

## Product Data Sheet

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Product Name: Human Serum Albumin-FITC

Cat. No.: GC81671

### Chemical Properties

Cas. No.

Formula

M.Wt

Solubility

Storage Store at -20°C, protect from light

General tips For obtaining a higher solubility, please warm the tube at 37 °C and shake it in the ultrasonic bath for a while. Stock solution can be stored below -20°C for several months.

Shipping Condition Evaluation sample solution : ship with blue ice All other available size: ship with RT, or blue ice upon request.

Structure

### Human Serum Albumin-FITC

### Background

Human Serum Albumin-FITC (HSA-FITC) is a green fluorescent probe prepared by covalent conjugation of FITC with human serum albumin (HSA), with excitation/emission wavelengths of 488/505-550 nm. Human Serum Albumin-FITC (HSA-FITC) can be used to track nanotube conjugates taken up via caveolae-mediated endocytosis initiated by Gp60 receptor binding, and can also non-covalently adsorb to oxidized multi-walled carbon nanotubes. Human Serum Albumin-FITC (HSA-FITC) is widely applied in macrophage imaging, scavenger receptor targeting, and hepatoma cell imaging [1] [2].

**Caution: Product has not been fully validated for medical applications. For research use only.**

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In Vitro, Guidelines (The following recommended experimental protocol is for guidance only and needs to be adjusted according to your specific requirements)

1. Stock Solution Preparation 1.1 Solvent: Anhydrous dimethyl sulfoxide (DMSO). 1.2 Recommended concentration: 10 mg/mL. 2. Working Solution Preparation 2.1 Diluent: Carbonate/bicarbonate buffer (pH 9) or 20 mM sodium buffer (pH 8.5). 2.2 Working concentration: 0.1  $\mu$ M, 1  $\mu$ M, 5  $\mu$ M or 1.25 mg/mL. 2.3 Notes: Adjust the working solution concentration as needed; prepare and use immediately. 3. Staining Procedure 3.1 Sample type description: 3.1.1 Adherent cells<sup>[1][2]</sup>: THP-1 macrophages, THP-1 foam cells, polarized primary human macrophages (M0, M1, M2), human carotid artery smooth muscle cells, human umbilical vein endothelial cells, HepG2 cells, immortalized hepatic epithelial cells (CRL-4020). 3.1.2 Notes on cell detachment: After staining and fixation, scrape cells from plastic culture plates using a scraper<sup>[1]</sup>; or detach and transfer cells using trypsin digestion<sup>[2]</sup>. 3.2 Incubation conditions: 3.2.1 Incubate with 0.1  $\mu$ M, 1  $\mu$ M, or 5  $\mu$ M HSA-FITC for 1 h, 3 h (at 37°C in a 5% CO<sub>2</sub>, 95% air humidified atmosphere), or for up to 24 h<sup>[1]</sup>. 3.2.2 Incubate with 1-50 mg/L HSA-FITC-MWCNTs for durations ranging from 1 minute to 24 h, depending on experimental design<sup>[2]</sup>. 3.3 Washing steps: 3.3.1 After live/dead staining, wash with 1% BSA first, followed by PBS<sup>[1]</sup>. 3.3.2 After probe incubation and before fixation, wash with PBS<sup>[1]</sup>. 3.3.3 Wash cells 3 times with 1 $\times$  PBS<sup>[2]</sup>. 4. Control Setup 4.1 Negative controls: Unmodified HSA-FITC, non-radioactively labeled probe blocking, IgG1 isotype control for SR-A1 blocking<sup>[1]</sup>. 4.2 Competition controls: Modified LDL particles (MDA-LDL, oxLDL)<sup>[1]</sup>. 4.3 Inhibition controls: Dextran sulfate, cytochalasin D<sup>[1]</sup>. 4.4 Untreated cell control, non-functionalized MWCNTs-treated cell control, and background control for apoptosis detection<sup>[2]</sup>. 4.5 Positive control (for validating the experimental system)<sup>[2]</sup>; blank control (to exclude reagent fluorescence interference)<sup>[2]</sup>. 5. Detection and Analysis 5.1 Instrument types: Flow cytometer, confocal microscope, real-time imaging microscope, confocal laser scanning microscope. 5.2 Excitation/emission wavelengths: For FITC, excitation at 488 nm and emission at 525 nm; or excitation at 488 nm and emission at BP 505-550 nm. 5.3 Result analysis: 5.3.1 Changes in fluorescence intensity: Modified HSA-FITC (Mal-HSA-FITC, Aco-HSA-FITC) shows significantly higher fluorescence intensity in macrophages than unmodified HSA-FITC, and the uptake increases with prolonged incubation time and increased probe concentration<sup>[1]</sup>; Green fluorescence is observed in HepG2 cells that endocytose HSA-FITC-MWCNTs, whereas only minimal or no green fluorescence is detected in CRL-4020 cells under the same conditions<sup>[2]</sup>. 5.3.2 Fluorescence localization: Modified HSA-FITC localizes to granular cytoplasmic structures and colocalizes with the

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lysosome marker LAMP-2<sup>[1]</sup>; HSA-FITC-MWCNTs localize to punctate intracellular structures and plasma membrane vesicles in HepG2 cells, and colocalize with Cy3-labeled anti-Gp60 antibody and anti-caveolin-1 antibody<sup>[2]</sup>. 5.3.3 Fluorescence visualization: Green fluorescence emitted by FITC is observable<sup>[1]</sup>; Green fluorescence from HSA-FITC, red fluorescence from Cy3-labeled antibodies, and blue fluorescence from DAPI (nuclear dye) are observable, and the colocalized regions of green and red fluorescence appear as yellow signals in merged images<sup>[2]</sup>.

### References:

- [1]. Ahmed M, et al. Human serum albumin-based probes for molecular targeting of macrophage scavenger receptors. International journal of nanomedicine. 2019;14:3723-3741.
- [2]. Iancu C, et al. Enhanced laser thermal ablation for the in vitro treatment of liver cancer by specific delivery of multiwalled carbon nanotubes functionalized with human serum albumin. International journal of nanomedicine. 2011 Jan 17;6:129-41.

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