Product Information

Aniline, 10X acetate buffer

Catalog Number: 91057

Storage: 4°C

Unit Size: 10mL

Product Description

Aniline, 10X in acetate buffer is designed to use as catalytic reagent for the optimal coupling of protein carbonyls with aminooxy compounds.

General protocol for labeling protein aldehyde or protein ketone with dye / biotin aminooxy

1. Prepare a 5 mM stock solution of dye / biotin aminooxy in water, DMSO or DMF.

2. Prepare a stock solution of protein aldehyde or protein ketone in 1XPBS buffer, preferably reaching a concentration between 20 uM-100 uM. Low concentration may result in poor protein recovery yield or inefficient labeling.

3. Add 50 molar equivalents of dye / biotin aminooxy to the solution prepared in step 2. For example, if you have 100 uL of protein aldehyde or protein ketone at 50 uM (amount of protein aldehyde or protein ketone is 5 nmole, add 250 nmole of dye / biotin aminooxy, which means 50 uL of 5 mM stock solution).

4. Initiate the ligation by adding 1/10 volume of aniline acetate catalyst. For example, if the mixture from step 3 is 150 uL in total, add 15uL of catalyst.

5. Vortex the solution and allow the reaction to proceed at room temperature with agitation for 2 hrs for protein aldehyde or 5 to 10 hrs for protein ketone in the dark.

6. Purify the CF labeled protein conjugate by Sephadex G25 column or centrifugal protein concentrator. Biotium offers ultrafiltration vials with molecular weight cut-off of 10kDa (**Cat#:22004**) or 3kDa (**Cat#:22018**). To remove free dye by ultrafiltration, choose a molecular weight cut-off that is at least three times larger than your labeled protein, and follow the instructions provided with the ultrafiltration vial.

7. Confirm the formation of product by SDS-PAGE analysis, MALDI-MS analysis, or LC-MS analysis.

Related Products

Catalog number	Product
90103	Aminooxy-5(6)-FAM
90104	Aminooxy-5(6)-ROX
90105	Aminooxy-5(6)-TAMRA
90113	Aminooxy-5(6)-biotin
92050	CF™350, aminooxy
92055	CF™405S, aminooxy
92056	CF™405M, aminooxy
92051	CF™488A, aminooxy
92057	CF™568, aminooxy
92052	CF™594, aminooxy
92053	CF™633, aminooxy
92058	CF™640R, aminooxy
92059	CF™660R, aminooxy
92054	CF™680R, aminooxy

Please visit www.biotium.com to view our full selection of products featuring bright and photostable fluorescent CF™ dyes, including secondary antibodies and Mix-n-Stain™ antibody labeling kits. Biotium also offers a variety of apoptosis and cell viability assays for flow cytometry analysis, including mitochondrial membrane potential dyes, fluorescent Annexin V conjugates, and NucView™488 Caspase-3 Substrate for live cells.

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