

PhosphoBLOCKER™ Blocking Reagent

CATALOG NUMBER: AKR-104

STORAGE: Room Temperature

QUANTITY AND CONCENTRATION: 200 g dry blend; 5% concentration after reconstitution in 4L

Background

Protein phosphorylation-dephosphorylation is one of the major signaling mechanisms for modulating the functional properties of proteins involved in gene expression, cell adhesion, cell cycle, cell proliferation, and differentiation. Proteins can be phosphorylated by protein kinases on specific serine, threonine, or tyrosine residues. The utilization of anti-phosphoprotein antibodies in western blotting has become a commonly used tool for signal transduction research. Unfortunately, low levels of endogenous phosphoprotein in various cell lysates often can not be detected, even with high concentrations of antibody and long exposure times. Most commercially available western blot blockers (e.g. dry milk, serum) are sufficient to block the unreacted sites on the membrane, reducing the amount of nonspecific antibody binding during the assay; however, they are not designed to preserve phosphoprotein antigens during blotting.

Cell Biolabs' PhosphoBLOCKER™ contains a proprietary formulation that provides several advantages over conventional blockers:

- Designed specifically for phosphoprotein blotting
- Enhances low level phosphoprotein signal without increasing background
- Premixed dry blend, easy to use

Methods

Freshly prepare 5% PhosphoBLOCKER™ solution in TBST or PBST. Use the 5% PhosphoBLOCKER™ solution to block the blot. When probing the blot, use the 5% PhosphoBLOCKER™ solution to dilute primary and secondary antibodies.

Notes:

- *Reconstituted PhosphoBLOCKER™ solution is only good for one week at 4°C.*
- *The presence of dark-colored particles is a normal artifact of our manufacturing process and will not adversely affect the performance of the product. If desired, the particles may be removed following reconstitution by filtration using standard laboratory filter paper.*

Example of results

The following figures demonstrate typical titration results. One should use the data below for reference only. This data should not be used to interpret actual results.

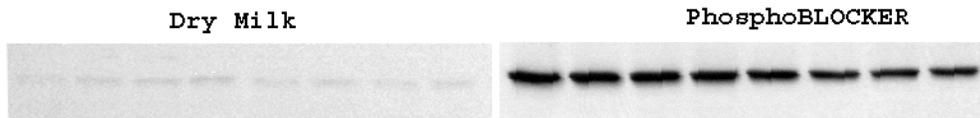


Figure 1. Western Blot of Phospho-p38 in A549 cell lysate.

Recent Product Citations

1. Zalesna, I., et al. (2017). Exogenous growth factors bFGF, EGF and HGF do not influence viability and phenotype of V600EBRAF melanoma cells and their response to vemurafenib and trametinib in vitro. *PLoS One*. **12**(8):e0183498. doi: 10.1371/journal.pone.0183498. eCollection 2017.
2. Surdo, N.C., et al. (2017). FRET biosensor uncovers cAMP nano-domains at β -adrenergic targets that dictate precise tuning of cardiac contractility. *Nat Commun*. **8**:15031. doi: 10.1038/ncomms15031.
3. Hartman, M.L., et al. (2017). Vemurafenib and trametinib reduce expression of CTGF and IL-8 in V600EBRAF melanoma cells. *Lab Invest*. **97**(2):217-227. doi: 10.1038/labinvest.2016.140.
4. Khan, R.S., et al. (2017). Intranasal Delivery of A Novel Amnion Cell Secretome Prevents Neuronal Damage and Preserves Function In A Mouse Multiple Sclerosis Model. *Sci Rep*. **7**:41768. doi: 10.1038/srep41768.
5. Tisdale, E.J. et al. (2016). GAPDH binds Akt to facilitate cargo transport in the early secretory pathway. *Exp. Cell Res*. **349**:310-319.
6. Barkan, D.T. et al. (2016). Clustering of disulfide-rich peptides provides scaffolds for hit discovery by phage display: application to interleukin-23. *BMC Bioinform*. **17**:481.
7. Hirozane, T. et al. (2016). Conditional abrogation of Atm in osteoclasts extends osteoclast lifespan and results in reduced bone mass. *Sci Rep*. doi:10.1038/srep34426.
8. Treda, C. et al. (2016). EGFR activation leads to cell death independent of PI3K/AKT/mTOR in an AD293 cell line. *PLoS One*. **11**:e0155230.
9. Ye, J. X. et al. (2016). The suppression of PGC-1 α is associated with hypoxia-induced endothelial dysfunction and provides a new therapeutic target in pulmonary arterial hypertension. *Am J Physiol Lung Cell Mol Physiol*. doi:10.1152/ajplung.00356.2015.
10. Fukuda, S. et al. (2016). Reversible interconversion and maintenance of mammary epithelial cell characteristics by the ligand-regulated EGFR system. *Sci Rep*. doi:10.1038/srep20209.
11. Shinoda, K. et al. (2015). Pin1 facilitates NF- κ B activation and promotes tumour progression in human hepatocellular carcinoma. *Br J Cancer*. doi: 10.1038/bjc.2015.272.
12. Kuroda, Y. et al. (2015). Suppressive effect of membrane-permeable peptides derived from autophosphorylation sites of the IGF-1 receptor on breast cancer cells. *Eur J Pharmacol*. doi:10.1016/j.ejphar.2015.08.004.
13. Matsushima, M. et al. (2015). Intravesical dual PI3K/mTOR complex 1/2 inhibitor NVP-BEZ235 therapy in an orthotopic bladder cancer model. *Int J Oncol*. doi: 10.3892/ijo.2015.2995.
14. Obis, T. et al. (2015). The novel protein kinase C epsilon isoform at the adult neuromuscular synapse: location, regulation by synaptic activity-dependent muscle contraction through TrkB signaling and coupling to ACh release. *Molecular Brain*. **8**:8.

15. Bemben, M. A. et al. (2015). Autism-associated mutation inhibits protein kinase C-mediated neuroligin-4X enhancement of excitatory synapses. *Proc Natl Acad Sci U S A*. **112**:2551-2556.
16. F. de Castro, L. et al. (2015). VEGF Receptor 2 (VEGFR2) activation is essential for osteocyte survival induced by mechanotransduction. *J Cell Physiol*. **230**:278-285.
17. Ohba, K. et al. (2014). Expression of (pro) renin receptor in breast cancers and its effect on cancer cell proliferation. *Biomedical Res*. **35**:117-126.
18. Shimura, T. et al. (2014). AKT-mediated enhanced aerobic glycolysis causes acquired radioresistance by human tumor cells. *Radiother Oncol*. **112**:302-307.
19. Shimura, T. et al. (2014). DNA damage signaling guards against perturbation of cyclin D1 expression triggered by low-dose long-term fractionated radiation. *Oncogenesis*. **3**:e132.
20. Inserte, J. et al. (2014). Delayed phospholamban phosphorylation in post-conditioned heart favours Ca²⁺ normalization and contributes to protection. *Cardiovasc Res*. **103**:542-553.
21. Lan, F. et al. (2014). LECT2 functions as a hepatokine that links obesity to skeletal muscle insulin resistance. *Diabetes*. **63**:1649-1664.
22. Nakamura, Y. et al. (2014). Oligomerization-induced conformational change in the C-terminal region of Nel-like molecule 1 (NELL1) protein is necessary for the efficient mediation of murine MC3T3-E1 cell adhesion and spreading. *J. Biol. Chem*. **289**:9781-9794.
23. Marques, J. et al. (2013). CRMP2 tethers kainate receptor activity to cytoskeleton dynamics during neuronal maturation. *J. Neurosci*. **33**:18298-18310.
24. Ferreira, E. et al. (2013). Inflammatory cytokines induce a unique mineralizing phenotype in mesenchymal stem cells derived from human bone marrow. *J. Biol. Chem*. **288**:29550-29561.
25. Ishii, K. et al. (2012). *Serratia marcescens* induces apoptotic cell death in host immune cells via a lipopolysaccharide- and flagella-dependent mechanism. *J. Biol. Chem*. **287**: 36582-36592.

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