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AS160 [pT642] Rabbit Polyclonal Antibody

Store at 2°C to 8°C (short-term), or –20°C (long-term)

Catalog Number: 44-1071G

Pub. No. MAN0005662 **Rev.** 1.0

Clonality: Polyclonal **Quantity**: 10 mini-blot size **Volume**: 100 µL

Host/Class: Rabbit IgG **Reactivity**: Human AS160 [pT642] **Predicted Reactivity**: Human, Mouse

Product description

AS160 is a substrate of Akt containing a Rab GTPase-activating protein domain (GAP). It plays a role in membrane trafficking by regulating insulin-stimulated exocytosis of glucose transporter GLUT4. Insulin-stimulated phosphorylation of AS160 on threonine 642 is critical for mediating GLUT4 translocation in fat and muscle cells, by blocking Rab GAP function. Phosphorylation of threonine 642 is also important in mediating AS160 redistribution from low density microsomes to the cytosol in adipocytes.

Product specifications

Immunogen:	A chemically synthesized phosphopeptide derived from the region of human AS160 containing threonine 642
Purification:	Antibody negatively preadsorbed using a non-phosphopeptide then purified by epitope-specific affinity chromatography
Apparent MW:	160 kDa
Sequence Identity:	Human
Sequence Homology:	Mouse
Lot:	See product label

Product applications

The antibody has been used in western blotting (1:1000 dilution). Other applications may work but have not been tested.

Because conditions may vary, it is recommended that each investigator determine the optimal amount of antibody to be used for each application.

Storage and handling

Store the antibody at 2°C to 8°C for up to 1 week, or apportion into working aliquots and keep at –20°C for long-term storage. Avoid repeated freezing and thawing.

Stability

When stored as instructed, expires one year from date of receipt unless otherwise indicated on the Certificate of Analysis.

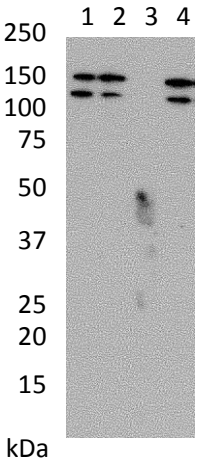

Figure 1 Peptide Competition

Extracts HeLa cells treated with 180 nM insulin for 15 minutes were resolved on a 10% Tris-glycine gel and transferred to PVDF. Membranes were blocked with 5% BSA-TBST for one hour at room temperature, then incubated with the AS160 [pS642] antibody for 2 hrs at room temperature in 3% BSA-TBST, following prior incubation with: no peptide (lane 1), the phosphopeptide immunogen (lane 2), a non-phosphorylated peptide corresponding to the immunogen (lane 3), or a generic phosphothreonine-containing peptide (lane 4). The blots were developed using chemiluminescence (ECL) method with a goat $F(ab')_2$ anti-rabbit IgG HRP conjugate (Cat. no. ALI4404).

Only the phosphopeptide corresponding to AS160 [pS642] blocks the antibody signal (lane 2) demonstrating the specificity of the antibody.

Positive controls used

HeLa cells treated with 180 nM insulin for 15minutes.

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Storage buffer

Dulbecco's phosphate buffered saline (without Mg^{2+} and Ca^{2+}), pH 7.3 (+/– 0.1), 50% glycerol with 1.0 mg/mL BSA (IgG, protease free) as a carrier, and 0.05% sodium azide.

CAUTION! Sodium azide is extremely toxic and may react with lead and copper plumbing to form highly explosive metal azides. Properly dispose of solutions containing sodium azide. Read the Safety Data Sheet (SDS) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. SDSs are available at **www.lifetechnologies.com/support**.

References

1. Zeigerer, A. et al. (2004) Insulin stimulation of GLUT4 exocytosis, but not its inhibition of endocytosis, is dependent on RabGAP AS160. *Mol. Biol. Cell*. 15(10):4406-4415.
2. Sano, H. et al. (2003) Insulin-stimulated phosphorylation of a Rab GTPase-activating protein regulates GLUT4 translocation. *J. Biol. Chem*. 278(17):14599-14602.
3. Kane, S. et al. (2002) A method to identify serine kinase substrates. Akt phosphorylates a novel adipocyte protein with a Rab GTPase-activating protein (GAP) domain. *J. Biol. Chem*. 277(25):22115-22118.
4. Lavan, B.E. et al (1997) A novel 160-kDa phosphotyrosine protein in insulin-treated embryonic kidney cells is a new member of the insulin receptor substrate family. *J. Biol. Chem*. 272(34):21403-21407.

Related products

Product Name	Quantity	Cat. No.
AKT/PKB1 [pS473] Rabbit pAb	100 µL	44623G
AKT/PKB [pT308] Rabbit pAb	10 blots	44602G
AKT (Pan) Rabbit pAb	200 µL	44609G
IRS-1 [pS312] Rabbit Polyclonal Antibody	100 µL	44814G
IRS-1 [pY612] Polyclonal Antibody, Rabbit	100 µL	44816G
IR/IGF1R [pY1158] Rabbit Polyclonal Antibody	10 blots	44802G
IR/IGF1R [pYpYpY1158/1162/1163] Rabbit Polyclonal Antibody	10 blots	44806G
PRAS40 [pT246] Polyclonal Antibody, Rabbit	10 blots	441100G
PTEN [pS370] Rabbit Polyclonal Antibody	10 blots	441060G
PTEN [pSpTpS380/382/385] Rabbit Polyclonal Antibody	10 blots	441066G
Glycogen Synthase [pSpS641/645] Rabbit Polyclonal Antibody	10 blots	441092G
PED/PEA-15 [pS116] Rabbit Polyclonal Antibody	10 blots	44836G

Product documentation

To obtain a Certificate of Analysis or Safety Data Sheets (SDSs), visit **www.lifetechnologies.com/support.**

Important licensing information

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Explanation of symbols

Symbol	Description	Symbol	Description
	Manufacturer		Catalog number
	Use by		Temperature limitation
	Consult instructions for use		Caution, consult accompanying documents
	Batch code		