



Zinc Microplate Assay Kit

User Manual

Catalog # CAK1110

(Version 1.3D)

Detection and Quantification of Zinc (Zn^{2+}) Content in Serum, Urine,
Saliva and Other biological fluids Samples.

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

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I. INTRODUCTION

Zinc, a metallic chemical element, symbol Zn and atomic number 30 is chemically similar to Magnesium due to its similar size and sole oxidation state of $^{2+}$. Zinc is an essential mineral of great biological significance, because many enzymes require it as an essential cofactor. Examples of zinc's biological roles include signal transduction, gene expression, regulation of apoptosis, synaptic plasticity and prostate gland function.

The reaction products can be measured at a colorimetric readout at 558 nm.

II. KIT COMPONENTS

Component	Volume	Storage
96-Well Microplate	1 plate	
Assay Buffer I	15 ml x 1	4 °C
Assay Buffer II	15 ml x 1	4 °C
Reaction Buffer	15 ml x 1	4 °C
Dye Reagent	Powder x 1	4 °C
Standard (400 µmol/L)	1 ml x 1	4 °C
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Note:

Dye Reagent: add 1 ml ethanol to dilute before use.

III. MATERIALS REQUIRED BUT NOT PROVIDED

1. Microplate reader to read absorbance at 558 nm
2. Distilled water
3. Pipettor, multi-channel pipettor
4. Pipette tips
5. Ethanol
6. Centrifuge
7. Timer

IV. SAMPLE PREPARATION

1. For liquid sample

Add 40 µl serum and 40 µl Assay Buffer I into the microcentrifuge tube, mix, centrifuged at 10,000g 4 °C for 10 minutes, transfer the supernatant into a new centrifuge tube; then add 40 µl Assay Buffer II into the microcentrifuge tube, mix, centrifuged at 10,000g 4 °C for 10 minutes, transfer the supernatant into a new centrifuge tube for detection.

V. ASSAY PROCEDURE

Add following reagents into the microplate:

Reagent	Blank	Standard	Sample
Assay Buffer I	20 µl	20 µl	--
Assay Buffer II	20 µl	20 µl	--
Distilled water	20 µl	--	--
Standard	--	20 µl	--
Sample	--	--	60 µl
Reaction Buffer	130 µl	130 µl	130 µl
Dye Reagent	10 µl	10 µl	10 µl
Mix, wait for 2 minutes, measured at 558 nm and record the absorbance.			

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 serum sample

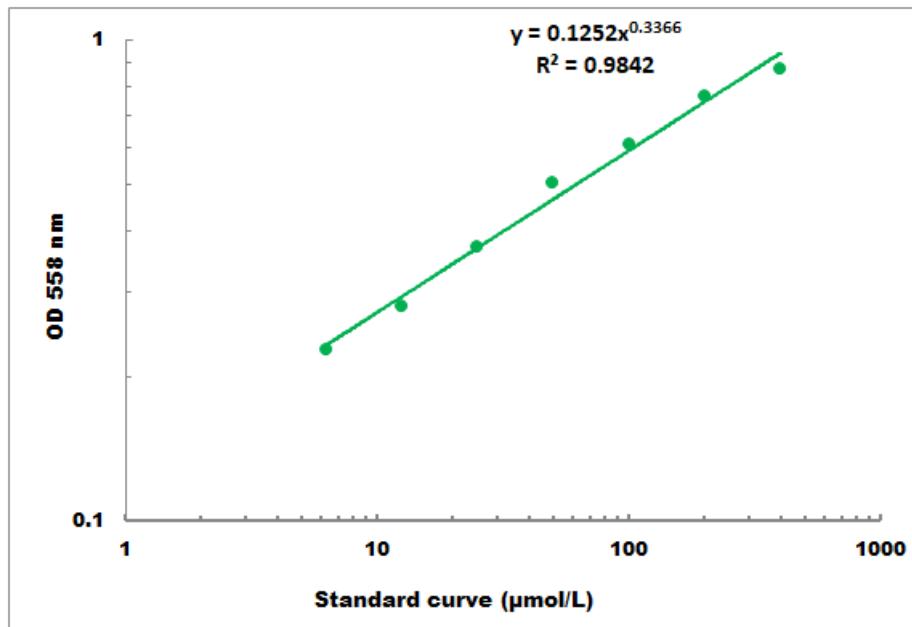
$$\begin{aligned} \text{Zn}^{2+} (\mu\text{mol/L}) &= C_{\text{Standard}} \times (\text{OD}_{\text{Sample}} - \text{OD}_{\text{Blank}}) / (\text{OD}_{\text{Standard}} - \text{OD}_{\text{Blank}}) \times n \\ &= 1200 \times (\text{OD}_{\text{Sample}} - \text{OD}_{\text{Blank}}) / (\text{OD}_{\text{Standard}} - \text{OD}_{\text{Blank}}) \end{aligned}$$

C_{Standard} : the concentration of standard, 400 $\mu\text{mol/L}$.

n: sample dilution factor, n=3

VII. TYPICAL DATA

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



Detection Range: 4 μmol/L - 400 μmol/L

VIII. TECHNICAL SUPPORT

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

IX. NOTES