CheKine™ Micro Uric Acid (UA) Assay Kit

Cat #: KTB1510 Size: 48 T/96 T

	Micro Uric Acid (UA) Assay Kit		
REF	**Cat #:** KTB1510	LOT	**Lot #:** Refer to product label
	Applicable samples: Serum, Plasma or Urine Sample, Animal Tissues		
	Storage: Stored at 4℃ for 6 months		

Assay Principle

CheKine™ Micro Uric Acid (UA) Colorimetric Assay Kit provides a simple method for detecting Uric Acid (UA) concentration in a variety of biological samples such as Animal Tissues, Serum, Plasma, Urine and other biological Fluids. In the assay, Uricase can catalyze UA to produce allantoin, CO_2 and H_2O_2; H_2O_2 oxidizes Fe^{2+} in potassium ferrocyanide to produce Fe^{3+}; Fe^{3+} further condenses with phenol and 4-aminoantipyrine to form red quinone compounds which has a characteristic absorption peak at 505 nm. The UA content can be calculated by measuring the light absorption at this wavelength.

Materials Supplied and Storage Conditions

Kit components	Size		Storage conditions
	48 T	96 T	
Extraction Buffer	60 mL	60 mL×2	4℃
Reagent Ⅰ A	0.65 mL	1.3 mL	4℃, protected from light
Reagent Ⅰ B	0.35 mL	0.7 mL	4℃, protected from light
Standard	1	1	4℃

Materials Required but Not Supplied

- Microplate Readeror Visible Spectrophotometer capable of measuring absorbance at 505 nm
- Precision pipettes, Disposable Pipette Tips
- Deionized Water, Centrifuge, Incubator, 96 well plate or Microglass cuvette
- Ice Maker, Homogenizer (for tissue samples)

Reagent Preparation

Extraction Buffer: Ready to use as supplied. Equilibrate to room temperature before use. Store at 4℃.

Working Reagent Ⅰ A : Prepared before use; For standard tubes and test tubes, it needs to dilute this reagent at 1:10 before use as follows: For the product size of 48 T, add 5.85 mL of Extraction Buffer, and then get 6.5 mL of Working Reagent Ⅰ A; For the product size of 96 T, add 11.7 mL of Extraction Buffer, and then get 13 mL of Working Reagent Ⅰ A.

Working Reagent Ⅰ B: Prepared before use; For blank tubes, it needs to dilute this reagent at 1:10 before use as follows: For the product size of 48 T, add 3.15 mL of Extraction Buffer, and then get 3.5 mL of Working Reagent Ⅰ B; For the product size of 96 T,

add 6.3 mL of Extraction Buffer, and then get 7 mL of Working Reagent ⅰ B.

Standard: Add 20 mL deionized water to the tube before use, and heat to dissolve at 60℃.

Sample preparation

Note: Fresh samples are recommended, If not assayed immediately, samples can be stored at -80℃ for one month.

1. Serum, Plasma or Urine Sample: No need to deal with Serum (Plasma) or Urine sample.

2. Tissues: According to the ratio of Tissue weight (g): Extraction Buffer volume (mL) at 1:5~10, it is recommended to weigh about 0.1 g tissue and add 1 mL Extraction Buffer. Homogenize on ice. Centrifuge at 10,000 rpm for 10 min at 4℃, aspirating the supernatant, place it on ice to be tested.

Assay procedure

1. Preheat the Microplate Reader or Visible Spectrophotometer for more than 30 min, and adjust the wavelength to 505 nm. Visible Spectrophotometer deionized water zero.

2. Sample measurement (Add the following reagents respectively into the 96 well plate or Microglass cuvette):

Reagent	Blank well (μL)	Standard well (μL)	Test well (μL)
Working Reagent ⅰ A	0	150	150
Working Reagent ⅰ B	150	0	0
Standard	0	60	0
H_2O	60	0	0
Sample	0	0	60
Mix well, and then incubated at 37℃ for 30 min. Add 200 μL of mixture into a 96 well plate or Microglass cuvette, measure the absorbance values of blank well, standard well and test well at 505 nm, and recorded them as A_{Blank}, $A_{Standard}$, A_{Test}.			

Data Analysis

Note: We provide you with calculation formulae, including the derivation process and final formula. The two are exactly equal. It is suggested that the concise calculation formula in bold is final formula.

1. Calculation of UA concentration in tissues

UA (μmol/g fresh weight) $=C_{Standard}\times(A_{Test}-A_{Blank})\div(A_{Standard}-A_{Blank})\div(W\div V_{Sample\ Total})$**$=5\times(A_{Test}-A_{Blank})\div(A_{Standard}-A_{Blank})\div W$**

2. Calculation of UA concentration in Serum, Plasma, Urine or other biological Fluids

UA (μmol/mL) $=C_{Standard}\times(A_{Test}-A_{Blank})\div(A_{Standard}-A_{Blank})$**$=5\times(A_{Test}-A_{Blank})\div(A_{Standard}-A_{Blank})$**

Where: $C_{Standard}$: the concentration of Standard,5 μmol/mL; W: the fresh weight, g; A_{Test}: the Absorbance of the Test well; A_{Blank}: the Absorbance of the Blank well; $A_{Standard}$: the Absorbance of the Standard well; $V_{Sample\ Total}$: the volume of Extraction Buffer added to Samples, 1 mL.

Recommended Products

Catalog No.	Product Name
KTB1500	CheKine™ Micro Total Antioxidant Capacity (TAC) Assay Kit
KTB1080	CheKine™ Micro Superoxide anion Scavenging Capacity Assay Kit
KTB1091	CheKine™ Micro Hydroxyl Free Radical Scavenging Capacity Assay Kit

Disclaimer

The reagent is only used in the field of scientific research, not suitable for clinical diagnosis or other purposes.