

# Product Datasheet

## GADD153/CHOP Antibody (9C8)

### NB600-1335SS

Unit Size: 0.025 ml

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

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**NB600-1335SS**

GADD153/CHOP Antibody (9C8)

Product Information	
Unit Size	0.025 ml
Concentration	1 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Clone	9C8
Preservative	0.05% Sodium Azide
Isotype	IgG2b Kappa
Purity	Protein A purified
Buffer	Tris-Glycine and 0.15M NaCl
Target Molecular Weight	29 kDa
Product Description	
Host	Mouse
Gene ID	1649
Gene Symbol	DDIT3
Species	Human, Mouse, Rat, Primate
Reactivity Notes	Human, mouse, rat and primate.
Marker	ER Stress Marker
Immunogen	Full length mouse CHOP/GADD153 [Swiss-Prot# P35639]
Product Application Details	
Applications	Western Blot, Simple Western, Gel Super Shift Assays, Immunocytochemistry/Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin, Immunoprecipitation
Recommended Dilutions	Western Blot 1:500-1:1000, Simple Western 1:250, Immunohistochemistry 1:100, Immunocytochemistry/Immunofluorescence 1:100, Immunoprecipitation 1:10-1:500, Immunohistochemistry-Paraffin 1:100, Gel Super Shift Assays
Application Notes	<p>This CHOP/GADD153 Antibody (9C8) is useful for Immunoprecipitation, Immunocytochemistry/Immunofluorescence, Immunohistochemistry on paraffin-embedded sections and Western blot, where a band can be seen at approx. 29 kDa. Use in Rat reported in customer review. Gel Super Shift Assays was reported in scientific literature.</p> <p>In Simple Western only 10 - 15 uL of the recommended dilution is used per data point. Separated by Size-Wes, Sally Sue/Peggy Sue. The observed molecular weight of the protein may vary from the listed predicted molecular weight due to post translational modifications, post translation cleavages, relative charges, and other experimental factors.</p>

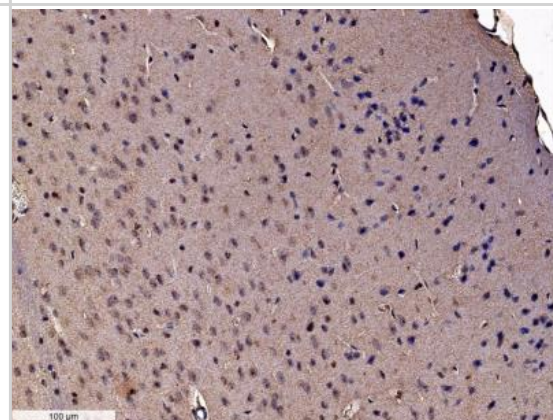


## Images

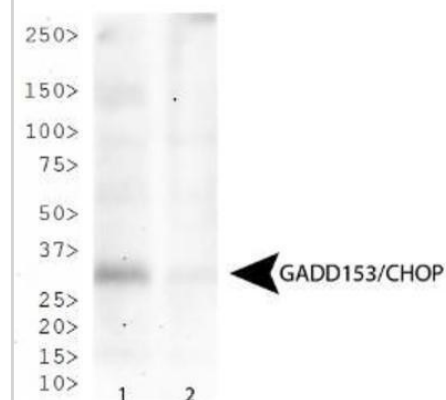
Western Blot: GADD153/CHOP Antibody (9C8) [NB600-1335] - WB analysis of CHOP in rat heart tissue lysate. Image courtesy of product review submitted by Lee Hsiao-Wei.



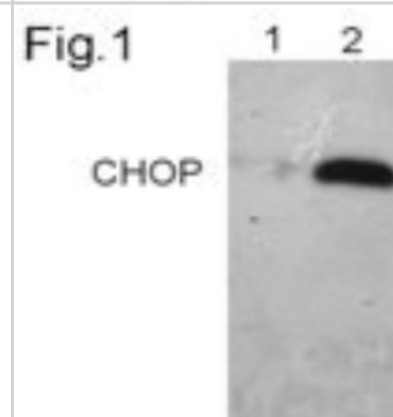
Immunohistochemistry-Paraffin: GADD153/CHOP Antibody (9C8) [NB600-1335] - IHC analysis of a formalin fixed and paraffin embedded (FFPE) tissue section of mouse brain using 1:100 dilution of Calreticulin antibody. The signal was developed using HRP-DAB based detection method which followed counterstaining of the nuclei with hematoxylin. The antibody generated a cytoplasmic and nuclear staining of CHOP in various cell types in the tested section.



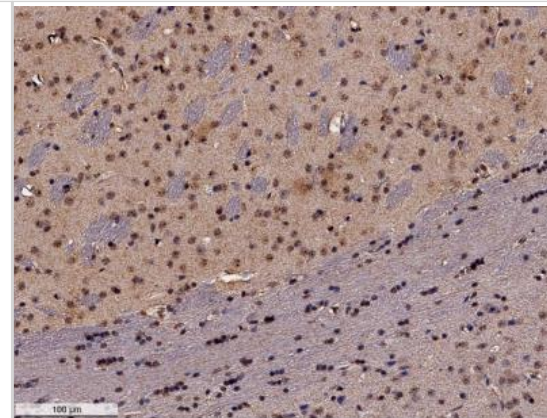
Western Blot: GADD153/CHOP Antibody (9C8) [NB600-1335] - Western blot analysis of GADD153/CHOP expression in HeLa cells treated with 2.5ug/ml tunicamycin for 4 hours (Lane 1) and untreated (Lane 2).



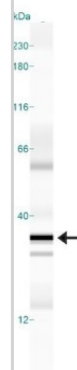
Western Blot: GADD153/CHOP Antibody (9C8) [NB600-1335] - Figure 1 shows a Western blot of endogenous CHOP/GADD153 from primary human fibroblasts using NB600-1335. Lane 1: Untreated cells, Lane 2: Cells treated with tunicamycin for 10 hours.



**Immunohistochemistry-Paraffin: GADD153/CHOP Antibody (9C8) [NB600-1335]** - IHC analysis of a formalin fixed and paraffin embedded (FFPE) tissue section of mouse brain using 1:100 dilution of Calreticulin antibody. The signal was developed using HRP-DAB based detection method which followed counterstaining of the nuclei with hematoxylin. The antibody generated a cytoplasmic and nuclear staining of CHOP in various cell types in the tested section.



**Simple Western: GADD153/CHOP Antibody (9C8) [NB600-1335]** - Simple Western lane view shows a specific band for CHOP/GADD153 in 1.0 mg/ml of HeLa lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.



## Publications

Kilaparty SP, Agarwal R, Singh P et al. Endoplasmic reticulum stress-induced apoptosis accompanies enhanced expression of multiple inositol polyphosphate phosphatase 1 (Minpp1): a possible role for Minpp1 in cellular stress response. *Cell Stress Chaperones*. 2016 Apr 02 [PMID: 27038811] (WB, Mouse)

### Details:

Mouse monoclonal CHOP antibody was used for WB analysis of lysates from MC3T3-E1 cells which were incubated or not with ER stress inducers brefeldin A and thapsigargin.

Ying R, Wang XQ, Yang Y et al. Hydrogen sulfide suppresses endoplasmic reticulum stress-induced endothelial-to-mesenchymal transition through Src pathway. *Life Sci*. 2016 Jan 01 [PMID: 26656263] (WB, Human)

Djelti F, Braudeau J, Hudry E et al. CYP46A1 inhibition, brain cholesterol accumulation and neurodegeneration pave the way for Alzheimer's disease *Brain* 2015 Aug 01 [PMID: 26141492] (IHC-P, Mouse)

### Details:

CHOP antibody was used for IHC-P on brain tissues from wild-type C57Bl/6 and 5-month-old APP23 transgenic mice (Thy1-hAPPswe) that were subjected to injection of AAV-scramble or AAV-shCYP46A1 vectors. The IHC-P assay implicated 4% paraformaldehyde fixation at 4C for 24 hours, 6 um paraffin sections, sodium citrate based heat induced antigen retrieval, permeabilization in PBS/0.1% Triton X 100, use of primary at 1:500 dilution with ON 4C incubation, detection using fluorescence labelled secondary (Figure 3A).

Lee YJ, Ha YJ, Na Kang Y et al. The Autophagy-Related Marker LC3 Can Predict Prognosis in Human Hepatocellular Carcinoma. *PLoS One*. 2013 Nov 25 [PMID: 24282606] (IHC-P, Human)

Lin CJ, Lee CC, Shih YL et al. Inhibition of mitochondria- and endoplasmic reticulum stress-mediated autophagy augments temozolomide-induced apoptosis in glioma cells. *PLoS One* 2012 [PMID: 22745676] (WB, Human)

Hull RL, Zraika S, Udayasankar J, Aston-Mourney K, Subramanian SL, Kahn SE. Amyloid formation in human IAPP transgenic mouse islets and pancreas, and human pancreas, is not associated with endoplasmic reticulum stress. *Diabetologia*. 2009 Jun. [PMID: 19352619]

Sok J, Wang XZ, Batchvarova N, Kuroda M, Harding H, Ron D. CHOP-Dependent stress-inducible expression of a novel form of carbonic anhydrase VI. *Mol Cell Biol*;19(1):495-504. 1999 Jan. [PMID: 9858573] (WB)

Zinszner H, Sok J, Immanuel D, Yin Y, Ron D. TLS (FUS) binds RNA in vivo and engages in nucleo-cytoplasmic shuttling. *J Cell Sci*. 1997 Aug. [PMID: 9264461] (IP, ICC/IF, Human)

Saisanit S, Sun XH. Regulation of the pro-B-cell-specific enhancer of the *Id1* gene involves the C/EBP family of proteins. *Mol Cell Biol* 17(2):844-50. 1997 Feb. [PMID: 9001238] (WB, IP, GS, Mouse)

Wang XZ, Lawson B, Brewer JW, Zinszner H, Sanjay A, Mi LJ, Boorstein R, Kreibich G, Hendershot LM, Ron D. Signals from the stressed endoplasmic reticulum induce C/EBP-homologous protein (CHOP/GADD153). *Mol Cell Biol* 16(8):4273-80. 1996 Aug. [PMID: 8754828] (WB, Human, Mouse)

Wang XZ, Harding HP, Zhang Y, Jolicoeur EM, Kuroda M, Ron D. Cloning of mammalian Ire1 reveals diversity in the ER stress responses. *EMBO J*;17(19):5708-17. 1998 Oct 1. [PMID: 9755171] (ICC/IF, Human, Primate)

Wang XZ, Kuroda M, Sok J, Batchvarova N, Kimmel R, Chung P, Zinszner H, Ron D. Identification of novel stress-induced genes downstream of chop. *EMBO J*;17(13):3619-30. 1998 Jul 1. [PMID: 9649432] (WB, IP, Mouse)

More publications at <http://www.novusbio.com/NB600-1335>



**Procedures****Western Blot Protocol for NB600-1335**

## Western Blot Protocol

1. Perform SDS-PAGE on samples to be analyzed, loading 40 ug of total protein per lane.
  2. Transfer proteins to membrane according to the instructions provided by the manufacturer of the membrane and transfer apparatus.
  3. Stain according to standard Ponceau S procedure (or similar product) to assess transfer success, and mark molecular weight standards where appropriate.
  4. Rinse the blot.
  5. Block the membrane using standard blocking buffer for at least 1 hour.
  6. Wash the membrane in wash buffer three times for 10 minutes each.
  7. Dilute primary antibody in blocking buffer and incubate 1 hour at room temperature.
  8. Wash the membrane in wash buffer three times for 10 minutes each.
  9. Apply the diluted HRP conjugated secondary antibody in blocking buffer (as per manufacturers instructions) and incubate 1 hour at room temperature.
  10. Wash the blot in wash buffer three times for 10 minutes each (this step can be repeated as required to reduce background).
  11. Apply the detection reagent of choice in accordance with the manufacturers instructions.
- Note: Tween-20 can be added to the blocking or antibody dilution buffer at a final concentration of 0.05-0.2%.

\*The above information is only intended as a guide. The researcher should determine what protocol best meets their needs. Please follow safe laboratory procedures.





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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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