

Sheep anti-human GRP78

Affinity-Purified IgG

0.1 mg

Product #: SAGRP78-AP

Lot #:

Expiry date:

Store at -10 to -20°C

For Research Use Only. Not for use in diagnostic procedures.

Description of GRP78

The 78 kDa glucose-regulated protein (GRP78), a highly conserved ATP-binding protein and member of the heat shock protein family^{1,3}. GRP78, also known as binding immunoglobulin protein (BiP), is encoded by the HSPA5 gene^{4,5}. GRP78 transiently associates with efficiently folded proteins, while stably complexing with misfolded or incompletely assembled proteins. Misfolded proteins bound to GRP78 are subsequently translocated from the ER into the cytosol for proteasomal degradation^{6,7}. The observation that GRP78 is induced by a variety of conditions that cause ER stress^{1,2,3} provides further evidence that it is a multifunctional stress response protein that plays a critical role in quality control processes during protein folding and processing. GRP78 contains two functional domains: a nucleotide-binding domain (NBD) and a substrate-binding domain (SBD). The NBD binds and hydrolyses ATP, and the SBD binds polypeptides^{8,9}. Current evidence suggests that GRP78 can protect against a variety of pathological conditions that cause ER stress, a cellular stress condition known to contribute to a wide range of human diseases, including cardiometabolic diseases, diabetes, diabetic nephropathy as well as neurodegenerative, autoimmune, and infectious diseases^{8,9,10}. However, ER stress can localize GRP78 to the cell surface whereby it acts as a unique signalling receptor and neoantigen leading to the development of anti-GRP78 autoantibodies that further contribute to disease progression^{11,12}.

Product Specifications

Description:

Vial containing ml of IgG purified by affinity chromatography on immobilized recombinant human GRP78. Total protein is 0.1 mg.

Format:

Affinity-purified IgG (APiG), clear liquid.

Host Animal:

Sheep

Immunogen:

Recombinant human GRP78

Concentration:

APiG concentration is mg/ml, determined by absorbance using an extinction coefficient ($E^{1\%_{280}}$) of 13.4.

Buffer:

10 mM HEPES, pH 7.4, 150 mM NaCl, 50% (v/v) glycerol.

Storage:

Store between -10 and -20°C. Product will become viscous but will not freeze. Avoid storage in frost-free freezers. Keep vial tightly capped. Allow product to warm to room temperature and gently mix before use.

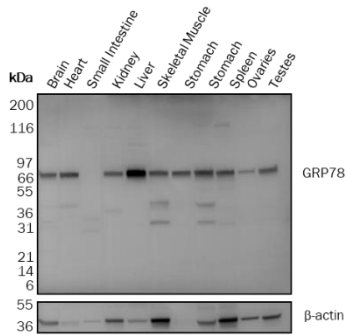
Specificity:

This antibody is specific for human GRP78 as demonstrated by direct ELISA and Western blot.

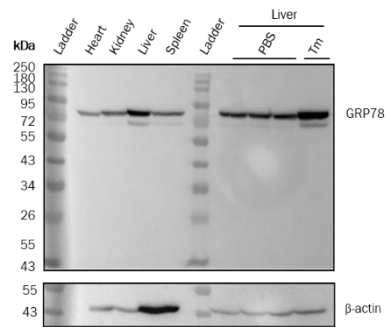
Applications:

Suitable as a source of enriched antibodies to human GRP78. Antibodies for the detection of GRP78 may be used in several applications including Western Blot, immunofluorescence, immunocytochemistry and ELISA. The following images for SAGRP78-AP are representative examples of product applications. The indicated dilutions are lot specific, therefore, optimal dilutions/concentrations should be determined by the end user in their application.

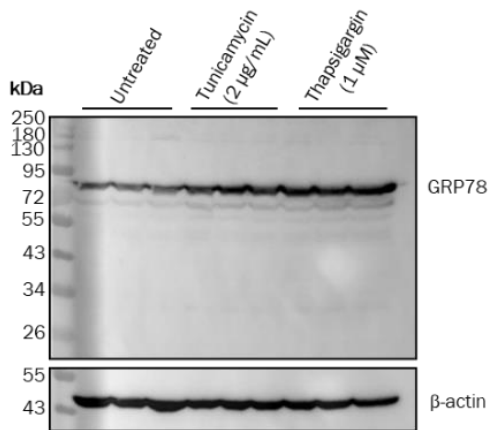
Western Blots:



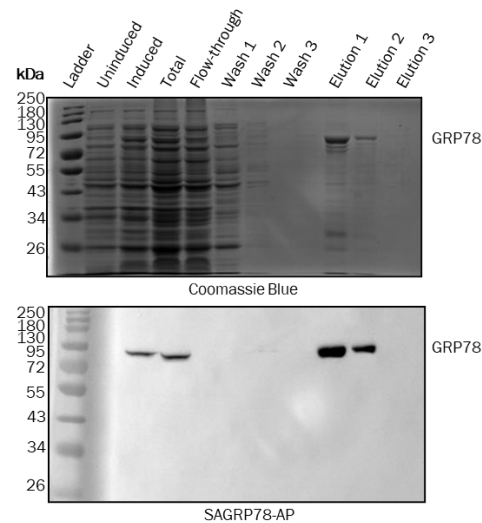
Immunoblot analysis of GRP78 expression in human tissue extracts. Whole cell lysates (50 µg/lane) from a variety of human tissues. (SAGRP78-AP; 1:2000) and β-actin (Millipore-Sigma; 1:2000) expression was examined.



Immunoblot analysis of GRP78 expression in mouse tissue extracts. (Left) Whole tissue lysates (50 µg/lane) from mouse, heart, kidney, liver, spleen. (Right) Liver lysates from mice which were treated with PBS or tunicamycin (Tm; 2 µg/mL). GRP78 (SAGRP78-AP; 1:2000) and β-actin (Millipore-Sigma; 1:2000) expression was examined.

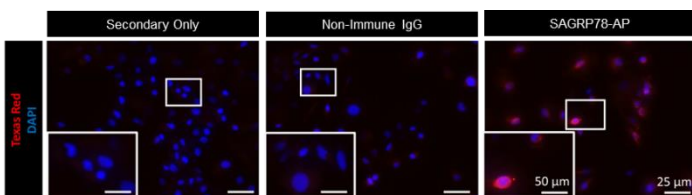


Increased expression of GRP78 in human cells exposed to thapsigargin or tunicamycin. DU145 prostate cancer cells treated with tunicamycin or thapsigargin at indicated doses for 24 hours and lysed. GRP78 (SAGRP78-AP; 1:2000) and β-actin (Millipore-Sigma; 1:2000) expression was examined.

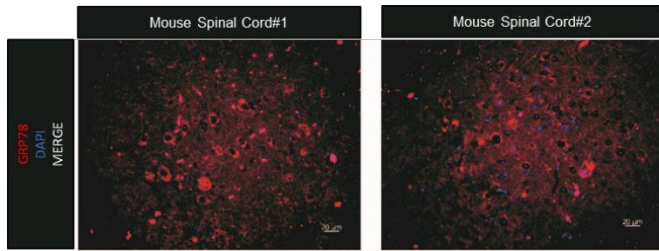


Detection of recombinant human GRP78 from bacterial lysate. *E. coli* bacterial cell lysate containing recombinant human GRP78 was separated and either stained with Coomassie blue or transferred to a nitrocellulose membrane. GRP78 expression was examined (SAGRP78-AP; 1:2000).

Immunofluorescence:

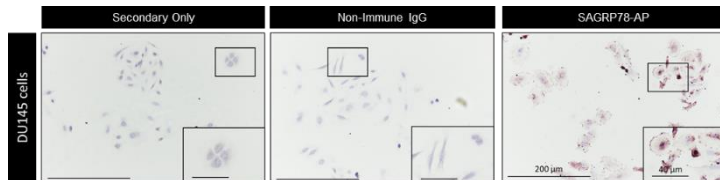


Immunofluorescence staining of GRP78 in human cells. Fixed DU145 cells were blocked and incubated with either SAGRP78-AP (1:200), non-immune sheep IgG (1:200), or BSA alone. Fluorescently labelled donkey anti-sheep IgG (Novus Biologicals; 1:200) was used and visualized using a fluorescent microscope.



Immunofluorescence staining of GRP78 in mouse spinal cord. Mice spinal cords were fixed, blocked, and incubated with either SAGRP78-AP (1:200), non-immune sheep IgG (1:200), or BSA alone. Fluorescently labelled donkey anti-sheep IgG (Novus Biologicals; 1:200) was used and visualized using a fluorescent microscope.

Immunocytochemistry:



Immunocytochemical staining of GRP78 in human cells. Fixed DU145 cells were blocked and incubated with either SAGRP78-AP (1:200), non-immune sheep IgG (1:200), or BSA alone. Fluorescently labelled donkey anti-sheep IgG (Novus Biologicals; 1:200) was used and visualized using a light microscope.

Related Products:

Cat #: **SAGRP78-APHRP** Sheep anti-human GRP78, affinity purified IgG-peroxidase

Cat #: **SAGRP78-APFTC** Sheep anti-human GRP78, affinity purified IgG-FITC

Cat #: **DAS-APHRP** Donkey anti-sheep, affinity purified IgG-peroxidase – Secondary Antibody

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References

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Manufactured in Canada by:

Affinity Biologicals Inc. **OFFICE** 905.304.9896
 1348 Sandhill Drive **FAX** 905.304.9897
 Ancaster, Ontario **TOLL FREE** 800.903.6020
 L9G 4V5 Canada **AFFINITYBIOLOGICALS.COM**