

Anti-DHFR Antibody Picoband™ (monoclonal, 3C8)

Catalog # ABO14841

Product Information

Application	WB, IHC
Primary Accession	P00374
Host	Mouse
Isotype	Mouse IgG2b
Reactivity	Rat, Human
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-DHFR Antibody Picoband™ (monoclonal, 3C8) . Tested in IHC, WB applications. This antibody reacts with Human, Rat.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500 µg/ml.

Additional Information

Gene ID	1719
Other Names	Dihydrofolate reductase, 1.5.1.3, DHFR
Calculated MW	21453
Application Details	Western blot, 0.1-0.5 µg/ml Immunohistochemistry (Paraffin-embedded Section), 0.5-1 µg/ml
Subcellular Localization	Mitochondrion. Cytoplasm
Tissue Specificity	Widely expressed in fetal and adult tissues, including throughout the fetal and adult brains and whole blood. Expression is higher in the adult brain than in the fetal brain.
Source	Eukaryota
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na ₂ HPO ₄ , 0.05mg NaN ₃ .
Clone Names	Clone: 3C8
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.
Cross Reactivity	No cross-reactivity with other proteins.
Storage	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.

Protein Information

Name	DHFR (HGNC:2861)
Function	Catalyzes the reduction of 7,8-dihydrofolate (DHF) to 5,6,7,8-tetrahydrofolate in a NADPH-dependent manner (PubMed: 12096917 , PubMed: 15039552 , PubMed: 17569517 , PubMed: 19196009 , PubMed: 19478082 , PubMed: 21876184 , PubMed: 9719595). Key enzyme in folate metabolism. Contributes to the nuclear and mitochondrial de novo thymidylate biosynthesis pathway (PubMed: 21876188 , PubMed: 22235121). Catalyzes an essential reaction for de novo glycine and purine synthesis, and for DNA precursor synthesis. Binds its own mRNA and that of DHFR2.
Cellular Location	Mitochondrion {ECO:0000250 UniProtKB:P00375}. Cytoplasm {ECO:0000250 UniProtKB:P00375}. Nucleus. Note=Localized to the nucleus during S and G2/M phases of the cell cycle. As a component of the de novo thymidylate synthesis complex, localizes specifically to replication forks during DNA synthesis (PubMed:22235121)
Tissue Location	Widely expressed in fetal and adult tissues, including throughout the fetal and adult brains and whole blood Expression is higher in the adult brain than in the fetal brain

Images

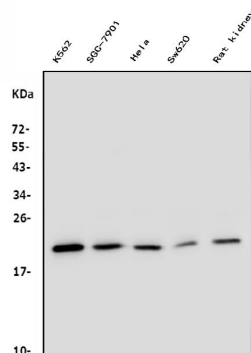


Figure 1. Western blot analysis of DHFR using anti-DHFR antibody (M00813-1).

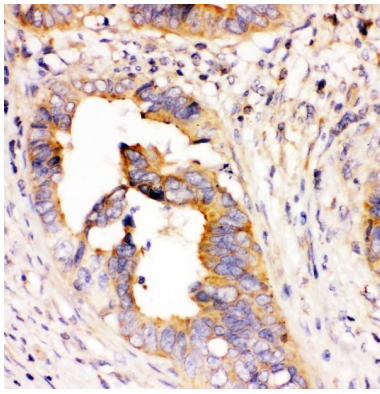
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.

Lane 1: human K562 whole cell lysates,
Lane 2: human SGC-7901 whole cell lysates,
Lane 3: human HeLa whole cell lysates,
Lane 4: human SW620 whole cell lysates,
Lane 5: rat kidney tissue lysates.

After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-DHFR antigen affinity purified monoclonal antibody (Catalog # M00813-1) at 0.5 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-mouse 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 (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for DHFR at approximately 22KD. The expected band size for DHFR is at 22KD.

Figure 2. IHC analysis of DHFR using anti-DHFR antibody (M00813-1).

DHFR was detected in paraffin-embedded section of



human rectal 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 µg/ml mouse anti-DHFR Antibody (M00813-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

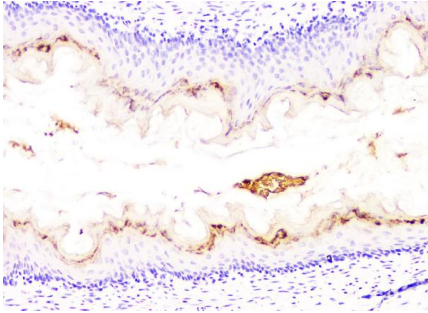


Figure 3. IHC analysis of DHFR using anti-DHFR antibody (M00813-1).

DHFR was detected in paraffin-embedded section of rat gaster 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 mouse anti-DHFR Antibody (M00813-1) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC) (Catalog # SA1021) with DAB as the chromogen.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.