

CFSE Cell Proliferation Kit

Table 1 Contents and storage

Material	Amount	Concentration	Storage	Stability
CFSE	20 vials 25mg	5 mM stock solution in anhydrous DMSO	<ul style="list-style-type: none"> • $\leq -20^{\circ}\text{C}$ • Desiccate • Protect from light 	

Spectral characteristic of the fluorescent probe: **Ex~494, Em~521**

Introduction

5,6-Carboxyfluorescein diacetate succinimidyl ester (CFSE, Fig. 1) is widely used for fluorescently labeling of living cells. CFSE is readily deacetylated by intracellular esterases, leading to accumulation of 5,6-carboxyfluorescein succinimidyl ester inside cells and subsequent covalent labeling of lysine residues of the intracellular proteins. The fluorescent labeling allows stable tracking of cells.

During cell division and proliferation, the fluorescence intensity of CFSE-labeled cells decreases with the division of cells, owing to fluorescence distributed to the two daughter cells. This characteristic allows detection of cell proliferation, cell cycle estimation and cell division.

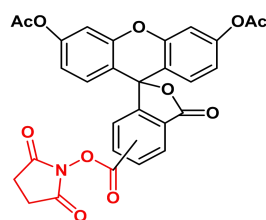


Fig. 1 Chemical structures of CFSE

Before opening, allow the vial to warm to room temperature and then briefly centrifuge the vial in a micro centrifuge to deposit the DMSO solution at the bottom of the vial.

Guidelines for Use

The concentration of probe for optimal staining will vary depending on the application. Here we suggest some initial conditions to use as a guideline. The preparation of different concentrations of CFSE

can refer to Table 2. The staining conditions may need to be modified depending on specific cell lines.

	0.1 mg	0.5 mg	1 mg	5 mg
1 mM	179.39 μL	896.93 μL	1.794 mL	8.969 mL
5 mM	35.88 μL	179.39 μL	358.77 μL	1.794 mL
10 mM	17.94 μL	89.69 μL	179.39 μL	896.93 μL

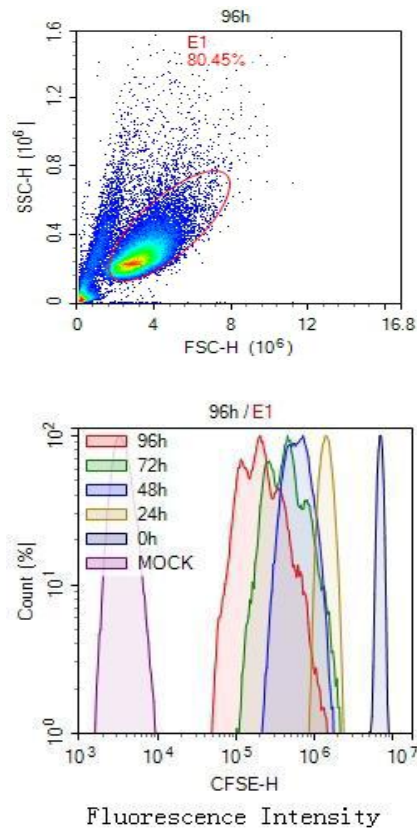
Table 2. Volume of DMSO needed to reconstitute

CFSE Labeling Procedure:

1. Prepare CFSE at a concentration of 5 mM
2. Prepare a 5 μM working solution by diluting 1 μL of 5 mM CFSE stock solution in 1 mL PBS for every 1 mL of cell suspension (or at an optimal working concentration as determined by titration).
3. Spin down and resuspend cells at 10-100 x 10⁶ cells/mL in the CFSE working solution.
4. Incubate cells for 20 minutes at room temperature or 37°C and keep protected from light.
5. Quench the staining by adding 5 times the original staining volume of cell culture medium containing 10% FBS.
6. Pellet cells and resuspend in pre-warmed cell culture medium.
7. Incubate cells for 10 minutes.
8. After incubation, CFSE labeled cells are ready for downstream applications or analysis.



Product Data



Suspension cells were stained with CFSE Cell Division Tracking Kit, cells were harvested and the CFSE fluorescent staining was analyzed by flow cytometry.

Fluorescence spectrum

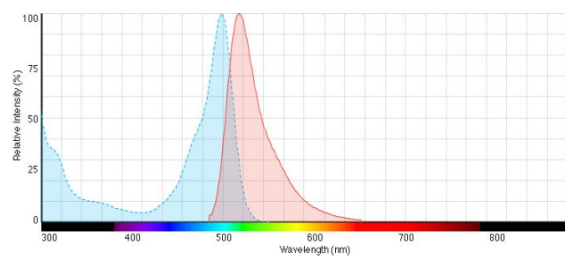


Fig 2 Fluorescence Ex/Em spectra of CFSE

References

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