

# Anti-GAD65/GAD2 Antibody Picoband™

Catalog Number: A03142

### **About GAD2**

Glutamate decarboxylase 2, also known as GAD65, is an enzyme that in humans is encoded by the GAD2 gene. This gene encodes one of several forms of glutamic acid decarboxylase, identified as a major autoantigen in insulin-dependent diabetes. The enzyme encoded is responsible for catalyzing the production of gamma-aminobutyric acid from L-glutamic acid. A pathogenic role for this enzyme has been identified in the human pancreas since it has been identified as an autoantibody and an autoreactive T cell target in insulin-dependent diabetes. This gene may also play a role in the stiff man syndrome. Alternative splicing results in multiple transcript variants that encode the same protein.

#### Overview

Product Name	Anti-GAD65/GAD2 Antibody Picoband™
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-GAD65/GAD2 Antibody Picoband™ catalog # A03142. Tested in IHC, WB applications. This antibody reacts with Human, Mouse, Rat.
Application	IHC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.
Storage Instructions	Store at -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freeze-thaw cycles.
Host	Rabbit
Uniprot ID	Q05329

### **Technical Details**

Immunogen	A synthetic peptide corresponding to a sequence at the N-terminus of human GAD65, different from the related mouse and rat sequences by one amino acid.
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P).
Cross Reactivity	No cross-reactivity with other proteins
Isotype	Rabbit IgG
Form	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.



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Suggested Dilutions	Dilute the sample so that the expected range of concentrations fall within the detection range of this kit.  If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples.  Some PubMed article(s) citing the expression level of this target are as follows:  Boster Bio's internal QC testing used:  Western blot, 0.1-0.5ug/ml, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 0.5-1ug/ml, Human, Mouse, Rat, By Heat
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### Anti-GAD65/GAD2 Antibody Picoband™ (A03142) Images

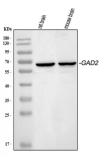


Figure 1. Western blot analysis of GAD65 using anti-GAD65 antibody (A03142).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

Lane 1: rat brain tissue lysates,

Lane 2: mouse brain tissue lysates.

After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-GAD65 antigen affinity purified polyclonal antibody (Catalog # A03142) at 0.5 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for GAD65 at approximately 65 kDa. The expected band size for GAD65 is at 65 kDa.

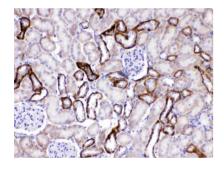


Figure 2. IHC analysis of GAD65 using anti-GAD65 antibody (A03142).

GAD65 was detected in paraffin-embedded section of rat kidney tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-GAD65 Antibody (A03142) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

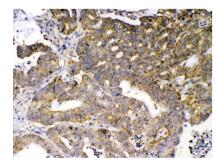


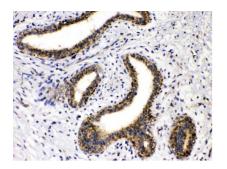
Figure 3. IHC analysis of GAD65 using anti-GAD65 antibody (A03142).

GAD65 was detected in paraffin-embedded section of human intetsinal cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-GAD65 Antibody (A03142) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

Figure 4. IHC analysis of GAD65 using anti-GAD65 antibody (A03142).

GAD65 was detected in paraffin-embedded section of human mammary cancer tissues. Heat mediated antigen retrieval





was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-GAD65 Antibody (A03142) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

# **3 Publications Citing This Product**

- 1. PubMed ID: 10.1007/s00540-011-1199-z, Propofol protects against impairment of learning-memory and imbalance of hippocampal Glu/GABA induced by electroconvulsive shock in depressed rats
- 2. PubMed ID: 21444355, Shi J, Ma Y, Zheng M, Ruan Z, Liu J, Tian S, Zhang D, He X, Li G. Toxicol Ind Health. 2012 Feb;28(1):10-20. Doi: 10.1177/0748233711401264. Epub 2011 Mar 28. Effect Of Sub-Acute Exposure To Acrylamide On Gabaergic Neurons And Astrocytes In Weaning ...
- 3. PubMed ID: 26199611, Acrylamide neurotoxicity on the cerebrum of weaning rats

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