Data Sheet (Cat.No.T209457)

TargetMol

Vari Fluor 425 SE

Chemical Properties

CAS No. :

Formula:

Molecular Weight:

Storage: Keep away from direct sunlight
Store at -20°C
Actual storage temperature shall be subject to the COA.

Biological Description

Description	Vari Fluor 425 SE (VF 425 SE) is a labeling dye from the Vari Fluor SE series, characterized by excitation/emission wavelengths of 430 nm/475 nm. This series features fluorescent dyes with NHS ester groups, designed for labeling antibodies, proteins, peptides, amine-modified oligonucleotides, and other biomolecules containing free amines (-NHX).
Targets(IC50)	Others
In vitro	Preparation of Stock Solution: 1. Protein Preparation: For optimal labeling, adjust the protein (antibody) concentration to 2 mg/mL. 1) Ensure the protein solution has a pH of 8.5±0.5; if below 8.0, adjust with 1 M sodium bicarbonate. 2) A protein concentration below 2 mg/mL significantly reduces labeling efficiency; for best results, aim for a final protein concentration between 2-10 mg/mL. 3) Use buffers free of primary amines (such as Tris or glycine) and ammonium ions to avoid affecting labeling efficiency. 2. Dye Preparation: Dilute VF dye in anhydrous DMSO to create a 10 mg/mL stock solution, mixing thoroughly by pipetting or vortexing. Note: Aliquots of the VF stock solution should be stored protected from light at -20°C or -80°C. 3. Calculation for the Dye Working Solution: The amount of VF dye needed for the labeling reaction depends on the protein quantity, with an ideal molar ratio of VF dye to protein around 10. Example: For labeling 500 µL of 2 mg/mL IgG (MW=150,000), dissolve 1 mg of VF dye in 100 µL DMSO; the required VF volume is 3.95 µL, calculated as follows (using VF 488 as an example): 1) mmol (IgG) = mg/mL (IgG)×mL (IgG) / MW (IgG) = 2 mg/mL × 0.5 mL / 150,000 mg/mmol = 6.7×10^-6 mmol 2) mmol (VF 488) = mmol (IgG) × 10 = 6.7×10^-6 mmol × 10 = 6.7×10^-5 mmol 3) µL (VF 488) = mmol (VF 488) × MW (VF 488) / mg/µL (VF 488) = 6.7×10^-5 mmol × 834 mg/mmol / 0.01 mg/µL = 5.6 µL (VF 488). Usage Instructions: 1. Labeling Reaction: Add the freshly prepared 10 mg/mL VF dye slowly to the 0.5 mL protein sample solution, mix gently, and briefly centrifuge to collect the sample at the bottom of the reaction tube. Avoid vigorous mixing to prevent protein denaturation. 2. Incubate the reaction tube in the dark at room temperature, gently shaking for 60 minutes. Invert the tube occasionally every 10-15 minutes to ensure thorough mixing and enhance labeling efficiency. 2. Protein Purification and Desalting: Purify the dye-protein conjugate using a Sephadex G-25 column, following these steps: 1) Prepare the Sephadex G-25 column according to the manufacturer's instructions. 2) Apply the reaction mixture to the top of the G-25 column. 3) Once the sample reaches below the resin surface, add PBS (pH 7.2-7.4). 4) Elute with additional PBS (pH 7.2-7.4)

In vitro	to collect the desired dye-protein conjugate fraction. Storage Conditions: Store at -20℃ protected from light. Precautions: 1. VF dye is sensitive to light and moisture; prepare VF solutions freshly and discard unused portions. 2. Low concentrations of sodium azide (≤3 mM or 0.02%) or thimerosal (≤0.02 mM or 0.01%) do not significantly interfere with protein labeling; however, 20-50% glycerol decreases labeling efficiency. 3. Avoid buffers containing primary amines (e.g., Tris, glycine) or ammonium ions, as they compete with the protein for labeling. 4. For research use only by professionals and not for clinical or therapeutic applications, food, or medications. 5. Wear lab coats and disposable gloves for safety and health. The above information is based on published literature. Experimental procedures should be appropriately modified to meet specific research demands.