

Smart Screen – PurePEGs

The PurePEGs screen features conditions with a mixture of purified PEGs, ranging from 300 MW to 8,000 MW. At a total PEG concentration of 22.5 % [1], combined with various salts and buffers, the conditions are designed to give the most crystallization hits spanning a wide pH range (1.1 - 9.8).

Features:

- **Ultra pure PEGs – USP grade**
- **Optimized for diffusive mixing; capillary crystallization**
- **Polydispersed PEGs; 5 PEGs in each condition**
- **pH measured and recorded for final solution**
- **Better control of manufacturing impurities in the PEGs**

A variety of successful crystallization conditions are a combination of poly-ethylene-glycols (PEG's) and salts adjusted to a particular pH [2]. However, in an initial sparse screen, sampling the vast space of different salts versus different molecular weight (MW) PEGs, at a range of pH's, is challenging. Interestingly, there is evidence to suggest that protein crystallization hits obtained using a particular MW PEG can be repeated when using a mixture of PEGs [1]. Consequently, a more rational screening strategy should use mixtures of PEGs rather than individual conditions. Microlytic has now developed such a PEG cocktail contain PEGs ranging from 300 – 8,000 MW.

Additionally, the pH of various sparse screen solutions has been observed to significantly differ from what can be inferred from the precipitant composition [3, 4]. A common misconception is that the presence of a buffer adjusted to a particular pH, using HCl or NaOH, will have sufficient buffering capacity to overcome the acidic or basic nature of the other components of the condition [3, 4]. Since most crystallization screens are manufactured simply by adding individual pre-formulated components to generate a final solution, without adjusting or even measuring the pH of the final solution, there are bound to be significant discrepancies between the expected and actual pH of the precipitant condition. Particularly for conditions containing technical grade PEG's, variable concentrations of sodium sulfate, sodium hydroxide or sulfuric acid, will affect the pH and the salts in the final solution. Anecdotally, there has been an ongoing issue with Lithium Sulfate and PEG solutions producing crystals of salts; we believe that the precipitation is due to residual Sodium Sulfate from the manufacturing process causing super saturation of salts in mixture. As a consequence, protein crystallographers trying to optimize a crystallization hit from such a condition may have an incorrect starting point for their efforts.

To address these issues Microlytic is introducing a new screen containing a new series of PEGs that are USP grade, highly purified to avoid unwanted contaminants and improve reproducibility of experiments. Further, Microlytic lists the final pH of the crystallization conditions rather than the pH of the concentrated buffer components to provide protein crystallographers with as precise a starting point for optimization as possible.

References:

- [1] Newman *et al.* (2005) Towards rationalization of crystallization screening for small- to medium-sized academic laboratories: the PACT/JCSG+ strategy. *Acta Cryst.* **D61**:1426.
- [2] Page *et al.* (2003) Shotgun crystallization strategy for structural genomics: an optimized two-tiered crystallization screen against the *Thermotoga maritima* proteome. *Acta Cryst.* **D59**:1028.
- [3] Wooh *et al.* (2003) Comparison of three commercial sparse-matrix crystallization screens. *Acta Cryst.* **D59**:769.
- [4] Bukrinsky *et al.* (2001) pH, conductivity and long-term stability in the Crystal Screen solutions. *J. Appl. Cryst.* **34**:533.