



Qty: 100 µg/200 µL

Mouse anti-Ubiquitin

Catalog No. 13-1600

Lot No. See product label

## Mouse anti-Ubiquitin

### FORM

This monoclonal antibody is supplied as a 200 µL aliquot at 0.5 mg/mL in phosphate buffered saline (PBS), pH 7.4 containing 0.1% sodium azide (NaN<sub>3</sub>).

CLONE: Ubi-1

ISOTYPE: IgG<sub>1</sub>-kappa

### IMMUNOGEN

Purified bovine ubiquitin conjugated to carrier protein

### SPECIFICITY

This antibody recognizes ubiquitin, both conjugated and unconjugated. It reacts with a single chain 8.5 kDa protein. The ubiquitin molecule appears to be present in all eukaryotic cells and has an identical primary structure in all animals. Reactivity has been shown in human, bovine, chicken, *Drosophila*, and *C. elegans*. Ubiquitin is present in the nucleus, cytoplasm, and on cell surface membranes.

REACTIVITY: Human, bovine, chicken, mouse<sup>15</sup>, *C. elegans* and *Drosophila*<sup>16</sup>

### USAGE

This antibody is suitable for immunohistochemical staining of alcohol or paraformaldehyde fixed paraffin-embedded or frozen tissue sections. Heat-induced epitope retrieval with citrate buffer, pH 6.0, is required for specific staining of formalin-fixed, paraffin-embedded tissue sections. To stain, incubate 30-60 minutes at room temperature or overnight at 4°C. This antibody may also be used in ELISA and Western blotting.

ELISA: 0.1-1 µg/mL  
Western Blotting\*<sup>6-15, 18-19</sup>: 1-3 µg/mL  
Immunofluorescence<sup>16-17</sup>: 2-5 µg/mL

**\*Important:** When performing Western Blotting, prepare lysis buffer in 10mM N-Ethylmaleimide to inhibit ubiquitin-conjugating enzymes. N-Ethylmaleimide inactivates certain enzymes by blocking free sulfhydryls. After electrophoresis and transfer, pre-incubate transferred membranes in denaturing buffer (6 M guanidine-HCl 20 mM Tris-HCl, pH 7.5, 5 mM beta-mercaptoethanol, 1 mM PMSF)<sup>15</sup> for 30-60 minutes at 4°C, followed by extensive PBS washing.

### STORAGE

Store at 2-8°C for up to one month. Store at -20°C for long-term storage. Avoid repeated freezing and thawing.

(cont'd)

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**BACKGROUND**

Ubiquitin is a multifunctional and extremely highly conserved protein of ~8.5 kDa. An unusual property of ubiquitin is that it can become linked to a lysine  $\epsilon$ -amino group in another protein to produce a covalent ubiquitin-conjugate. Such conjugates are usually very unstable, as they are recognized by a specific ubiquitin-dependent protease and rapidly degraded. Ubiquitin therefore appears to target such proteins for rapid degradation.

There are also examples of stable ubiquitin conjugates, including histone H2a and some unusual forms of actin. In the last few years several groups have noted that a variety of pathological inclusions found in many human disease states exist as stable ubiquitin conjugates. These inclusions include the neurofibrillary tangles of Alzheimer's disease and progressive supranuclear palsy, Pick bodies of Pick's disease, Lewy bodies of Parkinson's disease, Mallory bodies of alcoholic liver disease, Rosenthal fibers of Alexander's disease, and the inclusion bodies in inclusion myositis and oculopharyngeal muscular dystrophy. Such inclusions can be visualized efficiently with antibodies to ubiquitin. Why ubiquitin is found in this variety of inclusions is not currently known, although it seems likely that the cell has tried and failed to remove these insoluble inclusions via the ubiquitin-dependent proteolysis pathway.

**REFERENCES**

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