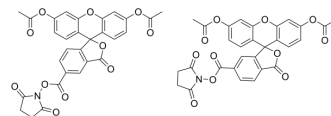


## CFDA-SE

<b>Cat. No.:</b>	HY-D0938
<b>CAS No.:</b>	150347-59-4
<b>Molecular Formula:</b>	C <sub>58</sub> H <sub>38</sub> N <sub>2</sub> O <sub>22</sub>
<b>Molecular Weight:</b>	557.46
<b>Target:</b>	Fluorescent Dye
<b>Pathway:</b>	Others
<b>Storage:</b>	-20°C, sealed storage, away from moisture and light * In solvent : -80°C, 6 months; -20°C, 1 month (sealed storage, away from moisture and light)



## SOLVENT & SOLUBILITY

<b>In Vitro</b>	DMSO : 50 mg/mL (89.69 mM; Need ultrasonic)					
	<b>Preparing Stock Solutions</b>	<b>Solvent</b>	<b>Mass</b>	<b>1 mg</b>	<b>5 mg</b>	<b>10 mg</b>
		<b>Concentration</b>				
		<b>1 mM</b>		1.7939 mL	8.9693 mL	17.9385 mL
		<b>5 mM</b>		0.3588 mL	1.7939 mL	3.5877 mL
<b>10 mM</b>		0.1794 mL	0.8969 mL	1.7939 mL		
Please refer to the solubility information to select the appropriate solvent.						
<b>In Vivo</b>	<ol style="list-style-type: none"> <li>Add each solvent one by one: 10% DMSO &gt;&gt; 40% PEG300 &gt;&gt; 5% Tween-80 &gt;&gt; 45% saline Solubility: ≥ 2.08 mg/mL (3.73 mM); Clear solution</li> <li>Add each solvent one by one: 10% DMSO &gt;&gt; 90% corn oil Solubility: ≥ 2.08 mg/mL (3.73 mM); Clear solution</li> </ol>					

## BIOLOGICAL ACTIVITY

<b>Description</b>	CFSE is a fluorescent dye that can penetrate the cell membrane. It can react with the free amine group in the cytoskeleton protein inside the cell, and finally form a protein complex with fluorescence. After entering the cell, CFSE locates in the cell membrane, cytoplasm and nucleus, and the fluorescence staining is strongest in the nucleus <sup>[1]</sup> . CFSE dye can be uniformly inherited by the cells with cell division and proliferation, and its attenuation is proportional to the number of cell divisions. This phenomenon can be detected and analyzed by flow cytometry under the excitation light of 488nm, and can be used to detect the proliferation of cells <sup>[1]</sup> .
<b>In Vitro</b>	Preparation of CFSE working solution 1.1 Preparation of the stock solution Dissolve 1 mg of CFSE in 0.1794 mL of DMSO to obtain 10 mM of CFSE. Note: It is recommended to store the stock solution at -20 °C or -80 °C away from light and avoid repetitive freeze-thaw cycles.

### 1.2 Preparation of CFSE working solution

Dilute the stock solution in serum-free cell culture medium or PBS to obtain 5-10  $\mu\text{M}$  of CFSE working solution.

Note: Please adjust the concentration of CFSE working solution according to the actual situation.

### Cell staining

2.1 For suspension cells: Centrifuge at 1000 g at 4 $\times$  for 3-5 minutes and then discard the supernatant. Wash twice with PBS, 5 minutes each time.

For adherent cells: Discard the cell culture medium, and add trypsin to dissociate cells to make a single-cell suspension. Centrifuge at 1000 g at 4 $\times$  for 3-5 minutes and then discard the supernatant. Wash twice with PBS, 5 minutes each time.

2.2 Add 1 mL of CFSE working solution, and then incubate at room temperature for 30 minutes.

2.3 Centrifuge at 400 g at 4 $\times$  for 3-4 minutes and then discard the supernatant.

2.4 Wash twice with PBS, 5 minutes each time.

2.5 Resuspend cells with serum-free cell culture medium or PBS, and then detect by fluorescence microscope or flow cytometer.

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

## PROTOCOL

### Cell Assay <sup>[2]</sup>

Single-cell suspensions of splenocytes (or PBMC) are stained with 1  $\mu\text{M}$  CFSE in PBS for 9 min at 37°C, combined with 20 mL of 10% FBS RPMI-1640 medium at RT for 2 min, centrifuged, washed, and counted.  $2 \times 10^6$  CFSE-labeling splenocytes are added to the top of the slices in 24-well membrane culture insert<sup>[2]</sup>.

MCE has not independently confirmed the accuracy of these methods. They are for reference only.

## CUSTOMER VALIDATION

- Cell Res. 2023 May 4.
- Nature Cancer. 222-234 (2020).
- Autophagy. 2021 Nov;17(11):3592-3606.
- Proc Natl Acad Sci U S A. 2022 Aug 9;119(32):e2201899119.
- Theranostics. 2021 Feb 20;11(9):4251-4261.

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## REFERENCES

[1]. Banks HT, et al. Quantifying CFSE Label Decay in Flow Cytometry Data. Appl Math Lett. 2013 May 1;26(5):571-577.

[2]. Cui J, et al. Prostaglandin E3 attenuates macrophage-associated inflammation and prostate tumour growth by modulating polarization. J Cell Mol Med. 2021 Jun;25(12):5586-5601.

[3]. A Bruce Lyons, et al. Flow cytometric analysis of cell division by dilution of CFSE and related dyes. Curr Protoc Cytom. 2013;Chapter 9:Unit9.11.

[4]. Xiuyun Jiang et al. Long-lived pancreatic ductal adenocarcinoma slice cultures enable precise study of the immune microenvironment. Oncoimmunology. 2017; 6(7): e1333210.

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

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