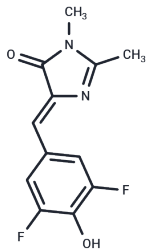
Data Sheet (Cat.No.T15106)

TargetMol

DFHBI

Chemical Properties



CAS No. : 1241390-29-3

Formula: C12H10F2N2O2

Molecular Weight: 252.22

Storage: Keep away from direct sunlight
Powder: -20°C for 3 years | In solvent: -80°C for 1 year
Actual storage temperature shall be subject to the COA.

Biological Description

Description	DFHBI, a small molecule resembling the chromophore of green fluorescent protein (GFP), is essentially nonfluorescent when unbound. The Spinach-DFHBI complex, however, exhibits bright fluorescence both in vitro and in living cells.
Targets(IC50)	Others
In vitro	Spinach and Spinach2 bind to DFHBI have fluorescence excitation maxima of 447 nm and peak fluorescence emission of 501 nm[1] and they are RNA aptamers that can be used for the genetic encoding of fluorescent RNA. Spinach is a 98-nt-long RNA aptamer that binds to and switches on the fluorescence of DFHBI. Both Spinach and DFHBI are essentially nonfluorescent when unbound, whereas the Spinach-DFHBI complex is brightly fluorescent both in vitro and in living cells. Spinach2 binds and activates the fluorescence of DFHBI, allowing the dynamic localizations of Spinach2-tagged RNAs to be imaged in live cells. The spectral properties of Spinach2 are limited by DFHBI, which produces fluorescence that is bluish-green and is not optimized for filters commonly used in fluorescence microscopes. DFHBI should be shielded from light. All stock solutions of DFHBI should be maintained in dark tubes or wrapped in foil. Plates containing cultures incubated with DFHBI should be kept in the dark by using a foil overwrap[2].
Cell Research	I. RNA labeling and imaging 1. Prepare Spinach RNA: synthesize target RNA containing Spinach aptamer sequence by in vitro transcription or RNA synthesis method. 2. Dissolve DFHBI: dissolve DFHBI in DMSO or water to make a 1-10 mM stock solution, and dilute to 1-50 μM range when used. 3. Complex formation: mix Spinach RNA with DFHBI and incubate under appropriate buffer conditions (such as Tris-HCl, $MgCl_2$, etc.) to form a highly fluorescent complex. 3. Imaging: observe using a fluorescence microscope, with an excitation wavelength of 470-490 nm and an emission wavelength of 510-530 nm. II. Live cell experiment 1. Spinach RNA expression: transfect cells with a plasmid containing Spinach sequence, or express target RNA through an in vivo RNA synthesis system. 2. Add DFHBI: add DFHBI (final concentration is usually 10-50 μM) to the cell culture medium and incubate for 10-30 minutes. 3. Fluorescence detection: Observe under a fluorescence microscope or fluorescence

Cell Research	imager to monitor the localization and dynamics of RNA. 4 Fluorescence detection and quantification 1) Fluorescence measurement: Use a fluorescence spectrophotometer to quantify the fluorescence intensity of the Spinach-DFHBI complex to analyze the amount or dynamic changes of RNA. 2) Optimization conditions: Optimize the buffer, ion concentration (especially Mg^{2+} concentration) and pH in the experiment to obtain the best fluorescence signal. The above information is based on published literature. Experimental procedures should be appropriately modified to meet specific research demands.

Solubility Information

Solubility	DMSO: 85 mg/mL (337.01 mM),Sonication is recommended. (< 1 mg/ml refers to the product slightly soluble or insoluble)
In vivo Formulation	10% DMSO+40% PEG300+5% Tween-80+45% Saline: 1 mg/mL (3.96 mM),Sonication is recommended. *Please add the solvents sequentially, clarifying the solution as much as possible before adding the next one. Dissolve by heating and/or sonication if necessary. Working solution is recommended to be prepared and used immediately. The formulation provided above is for reference purposes only. In vivo formulations may vary and should be modified based on specific experimental conditions.*

Preparing Stock Solutions

	1mg	5mg	10mg
1 mM	3.9648 mL	19.824 mL	39.6479 mL
5 mM	0.793 mL	3.9648 mL	7.9296 mL
10 mM	0.3965 mL	1.9824 mL	3.9648 mL
50 mM	0.0793 mL	0.3965 mL	0.793 mL

Please select the appropriate solvent to prepare the stock solution, according to the solubility of the product in different solvents. Please use it as soon as possible.

Note: The dilution table applies only to solid products. For liquid products, please calculate the stock solution based on the stated concentration and/or density.

Reference

Qin W, et al. High-throughput iSpinach fluorescent aptamer-based real-time monitoring of in vitro transcription. Bioresour Bioprocess. 2022 Oct 27;9(1):112.
Liu XW, et al. Demethylation-activated light-up dual-color RNA aptamersensor for label-free detection of multiple demethylases in lung tissues. Biosens Bioelectron. 2024 Mar 1;247:115966.
Zhao NN, et al. Construction of Genetically Encoded Light-Up RNA Aptamers for Label-free and Ultrasensitive Detection of CircRNAs in Cancer Cells and Tissues. Anal Chem. 2023 Jun 6;95(22):8728-8734.

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