

# AT<sub>1</sub> siRNA (h): sc-29750

## BACKGROUND

Angiotensin II (Ang II) is an important physiological effector of blood pressure and volume regulation through vasoconstriction, aldosterone release, sodium uptake and thirst stimulation. Although Ang II interacts with two types of cell surface receptors, AT<sub>1</sub> and AT<sub>2</sub>, most of the major cardiovascular effects seem to be mediated through AT<sub>1</sub>. Molecular cloning of the AT<sub>1</sub> protein has shown it to be a member of the G protein-associated seven transmembrane protein receptor family. Ang II treatment of cells results in activation of several signal transduction pathways as evidenced by tyrosine phosphorylation of several proteins and induction of others. PLC $\gamma$  is phosphorylated after 30 seconds of treatment with Angiotensin II, indicating this as an early signal transduction event. Ang II treatment also stimulates phosphorylation of Shc, FAK and MAP kinases, and induces MKP-1, indicating stimulation of growth factor pathways. Ang II stimulation through AT<sub>1</sub> has been shown to activate the JAK/Stat pathway involving a direct interaction between JAK2 and AT<sub>1</sub> as demonstrated by coimmunoprecipitation. The AT<sub>1</sub> receptor has no cytoplasmic kinase domain, but is able to function as a substrate for Src kinases and has several putative phosphorylation sites.

## CHROMOSOMAL LOCATION

Genetic locus: AGTR1 (human) mapping to 3q24.

## PRODUCT

AT<sub>1</sub> 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 AT<sub>1</sub> shRNA Plasmid (h): sc-29750-SH and AT<sub>1</sub> shRNA (h) Lentiviral Particles: sc-29750-V as alternate gene silencing products.

For independent verification of AT<sub>1</sub> (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-29750A, sc-29750B and sc-29750C.

## 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

AT<sub>1</sub> siRNA (h) is recommended for the inhibition of AT<sub>1</sub> expression in human cells.

## PROTOCOLS

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

## 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.

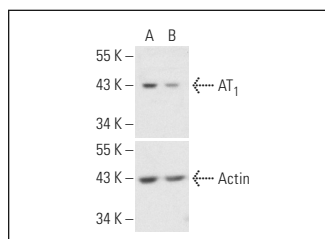
## GENE EXPRESSION MONITORING

AT<sub>1</sub> (G-3): sc-515884 is recommended as a control antibody for monitoring of AT<sub>1</sub> 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).

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor AT<sub>1</sub> gene expression knockdown using RT-PCR Primer: AT<sub>1</sub> (h)-PR: sc-29750-PR (20  $\mu$ l, 452 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## DATA



AT<sub>1</sub> siRNA (h): sc-29750. Western blot analysis of AT<sub>1</sub> expression in non-transfected control (A) and AT<sub>1</sub> siRNA transfected (B) HeLa cells. Blot probed with AT<sub>1</sub> (N-10): sc-1173. Actin (I-19): sc-1616 used as specificity and loading control.

## SELECT PRODUCT CITATIONS

- Li, X.C., et al. 2006. AT<sub>1</sub> receptor-mediated accumulation of extracellular Angiotensin II in proximal tubule cells: role of cytoskeleton microtubules and tyrosine phosphatases. *Am. J. Physiol. Renal Physiol.* 291: F375-F383.
- Haas, M.J., et al. 2016. Angiotensin II receptor one (AT<sub>1</sub>) mediates dextrose induced endoplasmic reticulum stress and superoxide production in human coronary artery endothelial cells. *Int. J. Cardiol.* 220: 842-850.
- Liu, J., et al. 2021. Activation of RAS contributes to peritoneal fibrosis via dysregulation of low-density lipoprotein receptor. *Am. J. Physiol. Renal Physiol.* 320: F273-F284.

## RESEARCH USE

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