

# Protein Crystallisation

An important method for the determination of protein structures is x-ray analysis of protein crystals. The determination of the three-dimensional structure of proteins has contributed towards major advances in basic research, particularly in the fields of structural genomics and structure-based drug design.

The most commonly used method for the crystallisation of proteins is vapour diffusion which comprises both the sitting drop and hanging drop methods (Fig. 1a and Fig. 1b). One drop of protein solution is mixed with one drop of reagent solution and incubated together with a larger volume of reagent solution in a sealed well.

Concentration gradients between the sample drop and the reservoir solution are balanced out by diffusion, which induces the crystallisation process if the correct conditions have been selected.

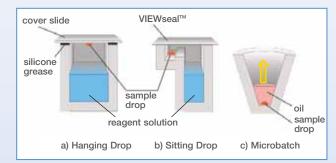


Figure 1: Crystallisation methods a) Hanging Drop b) Sitting Drop c) Microbatch The microbatch method (Fig. 1c) in which the sample drop is covered with oil is also widely used, and in this technique the choice of oil determines the rate of diffusion of the water in the sample drop through the oil.

Numerous factors affect the crystallisation of proteins. Since the optimal crystallisation conditions generally cannot be predicted, a large number of attempts is often necessary in order to determine and optimise the appropriate conditions. Protein crystallisation therefore still represents a major bottleneck in structure analysis. The use of high-throughput technologies, such as pipetting robots and standardised microplates, makes it possible to test a large number of crystallisation conditions in a short period of time and with relatively small amounts of protein.

The CrystalStar™ product range from Greiner Bio-One is a family of crystallisation plates and accessories designed specifically for high-throughput crystallisation.

## Format

We place great value on the suitability of our protein crystallisation plates for use with automated systems. Therefore, with the exception of Terasaki plates, all crystallisation plates have a footprint conforming to the ANSI 1-2004 standard. For further information please visit our website:

www.gbo.com/bioscience/technical\_information

6 Liquid Handling

7 Molecular Biology

Accessories

#### Barcode Labelling

Customer-specific barcode labelling is available on request for all crystallisation plates, with the exception of Terasaki plates.

## Material

All Greiner Bio-One protein crystallisation plates, with the exception of the LBR plates (see below), are made from polystyrene. This is characterised by high clarity and excellent optical properties.

#### **Hydrophobic Plates**

Plates with a hydrophobic surface are particularly well suited for nanolitre crystallisation of membrane proteins. The surface properties of hydrophobic plates efficiently counteract the spreading of detergent-containing drops, respectively of drops with surfactant precipitants, such as MPD (Fig. 2). Moreover, the meniscus of the screening solution in the reservoir is substantially reduced, so that contaminations through creeping of the screening solution into the crystallisation well are avoided.

## LBR Plates

LBR (low birefringence) plates are specifically designed for the use of polarised light. LBR plates for sitting drop applications are made from polyolefin which is characterised by very low birefringence in comparison with polystyrene plates (Fig. 3). Extreme transparency, high chemical resistance and low water absorption are further characteristics of LBR plates.



Figure 2: Comparison of (a) CrystalQuick™Plus (hydrophobic surface) and (b) CrystalQuick™ standard. Images of 100 nl drops containing 50 mM n-Octyl-Glucoside are courtesy of Karl Harlos, The Wellcome Trust Centre for Human Genetics, Oxford, UK.

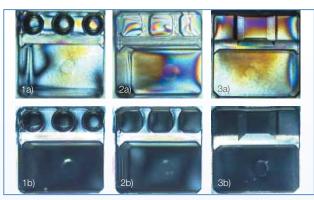


Figure 3: CrystalQuick™ plates in polarised light: (a) Standard versions with strong birefringence, (b) LBR versions with lower birefringence. (1) CrystalQuick™ RW (2) CrystalQuick™ SW (3) CrystalQuick™ LP

Further information on protein crystallisation → Forum No. 7: Advanced high-throughput platforms for protein crystallisation (F073 016)

CatNo.	Description	Number of sample wells	Number of reservoirs	Special features
		Vapour Diffusion		
662 150	ComboPlate™	-	24	-
662 050	ComboPlate™	-	24	pre-greased
609 101	CrystalQuick™ SW (Square Wells)	288	96	-
609 801	CrystalQuick™ SW (Square Wells)	288	96	LBR
609 130	CrystalQuick™ Plus SW (Square Wells)	288	96	hydrophobic
609 830	CrystalQuick™ Plus SW (Square Wells)	288	96	LBR, hydrophobic
609 120	CrystalQuick™ RW (Round Wells)	288	96	-
609 820	CrystalQuick™ RW (Round Wells)	288	96	LBR
609 171	CrystalQuick™ LP (Low Profile)	96	96	-
609 871	CrystalQuick™ LP (Low Profile)	96	96	LBR
609 180	CrystalQuick™ Plus LP (Low Profile)	96	96	hydrophobic
		Microbatch		
653 102	Terasaki Plate	60	-	-
654 102	Terasaki Plate	72	-	-
673 170	IMP@CT™ Plate	96	-	-
790 801	IMP@CT™ Plate	1536	-	LBR
		Accessories		
676 070	VIEWseal™	-	-	-
676 040	AMPLIseal™	-	-	-
662 145	CrystalBridge™	-	1	-
501 870	Coverslip, 18 mm ø, thickness 2 (0.19 - 0.22 mm)	-	-	glass, siliconised
503 870	Coverslip, 22 mm ø, thickness 2 (0.19 - 0.22 mm)	-	-	glass, siliconised
503 850	Coverslip, 22 mm ø, thickness 5 (0.5 - 0.6 mm)	-		glass, siliconised

Table 1: Overview of CrystalStar™ crystallisation plates and accessories.

11 Cryo-Technics