Supporting Information

Evaluation of Imidazolium-based Ionic Liquids towards Vermicidal Activity: *In vitro & In silico studies*

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Spectral details of the synthesized ILs

The ^{1}H NMR and mass spectral data of the synthesized ILs were given bellow and the data is matching with literature reports (see supporting information for ^{1}H NMR and mass spectra of the synthesized ILs).

1-Ethyl-3-methylimidazolium bromide [EMIM]Br, Golden yellow flakes when it is dry, hygroscopic in nature. ^{1}H NMR ($CDCl_3$, 400 MHz) δ (ppm): 9.66 (1 H, s, N-C*H*-N), 7.36 (2 H, dq, *J* 6.2, 2.1, NC*H*CHN), 4.03 (2 H, q, *J* 7.4, N-C*H*$_2$CH$_3$), 3.72 (3 H, d, *J* 2.2, N-CH$_3$), 1.20 (3 H, t, *J* 7.4, CH$_2$-C*H*$_3$). ^{13}C NMR ($CDCl_3$, 100 MHz) δ (ppm): 133.79, 121.44, 119.98, 42.59, 34.33, 13.46. LC-MS: 111.1(M+-Br), 83.2 (M+-Br-C_2H_5).

1-Ethyl-3-methylimidazolium hydroxide [EMIM]OH, Golden yellow liquid. ^{1}H NMR ($CDCl_3$, 400 MHz) δ (ppm): 10.23 (1 H, s, N-C*H*-N), 7.57 (2 H, dt, *J* 6.0, 1.9, NC*H*C*H*N), 4.41 (2 H, q, *J* 7.4, N-C*H*$_2$CH$_3$), 4.11 (3 H, d, *J* 1.5, NC*H*$_3$), 1.60 (3 H, td, *J* 7.4, 1.4, CH$_2$-C*H*$_3$).^{13}C NMR ($CDCl_3$, 100 MHz) δ (ppm): 135.02, 122.49, 120.92, 43.78, 35.32, 14.42. LC-MS: 111.1(M+ -OH), 83.2 (M+-OH-C_2H_5).

1-Butyl-3-methylimidazolium bromide [BMIM]Br, Dark yellow solid when it is dry, hygroscopic in nature. ^{1}H NMR ($CDCl_3$, 400 MHz) δ (ppm): 10.03 (1 H, s, N-C*H*-N), 7.60 (1 H, t, *J* 1.8,

NC<u>*H*</u>CHN), 7.49 (1 H, t, *J* 1.8, NCHC<u>*H*</u>N), 4.23 (2 H, t, *J* 7.3, N-C<u>*H*</u>$_2$CH$_2$), 4.01 (3 H, s, NC<u>*H*</u>$_3$), 1.79 (2 H, tt, *J* 9.4, 6.8, NCH$_2$C<u>*H*</u>$_2$CH$_2$CH$_3$), 1.32-1.20 (2 H, m, NCH$_2$CH$_2$C<u>*H*</u>$_2$CH$_3$), 0.83 (3 H, t, *J* 7.3, NCH$_2$CH$_2$CH$_2$C<u>*H*</u>$_3$).^{13}C NMR (CDCl$_3$, 100 MHz) δ (ppm): 135.82, 123.35, 121.95, 48.94, 36.16, 31.44, 18.69, 12.91. LC-MS: 139.1 (M$^+$-Br), 111.1 (M+-Br-C$_2$H$_5$), 83.2 (M+-Br-C$_4$H$_9$).

1-Butyl-3-methylimidazolium hydroxide [BMIM]OH, Dark yellow liquid. ^{1}H NMR (CDCl$_3$, 400 MHz) δ (ppm): 10.11 (1 H, s, N-C<u>*H*</u>-N), 7.61 (1 H, t, *J* 1.8, NC<u>*H*</u>CHN), 7.48 (1 H, t, *J* 1.8, NCHC<u>*H*</u>N), 4.24 (2 H, t, *J* 7.4, N-C<u>*H*</u>$_2$CH$_2$), 4.02 (3 H, s, NC<u>*H*</u>$_3$), 1.85-1.74 (2 H, m, NCH$_2$C<u>*H*</u>$_2$CH$_2$CH$_3$), 1.27 (2 H, h, *J* 7.4, NCH$_2$CH$_2$C<u>*H*</u>$_2$CH$_3$), 0.84 (3 H, t, *J* 7.4, NCH$_2$CH$_2$CH$_2$C<u>*H*</u>$_3$). ^{13}C NMR (CDCl$_3$, 100 MHz) δ (ppm): 136.31, 123.59, 122.17, 49.29, 36.57, 31.95, 19.21, 13.29. LC-MS: 139.1 (M+-OH), 83.2 (M+-OH-C$_4$H$_9$).

1-Octyl-3-methylimidazolium bromide [OMIM]Br, Golden yellow liquid. ^{1}H NMR (CDCl$_3$, 400 MHz) δ (ppm): 10.37 (1 H, s, N-C<u>*H*</u>-N), 7.57 (1 H, s, NC<u>*H*</u>CHN), 7.41 (1 H, s, NCHC<u>*H*</u>N), 4.32 (2 H, t, *J* 7.4, N-C<u>*H*</u>$_2$CH$_2$), 4.14 (3 H, d, *J* 2.3, NC<u>*H*</u>$_3$), 1.92 (2 H, p, *J* 7.3, NCH$_2$C<u>*H*</u>$_2$ C$_5$H$_{10}$CH$_3$), 1.42-1.16 (10 H, m, NCH$_2$CH$_2$C$_5$<u>*H*</u>$_{10}$CH$_3$), 0.87 (3 H, dd, *J* 7.7, 5.3, NCH$_2$CH$_2$C$_5$H$_{10}$C<u>*H*</u>$_3$).^{13}C NMR (CDCl$_3$, 100 MHz) δ (ppm): 136.50, 123.69, 121.91, 49.80, 36.50, 31.41, 30.01, 28.77, 25.97, 22.30, 17.97, 13.91. LC-MS: 195.2 (M+-Br).

1-Octyl-3-methylimidazolium hydroxide [OMIM]OH, Dark yellow liquid. ^{1}H NMR (CDCl$_3$, 400 MHz) δ (ppm): 10.17 (1 H, d, *J* 2.7, N-C<u>*H*</u>-N), 7.62 (1 H, s, NC<u>*H*</u>CHN), 7.45 (1 H, s, NCHC<u>*H*</u>N), 4.27 (2 H, t, *J* 7.4, N-C<u>*H*</u>$_2$CH$_2$), 4.08 (3 H, s, NC<u>*H*</u>$_3$), 1.86 (2 H, p, *J* 7.4, NCH$_2$C<u>*H*</u>$_2$ C$_5$H$_{10}$CH$_3$), 1.34-1.12 (11 H, m, NCH$_2$CH$_2$C$_5$<u>*H*</u>$_{10}$CH$_3$), 0.81 (3 H, t, *J* 6.7, NCH$_2$CH$_2$C$_5$H$_{10}$C<u>*H*</u>$_3$). ^{13}C NMR (CDCl$_3$, 100 MHz) δ (ppm): 135.50, 122.76, 121.17, 48.72, 35.47, 30.36, 29.07, 27.72, 27.66, 24.91, 21.26, 12.82. MS (ESI): 196.20 (M+-OH), 182.27 (M+-OH-CH$_3$), 167.80 (M+-OH-C$_2$H$_5$), 153.13 (M+-OH-C$_3$H$_7$).

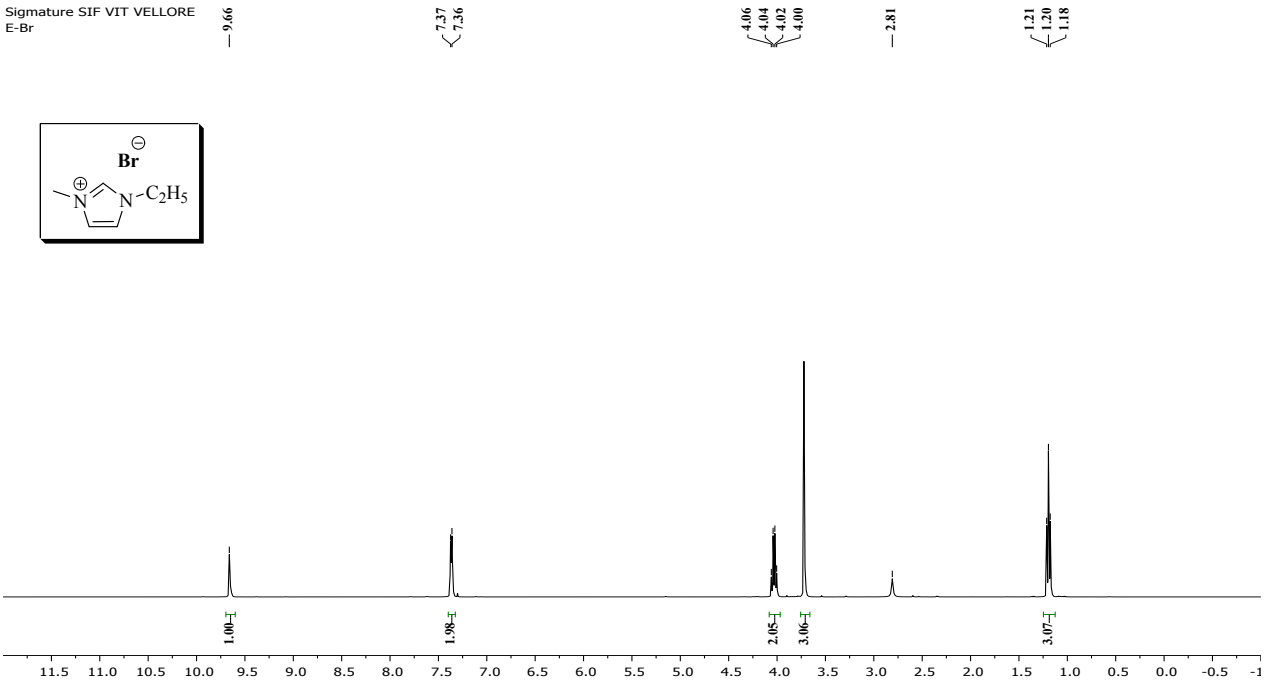

Fig. S1. ^{1}H NMR spectrum of 1-ethyl-3-methylimidazolium bromide.

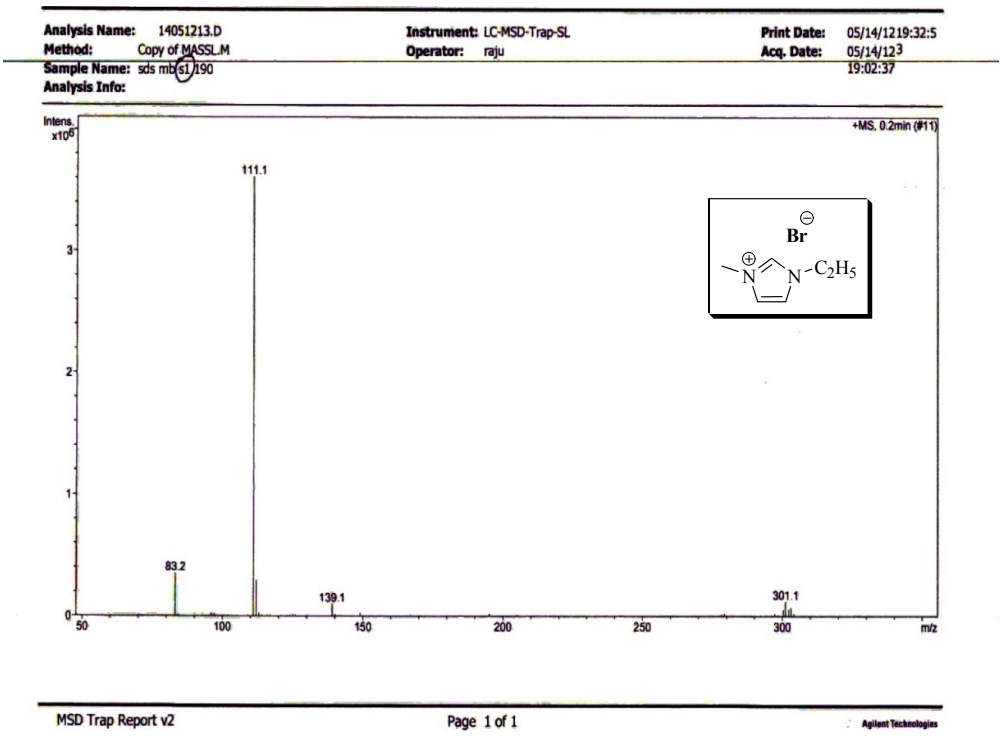

Fig. S2. Mass spectrum of 1-ethyl-3-methylimidazolium bromide.

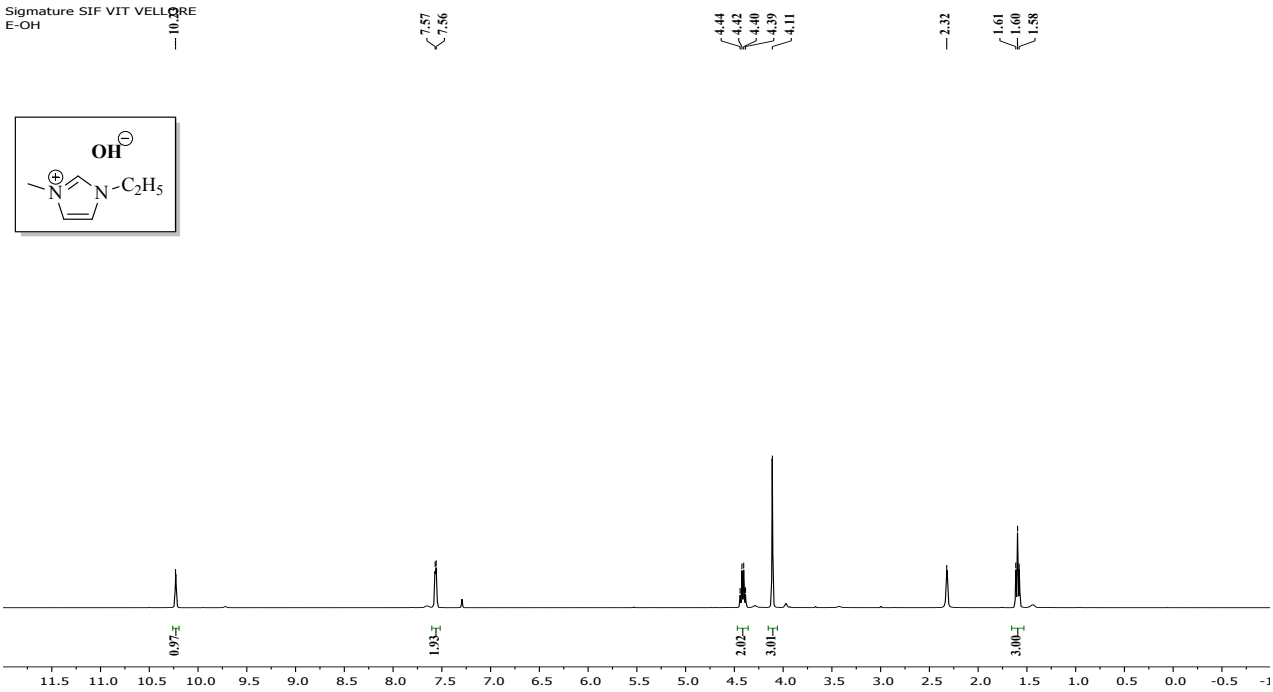

Fig. S3. ^{1}H NMR spectrum of 1-ethyl-3-methylimidazolium hydroxide.

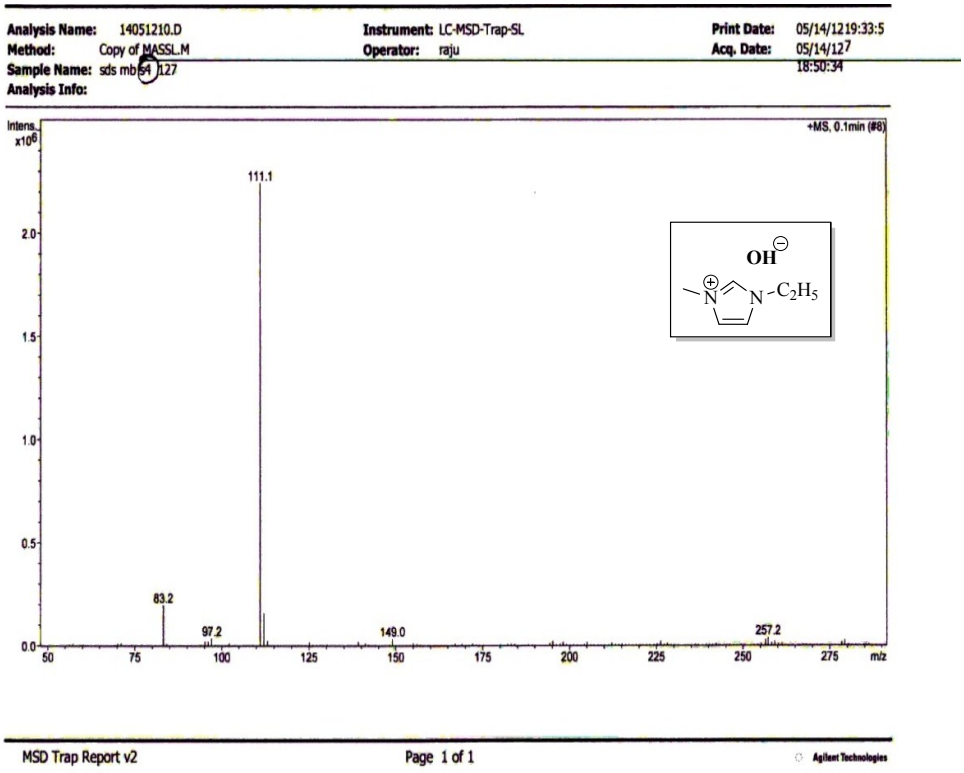

Fig. S4. Mass spectrum of 1-ethyl-3-methylimidazolium hydroxide.

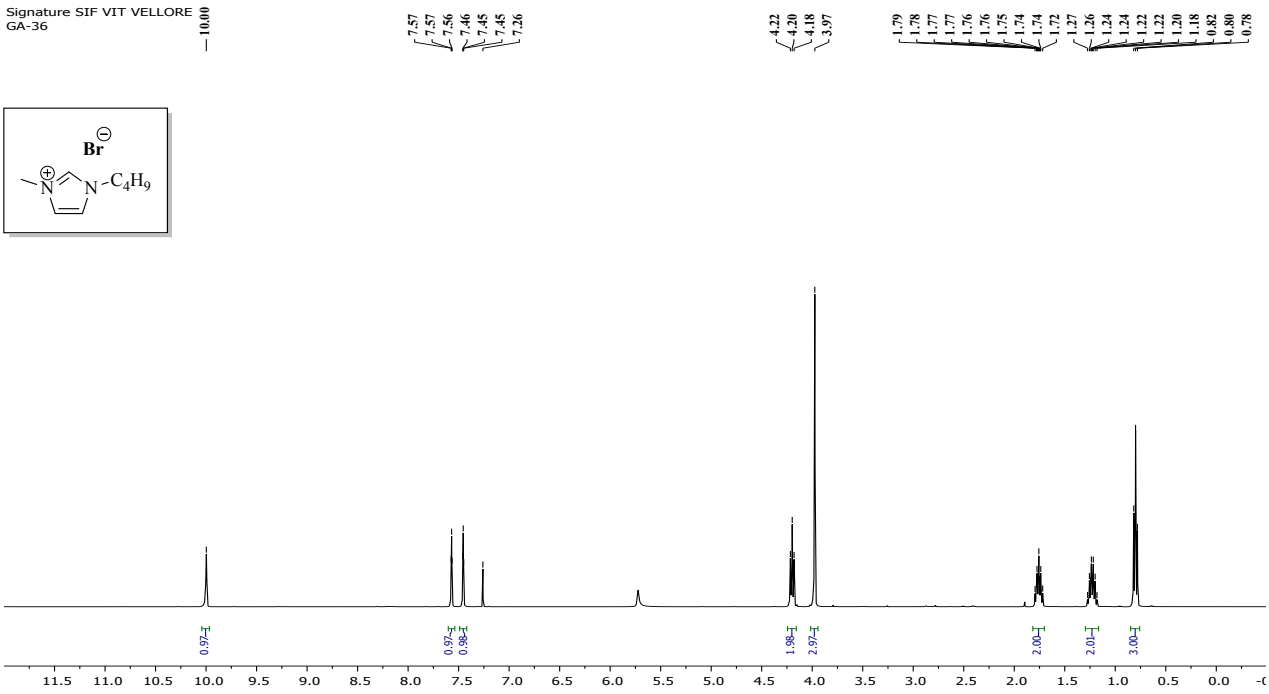

Fig. S5. ^{1}H NMR spectrum of 1-butyl-3-methylimidazolium bromide.

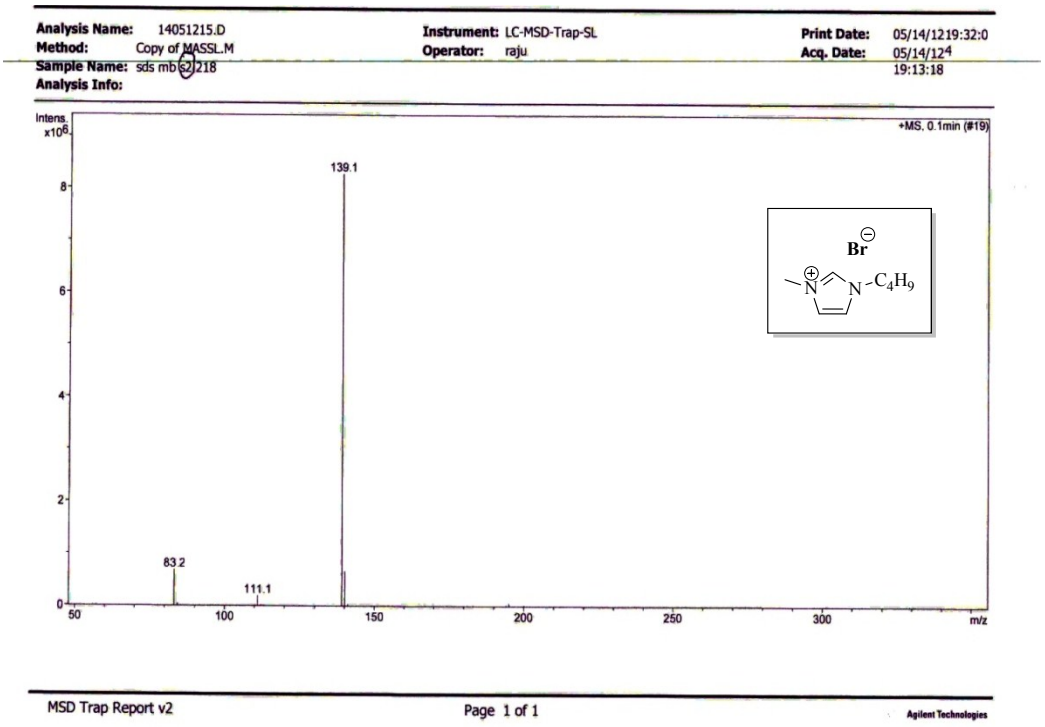

Fig. S6. Mass spectrum of 1-butyl-3-methylimidazolium bromide.

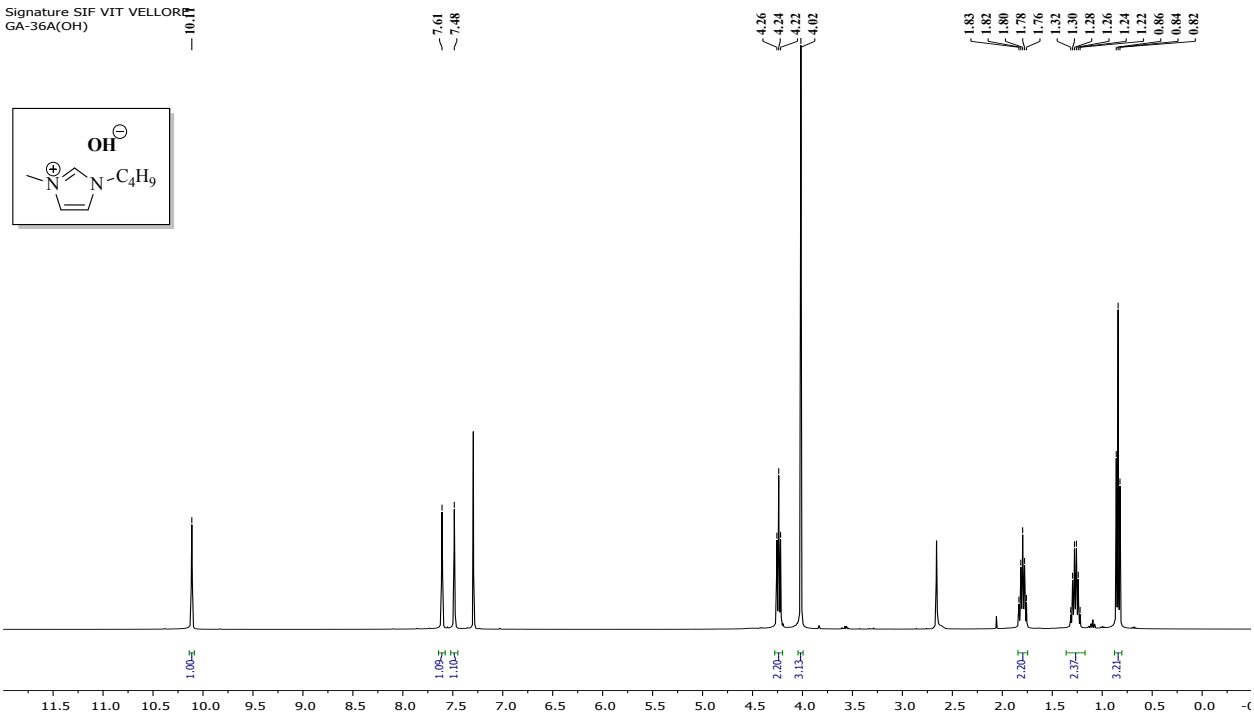

Fig. S7. ^{1}H NMR spectrum of 1-butyl-3-methylimidazolium hydroxide.

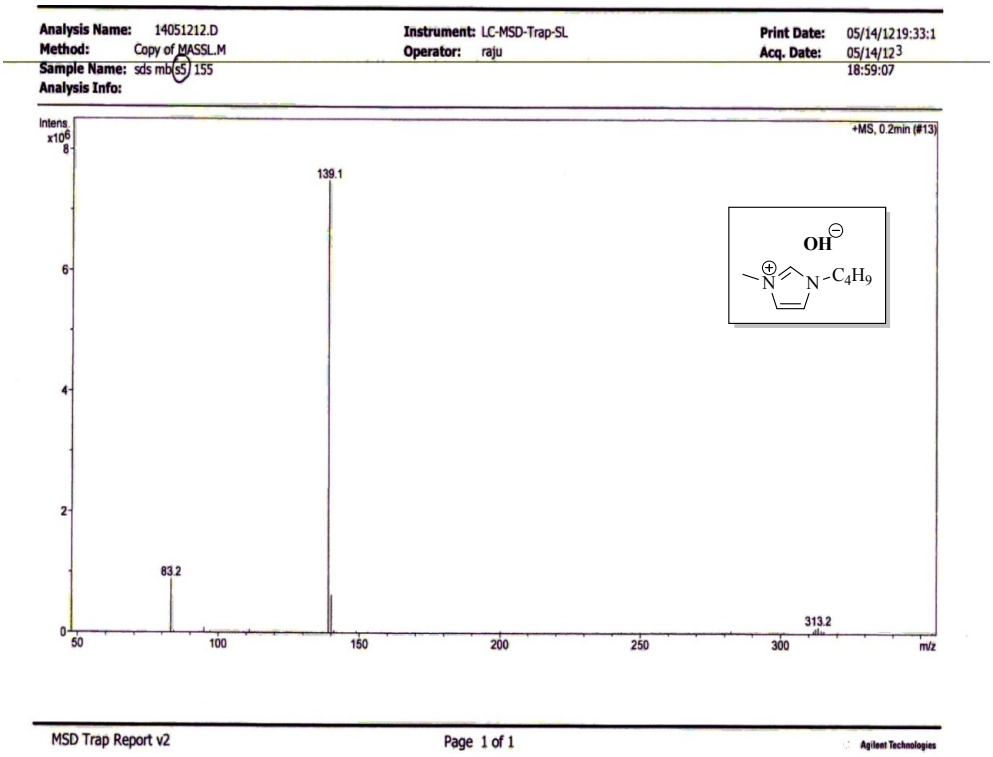

Fig. S8. Mass spectrum of 1-butyl-3-methylimidazolium hydroxide.

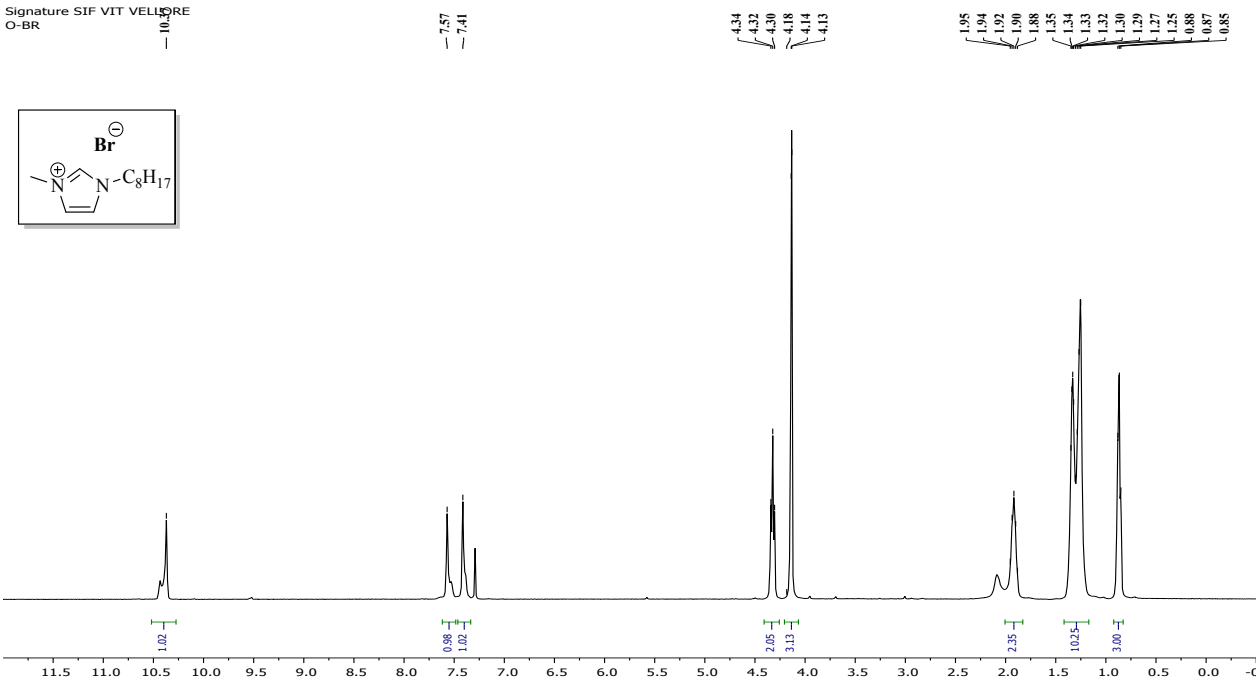

Fig. S9. ^{1}H NMR spectrum of 1-octyl-3-methylimidazolium bromide.

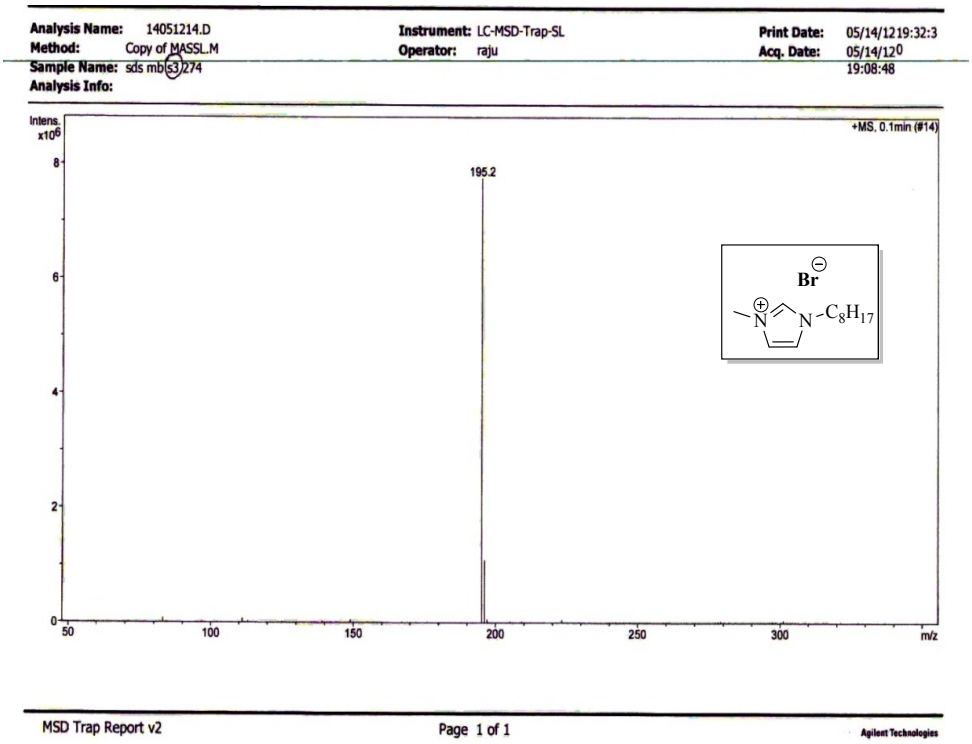

Fig. S10. Mass spectrum of 1-octyl-3-methylimidazolium bromide.

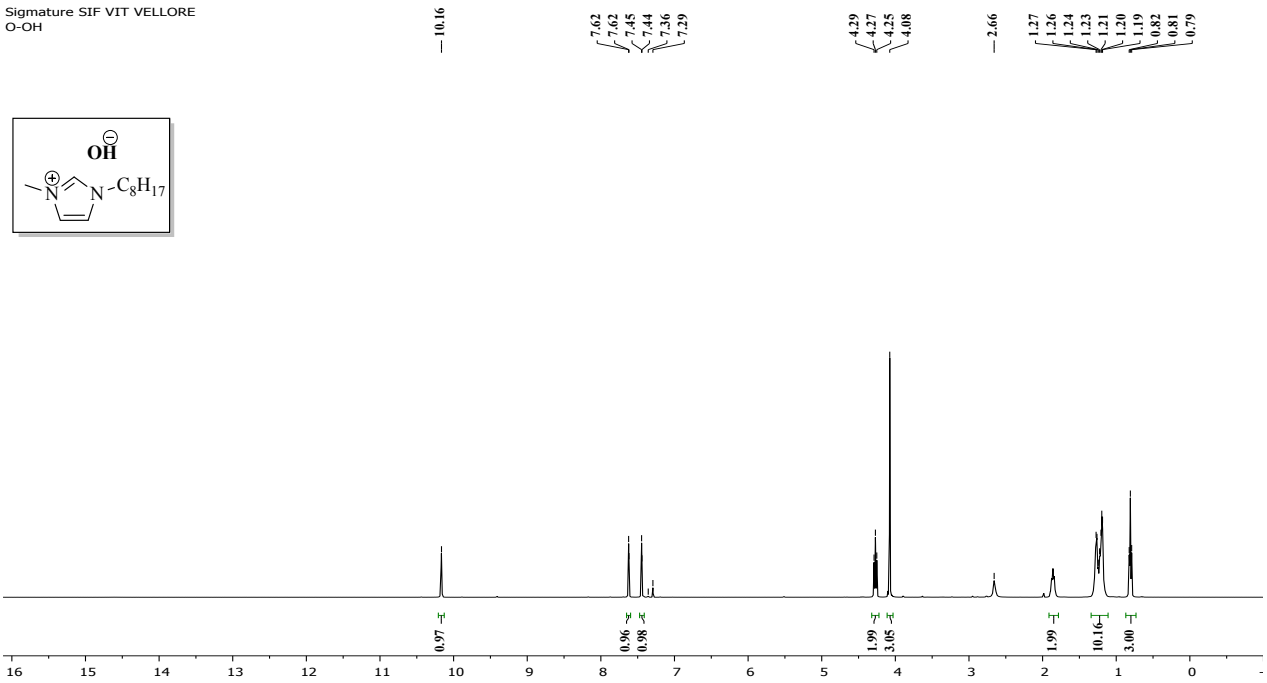

Fig. S11. ^{1}H NMR spectrum of 1-octyl-3-methylimidazolium hydroxide.

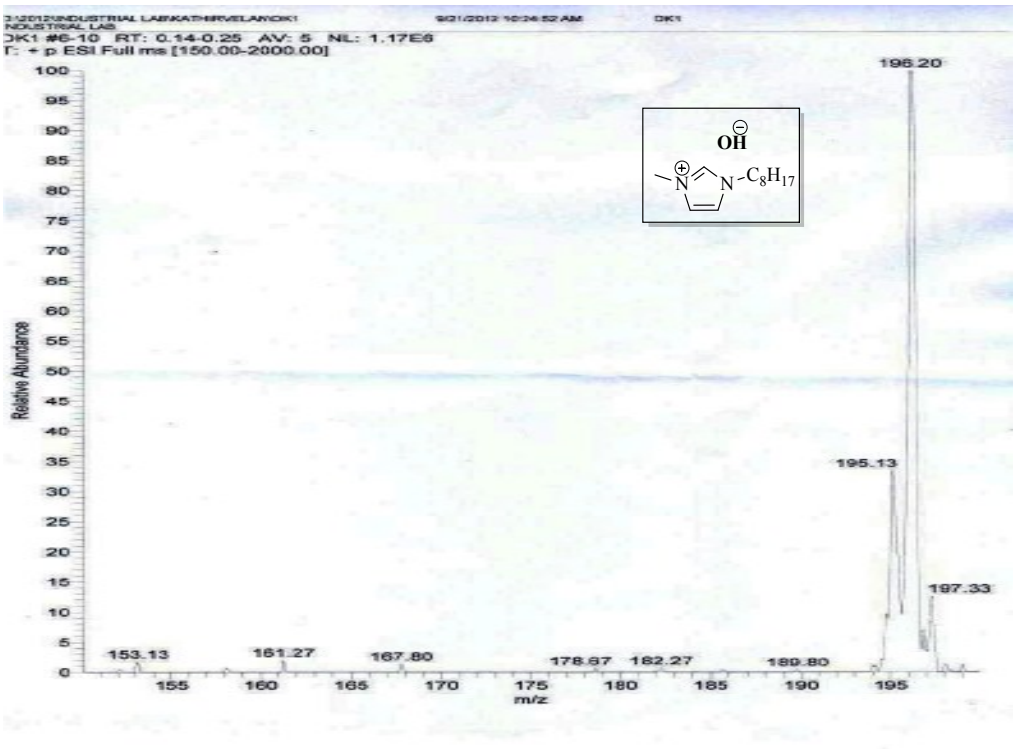

Fig. S12. Mass spectrum of 1-octyl-3-methylimidazolium hydroxide.

Table S1.

Percentage of paralysis and percentage of mortality of earthworms against six different ionic liquids and Albendazole at different time intervals.

S. No.	ILs or Drug	Time (min)	% Paralysis of earthworms at different concentration of ILs												% Mortality of earthworms at different concentration of ILs											
			2 mM			4 mM			8 mM			16 mM			2 mM			4 mM			8 mM			16 mM		
1.	[EMIM] Br	15	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
		30	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
		45	0	0	0	0	0	0	20	0	0	20	20	20	0	0	0	0	0	0	0	0	0	0	0	0	0
		60	0	0	0	0	0	0	20	20	0	40	40	40	0	0	0	0	0	0	0	0	0	0	0	0	0
		75	0	0	0	0	0	0	20	20	20	40	40	40	0	0	0	0	0	0	0	0	0	0	0	0	0
		90	0	0	0	0	0	0	20	20	20	40	80	40	0	0	0	0	0	0	0	0	20	0	0	20
		120	20	20	20	20	20	20	40	40	40	60	60	60	0	0	0	0	0	0	20	20	20	40	20	40
2.	[EMIM] OH	15	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
		30	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
		45	0	0	0	0	0	0	0	0	0	20	20	20	0	0	0	0	0	0	0	0	0	0	0	0	0
		60	20	20	20	20	20	20	20	20	20	40	40	40	0	0	0	0	0	0	0	0	0	0	0	0	0
		75	20	20	20	20	20	20	20	20	20	40	40	40	0	0	0	0	0	0	0	0	0	0	0	0	0
		90	20	20	20	40	40	40	40	40	40	60	60	60	0	0	0	40	40	40	40	40	40	40	40	40	40
		120	40	40	60	60	60	60	60	60	60	60	60	60	0	0	0	40	40	40	40	40	40	40	40	40	40

3.	**[BMIM] Br**	**15**	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
		30	0	0	0	0	0	0	20	20	0	20	20	0	0	0	0	0	0	0	0	0	0	0	0	0
		45	0	0	0	0	0	0	40	40	0	40	40	0	0	0	0	0	0	0	0	0	0	0	0	0
		60	0	0	0	20	40	40	40	40	20	40	40	40	0	0	0	0	0	0	0	0	0	0	0	0
		75	0	0	20	60	60	60	60	60	60	60	60	60	0	0	0	0	0	0	0	0	0	0	0	0
		90	60	40	20	100	100	100	100	100	100	100	100	100	0	0	0	20	20	20	20	20	20	20	20	20
		120	100	100	100	100	100	100	100	100	100	100	100	100	0	0	0	20	20	20	40	40	40	60	60	60
4.	**[BMIM] OH**	**15**	0	0	0	0	0	0	0	0	0	20	20	20	0	0	0	0	0	0	0	0	0	0	0	0
		30	0	0	0	0	0	0	0	0	0	20	20	20	0	0	0	0	0	0	0	0	0	0	0	0
		45	0	0	0	20	20	20	20	20	20	40	40	40	0	0	0	0	0	0	0	0	0	0	0	0
		60	20	20	20	20	20	20	40	40	40	60	60	60	0	0	0	0	0	0	0	0	0	0	0	0
		75	60	40	40	40	40	40	60	60	60	100	100	100	0	0	0	0	0	0	0	0	0	20	20	20
		90	60	60	60	80	80	80	80	80	80	100	100	100	0	0	0	20	20	20	20	20	20	40	40	40
		120	80	80	80	80	80	80	100	100	100	100	100	100	20	20	20	20	20	20	20	20	20	40	40	40
5.	**[OMIM] Br**	**15**	0	0	0	0	0	0	20	20	20	40	40	40	0	0	0	0	0	0	0	0	0	0	0	0
		30	0	0	0	20	0	0	20	20	20	100	100	100	0	0	0	0	0	0	0	0	0	0	0	0
		45	0	0	0	40	20	20	100	100	100	100	100	100	0	0	0	0	0	0	0	0	0	60	60	60
		60	40	40	40	40	40	40	100	100	100	100	100	100	0	0	0	0	0	0	40	80	80	100	100	100
		75	60	60	60	60	60	60	100	100	100	100	100	100	0	0	0	0	0	0	60	100	80	100	100	100
		90	100	100	100	100	100	100	100	100	100	100	100	100	20	20	20	20	20	20	100	100	100	100	100	100

		120	100	100	100	100	100	100	100	100	100	100	100	100	40	40	40	60	60	60	100	100	100	100	100	100
6.	**[OMIM] OH**	**15**	20	20	20	20	20	20	40	40	40	60	60	60	0	0	0	0	0	0	0	0	0	0	0	0
		30	40	40	40	80	80	80	40	40	40	100	100	100	0	0	0	20	20	20	20	20	20	80	100	100
		45	100	100	100	100	100	100	100	100	100	100	100	100	40	40	40	40	40	40	80	80	80	100	100	100
		60	100	100	100	100	100	100	100	100	100	100	100	100	80	60	80	80	80	80	100	100	100	100	100	100
		75	100	100	100	100	100	100	100	100	100	100	100	100	100	80	80	80	80	80	100	100	100	100	100	100
		90	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
		120	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100	100
7.	**Albendazole**	**15**	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
		30	0	0	0	0	0	0	0	0	0	60	60	60	0	0	0	0	0	0	0	0	0	0	0	0
		45	0	0	0	0	0	0	20	20	20	80	80	80	0	0	0	0	0	0	0	0	0	0	0	0
		60	0	0	0	20	20	20	40	40	20	80	80	80	0	0	0	0	0	0	0	0	0	0	0	0
		75	0	0	20	40	40	40	40	40	40	80	80	80	0	0	0	0	0	0	0	0	0	0	0	0
		90	20	20	20	60	60	60	80	80	80	100	100	100	0	0	0	20	20	20	20	20	20	20	20	20
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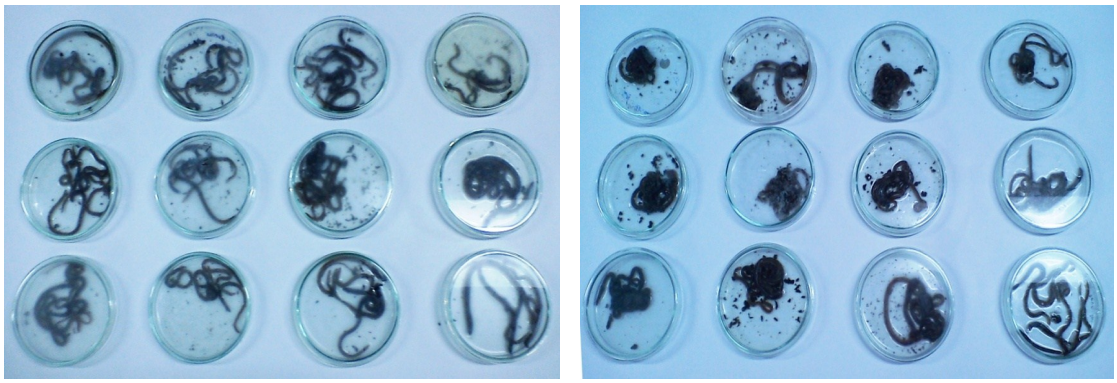

Fig. S14. The state of earthworms on treatment with ionic liquids.

Molecular Docking Protocol:

The program Glide (Grid-Based Ligand Docking with Energetics) incorporated in the Schrödinger molecular modeling package (Schrödinger, Inc., USA, *version- 2014-14*) installed on a Windows workstation with an Intel (R) xenon 2.8 GHz processor and 32 GB physical memory was used to perform the molecular docking study.

The protein structure was cleaned and optimized using the *Protein Preparation Wizard*. The crystallographic water molecules were removed as no water molecule was found to be conserved and hydrogen atoms were added to the protein structure corresponding to pH 7.0 considering the appropriate ionization states for both the acidic and basic amino acid residue. Following the assignment of charge and protonation state, energy minimization with root mean square deviation (RMSD) value of 0.30Å was carried out using the Optimized Potentials for Liquid Simulations (OPLS-2005) force field. On the other hand, the 3D geometries of the IL structures were optimized using LigPrep module and the partial charges were ascribed using the OPLS-2005 force-field. The binding pocket of β-tubulin was defined by a 14×14×14Å box that was centered on the geometric centroid of the co-crystallized Albendazole structure in the PDB complex. Flexibility of the IL as well as the β-tubulin was considered during docking, allowing for a flip of 5- and 6-membered rings. Extra-precision scoring function in Glide was used to rank the docking poses and to measure the relative binding affinities of the ILs to β-tubulin.