

Angiotensinogen siRNA (h): sc-37196

BACKGROUND

Angiotensin is formed from a precursor, angiotensinogen, which is produced by the liver and found in the α -globulin fraction of plasma. The lowering of blood pressure is a stimulus to secretion of Renin by the kidney into the blood. Renin cleaves from angiotensinogen a terminal decapeptide, Angiotensin I (Ang I). This is further altered by the enzymatic removal of a dipeptide to form Angiotensin II (Ang II). Screening a panel of human-mouse somatic cell hybrids confirmed the assignment of the AGT locus to human chromosome 1. Angiotensin, an octapeptide hormone, is an important physiological effector of blood pressure and volume regulation through vasoconstriction, aldosterone release, sodium uptake and thirst stimulation. It has been shown that mechanical stress causes release of Angiotensin from cardiac myocytes and that Angiotensin acts as an initial mediator of the hypertrophic response. Angiotensin treatment also stimulates phosphorylation of Shc, FAK and MAP kinases and induces MKP-1, indicating stimulation of growth factor pathways. Angiotensin stimulation through AT1 has been shown to activate the JAK/Stat pathway involving a direct interaction between JAK2 and AT1 as demonstrated by co-immunoprecipitation.

REFERENCES

1. Tsuda, T., et al. 1991. Vasoconstrictor-induced protein-tyrosine phosphorylation in cultured vascular smooth muscle cells. *FEBS Lett.* 285: 44-48.
2. Abonia, J.P., et al. 1993. Linkage of Agt and Actsk-1 to distal mouse chromosome 8 loci: a new conserved linkage. *Mamm. Genome* 4: 25-32.
3. Sadoshima, J., et al. 1993. Autocrine release of angiotensin II mediates stretch-induced hypertrophy of cardiac myocytes *in vitro*. *Cell* 75: 977-984.
4. Duff, J.L., et al. 1993. Angiotensin II induces 3CH134, a protein-tyrosine phosphatase, in vascular smooth muscle cells. *J. Biol. Chem.* 268: 26037-26040.
5. Schorb, W., et al. 1994. Angiotensin II-induced protein tyrosine phosphorylation in neonatal rat cardiac fibroblasts. *J. Biol. Chem.* 269: 19626-19632.
6. Marrero, M.B., et al. 1995. Direct stimulation of Jak/Stat pathway by the angiotensin II AT1 receptor. *Nature* 375: 247-250.

CHROMOSOMAL LOCATION

Genetic locus: AGT (human) mapping to 1q42.2.

PRODUCT

Angiotensinogen 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Angiotensinogen shRNA Plasmid (h): sc-37196-SH and Angiotensinogen shRNA (h) Lentiviral Particles: sc-37196-V as alternate gene silencing products.

For independent verification of Angiotensinogen (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-37196A, sc-37196B and sc-37196C.

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

APPLICATIONS

Angiotensinogen siRNA (h) is recommended for the inhibition of Angiotensinogen 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 μ M in 66 μ 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

Angiotensinogen (H-12): sc-374511 is recommended as a control antibody for monitoring of Angiotensinogen 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 κ BP-HRP: sc-516102 or m-IgG κ 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 κ BP-FITC: sc-516140 or m-IgG κ 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 Angiotensinogen gene expression knockdown using RT-PCR Primer: Angiotensinogen (h)-PR: sc-37196-PR (20 μ l, 451 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

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

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.