



---

<b>Catalog Number:</b>	RA30048	<b>Host:</b>	Rabbit
<b>Product Type:</b>	Affinity Purified Antibody	<b>Species Reactivity:</b>	Human
<b>Immunogen Sequence:</b>	Synthetic-KLH couple peptide from the 3rd extracellular domain of human GPR3	<b>Format:</b>	PBS, 0.1% sodium azide. Concentration 1 mg/ml.
<b>Applications:</b>	Immunohistochemistry: 40 µg/ml (Paraffin Embedded Only).  <i>Dilutions listed only as a recommendation. Optimal dilution should be determined by researcher.</i>		
<b>References:</b>	Long term: -70°C; Short term: +4°C. . Repeated freeze/thaw cycles compromise the integrity of the antibody.		

---

### Application Notes

#### Immunohistochemistry Protocol

*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 biotinylated anti-rabbit IgG (H+L) secondary – 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

*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

### FOR RESEARCH USE ONLY –V1-01/10

NEUROMICS REAGENTS ARE FOR IN VITRO AND CERTAIN NON-HUMAN IN VIVO EXPERIMENTAL USE ONLY AND NOT INTENDED FOR USE IN ANY HUMAN CLINICAL INVESTIGATION, DIAGNOSIS, PROGNOSIS, OR TREATMENT. THE ABOVE ANALYSES ARE MERELY TYPICAL GUIDES. THEY ARE NOT TO BE CONSTRUED AS BEING SPECIFICATIONS. ALL OF THE ABOVE INFORMATION IS, TO THE BEST OF OUR KNOWLEDGE, TRUE AND ACCURATE. HOWEVER, SINCE THE CONDITIONS OF USE ARE BEYOND OUR CONTROL, ALL RECOMMENDATIONS OR SUGGESTIONS ARE MADE WITHOUT GUARANTEE, EXPRESS OR IMPLIED, ON OUR PART. WE DISCLAIM ALL LIABILITY IN CONNECTION WITH THE USE OF THE INFORMATION CONTAINED HEREIN OR OTHERWISE, AND ALL SUCH RISKS ARE ASSUMED BY THE USER. WE FURTHER EXPRESSLY DISCLAIM ALL WARRANTIES OF MERCHANTABILITY AND FITNESS FOR A PARTICULAR PURPOSE.

**www.neuromics.com**

Neuromics • 5325 West 74<sup>th</sup> Street, Suite 8 • Edina, MN 55439  
phone 866-350-1500 • fax 612-677-3976 • e-mail [pshuster@neuromics1.com](mailto:pshuster@neuromics1.com)