NEUROMICS

5-HIAA (5-Hydroxyindoleacetic Acid) Data Sheet

Catalog Number:	RA24274	**Host:**	Rabbit
Product Type:	Whole Serum	**Species Reactivity:**	Mollusca, Rat, Sea Slug
Immunogen Sequence:	5-HIAA coupled to bovine serum albumin (BSA) with paraformaldehyde.	**Format:**	100ul Lyophilized, ≤ 0.09% sodium azide
Applications:	**Immunohistochemistry:** 1:4,000–1:8,000 in PBS/0.3% Triton X-100 – Bn-AV/HRP **Immunofluorescence:** Recommended Dilution: 1:200-1:400 for indirect immunofluorescence and 1:4,000–1:8,000 for biotinstreptavidin/HRP technique.		
Storage and Preparation:	Storage: Dilute with phosphate buffer or Tris buffer at dilutions no higher than 1/10, aliquot and freeze at -15° C or lower. Antibody can be stored for up to six months if handled as described above. It is strongly recommended that the customer perform a primary antibody dilution series using our dilution recommendations as a guideline. Note that a change in the fixation or buffering system as used in our protocol may change the configuration of the protein and, therefore, may alter the reactivity with the tissue tested.		

Application Notes for Immunohistochemistry

Tissue: Rat dorsal and median raphe neuronal cell bodies. Serotonergic system may be activated by salt loading which is achieved by 2% NaCl placed in drinking water for 48 hours prior to perfusion.

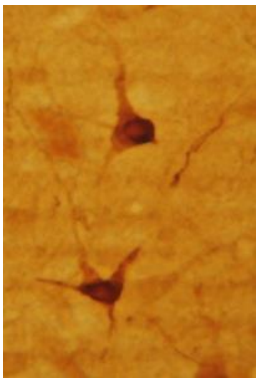
Perfusion Fixation • Fixation: 4% paraformaldehyde in 0.1M phosphate buffer, pH 7.4; 500 mL over 20 min. • Post Fixation: 1.5 hour at 4°C in 4% paraformaldehyde in 0.1M phosphate buffer, pH 7.4. • Note: Paraformaldehyde is a necessary component of fixation for this antiserum. If needed for other applications, glutaraldehyde may be used at low levels (0.1–0.3%) in conjunction with paraformaldehyde.

Sections 10 μm cryostat or 50 μm vibratome

Tissue Incubation 18–24 hours at 2°–8°C.

Detection System Use IF or Bn-AV/HRP reagents at dilutions recommended by the manufacturers.

Image: HC image of neurons staining for 5-H1AA in the raphe nucleus of the rat brainstem. The tissue was fixed with 4% formaldehyde in phosphate buffer, before being removed and prepared for vibratome sectioning. Floating sections were incubated at RT in 10% goat serum in PBS, before standard IHC procedure. Primary antibody was incubated at 1:5000 for 48 hours, goat anti-rabbit secondary was subsequently added for 1 hour after washing with PBS. Light microscopy staining was achieved with standard biotin-streptavidin/HRP procedure and DAB chromogen.



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www.neuromics.com

Neuromics Antibodies • 5325 West 74th Street, Suite 8 • Edina, MN 55439
phone 866-350-1500 • fax 612-677-3976 • e-mail: pshuster@neuromics.com