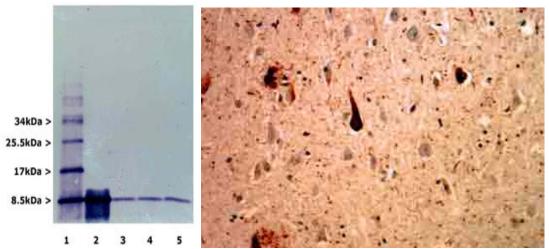


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Catalogue# MCA-Ubi-1: Ubiquitin Monoclonal Antibody Ubi-1

The Immunogen: Ubiquitin is a highly conserved globular 76 amino acid protein of about 8.5 kDa molecular weight. It has a important role in the targeting of proteins for proteolytic degradation. Proteins to be degraded are covalently coupled to the C-terminus of ubiquitin by means of ubiquitin ligases. The ubiquitin itself is frequently also ubiquitinated, producing a polyubiquitin chain. The polyubquitinated complex is then recognized by a complex of degradative enzymes which together form the proteosome. Interestingly, ubiquitin also becomes covalently bonded to many types of pathological inclusions seen in serious human disease states which appear to be resistant to normal degradation, so that ubiquitin antibodies are very useful for studies of these inclusions. For example the neurofibrillary tangles and paired helical filaments diagnostic of Alzheimer's disease, the Lewy bodies seen in Parkinson's disease, and Pick bodies found in Pick's disease are all heavily ubiquitinated and can all be readily visualized with ubiquitin antibodies of appropriate specificity. Ubi-1 has become very widely used for such studies.



Left: Blots of mono and polyubiquitin (lane 1), monoubiquitin only (2), and 100 µg total wet weight of cerebellum, cortex and brain stem respectively (lane 3-5). Material was run out on 20% SDS-PAGE and transferred electrophoretically to PVDF. **Right:** MCA-Ubi-1 staining of cerebral cortex of an Alzheimer patient. Neurofibrillary tangles and dystrophic neurites associated with senile plaques stain strongly with this antibody. In the center is a typical neurofibrillary tangle containing neuron.

Antibody characteristics: MCA-Ubi-1 was made in the University of Florida in 1987, and has been on the market since 1989. It has become widely used to study ubiquitinated inclusions seen in Alzheimer's and other kinds of disease. MCA-Ubi-1 was raised against purified ubiquitin conjugated with glutaraldehyde to keyhole limpet hemocyanin. The clone was initially screened on ELISA of the immunogen, and subsequently tested on sections of Alzheimer brain. MCA-Ubi-1 was one of several clones which stained neurofibrillary tangles in frozen sectioned material strongly and specifically. Subsequent studies indicated that MCA-Ubi-1 is relatively insensitive to formalin fixation and so can be used on mildly fixed sections of human brain for studies of Alzheimer's disease. It also recognizes other ubiquitinated inclusion bodies such as the Lewy bodies of Parkinson's and the Pick bodies in Pick's disease in formalin fixed tissues. MCA-Ubi-1 will recognize human, bovine, chicken, *Drosophila*, and *C. elegans* in ELISA. MCA-Ubi-1 works on Western blots, producing a strong band at ~8.5kDa corresponding to mono-ubiquitin. As shown in the blot, the antibody also stain poly-ubiquitin chains. MCA-Ubi-1 is a mouse IgG1 with a κ light chain. We supply this antibody as ascites fluid or affinity purified preparation at 1 mg/mL. The preparation contains 10 mM sodium azide as a preservative. Store at 4°C or -20°C. Avoid repeated freezing and thawing, store for short term at 4°C for longer term at -20°C or -80°C.

Suggestions for use: MCA-Ubi-1 is excellent for the detection of ubiquitinated inclusions seen in human neurodegenerative diseases such as the neurofibrillary tangles of Alzheimer's disease. We recommend a starting

dilution of 1:1,000 for this purposes, using ABC or other enzymatic amplification procedures. For immunofluorescence try a dilution of 1:500. Ubi-1 will also bind to monomeric and polymeric forms on ubiquitin in western blots, use at 1:1,000 – 1:10,000 for this.

Limitations:

This product is for research use only and is not approved for use in humans or in clinical diagnosis.

Older References:

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Recent Papers using Ubi-1:

Recent Papers using MCA-Ubi-1:

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- 2. Boutajangout A, Authelet M, Blanchard V, Touchet N, Tremp G, Pradier L, Brion JP. Characterisation of cytoskeletal abnormalities in mice transgenic for wild-type human tau and familial Alzheimer's disease mutants of APP and presenilin-1. Neurobiol Dis. 15:47-60 (2004).
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- 4. He CZ, Hays AP. Expression of peripherin in ubiquinated inclusions of amyotrophic lateral sclerosis. <u>J. Neurol.</u> Sci. 217:47-54 (2004).
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- 7. Harris KF, Shoji I, Cooper EM, Kumar S, Oda H, Howley PM. Ubiquitin-mediated degradation of active Src tyrosine kinase. Proc Natl Acad Sci U S A. 96:13738-43 (1999).
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