

## PAR-2 siRNA (h): sc-36188

### BACKGROUND

Thrombin receptor (also designated PAR-1, for protease-activated receptor-1), PAR-2 and PAR-3 compose a distinct class of G protein-coupled receptors that are activated by proteolysis. Cleavage of this class of seven transmembrane receptors occurs at the amino-terminal extracellular domain by proteases including Thrombin and Trypsin. Thrombin is a serine protease involved in platelet aggregation and blood coagulation that activates the Thrombin receptor, resulting in elevated intracellular calcium levels in platelets. Thrombin has also been demonstrated to cleave PAR-3 *in vitro*, suggesting that PAR-3 may be involved in thrombosis or mitogenesis. It has been demonstrated that Thrombin receptor and PAR-4 account for most Thrombin signaling in platelets. Activation of PAR-2 *in vitro* is induced by Trypsin and to a lesser extent by Thrombin, suggesting that PAR-2 is not an alternative Thrombin receptor. Cytokines including TNF $\alpha$  and IL-1 $\beta$  increase PAR-2 expression, indicating that PAR-2 may be involved in the acute inflammatory response.

### REFERENCES

1. Santulli, R.J., et al. 1995. Evidence for the presence of a protease-activated receptor distinct from the thrombin receptor in human keratinocytes. *Proc. Natl. Acad. Sci. USA* 92: 9151-9155.
2. Lerner, D.J., et al. 1996. Agonist recognition by proteinase-activated receptor 2 and thrombin receptor. Importance of extracellular loop interactions for receptor function. *J. Biol. Chem.* 271: 13943-13947.

### CHROMOSOMAL LOCATION

Genetic locus: F2RL1 (human) mapping to 5q13.3.

### PRODUCT

PAR-2 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 PAR-2 shRNA Plasmid (h): sc-36188-SH and PAR-2 shRNA (h) Lentiviral Particles: sc-36188-V as alternate gene silencing products.

For independent verification of PAR-2 (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-36188A, sc-36188B and sc-36188C.

### 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

PAR-2 siRNA (h) is recommended for the inhibition of PAR-2 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

PAR-2 (SAM11): sc-13504 is recommended as a control antibody for monitoring of PAR-2 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 PAR-2 gene expression knockdown using RT-PCR Primer: PAR-2 (h)-PR: sc-36188-PR (20  $\mu$ l, 107 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

### SELECT PRODUCT CITATIONS

1. Uehara, A., et al. 2005. Arginine-specific gingipains from *Porphyromonas gingivalis* stimulate production of hepatocyte growth factor (scatter factor) through protease-activated receptors in human gingival fibroblasts in culture. *J. Immunol.* 175: 6076-6084.
2. Kuckleburg, C.J. and Newman, P.J. 2013. Neutrophil proteinase 3 acts on protease-activated receptor-2 to enhance vascular endothelial cell barrier function. *Arterioscler. Thromb. Vasc. Biol.* 33: 275-284.
3. Chen, K.D., et al. 2014. Interconnections between autophagy and the coagulation cascade in hepatocellular carcinoma. *Cell Death Dis.* 5: e1244.
4. Bach, N., et al. 2015. Cytokine responses induced by diesel exhaust particles are suppressed by PAR-2 silencing and antioxidant treatment, and driven by polar and non-polar soluble constituents. *Toxicol. Lett.* 238: 72-82.
5. Nieuwenhuizen, L., et al. 2016. Silencing of protease-activated receptors attenuates synovitis and cartilage damage following a joint bleed in haemophilic mice. *Haemophilia* 22: 152-159.
6. Huang, K.T., et al. 2017. Factor VII-induced microRNA-135a inhibits autophagy and is associated with poor prognosis in hepatocellular carcinoma. *Mol. Ther. Nucleic Acids* 9: 274-283.
7. Natarajan, K., et al. 2019. Organic dust induces inflammatory gene expression in lung epithelial cells via Ros-dependent Stat3 activation. *Am. J. Physiol. Lung Cell. Mol. Physiol.* 317: L127-L140.

### RESEARCH USE

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