# SANTA CRUZ BIOTECHNOLOGY, INC.

# GABA<sub>A</sub> Ry2 siRNA (h): sc-42449



## BACKGROUND

GAD-65 and GAD-67, glutamate decarboxylases, function to catalyze the production of GABA ( $\gamma$ -aminobutyric acid). In the central nervous system, GABA functions as the main inhibitory transmitter by increasing a Cl<sup>-</sup> (chloride) conductance that inhibits neuronal firing. GABA has been shown to activate both ionotropic (GABA<sub>A</sub>) and metabotropic (GABA<sub>B</sub>) receptors, as well as a third class of receptors called GABA<sub>C</sub>. The  $\gamma$  subunit of GABA<sub>A</sub> receptors are important for benzodiazepine binding and modulation of GABA-mediated Cl<sup>-</sup> current. GABA<sub>A</sub> R $\gamma$ 2 is a 467 amino acid mulit-pass membrane protein localized to the postsynaptic cell membrane. Present as a pentamer with other GABA<sub>A</sub> receptor chains ( $\alpha$ ,  $\beta$ ,  $\gamma$ ,  $\delta$  and  $\rho$ ), the GABA<sub>A</sub> ligand-gated Cl<sup>-</sup> channels selectively complex with D5DR to enable mutual inhibitory functional interactions between the two receptor systems. Defects in the gene encoding GABA<sub>A</sub> R $\gamma$ 2 have been found to be the cause of childhood absence epilepsy type 2, familial febrile convulsions type 8, generalized epilepsy with febrile seizures plus type 3 and severe myoclonic epilepsy in infancy.

## REFERENCES

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- 3. Kucken, A.M., et al. 2000. Identification of benzodiazepine binding site residues in the  $\gamma$ 2 subunit of the GABA<sub>A</sub> receptor. Mol. Pharmacol. 57: 932-939.
- Liu, F., et al. 2000. Direct protein-protein coupling enables cross-talk between dopamine D5 and GABA<sub>A</sub> receptors. Nature 403: 274-280.
- 5. Baulac, S., et al. 2001. First genetic evidence of  $GABA_A$  receptor dysfunction in epilepsy: a mutation in the  $\gamma 2$  subunit gene. Nat. Genet. 28: 46-48.
- Kananura, C., et al. 2002. A splice-site mutation in GABRG2 associated with childhood absence epilepsy and febrile convulsions. Arch. Neurol. 59: 1137-1141.
- 7. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 137164. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
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- 9. Audenaert, D., et al. 2006. A novel GABRG2 mutation associated with febrile seizures. Neurology 67: 687-690.

## CHROMOSOMAL LOCATION

Genetic locus: GABRG2 (human) mapping to 5q34.

## PROTOCOLS

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

## PRODUCT

GABA<sub>A</sub> R<sub>Y</sub>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 GABA<sub>A</sub> R<sub>Y</sub>2 shRNA Plasmid (h): sc-42449-SH and GABA<sub>A</sub> R<sub>Y</sub>2 shRNA (h) Lentiviral Particles: sc-42449-V as alternate gene silencing products.

For independent verification of GABA<sub>A</sub> Ry2 (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-42449A, sc-42449B and sc-42449C.

## 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**

GABA\_A Ry2 siRNA (h) is recommended for the inhibition of GABA\_A Ry2 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.

## **RT-PCR REAGENTS**

Semi-quantitative RT-PCR may be performed to monitor GABA<sub>A</sub> R<sub>Y</sub>2 gene expression knockdown using RT-PCR Primer: GABA<sub>A</sub> R<sub>Y</sub>2 (h)-PR: sc-42449-PR (20  $\mu$ I). 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.