

# ADPN siRNA (h): sc-60129

## BACKGROUND

ADPN, a member of the Adiponutrin family, displays lipase activity that is dependent upon the presence of an activated serine residue. D-glucose elicits a seven-fold increase in ADPN mRNA levels, and Insulin has a slight effect on ADPN expression in the presence or absence of glucose. The glucose-induced increase in ADPN expression can be reversed by factors known to raise intracellular cAMP. mRNA ADPN levels are negatively correlated with fasting glucose levels and subjects with high ADPN mRNA levels have increased Insulin sensitivity, implicating ADPN in obesity and diabetes. ADPN gene expression in humans is highly regulated by changes in energy balance. In mice adipocytes, ADPN parallels the expression of fatty acid synthase (FAS) and Srebp1c, a variant of Srebp1.

## REFERENCES

1. Baulande, S., et al. 2001. Adiponutrin, a transmembrane protein corresponding to a novel dietary- and obesity-linked mRNA specifically expressed in the adipose lineage. *J. Biol. Chem.* 276: 33336-33344.
2. Polson, D.A. and Thompson, M.P. 2003. Adiponutrin mRNA expression in white adipose tissue is rapidly induced by meal-feeding a high-sucrose diet. *Biochem. Biophys. Res. Commun.* 301: 261-266.
3. Polson, D. and Thompson, M. 2003. Adiponutrin gene expression in 3T3-L1 adipocytes is downregulated by troglitazone. *Horm. Metab. Res.* 35: 508-510.
4. Polson, D.A. and Thompson, M.P. 2004. Macronutrient composition of the diet differentially affects leptin and adiponutrin mRNA expression in response to meal feeding. *J. Nutr. Biochem.* 15: 242-246.

## CHROMOSOMAL LOCATION

Genetic locus: PNPLA3 (human) mapping to 22q13.31.

## PRODUCT

ADPN siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ADPN shRNA Plasmid (h): sc-60129-SH and ADPN shRNA (h) Lentiviral Particles: sc-60129-V as alternate gene silencing products.

For independent verification of ADPN (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-60129A, sc-60129B and sc-60129C.

## STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

ADPN siRNA (h) is recommended for the inhibition of ADPN expression in human cells.

## SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10  $\mu$ M in 66  $\mu$ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

## GENE EXPRESSION MONITORING

ADPN (D-5): sc-390251 is recommended as a control antibody for monitoring of ADPN gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ADPN gene expression knockdown using RT-PCR Primer: ADPN (h)-PR: sc-60129-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## SELECT PRODUCT CITATIONS

1. Bruschi, F.V., et al. 2017. The PNPLA3 I148M variant modulates the fibrogenic phenotype of human hepatic stellate cells. *Hepatology* 65: 1875-1890.

## RESEARCH USE

For research use only, not for use in diagnostic procedures.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.