INSTRUCTIONS



ProFoldin Detergent Critical Micelle Concentration (CMC) Assay Kit

CATALOG NUMBER CMC1000

INTRODUCTION

The critical micelle concentration (CMC) value of a detergent depends on the solution compositions such as salt concentrations. Knowing the CMC value of the detergent in a particular buffer is essential in membrane protein extraction, purification and crystallization. Membrane proteins tend to aggregate when the detergent concentration is below the CMC. A too high detergent concentration may cause many problems including protein instability, difficulty in purification and crystallization. A low concentration of a detergent may be used for enzyme assays where formation of detergent micelles should be avoided in testing enzyme inhibitors.

The CMC assay kit is based on the principle that the detergent interacts with the fluorescence dye and enhances the fluorescence intensity at 465 nm (excitation at 360 nm). The kit can be used for measurement of the CMC values of known detergents in solutions with different salt concentrations that affect the CMC values. It can also be used to measure the CMC values of new detergents or to test if a new molecule is a detergent that should have a CMC value.

The CMC assay kit includes $100 \ \mu$ l of $1000 \ x$ CMC dye. It is for measurement of $1000 \ samples$ using 96-well plates. It can also be used for determination of detergent CMC values using cuvettes.



INSTRUCTIONS



ASSAY PROTOCOL

The following protocol is for assays using a standard black 96-well plate (Greiner 655076). Adjust the assay reagent volumes proportionally for measurements using cuvettes.

1. Prepare standard detergent solutions by a serial dilution of the detergent from 0.5 % to 0.0005% in water or a buffer. Set a control sample without detergent.

Note: This concentration range applies to most detergents. A different concentration range may be used for detergents with unusually high or low CMC values.

- 2. For each 100 samples, dilute 10 μ l of the 1000 x CMC dye with 10 ml of water.
- 3. Mix 100 μ l of the detergent solution with 100 μ l of the 1 x dye and incubate the solution in the dark for 30 min.
- 4. Read the fluorescence at 465 nm (excitation at 360 nm).

DATA ANALYSIS

- 1. Calculate the ratio between the fluorescence of the samples with detergent and the control sample without detergent.
- 2. Plot the fluorescence ratio versus the detergent concentration. The transition point is the detergent CMC value.

RELATED PRODUCTS

DAK1000	Detergent Assay Kit
LIP1000	MicroGram Lipid Assay Kit
NPA1000	NanoMolar Phosphate Assay Kit
PPD1000	MicroMolar Polyphosphate Assay Kit
EPA001	Easy Protein Assay Reagent
HIS200	MicroMolar Histidine Assay Kit
CYS200	MicroMolar Cysteine Assay kit
PEP200	Peptide Assay Kit
PAA100K	MicroMolar Primary Amine Assay Kit
CAK1000	Coenzyme A Assay Kit
EDTA200	MicroMolar EDTA Assay kit
DTT200	MicroMolar DTT Assay kit
MAD100K	MicroMolar ADP Assay kit
MUD100K	MicroMolar UDP assay kit
MCA1000	MicroMolar Copper Assay Kit
NZA1000	NanoMolar Zinc Assay Kit
NMA1000	NanoMolar Nickel / Cobalt Assay Kit
MSA200	MicroMolar Sulfate Assay Kit

For more information of concentration assays, please visit <u>www.profoldin.com</u>.