

## Product Datasheet

### ENO1 Rabbit Polyclonal Antibody (orb654297)

<b>Catalog Number</b>	orb654297
<b>Category</b>	Antibodies
<b>Description</b>	Anti-ENO1 Antibody. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Monkey, Mouse, Rat.
<b>Target</b>	Alpha-enolase
<b>Clonality</b>	Polyclonal
<b>Species/Host</b>	Rabbit
<b>Isotype</b>	Rabbit IgG
<b>Conjugation</b>	Unconjugated
<b>Reactivity</b>	Human, Monkey, Mouse, Rat
<b>Form/Appearance</b>	Lyophilized
<b>Concentration</b>	500 µg/ml
<b>Buffer/Preservatives</b>	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na <sub>2</sub> HPO <sub>4</sub> , 0.05mg NaN <sub>3</sub> .
<b>Reconstitution</b>	Add 0.2ml of distilled water will yield a concentration of 500ug/ml.
<b>Purification</b>	Immunogen affinity purified.
<b>Immunogen</b>	A synthetic peptide corresponding to a sequence in the middle region of human ENO1, which shares 90.9% amino acid (aa) sequence identity with rat ENO1.
<b>UniProt ID</b>	<b>P06733</b>
<b>MW</b>	47 kDa

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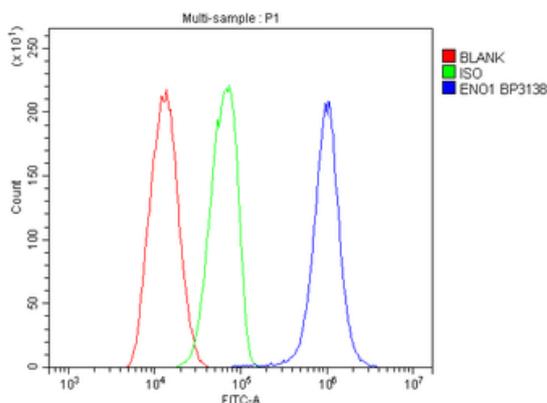
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<b>Tested applications</b>	FC, ICC, IF, IHC, IP, WB
<b>Dilution range</b>	Western blot, 0.1-0.25µg/ml, Human, Mouse, Monkey, Rat Immunohistochemistry (Paraffin-embedded Section), 0.5-1µg/ml, Human, Mouse, Rat Immunocytochemistry/Immunofluorescence, 2µg/ml, Human Immunoprecipitation, 0.5-2 µg/ml, Human Flow Cytometry (Fixed), 1-3µg/1x10 <sup>6</sup> cells, Human
<b>Specificity</b>	No cross reactivity with other proteins.
<b>Cross Reactivity</b>	No cross-reactivity with other proteins.
<b>Antibody Type</b>	Primary Antibody
<b>Storage</b>	Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at -20°C in small aliquots to prevent freeze-thaw cycles.
<b>Note</b>	For research use only
<b>Expiration Date</b>	12 months from date of receipt.



Flow Cytometry analysis of HL-60 cells using anti-ENO1 antibody. Overlay histogram showing HL-60 cells (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-ENO1 Antibody (1 µg/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (5-10 µg/1x10<sup>6</sup> cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 µg/1x10<sup>6</sup>) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

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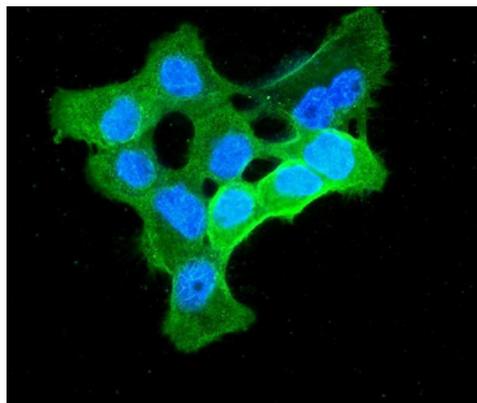
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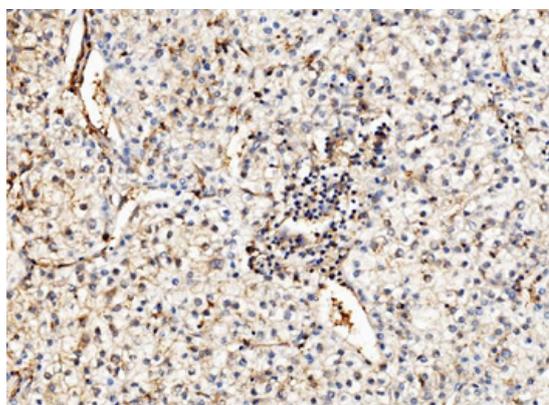
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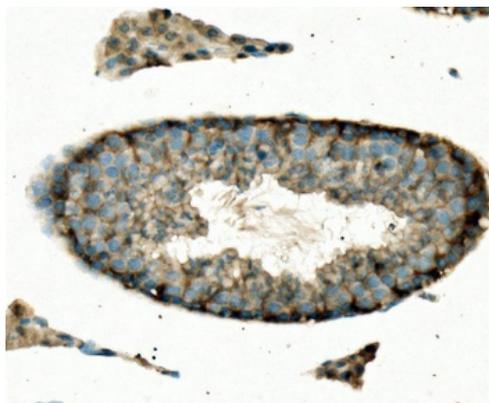
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IF analysis of ENO1 using anti-ENO1 antibody. ENO1 was detected in immunocytochemical section of A431 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2  $\mu\text{g}/\text{mL}$  rabbit anti-ENO1 Antibody overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.



IHC analysis of ENO1 using anti-ENO1 antibody. ENO1 was detected in paraffin-embedded section of human liver cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1  $\mu\text{g}/\text{ml}$  rabbit anti-ENO1 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.



IHC analysis of ENO1 using anti-ENO1 antibody. ENO1 was detected in paraffin-embedded section of mouse testis tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1  $\mu\text{g}/\text{ml}$  rabbit anti-ENO1 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.

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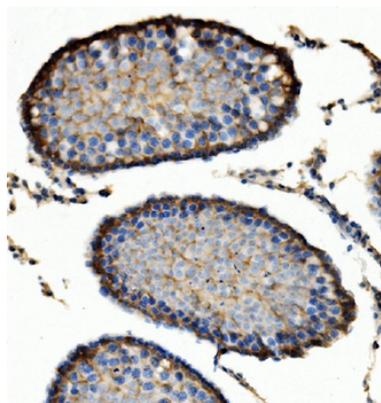
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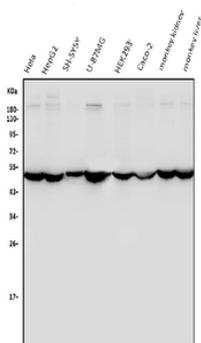
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IHC analysis of ENO1 using anti-ENO1 antibody. ENO1 was detected in paraffin-embedded section of rat testis tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml rabbit anti-ENO1 Antibody overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) with DAB as the chromogen.



Western blot analysis of ENO1 using anti-ENO1 antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50 µg of sample under reducing conditions. Lane 1: human Hela whole cell lysates, Lane 2: human HepG2 whole cell lysates, Lane 3: human SH-SY5Y whole cell lysates, Lane 4: human U-87MG whole cell lysates, Lane 5: human HEK293 whole cell lysates, Lane 6: human Caco-2 whole cell lysates, Lane 7: monkey kidney tissue lysates, Lane 8: monkey liver tissue lysates. After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-ENO1 antigen affinity purified polyclonal antibody at 0.25 µg/mL overnight at 4°C, then washed with TBS-0.1% Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit with Tanon 5200 system. A specific band was detected for ENO1 at approximately 47 KD. The expected band size for ENO1 is at 47 KD.

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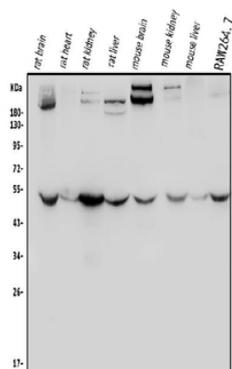
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