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Sulfate Assay Kit

10/18

(Catalog # K415-200; 200 assays, Store kit at Room Temperature)

I. Introduction:

Inorganic sulfate ion is one of the most abundant polyatomic anions in mammalian body fulids and it is essential for the synthesis of many biomolecules such as glycosaminoglycans, choline sulfate, steroid sulfate, cerebroside sulfate and heparin sulfate. Some biomolecules having a sulfate moiety can activate or assist detoxification processes of drugs, steroids, neurotransmitters, bile acids, food additives and heavy metals. In humans, sulfate ions are mostly absorbed by kidneys and little is excreted into urine. Sulfate deficiency can lead to fetal underdevelopment, autism or osteochondrodysplasia disorder. Therefore, the estimation of physiological concentrations of sulfate in human is critical. BioVision's Sulfate Assay is a simple, fast and accurate assay to determine sulfate concentration in biological fluids such as serum and urine samples. The detection reagent in the kit specifically reacts with free sulfate ions present in a variety of biological samples. The signal is measured at OD 600 nm and is proportional to the amount of sulfate up to 2 mM. BioVision's Sulfate Assay is suitable for the detection sulfate ion in serum and urine samples and it can detect as low as 0.1 mM of this ion.

Sulfate + Detection Reagent ⟶ White Suspension (measurement at 600 nm)

II. Application:

- Measurement of inorganic sulfate biological and non-biological samples

III. Sample Type:

- Urine, serum and plasma

IV. Kit Contents:

Components	K415-200	Cap Code	Part Number
Detection Reagent	1 vial	Red	K415-200-1
Detection Buffer	25 ml	NM	K415-200-2
Treatment Solution	10 ml	NM	K415-200-3
2 mM Sulfate Standard	25 ml	WM	K415-200-4

V. User Supplied Reagents and Equipment:

- 96-well clear flat-bottom plate
- Multi-well spectrophotometer

VI. Storage Conditions and Reagent Preparation:

Store kit components at room temperature. The kit components are stable for one year when stored as recommended. Briefly centrifuge small vials at low speed prior to opening. Read the entire protocol before performing the experiment.

- **Detection Reagent**: For 10 assay wells, weigh out 100 mg of Detection Reagent and dissolve in 1 ml of Detection Buffer to prepare Detection Solution (100 mg/ml). Vortex for 1 min to dissolve the reagent completely and let it stay at room temperature for 5 min before use. Discard unused Detection Solution after 3 hrs.
- **Detection Buffer, Treatment Solution and 2 mM Standard**: Ready to use.

VII. Sulfate Assay Protocol:

1. **Sample Preparation:** Urine samples need to be diluted 10-fold using deionized water. After dilution, add 200 µl of diluted urine into the desired wells for assay. For serum or plasma, vortex 500 µl of sample briefly with 50 µl of Treatment Solution and spin it at 10,000 x g for 10 min. After centrifugation, transfer clear supernatant to a new Eppendorf tube and add 200 µl of the supernatant into desired wells for assay.
2. **Standard Curve:** Add 0, 50, 100, 150 and 200 µl of 2 mM Standard into the desired wells. Add water to adjust the final volume to 200 µl to generate 0, 0.5, 1.0, 1.5 and 2.0 mM standards respectively.
3. **Measurement:** Add 100 µl of prepared Detection Solution (100 mg/ml) into each assay well. Incubate the plate at room temperature for 5 min and then measure O.D. at 600 nm.
4. **Calculation:** Correct the background by subtracting the O.D. of the control (S0) from all readings. Plot the net O.D. at 600 nm vs. sulfate concentration. Sulfate concentration in the sample can be determined using the formula below:

$$\text{Sulfate (mM)} = \frac{\text{O.D.}_{\text{sample}}}{\text{Slope}} \times D$$

Where: $\text{O.D.}_{\text{sample}}$ is the O.D. of the sample after background correction
D is the dilution factor (serum is 1 and urine is 10)

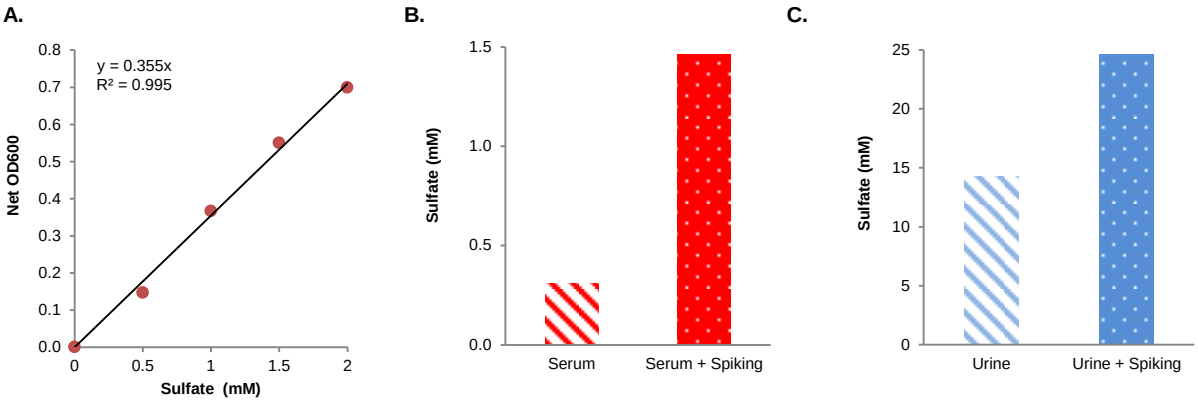

Figure A. Sulfate Standard Curve. **Figures B** Sulfate concentration determined in non-spiked and spiked (1.2 mM sulfate) serum samples **C.** Sulfate concentration determined in non-spiked and spiked (10 mM sulfate) urine samples. A spike recovery higher than 97% was obtained in both samples.

VIII. RELATED PRODUCTS:

Ethanolamine Kinase 2, human recombinant (P1158)
Phospholipid Assay Kit (Colorimetric/Fluorometric) (K351)
Phospholipase D (PLD) Activity Colorimetric Assay Kit (K725)
Pyruvate Colorimetric/Fluorometric Assay Kit (K609)
Citrate Colorimetric/Fluorometric Assay Kit (K655)
Citrate Synthase Activity Colorimetric Assay Kit (K318)
Succinate (Succinic Acid) Colorimetric Assay Kit (K649)
Choline Kinase B, Human Recombinant (P1220)
Phospholipase A2 Activity Assay Kit (Fluorometric) (K400)
Malate Colorimetric Assay Kit (K637)
PicoProbe™ Acetyl-CoA Fluorometric Assay Kit (K317)
Oxaloacetate Colorimetric/Fluorometric Assay kit (K659)
Isocitrate Colorimetric Assay Kit (K656)

FOR RESEARCH USE ONLY! Not to be used on humans.