

# Anti-5-HT (Serotonin) Antibody [PSH06-04] - BSA and Azide free

## HA751044



<b>Product Type:</b>	Recombinant Rabbit monoclonal IgG, primary antibodies
<b>Species reactivity:</b>	Species independent
<b>Applications:</b>	WB, IHC-P, IF-Tissue, IHC-Fr, ELISA
<b>Clone number:</b>	PSH06-04

**Description:** Serotonin or 5-hydroxytryptamine (5-HT) is a monoamine neurotransmitter. Its biological function is complex, touching on diverse functions including mood, cognition, reward, learning, memory, and numerous physiological processes such as vomiting and vasoconstriction. Serotonin is produced in the central nervous system (CNS), specifically in the brainstem's raphe nuclei, the skin's Merkel cells, pulmonary neuroendocrine cells and the tongue's taste receptor cells. Approximately 90% of the serotonin the human body produces is in the gastrointestinal tract's enterochromaffin cells, where it regulates intestinal movements. Additionally, it is stored in blood platelets and is released during agitation and vasoconstriction, where it then acts as an agonist to other platelets. About 8% is found in platelets and 1–2% in the CNS. The serotonin is secreted lumenally and basolaterally, which leads to increased serotonin uptake by circulating platelets and activation after stimulation, which gives increased stimulation of myenteric neurons and gastrointestinal motility. The remainder is synthesized in serotonergic neurons of the CNS, where it has various functions, including the regulation of mood, appetite, and sleep.

**Immunogen:** 5-HT-OVA

**Recommended Dilutions:**

<b>ELISA</b>	1:1,000
<b>WB</b>	1:2,000
<b>IHC-P</b>	1:200-1:1,000
<b>IF-Tissue</b>	1:200
<b>IHC-Fr</b>	1:200-1:500

**Storage Buffer:** PBS (pH7.4).

**Storage Instruction:** Store at +4℃ after thawing. Aliquot store at -20℃. Avoid repeated freeze / thaw cycles.

**Purity:** Protein A affinity purified.

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Orders:0086-571-88062880

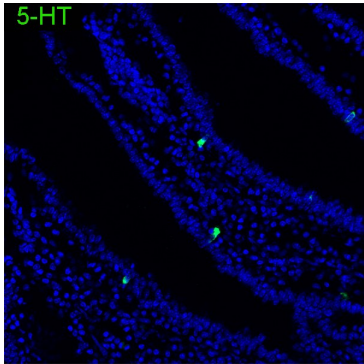
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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images



**Fig1:** Application: IHC-Fr

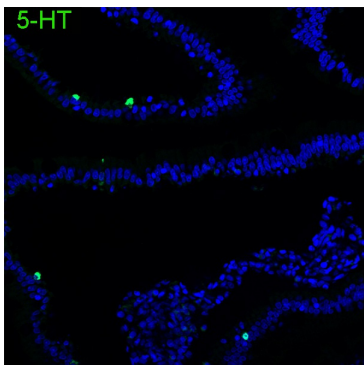
Species: Mouse

Site: Small intestine

Sample: Frozen section

Antibody concentration: 1:200

Antigen retrieval: The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven.



**Fig2:** Application: IHC-Fr

Species: Rat

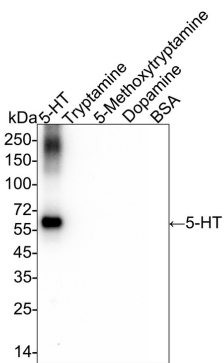
Site: Small intestine

Sample: Frozen section

Antibody concentration: 1:200

Antigen retrieval: The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for about 2 minutes in microwave oven.

**Fig3:** Western blot analysis of 5-HT (Serotonin) on different peptides with Rabbit anti-5-HT (Serotonin) antibody (HA751044) at 1/2,000 dilution.



Lane 1: 5-HT-BSA (positive) (30 ng/Lane)

Lane 2: Tryptamine-BSA (negative) (30 ng/Lane)

Lane 3: 5-Methoxytryptamine-BSA (negative) (30 ng/Lane)

Lane 4: Dopamine-BSA (negative) (30 ng/Lane)

Lane 5: BSA (negative) (30 ng/Lane)

Predicted band size: 66 kDa

Observed band size: 66 kDa

Exposure time: 1 minute; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA751044) at 1/2,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

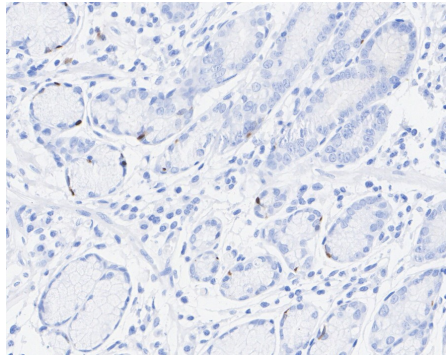
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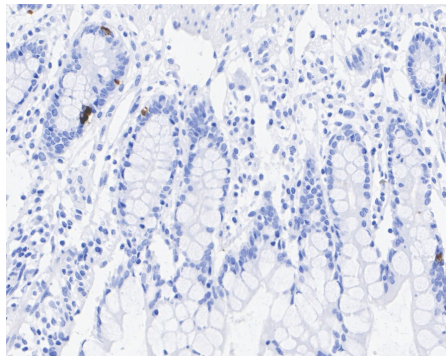
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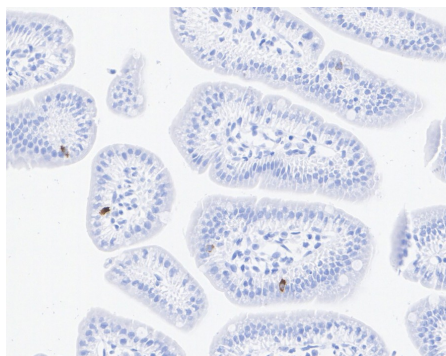
**Fig4:** Immunohistochemical analysis of paraffin-embedded human stomach tissue with Rabbit anti-5-HT (Serotonin) antibody (HA751044) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (HA751044) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



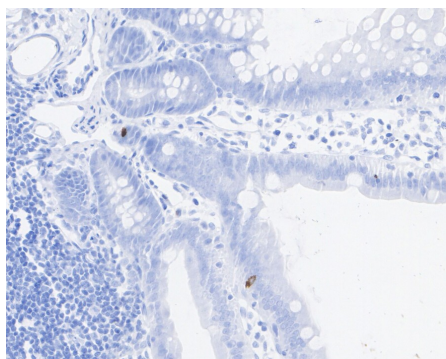
**Fig5:** Immunohistochemical analysis of paraffin-embedded human small intestine tissue with Rabbit anti-5-HT (Serotonin) antibody (HA751044) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (HA751044) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



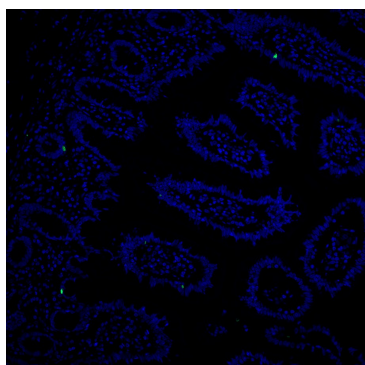
**Fig6:** Immunohistochemical analysis of paraffin-embedded mouse small intestine tissue with Rabbit anti-5-HT (Serotonin) antibody (HA751044) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (HA751044) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



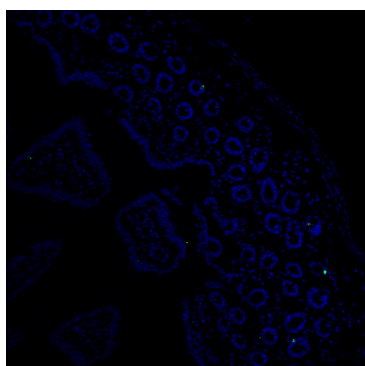
**Fig7:** Immunohistochemical analysis of paraffin-embedded rat small intestine tissue with Rabbit anti-5-HT (Serotonin) antibody (HA751044) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (HA751044) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



**Fig8:** Immunofluorescence analysis of paraffin-embedded human small intestine tissue labeling 5-HT (Serotonin) with Rabbit anti-5-HT (Serotonin) antibody (HA751044) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA751044, green) at 1/200 dilution overnight at 4 °C, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).



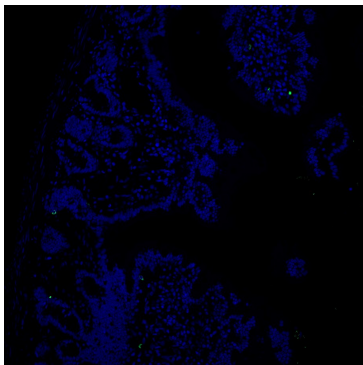
**Fig9:** Application: IF-tissue

Species: Mouse

Site: Small intestine

Sample: Paraffin-embedded section

Antibody concentration: 1:200

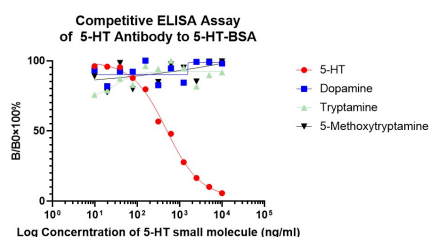
**Fig10:** Application: IF-tissue

Species: Rat

Site: Small intestine

Sample: Paraffin-embedded section

Antibody concentration: 1:200



**Fig11:** Competitive ELISA analysis of 5-HT was performed by coating wells of a 96-well plate with 50  $\mu$ L per well of 5-HT-BSA diluted in carbonate/bicarbonate buffer, at a concentration of 1  $\mu$ g/mL overnight at 4 $^{\circ}$ C. Wells of the plate were washed, blocked with 1% BSA blocking buffer, and incubated with 50  $\mu$ L per well of 5-HT monoclonal antibody at concentration of 1  $\mu$ g/mL with serial diluted 5-HT starting from a concentration of 10  $\mu$ g/mL for 45 min at 37 $^{\circ}$ C. The plate was washed and incubated with 50  $\mu$ L per well of an HRP-conjugated goat anti-rabbit IgG secondary antibody at a dilution of 1:70,000 for 30 min at 37 $^{\circ}$ C. Detection was performed using an Ultra TMB Substrate for 10 minutes at 37 $^{\circ}$ C in the dark. The reaction was stopped with sulfuric acid and absorbances were read on a spectrophotometer at 450 nm.

**Note:** All products are “FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE”.

### Background References

1. Shah PA et al. Serotonin as a Mitogen in the Gastrointestinal Tract: Revisiting a Familiar Molecule in a New Role. Cell Mol Gastroenterol Hepatol. 2021
2. Alvarez BD et al. Impact of specific serotonin receptor modulation on behavioral flexibility. Pharmacol Biochem Behav. 2021 Oct

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