Potassium Assay Kit

(Cat/No.:BC062 Size:96T)

1. Determination principle (microplate method)

In alkaline medium, potassium ions in serum samples treated with protein precipitants react with NA-TPB to produce turbid and stable suspensions. The turbidity is proportional to the potassium ion concentration in the sample.

2. Composition and preparation (The kit is valid for 6 months)

96T	Components	Specification	save
Reagent 1	Nail Liquid	20mL×1 bottle	at 4 ℃ for 6 months
	Liquid B	2.5mL×1 bottle	at 4 ℃ for 6 months
	Preparation of protein precipitation agent : Prepare according to the ratio of Liquid A : Liquid B = 8:1 and prepare it before use.		
Reagent 2	NA-TPB working fluid	25mL×1 bottle	at 4 ℃ for 6 months
Reagent 3	0.8mmol / L potassium standard solution	1mL×1 tube	at 4 ℃ for 6 months
	Preparation of 0.4mmol/L potassium standard solution: Mix 0.8mmol /L potassium standard solution **and** deionized water in a ratio of **1 : 1.**		

3.Sample and Instrument Requirements

1. Collect and process samples according to routine testing requirements. Samples can be serum (plasma), tissue homogenate, cells, or culture supernatant.

2. Samples are stable for 3-4 days at 2-8℃ and for several months below -20℃.

3. This method is for assays using an ELISA reader (440±10nm).

4. Operation steps

1. Sample pretreatment:

Serum (plasma) sample : Take 20μL of serum (plasma) and add 180μL of protein

precipitant , centrifuge at 3500 rpm for 5 minutes , and take 50μL of supernatant for measurement.

Tissue samples : Accurately weigh the tissue , add 9 times **deionized water** at a ratio of weight (g): volume (mL) = 1:9, homogenize in an ice water bath, centrifuge at 2500 rpm for 10 minutes , take 20μL of the supernatant and add 180μL of protein precipitant , centrifuge at 3500 rpm for 5 minutes, and take 50μL of the supernatant for measurement.

2. Operation table:

	Blank well	Standard well	Measurement well
Deionized water (μL)	50		
0.4mmol / L potassium standard solution (μL)		50	
Supernatant of sample to be tested (μL)			50
Working solution (μL)	200	200	200
Mix well, let stand for 5 minutes and measure the absorbance of each well using a microplate reader at 440nm.			

Note : Before the experiment, you can read and record the OD value of the empty plate using a microplate reader. To ensure the accuracy of the experiment.

5. Calculation formula

1. Serum calculation formula:

Serum (Plasma) Potassium Concentration (mmol/L) =

$$\frac{A_{measurement} - A_{blank}}{A_{standard} - A_{blank}} \times C_{standard} \times N \times 10$$

C standard : standard concentration, 0.4mmol/ L;

N: dilution multiple of the sample before testing (after adding the precipitant and before mixing with the working solution);

10: dilution factor when the sample is added with protein precipitant;

Cpr: protein concentration of tissue homogenate, gprot/ L (prot refers to protein);

W: tissue weight, g;

V extract : the total volume of extract added during tissue homogenization, L.

6. Technical parameters

Linear range	0.01 ~ 0.8mmol/L	Intra-batch variation	≤4%
Batch Difference	≤6%	Kit Shelf Life	6 months

7. Note

1. Red blood cells contain high concentrations of potassium ions, so hemolyzed samples are not acceptable.

2. Ammonia , mercury and chlorine may interfere with the determination of potassium.

3. When preparing tissue homogenate , it is best to use deionized water as the homogenate medium and

potassium contamination should be avoided .

4. This kit can be used on fully automatic / semi - automatic biochemical analyzers .

5. Determine the potassium ion content in tissues or cells . It is recommended to simultaneously determine the total protein concentration

6. This kit is for scientific research only.

Appendix I: Preparation of Potassium Standard Curve

1. Pre-treatment :

Use deionized water to gradient dilute the 0.8mmol/ L potassium standard solution into different

concentrations : 0.025mmol/ L , 0.05mmol/ L , 0.1 mmol/ L , 0.2 mmol/ L , and 0.4 mmol/ L .

2. Operation table:

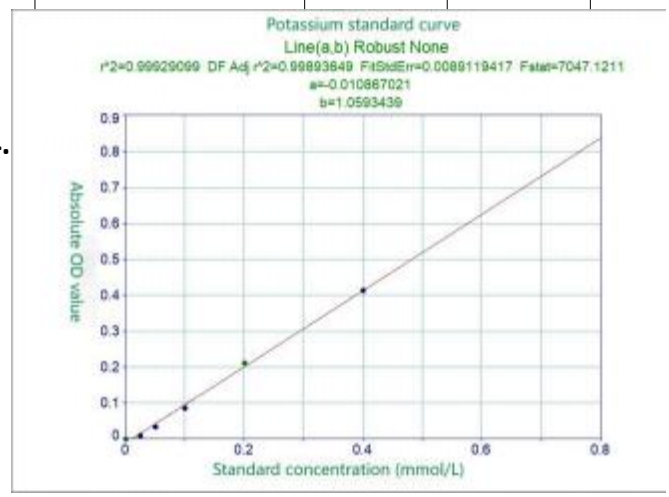
	Blank well	Standard well
Deionized water (μL)	50	
Potassium standard solutions of different concentrations (μL)		50
Working solution (μL)	200	200

Mix well, let stand for 5 minutes , and measure the absorbance of each well using a microplate reader at 450nm .

3. Measurement results :

Standard concentration (mmol/L)	Determination of OD value	Absolute OD value
0	0.0697	0
0.025	0.0802	0.0105
0.05	0.1042	0.0345
0.1	0.1564	0.0867
0.2	0.2818	0.2121

4.



(Users do not need to make a standard curve, and can use the calculation formula to calculate)

Appendix II: Determination of potassium in serum samples

1. Pre-treatment :

Take 20μL of serum (plasma) , add 180μL of protein precipitant , centrifuge at 3500 rpm for 5 minutes, and take 50μL of supernatant for measurement.

2. Operation table:

	Blank well	Standard well	Measurement well
Deionized water (μL)	50		
0.4mmol / L potassium standard solution (μL)		50	
Supernatant of sample to be tested (μL)			50
Working solution (μL)	200	200	200
Mix well, let stand for 5 minutes , and measure the absorbance value of each well using an enzyme-labeled colorimeter at 450nm .			

Note: Before the experiment, the 96- well plate can be read and recorded with an ELISA reader . To ensure the accuracy of the experiment.

3. Calculation formula :

$$\text{Serum (Plasma) Potassium Concentration (mmol/L)} = \frac{A_{measurement} - A_{blank}}{A_{standard} - A_{blank}} \times C_{standard} \times N \times 10$$

C standard : standard concentration, 0.4mmol/ L;

N: dilution multiple of the sample before testing after adding the precipitant and before mixing with the working solution)

Dilution factor of the sample when protein precipitant is added

4. Calculation example :

Example 1: Take 20 μL of rat serum (plasma) and measure according to the instructions : the OD value of the blank well is 0.0697, the OD value of the standard well is 0.4830, and the OD value of the test well is 0.5763

The calculation result is :

$$\text{Serum (Plasma) Potassium Concentration} = \frac{0.5763 - 0.0697}{0.4830 - 0.0697} \times 0.4 \times 10 \times 1 = 4.9034 \text{ mmol/L}$$

Example 2 : Take 20 μL of fish serum (plasma) and measure according to the instructions : the OD value of

the blank well is 0.0697, the OD value of the standard

well is 0.4830, and the OD value of the test well is 0.2621 . The calculation result is :

$$\text{Serum (Plasma) Potassium Concentration} = \frac{0.2621-0.0697}{0.4830-0.0697} \times 0.4 \times 10 \times 1$$

$$= 1.8619 \quad \text{mmol/L}$$

Appendix III : Potassium Determination in Tissue Samples

1. Pre-treatment :

Accurately weigh the tissue , add 9 times deionized

water at a ratio of weight (g): volume (mL) = 1:9 ,

homogenize in an ice water bath, centrifuge at 2500 rpm for 10 minutes , take 20μL of the supernatant, add 180μL of protein precipitant , centrifuge at 3500 rpm for 5

minutes , and take 50μL of the supernatant for measurement.

2. Operation table:

	Blank hole	Standard hole	Sample well
Deionized water (μL)	50		
0.4mmol / L potassium standard solution (μL)		50	
Supernatant of sample to be tested (μL)			50
Working solution (μL)	200	200	200
Mix well, let stand for 5 minutes , and measure the absorbance of each well using a microplate reader at 450nm .			

Note : Before the experiment, the 96- well plate can be read and recorded with an ELISA reader . To ensure the accuracy of the experiment.

4. Calculation formula :

$$\text{Potassium Content in Tissue (mmol/gprot)} = \frac{A_{measurement} - A_{blank}}{Cpr\ A_{standard} - A_{blank}} \times C_{standard} \times 10 \div$$

$$\text{Potassium Content in Tissue (mmol/gtissue)} = \frac{A_{measurement} - A_{blank}}{A\quad\quad A} \times C_{standard} \times 10 \div \frac{W_{extract}}{V}$$

C standard : standard concentration, 0.4mmol/ L;

10: dilution factor of the sample when the protein precipitant is added;

Cpr: protein concentration of tissue homogenate, gprot/ L (prot refers to protein);

W: tissue weight, g; V : total volume of extract added during tissue homogenization, L.

4. Calculation example：

Example 1: Take 20 μL of 10% homogenate supernatant of a certain tissue and measure it according to the instructions : the OD value of the blank well is 0.0697, the OD value of the standard well is 0.4830, the OD value of the test well is 0.6699 , and the protein concentration of the 10% squid gill homogenate supernatant is 4.5851 ; the calculation result is :

$$\text{Potassium Content in Tissue (mmol/gprot)} = \frac{0.6699-0.0697}{0.4830-0.0697} \times 0.4 \times 10 \div 4.5851$$

$$= 1.2670 \quad \text{mmol/gprot}$$

Example 2: Take 20 μL of 10% brain tissue homogenate supernatant and measure according to the instructions : the OD value of the blank well is 0.0697, the OD value of the standard well is 0.4830, the OD value of the test well is 0.4350 , and the protein concentration of the 10% rabbit brain tissue homogenate supernatant is 3.7909 ; the calculation result is :

$$\text{Potassium Content in Tissue (mmol/gprot)} = \frac{0.4350-0.0697}{0.4830-0.0697} \times 0.4 \times 10 \div 3.7909$$

$$= 0.9326 \quad \text{mmol/gprot}$$