TUBE2 Agarose
Cat. # UM402

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Background

Based on protein domains known to possess an affinity for ubiquitin, Tandem Ubiquitin Binding Entities (TUBEs) have been developed for the isolation and identification of ubiquitinated proteins. TUBEs display up to a 1000-fold increase in affinity for polyubiquitin moieties over the single ubiquitin-binding associated domain (UBA). In addition, TUBEs display a protective effect on polyubiquitinated proteins, allowing for detection at relatively low abundance. These properties effectively "capture" proteins in their polyubiquitin state.

The affinity of solution phase TUBE 2 for K63 linked tetra-ubiquitin is approximately equal to K48 linked tetra-ubiquitin (5-10nM).

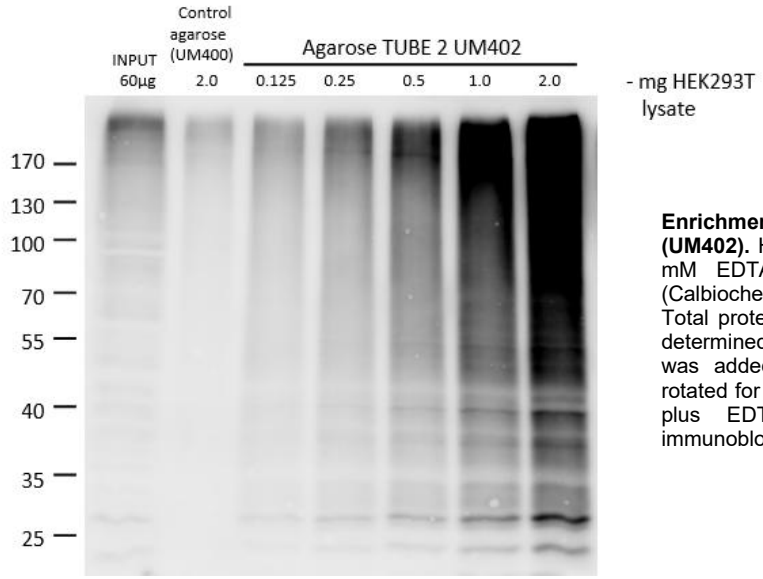
Application(s)

- Pulldown of polyubiquitinated proteins from cell lines, tissues, and organs
- Protection of polyubiquitinated proteins from both deubiquitination and degradation by the proteasome

Product Specifications

Affinity Tag	None
Purity	(prior to coupling) ≥ 95% by SDS-PAGE
Quantity	1ml (0.5ml resin)
Expression System	*E. coli*
Physical State	Liquid (50% resin slurry)
Buffer	PBS, pH 7.5, 20% ethanol
Stability & Storage	≥1 year at -20°C. Avoid freeze/thaw cycles and storage at lower temperatures

Product QC



Enrichment for polyubiquitin with Agarose-TUBE2 (UM402). HEK293T cells were lysed in TBS containing 5 mM EDTA, 1% NP-40, Protease Cocktail Inhibitor (Calbiochem), 50 µM PR619, and 5 mM *o*-phenanthroline. Total protein content of pre-cleared (UM400) lysate was determined by Bradford assay, and 10 µl of UM402 resin was added to the indicted amounts. Reactions were rotated for 3.5 hours (4°C), washed three times with TBS plus EDTA, and analyzed by SDS-PAGE and immunoblotting for ubiquitin (VU101, 1:1000).

References

1. Garadi Suresh H et al., Mol Cell, 2024;84(12):2337-2352.
2. Kadimisetty K., et al., Methods Mol Biol, 2021;2365:185-202.
3. Hjerpe, R, et al., EMBO Rep., 2009; 10,1250-1258.