

## Product Information

### Anti-GABA<sub>A</sub> Receptor (β3 subunit), Cytosolic Loop

Developed in Rabbit, Affinity Isolated Antibody

Product Number **G 9669**

#### Product Description

Anti-GABA<sub>A</sub> Receptor (β3 subunit), cytosolic loop, is developed in rabbit using a fusion protein with the amino acid sequence representing the cytosolic loop of the rat GABA<sub>A</sub> receptor (β3 subunit) as immunogen. The antiserum is affinity purified using affinity columns containing the appropriate amino acid sequence of the antigen.

The antibody specifically detects GABA<sub>A</sub> receptor β3 subunit (protein with apparent molecular mass of 50-56 kDa) in rat brain membrane fractions. It has been used in immunoblotting applications.

GABA is the major inhibitory neurotransmitter in the mammalian brain where it acts at GABA<sub>A</sub> receptors, which are ligand-gated chloride channels. Chloride conductance of these channels can be modulated by agents such as benzodiazepines that bind to the GABA<sub>A</sub> receptor. The inhibitory neurotransmitter GABA (γ-aminobutyric acid) signals through two distinct types of pre- and postsynaptic receptors, GABA<sub>A</sub> and GABA<sub>B</sub>. Both GABA receptors can mediate depression of synaptic transmission and contribute to the inhibition controlling neuronal excitability. GABA<sub>A</sub> and GABA<sub>B</sub> receptors differ with regard to their ionic characteristics and pharmacological properties. The GABA<sub>A</sub> receptor is an ionotropic receptor that forms the GABA gated chloride channel and consists of several heterogeneous subunits with membrane recognition sites for benzodiazepines. Over the past decade, a family of GABA<sub>A</sub> receptor subtypes has been delineated. These subtypes are generated by the co-assembly of five polypeptides selected from the α1-α6, β1-β3, γ1-γ3, δ, ε, π, and θ subunits.

Zinc ions regulate GABA<sub>A</sub> receptors by inhibiting receptor function via an allosteric mechanism that is critically dependent on the receptor subunit composition. Three discrete sites mediate zinc inhibition: one is located within the ion channel and comprises subunit β-3 His267 and Glu270, and the other 2 are on the external N-terminal face of the receptor and require the coordination of subunit α-1 Glu137 and His141 and β-3 Glu182. The

characteristically low zinc sensitivity of GABA<sub>A</sub> receptors containing the γ-2 subunit results from disruption of 2 of the 3 sites after subunit assembly. It has been demonstrated that 3 human GABA<sub>A</sub> receptor subunit genes, GABRB3, GABRA5, and GABRG3, are expressed exclusively from the paternal allele and that UBE3A is biallelically expressed. Analysis for a marker of GABRB3 called 155CA-2 in autistic families demonstrated association between autistic disorder and 155CA-2. β-3 subunit had been also implicated in sleep processes and the mice lacking β-3 lose the hypnotic response to oleamide.

#### Reagent

The antibody is supplied in 10 mM HEPES, pH 7.5, 150 mM NaCl, 100 μg/ml BSA and 50% glycerol.

#### Storage/Stability

Store at -20 °C. Due to the presence of 50% glycerol the antibody will remain in solution. For extended storage, centrifuge the vial briefly before opening and prepare working aliquots. The antibody is stable for at least 24 months when stored at -20 °C. Defrosted aliquots in use should be stored at 4 °C. Avoid repeated freezing and thawing.

#### Product Profile

The supplied reagent is sufficient for 10 immunoblots.

A recommended working dilution of 1:1000 is determined by immunoblotting using rat brain membrane fractions

**Note:** In order to obtain best results in different techniques and preparations we recommend determining optimal working concentration by a titration test.

## Results

In immunoblot analysis, the Anti-GABA<sub>A</sub>-R  $\beta$ 3 subunit labeled the 50-56 kDa  $\beta$ 3 subunit of GABA<sub>A</sub>-R in both a control brain lysate and in a sample of the purified receptor. However, immunolabeling was absent in a brain lysate prepared from animals in which the  $\beta$ 3 subunit had been knocked out.

## References

1. Buhr, A., et al., Functional characterization of the new human GABA(A) receptor mutation beta-3(R192H). *Hum. Genet.*, **111**, 154-160, (2002).
2. Buxbaum, J. D. Association between a GABRB3 polymorphism and autism. *Molec. Psychiat.*, **7**, 311-316, (2002).
3. Hosie, A. M. et al., Zinc-mediated inhibition of GABA-A receptors: discrete binding sites underlie subtype specificity. *Nature Neurosci.*, **6**, 362-369 (2003).
4. Laposky, A. D. et al., Deletion of the GABA(A) receptor beta 3 subunit eliminates the hypnotic actions of oleamide in mice. *Neuroreport*, **12**, 4143-4147 (2001).

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