# Affinity Purified Rabbit Anti-Phospho-14-3-3 Protein (S58) Certificate of Analysis

#### **ORDERING INFORMATION**

Catalog Number: PPS042

Lot Numbers: 1554473

Size: 100 L (sufficient for 10 mini-blots)

Storage: -20° C

- Specificity: Human, mouse, rat, and Xenopus ~29 kDa 14-3-3 phosphorylated at S58
- Immunogen: Phosphopeptide corresponding to amino acid residues surrounding the phospho-S58 of rat 14-3-3 protein

Ig Type: rabbit IgG

Applications: Western blot



#### Western blot of rat brainstern lysate showing specific immunolabeling of the approximately 29 kDa 14-3-3 protein phosphorylated at S58 (Control). The immunolabeling is blocked by the phosphopeptide used as the antigen (Pep. block) but not by the corresponding dephosphopeptide (not shown).

#### Description

14-3-3 is a general name given to seven ubiquitously expressed, 28 kDa proteins that are the products of related, but separate genes. They are considered isoforms and are named eta ( ), gamma ( ), delta/zeta ( / ), alpha/beta ( / ), theta/tau ( / ), sigma ( ), and epsilon (). The first molecule was named for its 14th DEAE elution fraction and 3.3 migration pattern on gel electrophoresis. All seven molecules range from 245 - 255 amino acids in length and exhibit nine -helices arranged in an antiparallel fashion. Overall amino acid sequence identity is approximately 49%, with most identity limited to six blocks of six or more amino acids. The invariant amino acids create a negatively-charged channel that is common to all isoforms. 14-3-3 proteins are known to bind over 200 client (mostly phosphorylated) proteins. The 14-3-3s are highly versatile, inhibiting, activating, linking and transporting a myriad of unrelated targets. They participate in all cellular processes. 14-3-3 proteins are both homodimers and heterodimers and also exist as monomers. Dimeric 14-3-3 generally binds to phosphorylated targets, generating some type of response. Monomeric 14-3-3 also appears to bind to the same targets, but without generating a response. The transition from dimer to monomer is controlled by phosphorylation, particularly by SKD1. The favored site for this phosphorylation is S59 on the eta and gamma isoforms, S58 on the zeta/delta isoform, and S60 on the alpha/beta isoform; tau, sigma and epsilon do not contain favorable motifs for phosphorylation.

# Preparation

Prepared from rabbit serum by affinity purification via sequential chromatography on phospho and dephosphopeptide coupled columns.

### Formulation

100 L in 10 mM HEPES (pH 7.5), 150 mM NaCl, 100 g/mL BSA and 50% glycerol.

# Storage

For long-term storage, -20° C is recommended. Product is stable at -20° C for at least 1 year.

### Specificity

This antibody is specific for the 29 kDa 14-3-3 phosphorylated at S58 in Western blots of rat brain stem lysates.

## Applications

Western blot - 1:1000

Optimal dilutions should be determined by each laboratory for each application.

#### References

- 1. Aitken, A. (2006) Semin. Cancer Biol. 16:162.
- 2. Woodcock, J.M. et al. (2003) J. Biol. Chem. 278:36323.
- 3. Muslin, A.J. et al. (1996) Cell 84:889.
- 4. Hamaguchi, A. et al. (2003) Biochem. Biophys. Res. Commun. 307:589.
- 5. Aitken, A. *et al.* (2002) Biochem. Soc. Trans. **30**:351.

Shelley Falvey

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