Store at -20°C

PhosphoPlus® MLKL (Ser358) Antibody Duet

#74921

Support: +1-978-867-2388 (U.S.)
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New 04/18

Entrez-Gene ID #197259
UniProt ID #Q8NB16

For Research Use Only. Not For Use In Diagnostic Procedures.

Products Included	Product #	Quantity	Mol. Wt.	Isotype/Source
MLKL (D2I6N) Rabbit mAb	14993	100 µl	54 kDa	Rabbit IgG
Phospho-MLKL (S358) (D6H3V) Rabbit mAb	91689	100 µl	54 kDa	Rabbit IgG

See www.cellsignal.com for individual component applications, species cross-reactivity, dilutions, and additional application protocols.

Description: PhosphoPlus® Duets from Cell Signaling Technology (CST) provide a means to assess protein activation status. Each Duet contains an activation-state and total protein antibody to your target of interest. These antibodies have been selected from CST's product offering based upon superior performance in specified applications.

Background: Necroptosis, a regulated pathway for necrotic cell death, is triggered by a number of inflammatory signals including cytokines in the tumor necrosis factor (TNF) family, pathogen sensors such as toll-like receptors (TLRs), and ischemic injury (1,2). The process is negatively regulated by caspases and is initiated through a complex containing the RIP1 and RIP3 kinases, typically referred to as the necrosome. Mixed lineage kinase domain-like protein (MLKL) is a pseudokinase that was identified as downstream target of RIP3 in the necroptosis pathway (3,4). During necroptosis RIP3 is phosphorylated at Ser227, which recruits MLKL and leads to its phosphorylation at Thr357 and Ser358 (3). Knockdown of MLKL through multiple mechanisms results in inhibition of necroptosis (3-5). While the precise mechanism for MLKL-induced necroptosis is unclear, some studies have shown that necroptosis leads to oligomerization of MLKL and translocation to the plasma membrane, where it effects membrane integrity (6-9).

Specificity/Sensitivity: MLKL (D2I6N) Rabbit mAb recognizes endogenous levels of total MLKL protein. This antibody also cross-reacts with an unidentified protein of 130 kDa in some cell lines. Phospho-MLKL (Ser358) (D6H3V) Rabbit mAb recognizes endogenous levels of MLKL protein only when phosphorylated at Ser358. This antibody may also bind to MLKL when dually phosphorylated at Thr357 and Ser358.

Source/Purification: Monoclonal antibodies are produced by immunizing animals with a synthetic peptide corresponding to residues near the carboxy terminus of human MLKL protein and a phosphopeptide corresponding to residues surrounding Ser358 of human MLKL.

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. *Do not aliquot the antibody.*

For product specific protocols and a complete listing of recommended companion products please see the product web page at www.cellsignal.com.

Background References:

(1) Christofferson, D.E. and Yuan, J. (2010) *Curr Opin Cell Biol* 22, 263-8.

(2) Kaczmarek, A. et al. (2013) *Immunity* 38, 209-23.

(3) Sun, L. et al. (2012) *Cell* 148, 213-27.

(4) Wang, Z. et al. (2012) *Cell* 148, 228-43.

(5) Wu, J. et al. (2013) *Cell Res* 23, 994-1006.

(6) Cai, Z. et al. (2014) *Nat Cell Biol* 16, 55-65.

(7) Chen, X. et al. (2014) *Cell Res* 24, 105-21.

(8) Wang, H. et al. (2014) *Mol Cell* 54, 133-46.

(9) Dondelinger, Y. et al. (2014) *Cell Rep* 7, 971-81.

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Applications: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide **Species Cross-Reactivity:** **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **Dm**—D. melanogaster **X**—Xenopus **Z**—zebrafish **B**—bovine **Dg**—dog **Pg**—pig **Sc**—S. cerevisiae **Ce**—C. elegans **Hr**—Horse **All**—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.