

## Amplite® Fluorimetric Lithium Ion Quantification Kit

Catalog number: 21351  
Unit size: 100 Tests

Component	Storage	Amount (Cat No. 21351)
Component A: Lithiummighty™ 520	Freeze (< -15 °C), Minimize light exposure	1 vial
Component B: Assay Buffer	Minimize light exposure, Refrigerated (2-8 °C)	1 bottle (20 mL)
Component C: Lithium Standard	Refrigerated (2-8 °C), Minimize light exposure	1 vial
Component D: DMSO	Minimize light exposure, Refrigerated (2-8 °C)	1 vial (200 µL)

### OVERVIEW

Quantifying lithium ions is important in various scientific fields and industries, including biochemistry, medicine, environmental analysis, and food science, etc. The rapid and accurate determination of lithium ions is particularly important in the battery industry. There are several methods commonly used to quantify lithium ions, including flame photometry, ion-selective electrodes (ISE), atomic absorption spectroscopy, and fluorescence spectrophotometry. Flame photometry and atomic absorption spectroscopy require the inflammation of the samples. They are tedious to use and require expensive and sophisticated instrumentation. Ion-selective electrodes require large volumes of samples and often have low selectivity. Among all the methods, fluorescence spectrophotometry is the most convenient method for quantifying lithium ions. Fluorescence spectrophotometry involves complexing lithium ions with specific reagents and measuring the resulting fluorescence changes. However, there is still lacking a fluorescence-based lithium ion assay kit in the commercial market due to the absence of a robust fluorescence lithium ion indicator. Amplite® Fluorimetric Lithium Ion Quantification Kit uses our new robust lithium-ion indicator dye, Lithiummighty™ 520, which exhibits great fluorescence intensity enhancement upon binding to lithium ions. Lithiummighty™ 520 is perhaps the most robust lithium-ion indicator with high selectivity. It enables the kit to be useful for the rapid determination of lithium concentrations in a variety of samples compared to the other commercial lithium-ion assays. This microplate-based assay kit requires an extremely small amount of sample. It is particularly suitable for the determination of lithium ion concentration in a microvolume format.

### AT A GLANCE

#### Protocol Summary

1. Add 50 µL Lithium Standards or test samples
2. Add 50 µL Lithiummighty™ 520 working solution.
3. Incubate at RT for 5-10 minutes
4. Monitor the fluorescence at Ex/Em=490/525 nm

#### Important

The following protocol is an example for quantifying lithium content using Lithiummighty™ 520. Allow all the components to warm to room temperature before opening. The DMSO stock solution should be handled with particular caution as DMSO is known to facilitate the entry of organic molecules into tissues.

### KEY PARAMETERS

#### Fluorescence microplate reader

Cutoff	515 nm
Emission	525 nm
Excitation	490 nm
Recommended plate	Solid black

### PREPARATION OF STOCK SOLUTIONS

*Unless otherwise noted, all unused stock solutions should be divided into single-use aliquots and stored at -20 °C after preparation. Avoid repeated freeze-thaw cycles*

#### Prepare Lithiummighty™ 520 stock solution

1. Add 100 µL of DMSO (Component D) into Lithiummighty™ 520 vial (Component A).

**Note:** Make a single unused Lithiummighty™ 520 stock solution aliquot and store at ≤ -20 °C. Protect from light and avoid repeated freeze-thaw cycles.

### PREPARATION OF STANDARD SOLUTIONS

For convenience, use the Serial Dilution Planner:  
<https://www.aatbio.com/tools/serial-dilution/21351>

#### Lithium Standard

Add 350 µL of distilled water to the Lithium Standard vial (Component C) to make a 1M standard stock solution. Next, dilute this 1M stock solution using Assay Buffer (Component B) to make a 300 mM (LS1). Then perform 1:2 serial dilutions to get serially diluted Lithium Standard (LS2 – LS7).

### PREPARATION OF WORKING SOLUTION

#### Prepare Lithiummighty™ 520 working solution

1. Add 100 µL of Lithiummighty™ 520 (Component A) into 5 mL of Assay Buffer (Component B). Protect the working solution from light by covering it with foil or placing it in the dark.

**Note:** For best results, this solution should be used within a few hours of its preparation.

**Note:** 5 mL of working solution is enough for 100 tests.

### SAMPLE EXPERIMENTAL PROTOCOL

#### Important

The following protocol only provides a guideline and should be modified according to your specific needs.

**Table 1. Layout of Lithium standards and test samples in a solid black 96-well microplate.**

LS=Lithium Standards (LS1 - LS7, 300 to 4.69 mM, 2X dilutions);  
BL=Blank Control; TS=Test Samples

BL	BL	TS	TS
LS1	LS1	...	...
LS2	LS2	...	...
LS3	LS3	...	...
LS4	LS4	...	...
LS5	LS5	...	...
LS6	LS6	...	...
LS7	LS7	...	...

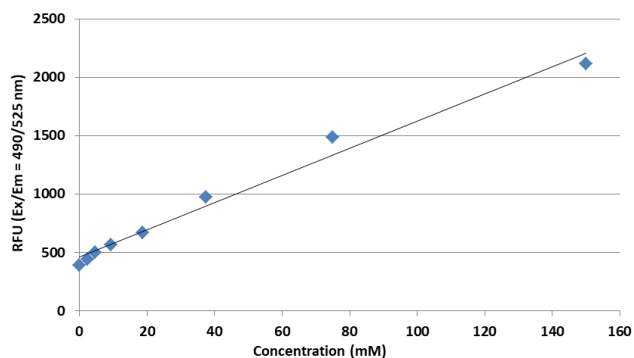
**Table 2. Reagent composition for each well.**

Well	Volume	Reagent
LS1-LS7	50 $\mu$ L	Serial dilutions (300 to 4.69 mM)
BL	50 $\mu$ L	Assay Buffer
TS	50 $\mu$ L	Sample

#### Protocol

1. Prepare Lithium Standards (LS), blank controls (BL), and test samples (TS) according to the layout provided in Tables 1 and 2. For a 384-well plate, use 25  $\mu$ L of reagent per well instead of 50  $\mu$ L.
2. Add 50  $\mu$ L of Lithiummighty™ 520 working solution to each well of Lithium Standards, blank control, and test samples to make the assay volume of 100  $\mu$ L/well. For a 384-well plate, add 25  $\mu$ L into each well instead, for a total volume of 50  $\mu$ L/well.
3. Incubate the reaction at room temperature for 5 to 10 minutes, protected from light.
4. Monitor the fluorescence increase with a fluorescence microplate reader at Ex/Em = 490/525 nm (cut off at 515 nm).

#### EXAMPLE DATA ANALYSIS AND FIGURES



**Figure 1.** Lithium dose response was measured with Amplite® Fluorimetric Lithium Ion Kit in a 96-well solid black plate.

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