

Hyaluronic Acid, BODIPY® FL Conjugate

Table 1. Contents and Storage Information.

Material	Amount	Concentration	Storage	Stability
Hyaluronic Acid, BODIPY® FL conjugate *	500 µg, lyophilized	NA	<ul style="list-style-type: none"> • ≤-20°C • Desiccate • Protect from light 	When stored as directed, product will remain stable for 6 months.

* The molecular weight of the conjugate is ~200,000, as determined by gel permeation chromatography.

Spectral Data: 504/511 nm

Introduction

Hyaluronic acid (HA), a major glycosaminoglycan (GAG) constituent of the extracellular matrix, participates in the adhesion, motility, proliferation, and differentiation of cells, and is involved in inflammation and cancer.¹ The numerous biological effects of this relatively simple unbranched polysaccharide are exerted through interactions with proteins including the cell surface receptors CD44 and RHAMM (receptor for HA-mediated motility).^{2,3} CD44, a 90,000 molecular weight transmembrane adhesion glycoprotein, is frequently overexpressed by tumor cells.^{4,5} Molecular Probes offers a BODIPY® FL dye-labeled HA derivative with many potential applications for direct detection of HA-protein interactions.⁶ Receptor-mediated internalization of a similar BODIPY® FL-HA conjugate by human ovarian cancer cells has been visualized by confocal microscopy.⁷

Before You Begin

Materials Required but not Provided

- Culture medium
- Balanced salt solution (e.g., Dulbecco's PBS)
- 3% paraformaldehyde
- FITC filter set (485 nm excitation filter, 505 nm dichroic beamsplitter, 530 nm emission filter, or similar specifications)
- ProLong® Gold antifade reagent (P36930)

Preparing the BODIPY® FL–HA

Stock Solution

Stock solutions of BODIPY® FL–HA at 1–5 mg/mL can be prepared by dissolving the contents of the vial (500 µg) in 500–100 µL of buffer or distilled water. Stock solutions may be stored at either 4°C or –20°C, protected from light.

Note: The degree of labeling has been optimized to maintain water solubility and biological activity of the conjugate.

Experimental Protocol

This protocol for visualization of receptor-mediated uptake of BODIPY® FL–HA is adapted from that described by Luo and Prestwich.⁷ It is provided as an initial guideline that may require further optimization for other applications.

- 1.1 Dilute BODIPY® FL–HA stock solution to a concentration of 100 µg/mL in culture medium. Incubate cells in this staining medium for 2–24 hours.
- 1.2 Wash cells three times with balanced salt solution (e.g., Dulbecco's PBS).
- 1.3 Fix cells with 3% paraformaldehyde for 10 minutes and wash again with Dulbecco's PBS.
- 1.4 Mount cells in ProLong® Gold antifade reagent and examine by fluorescence microscopy using an FITC filter set.

References

1. Curr Opin Cell Biol 12, 581 (2000); 2. Proteins 39, 103 (2000); 3. J Biol Chem 277, 4585 (2002); 4. Bioconjug Chem 10, 755 (1999); 5. Clin Cancer Res 1, 333 (1995); 6. J Biol Chem 277, 4585 (2002); 7. Bioconjug Chem 12, 1085 (2001).

Product List

Current prices may be obtained from our website or from our Customer Service Department.

Cat #	Product Name	Unit Size
H23379	hyaluronic acid, BODIPY® FL conjugate (BODIPY® FL hyaluronic acid).....	500 µg
P36930	ProLong® Gold antifade reagent	10 mL

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