# Anti-Lactate Dehydrogenase Antibody [SU39-06] ET1608-57

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat, Zebrafish
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, FC, IP
Molecular Wt:	Predicted band size: 37 kDa
Clone number:	SU39-06
Description:	Lactate dehydrogenase (LDH or LD) is an enzyme found in nearly all living cells. LDH catalyzes the conversion of lactate to pyruvate and back, as it converts NAD+ to NADH and back. A dehydrogenase is an enzyme that transfers a hydride from one molecule to another. LDH exists in four distinct enzyme classes. LDH is expressed extensively in body tissues, such as blood cells and heart muscle. Because it is released during tissue damage, it is a marker of common injuries and disease such as heart failure.
lmmunogen:	Synthetic peptide within human LDHB aa 270-310.
Positive control:	HeLa cell lysate, HEK-293 cell lysate, A549 cell lysate, hybrid fish (crucian-carp) brain tissue lysates, A549, A431, human liver tissue, human liver carcinoma tissue, human breast carcinoma tissue, mouse liver tissue, mouse testis tissue, mouse skeletal muscle tissue, Hela, LOVO.
Subcellular location:	Cytoplasm.
Database links:	SwissProt: P07195 Human   P00338 Human   P16125 Mouse   P06151 Mouse   P42123 Rat   P04642 Rat
Recommended Dilutions:	
WB	1:1,000-1:5,000
IF-Cell	1:50-1:200
IF-Tissue	1:50-1:200
IHC-P	1:200-1:2,000
	1.50-1.100 Use at an assay dependent concentration
Storage Buffer:	1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Shipped at 4 $^\circ\!\mathrm{C}$ . Store at +4 $^\circ\!\mathrm{C}$ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20 $^\circ\!\mathrm{C}$ long term.
Purity:	Protein A affinity purified.

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#### Images





**Fig1:** Western blot analysis of Lactate Dehydrogenase on different lysates with Rabbit anti-Lactate Dehydrogenase antibody (ET1608-57) at 1/5,000 dilution.

Lane 1: HeLa cell lysate (15 µg/Lane) Lane 2: HEK-293 cell lysate (15 µg/Lane) Lane 3: A549 cell lysate (15 µg/Lane)

Predicted band size: 37 kDa Observed band size: 37 kDa

Exposure time: 1 minute 36 seconds;

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 (ET1608-57) at 1/5,000 dilution was used in 5% NFDM/TBST at  $4^{\circ}$ C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:50,000 dilution was used for 1 hour at room temperature.

**Fig2:** Western blot analysis of Lactate Dehydrogenase on different lysates with Rabbit anti-Lactate Dehydrogenase antibody (ET1608-57) at 1/1,000 dilution.

Lane 1: Hela-si NT cell lysate (10 µg/Lane) Lane 2: Hela-si Lactate Dehydrogenase cell lysate (10 µg/Lane)

Predicted band size: 37 kDa Observed band size: 37 kDa

Exposure time: 25 seconds;

4-20% SDS-PAGE gel.

ET1608-57 was shown to specifically react with Lactate Dehydrogenase in Hela-si NT cells. Weakened band was observed when Hela-si Lactate Dehydrogenase sample was tested. Hela-si NT and Hela-si Lactate Dehydrogenase samples were subjected to SDS-PAGE. Proteins were transferred to a PVDF membrane and blocked with 5% NFDM in TBST for 1 hour at room temperature. The primary antibody (ET1608-57, 1/1,000) and Loading control antibody (Rabbit anti-GAPDH, ET1601-4, 1/10,000) were used in 5% BSA at room temperature for 2 hours. Goat Anti-rabbit IgG-HRP Secondary Antibody (HA1001) at 1:100,000 dilution was used for 1 hour at room temperature.

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**Fig3:** Western blot analysis of Lactate Dehydrogenase on hybrid fish (crucian-carp) brain tissue lysates. Proteins were transferred to a PVDF membrane and blocked with 5% BSA in PBS for 1 hour at room temperature. The primary antibody (ET1608-57, 1/500) was used in 5% BSA at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:200,000 dilution was used for 1 hour at room temperature.



**Fig4:** ICC staining of Lactate Dehydrogenase in A549 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 10% negative goat serum for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1608-57, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 conjugate-Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).



**Fig5:** ICC staining of Lactate Dehydrogenase in A431 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 10% negative goat serum for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1608-57, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 conjugate-Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).



**Fig6:** Immunohistochemical analysis of paraffin-embedded human liver tissue using anti-Lactate Dehydrogenase antibody. 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 5% BSA for 30 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1608-57, 1/50) for 30 minutes 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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**Fig7:** Immunohistochemical analysis of paraffin-embedded human liver carcinoma tissue using anti-Lactate Dehydrogenase antibody. 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 5% BSA for 30 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1608-57, 1/50) for 30 minutes 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:** Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue with Rabbit anti-Lactate Dehydrogenase antibody (ET1608-57) at 1/2,000 dilution.





**Fig9:** Immunohistochemical analysis of paraffin-embedded mouse liver tissue using anti-Lactate Dehydrogenase antibody. 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 5% BSA for 30 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1608-57, 1/50) for 30 minutes 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.



**Fig10:** Immunohistochemical analysis of paraffin-embedded mouse testis tissue using anti-Lactate Dehydrogenase antibody. 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 5% BSA for 30 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1608-57, 1/50) for 30 minutes 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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Lactate Dehydrogenase DAPI

**Fig11:** Immunohistochemical analysis of paraffin-embedded mouse skeletal muscle tissue using anti-Lactate Dehydrogenase antibody. 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 5% BSA for 30 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (ET1608-57, 1/50) for 30 minutes 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.

**Fig12:** Flow cytometric analysis of Lactate Dehydrogenase was done on Hela cells. The cells were fixed, permeabilized and stained with the primary antibody (ET1608-57, 1/50) (red). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor®488 conjugate-Goat anti-Rabbit IgG Secondary antibody at 1/1,000 dilution for 30 minutes.Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

**Fig13:** Immunocytochemistry analysis of LOVO cells labeling Lactate Dehydrogenase with Rabbit anti-Lactate Dehydrogenase antibody (ET1608-57) at 1/50 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37  $^{\circ}$ C, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-Lactate Dehydrogenase antibody (ET1608-57) at 1/50 dilution in 2% negative goat serum overnight at 4  $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluor <sup>TM</sup> 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

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

#### **Background References**

- 1. Rocha CS et al. Melatonin alters the glycolytic profile of Sertoli cells: implications for male fertility. Mol Hum Reprod 20:1067-76 (2014).
- 2. Mallat Y et al. Proteome modulation in H9c2 cardiac cells by microRNAs miR-378 and miR-378. Mol Cell Proteomics 13:18-29 (2014).

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