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Product Information

ANTI-CALRETICULIN

Developed in Rabbit
IgG Fraction of Antiserum

Product Number **C 4606**

Product Description

Anti-Calreticulin is developed in rabbit using a synthetic peptide corresponding to the C-terminus of human calreticulin (amino acids 401-417) conjugated to KLH as immunogen. This sequence has limited homology (65%) to rat and mouse calreticulin. Whole antiserum is fractionated and then further purified by ion-exchange chromatography to provide the IgG fraction of antiserum that is essentially free of other rabbit serum proteins.

Anti-Calreticulin recognizes human and dog calreticulin (55-60 kDa). Applications include the detection and localization of calreticulin by immunoblotting, immunoprecipitation, and immunofluorescence. Staining of calreticulin in immunoblotting is specifically inhibited with the calreticulin immunizing peptide (human, amino acids 401-417).

Calreticulin (Calregulin) is a 55 kDa, calcium-binding chaperone, that is localized primarily in the lumen of the endoplasmic reticulum (ER).¹⁻⁵ Newly synthesized cellular and extracellular proteins must be correctly folded and assembled in the ER before they progress to the cytosol or cell surface. This process is facilitated by transient interaction with a specific set of molecular chaperones that reside in the ER lumen including calnexin, calreticulin, protein disulfide isomerase (PDI), and molecular chaperones of the Hsp60, Hsp70, and Hsp90 families. Calreticulin acts as a lectin-like chaperone binding oligosaccharide residues of newly synthesized N-linked glycoproteins, and misfolded proteins.³⁻⁶ It is believed to play a critical role in quality control processes during protein synthesis and folding. The lectin specificity of calreticulin has been identified as high mannose oligosaccharides terminating in monoglucosyl residues linked through α 1-3.³⁻⁵ Increased expression of calreticulin increases Ca^{2+} storage capacity of the ER. It also appears to modulate store-

operated Ca^{2+} -influx, and to alter Ca^{2+} transport by the sarcoplasmic/ER Ca^{2+} -ATPase (SERCA).^{4, 7-9} Over-expression of calreticulin results in increased sensitivity of HeLa cells to drug-induced apoptosis.¹⁰ In contrast, calreticulin-deficient cells show increased resistance to apoptosis.

Calreticulin gene disruption in mice (*crt^{-/-}*), is embryonic lethal showing a marked impairment of cardiac development, indicating that calreticulin is essential for proper cardiac development.^{11, 12} In *crt^{-/-}* cells, agonist-induced Ca^{2+} release via the inositol 1,4,5-triphosphate (InsP3) pathway is also inhibited, and the ER has lower capacity for Ca^{2+} storage^{11, 13} indicating that calreticulin, in addition to its chaperone activity, plays a critical role in Ca^{2+} homeostasis.

Reagent

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

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 prolonged storage, freeze in working aliquots at -20 °C. 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 dilution samples should be discarded if not used within 12 hours.

Product Profile

A minimum working dilution of 1:2,000 is determined by immunoblotting using a RIPA lysate of the human hepatocytoma HepG2 cell line or the human epitheloid carcinoma HeLa cell line.

The antibody (minimum of 40 µg of IgG) immunoprecipitates calreticulin from a RIPA lysate of the human epitheloid carcinoma HeLa cell line.

A minimum working dilution of 1:100 is determined by immunofluorescence staining using the Madin-Darby canine kidney MDCK cell line.

Note: In order to obtain best results in different techniques and preparations we recommend determining optimal working dilutions by titration test.

References

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