DOI: 10.1002/cphc.201000645

DNA Molecular Solvation in Neat Ionic Liquids

Lena Cardoso and Nuno M. Micaelo*[a]

The structural and molecular mechanism of double-stranded DNA (dsDNA) and single-stranded DNA (ssDNA) solvation in ten room-temperature ionic liquids (RTILs) is disclosed herein. We show that the Drew Dickerson B DNA structure solvated in neat RTILs based on imidazolium, oxazolium, pyrrolidinium, pyrimidinium and quaternary ammonium cations, combined with hexafluorophosphate and tetrafluorobromide anions (Figure 1), is able to maintain the native dsDNA B form conformation. We show that cations are preferentially located near the DNA main chain, establishing strong electrostatic interactions with the phosphate groups. On the other hand, anions are predominantly involved in hydrogen-bond interactions with cytosine, adenine and guanine bases. Furthermore, cations are also able to establish hydrogen-bonding and edge-to-face NH···π interactions with the DNA bases. We also show that ssDNA bases in RTILs are preferentially solvated by the anions via hydrogen bonding with the fluor atoms. Our study provides the first fundamental structural insights of DNA in RTILs that can support the future development of novel RTILs, specific for nucleic acids solutes.

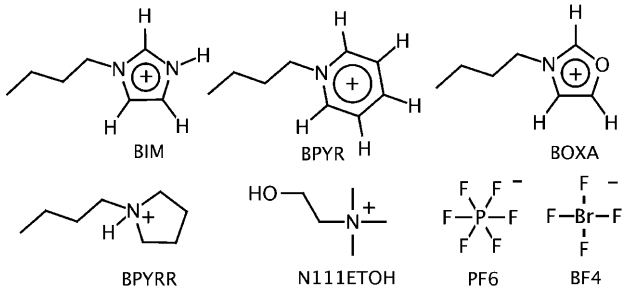

Figure 1. 2D structures[4] of the cations and anions used herein: 1-butyl-imidazolium ($[BIM]^+$), 1-butylpyridinium ($[BPYR]^+$), 1-butyl-oxazolium ($[BOXA]^+$), 1-butylpyrrolidinium ($[BPYRR]^+$), (2-hydroxyethyl)trimethylamonium ($[N111ETOH]^+$), hexafluorophosphate ($[PF_6]^-$) and tetrafluorobromide ($[BF_4]^-$).

RTILs are becoming ubiquitous in many chemical and biochemical processes,[1] and are replacing organic solvents traditionally used in non-aqueous enzymology[1c] and separation processes.[1a] Recently, the use of RTILs with DNA has also been investigated.[2] DNA separation using a capillary coated with imidazolium-based RTILs was reported,[2c] as well as the use of RTILs to extract trace amounts of DNA from aqueous solutions.[2e] Other applications of DNA in RTILs have also been reported, such as, long-term storage[2d] and DNA nanopore translocation.[2a]

The molecular mechanism of interaction between RTILs and DNA is still not clearly understood. Fundamental chemical and physical information about the interaction between a few RTILs and DNA have been reported.[3] The 31P NMR and IR spectra indicate that the molecular interaction between dsDNA and 1-butyl-3-methylimidazolium ($[BMIM]^+$) chloride is accomplished via strong electrostatic interactions between the cation head group of $[BMIM]^+$ with the phosphate groups of DNA,[2f,3] and hydrophobic association between the aliphatic chain of $[BMIM]^+$ with the DNA bases.[3] Furthermore, DNA in [BMIM]Cl maintains its B form, has shown by the similar circular dichroism (CD) spectra of DNA in this RTIL and in tris(hydroxymethyl)aminomethane hydrochloride (Tris-HCl). The same behaviour was also observed using choline-based RTILs.[2d] So far, we have some direct and indirect evidence of the interaction of the cation RTILs with DNA and no clear role for the anionic species in the solvation mechanism of DNA.

Our modeling and molecular dynamics simulation studies show that the DNA structure is highly stabilized by the RTILs, in agreement with the Vijayaraghavan et al. evidence of the long-term DNA stabilization by [BMIM]Cl.[2d] The root mean square deviation (RMSD) of the DNA structure solvated in neat RTILs is low (Table 1) in comparison to the same dsDNA and ssDNA structures in aqueous media (0.53 and 0.70 nm, respectively). dsDNA in RTILs have a RMSD in the range of 0.14 to 0.20 nm. These figures are significantly lower than the observed 0.53 nm deviation of dsDNA in water, and translate the fact that RTILs are able to retain the native B-form dsDNA crystallographic structure. The RTILs that perform best in retaining the native structure of dsDNA are the quaternary ammonium and the pyrimidine RTILs with $[PF_6]^-$. The change of anion from $[PF_6]^-$ to $[BF_4]^-$ seems to have a slight detrimental effect on the dsDNA structure. The dsDNA structure is insensitive to

Table 1. Average root mean square deviation [nm] of the dsDNA and ssDNA main-chain, relative to the X-ray structure, in the different RTILs.

	dsDNA		ssDNA	
	$[PF_6]^-$	$[BF_4]^-$	$[PF_6]^-$	$[BF_4]^-$
$[BOXA]^+$	0.20	0.19	0.18	0.11
$[BIM]^+$	0.18	0.18	0.17	0.17
$[BPYR]^+$	0.15	0.21	0.14	0.14
$[BPYRR]^+$	0.18	0.22	0.27	0.20
$[N111ETOH]^+$	0.14	0.17	0.20	0.16

[a] *L. Cardoso, Dr. N. M. Micaelo*
Chemistry Centre
Minho University
Campus Gualtar, 4710-057 Braga (Portugal)
Fax: (+351) 253 604 386
E-mail: micaelo@quimica.uminho.pt

Supporting information for this article is available on the WWW under http://dx.doi.org/10.1002/cphc.201000645.

the change of anion in imidazolium and oxazolium RTILs, however, in pyrrolidinium, pyridinium and quaternary ammonium cations, there is a slight increase of the dsDNA RMSD. In these RTILs, the $[BF_4]^-$ anions have higher tendency to establish, on average, more hydrogen bonds with the dsDNA bases than the $[PF_6]^-$ anion (Table 2).

Table 2. Average number of hydrogen bonds between the RTILs (cations and anions) and the DNA bases.[a]

	dsDNA		ssDNA	
	Anion	Cation	Anion	Cation
[BOXA][PF_6]	12.0	0.1	19.1	0.1
[BIM][PF_6]	12.1	5.7	19.0	4.6
[BPYR][PF_6]	11.5	0.2	15.6	0.0
[BPYRR][PF_6]	8.7	2.0	17.8	0.0
[N111ETOH][PF_6]	12.2	11.1	17.6	3.6
[BOXA][BF_4]	10.9	0.2	21.8	7.5
[BIM][BF_4]	10.0	1.8	21.1	0.0
[BPYR][BF_4]	14.9	0.0	17.3	0.1
[BPYRR][BF_4]	11.0	0.2	20.6	0.0
[N111ETOH][BF_4]	12.9	3.2	18.9	0.0

[a] Fluorine atoms were considered as hydrogen bond acceptors. A hydrogen bond is considered to exist in one conformation if the distance between the hydrogen atom and the acceptor is less than 0.35 nm, and the angle formed by acceptor-donor-hydrogen is less than 30°.

The RTILs also have the ability to retain the ssDNA structure close to its starting conformation (Table 1). ssDNA bases are more accessible to the solvent, in particular to the $[BF_4]^-$ anions. This anion induces lower structural changes on the ssDNA structure than $[PF_6]^-$ (Table 1). These facts are also correlated with the evidence that $[BF_4]^-$ anions are able to establish, on average, more hydrogen bonds with the ssDNA bases than the $[PF_6]^-$ anion (Table 2).

The radial distributions (RDF) of the cations show a preferential localization of the cation "head" charge group located at 0.5 nm from the DNA phosphate groups (Figure 2a) and a complete exclusion of anions from this region (Figure 2b). The same behavior is observed in the presence of the $[BF_4]^-$ anion (data not shown). These observations are in agreement with the experimental ^{31}P NMR and IR spectra studies that demonstrate the same type of cation DNA interaction.[2f,3] The quaternary ammonium cation (2-hydroxyethyl)trimethylamonium ($[N111EtOH]^+$) also exhibits a second peak at 0.7 nm (Figure 2a), corresponding to a conformation of the cation with the hydroxyl group in a hydrogen-bond interaction with an oxygen of the DNA phosphate groups. Apparently, Figure 2 suggests that the cation's RDF is insensitive to the type of cation or functional group. However, it is also clear that, if the cation is functionalized with other chemical groups with hydrogen donor/acceptor capability, such as the hydroxyl located at the end of the aliphatic chain of $[N111EtOH]^+$, this functional group can compete with the positively charged region of the cation for the DNA phosphate groups.

Cations also seem to interact, not only with the DNA main chain, but also with the DNA bases. This fact has been partially described with experimental techniques, but not yet fully understood.[3] All cations studied have hydrogen-bond donor and/or acceptor capabilities (Figure 1). 1-butyl-imidazolium ($[BIM]^+$) is seen to establish hydrogen bonds with the oxygen and nitrogen acceptor atoms of guanine in Figure 3a. Figure 3b shows the hydrogen bonding mechanism of the hydroxyl group of $[N111ETOH]^+$ to function, simultaneously, as a hydrogen-bond donor and acceptor with an adenine. Furthermore, the $[BPYRR]^+$ cation can act as a hydrogen donor group towards guanine bases as shown in Figure 3c. These examples clearly show that cations can participate in the solvation mechanism of DNA bases trough hydrogen bonding interactions, which have not yet been described experimentally.

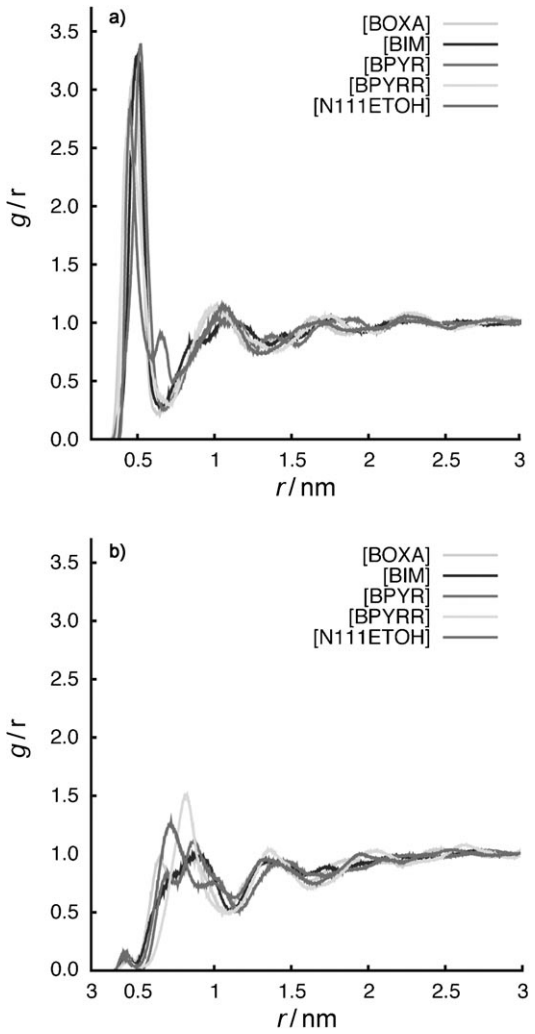

Figure 2. a) RTIL cations' (head charge group) radial distribution function around dsDNA main-chain phosphate groups. b) RTIL anions' radial distribution function around dsDNA main-chain phosphate groups. Seethe Supporting Information for details about radial distribution calculations.

We have also identified the occurrence of another type of nonbonded interaction known as edge-to-face NH···π.[5] This type of interaction is found between the hydrogen of amino groups of adenine and guanine bases facing the aromatic ring systems of oxazolium, imidzolium and pyrimidium cations (Figures 3d–f respectively).

Figures 3 g–i illustrates two common interactions between $[PF_6]^-$ and DNA bases. Figure 3 g shows the simultaneous in-

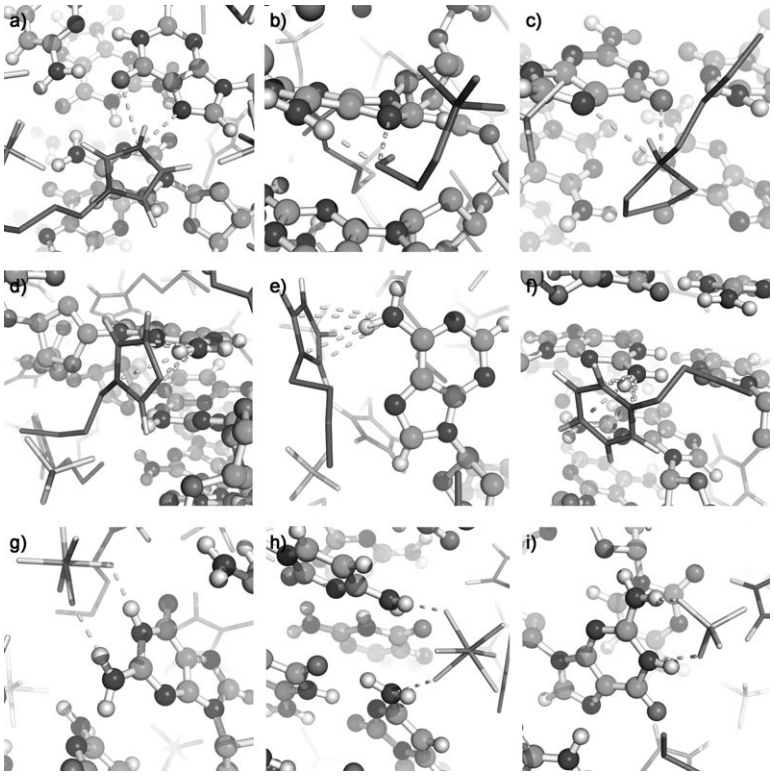

Figure 3. Figure composite of cation and anion non-bonded interactions with DNA bases. Hydrogen bonds in (a)–(c) and (g)–(i) are rendered in dashed lines. In (d)–(f), dashed lines connect the aromatic ring atoms with the NH hydrogen bond donor group in edge-to-face type interactions.

teraction of two fluor atoms of $[PF_6]^-$ with two hydrogen donors of a guanine. Figure 3 h shows the simultaneous interaction of two fluorine atoms with hydrogen donors from two close cytosines. Furthermore, $[BF_4]^-$ anions also have similar hydrogen bonds arrangements with guanine bases (Figure 3 i).

RTILs provide a molecular environment for biomolecular systems distinct from common molecular solvents such as water. Our observations are quite revealing about the mechanism of DNA solvation and stabilization by RTIL cations and anions. We stress the important role of anions and the hydrogen-bonding interactions of the cations with the DNA bases. Given the plethora of RTIL cations and anions available, it is possible that, with the structural insights provided herein, novel RTILs could be designed with tailor-made features specific for nucleic acids solutes.

Experimental Section

The RTILs were modeled using a similar parameterization approach to that used by us previously.[6] Details of the RTIL parameterization and validation are described in the Supporting Information. The Drew Dickerson B DNA structure with protein databank code 1BNA[7] was used herein. Both dsDNA and ssDNA structure were investigated, the latter structure was obtained by using only the chain A of the DNA molecule. The DNA was solvated in different RTIL systems and in a 100 mM aqueous system. Molecular mechanics/dynamics (MM/MD) simulations were performed with the GROMOS96 53 A6 biomolecular force field[8] and the GROMACS[9] simulation package. MD simulations were performed for 20 ns for each system, and structural analysis was done from the last 10 ns. Full details of the MM/MD simulation details and analysis are described in the Supporting Information.

Acknowledgements

The authors would like to acknowledge the access to the Minho University GRIUM cluster. Nuno M. Micaelo acknowledges the contract research program "Compromisso com a Ciência" reference: C2008-UMINHO-CQ-03.

Keywords: DNA · ionic liquid · molecular dynamics · molecular modeling · solvent effects

[1] a) A. Berthod, M. J. Ruiz-Ángel, S. Carda-Broch, *J. Chromatogr. A* **2008**, *1184*, 6; b) V. I. Pârvulescu, C. Hardacre, *Chem. Rev.* **2007**, *107*, 2615–2665; c) M. Sureshkumar, C. K. Lee, *J. Mol. Catal. B* **2009**, *60*, 1.

[2] a) R. S. S. de Zoysa, D. A. Jayawardhana, Q. T. Zhao, D. Q. Wang, D. W. Armstrong, X. Y. Guan, *J. Phys. Chem. B* **2009**, *113*, 13332; b) N. Nishimura, Y. Nomura, N. Nakamura, H. Ohno, *Biomaterials* **2005**, *26*, 5558; c) W. D. Qin, S. F. Y. Li, *Analyst* **2003**, *128*, 37; d) R. Vijayaraghavan, A. Izgorodin, V. Ganesh, M. Surianarayanan, D. R. MacFarlane, *Angew. Chem.* **2010**, *122*, 1675–1677; *Angew. Chem. Int. Ed.* **2010**, *49*, 1631–1633; e) J. Wang, D. Cheng, X. Chen, Z. Du, Z. Fang, *Anal. Chem.* **2007**, *79*, 620; f) Y. Xie, S. Wang, Z. Zhang, D. Pang, *J. Phys. Chem. B* **2008**, *112*, 9864; g) Y. Zhang, X. Chen, J. Lan, J. You, L. Chen, *Chem. Biol. Drug Des.* **2009**, *74*, 282.

[3] Y. H. Ding, L. Zhang, J. Xie, R. Guo, *J. Phys. Chem. B* **2010**, *114*, 2033.

[4] The cations were modelled using a united-atom description for the CH_2 and CH_3 groups of the aliphatic tails and for the hydrogens of the non-aromatic ring system of $[BPYRR]^+$. Marvin 5.2.6 ChemAxon was used for chemical drawing.

[5] a) S. Tsuzuki, K. Honda, T. Uchimaru, M. Mikami, K. Tanabe, *J. Am. Chem. Soc.* **2000**, *122*, 11450; b) R. S. Prajapati, M. Sirajuddin, V. Durani, S. Sreeramulu, R. Varadarajan, *Biochemistry* **2006**, *45*, 15000.

[6] N. M. Micaelo, A. M. Baptista, C. M. Soares, *J. Phys. Chem. B* **2006**, *110*, 14444.

[7] H. R. Drew, R. M. Wing, T. Takano, C. Broka, S. Tanaka, K. Itakura, R. E. Dickerson, *P. Natl. Acad. Sci. USA* **1981**, *78*, 2179.

[8] C. Oostenbrink, A. Villa, A. E. Mark, W. F. Van Gunsteren, *J. Comput. Chem.* **2004**, *25*, 1656.

[9] B. Hess, C. Kutzner, D. van der Spoel, E. Lindahl, *J. Chem. Theory Comput.* **2008**, *4*, 435.

Received: August 10, 2010
Revised: November 30, 2010
Published online on December 22, 2010

