Sodium (Na^+) Assay Kit

Catalog No.: abx295103

Size: 100 tests

Detection Range: 0.02 mmol/L – 10 mmol/L

Sensitivity: 0.02 mmol/L

Storage: Store all components at 4°C in the dark.

Application: For the detection and quantification of Sodium concentration in serum, plasma and tissue homogenates.

Principle of the Assay

Abbexa's Sodium Assay Kit is a quick, convenient, and sensitive method for measuring and calculating Sodium concentration. In the presence of sodium, the nitropyranoside substrate is catalyzed to form nitrophenol, with an absorbance maximum at 405 nm. The intensity of the color is proportional to the Sodium concentration, which can then be calculated.

Kit components

1. Standard (5 mmol/L): 1.6 ml
2. Chromogenic Reagent: 50 ml
3. Enzyme Stock Solution: 2 × 40 ml
4. Enzyme Reagent: 4 vials

Materials required but not provided

1. Spectrophotometer (405 nm) and 1 cm optical path quartz cuvette
2. Double-distilled water
3. Pipette and pipette tips
4. Centrifuge and centrifuge tubes
5. Mechanical homogenizer
6. Vortex mixer
7. Incubator
8. Timer
9. Ice

Protocol

A. Preparation of samples and reagents

1. Reagents

- **Reaction Working Solution:** Reconstitute 1 vial of Enzyme Reagent with 15 ml of Enzyme Stock Solution and mix thoroughly. Unused solution can be stored at 4°C for up to 2 days.

Note:

- Allow all reagents to equilibrate to room temperature before use.

2. Samples

Isolate the test samples soon after collecting and analyze immediately or aliquot and store at -20°C or -80°C for long-term storage. Avoid multiple freeze-thaw cycles.

The following sample preparation methods are intended as a guide and may be adjusted as required depending on the specific samples used.

- **Serum and plasma**: Serum and plasma samples can be tested directly. Samples can be stored at -80°C for up to 1 month.
- **Tissue homogenates:** Carefully weigh at least 20 mg of tissue. Per 20 mg of tissue, homogenize manually in 180 µl of double-distilled water, using a mechanical homogenizer, at 4°C. Centrifuge at 10,000 × g for 10 minutes at 4°C. Carefully take the supernatant for detection. Keep on ice and assay immediately.

Note: To calculate Sodium concentration in tissue homogenates using the formula in section **C. Calculation of Results**, the total protein concentration of the supernatant must be determined separately (**abx097193**)

It is recommended to carry out a preliminary experiment to determine the optical dilution factor of samples before carrying out the formal experiment. Dilute samples into different concentrations with double-distilled water, then carry out the assay procedure. The recommended dilution factor for different samples is as follows (for reference only):

Sample Type	Dilution Factor
Human serum	15 – 25
Mouse serum	15 – 25
Human plasma	15 – 25
Rat plasma	15 – 25
10% Mouse liver tissue homogenate	15 – 25
10% Rat heart tissue homogenate	15 – 25

Note:

- Fresh samples or recently obtained samples are recommended to prevent degradation and denaturalization that may lead to erroneous results.
- Lysis buffers may interfere with the performance of this kit. It is recommended to use mechanical lysis methods only for tissue homogenates.

B. Assay Procedure

Preheat the incubator and ensure it has reached a stable temperature before use.

1. Label tubes for each standard, blank and sample. *It is strongly recommended to test all tubes in duplicate.*
2. Add 50 µl of 5 mmol/L standard to the standard tubes.
3. Add 50 µl of double-distilled water to the blank tube.
4. Add 50 µl of sample to the sample wells.
5. Add 400 µl of Chromogenic Reagent to all tubes and mix thoroughly.
6. Add 600 µl of Reaction Working Solution to all tubes and mix thoroughly.
7. Using double-distilled water, set the spectrophotometer to 0.
8. Using a 1 cm optical path quartz cuvette, measure the absorbance of each tube at 405 nm. Record these OD values as A_1.
9. Incubate all tubes at 37°C for 3 minutes. *The incubation time should start immediately from the measurement of A_1. As the incubation time is short, it is recommended to assay one tube at a time.*
10. Using a 1 cm optical path quartz cuvette, measure the absorbance of each tube at 405 nm. Record these OD values as A_2.

C. Calculation of Results

1. Serum and plasma samples:

$$\text{Sodium (U/ml)} = \frac{(\Delta A_{\text{Sample}} - \Delta A_{\text{Blank}})}{(\Delta A_{\text{Standard}} - \Delta A_{\text{Blank}})} \times C \times F$$

2. Tissue samples:

$$\text{Sodium (U/mg protein)} = \frac{(\Delta A_{\text{Sample}} - \Delta A_{\text{Blank}})}{(\Delta A_{\text{Standard}} - \Delta A_{\text{Blank}})} \times \frac{C}{C_{\text{Protein}}} \times F$$

where:

ΔA_{Sample}	$A_{2\ \text{Sample}} - A_{1\ \text{Sample}}$
ΔA_{Blank}	$A_{2\ \text{Blank}} - A_{1\ \text{Blank}}$
$\Delta A_{\text{Standard}}$	$A_{2\ \text{Standard}} - A_{1\ \text{Standard}}$
C	Concentration of Standard (5 mmol/L)
C_{Protein}	Concentration of protein in sample (g/L)
F	Dilution factor of the sample

Technical Support

For troubleshooting and technical assistance, please contact us at support@abbexa.com.