

Anti-Pan Ubiquitin Monobody (Biotin-labelled)

Hold me closer,
tiny binder!



14.3 kDa Pan Ubiquitin Monobody Punches in Above its Molecular Weight

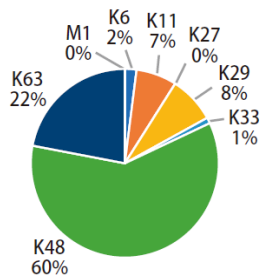
Cosmo Bio's new, exclusive, first in class Pan-Ubiquitin **Monobody (biotin labeled)** is a fibronectin FN3 domain-based recombinant-selected binding protein (monobody) recognizing ubiquitin monomers and mono- and poly-ubiquitinated proteins and featuring high affinity, low ubiquitin linkage site bias, and small size. Validation experiments point to superiority over commonly used pan-ubiquitin monoclonal antibodies such as FK2 and P4D1. Pan Ubiquitin Monobody is ideal for immunoprecipitation (IP), immunofluorescence (IF), immunohistochemistry (IHC), immunocytochemistry (ICC), Western blot (WB) and ELISA.

Anti pan-Ubiquitin Monobody was developed at Nagoya University by researchers expert in FN3 monobody screening (Profs. Hiroshi Murakami and Gosuke Endo) and characterized at Tokyo Metropolitan Institute of Medical Science by researchers with long-standing expertise in ubiquitin-proteasome system related research (Profs. Yasushi Saeki and Akinori Endo).

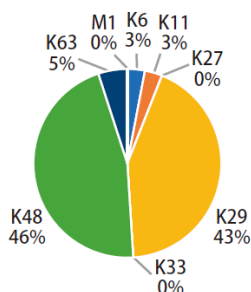
*This product is sold under license from the Tokyo Metropolitan Institute of Medical Science, Tokai National University, and Kyoto University.

Product	Applications	Cat. No.
Anti-Pan Ubiquitin Monobody, Biotin Conjugated	ELISA, WB, IP, IHC, ICC/IF	CAC-MB-001-B

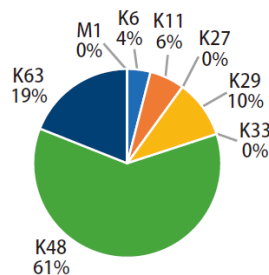
Input



IgG, P4D1



Anti-Ub Mb



Anti Pan-Ubiquitin Monobody captures ubiquitinated Low Ubiquitin Linkage Bias
Distribution of ubiquitin linkages immunoprecipitated from HCT-116 cell lysates with Pan-Ubiquitin Monobody closely mirror that of input lysate (determined by LC-MS/MS). Compare to distorted distribution by IP with monoclonal IgG P4D1.

Pan-Ubiquitin Monobody Specifications

Size	50ug
Product type	Monobody, biotin conjugated
Theoretical MW	14.3 kDa (not including biotin moiety)
Fusion tag	C-terminal His-tag
Specificity/target	Ubiquitin monomer, mono/poly (K6, K11, K27, K29, K33, K48, K63, M1)-ubiquitinated proteins.
Species Reactivity	All species expected
Affinity (Kd)	~0.88 nM
Tested applications	IP, IHC, ICC/IF, WB, ELISA
Purification	Immobilized Metal Chromatography
Format	0.5 mg/ml in 25 mM Na-Phosphate (pH8.0), 250 mM NaCl, 50% Glycerol, 0.05% ProClin 300
Storage condition	For continuous usage, store at -20°C for up to six months. For long-term storage, freeze at -80°C. Avoid repeated freeze/thaw cycles

Pan-Ubiquitin Monobody Reactivity

Application	Pan Ubi-Monobody Reactivity		
	Ubiquitin monomer	Mono-ubiquitinated	Poly-ubiquitinated
ELISA	+	?	?
WB	-	+	+
IP	+	+	+
ICC/IF	+	+	+
IHC	+	+	+



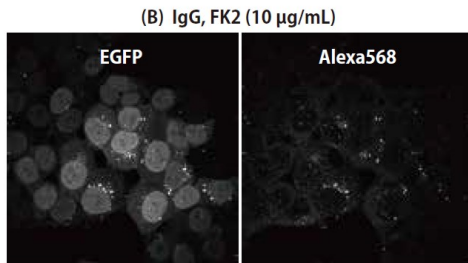
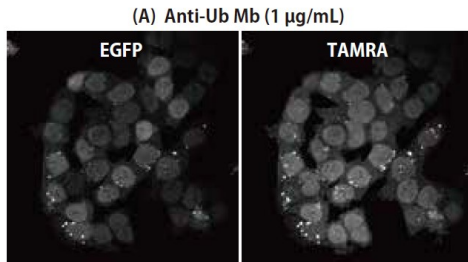
COSMO BIO USA
Inspiration for Life Science

Cosmo Bio USA, Inc.
Carlsbad, CA 92010

+1 760-431-4600

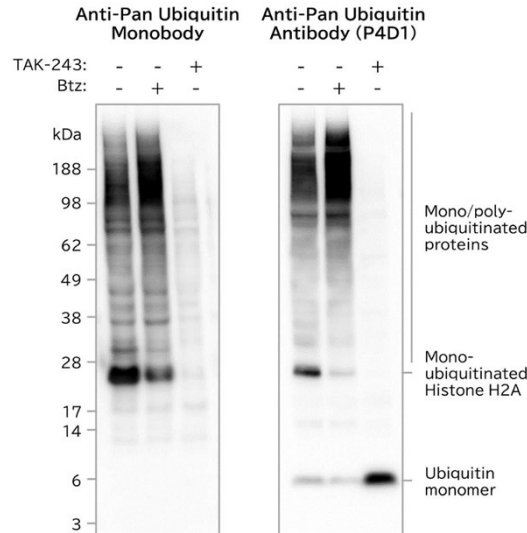
www.cosmobiousa.com

Immunocytochemistry



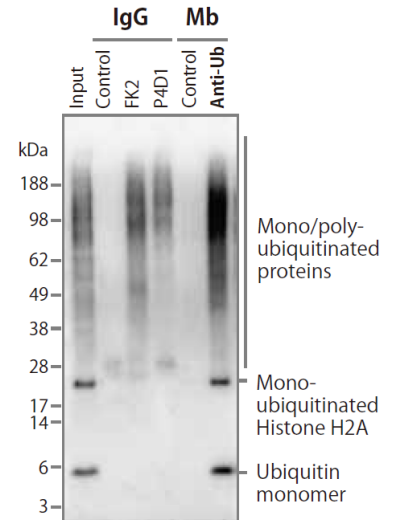
EGFP-ubiquitin fusion protein expressing HCT-116 cells were treated with puromycin to induce ubiquitin-positive aggregates were stained with TAMRA-labeled Pan Ubiquitin Monobody (A) or mouse mAb FK2 and Alexa568-labeled secondary antibody (B). Data provided by Tokyo Metropolitan Institute of Medical Science.

Western blot



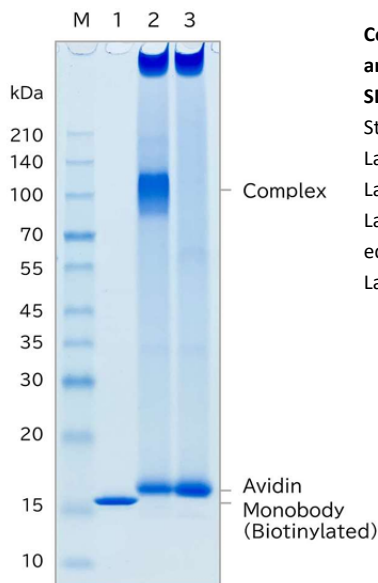
Cell lysates of TAK-243 (ubiquitination inhibitor) or Bortezomide/Btz (proteasome inhibitor) treated HCT-116 cells were Western blotted with Anti-Pan Ubiquitin Monobody (left) or anti-pan ubiquitin mAb clone P4D1, an IgG (right) and visualized by HRP/chemiluminescence. Mono-ubiquitinated histone H2A and low molecular weight mono/poly-ubiquitinated proteins were detected more strongly by Monobody than by P4D1. Monobody did not detect Ubiquitin monomers in Western blots despite doing so in ELISA.

Immunoprecipitation



Immunoprecipitation of HCT-116 cell extracts using Anti-pan Ubiquitin Monobody and two mAb IgG anti-Ubiquitinating antibodies, followed by Western blot with clone P4D1 show that only Pan-Ubiquitin Monobody could immunoprecipitate ubiquitin monomers and monoubiquitinated histones.

Validation Data



Confirmation of Monobody purity and high degree of biotinylation by SDS-PAGE.

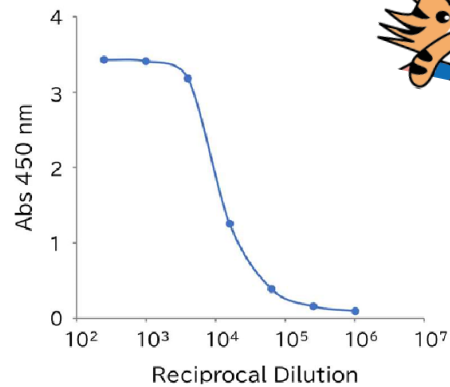
Stain: Coumassie Brilliant Blue.

Lane M: MW markers

Lane 1: 1 µg of Monobody.

Lane 2: 1 µg of Monobody incubated with 5 µg of avidin.

Lane 3: 5 µg avidin.



Monobody binds recombinant human ubiquitin monomer at high titer.

Serial dilutions of 0.5 mg/mL Monobody were allowed to react with immunoassay plate wells coated with 1 µg/well recombinant human ubiquitin monomer. Bound Monobody was detected using NeutrAvidin-HRP and TMB substrate and quenched with sulfuric acid.



Distributed Worldwide by:

COSMO BIO USA

2792 Loker Ave W, Suite 101
Carlsbad, CA 92010

TEL & FAX: 760-431-4600
email : info@cosmobiousa.com
web : www.cosmobiousa.com