

## Product Information

# Monoclonal Anti-GAD65, Clone N-GAD65

Produced in Mouse, Purified Immunoglobulin

**SAB4200232**

## Product Description

Monoclonal Anti-GAD65 (mouse IgG1 isotype) is derived from the hybridoma N-GAD65 produced by the fusion of mouse myeloma cells and splenocytes from BALB/c mice immunized with a synthetic peptide corresponding to a fragment of human GAD65 (GeneID: 2572), conjugated to KLH. The corresponding sequence differs by one amino acid in rat and two amino acids in mouse and monkey. The isotype is determined by ELISA using Mouse Monoclonal Antibody Isotyping Reagents, Product Number ISO2. The antibody is purified from culture supernatant of hybridoma cells grown in a bioreactor.

Monoclonal Anti-GAD65 recognizes human, rat, mouse, and monkey GAD65. The antibody may be used in several immunochemical techniques including immunoblotting (~65 kDa), immunoprecipitation, immunohistochemistry, and radioligand binding assay.<sup>1-3</sup>

Glutamic Acid Decarboxylase (GAD) catalyzes the conversion of L-glutamate to  $\gamma$ -aminobutyric acid (GABA), the principal inhibitory neurotransmitter in the brain, and a putative paracrine signal molecule in pancreatic islets. GAD has a restricted tissue distribution. It is highly expressed in the cytoplasm of GABAergic neurons in the central nervous system (CNS) and pancreatic  $\beta$ -cells, but is also present in other non-neuronal tissues such as testis, oviduct, and ovary. GAD is also transiently expressed in non-GABAergic cells of the embryonic and adult nervous system, suggesting its involvement in development and plasticity. GAD exists as two isoforms, GAD65 and GAD67 (molecular masses of 65 kDa and 67 kDa, respectively) that are encoded by two different genes. GAD65 is an amphiphilic, membrane anchored protein, (585 amino acid residues) and is encoded on human chromosome 10. GAD67 is a cytoplasmic protein (594 amino acid residues) and is encoded on chromosome 2. There is 64% amino acid identity between the two isoforms, with the highest diversity located at the N-terminus,

which in GAD65 is required for targeting the enzyme to GABA-containing secretory vesicles.

The two isoforms appear to have distinct intraneuronal distribution in the brain.<sup>4-6</sup> GAD65 has been identified as an autoantigen in insulin dependent diabetes mellitus (IDDM) and stiff-man syndrome (SMS).<sup>3</sup> Antibodies that react specifically with GAD65 are useful for the study of the differential tissue expression and intracellular localization of this isoform in normal and disease conditions.

## Reagent

Supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide as a preservative.

Antibody concentration: ~1.0 mg/mL

## Precautions and Disclaimer

For research use only. Not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

## Storage/Stability

Store at  $-20^{\circ}\text{C}$ . For continuous use, store at  $2-8^{\circ}\text{C}$  for up to one month. For extended storage, freeze at  $-20^{\circ}\text{C}$  in working aliquots. Repeated freezing and thawing, or storage in "frost-free" freezers, is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilution samples should be discarded if not used within 12 hours.

## Product Profile

### Immunoblotting:

A working antibody concentration of 0.25-0.5  $\mu\text{g/mL}$  is recommended using whole extracts of rat brain.

**Note:** In order to obtain best results in various techniques and preparations, it is recommended to determine optimal working dilutions by titration.

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## References

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3. Raju, R., et al., *J. Immunol.*, **175**, 7755-7762 (2005).
4. Reetz, A., et al., *EMBO J.*, **10**, 1275-1284 (1991).
5. Tillakaratne, N.J., et al., *J. Neurochem.*, **58**, 618-627 (1992).
6. Wei, J., and Wu, J.Y., *Neurochem. Res.*, **33**, 1459-1465 (2008).

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