

## XFD532-Wheat Germ Agglutinin (WGA) Conjugate

Catalog number: 25502  
Unit size: 1 mg

Component	Storage	Amount (Cat No. 25502)
XFD532-Wheat Germ Agglutinin (WGA) Conjugate	Freeze (< -15 °C), Minimize light exposure	1 mg

### OVERVIEW

XFD532, manufactured by AAT Bioquest, has a similar chemical structure to Alexa Fluor® 532, a trademark of Thermo Fisher. Wheat germ agglutinin (WGA) is a well-researched lectin known for its valuable biological applications. Due to its ability to bind to glycoconjugates, WGA derivatives, and conjugates are widely used to label cell membranes and fibrotic scar tissue for fluorescence imaging and analysis. WGA specifically targets sequences of  $\beta$ -1,4-GlcNAc-linked residues known as chitodextrins. Each monomer possesses two identical, non-interacting binding sites that complement 3 or 4  $\beta$ -1,4-GlcNAc units. Among the tested monosaccharides, only GlcNAc exhibits binding to WGA, while ManNAc does not bind, and GalNAc demonstrates weak binding. XFD532 WGA conjugate, like its counterpart Alexa Fluor® 532 WGA conjugate, emits a bright yellow fluorescence and is useful in a variety of applications, including immunofluorescence (IF), immunohistochemistry (IHC), and flow cytometry (FC).

### KEY PARAMETERS

#### Fluorescence microscope

Emission	Cy3/TRITC filter set
Excitation	Cy3/TRITC 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*

#### XFD532-Wheat Germ Agglutinin (WGA) Conjugate stock solution (200X)

Add 500  $\mu$ L of ddH<sub>2</sub>O into the powder form to make a 2 mg/mL stock solution.

**Note:** The reconstituted conjugate solution can be stored at 2-8 °C for short-term storage or at -20 °C for long-term storage.

### PREPARATION OF WORKING SOLUTION

#### XFD532-Wheat Germ Agglutinin (WGA) Conjugate working solution (1X)

Add 5  $\mu$ L of 200X WGA conjugate solution to 1 mL HHBS Buffer.

**Note:** The optimized staining concentration may be different with different cell lines. The recommended starting concentration is 5-10  $\mu$ g/mL for live cells.

### SAMPLE EXPERIMENTAL PROTOCOL

Warm the vial to room temperature centrifuge briefly before opening. Staining protocols vary with applications. Appropriate dilution of conjugates should be determined experimentally.

#### Live Cells Stain

1. Wash cells twice with a HHBS buffer.
2. Add 100  $\mu$ L XFD532-WGA working solution.
3. Incubate cells with working solution for 10-30 minutes at 37 °C.
4. Wash the cells twice with HHBS buffer.
5. Image cells on a fluorescence microscope using Cy3/TRITC filter set.

#### Fixed Cells Stain

WGA conjugates can be also used to stain fixed cells.

1. Fix cells with 4% formaldehyde in PBS.

**Note:** For fixed cell membrane staining, it is recommended to stain without the permeabilization step. A permeabilization step after fixation can facilitate staining intracellular compartments such as Golgi and Endoplasmic Reticulum (ER) structures.

2. Add 100  $\mu$ L XFD532-WGA working solution.
3. Incubate cells with working solution for 10-30 minutes at room temperature.
4. Wash the cells twice with HHBS buffer.
5. Image cells on a fluorescence microscope using Cy3/TRITC filter set.

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