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TAMRA-Lys(Ub)-Gly

Cat. # M3030

Quantity: 50 µg
Species: Human
Source: Synthetic
MW: 9163 Da
Form: Lyophilized powder
Quality Assurance: >95% by RP-HPLC
Description: This products is a fluorescence polarization (FP) assay reagent, which is based on a 5-carboxytetramethylrhodamine (TAMRA) modified Lys-Gly sequence that is linked to ubiquitin via a native isopeptide bond with the lysine side-chain. Typical substrate concentrations range from 10–100 nM. DUB concentrations can range from 0.01-10 nM but depend on specific assay conditions and method of detection. FP assays can be performed with any fluorescence polarization reader with a 530 nm excitation filter and two 580 nm emission filters.

Images:

SDS-PAGE analysis.
Left: coommassie blue staining. Right: fluorescence scan (exc 550 nm, emi 590 nm).

Storage: Powder at −20°C; solution at −80°C. Protect from light and avoid multiple freeze/thaw cycles.

Reconstitution recommendation (Important!):
1) Centrifuge the tube at 10,000 xg for 2 min to pellet the powder.
2) Dissolve the powder in a small amount of DMSO (e.g. 50 µg powder in 2 µL DMSO). Vortex the tube to completely dissolve the powder. Keep under room temperature for 5 min, and then centrifuge under room temperature at 10,000 xg for 2 min to collect solution to the tube bottom.
3) Add 98 µL colde buffer (such as 20 mM Tris, pH 7.2, 150 mM NaCl and 10% glycerol) directly into the tube bottom in once, and pipette up and down to mix (avoid generating bubbles and note the order of addition).
4) The stock solution is 0.5 µg/µL (55 µM). Working concentrations vary from 10 nM – 200 nM.

Literature:
1. Tirat, A. *et al* ., (2005) Anal. Biochem. 343, 244.
2. Huang *et al* ., (2009) Methods in Molecular Biology 565, 127.
3. Levine *et al* ., (1997) Anal. Biochem. 247, 83.
4. Geurink and El Oualid *et al* ., (2012) ChemBiochem 13, 293.