

Saint Louis, Missouri 63103 USA Telephone (800) 325-5832 (314) 771-5765 Fax (314) 286-7828 email: techserv@sial.com sigma-aldrich.com

ProductInformation

Anti-GRP75 (SQ-15)

Produced in Rabbit Affinity Isolated Antibody

Product Number G 4045

Product Description

Anti-GRP75 (SQ-15) is developed in rabbit using as immunogen a synthetic peptide corresponding to amino acids 665-679 located at the C-terminus of human GRP75, conjugated to KLH. This sequence is identical in mouse GRP75 (mot-1 and mot-2 isoforms) and highly conserved in rat GRP75 (1 amino acid substitution). The antibody is affinity-purified using the immunogenic peptide immobilized on agarose.

Anti-GRP75 (SQ-15) specifically recognizes GRP75. Applications include the detection of GRP75 by immunoblotting (75 kDa) and immunocytochemistry. Staining of the GRP75 band in immunoblotting is specifically inhibited by the GRP75 immunizing peptide (human, amino acids 665-679).

Heat shock proteins (HSP) are a class of stress proteins, which include HSP20, HSP60, HSP70, and HSP90. These proteins are considered to function as molecular chaperones by transiently binding to newly synthesized proteins to facilitate their correct folding and assembly. GRP75 (glucose-regulated protein 75, mitochondrial HSP70, mortalin, HSP70-9B, PBP74, 75kDa) belongs to a subfamily of the heat shock proteins Hsp70.^{1, 2} Other members of the GRPs family include GRP78/BiP and GRP94. GRPs are unresponsive to heat stress and are induced by stress related to glucose starvation or defects in glycoprotein processing. GRP75/mortalin has been localized mainly to the endoplasmic reticulum (ER), but also to various cellular compartments including mitochondria and cytoplasmic vesicles. 3-5 It has multiple functions ranging from stress responses to intracellular trafficking, antigen processing, and control of cell proliferation, differentiation, and tumorigenesis.

GRP75 levels correlate with muscle activity, mitochondrial activity, and biogenesis. It is induced by low levels of ionizing radiation, glucose deprivation, calcium ionophore, and ozone. Two murine mortalin genes have been cloned (mot-1 and mot-2), which encode two proteins that differ by two amino acids in their C-terminal region. Mot-1 is expressed in normal and embry onic mouse cells and has a pancytosolic cellular distribution. whereas mot-2 is found in the perinuclear region of immortal cells. Expression of the cytosolic form (mot-1) induces senescence of NIH3T3 cells that normally harbor in the perinuclear form. 6 Mot-2 expression is upregulated in human transformed and tumors cell lines and tumors. 7, 8 Mot-2 but not mot-1 has been shown to bind to p53 resulting in the inactivation of p53 function. Mot-2 overexpression in NIH3T3 and MRC5 cells leads to their malignant transformation and life span extension, respectively, an effect thought to be mediated, at least in part by inactivation of p53 by mot-2.7, 9, 10, 11

Reagent

The antibody is supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide.

Antibody Concentration: ~1.0 mg/mL

Precautions and Disclaimer

Due to the sodium azide content, a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazardous and safe handling practices.

Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For extended storage, freeze in working aliquots. Repeated freezing and thawing is not recommended. Storage in "frost-free" freezers is also not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilutions should be discarded if not used within 12 hours.

Product Profile

By immunoblotting, a working antibody concentration of 0.2-0.4 μ g/mL is recommended using whole cell extracts of the human epitheloid carcinoma HeLa cells, Madin-Darby canine kidney (MDCK) cells, and mouse fibroblasts NIH3T3 cells.

By indirect immunofluorescence, a working antibody concentration of 1-2 μ g/mL is recommended using HeLa cells.

Note: In order to obtain the best results using various techniques and preparations, we recommend determining the optimal working dilutions by titration.

References

- 1. Lee, A.S., Trends Biochem. Sci., **26**, 504-510 (2001).
- Wadhwa, R., et al., Cell Stress Chaperones, 7, 309-316 (2002).
- Wadhwa, R., et al., J. Biol. Chem., 268, 6615-6621 (1993).
- 4. Bhattacharyya, T., et al., J. Biol. Chem., **270**, 1705-1710 (1995).
- 5. Ran, Q., et al., Biochem. Biophys. Res. Commun., **275**, 174-179 (2000).
- Wadhwa, R., et al., J. Biol. Chem., 268, 22239-22242 (1993).
- 7. Kaul, S.C., et al., Oncogene, **17**, 907-911 (1998).
- 8. Takano, S., et al., Exp. Cell Res., **237**, 38-45 (1997).
- Wadhwa, R., et al., J. Biol. Chem., 273, 29586-29591 (1998).
- Kaul, S.C., et al., Exp. Cell Res., 286, 96-101 (2003).
- 11. Wadhwa, R., et al., Cell Res., **9**, 261-269 (1999).

KAA/EK 08/05