NEUROMICS **Data Sheet**

			Bata Choot
Catalog Number:	GT41028	Host:	Goat
Product Type:	Affinity Purified Antibody	Species Reactivity:	Human, Mouse, Rat
Immunogen Sequence:	Peptide with sequence DSGNHCFKVYRYLQ, from the C Terminus of the TRIM2 protein sequence according to NP_056086.2; NP_001123539.1.	Format:	Liquid 200 ul. (0.5 mg/ml). Tris saline, 0.02% sodium azide, pH 7.3, 0.5% BSA
Applications:	Immunohistochemistry: 6.0-12.0 μg/ml (paraffin embedded tissue only). Western Blot: 0.5-2.0 μg/ml. Immunoprecipitation* Peptide ELISA: 1:16,000.		
	*Dilutions listed as a recommendation. Optimal dilution should be determined by investigator.		
Storage:	Aliquot and store at -20°C. Avoid repeated freeze-thaw cycles.		

Application Notes

This antibody is expected to recognise both reported isoforms NP_003072 and NP_570824.

Western Blot:

TRIM2

Approximately 80+85kDa bands observed in Rat Brain lysates (calculated MW of 81.5kDa according to Human NP_001123539.1, 84.4 accorindg to Human NP_056086.2 and 83.3 according to Rat NP_001102022.1).

Immunohistochemistry:

Tissue Preparation: Formalin fixation and embedding in paraffin wax.

Tissue Sectioning: Make 4-µm sections and place on pre-cleaned and charged microscope slides. Heat in a tissue-drying oven for 45 minutes at 60°C.

Deparaffinization: Wash dry slides in 3 changes of xylene - 5 minutes each at Room Temperature.

Rehydration: Wash slides in 3 changes of 100% alcohol - 3 minutes each at Room Temperature. Wash slides in 2 changes of 95% alcohol – 3 minutes each at Room Temperature Wash slides in 1 change of 80% alcohol – 3 minutes at Room Temperature. Rinse slides in gentle running distilled water - 5 minutes at Room Temperature.

Antigen retrieval: Steam slides in 0.01 M sodium citrate buffer, pH 6.0 at 99-100°C - 20 minutes. Remove from heat and let stand at room temperature in buffer - 20 minutes. Rinse in 1X TBS with Tween (TBST) - 1 minute at Room Temperature.

Immunostaining: (Do not allow tissues to dry at any time during the staining procedure). Apply a universal protein block - 20 minutes at Room Temperature. Drain protein block from slides, apply diluted primary antibody - 45 minutes at Room Temperature. Rinse slides in 1X TBST - 1 minute at Room Temperature. Apply a horse-anti-goat IgG , biotin secondary (HO30002)*- 30 minutes at Room Temperature. Rinse slides in 1X TBST - 1 minute at Room Temperature. Apply alkaline phosphatase streptavidin - 30 minutes at Room Temperature. Rinse slides in 1X TBST - 1 minute at Room Temperature. Apply alkaline phosphatase chromogen substrate - 30 minutes at Room Temperature. Wash slides in distilled water - 1 minute at Room Temperature

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Dehydrate: (This method should only be used if the chromogen substrate is alcohol insoluble (e.g. Vector Red, DAB). Wash slides in 2 changes of 80% alcohol – 1 minute each at Room Temperature. Wash slides in 2 changes of 95% alcohol – 1 minute each at Room Temperature. Wash slides in 3 changes of 100% alcohol – 1 minute each at Room Temperature. Wash slides in 3 changes of xylene – 1 minute each at Room Temperature. Apply coverslip

* Horse anti-goat IgG, Biotin (catalog# HO30002)

Image: TRIM2 (dilution: $6.3 \ \mu g/ml$) staining of paraffin embedded Human Cortex. Steamed antigen retrieval with citrate buffer pH 6, AP-staining. Note cytoplasm staining in neuronal cell bodies.

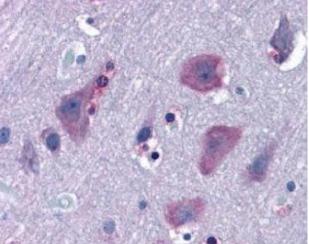
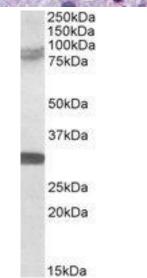


Image: TRIM2 (dilution: $0.5 \mu g/ml$) staining of Rat Brain lysate ($35\mu g$ protein in RIPA buffer). Primary incubation was 1 hour. Detected by chemiluminescence.



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