



Natrium

Colorimetric Microplate Assay Kit

User Manual

Catalog # CAK1109

(Version 1.4C)

Detection and Quantification of Natrium (Na^+) 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

Sodium (Na^+) is one of the most important electrolytes along with chloride, calcium and potassium. Na plays vital roles in the maintenance of plasma volume, pH balance, transmission of nerve impulses, and normal cell functions. Healthy individuals can absorb sodium ingested in food, and kidneys maintain proper sodium balance by excreting its excess in urine. Sodium sources include table salt, milk, meat, shellfish, bread, snack food, etc. Normal Sodium intake has been defined to be between 200-500 mg/day. Patients suffering high blood pressure, hypertension, chronic kidney disease, and people suffering salt sensitivity require restricted low-sodium diets due to those conditions. Hyponatremia (low sodium concentration in blood) can occur in patients with nephrotic syndrome, excessive vomiting and diarrhea, while Hypernatremia (high sodium concentration in blood) is developed in patients suffering from liver diseases, burns, and pregnancy.

NatriumColorimetric Microplate Assay Kit is designed to directly measure natrium content in a variety of samples. It is based on the enzyme kinetic reaction β -galactosidase catalyzed substrate O-NPG, measured at 405 nm is proportional to the natrium concentration in the sample.

II.KIT COMPONENTS

Component	Volume	Storage
96-Well Microplate	1 plate	
Reaction Buffer	20 mlx 1	4 °C
Enzyme	Powderx 1	4 °C
Substrate	Powderx 1	4 °C
Standard	Powderx 1	4 °C
Technical Manual	1 Manual	

III. MATERIALS REQUIRED BUT NOT PROVIDED

1. Microplate reader to read absorbance at 405 nm
2. Distilled water
3. Pipettor, multi-channel pipettor
4. Pipette tips
5. Mortar
6. Centrifuge
7. Timer

IV. REAGENT PREPARATION

Standard: Briefly centrifuge prior to opening. Dissolve in 1 ml distilled water to generate 1000 mmol/L of standard top solution, store at 4 °C for 1 month after reconstitution. Perform 2-fold serial dilutions of the top standard solution using distilled water to make the standard curve. The concentration of standard curve could be 1000/500/250/125/62.5 /31.2/15.6 mmol/L.

Enzyme: Briefly centrifuge prior to opening. Add 1 ml Reaction Buffer to dissolve before use. Store at 4 °C for 1 day or -80°C for 1 month.

Substrate: Briefly centrifuge prior to opening. Add 1 ml Reaction Buffer to dissolve before use. Store at 4 °C for 1 week or -80°C for 3 months.

Note: Divide into small aliquots to avoid repeated freeze-thaw cycles.

V. SAMPLE PREPARATION

1. For serum and other liquid samples

Detect directly.

2. For tissue samples

Weigh 0.1 g tissue, homogenize with 1 ml distilled water, centrifuged at 10000g for 20 minutes, take the supernatant into a new centrifuge tube for detection.

VI. ASSAY PROCEDURE

Add following reagents into the microcentrifuge tube:

Reagent*	Sample**	Standard	Blank
Reaction Buffer	170 μ l	170 μ l	170 μ l
Sample	10 μ l	--	--
Standard	--	10 μ l	--
Distilled water	--	--	10 μ l
Substrate	10 μ l	10 μ l	10 μ l
Enzyme	10 μ l	10 μ l	10 μ l
Mix, incubate at 37 °C for 10 mins, record absorbance measured at 405 nm.			

Note:

*Reagents must be added sequentially and should not be premixed prior to addition.

**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.

**For colored samples, we recommend setting a parallel sample background control well with same volume of sample only. Other reagents were replaced by distilled water to the same total volume. Subtract the OD value of the sample background control from the OD value of the sample to correct for interference from the sample's own color.

VII. CALCULATION

1. Calculate the sample concentration in ASSAY PROCEDURE according to the slope of the standard curve

$$C = \frac{(OD_{\text{Sample}} - OD_{\text{Blank}}) - \text{Intercept}}{\text{Slope}} \times n (\text{mmol/L})$$

Calculate the initial concentration according to sample preparation procedure.

2. According to one point of the standard OD and concentration

2.1 According to the weight of sample

$$C = \frac{(C_{\text{Standard}} \times V_{\text{Standard}}) \times (OD_{\text{Sample}} - OD_{\text{Blank}})}{(OD_{\text{Standard}} - OD_{\text{Blank}}) \times W \times (V_{\text{Sample}} / V_{\text{Assay}})} (\mu\text{mol/g})$$

2.2 According to the volume of sample

$$C = \frac{(C_{\text{Standard}} \times V_{\text{Standard}}) \times (OD_{\text{Sample}} - OD_{\text{Blank}})}{(OD_{\text{Standard}} - OD_{\text{Blank}}) \times V_{\text{Sample}}} (\mu\text{mol/ml})$$

Slope: the absorbance slope of standard curve

n: the dilution factor

C_{Standard} : the standard concentration, mmol/L = $\mu\text{mol/ml}$

V_{Standard} : the volume of standard in assay procedure, μl

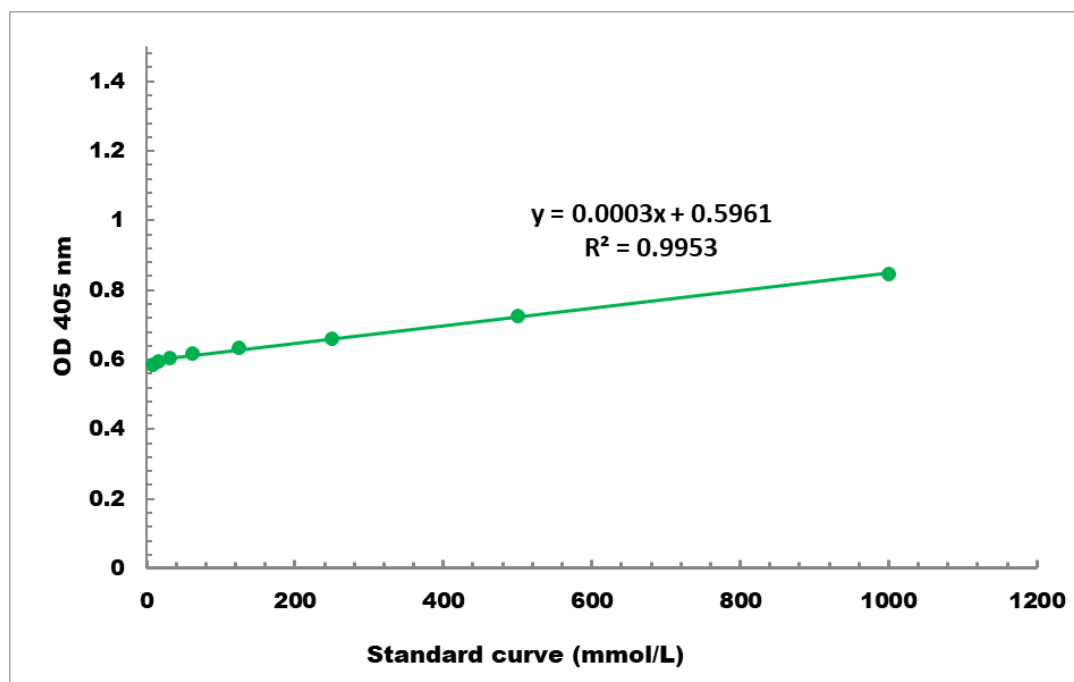
V_{Sample} : the volume of sample in assay procedure, μl

V_{Assay} : the volume of Assay Buffer, μl

W: the weight of sample, g

VIII. TYPICAL DATA

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



Detection Range: 10mmol/L - 1000mmol/L