



ProFoldin

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INSTRUCTIONS

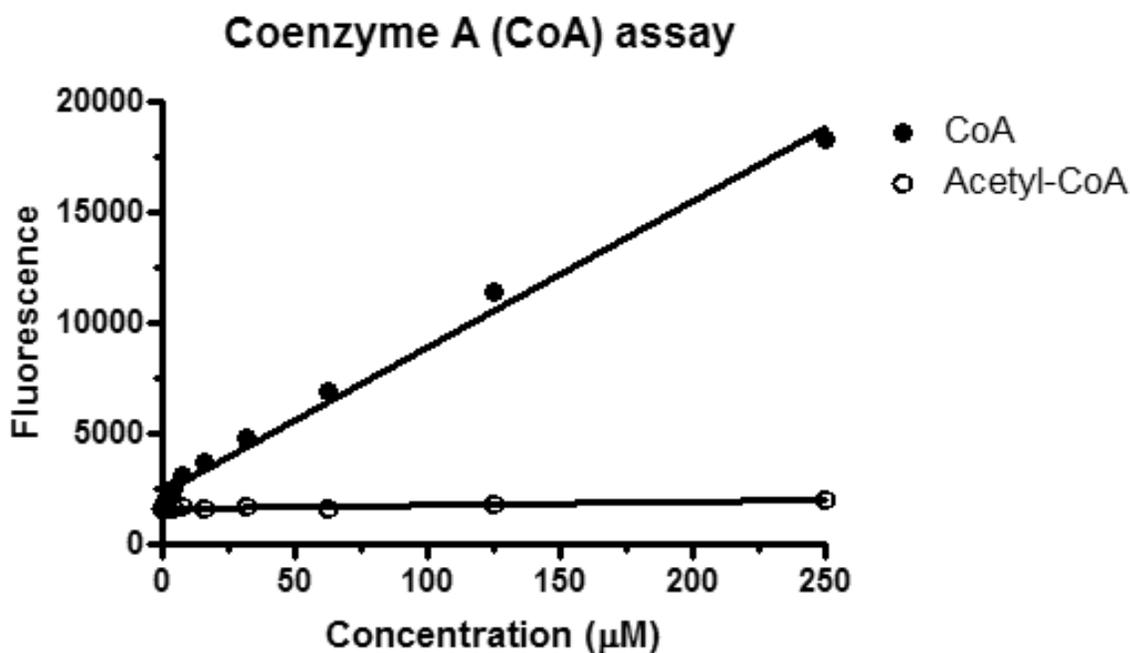
ProFoldin Coenzyme A Assay Kit

CATALOG NUMBER **CAK1000**

INTRODUCTION

The Coenzyme A Assay Kit is for measurement of micromolar concentrations of Coenzyme A (CoA). The assay is based on the principle that the thiol group of CoA interacts with the CAK reagent and enhances the fluorescence intensity at 535 nm (excitation at 485 nm). Acetyl-CoA does not interact with the CAK reagent. The assay reagent is sufficient for measurement of 1000 samples using 384-well plates. The assay kit can be used for measurement of other thiols.

The assay is compatible with HEPES buffer, low concentrations of non-ionic detergent (<0.01%), sodium and potassium salt, MgCl₂ (< 5 mM), CaCl₂ (<5 mM) and phosphate (< 1 mM). The assay is not compatible with ammonium, Tris buffer or other primary amine buffers. It is not compatible with amino acids, DTT or EDTA.



The Coenzyme A Assay Kit (catalog number CAK1000) includes 750 µl 10 x CAK reagent. It is for 1000 assays using 384-well plates or 250 assays using 96-well plates. Cuvettes may also be used for measurements.



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PROTOCOL

CoA Standard Curve

1. Prepare standard CoA solutions with a series of concentrations from 1 to 200 μM in 10 mM HEPES, pH7.5.
2. Prepare 1 x CAK reagent by dilution of the 10 x CAK reagent with water (10-fold dilution).
3. Mix 120 μl of the CoA solution with 30 μl of 1 x CAK reagent in the wells of a 96-well plate for 10 min and read the fluorescence at 535 nm (excitation at 485 nm).

Data Analysis

Plot the fluorescence intensity **F_c** and the CoA concentration [**CoA**] to generate the linear standard curve.

$$\mathbf{F_c} = \mathbf{a} [\mathbf{CoA}] + \mathbf{b}$$

Where the **F_c** values are from experimental data, the **a** and **b** values are from the linear fitting between the **F_c** values and the CoA concentrations.

UNKNOWN SAMPLES

Follow the same procedure to measure the fluorescence intensity **F_c** values from the unknown samples. Calculate the CoA concentrations in the unknown samples using the **F_c** values from the unknown samples and the **a** and **b** values from the standard curve.

$$[\mathbf{CoA}] = (\mathbf{F_c} - \mathbf{b}) / \mathbf{a}$$

RELATED PRODUCTS

NPA1000	NanoMolar Phosphate Assay Kit
PPD1000	MicroMolar Polyphosphate Assay Kit
HIS200	MicroMolar Histidine Assay Kit
CYS200	MicroMolar Cysteine Assay kit
AAK1000	Amino acid assay kit
PEP200	Peptide Assay Kit
PAA100K	MicroMolar Primary Amine Assay Kit
EDTA200	MicroMolar EDTA Assay kit
DTT200	MicroMolar DTT Assay kit
DAK1000	Detergent assay kit
LIP1000	MicroGram Lipid Assay Kit
MAD100K	MicroMolar ADP Assay kit
MCA1000	MicroMolar Copper Assay Kit
NZA1000	NanoMolar Zinc Assay Kit
CLA100	MicroMolar Chloride Assay Kit
MSA200	MicroMolar Sulfate Assay Kit

For more information of concentration assays and enzyme essays, please visit www.profoldin.com.