

## Dyes and Stains

### Wheat Germ Agglutinin (WGA) iFluor™ Conjugates

Fluorescent dye conjugates for staining cell membranes of gram-positive bacteria, mammalian cells, and yeast bud scars

Catalog #100-0816	1000 µg
Catalog #100-0817	1000 µg
Catalog #100-0818	1000 µg



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## Product Description

Wheat germ agglutinins (WGAs) are lectins—or carbohydrate-binding proteins—that have a high affinity for N-acetyl-D-glucosamine and sialic acid present in cell membranes. WGA iFluor™ conjugates are used to stain cell membranes and fibrotic scar tissue for fluorescence imaging and analysis.

## Product Information

PRODUCT NAME	CATALOG #	SIZE	STORAGE	SHELF LIFE
Wheat Germ Agglutinin (WGA), iFluor™ 488	100-0816	1000 µg	Store at -20°C. Protect product from prolonged exposure to light.	Stable until expiry date (EXP) on label.
Wheat Germ Agglutinin (WGA), iFluor™ 555	100-0817	1000 µg	Store at -20°C. Protect product from prolonged exposure to light.	Stable until expiry date (EXP) on label.
Wheat Germ Agglutinin (WGA), iFluor™ 647	100-0818	1000 µg	Store at -20°C. Protect product from prolonged exposure to light.	Stable until expiry date (EXP) on label.

## Spectral Properties

PRODUCT	EXCITATION WAVELENGTH (nm)	EMISSION WAVELENGTH (nm)	EXTINCTION COEFFICIENT (cm <sup>-1</sup> M <sup>-1</sup> )*	QUANTUM YIELD*	CORRECTION FACTOR (280 nm)	CORRECTION FACTOR (260 nm)
WGA, iFluor™ 488	491	516	75,000	0.9	0.11	0.21
WGA, iFluor™ 555	557	570	100,000	0.64	0.14	0.23
WGA, iFluor™ 647	656	670	250,000	0.24	0.3	0.3

\*Measured with aqueous buffer (pH 7.2).

## Directions for Use

Please read the entire protocol before proceeding. The following protocol is for staining cells in a black-wall/clear-bottom 96-well plate. If using other cultureware, adjust volumes accordingly.

### Preparation of WGA iFluor™ Stock and Working Solutions

1. Warm the Wheat Germ Agglutinin (WGA), iFluor™ vial to room temperature (15 - 25°C) and centrifuge briefly before opening.
2. To prepare a 2 mg/mL (200X) stock solution, add 500 µL of ddH<sub>2</sub>O to the WGA iFluor™ vial. Mix thoroughly.  
NOTE: If not used immediately, aliquot and store at -20°C. After thawing aliquots, use immediately; do not re-freeze.
3. To prepare a WGA iFluor™ working solution, add 5 µL of stock solution to 1 mL of Hanks' Balanced Salt Solution with 20 mM HEPES (HHBS). Use the working solution immediately; do not store.

NOTE: The optimal concentration of working solution may need to be determined for different cell lines; for live cells, the recommended starting concentration is 5 - 10 µg/mL.

### Staining Cells

Refer to section A for staining live cells or section B for staining fixed cells.

#### A. LIVE CELLS

1. Wash cells 2X with HHBS.
2. Add 100 µL/well of WGA iFluor™ working solution to the cells. Incubate at 37°C for 10 - 30 minutes; protect from light.
3. Remove WGA iFluor™ working solution and wash cells 2X with HHBS.

#### B. FIXED CELLS

1. Add 4% formaldehyde in phosphate-buffered saline (PBS) to cells and incubate at room temperature (15 - 25°C) for 10 - 30 minutes.  
NOTE: For fixed cell membrane staining, it is recommended to stain without a permeabilization step. Permeabilization after fixation can result in staining of intracellular compartments such as Golgi apparatus and endoplasmic reticulum.
2. Remove fixative and add 100 µL/well of WGA iFluor™ working solution to fixed cells. Incubate at room temperature for 10 - 30 minutes; protect from light.
3. Wash cells 2X with HHBS.

### Imaging Stained Cells

Observe stained cells using a fluorescence microscope with the appropriate filter set, as follows:

- iFluor™ 488 conjugate: FITC
- iFluor™ 555 conjugate: Cy3/TRITC
- iFluor™ 647 conjugate: Cy5

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