

# ATGL siRNA (m): sc-60224

## BACKGROUND

The adiponutrin family members, which have been implicated in obesity and diabetes, consist of adiponutrin (ADPN), GS1, GS2, GS2-like, PNPLA1, and adipose triglyceride lipase (ATGL), also designated desnutrin or patatin-like phospholipase domain-containing protein 2 (PLNPA2). ATGL is a 486-amino acid protein that is highly expressed in mouse and human adipose tissue. It contains a highly conserved 180-amino acid N-terminal patatin domain common to plant acyl-hydrolases with a glycine-rich region, an aspartate active site motif, and an active serine hydrolase motif. Along with hormone-sensitive lipase, ATGL catabolizes stored triglycerides in mammalian adipose tissue. The lipase activity of ATGL is dependent upon the presence of an activated serine residue. ADPN and ATGL are oppositely regulated by Insulin, where upregulation of ATGL and downregulation of ADPN occurs when fasting.

## REFERENCES

1. Zimmermann, R., et al. 2004. Fat mobilization in adipose tissue is promoted by adipose triglyceride lipase. *Science* 306: 1383-1386.
2. Smirnova, E., et al. 2005. ATGL has a key role in lipid droplet/adiposome degradation in mammalian cells. *EMBO Rep.* 7: 106-113.
3. Langin, D., et al. 2005. Adipocyte lipases and defect of lipolysis in human obesity. *Diabetes* 54: 3190-3197.
4. Lake, A.C., et al. 2005. Expression, regulation, and triglyceride hydrolase activity of adiponutrin family members. *J. Lipid Res.* 46: 2477-2487.
5. Gronke, S., et al. 2005. Brummer lipase is an evolutionary conserved fat storage regulator in *Drosophila*. *Cell Metab.* 1: 323-330.

## CHROMOSOMAL LOCATION

Genetic locus: Pnpla2 (mouse) mapping to 7 F5.

## PRODUCT

ATGL siRNA (m) 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 ATGL shRNA Plasmid (m): sc-60224-SH and ATGL shRNA (m) Lentiviral Particles: sc-60224-V as alternate gene silencing products.

For independent verification of ATGL (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-60224A, sc-60224B and sc-60224C.

## 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

ATGL siRNA (m) is recommended for the inhibition of ATGL expression in mouse 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

ATGL (F-7): sc-365278 is recommended as a control antibody for monitoring of ATGL 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™ Molecular Weight Standards: sc-2035, UltraCruz® 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® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ATGL gene expression knockdown using RT-PCR Primer: ATGL (m)-PR: sc-60224-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. Lettieri Barbato, D., et al. 2014. Proline oxidase-adipose triglyceride lipase pathway restrains adipose cell death and tissue inflammation. *Cell Death Differ.* 21: 113-123.
2. Lettieri Barbato, D., et al. 2014. Inhibition of age-related cytokines production by ATGL: a mechanism linked to the anti-inflammatory effect of resveratrol. *Mediators Inflamm.* 2014: 917698.
3. Lettieri-Barbato, D., et al. 2018. Time-controlled fasting prevents aging-like mitochondrial changes induced by persistent dietary fat overload in skeletal muscle. *PLoS ONE* 13: e0195912.

## RESEARCH USE

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