Product Data Sheet

Cyclodextrin Screening Kit

Catalog Number: 0602

Product Description

The AthenaES™ Cyclodextrin Screening Kit is intended for identification of the best cyclodextrin for use in the refolding of proteins. Cyclodextrins' unique molecular structure greatly promotes protein refolding. Cyclodextrins can suppress protein aggregation, protect against degradation, and alter the functions of proteins. They also bind well to detergents (aggregation suppressors), which allows for easy separation of the detergent from the protein, encouraging proper refolding. The cyclodextrins remove the detergent from partially refolded protein solutions, allowing the protein to complete the refolding process.

Different cyclodextrins act and respond to different protein structures. Since no universal protein refolding condition has been identified, the best conditions for a given protein must be determined empirically. Because of this, AthenaES™ provides 3 of the most commonly used cyclodextrins in a simple screening kit so that the ideal conditions for the refolding of a given protein can be found quickly and easily in a rapid screening format.

The Cyclodextrin Screening Kit can prove particularly useful when paired with detergents and can be easily optimized with the AthenaES™ Detergent Screening Kit. Individual cyclodextrins and detergents are also available.

Instructions for Use

Cyclodextrins are typically used in a two-step protein refolding procedure. In the first step the protein is partially refolded in the presence of detergent. It is recommended that a suitable detergent be identified or that an experimental design which screens both the detergent and the cyclodextrin simultaneously be used. The following algorithm is suggested for identifying a suitable cyclodextrin:

- 1. Perform a preliminary screen of the physical-chemical parameters which are known to affect protein refolding. Parameters to test include pH, ionic strength, excipitents (i.e., detergents, polyols, chaotropic agents) redox state, temperature, and protein concentration. Refolding can be done by dilution, dialysis, or immobilization on a resin. Several commercial kits are available, such as Athena's Protein Refolding Kit (Cat. No. 0600), which simplify the screening process by providing the pre-mixed buffers along with straightforward statistical analyses of the results. A detailed method can be found in the AthenaES[™]'s Protein Refolding Kit Applications Manual at www.athenaes.com.
- 2. Identify a suitable detergent. Several different detergents should be tested to determine the best one for the given target protein. Optimization of the detergent



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Product Specifications

Unit Size	1 Kit
	Store at Room Temperature
Storage	DO NOT FREEZE

Kit Contents

Reorder No.	Component	Amt.
0609	100mM β- cyclodextrin	10mL
0610	100mM methyl-β-cyclodextrin	10mL
0611	50mM α-cyclodextrin	10mL

Material Safety Data

FOR RESEARCH USE ONLY. NOT INTENDED OR AP-PROVED FOR HUMAN, DIAGNOSTICS OR VETERINARY USE. Do not ingest, swallow or inhale. Do not get in eyes, on skin, or on clothing. Wash thoroughly after handling. For complete safety information see full Material Safety Data Sheet.

concentration is not needed at this stage, but should be determined once the cyclodextrin has been selected.

- 3. Perform the cyclodextrin screen. Perform the first step in the refolding process by employing the buffer composition and detergent identified in steps 1 and 2. The following protocol is recommended as it is one of the simplest to perform.
 - a. Prepare three reactions of the refolding buffer with the selected detergent and add the denatured protein to each solution slowly while mixing gently to give a final protein concentration of 50 μ g/ml.
 - b. Incubate for 1 h on ice.
 - c. Add each cyclodextrin to a final concentration of 5 mM, one cyclodextrin per reaction. Mix gently.
 - d. Incubate on ice for 1-24 h.
 - e. Assay the solution for refolded protein (by activity or physical means).
- 4. Once the cyclodextrin has been selected, the optimum concentration of the detergent and cyclodextrin should be determined. The process can then be adopted for an alternate refolding scheme such as by dialysis, diafiltration, or chromatography.

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