

Molecular
Dimensions

Membrane Protein Crystallography



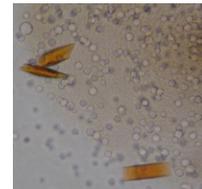
Contents: **Crystallization Screens**
 Additive Kits
 Detergent Kits
 Crystallization Plates

Crystallization Screens

LCP - MemMeso™

Developed in conjunction with the successful laboratory of Prof. Osamu Nureki at the University of Tokyo, Japan and optimized to work in synergy with Lipidic Cubic Phase (LCP) and the LCP crystallization method. Screening both LCP and Sponge Phase conditions are data-mined from current reported GPCR crystal structures.

Proven success – used to solve the crystal structures of at least eight membrane proteins, including channelrhodopsin¹.



Wizard™ Cubic LCP

Wizard Cubic LCP Kit™ kit contains all tools, formulations, and lipid to prepare micro-crystallization experiments according to the LCP micro method. This LCP random sparse matrix screen is designed for the crystallization of biological macromolecules that are embedded within a lipidic cubic phase (LCP) host matrix. Ideal for low-protein experiments: effective protein volume for a single crystallization experiment is about 80 nanoliters. Lipidic cubic phase has worked well for the crystallization of 7TM membrane proteins, and GPCRs.

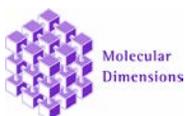
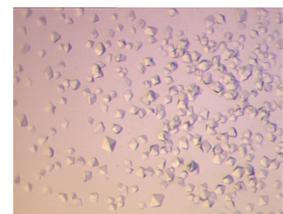
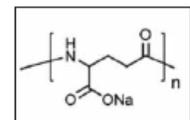


PGA Screen

A revolutionary new systematic screen based on poly-γ-glutamic acid (PGA), the first of the new chemical polymers that exploit poly-amino acids to unlock new areas of crystallization space. The high nucleation-precipitation potential of PGAs enables use at very low concentrations and in combination with classical precipitants, thus scaling down the amount of precipitant necessary for crystal appearance and growth.

Developed and utilized in the Structural Biology Lab, University of York, UK, and manufactured under an exclusive license.²

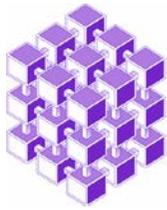
- Novel precipitant
- Totally New Crystallization Space
- Easy mixing properties with other PEGs
- Applicable to both globular and membrane protein crystallization
- Especially useful for labile, easily precipitating proteins



Molecular
Dimensions

moleculardimensions.com





Molecular
Dimensions

Membrane Protein Crystallography

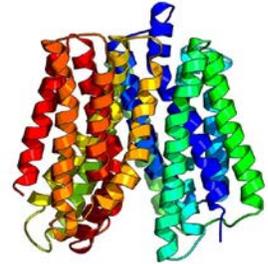


MemGold™ and MemGold™2

The MemGold screens were based on extensive data-mining of membrane protein structures in the pdb, and were carefully designed to include the most effective combinations of precipitants, pH, salt, and buffer components.³⁻⁴

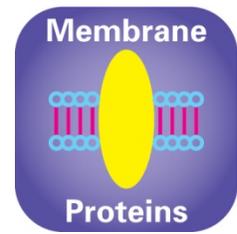
- Particularly suited for Prokaryotic and Eukaryotic alpha helical membrane proteins
- Screening over a wider range of pH's (4 - 10)
- Addition of small MW PEGs
- Can be used in conjunction with Lipidic Cubic Phase and/or Lipidic Sponge Phase

MemGold and MemGold 2 are exclusively supplied by Molecular Dimensions under a license from Imperial College of Science Technology and Medicine, London, and the University of Oxford respectively.



The "MEM Suite"

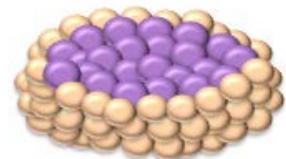
MemStart™, MemSys™, and MemPlus™ are exclusively supplied by Molecular Dimensions under license from Imperial College of Science Technology & Medicine, London. Designed and developed by members of the So Iwata laboratory at Imperial College, these crystal growth screens are widely acknowledged as the gold standard as the starting point for membrane protein crystal growth. The screens are available in 10ml tube and HT Block formats and fully supported by several publications.⁵⁻⁶



Bicelle Screening - MemMagic®

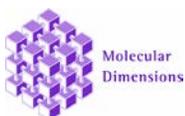
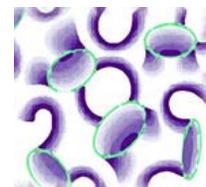
Membrane proteins can be readily reconstituted into bicelles and are maintained in a native-like bilayer environment, which can be manipulated with almost the same ease as for detergent solubilized membrane proteins.⁷

- User friendly - handles like a detergent
- Behaves like a lipid - even at 4°C
- Easily dispensed by hand or by standard robotics allowing rapid set up
- Extended storage periods compared to pre-made LCP
- Proven method - multiple published structures in protein crystallography
- Versatile - also useful for NMR structural studies and functional assay development



Lipidic-Sponge™ Phase Screen

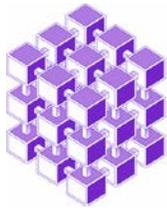
Market leading membrane protein screen, utilizing an expanded mesophase, yielding more accommodating aqueous channels than existing lipidic cubic phase methods. The Lipidic-Sponge Phase Screen is supplied as pre-mixed, easy-to-handle liquids, readily compatible with high-throughput crystallization approaches.⁸



Molecular
Dimensions

moleculardimensions.com





Molecular
Dimensions

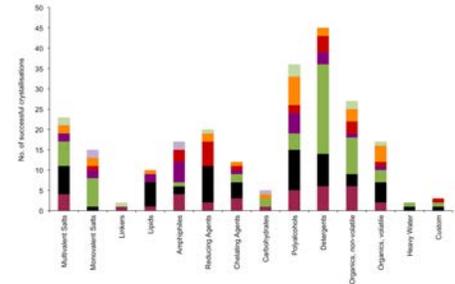
Membrane Protein Crystallography



Additive Kits

MemAdvantage™

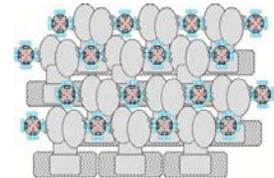
Additives may affect hydration and intermolecular interactions between protein molecules or between protein molecule and solvent or ligands. MemAdvantage is the first rational and intelligently designed additive screen targeted specifically for membrane proteins with easy screening of 96 different additives (12 different classes of polyalcohols, detergents, multivalent salts, non-volatile organics etc.) found to be the most successful in membrane protein crystallization⁴. It is particularly suited for Prokaryotic and Eukaryotic alpha helical membrane proteins. MemAdvantage™ is manufactured and distributed under an exclusive license with Dr. Simon Newstead and the University of Oxford.



New Calixar™ kit

*Calixarenes actively promote the stabilization and crystallization of membrane proteins by forming salt bridges across extra-membrane hydrophilic regions.*⁹

- New improved kit to explore even more additives
- Active promotion of crystallization via the formation of an organized aggregation state
- Form supramolecular clusters organizing CALIXAR derivatives into micelles leading to increased crystal formation
- Mini-micelles become intercalated between the membrane proteins via ionic interactions.
- Supramolecular clusters act like a molecular glue to increase crystalline organization



Detergent Kits

Wizard™ TIME screen

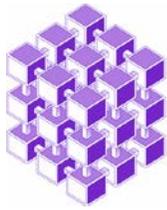
The Wizard™ Total Integral Membrane protein Extraction kit aids in high-throughput detergent screening. The kit consists of 84 different formulations for identifying those detergent reagents that successfully extract a membrane protein from a membrane preparation. Extraction yield and behavior of the protein can be characterized by the optimized combined ultracentrifugation and fluorescence size exclusion chromatography protocol¹⁰.



Molecular
Dimensions

moleculardimensions.com





Molecular
Dimensions

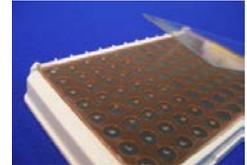
Membrane Protein Crystallography



Plates

Laminex™ and Laminex™HT

Laminex offers considerable advantages for viewing and imaging in lipidic and sponge phase experiment since the experiment is sandwiched between planar surfaces. The system is built up from lower support plates of glass or UV transparent plastic with an adhesive spacer or 100 or 200 microns. On to this base, glass, UV transparent plates or films can be applied to create 96 chambers for crystal growth. The whole assembly can be used as a sandwich or mounted on an SBS frame for robotic handling.



CrystalHarp™ Counter Diffusion Plate

This new SBS format plate with quartz capillaries, allows crystal growth and optimization with *in-situ* X-ray diffraction in the plate, or the individual capillaries can be mounted in standard CryoCaps.

- From screening to data collection, with minimum handling.
- Developed and patented at the Institute of Biochemistry, University of Zurich, Switzerland.
- 48 quartz capillaries per plate using the interface diffusion method.
- Proven experiment length (30mm) samples large amounts of crystallization phase space.
- Unique capillary material allows RT data collection or cryo-cooled in a liquid nitrogen stream (with or without the use of cryo-protectants).
- Gentle introduction of cryo-protectants or derivatives for phasing studies.



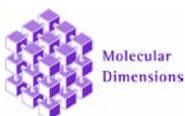
Sitting drop, Hanging drop and Microbatch Plates

Fourth generation plates inspired by crystallographers in polystyrene and UV transmissible polymer with micro-numbering.
See our Plates flier and catalogue.



References

1. Kato, H. et al. (2012) Crystal structure of the channelrhodopsin light-gated cation channel *Nature* **482** (7385):369-74.
2. Hu TC, Korczynska J, Smith DK, Brzozowski AM. (2008) High-molecular-weight polymers for protein crystallization: poly-*c*-glutamic acid-based precipitants *Acta Cryst D: Biological Crystallography* **D64**, 957-963.
3. Newstead S., Ferrandon S., Iwata S., (2008), Rationalizing α -helical membrane protein crystallization *Protein Sci.* **17**:466-472.
4. Parker S, & Newstead S. (2012) Current trends in α -helical membrane protein crystallization: An update *Protein Science* **21** (9): 1358-1365
5. Newstead S., Hobbs J., Jordan D., Carpenter E., Iwata S. (2008) *Molecular Membrane Biology* **25** :631-8
6. Methods and Results in the crystallization of Membrane proteins (2003) Ed. Iwata S. (International University Line).
7. Ujwal R. & Bowie JU. (2011) Crystallizing membrane proteins using lipidic bicelles *Methods* **55** 337-341
8. Wöhri A.W., et al., (2008) A lipidic-sponge phase screen for membrane protein crystallisation, *Structure* **16**,1003 - 1009.
9. Matar Merhab M et al. (2011) Structuring Detergents for Extracting and Stabilizing Functional Membrane Proteins *PLoS ONE* **6**(3)
10. Hattori M., Hibbs RE., & Gouaux E. (2012) A fluorescence-detection size-exclusion chromatography-based thermostability assay for membrane protein precrystallization screening. *Structure*. **20**(8): 1293-1299



Molecular
Dimensions

moleculardimensions.com

