Ammonia Assay Kit II (BA0207)

Product Datasheet | SKU BA0207

Description

Ammonia (NH3), or its ion form ammonium (NH4+), is an important source of nitrogen for living systems and is ubiquitously present in nature. Simple, direct and automation-ready procedures for measuring NH3 are highly desirable. This updated ammonia and ammonium assay is based on an improved o-phthalaldehyde method. This reagent reacts with ammonia and ammonium and forms a fluorescent product. The fluorescence intensity (lambda ex/em = 415/475 nm) is proportional to the ammonia concentration in the sample.

Specifications

Detection Method	Fluorimetric (lambda ex/em = 415/475 nm)
Detection Range	0.005 - 2 mM ammonia (detection limit 5.3 uM)
Sample Type	Urine, serum, plasma, environmental and other biological samples
Species Reactivity	All
Assay Time	20 minutes
Kit Size	100 Assays
Equipment Required	Microplate reader
Storage	Shipped at room temperature. Store kit at -20 degrees C.
Shelf Life	6 months after receipt
Shipping	Room Temperature

Kit Components

Component	Quantity	Storage
Detection Reagent	10 mL	-20 degrees C
Standard (NH4Cl)	100 uL	-20 degrees C

Ordering Information

Assay Genie Cat. No.	Pack Size
BA0207	100 Assays

Assay Procedure

Note: This is a sample protocol. Protocols are batch/lot specific - always follow the instructions supplied with your kit.

1. Use black flat-bottom 96-well plates and bring all reagents to room temperature prior to assay.
2. Prepare 200 uL of 1 mM Standard Premix by mixing 10 uL of 20 mM NH4Cl Standard and 190 uL of water.
3. Dilute the standards according to the standard dilution table to give 1.00, 0.50, 0.25 and 0 mM.
4. Transfer 10 uL of each standard into separate wells of the plate.
5. Transfer 10 uL of each sample into separate wells of the plate.
6. Add 90 uL of Detection Reagent to each well and immediately tap the plate to mix.
7. Incubate for 20 minutes in the dark at room temperature.
8. Measure fluorescence intensity at lambda ex/em = 415/475 nm on a plate reader.

Standard Dilution

No	Premix	+ H2O	Standard (mM)
1	100 uL	0 uL	1.00

No	Premix	+ H2O	Standard (mM)
2	50 uL	50 uL	0.50
3	25 uL	75 uL	0.25
4	0 uL	100 uL	0

Calculation

Plot the ammonia standard curve and determine its slope. The ammonia concentration of a sample is calculated as: [NH3] = (F_SAMPLE - F_BLANK) / Slope (mM), where F_SAMPLE and F_BLANK are the fluorescence intensity values of the sample and the blank (standard #4, water), respectively. If the ammonia concentration is higher than 2 mM, dilute the sample in water, repeat the assay and multiply the result by the dilution factor.

Notes

- This assay is compatible with most detergents, chelators and buffer components. Protein concentrations up to 17 ug/mL are tolerated and primary amine-containing buffers (e.g. Tris, glycine) should be kept below 10 mM where possible. For best results, include the same concentration of sample buffer in the standards and blank.
- Samples should be clear and free of particles or precipitates. Particles or precipitates can be removed by centrifugation for 5 minutes at 14,000 rpm or by filtration.
- Urine samples should be diluted 50-fold in water prior to assay. Serum and plasma samples should be diluted 10-fold in water prior to assay.
- Reagents are for research use only. Refer to the MSDS for detailed safety information.