Mouse Adiponectin, Recombinant Protein

Product Data Sheet

Cat. No.: RD272023100

Introduction:

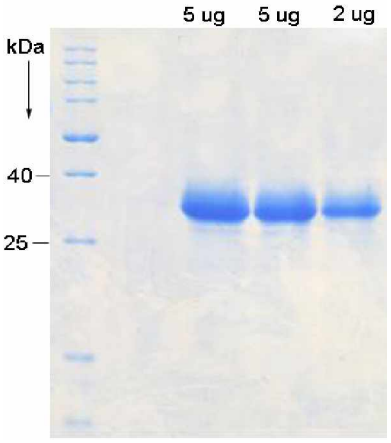
Adiponectin (also called ACRP30 and GBP28) is a protein exclusively secreted from adipose tissue. Full-length adiponectin was expressed in HEK293 cells stably transfected with pcdm-F vector encoding FLAG epitope-tagged mouse adiponectin. Recombinant adiponectin was purified using anti-FLAG M_2 affinity gel (Sigma-Aldrich) and was eluted with FLAG peptide. FLAG peptides were removed by an extensive wash with an excessive volume of saline. Highly sensitive matrix-assisted laser desorption/ionization time of flight (MALDI-TOF) mass-spectrometric analysis confirmed that FLAG peptide was not detectable in the protein solution.

Specificity: The recombinant mouse adiponectin is 100% homologous with the Mouse serum adiponectin

Source: HEK293 (Human embryonic kidney cell line)

Protein Content: 0.1 mg (determined by BCA method)

Purity: Purity of recombinant mouse adiponectin is >98% (HPLC and SDS PAGE analyzed).



SDS-PAGE separation of mouse adiponectin purified from HEK293 cells.

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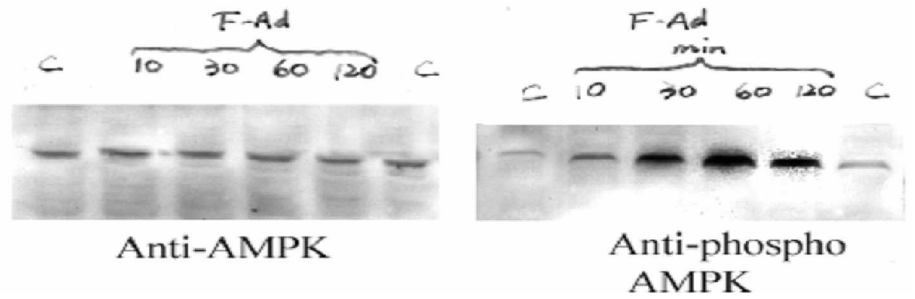
Activity *in vitro*:

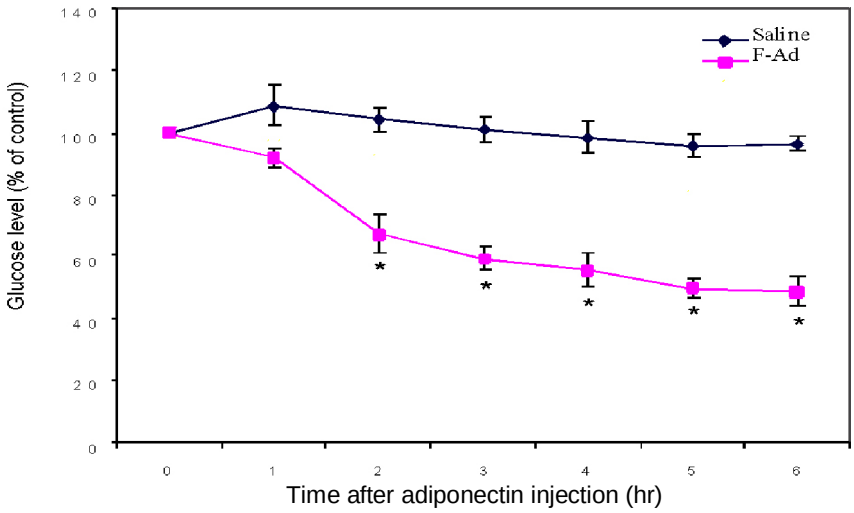
Full-length adiponectin has been shown to activate AMP-activated protein kinase in hepatocyte (Yamauchi T. *et al*., 2002). It can also activate AMPK in HepG2 human hepatocytes at the concentration of as low as 1.0 μg/ml.



Activation of AMP-activated kinase by full-length adiponectin purified from HEK293 cells. Hep G2 cells grown in a 24 well dish was starved in a serum-free medium for 16 hr, and then stimulated with 1 μg/ml adiponectin for different period. The cell lysates harvested were separated by 10% SDS-PAGE, and then probed with either anti-AMPK or anti-phospho AMPK. C: control; F-Ad: full-length adiponectin.

Activity *in vivo*:

Full-length adiponectin purified from mammalian cells can acutely decrease blood glucose levels (Berg A.H. *et al*., 2001). Single subcutaneous injection of adiponectin at the dose of 30 μg/g body weight significantly decreased the blood glucose levels in C57 mice.



The glucose-lowering effect of adiponectin on C57 mice. Adiponectin was injected into male C57 mice at 10 weeks as described in the text. At different times after injection, glucose glucose levels were measured via tail vain (n=3).

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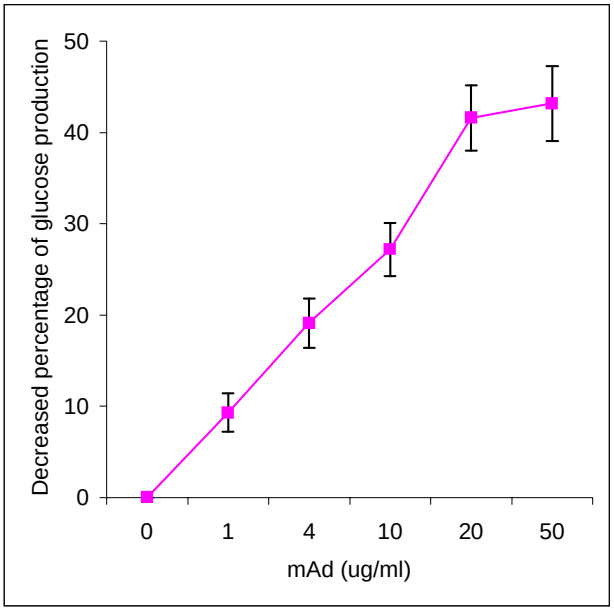
In vitro gluconeogenesis assay in primary rat hepatocytes was performed, showing the murine adiponectin derived from mammalian cells can inhibit glucose production.



Effect of murine adiponectin on gluconeogenesis in primary rat hepatocytes.
Primary rat hepatocytes were isolated and maintained as we described previously (Wang *et al.,* 2002). The cells were then treated with 50 pM insulin plus different concentration of murine adiponectin purified from HEK293 cells for 48 hours. At the end of treatment, the cells were incubated with a glucose-free medium for 6 hours, and glucose production was measured as we described previously (n=4-6).

Formulation:	Sterile filtered and lyophilized from 0.5 mg/ml in TBS buffer
Reconstitution:	Add 0.2 ml of deionized water and let the lyophilized pellet dissolve completely.
Storage:	Store lyophilized protein at -20°C. Aliquot the product after reconstitution to avoid repeated freezing/thawing cycles. Reconstituted protein can be stored at 4°C for a limited period of time; it does not show any change after two weeks at 4°C.
Stability/Shelf Life:	The lyophilized protein remains stable until the expiry date when stored at -20°C.

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Quality Control Test:	BCA - to determine content of the protein SDS PAGE - to determine purity of the protein
Applications:	ELISA, Western blotting, cell culture and/or animal studies
Note:	The recombinant mouse adiponectin is for research use only.

References:

- Xu A, Wang Y, Keshaw H, Xu LY, Lam KS, Cooper GJ. The fat-derived hormone adiponectin alleviates alcoholic and nonalcoholic fatty liver diseases in mice. J Clin Invest. 2003;112:91-100.

- Wang Y, Xu A, Knight C, Xu LY, Cooper GJ. Hydroxylation and glycosylation of the four conserved lysine residues in the collagenous domain of adiponectin. Potential role in the modulation of its insulin-sensitizing activity. J Biol Chem. 2002;277:19521-19529.

- Yamauchi T, Kamon J, Minokoshi Y, Ito Y, Waki H, Uchida S, Yamashita S, Noda M, Kita S, Ueki K, Eto K, Akanuma Y, Froguel P, Foufelle F, Ferre P, Carling D, Kimura S, Nagai R, Kahn BB, Kadowaki T. Adiponectin stimulates glucose utilization and fatty-acid oxidation by activating AMP-activated protein kinase. Nat Med. 2002;8:1288-1295.

- Berg AH, Combs TP, Du X, Brownlee M, Scherer PE. The adipocyte-secreted protein Acrp30 enhances hepatic insulin action. Nat Med. 2001;7:947-953.

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