BOSTER BIOLOGICAL TECHNOLOGY
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# Anti-Dihydrofolate reductase (DHFR) Antibody Picoband®

**Catalog Number:** PB9175

## About DHFR

Dihydrofolate reductase, or DHFR, is an enzyme that reduces dihydrofolic acid to tetrahydrofolic acid, using NADPH as electron donor, which can be converted to the kinds of tetrahydrofolate cofactors used in 1-carbon transfer chemistry. In humans, the DHFR enzyme is encoded by the DHFR gene. It is found in the q11→q22 region of chromosome 5. What's more, DHFR belongs to the dihydrofolate reductase family, and it converts dihydrofolate into tetrahydrofolate, a methyl group shuttle required for the de novo synthesis of purines, thymidylic acid, and certain amino acids. DHFR is the key enzyme in folate metabolism. In addition, DHFR catalyzes an essential reaction for de novo glycine and purine synthesis, and for DNA precursor synthesis.

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## Overview

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| Product Name | Anti-Dihydrofolate reductase (DHFR) Antibody Picoband® |
| Reactive Species | Human, Mouse, Rat |
| Description | Boster Bio Anti-Dihydrofolate reductase (DHFR) Antibody Picoband® catalog # PB9175. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat. The brand Picoband indicates this is a premium antibody that guarantees superior quality, high affinity, and strong signals with minimal background in Western blot applications. Only our best-performing antibodies are designated as Picoband, ensuring unmatched performance. |
| Application | Flow Cytometry, IF, IHC, ICC, WB |
| Clonality | Polyclonal |
| Formulation | Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.01mg NaN3. |
| Storage Instructions | Store at -20˚C for one year from date of receipt. After reconstitution, at 4˚C for one month. It can also be aliquotted and stored frozen at -20˚C for six months. Avoid repeated freeze-thaw cycles. |
| Host | Rabbit |
| Uniprot ID | P00374 |

## Technical Details

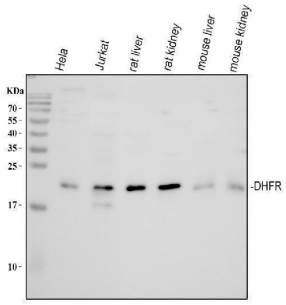
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| Immunogen | E.coli-derived human DHFR recombinant protein (Position: V2-D187). Human DHFR shares 90% amino acid (aa) sequence identity with both mouse and rat DHFR. |

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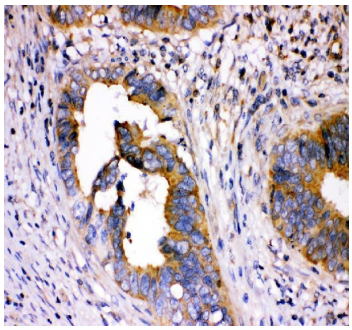
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| Recommended Detection Systems | Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P) and ICC. |
| Cross Reactivity | No cross-reactivity with other proteins |
| Isotype | Rabbit IgG |
| Form | Lyophilized |
| Concentration | Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml. |
| Purification | Immunogen affinity purified. |
| Suggested Dilutions | Western blot, 0.1-0.5ug/ml, Human, Mouse, Rat<br>Immunohistochemistry (Paraffin-embedded Section), 2-5ug/ml, Human, Mouse, Rat<br>Immunocytochemistry/Immunofluorescence, 5ug/ml, Human<br>Flow Cytometry (Fixed), 1-3ug/1x$10^6$ cells, Human<br><br>Note: For flow cytometry (FCM), suitable for fixed cells only. Not suitable for FACS or surface flow cytometry using live cells. |

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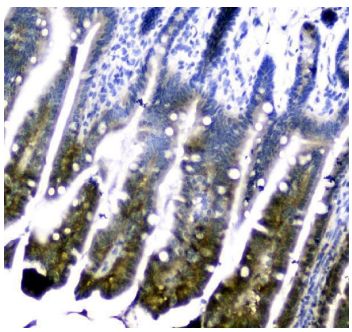
# Anti-Dihydrofolate reductase (DHFR) Antibody Picoband® (PB9175) Images




Western blot analysis of DHFR using anti-DHFR antibody (PB9175). 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 30 ug of sample under reducing conditions. Lane 1: human Hela whole cell lysates, Lane 2: human Jurkat whole cell lysates, Lane 3: rat liver tissue lysates, Lane 4: rat kidney tissue lysates, Lane 5: mouse liver tissue lysates, Lane 6: mouse kidney 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-DHFR antigen affinity purified polyclonal antibody (Catalog # PB9175) at 0.5 ug/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 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for DHFR at approximately 21 kDa. The expected band size for DHFR is at 21 kDa.



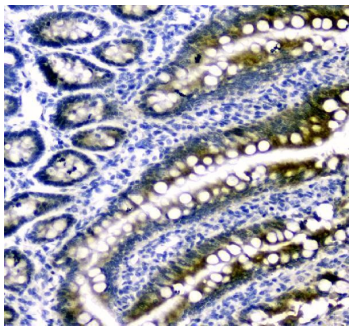
IHC analysis of DHFR using anti-DHFR antibody (PB9175). DHFR was detected in a paraffin-embedded section of human colorectal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-DHFR Antibody (PB9175) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



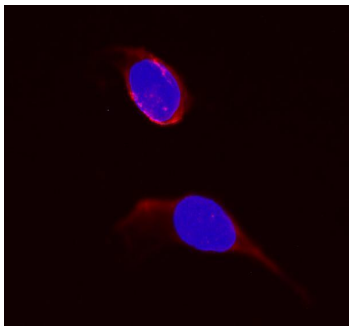
IHC analysis of DHFR using anti-DHFR antibody (PB9175). DHFR was detected in a paraffin-embedded section of mouse small intestine tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-DHFR Antibody (PB9175) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.

IHC analysis of DHFR using anti-DHFR antibody (PB9175). DHFR was detected in a paraffin-embedded section of rat

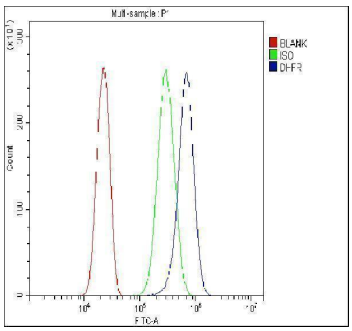
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small intestine tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-DHFR Antibody (PB9175) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



IF analysis of DHFR using anti-DHFR antibody (PB9175). DHFR was detected in immunocytochemical section of U20S cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5ug/mL rabbit anti-DHFR Antibody (PB9175) overnight at 4°C. DyLight®550 Conjugated Goat Anti-Rabbit IgG (BA1135) was used as secondary antibody at 1:500 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.




Flow Cytometry analysis of PC-3 cells using anti-DHFR antibody (PB9175). Overlay histogram showing PC-3 cells stained with PB9175 (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-DHFR Antibody (PB9175, 1ug/1x106 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10ug/1x106 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1ug/1x106) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

## 1 Publications Citing This Product

1. PubMed ID: 24297308, Tian L, Zhang J, Ge J, Xiao H, Lu J, Fu S, Liu M, Sun Y. Med Oncol. 2014 Jan;31(1):785. Doi: 10.1007/S12032-013-0785-3. Epub 2013 Dec 3. Microrna-205 Suppresses Proliferation And Promotes Apoptosis In Laryngeal Squamous Cell Carcinoma.

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