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Anti-nmt55 p54nrb NONO Antibody Picoband™ (monoclonal, 11E2)

Catalog # ABO14799

Product Information

Application	WB, IF, ICC, FC
Primary Accession	Q15233
Host	Mouse
Isotype	Mouse IgG1
Reactivity	Human
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-nmt55 p54nrb NONO Antibody Picoband™ (monoclonal, 11E2) . Tested in Flow Cytometry, IF, ICC, WB applications. This antibody reacts with Human.

Additional Information

Gene ID	4841
Other Names	Non-POU domain-containing octamer-binding protein, NonO protein, 54 kDa nuclear RNA- and DNA-binding protein, p54(nrb), p54nrb, 55 kDa nuclear protein, NMT55, DNA-binding p52/p100 complex, 52 kDa subunit, NONO {ECO:0000303|PubMed:9393982, ECO:0000312|HGNC:HGNC:7871}
Calculated MW	54232
Application Details	Western blot, 0.1-0.5 □g/ml Immunocytochemistry, 0.5-1 □g/ml Immunocytochemistry/Immunofluorescence, 5 □g/ml Flow Cytometry, 1-3 □g/1x10^6 cells
Subcellular Localization	Nucleus. Nucleus, nucleolus. Nucleus speckle. Detected in punctate subnuclear structures often located adjacent to splicing speckles, called paraspeckles.
Tissue Specificity	Heart, brain, placenta, lung, liver, skeletal muscle, kidney and pancreas. Also found in a number of breast tumor cell lines.
Source	Eukaryota
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.
Clone Names	Clone: 11E2
Immunogen	A synthetic peptide corresponding to a sequence at the N-terminus of human nmt55 p54nrb, identical to the related mouse and rat sequences.

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	NONO {ECO:0000303|PubMed:9393982, ECO:0000312|HGNC:HGNC:7871}
Function	DNA- and RNA binding protein, involved in several nuclear processes (PubMed:11525732, PubMed:12403470, PubMed:26571461). Binds the conventional octamer sequence in double-stranded DNA (PubMed:11525732, PubMed:12403470, PubMed:26571461). Also binds single- stranded DNA and RNA at a site independent of the duplex site (PubMed:11525732, PubMed:12403470, PubMed:26571461). Involved in pre- mRNA splicing, probably as a heterodimer with SFPQ (PubMed:11525732, PubMed:12403470, PubMed:26571461). Interacts with U5 snRNA, probably by binding to a purine-rich sequence located on the 3' side of U5 snRNA stem 1b (PubMed:12403470). Together with PSPC1, required for the formation of nuclear paraspeckles (PubMed:22416126). The SFPQ-NONO heteromer associated with MATR3 may play a role in nuclear retention of defective RNAs (PubMed:11525732). The SFPQ-NONO heteromer may be involved in DNA unwinding by modulating the function of topoisomerase I/TOP1 (PubMed:10858305). The SFPQ-NONO heteromer may be involved in DNA non-homologous end joining (NHEJ) required for double-strand break repair and V(D)J recombination and may stabilize paired DNA ends (PubMed:15590677). In vitro, the complex strongly stimulates DNA end joining, binds directly to the DNA substrates and cooperates with the Ku70/G22P1-Ku80/XRCC5 (Ku) dimer to establish a functional preligation complex (PubMed:15590677). NONO is involved in transcriptional regulation. The SFPQ-NONO-NR5A1 complex binds to the CYP17 promoter and regulates basal and cAMP-dependent transcriptional activity (PubMed:11897684). NONO binds to an enhancer element in long terminal repeats of endogenous intracisternal A particles (IAPs) and activates transcription (By similarity). Regulates the circadian clock by repressing the transcriptional activator activity of the CLOCK-BMAL1 heterodimer (By similarity). Important for the functional organization of GABAergic synapses (By similarity). Plays a specific and important role in the regulation of synaptic RNAs and GPHN/gephyrin scaffold structure, through the regulation of GABRA2 transcript (By similarity). Plays a key role during neuronal differentiation by recruiting TET1 to genomic loci and thereby regulating 5-hydroxymethylcytosine levels (By similarity). Plays a role in the regulation of DNA virus-mediated innate immune response by assembling into the HDP-RNP complex, a complex that serves as a platform for IRF3 phosphorylation and subsequent innate immune response activation through the cGAS-STING pathway (PubMed:28712728, PubMed:30270045). Promotes activation of the cGAS-STING pathway in response to HIV-2 infection: acts by interacting with HIV-2 Capsid protein p24, thereby promoting detection of viral DNA by CGAS, leading to CGAS-mediated inmmune activation (PubMed:30270045). In contrast, the weak interaction with HIV-1 Capsid protein p24 does not allow activation of the cGAS-STING pathway (PubMed:30270045).
Cellular Location	Nucleus. Nucleus, nucleolus. Nucleus speckle. Chromosome {ECO:0000250|UniProtKB:Q99K48}. Note=Detected in punctate subnuclear structures often located adjacent to splicing speckles, called paraspeckles.
Tissue Location	Heart, brain, placenta, lung, liver, skeletal muscle, kidney and pancreas. Also found in a number of breast tumor cell lines.

Background

Non-POU domain-containing octamer-binding protein is a protein that in humans is encoded by the NONO gene. This gene encodes an RNA-binding protein which plays various roles in the nucleus, including transcriptional regulation and RNA splicing. A rearrangement between this gene and the transcription factor E3 gene has been observed in papillary renal cell carcinoma. Alternatively spliced transcript variants have been described. Pseudogenes exist on Chromosomes 2 and 16.

Images

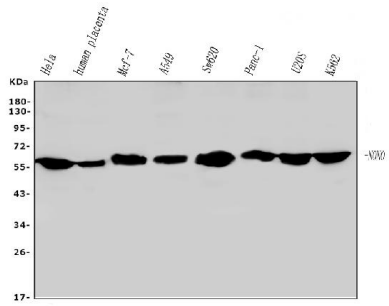


Figure 1. Western blot analysis of nmt55 p54nrb using anti-nmt55 p54nrb antibody (M03515).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: human HELA whole cell lysates,
Lane 2: human placenta tissue lysates,
Lane 3: human MCF-7 whole cell lysates,
Lane 4: human A549 whole cell lysates,
Lane 5: human SW620 whole cell lysates,
Lane 6: human PANC-1 whole cell lysates,
Lane 7: human U20S whole cell lysates,
Lane 8: human K562 whole cell 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-nmt55 p54nrb antigen affinity purified monoclonal antibody (Catalog # M03515) 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 nmt55 p54nrb at approximately 60KD. The expected band size for nmt55 p54nrb is at 60KD.

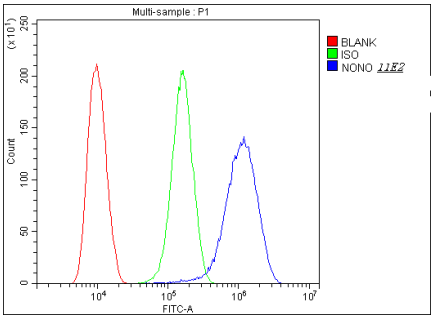
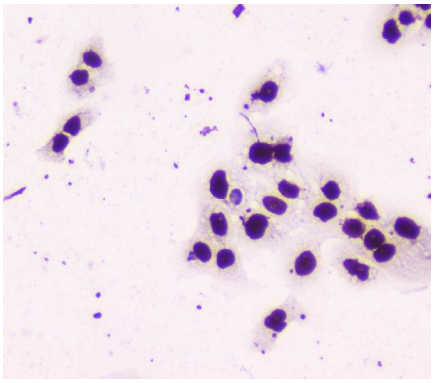


Figure 2. Flow Cytometry analysis of U20S cells using anti-nmt55 p54nrb antibody (M03515).
Overlay histogram showing U20S cells stained with M03515 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-nmt55 p54nrb Antibody (M03515,1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Figure 3. IHC analysis of nmt55 p54nrb using anti-nmt55 p54nrb antibody (M03515).
nmt55 p54nrb was detected in immunocytochemical section of A431 cell. 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 1 µg/ml mouse anti-nmt55 p54nrb Antibody (M03515) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1021) with DAB as the chromogen.

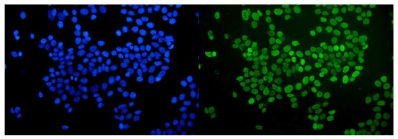


Figure 4. IF analysis of nmt55 p54nrb using anti-nmt55 p54nrb antibody (M03515).
nmt55 p54nrb was detected in immunocytochemical section of MCF-7 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 µg/mL mouse anti-nmt55 p54nrb Antibody (M03515) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Mouse IgG (BA1126) 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.

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