Mannitol Microplate Assay Kit

User Manual

Catalog # CAK1165

(Version 1.3A)

Detection and Quantification of Mannitol Content in Serum, Urine, Other biological fluids, Food, Beverage Samples.

For research use only. Not for diagnostic or therapeutic procedures.

I. INTRODUCTION

Mannitol is a sugar alcohol used in dietary supplement, sweetener, intestinal permeability test for leaky gut, etc. It also serves as a coating for hard candies, dried fruits, and chewing gums due to its low ability to attract and hold water molecules. In addition, it is an osmoprotectant for plants and is used clinically in osmotherapy to reduce intracranial pressure.

Mannitol Microplate Assay Kit is designed to measure mannitol in various samples in 96-well microplate. The color intensity at 413 nm is directly proportional to mannitol concentration in the sample.

II. KIT COMPONENTS

Component	Volume	Storage
96-Well Microplate	1 plate	
Reaction Buffer	2.5 ml x 1	4 °C
Stop Solution	Powder x 1	4 °C
Dye Reagent	10 ml x 1	4 °C
Standard	Powder x 1	4 °C
Plate Adhesive Strips	3 Strips	
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Note:

Standard: add 1 ml Distilled water to dissolve before use, then add 0.1 ml into 0.9 ml Distilled water, the concentration will be 200 µg/ml.

Stop Solution: add 5 ml distilled water to dissolve before use.

III. MATERIALS REQUIRED BUT NOT PROVIDED

1. Microplate reader to read absorbance at 413 nm
2. Distilled water
3. Pipettor, multi-channel pipettor
4. Pipette tips
5. Distilled water
6. Hot air circulation oven

IV. SAMPLE PREPARATION

1. For serum, urine and other biological fluids samples

Samples can be assayed directly.

V. ASSAY PROCEDURE

Add following reagents in the microplate:

Reagent	Sample	Standard	Blank
Sample	25 µl	--	--
Standard	--	25 µl	--
Distilled water	--	--	25 µl
Reaction Buffer	25 µl	25 µl	25 µl
Mix, incubate at room temperature for 10 minutes.			
Stop Solution	50 µl	50 µl	50 µl
Mix, incubate at room temperature for 5 minutes.			
Dye Reagent	100 µl	100 µl	100 µl
Mix, cover the plate adhesive strip, put the plate into the oven, incubate at 53 °C for 15 minutes, then put it on ice immediately, record absorbance measured at 413 nm.			

Note:

1) Perform 2-fold serial dilutions of the top standards to make the standard curve.

2) The concentrations can vary over a wide range depending on the different samples. For unknown samples, we recommend doing a pilot experiment & testing several doses to ensure the readings are within the standard curve range.

3) Reagents must be added step by step, can not be mixed and added together.

VI. CALCULATION

1. According to the volume of sample

$$\text{Mannitol } (\mu g/ml) = C_{Standard} \times V_{Standard} \times (OD_{Sample} - OD_{Blank}) / (OD_{Standard} - OD_{Blank}) / V_{Sample}$$

$$= 200 \times (OD_{Sample} - OD_{Blank}) / (OD_{Standard} - OD_{Blank})$$

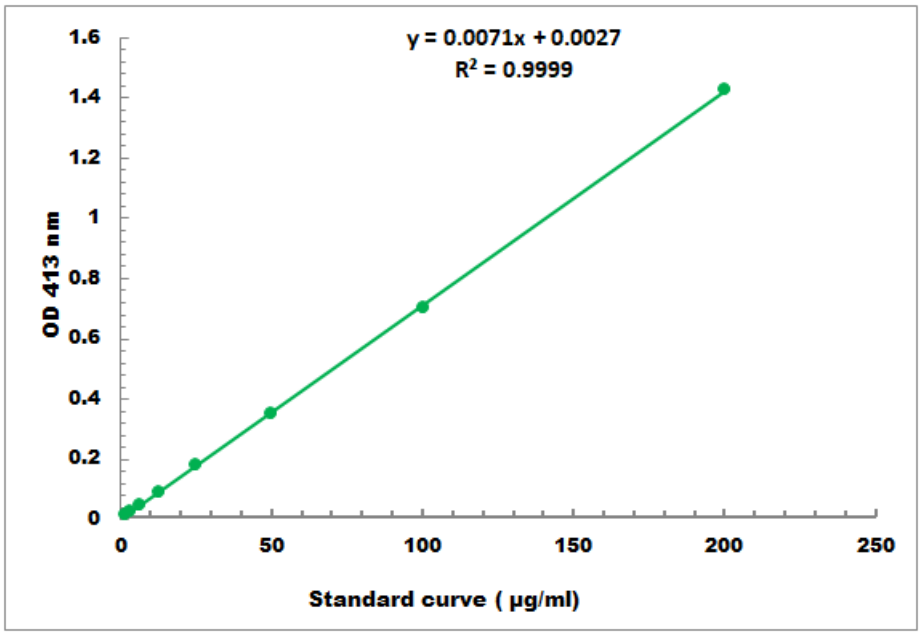
$C_{Standard}$: the standard concentration, 200 μg/ml;

$V_{Standard}$: the volume of standard, 0.025 ml;

V_{Sample}: the volume of sample, 0.025 ml.

VII. TYPICAL DATA

The standard curve is for demonstration only. A standard curve must be run with each assay.



Detection Range: 2 μg/ml - 200 μg/ml

VIII. TECHNICAL SUPPORT

For troubleshooting, information or assistance, please go online to www.cohesionbio.com or contact us at techsupport@cohesionbio.com

IX. NOTES