Why Is Water So Special?

Lipid Vesicle Formation in Ionic Liquids and Deep Eutectic Solvents

Saffron J. Bryant[1], Kathleen Wood[2], Rob Atkin[3], Gregory G. Warr[1]

[1]School of Chemistry F11, The University of Sydney, NSW 2006 Australia. [2]Bragg Institute, ANSTO, NSW 2232 Australia. [3]The University of Newcastle, Callaghan, NSW 2308 Australia

The search for extra-terrestrial life is often restricted to the 'Goldilocks Zone;' that is, to planetary objects with environments that support liquid water. This is because of the importance of water to life as we know it on Earth, however, using terrestrial limitations to search for extra-terrestrial life is short-sighted. Through evolution, life as we know it has come to rely on water, but is it possible for all the conditions for life to originate in a water-free environment?

Compartmentalisation is one of the features of prebiotic environments believed to be essential to the origin of life. Compartments create sequestered units that can undergo natural selection and evolution, and concentration of substrates. One of the simplest prebiotic compartments are vesicles which share a similar membrane structure to modern day cells, and can self assemble from simple lipids (Figure 1). In this research compartmentalisation is used as a model for the possibility of life.

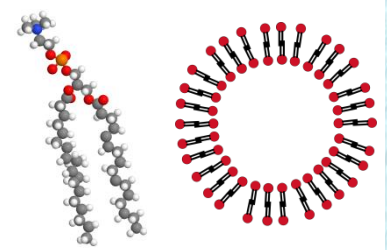

Figure 1. Phosphatidylcholine lipid and a vesicle.

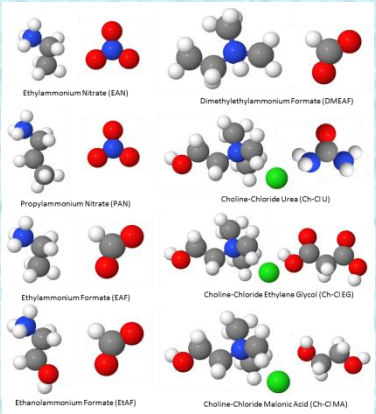

Figure 2. Solvent structures.

In the search for a non-aqueous environment that supports spontaneous self-assembly we must consider the relevant properties of water and then find other solvents that also possess these essential features. Hydrophobic interactions, mediated by the extended three-dimensional hydrogen-bond network of water, are the main driver for the self-assembly of lipids in water, so we focused on polar solvents that would similarly repel the non-polar tails of the lipids.

Obvious molecular hydrogen-bonding alternatives to water, such as glycerol and ethylene glycol, are liquid over temperatures similar to water. To explore the possibility of a drastically expanded Goldilocks Zone, we seek more exotic solvents with extreme melting points and very broad liquid stability ranges, including molten salts or *ionic liquids*.

Surfactants are known to self-assemble into micelles in ionic liquids, making them ideal prospects for this investigation.[1,2] In addition, ionic liquids can be specifically tuned for certain properties, and many exhibit extensive hydrogen bond networks akin to those in water.

Deep eutectic solvents (DES) are mixtures similar to ionic liquids that are also expected to repel hydrophobic lipid tails in the same manner as water.

These liquids could thus comprise a completely unexplored and truly alien prebiotic environment.

Terrestrial phosphatidylcholine (PC) lipids are good models for spontaneous vesicle self-assembly (Figure 1). The lipids tested have non-polar tails comprising 14 (DMPC), 16 (DPPC) and 18 (DSPC) carbons, and were compared with naturally-extracted egg PC.

Polarizing Optical Microscopy was used to examine how the various lipids are swollen[3] by contact with each solvent. In water, DESs, and formate ionic liquids, spontaneous vesicle formation was observed for all lipids. In nitrate ILs (EAN and PAN) spontaneous vesicle formation was only observed for eggPC.

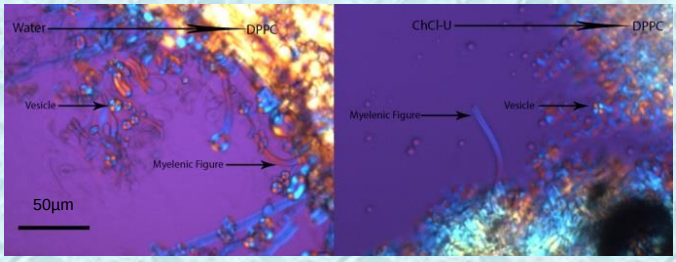

Figure 3. Concentration gradient experiments of DPPC with water and choline-chloride urea demonstrated lamellar phase formation with vesicles and myelenic figures as indicated

Small-Angle Neutron Scattering (SANS) determines structure in solutions in the form of intensity against scattering angle (expressed as scattering vector $q = (4\pi/\lambda)\sin(\theta/2)$) by comparison against known models.[5]

The SANS results confirmed the microscopy observations, and demonstrated that vesicles formed in all but two lipid/IL combinations. Both unilamellar and multilamellar vesicles were formed (see Figure 4), all leading to self-assembled compartments.

Only Egg PC and DMPC in PAN did not form vesicles. This may be because PAN has the longest hydrocarbon chain of all the ionic liquids tested and therefore has the smallest solvophobic effect. This highlights the importance of solvophobicity to self-assembled vesicles and will help identify other potential solvents.

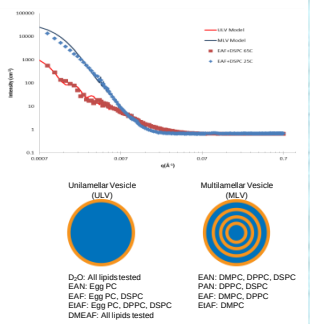

Figure 4. Example SANS data with representations of unilamellar and multilamellar vesicles.

DMEAF, which cannot form H-bond networks, also supported vesicles with all lipids examined. This suggests that hydrogen bonding is not a requirement of vesicle self-assembly and that electrostatic interactions are far more important, raising the prospect of prebiotic compartmentalisation in molten salts at extreme temperatures.

Conclusions and Prospects

The formation of vesicles by lipids has been shown to occur in a diverse range of ILs and DESs.

Compartmentalisation, one of the basic requirements for life, is thus possible in extreme, non-aqueous environments and therefore the search for extra-terrestrial life should not be bound by terrestrial limitations and preconceptions. It highlights the fact that water is not as unique to fundamental processes as previously thought and therefore should not be the sole priority in the search for extraterrestrial life.

The answer to the question 'Why is water so special?' appears to be; 'It's not.'

References

1. Atkin, R.; Bobillier, S. M. C.; Warr, G. G., Propylammonium nitrate as a solvent for amphiphile self-assembly into micelles, lyotropic liquid crystals, and microemulsions. The Journal of Physical Chemistry B 2009, 114 (3), 1350-1360.
2. Greaves, T. L.; Drummond, C. J., Ionic liquids as amphiphile self-assembly media. Chemical Society Reviews 2008, 37 (8), 1709-1726.
3.Carlton, R. (2011). Polarized Light Microscopy. Pharmaceutical Microscopy. Springer New York: 7-64.
4. Maitani, Y. (2010). Lipoplex Formation Using Liposomes Prepared by Ethanol Injection. Liposomes. V. Weissig, Humana Press. **605:** 393-403.
5. Kiselev, M. A.; Zemlyanaya, E. V.; Aswal, V. K.; Neubert, R. H. H., What can we learn about the lipid vesicle structure from the small-angle neutron scattering experiment? Eur Biophys J 2006, 35 (6), 477-493.
6. Jiang, H. J.; FitzGerald, P. A.; Dolan, A.; Atkin, R.; Warr, G. G., Amphiphilic self-assembly of alkanols in protic ionic liquids. The Journal of Physical Chemistry B 2014, 118 (33), 9983-9990