

# Product Datasheet

## MYH6 Antibody (3-48) - Azide and BSA Free NB300-284

Unit Size: 0.1 mg

Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.

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Updated 10/23/2024 v.20.1

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**NB300-284**

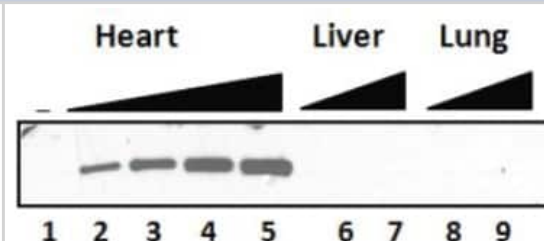
MYH6 Antibody (3-48) - Azide and BSA Free

Product Information	
Unit Size	0.1 mg
Concentration	1 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Clone	3-48
Preservative	No Preservative
Isotype	IgG1 Kappa
Purity	Protein G purified
Buffer	Lyophilized protein G purified in PBS pH7.4 (Full IgG)
Product Description	
Host	Mouse
Gene ID	4624
Gene Symbol	MYH6
Species	Human, Mouse, Rat, Canine
Reactivity Notes	Canine reactivity reported in (PMID: 23577316). Please note that this antibody is reactive to Mouse and derived from the same host, Mouse. Mouse-On-Mouse blocking reagent may be needed for IHC and ICC experiments to reduce high background signal. You can find these reagents under catalog numbers PK-2200-NB and MP-2400-NB. Please contact Technical Support if you have any questions. Predicted to react with Rabbit, Bovine.
Specificity/Sensitivity	This antibody recognizes alpha and beta type heavy chains of light meromyosin fragments (LMM) of cardiac myosin heavy chains. It also reacts with LMM fragments of cardiac myosin and beta heavy chain of slow human skeletal muscle. The antibody does not crossreact with human myosin light chains.
Immunogen	Human ventricular myosin
Product Application Details	
Applications	Western Blot, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin
Recommended Dilutions	Western Blot 1:500, Immunohistochemistry 1:300, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry-Paraffin 1:300
Application Notes	Please only use IgG specific secondaries with this product. Fab region specific secondaries may not detect this antibody. ICC/IF reactivity reported in (PMID: 23577316).

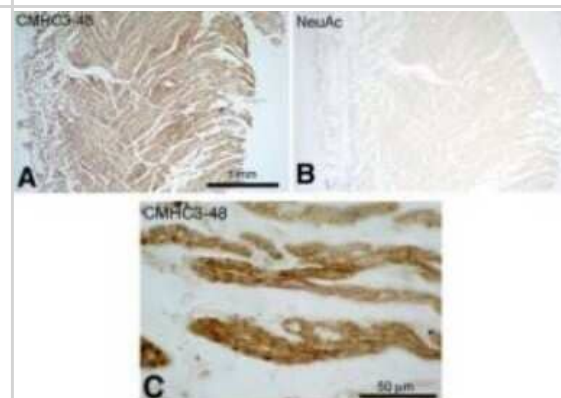


## Images

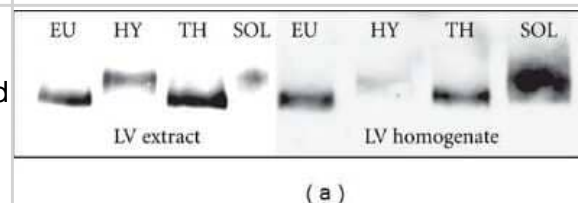
Western Blot: MYH6 Antibody (3-48) [NB300-284] - analysis of total cardiac myosin expression in tissue lysates of rat heart (lanes 1-5), liver (lanes 6-7), and lung (lanes 8-9). Increasing amounts of total protein were loaded as follow: lanes 1, 6 and 8: 0 ug; lane 2: 50 ug; lanes 3, 7 and 9: 250 ug; lane 5: 500 ug. (anti-cMHC was used at 1:500; secondary antibody: Goat anti-mouse IgG Fc-HRP; 1:5000)



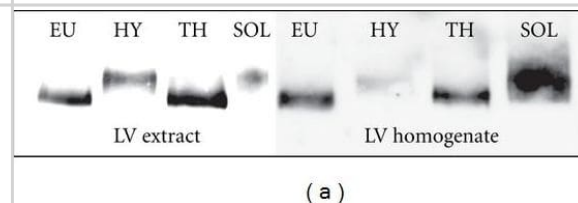
Immunohistochemistry-Paraffin: MYH6 Antibody (3-48) [NB300-284] - Formaldehyde-fixed paraffin-embedded (FFPE) Tissue Slides. Human heart ventricle following immunostaining with cMHC 3-48 and irrelevant NeuAc monoclonal antibody at low microscopical magnification.



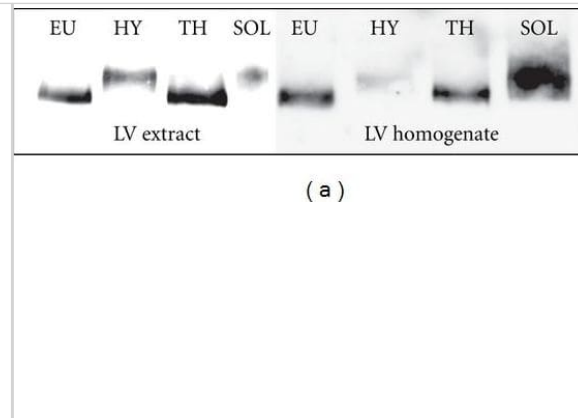
Western Blot: MYH6 Antibody (3-48) [NB300-284] - Western blot immunodetection of cardiac MyHC isoforms from left ventricle (LV) homogenates and extracts of adult euthyroid (EU), hypothyroid (HY), and hyperthyroid (TH) Lewis rats. The membranes were stained by NB300-284 antibody recognizing both MyHCalpha and MyHCbeta isoforms. Image collected and cropped by CiteAb from the following publication (<https://www.hindawi.com/journals/bmri/2011/634253/>), licensed under a CC-BY license.



Western blot immunodetection of cardiac MyHC isoforms from left ventricle (LV) homogenates and extracts of adult euthyroid (EU), hypothyroid (HY), and hyperthyroid (TH) Lewis rats. The membranes were stained by NB300-284 antibody recognizing both MyHCα and MyHCβ isoforms (a) or by BA.G5 antibody, which was solely specific for cardiac MyHCα isoform (b).



**Western Blot: MYH6 Antibody (3-48) - Azide and BSA Free [NB300-284]**  
 - Western blot immunodetection of cardiac MyHC isoforms from left ventricle (LV) homogenates & extracts of adult euthyroid (EU), hypothyroid (HY), & hyperthyroid (TH) Lewis rats. The membranes were stained by NB300-284 antibody recognizing both MyHC $\alpha$  & MyHC $\beta$  isoforms (a) or by BA.G5 antibody, which was solely specific for cardiac MyHC $\alpha$  isoform (b). Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/22187528>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



## Publications

Yefimova, M G, LefEvre, C Et al. Granulosa cells provide elimination of apoptotic oocytes through unconventional autophagy-assisted phagocytosis. Hum Reprod 2020-06-01 [PMID: 32531067]

Barrett P, Quick TJ, Mudera V, Player DJ. Neuregulin 1 Drives Morphological and Phenotypical Changes in C2C12 Myotubes: Towards De Novo Formation of Intrafusal Fibres In Vitro Frontiers in Cell and Developmental Biology 2022 -01-11 [PMID: 35087826] (Immunocytochemistry/ Immunofluorescence)

Barrett PL Nrg-1 effects on C2C12 differentiation: Towards generating Intrafusal muscle fibres in vitro Thesis 2022-01-01

Nonaka K, Akiyama J, Yoshikawa Y et al. 1,25-Dihydroxyvitamin D3 Inhibits Lipopolysaccharide-Induced Interleukin-6 Production by C2C12 Myotubes Medicina (Kaunas) 2020-09-04 [PMID: 32899782] (WB, Mouse)

Arnostova P, Jedelsky PL, Soukup T, Zurmanova J. Electrophoretic mobility of cardiac myosin heavy chain isoforms revisited: application of MALDI TOF/TOF analysis. J Biomed Biotechnol 2011-01-01 [PMID: 22187528] (Rat)

Ott HC, Matthiesen TS, Goh SK et al. Perfusion-decellularized matrix: using nature's platform to engineer a bioartificial heart. Nat Med 2008-02-01 [PMID: 18193059] (Rat)

Berkholz J, Kuzyniak W, Hoepfner M, Munz B. Overexpression of the skNAC gene in human rhabdomyosarcoma cells enhances their differentiation potential and inhibits tumor cell growth and spreading. Clin Exp Metastasis. 2014-09-11 [PMID: 25209525]

Berkholz J, Michalick L, Munz B. The E3 SUMO ligase Nse2 regulates sumoylation and nuclear-to-cytoplasmic translocation of skNAC-Smyd1 in myogenesis. J. Cell. Sci. 2014-07-07 [PMID: 25002400] (WB, Mouse)

### Details:

Myosin heavy chain antibody (clone 3-48) used in WB application as myogenic differentiation markers on C2C12 cells, a mouse myoblast cell line, transfected or not with Nse2-specific siRNA - siNse2 (Figure 3C).

Suzuki K, Kishiok Y, Wakamatsu J-I, Nishimura T. Decorin activates Akt downstream of IGF-IR and promotes myoblast differentiation Animal Science Journal 2013-03-27 [PMID: 23607268] (WB, Mouse)

Kang SB, Lee HS, Lim JY et al. Injection of porous polycaprolactone beads containing autologous myoblasts in a dog model of fecal incontinence J Korean Surg Soc 2013-04-01 [PMID: 23577316] (ICC/IF, IHC-P, Canine)

Broderick TL, Jankowski M, Wang D et al. Downregulation in GATA4 and Downstream Structural and Contractile Genes in the db/db Mouse Heart ISRN Endocrinology, vol 2012-01-01 [PMID: 22474596] (WB, Mouse)

Berger F, Berkholz J, Breustedt T et al. Skeletal muscle-specific variant of nascent polypeptide associated complex alpha (skNAC): Implications for a specific role in mammalian myoblast differentiation. European Journal of Cell Biology. 2011-12-09 [PMID: 22154550]

## Procedures

### Immunohistochemistry-Paraffin Protocol Specific for NB300-284: Myosin heavy chain Antibody (3-48)

Immunohistochemistry-Paraffin Protocol Specific for NB300-284: Myosin heavy chain Antibody (3-48):

#### Materials

- 1) 1 Phosphate buffered saline (pH 7.6): NaCl 137mmol/L, KCl 2.7mmol/L, Na<sub>2</sub>HPO<sub>4</sub> 4.3mmol/L, KH<sub>2</sub>PO<sub>4</sub> 1.4 mmol/L
- 2) Citrate buffer, 0.01 M, pH6.0, Sodium Citrate 3g, Citric acid 0.4g
- 3) 3% Hydrogen peroxide
- 4) Primary antibody
- 5) Blocking serum (normal serum)
- 6) Biotinylated secondary antibody
- 7) DAB staining kit

#### Methods

1. Dewax and hydration of slides using xylene and EtOH:

Dry slides for 20 min in a 60 C oven

Add Xylene, 2 x 10 min

100%, 95%, 80%, and 70% EtOH, 5 min each EtOH concentration

Rinse in PBS, 5'

- 2 Antigen retrieval method (only for paraffin slides)

- 1a. High-pressure antigen retrieval procedure (recommended method)

Place slides in a glass slide holder (ensure that the slide holder is completely filled with slides, slides without sections if necessary, to ensure even heating. The entire slide holder is immersed in 1000 ml of Citrate buffer (0.01M, pH6.0) within a pressure cooker

Once steam is produced, and ONLY when steam is visible, from the pressure cooker (usually 15-20 min), the required high-pressure will have been reached, and slides will be incubated for 2 min.

Turn off heat, and allow buffer and slides to cool to room temperature

Slides are then rinsed in PBS for 5 minutes

2. Add 3% hydrogen peroxide solution, 10'at RT, then PBS, 3X5'

3. Normal blocking serum, 20'at RT

4. Incubate with Primary Ab, 4C overnight or 1.5 hours at 37C

5. Rinse with PBS, 3 X 5' each rinse

6. Add Biotin-conjugated second antibody, 10'at RT

7. Rinse with PBS, 3 X 5' each rinse

8. Add Streptavidin-Peroxidase, 10'at RT

9. Rinse with PBS, 3 X 5' each rinse

10. Staining with DAB solution, 2-5'under microscope

11. Stop the reaction by washing in tap water

12. Counterstain in Haematoxylin for 3-5 minutes

13. 75%, 80%, 95% and 100% ethanol, 5x2', xylene 2 x 10'



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### **Products Related to NB300-284**

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HAF007	Goat anti-Mouse IgG Secondary Antibody [HRP]
NB720-B	Rabbit anti-Mouse IgG (H+L) Secondary Antibody [Biotin]
NBP1-43319-0.5mg	Mouse IgG1 Kappa Isotype Control (P3.6.2.8.1)
NBP2-36744PEP	MYH6 Recombinant Protein Antigen

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### **Limitations**

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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