10320 Camino Santa Fe, Suite G
San Diego, CA 92121
Tel: 858.875.1900 Fax: 858.875.1999

Ubiquitin Antibody
Catalog # ASM10385

Specification

Ubiquitin Antibody - Product Information

Application	**WB, IHC**
Primary Accession	P0CG53
Other Accession	NP_776558.1
Host	**Rabbit**
Reactivity	**Human, Mouse, Rat, Rabbit, Hamster, Monkey, Pig, Chicken, Bovine, Yeast, Xenopus, Dog, Fish, Sheep, Guinea Pig, Drosophila**
Clonality	**Polyclonal**

Description
Rabbit Anti-Bovine Ubiquitin Polyclonal

Target/Specificity
Detects ~10kDa. It also recognizes ubiquinated proteins.

Other Names
Polyubiquitin B Antibody, RPS27A Antibody, UBA52 Antibody, UBB Antibody, UBC Antibody, ubiquitin B Antibody

Immunogen
Native bovine Ubiquitin, conjugated to KLH

Purification
Peptide Affinity Purified

Storage	**-20ºC**

Storage Buffer
PBS pH7.4, 50% glycerol, 0.09% sodium azide

Shipping Temperature	**Blue Ice or 4ºC**

Certificate of Analysis
A 1:1000 dilution of SPC-119 was sufficient for detection of free ubiquitin in 15 µg of HeLa lysate by ECL immunoblot analysis using Donkey anti-rabbit IgG:HRP as the secondary antibody.

Cellular Localization
Cell Membrane | Cytoplasm | Nucleus

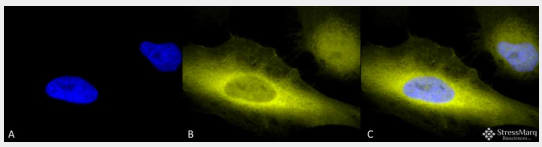
Ubiquitin Antibody - Protocols

Provided below are standard protocols that you may find useful for product applications.

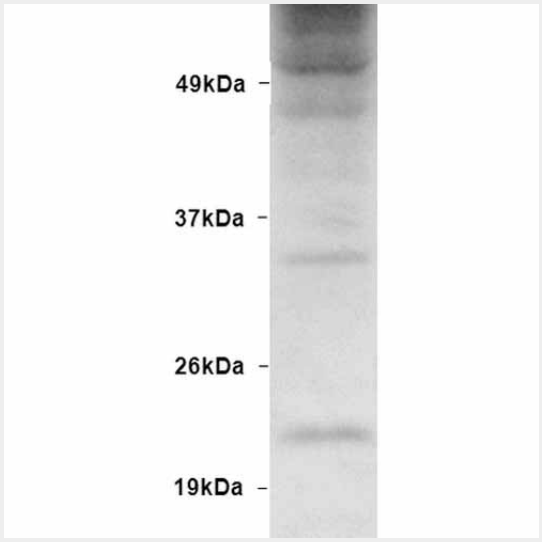
- Western Blot
- Blocking Peptides
- Dot Blot

- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- Cell Culture

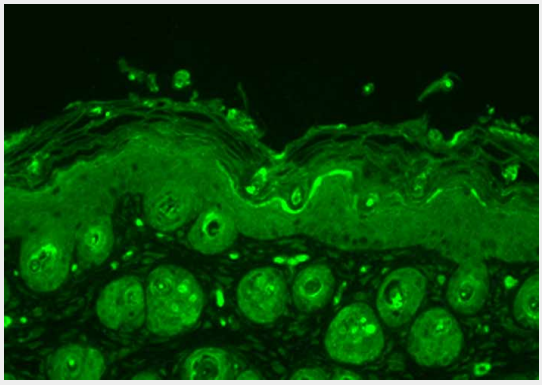
Ubiquitin Antibody - Images



Immunocytochemistry/Immunofluorescence analysis using Rabbit Anti-Ubiquitin Polyclonal Antibody (ASM10385). Tissue: Heat Shocked HeLa Cells. Species: Human. Fixation: 2% Formaldehyde for 20 min at RT. Primary Antibody: Rabbit Anti-Ubiquitin Polyclonal Antibody (ASM10385) at 1:100 for 12 hours at 4°C. Secondary Antibody: R-PE Goat Anti-Rabbit (yellow) at 1:200 for 2 hours at RT. Counterstain: DAPI (blue) nuclear stain at 1:40000 for 2 hours at RT. Localization: Cytoplasm. Magnification: 100x. (A) DAPI (blue) nuclear stain. (B) Anti-Ubiquitin Antibody. (C) Composite. Heat Shocked at 42°C for 1h.

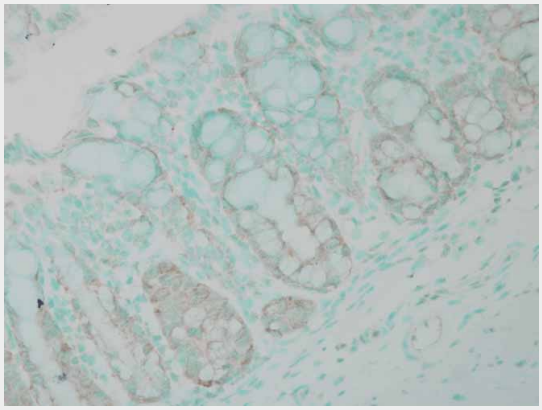


Western blot analysis of Human HEK93 lysates showing detection of Ubiquitin protein using Rabbit Anti-Ubiquitin Polyclonal Antibody (ASM10385). Primary Antibody: Rabbit Anti-Ubiquitin Polyclonal Antibody (ASM10385) at 1:1000.

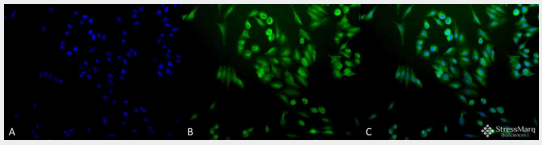


Immunohistochemistry analysis using Rabbit Anti-Ubiquitin Polyclonal Antibody (ASM10385).

Tissue: backskin. Species: Mouse. Fixation: Bouin's Fixative Solution. Primary Antibody: Rabbit Anti-Ubiquitin Polyclonal Antibody (ASM10385) at 1:100 for 1 hour at RT. Secondary Antibody: FITC Goat Anti-Rabbit (green) at 1:50 for 1 hour at RT. Localization: Cytoplasm.



Immunohistochemistry analysis using Rabbit Anti-Ubiquitin Polyclonal Antibody (ASM10385). Tissue: colon carcinoma. Species: Human. Fixation: Formalin. Primary Antibody: Rabbit Anti-Ubiquitin Polyclonal Antibody (ASM10385) at 1:100000 for 12 hours at 4°C. Secondary Antibody: Biotin Goat Anti-Rabbit at 1:2000 for 1 hour at RT. Counterstain: Methyl Green at 200uL for 2 min at RT.



Immunocytochemistry/Immunofluorescence analysis using Rabbit Anti-Ubiquitin Polyclonal Antibody (ASM10385). Tissue: HeLa Cells. Species: Human. Fixation: 2% Formaldehyde for 20 min at RT. Primary Antibody: Rabbit Anti-Ubiquitin Polyclonal Antibody (ASM10385) at 1:200 for 12 hours at 4°C. Secondary Antibody: FITC Goat Anti-Rabbit (green) at 1:200 for 2 hours at RT. Counterstain: DAPI (blue) nuclear stain at 1:40000 for 2 hours at RT. Localization: Cytoplasm. Magnification: 20x. (A) DAPI (blue) nuclear stain. (B) Anti-Ubiquitin Antibody. (C) Composite.

Ubiquitin Antibody - Background

Ubiquitin is a small protein that occurs in all eukaryotic cells. The ubiquitin protein itself consists of 76 amino acids and has a molecular mass of about 8.5 kDa. Key features include its C-terminal tail and the 7 Lys residues. It is highly conserved among eukaryotic species: Human and yeast ubiquitin share 96% sequence identity (1). The main function of Ubiquitin is to clear abnormal, foreign and improperly folded proteins by targeting them for degradation by the 26S proteosome (2). Ubiquitination represents an essential cellular process affected by a multi-enzyme cascade involving classes of enzymes known as ubiquitin-activating enzymes (E1s), ubiquitin-conjugating enzymes (E2s or Ubcs) and ubiquitin-protein ligases (E3s). Ubiquitin is activated in a two-step reaction by an E1 ubiquitin-activating enzyme in a process requiring ATP as an energy source. The initial step involves production of an ubiquitin-adenylate intermediate. The second step transfers ubiquitin to the E1 active site cysteine residue, with release of AMP. This step results in a thioester linkage between the C-terminal carboxyl group of ubiquitin and the E1 cysteine sulfhydryl group. The third step is a transfer of ubiquitin from E1 to the active site cysteine of a ubiquitin-conjugating enzyme E2 via a trans(thio)esterification reaction. And the final step of the ubiquitylation cascade creates an isopeptide bond between a lysine of the target protein and the C-terminal glycine of ubiquitin. In general, this step requires the activity of one of the hundreds of E3 ubiquitin-protein ligases (often termed simply ubiquitin ligase). E3 enzymes function as the substrate recognition modules of the system and are capable of interaction with both E2 and substrate(3, 4). Ubiquitination also participates in the internalization and degradation of plasma membrane proteins such as some of the TCR subunits while still ER-membrane associated (5). Ubiquitin also plays a role

in regulating signal transduction cascades through the elimination inhibitory proteins, such as IκBα and p27 (6).

Ubiquitin Antibody - References

1. Wilkinson K.D. (1995) Annu. Rev. Nutr. 15:161-189.
2. Smalle J., Vierstra R.D. (2004) Anu Rev Plant Biol. 55: 555-590.
3. Bonifacino J.S., et al. (1998) Annu Rev Cell Dev Biol. 14: 19-57.
4. Boston Biochem: "Ubiquitin Proteasome Pathway Overview"
http://www.bostonbiochem.com/upp.php
5. Yang M., et al. (1998) J Exp Med. 187: 1835-1846.
6. Chen Z.J., et al. (1996) Cell 84: 853-862.