

**Metal Fluor™ Zn-520, AM**

Catalog number: 21263

Unit size: 1 mg

Component	Storage	Amount (Cat No. 21263)
Metal Fluor™ Zn-520, AM	Freeze (< -15 °C), Minimize light exposure	1 vial (1 mg)

**OVERVIEW**

Metal Fluor™ Zn-520 is designed for detection of higher zinc ion concentrations that are present in synaptic vesicles and released in response to electrical stimulation or excitotoxic agonists (0.05-50  $\mu$ M) with minimal interfering calcium sensitivity. Metal Fluor™ Zn-520 has shown great fluorescence enhancement upon binding zinc ion (>250 folds). This cell permeant AM-ester is useful for detecting intracellular zinc ion levels.

**AT A GLANCE**
**Protocol summary**

1. Grow cells as desired
2. Prepare and add Metal Fluor™ Zn-520, AM working solution to samples
3. Incubate samples at 37 °C for 15 to 45 minutes
4. Monitor the fluorescence intensity using flow cytometer with 530/30 nm filter (FITC channel) or using fluorescence microscopy with FITC filter set

**KEY PARAMETERS**
**Flow cytometer**

Emission	530/30 nm filter
Excitation	488 nm laser
Instrument specification(s)	FITC channel

**Fluorescence microscope**

Emission	FITC filter set
Excitation	FITC filter set
Recommended plate	Black wall/clear bottom

**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

**Metal Fluor™ Zn-520, AM stock solution**

1. Reconstitute Metal Fluor™ Zn-520, AM by adding an appropriate volume of DMSO directly into the vial to achieve a final stock concentration of 2–5 mM. Mix thoroughly.

**PREPARATION OF WORKING SOLUTION**
**Metal Fluor™ Zn-520, AM working solution**

1. Prepare Metal Fluor™ Zn-520, AM working solution at a final concentration of 5–50  $\mu$ M in the buffer of choice.

**Note:** Metal Fluor™ Zn-520, AM working solution should be used promptly.

**Note:** The concentration of the Metal Fluor™ Zn-520, AM should

be optimized for different cell types and conditions.

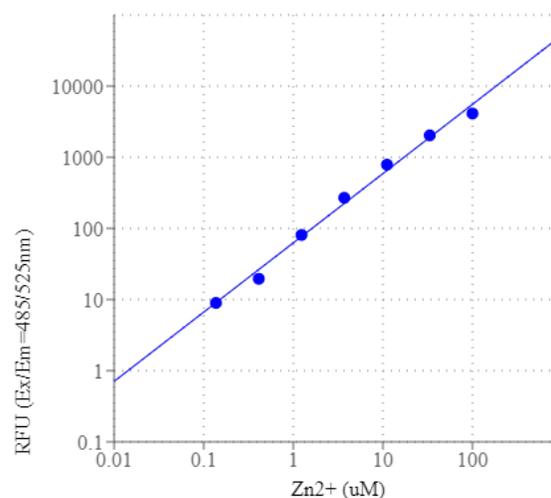
**SAMPLE EXPERIMENTAL PROTOCOL**

The following protocol can be used as a guideline and should be optimized according to the needs.

1. Grow cells as desired.
2. Remove the treatment and wash the cells with buffer of your choice such as DPBS.
3. Add Metal Fluor™ Zn-520, AM working solution and incubate the samples for 15-45 minutes at 37 °C incubator.

**Note:** Optimal time for incubation needs to be determined carefully.

4. Remove the working solution and wash cells with buffer of your choice.
5. Resuspend cells in buffer of your choice.
6. Add stimulant to stimulate the cells and monitor the fluorescence intensity with flow cytometer using 530/30 nm filter (FITC channel) or fluorescence microscope with FITC filter set.

**EXAMPLE DATA ANALYSIS AND FIGURES**


**Figure 1.** Zinc Chloride dose response was measured on a 96-well black plate with the Amplitude® Fluorimetric Zinc Quantitation Kit. As low as 0.2  $\mu$ M Zn<sup>2+</sup> could be detected with 5 min incubation time.

**DISCLAIMER**

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