abcam

ab14387

Anti-Mus81 antibody [MTA30 2G10/3]

Anti-Mus81 antibody [MTA30 2G10/3] (ab14387) is a mouse monoclonal antibody detecting Mus81 in **Western Blot, Flow Cytometry**. Suitable for **Human**.

- Over 50 publications
- Trusted since 2004

Key facts

Isotype	IgG2a
Host species	Mouse
Storage buffer	Preservative: 0.02% Sodium azide Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	Native Full Length Protein corresponding to Human Crossover junction endonuclease MUS81. Database link Q96NY9
Clone number	MTA30 2G10/3
Purification technique	Affinity purification Protein A
Concentration	1 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →
Purification notes	from TCS

Reactivity data

Flow Cyt

Tested

Species	Human
Dilution info	0.5 µg for 10^6 Cells
Notes	ab170191 - Mouse monoclonal IgG2a, is suitable for use as an isotype control with this antibody.

Not recommended

Species	Mouse
Dilution info	-
Notes	ab170191 - Mouse monoclonal IgG2a, is suitable for use as an isotype control with this antibody.

WB

Tested

Species	Human
Dilution info	1/1000
Notes	Tested on both recombinant protein and whole cell extract.

Not recommended

Species	Mouse
Dilution info	-
Notes	Tested on both recombinant protein and whole cell extract.

Target data

See full target information Crossover junction endonuclease MUS81

Function	Catalytic subunit of two functionally distinct, structure-specific, heterodimeric DNA endonucleases MUS81-EME1 and MUS81-EME2 that are involved in the maintenance of genome stability (PubMed:11741546, PubMed:12374758, PubMed:12686547, PubMed:12721304, PubMed:24371268, PubMed:24733841, PubMed:24813886, PubMed:35290797, PubMed:39015284). Both endonucleases have essentially the same substrate specificity though MUS81-EME2 is more active than its MUS81-EME1 counterpart. Both cleave 3'-flaps and nicked Holliday junctions, and exhibit limited endonuclease activity with 5' flaps and nicked double-stranded DNAs (PubMed:24371268, PubMed:24733841, PubMed:35290797). MUS81-EME2 which is active during the replication of DNA is

more specifically involved in replication fork processing (PubMed:24813886). Replication forks frequently encounter obstacles to their passage, including DNA base lesions, DNA interstrand cross-links, difficult-to-replicate sequences, transcription bubbles, or tightly bound proteins. One mechanism for the restart of a stalled replication fork involves nucleolytic cleavage mediated by the MUS81-EME2 endonuclease. By acting upon the stalled fork, MUS81-EME2 generates a DNA double-strand break (DSB) that can be repaired by homologous recombination, leading to the restoration of an active fork (PubMed:24813886). MUS81-EME2 could also function in telomere maintenance (PubMed:24813886). MUS81-EME1, on the other hand, is active later in the cell cycle and functions in the resolution of mitotic recombination intermediates including the Holliday junctions, the four-way DNA intermediates that form during homologous recombination (PubMed:11741546, PubMed:12374758, PubMed:14617801, PubMed:15805243, PubMed:24813886).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

What is this antibody validated in?
Anti-Mus81 antibody [MTA30 2G10/3] (ab14387) is a mouse monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Flow Cyt) in Human samples.

What is the molecular weight of Mus81?
Anti-Mus81 [MTA30 2G10/3] (ab14387) specifically detects a band for Mus81 (UniProt: Q96NY9) at a molecular weight of 66kDa.

Trusted by the scientific community
Anti-Mus81 [MTA30 2G10/3] (ab14387) was first used in a scientific publication in 2004 and has been cited over 50 times in peer-reviewed journals.

Reviewed by scientists
Anti-Mus81 [MTA30 2G10/3] (ab14387) has over 10 independent reviews from customers.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	Mus81 also known as methyl methanesulfonate 81 plays a significant role in DNA repair and genome stability. As a part of the XPF family of endonucleases Mus81 cuts DNA structures that arise during DNA replication and repair processes. It has a molecular mass of approximately 79 kDa and expresses in a range of organisms including humans. Mus81 is found in tissues with high cell turnover where DNA repair is necessary for maintaining genetic integrity.
Biological function summary	Mus81 functions as a component of the Mus81-Eme1 complex which facilitates the resolution of DNA junctions such as Holliday junctions stalled replication forks and other intermediates. This activity is important for the successful completion of homologous recombination and DNA repair safeguarding against mutations that may lead to genomic instability. Additionally Mus81 helps maintain the correct number of chromosomes during cell division by preventing or resolving DNA entanglement.
Pathways	Mus81 integrates into the pathway coordinating DNA damage response and repair. It is involved in the homologous recombination repair pathway essential for repairing double-strand breaks. Mus81 interacts with proteins such as Rad51 a pivotal protein in the homologous recombination process playing a collaborative role in restructuring damaged DNA. Mus81 provides flexibility and adaptability in orchestrating efficient DNA repair responses through these interactions.
Associated diseases and disorders	Mus81 links to cancer progression and genetic disorders where DNA repair is compromised. For instance alterations in Mus81 function can lead to increased susceptibility to cancers due to the buildup of genomic instability. Additionally deficiencies in the Mus81 pathway can contribute to disorders such as Fanconi anemia which involves defective DNA repair. Mus81 also connects with proteins like BRCA2 another critical component in the DNA repair machinery highlighting its potential impact on cancer therapy strategies.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

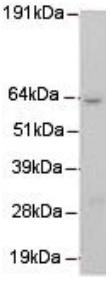
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

6 product images



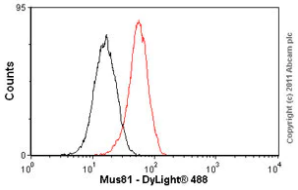
Western blot - Anti-Mus81 antibody [MTA30 2G10/3] (ab14387)

All lanes:
Western blot - Anti-Mus81 antibody [MTA30 2G10/3] (ab14387) at 1/1000 dilution

All lanes:
HeLa cell lysate

Predicted band size: 61 kDa

Observed band size: 64 kDa



Flow Cytometry - Anti-Mus81 antibody [MTA30 2G10/3] (ab14387)

Mus81 Flow Cytometry staining using mouse Anti-Mus81 antibody

Overlay histogram showing HeLa cells stained with ab14387 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab14387, 0.5µg/1x10^6 cells) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG2a [ICIGG2A] (ab91361, 1µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed.

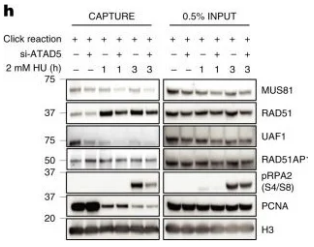


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Western blot - Anti-Mus81 antibody [MTA30 2G10/3] (ab14387)

Mus81 Western blot staining using mouse Anti-Mus81 antibody

Western Blotting using Anti-Mus81 antibody [MTA30 2G10/3], ab14387. Publication image from Park, S. H. et al., 2019, Nat Commun, 31844045. Legend direct from paper.

ATAD5 promotes generation of MUS81-mediated single-stranded DNA-associated breaks in response to replication stress.a, b U2OS cells transfected with ATAD5 siRNA under the Noco-APH condition were treated with 2 mM HU for 6 h before being collected for analysis by pulsed field gel electrophoresis. a Representative data from three independent experiments. Asterisk (*) indicates a DNA break. b DNA breaks were quantified and displayed (N = 4). c U2OS or HeLa cells transfected with ATAD5 siRNA under the Noco-APH condition were treated with 2 mM HU for 6 h before being collected for a neutral COMET assay. The tail moment was calculated from ~200 cells and plotted. d U2OS cells expressing ATAD5AID were pre-treated with auxin and treated with 2 mM HU for another 6 h before being collected for a neutral COMET assay. Two independent experiments were performed, and one representative result is displayed. e U2OS-TetOn-ATAD5 cells treated with doxycycline for 24 h before entering the Noco-APH condition were treated with 2 mM HU for 6 h

before collection for the neutral COMET assay. f U2OS cells transfected with siRNAs under the Noco-APH condition were treated with 2 mM HU for 6 h. Chromatin-bound proteins were fractionated and subjected to immunoblotting. g U2OS cells transfected with a combination of ATAD5 and MUS81 siRNAs under the Noco-APH condition were treated with 2 mM HU for 6 h before collection for the neutral COMET assay. The tail moment was calculated from ~300 cells and plotted. h HEK293T cells transfected with ATAD5 siRNA for 48 h were labeled with 10 μM EdU for 20 min, washed, and treated with 2 mM HU for the indicated times. Samples were processed for iPOND, and captured proteins were separated by SDS-PAGE and immunoblotted. b–e, g Error bars represent standard deviation of the mean. Statistical analysis: two-tailed Student’ s t test; *p < 0.05, **p < 0.005, ***p < 0.001, and ****p < 0.0001.

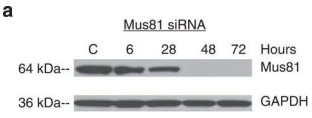


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Western blot - Anti-Mus81 antibody [MTA30 2G10/3] (ab14387)

Mus81 Western blot staining using mouse Anti-Mus81 antibody

Western Blotting using Anti-Mus81 antibody [MTA30 2G10/3], ab14387. Publication image from Pommier, Y. et al., 2015, Nat Commun, 25879486. Legend direct from paper.

Acute Mus81 depletion triggers slower DNA synthesis and elevated initiation frequencyHCT116 cells were transfected withsiRNA directed against Mus81 or with control siRNA. (A) Western-blot analysis showed that HCT116 cells transfected with siRNA against Mus81 exhibited a dramatic knockdown of Mus81 protein, compared to cells transfected with control siRNA 48 and 72 hours after transfection. Cells transfected with Mus81 siRNA or control siRNA for 48 hours were used for single fiber replication analyses: rates of replication fork progression (B) and inter-origin distances (C) in siRNA-treated cells. In comparisons between control- and Mus81-siRNA treated cells, the Mann Whitney test revealed significant differences in both replication fork velocity and inter-origin distance (Supplementary Table 2).

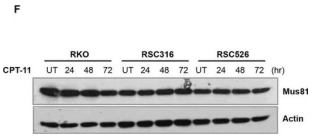


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Western blot - Anti-Mus81 antibody [MTA30 2G10/3] (ab14387)

Mus81 Western blot staining using mouse Anti-Mus81 antibody

Western Blotting using Anti-Mus81 antibody [MTA30 2G10/3], ab14387. Publication image from El-Khamisy, S. F. et al., 2017, Nucleic Acids Res, 28180300. Legend direct from paper.

Irinotecan-resistant cells display more fork restart events but no detectable change in HR factors. (A) Parental RKO and its derivative RSC316 irinotecan-resistant cells were pulse labeled with CldU for 20 min to label newly replicated DNA, followed by IdU for 30 min to measure replication fork speed. DNA fibres were spread on superfrost slides, fixed, and stored at 4°C. DNA fibers were subjected to acid denaturation using 2.5 M HCl. Slides were then stained with a mixture of rat anti-BrdU (clone BU1/75, Serotec) targeting CldU and mouse anti-BrdU (clone B44; Becton Dickinson) targeting IdU. DNA fibres were visualised using donkey anti-rat Alexafluor 488 and goat anti-mouse Alexafluor 555 secondary antibodies on an Olympus BX53 fluorescence upright microscope. Lengths of the fibres were measured, and speeds of the forks were determined using speed = length (kb)/time (min). Data are the average of three biological replicates ± SEM. Inset: Schematic depicting the DNA labelling procedure. (B) The average distribution of fork speeds is depicted from three biological replicates ± SEM. (C) A schematic showing the labelling procedure and the different replication fork structures; green-red signals represent on-going forks, green only represent stalled forks, and red only represent new origin firing. (D and E) Parental RKO and its derivative irinotecan-resistant cells were pulse labelled with CldU for 10 min to label newly replicated DNA, followed by treatment with DMSO ‘-CPT-11’ (D) or 50 μM CPT-11 ‘+CPT-11’ (E) for 30 min, followed by 1 h incubation with IdU. DNA fibres were spread on superfrost slides, fixed, and processed as described in ‘A’. At least 238 fork structures were counted per sample and the average number of stalled forks, fork restart ‘ongoing forks’, and new origin firing was quantified from three biological replicates ± SEM. Asterisks denote statistical significance; *P < 0.05, Student's t-test. (F and G) Cell lysates from the indicated cell lines were fractionated by SDS-

PAGE and analysed by immunoblotting. (H) Parental and resistant cells were exposed to CPT and mRNA extracted using RNeasy mini RNA purification kit. mRNA was subjected to microarray analysis using a HumanHT-12 v4 WG-GX Beadchip on the Illumina BeadArray system. Expression changes of HR genes in resistant cells were presented as fold change relative to the parental RKO cells. Data are the average of two independent biological repeats ± range.

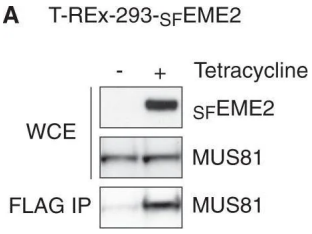


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Western blot - Anti-Mus81 antibody [MTA30 2G10/3] (ab14387)

Mus81 Western blot staining using mouse Anti-Mus81 antibody

Western Blotting using Anti-Mus81 antibody [MTA30 2G10/3], ab14387. Publication image from West, S. C. et al., 2014, Nucleic Acids Res, 24371268. Legend direct from paper.

Interactions of MUS81 with EME2 and EME2B. (A). Extracts were prepared from T-REx-293 cells expressing SFEME2 or SFEME2B from a tetracycline-inducible promoter and probed by western blotting for the indicated proteins. The SFEME1 or SFEME2 proteins were immunoprecipitated using FLAG antibodies and the immunoprecipitates were probed for the presence of MUS81. (B). Western blots showing the presence of EME2 in the indicated cell lines, with and without siRNA treatment. Beta-actin was used as a loading control. (C). The U2OS cells expressing SFEME2 from a tetracycline-inducible promoter were treated with cisplatin. The SFEME2 and MUS81 proteins were detected by immunofluorescence using anti-FLAG and anti-MUS81 antibodies, respectively. DNA was visualized by DAPI staining.

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