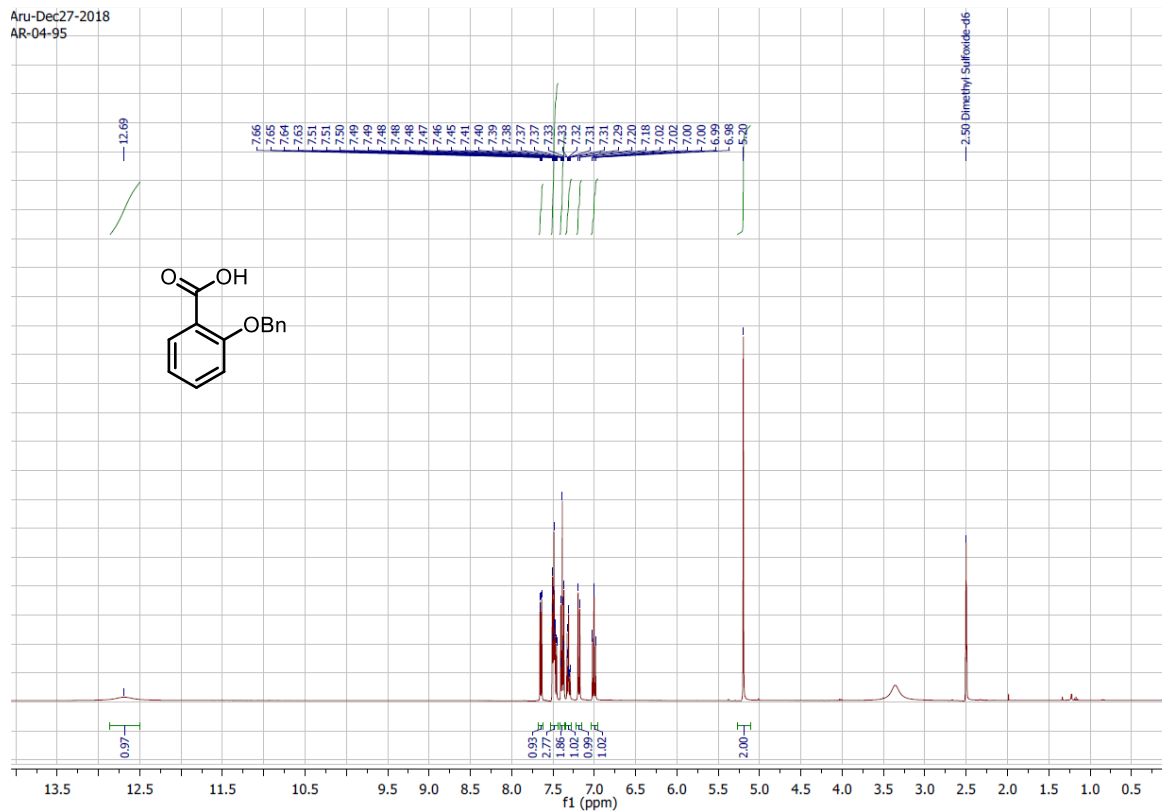




Aru-Dec27-2018  
AR-04-93



Aru-Dec27-2018  
AR-04-95



## References:

- [1] H. Zeng, R. S. Miller, R. A. Flowers, B. Gong, *J. Am. Chem. Soc.* **2000**, *122*, 2635-2644.
- [2] Y. Yang, Z.-Y. Yang, Y.-P. Yi, J.-F. Xiang, C.-F. Chen, L.-J. Wan, Z.-G. Shuai, *J. Org. Chem.* **2007**, *72*, 4936-4946.
- [3] C. Zhou, W. Cai, G. T. Wang, X. Zhao, Z. T. Li, *Macromol. Chem. Phys.* **2010**, *211*, 2090-2101.
- [4] E. Orłowska, A. Roller, H. Wiesinger, M. Pignitter, F. Jirsa, R. Krachler, W. Kandioller, B. K. Keppler, *RSC Adv.* **2016**, *6*, 40238-40249.
- [5] J. M. H. Cheng, L. Liu, D. G. Pellicci, S. J. J. Reddiex, R. N. Cotton, T.-Y. Cheng, D. C. Young, I. Van Rhijn, D. B. Moody, J. Rossjohn, D. P. Fairlie, D. I. Godfrey, S. J. Williams, *Chem. Eur. J.* **2017**, *23*, 1694-1701.
- [6] W. Humphrey, A. Dalke, K. Schulten, *J. Mol. Graph. Model.* **1996**, *14*, 33-38.
- [7] J. C. Phillips, R. Braun, W. Wang, J. Gumbart, E. Tajkhorshid, E. Villa, C. Chipot, R. D. Skeel, L. Kale, K. Schulten, *J. Comput. Chem.* **2005**, *26*, 1781-1802.
- [8] T. Darden, D. York, L. Pedersen, *J. Chem. Phys.* **1993**, *98*, 10089-10092.
- [9] aS. E. Feller, Y. Zhang, R. W. Pastor, B. R. Brooks, *J. Chem. Phys.* **1995**, *103*, 4613-4621; bG. J. Martyna, D. J. Tobias, M. L. Klein, *J. Chem. Phys.* **1994**, *101*, 4177-4189.
- [10] D. J. Sindhikara, S. Kim, A. F. Voter, A. E. Roitberg, *J. Chem. Theory Comput.* **2009**, *5*, 1624-1631.
- [11] J. B. Klauda, R. M. Venable, J. A. Freites, J. W. O'Connor, D. J. Tobias, C. Mondragon-Ramirez, I. Vorobyov, A. D. MacKerell Jr, R. W. Pastor, *J. Phys. Chem B* **2010**, *114*, 7830-7843.
- [12] D. Beglov, B. Roux, *J. Chem. Phys.* **1994**, *100*, 9050-9063.
- [13] P. Li, L. F. Song, K. M. Merz, *J. Chem. Theory Comput.* **2015**, *11*, 1645-1657.
- [14] S. Miyamoto, P. A. Kollman, *J. Comput. Chem.* **1992**, *13*, 952-962.
- [15] H. C. Andersen, *J. Comput. Phys.* **1983**, *52*, 24-34.
- [16] D. R. Roe, T. E. Cheatham, III, *J. Chem. Theory Comput.* **2013**, *9*, 3084-3095.
- [17] K. Vanommeslaeghe, E. Hatcher, C. Acharya, S. Kundu, S. Zhong, J. Shim, E. Darian, O. Guvench, P. Lopes, I. Vorobyov, *J. Comput. Chem.* **2010**, *31*, 671-690.
- [18] S. Jo, T. Kim, V. G. Iyer, W. Im, *J. Comput. Chem.* **2008**, *29*, 1859-1865.
- [19] W. L. Jorgensen, J. Chandrasekhar, J. D. Madura, R. W. Impey, M. L. Klein, *J. Chem. Phys.* **1983**, *79*, 926-935.
- [20] G. M. Torrie, J. P. Valleau, *J. Comput. Phys.* **1977**, *23*, 187-199.
- [21] Y. Sugita, A. Kitao, Y. Okamoto, *J. Chem. Phys.* **2000**, *113*, 6042-6051.
- [22] S. Kumar, J. M. Rosenberg, D. Bouzida, R. H. Swendsen, P. A. Kollman, *J. Comput. Chem.* **1992**, *13*, 1011-1021.
- [23] B. P. Benke, P. Aich, Y. Kim, K. L. Kim, M. R. Rohman, S. Hong, I.-C. Hwang, E. H. Lee, J. H. Roh, K. Kim, *J. Am. Chem. Soc.* **2017**, *139*, 7432-7435.