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Analysis of Magnetite Nanoparticle Biomineralization Proteins from Magnetotactic Bacteria

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Extended Abstract

Magnetic nanoparticles (MNP) represent a growing area of research due to their useful and varied applications across a range of different settings. These particles can be used in biomedical diagnostics, innovative therapies, magnetic resonance imaging (Pankhurst et al., 2003), as well as in the areas of data storage and biotechnology. The magnetic and physical properties of MNP are inherently linked to their suitability in each application, with a narrow size distribution and uniform morphology being two important considerations. Conventional synthetic routes to MNP often produce a mixture of differently sized particles with a wide range of different shapes. By the addition of high temperature incubations or capping agents the homogeneity of the particles can be improved. However, these methods come at a high cost and use harsh reagents and conditions. A key goal in MNP research is to achieve similar or improved particle control with ambient, mild conditions.

Magnetotactic bacteria (MTB) represent a novel group of organisms that have evolved to produce chains of single crystals of magnetite enveloped within lipid vesicles termed magnetosomes. Interestingly, each species displays a narrow distribution of size and shape of the formed particles. Previous studies have revealed many different proteins are associated with the magnetosome (Grünberg, K. et al, 2004), and a small number of these have been shown to play a role in controlling the morphology of the magnetite particles *in vivo*. One protein in particular, Mms6 from *Magnetospirillum magneticum* AMB-1 has been shown to interact strongly with the magnetite crystal and when purified and introduced into synthetic magnetite precipitation reactions it can produce improvements in MNP homogeneity (Arakaki et al., 2003). This highlights the potential to mimic the highly uniform biogenic MNP which the bacteria produce under ambient conditions by harnessing the activity of biomineralization proteins in so called biokleptic experiments (Rawlings et al., 2012).

To begin to understand how these proteins function we are investigating a range of suspected biomineralization proteins from *Magnetospirillum magneticum* AMB-1, including Mms6 and others from the same or neighbouring gene clusters (Murat et al., 2012) or those with significant homology to proteins known to affect magnetite morphology *in vivo*. Analysis of the primary sequences shows that all of these target proteins are predicted to span the magnetosome membrane at least once, and others multiple times. We have transferred the genes encoding these protein targets to *Escherichia coli* to allow high level protein production and have established purification routes for many of these proteins. We have discovered that a common hallmark of these proteins is their capability to be produced in a soluble form; an unusual finding considering their predominantly hydrophobic character. Using a range of biophysical techniques the oligomerisation and structural characteristics of these proteins have been probed. The ability of the protein assemblies to affect synthetic magnetite precipitation reactions is also being investigated with the resulting particles analysed by TEM, VSM, and XMCD to determine size, shape, mineral type and magnetization changes in response to protein addition. The sequence similarities and protein properties will be compared across the range of identified magnetite biomineralization proteins and their implications for synthetic MNP production will be discussed.

References

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