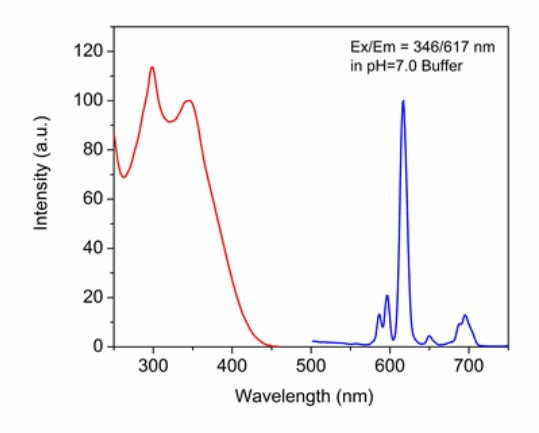
trFluor™ Eu maleimide

Ordering Information	*Storage Conditions*
Product Number: 1434 (100 µg)	Keep refrigerated and desiccated; Avoid light. Expiration date is 12 months from the date of receipt.

Chemical and Spectral Properties

Molecular Weight: 1116.96
Appearance: light yellow powder
Solvents: soluble in dimethylsulfoxide (DMSO)
Spectral Properties: Ex = ~346 nm; Em = ~617 nm (see the spectra below)



Introduction:

Many biological compounds present in cells, serum or other biological fluids are naturally fluorescent, and thus the use of conventional, prompt fluorophores leads to serious limitations in assay sensitivity due to the high background caused by the autofluorescence of the biological molecules to be assayed. The use of long-lived fluorophores combined with time-resolved detection (a delay between excitation and emission detection) minimizes prompt fluorescence interferences. Our trFluor™ Eu probes enable time-resolved fluorometry (TRF) for the assays that require high sensitivity. These trFluor™ Eu probes have large Stokes shifts and extremely long emission half-lives when compared to more traditional fluorophores such as Alexa Fluor® or cyanine dyes. Compared to the other TRF compounds, our trFluor™ Eu probes have relatively high stability, high emission yield and ability to be linked to biomolecules. Moreover, our trFluor™ Eu probes are insensitive to fluorescence quenching when conjugated to biological polymers such as antibodies.

Sample Protocol for Labeling Proteins with trFluor™ Eu Maleimide:

1) Dissolve your thiol-containing protein at concentration 1-10 mg/mL (3-10 mg is the optimal labeling concentration) using PBS buffer (20 mM, pH 7.2).
2) Dissolve the trFluor™ Maleimide in DMSO at the concentration of 10-15 mg/mL (For example, to the 100 µg trFluor™ Maleimide add 10 µL DMSO to make 7 mM dye labeling solution).
3) Mix the trFluor™ Maleimide (from Step 2) and protein solution (from Step 1) at 20:1 **molar** ratio of dye/protein, and shake the reaction mixture at room temperature for 2-4 hours in the dark.

4) Filter the reaction mixture through a protein spin column for 100 µg to 1 mg protein labeling reaction; or purify the conjugate using gel filtration on a properly sized Sephadex G-25 column if the reaction scale is larger than 1 mg.
5) Collect the desired fractions for your immediate use or freeze dry them for your future use.

Note: The trFluor™ conjugate need be used near neutral pH range (6.5 to 7.5). Either acidic or basic pH would reduce its fluorescence intensity.

Sample Protocol for Labeling Small Molecules with trFluor™ Eu Maleimide:

1) Dissolve trFluor™ Maleimide (10 -15 mg/mL) and your thiol-contain molecule in DMSO at 1:1.2 **molar** ratio of dye/ thiol-contain molecule (For example, to the 100 µg trFluor™ Maleimide add 10 µL DMSO to make 7 mM dye labeling solution).
2) Stir the reaction mixture at room temperature for 2-4 hours in the dark.
3) Purify the conjugate using HPLC (ammonium acetate/water and acetonitrile, pH 7.0).
4) Collect and pool the desired fractions.
5) Combine and freeze-dry the pooled fractions.

Note: The trFluor™ conjugate need be used near neutral pH range (6.5 to 7.5). Either acidic or basic pH would reduce its fluorescence intensity.

References:

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3. Paila YD, Kombrabail M, Krishnamoorthy G, Chattopadhyay A. (2011) Oligomerization of the serotonin(1A) receptor in live cells: a time-resolved fluorescence anisotropy approach. J Phys Chem B, 115, 11439.
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