

# Anti-Digoxin Antibody [PSH04-40] - BSA and Azide free

## HA750949



<b>Product Type:</b>	Recombinant Rabbit monoclonal IgG, primary antibodies
<b>Species reactivity:</b>	Species independent
<b>Applications:</b>	WB, ELISA, IHC-P, IHC-Fr, Dot Blot, IF-Tissue, IP
<b>Clone number:</b>	PSH04-40

**Description:** Digoxigenin (DIG) is a steroid found exclusively in the flowers and leaves of the plants *Digitalis purpurea*, *Digitalis orientalis* and *Digitalis lanata* (foxgloves), where it is attached to sugars, to form the glycosides (e.g. digoxin, lanatoside C). Digoxigenin is a hapten, a small molecule with high antigenicity, that is used in many molecular biology applications similarly to other popular haptens such as 2,4-Dinitrophenol, biotin, and fluorescein. Typically, digoxigenin is introduced chemically (conjugation) into biomolecules (proteins, nucleic acids) to be detected in further assays. Kd of the digoxigenin-antibody interaction has been estimated at ~12 nM (compare to Kd~0.1pM for the biotin-streptavidin interaction).

**Immunogen:** DIG-OVA

### Recommended Dilutions:

<b>WB</b>	1:2,000
<b>ELISA</b>	1:5,000-1:20,000
<b>IHC-P</b>	1:200-1:5,000
<b>IHC-Fr</b>	1:50-1:1,000
<b>Dot Blot</b>	1:2,000
<b>IF-Tissue</b>	1:50-1:1,000
<b>IP</b>	1-2µg/sample

**Storage Buffer:** 1\*PBS (pH7.4).

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

**Purity:** Protein A affinity purified.

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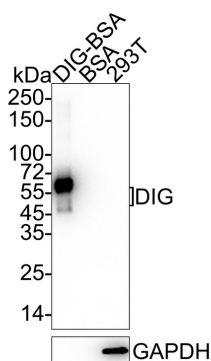
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## Images



**Fig1:** Western blot analysis of Digoxin on different lysates with Rabbit anti-Digoxin antibody (HA750949) at 1/2,000 dilution.

Lane 1: DIG-BSA (2 ng/Lane)

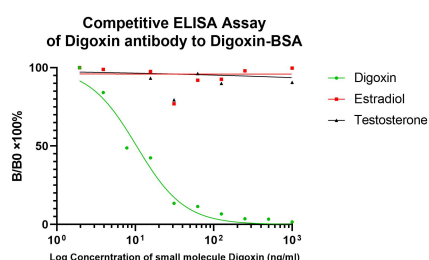
Lane 2: BSA (negative) (2 ng/Lane)

Lane 3: 293T cell lysate (negative) (20 µg/Lane)

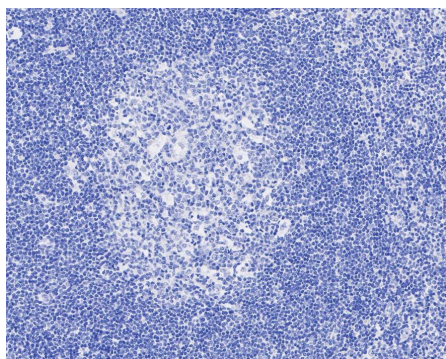
Exposure time: 20 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDN/TBST for 1 hour at room temperature. The primary antibody (HA750949) at 1/2,000 dilution was used in 5% NFDN/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.



**Fig2:** Competitive ELISA analysis of Digoxin / Estradiol / Testosterone was performed by coating wells of a 96-well plate with 50 µl per well of Digoxin-BSA diluted in carbonate/bicarbonate buffer, at a concentration of 1 µg/mL overnight at 4°C. Wells of the plate were washed, blocked with 1% BSA blocking buffer, and incubated with 100 µl per well of Digoxin monoclonal antibody at concentration of 1 µg/mL with serial diluted Digoxin / Estradiol / Testosterone starting from a concentration of 10 µg/ml for 1 hours at room temperature. The plate was washed and incubated with 50 µl per well of an HRP-conjugated goat anti-Rabbit IgG secondary antibody at a dilution of 1/5,000 for one hour at room temperature. Detection was performed using an Ultra TMB Substrate for 10 minutes at room temperature in the dark. The reaction was stopped with sulfuric acid and absorbances were read on a spectrophotometer at 450 nm.



**Fig3:** Immunohistochemical analysis of paraffin-embedded human tonsil tissue (negative) with Rabbit anti-Digoxin antibody (HA750949) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 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 (HA750949) 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.

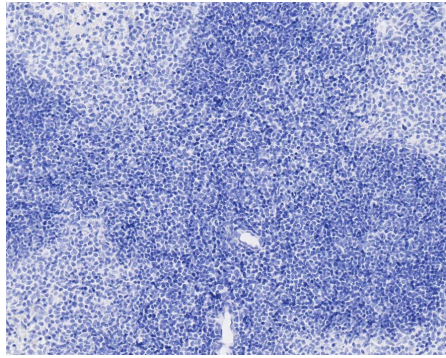
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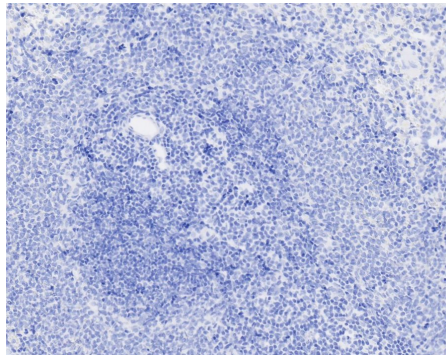
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**Fig4:** Immunohistochemical analysis of paraffin-embedded mouse spleen tissue (negative) with Rabbit anti-Digoxin antibody (HA750949) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 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 (HA750949) 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.



**Fig5:** Immunohistochemical analysis of paraffin-embedded rat spleen tissue (negative) with Rabbit anti-Digoxin antibody (HA750949) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 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 (HA750949) 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.

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

### Background References

1. Barratt KS et al. Production of Digoxigenin-Labeled Riboprobes for In Situ Hybridization Experiments. *Curr Protoc Mouse Biol.* 2020 Jun
2. Tanji M et al. Digoxigenin-labeled RNA probes for untranslated regions enable the isoform-specific gene expression analysis of myosin heavy chains in whole-mount in situ hybridization. *Dev Growth Differ.* 2023 Jan

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