Molecular Dimensions

memX biosciences

MemMagic® Bicelle Screen Kit MD1-81/MD1-82

MemMagic® Bicelle Screen Kit is based on the use of bicelles as an alternative method for the crystallization of membrane proteins in a lipidic environment.

MemMagic® Bicelle Screen kit provides four bicelle solutions of 40%, 35%, 30%, 25% DMPC:CHAPSO (2.8:1).

MD1- 81 is presented as 4 tubes of 100 µL.
MD1- 82 is presented as 4 tubes of 250 µL.

Features of MemMagic®:

- Easy-to-use kit.
- Handles like detergent.
- Behaves like lipid.
- Robotically compatible.
- Applications for:
  - Crystallography
  - NMR
  - Assay Development

Introduction

Membrane proteins can be readily reconstituted into bicelles (Figure 1) and are maintained in a native-like bilayer environment, which can be manipulated with almost the same ease as for detergent solublized membrane proteins, making them compatible with standard high-throughput screening methods. MemMagic® Bicelle Screen Kit provides four bicelle solutions of 40%, 35%, 30%, 25% DMPC:CHAPSO (2.8:1).

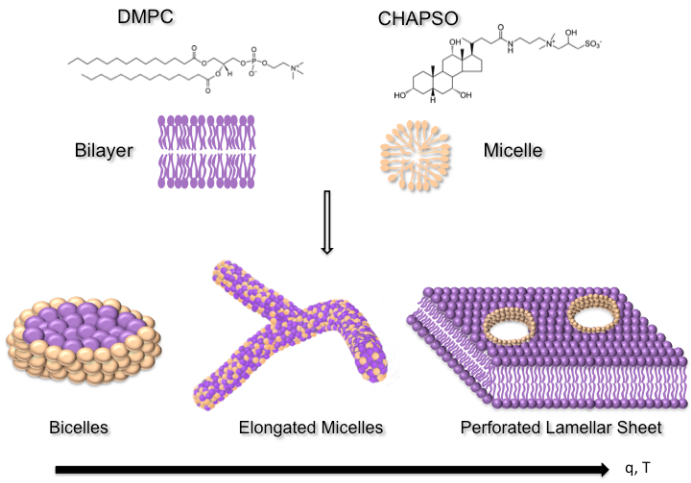
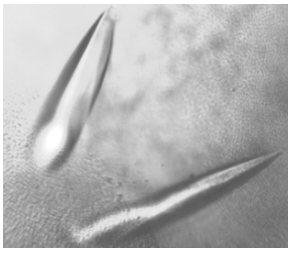


Figure 1. Phase behaviour of bicellar mixtures (Ujwal & Bowie (2011).

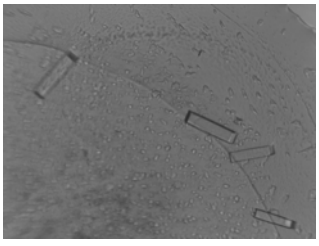
What are bicelles?

Bicelles are formed by a mixture of lipids and detergents and provide a native-like bilayer environment for membrane proteins. The bicelle discs can be described as patches of lipid bilayers with detergent molecules lining the apolar edges of each bilayer. Formed by the mixture of a phosphatidylcholine lipid such as Dimyristoyl-phosphatidylcholine (DMPC) and a detergent such as 3-[(3-cholamidopropyl) dimethylammonio]-2-hydroxy-1-propanesulfonate (CHAPSO), bicelles present a compromise between a rigid lipidic medium and an artificial detergent medium while offering beneficial aspects from both.

Recently, a significant number of membrane proteins have been successfully crystallized using the bicelle method, including Bacteriorhodopsin, β2 Adrenergic receptor/Fab, Voltage-Dependent Anion Channel and Xanthorhodopsin.



Crystals grown using MemMagic®



Materials provided (per kit)	Quantity
MD1-81	
40% DMPC:CHAPSO (2.8:1)	100 µL
35% DMPC:CHAPSO (2.8:1)	100 µL
30% DMPC:CHAPSO (2.8:1)	100 µL
25% DMPC:CHAPSO (2.8:1)	100 µL
MD1-82	
40% DMPC:CHAPSO (2.8:1)	250 µL
35% DMPC:CHAPSO (2.8:1)	250 µL
30% DMPC:CHAPSO (2.8:1)	250 µL
25% DMPC:CHAPSO (2.8:1)	250 µL

MemMagic® Bicelle Screen kit provides four bicelle solutions comprising of 40%, 35%, 30%, and 25% DMPC:CHAPSO (2.8:1). When mixed with protein in a ratio of 1:4 (bicelle:protein), these bicelle solutions enable the screening of 8%, 7%, 6% and 5% bicelle concentrations in the final protein-bicelle mixture, providing an optimal bicelle concentration range supporting successful crystal growth. MemMagic® Bicelle Screen Kit can be used with any of our available crystallization screening kits. MemMagic® Bicelle Screen Kit can also be used in conjunction with nanoliter liquid handling robotic systems.

References:

Ujwal R, Bowie JU. (2011). *"Crystallizing membrane proteins using lipidic bicelles".* Methods. Dec 2011. 55(4):337-41.

Faham, S., Ujwal, R., Abramson, J. and Bowie, J. U. (2009) *"Practical Aspects of Membrane Proteins Crystallization in Bicelles"* Current Topics in Membranes, Volume 63, Chapter 5, 111-127.

Faham, S., Boulting, G. L., Massey, E. A., Yohannan, S., Yang, D., & Bowie, J. U. (2005*). "Crystallization of bacteriorhodopsin from bicelle formulations at room temperature"*. Protein Science, 14, 836–840.

Faham, S., & Bowie, J. U. (2002*). "Bicelle crystallization: A new method for crystallizing membrane proteins yields a monomeric bacteriorhodopsin structure"*. Journal of Molecular Biology, 316, 1–6.

Quick Start Guide to MemMagic® Bicelle Kit (Figure 2).

- Thaw frozen bicelles completely at room temperature until a clear gel-like phase is observed.
- Place the clear gel on ice to transform it to liquid form. The bicelle solution may become cloudy when placed on ice
- Vortex 2-3 seconds to mix and immediately place it back on ice to maintain its liquid form.
- Add the bicelle solution to the protein (preferably protein concentration >10mg/ml) in a 1:4 (bicelle:protein) ratio (e.g. 10 µL bicelle + 40 µL protein) while keeping everything on ice. Only make enough protein/bicelle mixture for a single day experiment. Do not store protein/bicelle mixture for next day or future use.
- Mix by pipetting the contents up and down until the solution appears homogenous (do not vortex).
- Incubate the protein/bicelle mixture on ice for minimum 30 min before setting up crystallization trials.
- Do not incubate crystallization trays at 4˚C or below.

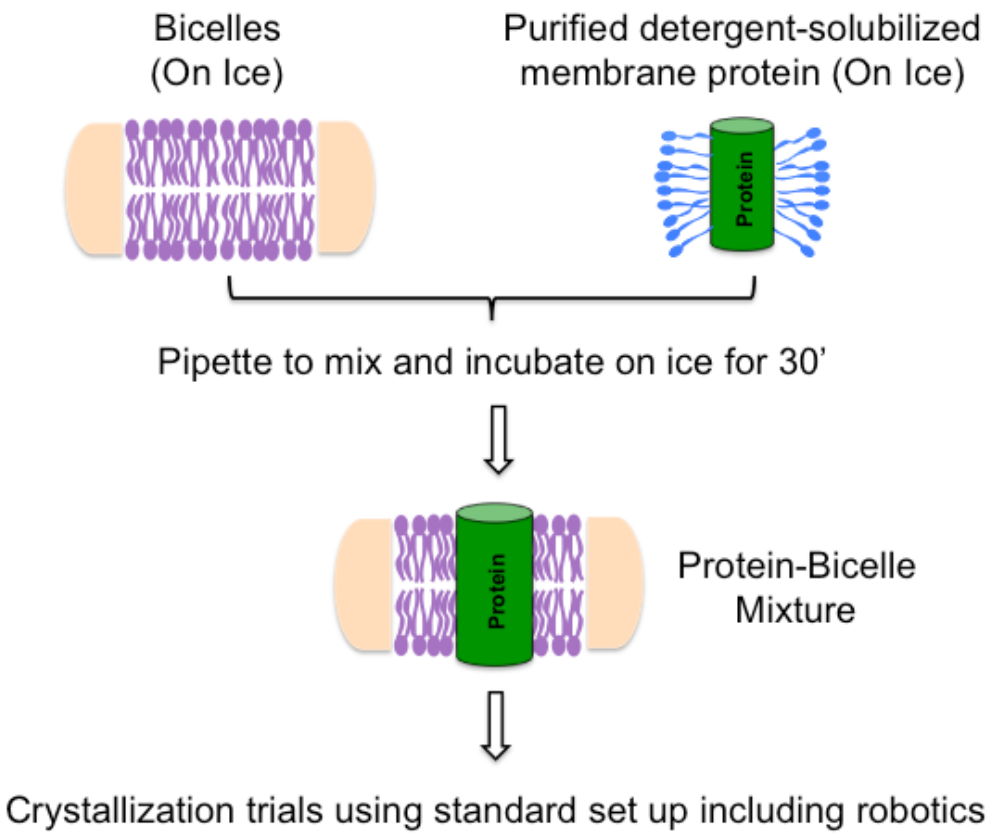


Figure 2. Bicelle crystallization. (Ujwal & Bowie (2011).

MemMagic® Bicelle Screen Kit Protocol

1. When ready to use, thaw the bicelle solution at room temperature until it becomes a clear gel. Place the clear gel on crushed ice (not on cold blocks) to allow it to transform completely to liquid form. Vortex for 2-3 seconds and immediately return it to crushed ice to maintain its liquid form. *MemMagic® Bicelle solution is stable under multiple freeze-thaw processes. The bicelle solution should be vortexed after each freeze-thaw to re-establish a homogenous bicelle phase. The bicelle solution may become cloudy when placed on ice.*

   Note: MemMagic® Bicelle solution has unique phase transition properties. It maintains liquid form at 4˚C or on ice. It transforms to a clear gel at room temperature or higher. It solidifies when below 0˚C.

2. Add the bicelle solution to the protein (preferably protein concentration >10 mg/ml) in a 1:4 (bicelle:protein) ratio (e.g. 10 µL bicelle + 40 µL protein) while keeping everything on ice.

   Note: Only make enough protein/bicelle mixture for a single day experiment.
   DO NOT store protein/bicelle mixture for next day or future use.

   Note: We recommend 1:4 (bicelle:protein) ratio to start. Other ratios can be performed as well, particularly in the optimization stage.

3. Mix by pipetting the contents up and down until the solution appears homogenous (**DO NOT VORTEX**).
4. Incubate the protein/bicelle mixture on ice for a minimum of 30 min before setting up crystallization trials in sitting drop or hanging drop format, using manual or standard high throughput robotic systems.

   Note: DO NOT incubate crystallization trays at 4˚C or below. Bicelles in the crystallization drops may precipitate under such temperature conditions.

   Note: Some conditions in many commercially available crystallization screens are not fully compatible with bicelle solutions, resulting in a high incidence of false positives (lipid crystals or salt crystals). We recommend using additional tools, such as a UV microscope, to rule out such false positives before proceeding to optimization.

The **Material Safety Data Sheet** (MSDS) information is provided on the Molecular Dimensions' website at *http://www.moleculardimensions.com*. Or can be downloaded via the QR Code.
MSDS documents are not included with product shipments.