



**PhosphoSolutions®**  
Antibodies that work™

Colorado Biosciences Park  
12635 East Montview Boulevard, #213  
Aurora, CO 80045-7337  
Tel: (888) 442-7100

## Anti-Phospho-Ser<sup>269</sup> Aquaporin 2

**Catalog Number:** p112-269

**Size:** 100 µl

**Product Description:** Affinity purified rabbit polyclonal antibody

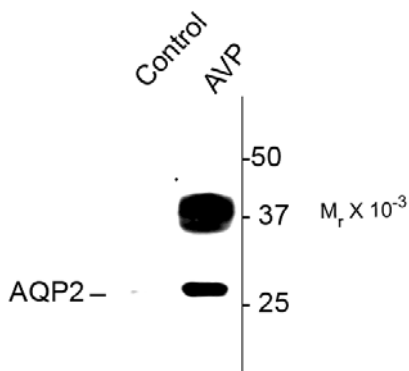
**Applications:** WB: 1:1000

**Antigen:** Phosphopeptide corresponding to amino acid residues surrounding the phospho-Ser<sup>269</sup> of rat aquaporin 2.

**Species reactivity:** The antibody has been directly tested for reactivity in Western blots with rat tissue. It is anticipated that the antibody will react with bovine, canine and mouse tissues based on the fact that these species have 100% homology with the amino acid sequence used as antigen.

**Biological Significance:** Aquaporin 2 (AQP2) is a hormonally regulated water channel located in the renal collecting duct. Mutations in the AQP2 gene cause hereditary nephrogenic diabetes insipidus in humans (Iolascon et al., 2007). A vasopressin induced cAMP increase results in the phosphorylation of AQP2 at serine-256 and its translocation from the intracellular vesicles to the apical membrane of principal cells (van Balkom et al., 2002). Serine-269 has been recently identified as a vasopressin-mediated phosphorylation site on AQP2 and as such has shown to potentiate plasma membrane retention of AQP2 (Hoffert JD et al., 2008).

### Anti-Phospho-Ser<sup>269</sup> Aquaporin 2



**Western blot** of vasopressin (AVP) treated rat kidney lysate showing specific immunolabeling of the ~ 29k and 37k glycosylated form of the AQP2 protein phosphorylated at Ser269.

**WB** = Western Blot   **IF** = Immunofluorescence   **IHC** = Immunohistochemistry   **IP** = Immunoprecipitation

**Packaging:** 100 µl in 10 mM HEPES (pH 7.5), 150 mM NaCl, 100 µg per ml BSA and 50% glycerol. Adequate amount of material to conduct 10-mini Western Blots.

**Storage and Stability:** Store at -20°C; stable for at least one year.

**Shipment:** Domestic - Blue Ice; International - Blue Ice or Dry Ice.

**Purification Method:** Prepared from rabbit serum by affinity purification via sequential chromatography on phospho- and dephosphopeptide affinity columns.

**Antibody Specificity:** Specific for the ~29k AQP2 protein phosphorylated at Ser<sup>269</sup>. Also recognizes the glycosylated form of AQP2 at ~ 37k. Immunolabeling of the AQP2 band is blocked by preadsorption with the phospho-peptide used as antigen but not by the corresponding dephospho-peptide.

**Quality Control Tests:** Western blots performed on each lot.

#### References:

- van Balkom BW, Savelkoul PJ, Markovich D, Hofman E, Nielsen S, van der Sluijs P, Deen PM (2002) The role of putative phosphorylation sites in the targeting and shuttling of the aquaporin 2 water channel. *J Biol Chem* 277(44):41473-9.
- Ford P, Rivarola V, Chara O, Blot-Chabaud M, Cluzeaud F, Farman M, Parisi M, Capurro C (2005) Volume regulation in cortical collecting duct cells: role of AQP2. *Biol Cell* 97(9):687-97.
- Hoffert JD, Nielsen J., Yu MJ., Pisitkun T., Schleicher SM., Nielsen Knepper MA (2007) Dynamics of aquaporin-2 serine-261 phosphorylation in response to short-term vasopressin treatment in collecting duct. *Am J Physiol Renal Physiol* 292: F691-F700.
- Iolascon A, Aglio V, Tamma G, D'Appolito M, Addabbo F, Procino G, Simonetti MC, Montini G, Gesulado L, Debler EW, Suelto M, Valenti G (2007) Characterization of two novel missense mutations in AQP2 gene causing nephrogenic diabetes insipidus. *Nephron Physiol.* 105(3): p33-41.
- Hoffert JD, Fenton RA, Moeller HB, Simons B, Tchapyjnikov D, McDill BW, Yu MJ, Pisitkun T, Chen F, Knepper MA. (2008) Vasopressin-stimulated increase in phosphorylation at serine 269 potentiates plasma membrane retention of aquaporin 2. *J Biol Chem.* 2008 Sep 5; 283(36):24617-27.

**WB** = Western Blot    **IF** = Immunofluorescence    **IHC** = Immunohistochemistry    **IP** = Immunoprecipitation

**Packaging:** 100 µl in 10 mM HEPES (pH 7.5), 150 mM NaCl, 100 µg per ml BSA and 50% glycerol. Adequate amount of material to conduct 10-mini Western Blots.

**Storage and Stability:** Store at -20°C; stable for at least one year.

**Shipment:** Domestic - Blue Ice; International - Blue Ice or Dry Ice.