

Anti-p53 acetyl-K120 antibody, monoclonal (10E5) 71-131 100 ug

Storage temperature: Ship at 4° C and store at -20° C.

Immunogen: synthetic peptide containing acetyl-Lys315 of human p53

Isotype: mouse IgG1

Applications

- 1. Western blotting (~1 ug/ml) (Fig.1)
- 2. Immuno-precipitation (Fig.2)
- 3. Immunofluorescence staining (Fig.3)
- 4. Flow-Cytmetry (1/100)
- 5. ELISA

Form: purified monoclonal antibody (IgG) 1mg/ml in PBS (pH 7.4), 50% glycerol Isotype: mouse IgG1 (κ)

Background: *p53* mutants are found in more than half of human cancers and are considered as the most important human cancer related gene. p53 is detected at 53kD position by electrophoresis and is composed of 393 amino acids. In the unstressed normal cells, the p53 level is low and it is inactive. However, with stress, especially with DNA damage, it is activated to promote arrest of cell cycle and repair of DNA damage, or induction of apoptosis. The functions and stability of p53 are regulated by phosphorylation of serine and threonine, and acetylation of lysine at various sites in the molecule.

Acetylation of lysine 120 (acetyl-K120) of p53 occurs rapidly after DNA damage and is catalyzed by the MYST family acetyltransferases hMOF and TIP60, and activates transcription of proapoptotic genes, BAX and PUMA.

Data Link UniProtKB/Swiss-Prot P04637 (P53_HUMAN)

Related Products: many antibodies specific to phosphorylated and acetylated oncogene products



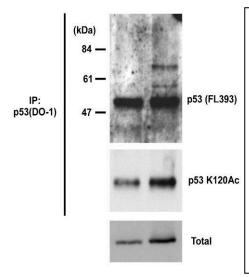


Fig. 1 Identification of p 5 3 protein, whose Lys 120 by Western blotting with 10E5 monoclonal antibody.

Samples are crude lysates of HCT116 cells: Left lanes are control. Right lanes are cells treated with siRNA to knockdown the expression of a Tip60 interacting protein, which results in increase in acetylation of p53 at Lys120. Total p53 was immuno-precipitated with omnipotent anti-p53 monoclonal antibody (DO-1) from the crude extracts and analyzed by Western blotting with anti-p53 antibody (FL393) (upper panel) or anti-p53 acetyl-K120 monoclonal antibody (10E5) (middle panel). The lower panel shows total p53.

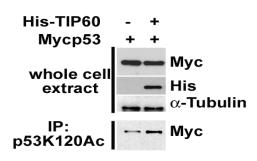


Fig.2 Immunoprecipitation of p53 acetylated at K120 by 10E5 monoclonal antibody.

Crude cell extracts were prepared from H129 cells (p53 negative cell line) expressing only Myc-p53 (first lane), and both Myc-p53 and His-Tip60. In the upper panel, the whole cell extracts were immuno-blotted with anti-Myc, anti-His-tag or anti-atublin antibodies. In the lower panel, the extracts were immuno-precipitated with anti-p53 Ac-K120 antibody

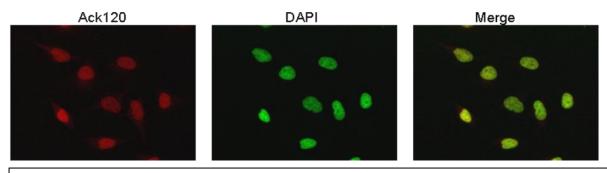


Fig.3. Immunofluorescence staining of p53 acetyl-K120 in nuclei of HeLa cells subjected to DNA damage.

HeLa cells were treated with 100 nM Doxorubicin for 24 hr, fixed with 4% paraformaldehyde overnight, permealized with 0.25% Triton X-100 in PBS for 10 min.

The antibody was used at 1/1,000 dilution

Nucleus (DNA) was stained with DAPI



References: This antibody has been used in the following publications.

- 1. Roe JS *et al.* Phosphorylation of von Hippel-Lindau protein by checkpoint kinase 2 regulates p53 transactivation. <u>Cell Cycle.</u> 2011 Nov 15;10(22):3920-8. PMID:<u>22071692</u> **WB 'human)**
- 2. Mittal S *et al.* The Ccr4a (CNOT6) and Ccr4b (CNOT6L) deadenylase subunits of the human Ccr4-Not complex contribute to the prevention of cell death and senescence. *Mol Biol Cell* 22:748-58 (2011). PMID:21233283 **WB (human)**
- 3. Naidu SR *et al.* PIASy-mediated Tip60 sumoylation regulates p53-induced autophagy Cell Cycle. 2012 Jul 15;11(14):2717-28. PMID:22751435 **WB (human)**
- 4. Gogna R *et al.* p53's choice of myocardial death or survival: Oxygen protects infarct myocardium by recruiting p53 on NOS3 promoter through regulation of p53-Lys(118) acetylation. *EMBO Mol Med* 5:1662-83 (2013). PMID:24096875 **WB (human)**
- 5. Liu N et al. ING5 is a Tip60 cofactor that acetylates p53 in response to DNA damage. Cancer Res 73:3749-60 (2013). PMID:23576563 WB (human)
- 6. Liu X. et al. NAT10 regulates p53 activation through acetylating p53 at K120 and ubiquitinating Mdm2. EMBO Rep. 2016 Mar;17(3):349-66. PMID:26882543 WB (human)
- 7. Itahana Y. *et al.* Histone modifications and p53 binding poise the p21 promoter for activation in human embryonic stem cells. <u>Sci Rep.</u> 2016 Jun 27;6:28112. PMID:<u>27346849</u> **WB (human)**