Dyes and Stains

CFSE

Cell proliferation and tracking dye

Catalog # 75003

10 mg

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Product Description

CFSE (carboxyfluorescein diacetate succinimidyl ester or CFDA-SE) is a stable, non-fluorescent, cell-permeable derivative of fluorescein containing two acetate groups and a succinimidyl ester functional group. Upon diffusion into the cell, intracellular esterases cleave the acetate groups to generate a highly fluorescent (green) dye that is impermeant to the cell membrane, and covalent binding of the succinimidyl ester to free amine groups forms a stable intracellular label. At appropriate concentrations, CFSE is not toxic to cells and as the cells divide, CFSE is partitioned approximately equally between the progeny so that cell division can be followed as a successive halving of the fluorescence intensity through multiple generational divisions. CFSE is most widely used for cell proliferation and motility assays, and in vivo cell tracking experiments (ex vivo labeling of cells for adoptive transfer). CFSE-labeled cells can be detected with any instrument/filter set compatible with fluorescein detection.

Chemical Name:	3',6'-bis(acetyloxy)-3-oxo-2,5-dioxo-1-pyrrolidinyl ester-spiro[isobenzofuran-1(3H),9'-[9H]xanthene]-ar-carboxylic acid
Alternative Names:	5(6)-Carboxyfluorescein succinimidyl ester; 6-{[(2,5-Dioxo-1-pyrrolidinyl)oxy]carbonyl}-3-oxo-3H-spiro[2-benzofuran-1,9'-xanthene]-3',6'-diyl diacetate; carboxyfluorescein diacetate succinimidyl ester; CFDA-SE
CAS Number:	150347-59-4
Chemical Formula:	$C_{29}H_{19}NO_{11}$
Molecular Weight:	557.5 g/mol
Excitation Wavelength:	492 nm (esterase-cleaved fluorescent derivative)
Emission Wavelength:	517 nm (esterase-cleaved fluorescent derivative)

Properties

Storage:	Store at -20°C.
Shelf Life:	Product stable until expiry date (EXP) on label. Protect product from prolonged exposure to light.
Format/Formulation:	A crystalline solid

Applications

Verified:	FA, FC, Fluorescence microscopy
Reported:	FA, FC, Fluorescence microscopy, Histochemistry, ICC, IF, In vivo cell tracking
Special Applications:	This product has been verified for analyzing cells isolated with EasySep™ kits, including EasySep™ Human T Cell Enrichment Kit (Catalog #19051) and EasySep™ Mouse CD11c Positive Selection Kit II (Catalog #18780), and for analyzing cells cultured in several media, including T cells cultured in ImmunoCult™-XF T Cell Expansion Medium (Catalog #10981).

Abbreviations: CellSep: Cell separation; ChIP: Chromatin immunoprecipitation; FA: Functional assay; FC: Flow cytometry; ICC: Immunocytochemistry; IF: Immunofluorescence microscopy; IHC: Immunohistochemistry; IP: Immunoprecipitation; RIA: Radioimmunoassay; WB: Western blotting

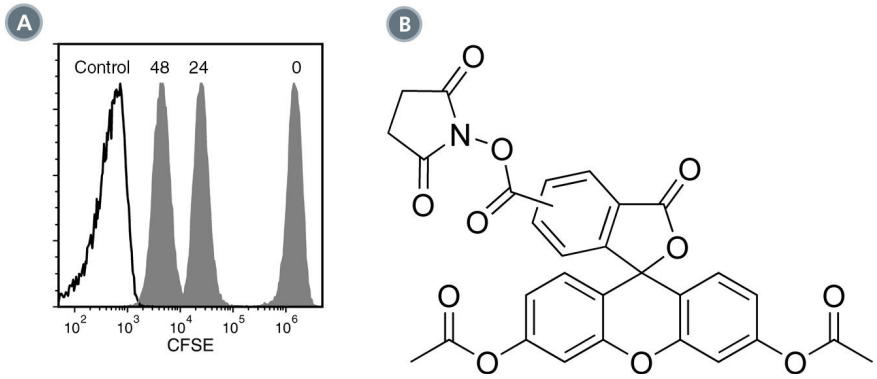
Handling/Directions for Use

PREPARATION
A stock solution may be made by dissolving the CFSE in the solvent of choice. CFSE is soluble in organic solvents. Guidelines for the solubility of CFSE are as follows:
· DMSO ≤ 20 mg/mL
· Dimethyl formamide ≤ 30 mg/mL
CFSE is sparingly soluble in aqueous buffers. For maximum solubility in aqueous buffers, CFSE should first be dissolved in an organic solvent and then diluted with the aqueous buffer of choice. If performing biological experiments, ensure the residual amount of organic solvent is insignificant, as organic solvents may have physiological effects at low concentrations.

Wherever possible, prepare and use the stock solutions on the same day. Protect stock solutions from prolonged exposure to light. If stock solutions must be made in advance, aliquot and store in tightly sealed vials at -20°C, protected from prolonged exposure to light. Generally these will be stable for up to 1 month.

FLOW CYTOMETRY (in vitro cell proliferation assay)
It is recommended to use CFSE at a final concentration of 0.5 - 10 µM.
1 Resuspend the cells at 1 x 10^7 - 1 x 10^8 cells/mL in phosphate-buffered saline (PBS).
2. Add an equal volume of CFSE as a 2X working stock to give a final concentration of 0.5 - 10 µM.
   Note: The dye should be titrated for optimal performance for each cell type and application.
3. Incubate cells with the dye for 5 - 10 minutes in the dark at 37°C or room temperature (15 - 25°C).
4. Add an equal volume of culture medium containing 10% fetal bovine serum (FBS) and incubate for 5 minutes to quench the staining.
5. Pellet the cells by centrifugation and wash once with an equal volume of culture medium.
Cells are now fluorescently labeled and ready to be cultured or analyzed.

Data/Structure



(A) Flow cytometry analysis of Sp2/0 mouse myeloma cells labeled with CFSE and analyzed by flow cytometry after being cultured for 0, 24, and 48 hours (filled histograms). Solid line histogram (control) shows unlabeled cells analyzed after 48 hours of cell culture.
(B) Chemical structure of CFSE.

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