

#3625 Store at -20°C

NUT (C52B1) Rabbit mAb



100 µl
 (10 western blots)

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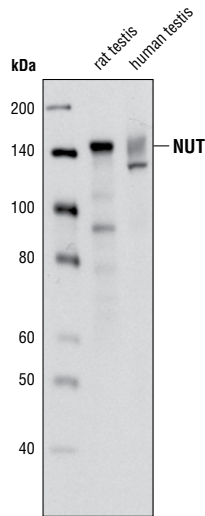
For Research Use Only. Not For Use In Diagnostic Procedures.

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IP, IHC-P, IF-F Endogenous	H, R, (Mk)	150 kDa	Rabbit IgG**

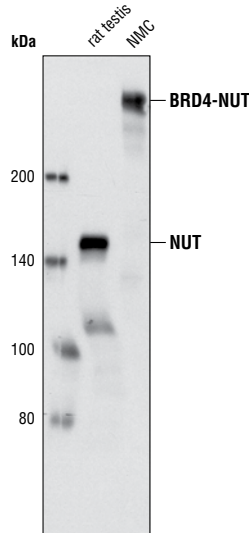
Background: Nuclear protein in testis (NUT) is normally confined to the germ cells of the testis and ovary (1,2). NUT midline carcinoma (NMC) is a recently recognized cancer that is defined by the presence of chromosomal rearrangements involving the NUT gene on chromosome 15q14 (3). In most cases the chromosomal translocation occurs between NUT and BRD4 on chromosome 19, resulting in the formation of a BRD4-NUT fusion protein. In the remaining tumors, variant NUT rearrangements are present involving BRD3, a very close homolog of BRD4. BRD4-NUT and BRD3-NUT encode fusion proteins that appear to contribute to carcinogenesis by blocking epithelial cell differentiation. NMCs, which are aggressive and highly lethal carcinomas, are morphologically indistinguishable from other poorly differentiated carcinomas. Given the limited expression of endogenous NUT protein, this antibody can be used to detect NUT fusion proteins in tissues by immunohistochemistry and immunofluorescence (2).

Specificity/Sensitivity: NUT (C52B1) Rabbit mAb detects endogenous levels of total NUT protein. The antibody also detects endogenous levels of the BRD4-NUT fusion protein found in NUT midline carcinoma (NMC).

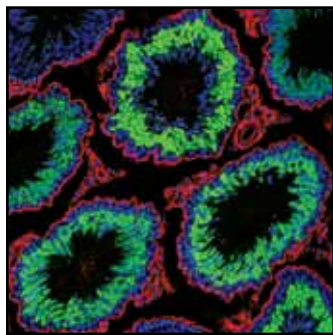
Source/Purification: Monoclonal antibody is produced by immunizing animals with a recombinant protein corresponding to the human NUT protein.



Western blot analysis of extracts from rat and human testis using NUT (C52B1) Rabbit mAb.



Western blot analysis of extracts from rat testis and NUT midline carcinoma (NMC) cells using NUT (C52B1) Rabbit mAb.



Confocal immunofluorescent analysis of rat testes using NUT (C52B1) Rabbit mAb (green) and Pan-Keratin (C11) Mouse mAb #4545 (red). Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye).

Entrez-Gene ID #256646
UniProt ID #Q5VZ89

Storage: Supplied in 10 mM HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. Do not aliquot the antibody.

***Species cross-reactivity is determined by western blot.**

****Anti-rabbit secondary antibodies must be used to detect this antibody.**

Recommended Antibody Dilutions:

Western blotting	1:1000
Immunoprecipitation	1:100
Immunohistochemistry (Paraffin)	1:45†
Unmasking buffer:	TE
Antibody diluent:	SignalStain® Antibody Diluent #8112
Detection reagent:	SignalStain® Boost (HRP, Rabbit) #8114
†Optimal IHC dilutions determined using SignalStain® Boost IHC Detection Reagent.	
Immunofluorescence (IF-F)	1:1600

For application specific protocols please see the web page for this product at www.cellsignaling.com.

Please visit www.cellsignaling.com for a complete listing of recommended companion products.

Selected Application References:

Haack, H. et al. (2009) Diagnosis of NUT Midline Carcinoma Using a NUT-specific Monoclonal Antibody. *Am J Surg Pathol* 33, 984-91. Application: IHC-P (paraffin).

Background References:

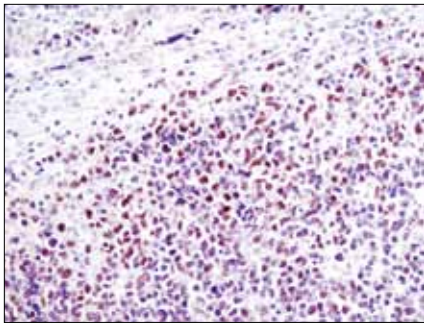
- (1) French, C.A. et al. (2003) *Cancer Res* 63, 304-7.
- (2) Haack, H. et al. (2009) *Am J Surg Pathol*, Epub ahead of print.
- (3) French, C.A. et al. (2008) *Oncogene* 27, 2237-42.

IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween®20 at 4°C with gentle shaking, overnight.

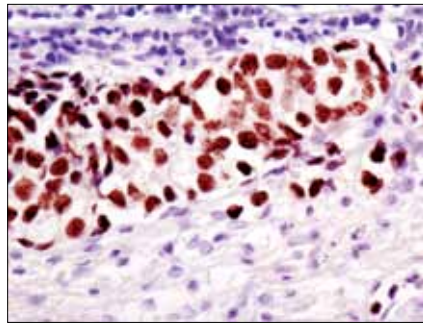
U.S. Patent No. 5,675,063

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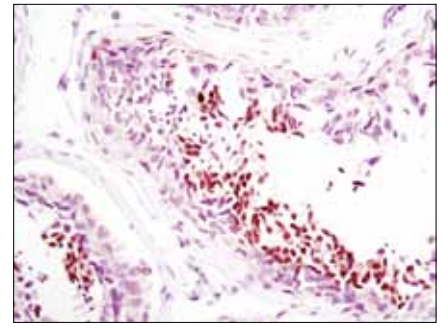
Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide
Species Cross-Reactivity Key: H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine
 Dg—dog Pg—pig Sc—S. cerevisiae Ce—C. elegans Hr—horse AI—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.



Immunohistochemical analysis of paraffin-embedded human midline carcinoma using NUT (C52B1) Rabbit mAb.



Immunohistochemical analysis of paraffin-embedded human midline carcinoma using NUT (C52B1) Rabbit mAb.



Immunohistochemical analysis of paraffin-embedded normal human testes using NUT (C52B1) Rabbit mAb.