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Datasheet for 200-301-A28

Hsc70 (Hsp73) Antibody

Overview

Description:	Anti-HSC70 (HSP73) (MOUSE) Monoclonal Antibody - 200-301-A28
Item No.:	200-301-A28
Size:	100 µg
Applications:	FC, IF, IHC, IP, WB
Reactivity:	Human, Mouse, Rat, Beluga, Bovine, C. elegans, Carp, Chicken, Cucumber, D. melanogaster, Dog, Guinea Pig, Hamster, Monkey, Pea, Pig, Rabbit, Salmon, Sheep, Trout, Xenopus
Host Species:	Mouse

Product Details

Background:	Hsp70 genes encode abundant heat-inducible 70-kDa hsps (hsp70s). In most eukaryotes, hsp70 genes exist as part of a multigene family. Hsp70s are found in most cellular compartments of eukaryotes, including nuclei, mitochondria, chloroplasts, the endoplasmic reticulum and the cytosol, as well as in bacteria. The genes show a high degree of conservation, having at least 50% identity (2). The N-terminal two-thirds of hsp70s are more conserved than the C-terminal one-third. Hsp70 binds ATP with high affinity and possesses a weak ATPase activity which can be stimulated by binding to unfolded proteins and synthetic peptides (3). When hsc70 (constitutively expressed) present in mammalian cells was truncated, ATP binding activity was found to reside in an N-terminal fragment of 44 kDa which lacked peptide binding capacity. Polypeptide binding ability therefore resided within the C-terminal half (4). The structure of this ATP binding domain displays multiple features of nucleotide binding proteins (5). All hsp70s, regardless of location, bind proteins, particularly unfolded ones. The molecular chaperones of the hsp70 family recognize and bind to nascent polypeptide chains as well as partially folded intermediates of proteins, preventing their aggregation and misfolding. The binding of ATP triggers a critical conformational change leading to the release of the bound substrate protein (6). The universal ability of hsp70s to undergo cycles of binding to and release from hydrophobic stretches of partially unfolded proteins determines their role in a great variety of vital intracellular functions such as protein synthesis, protein folding and oligomerization, and protein transport.
Synonyms:	Heat shock cognate protein 71-kDa antibody, Heat shock protein 8 antibody, Heat-shock70-KD protein 10, formerly antibody, HSC54 antibody, HSC71 antibody, Hsc54, Hsc71, Hsc73, Hsp71, Hsp73, HspA10, HspA8, LAP1, NIP71, Heat shock cognate 71 kDa protein, Heat shock 70 kDa protein 8
Host Species:	Mouse

Clonality:	Monoclonal
Clone ID:	N27F3-4
Format:	IgG1

Target Details

Gene Name:	HSPA8
Reactivity:	Human, Mouse, Rat, Beluga, Bovine, C. elegans, Carp, Chicken, Cucumber, D. melanogaster, Dog, Guinea Pig, Hamster, Monkey, Pea, Pig, Rabbit, Salmon, Sheep, Trout, Xenopus
Immunogen Type:	Conjugated Peptide
Immunogen:	This Protein G purified monoclonal antibody was prepared using conventional hybridoma technology after repeated immunizations with a synthetic peptide corresponding to a region of human Hsc70 (Hsp73) protein.
Purity/Specificity:	This Protein G purified monoclonal antibody reacts with human Hsc70 (Hsp73) protein. A BLAST analysis was used to suggest cross-reactivity with Hsc70 from human, bovine, mouse, rat, C.elegans, beluga, dog, chicken, Drosophila, carp, salmon, trout, guinea pig, hamster, monkey, pig, cucumber, pea, rabbit, sheep and Xenopus sources based on 100% homology with the immunizing sequence. This antibody recognizes both the inducible hsp and the constitutively expressed hsc as 72 kDa and 73 kDa proteins, respectively. Cross-reactivity with Hsp70 from other sources has not been determined.
Relevant Links:	• NCBI - NP_006588.1 • UniProtKB - P11142 • GeneID - 3312

Application Details

Tested Applications:	FC, IF, IHC, IP, WB
Application Note:	This Protein G purified antibody has been tested for use in western blotting, immunoelectron microscopy, immunohistochemistry-P, and immunoprecipitation. Specific conditions for reactivity should be optimized by the end user. Expect a doublet band approximately 72/73 kDa in size corresponding to Hsc70 (Hsp73) by western blotting in the appropriate cell lysate or extract. In general, a 1:1,000 dilution is suggested for most applications and is suitable to detect Hsc70 (Hsp73) in 20 µg of heat shocked HeLa cell lysate by western blotting.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ELISA:	1:10,000 - 1:50,000
IHC:	1:200 - 1:1,000

WB:	1:500 - 1:2,000

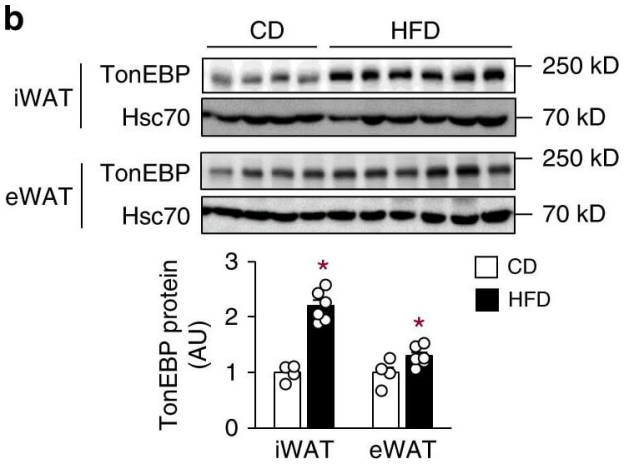
Formulation

Physical State:	Liquid (sterile filtered)
Concentration:	1.0 mg/mL by UV absorbance at 280 nm
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.09% (w/v) Sodium Azide
Stabilizer:	50% (v/v) Glycerol

Shipping & Handling

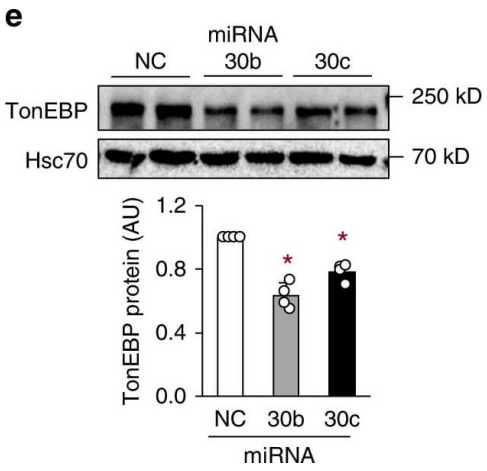
Shipping Condition:	Wet Ice
Storage Condition:	Store vial at -20° C prior to opening. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use. For extended storage aliquot contents and freeze at -20° C or below. Avoid cycles of freezing and thawing.
Expiration:	Expiration date is one (1) year from date of receipt.

Images



Western Blot

Adipocyte TonEBP expression is elevated in obesity and TonEBP-deficient mice resist obesity. aTonEBP mRNA levels in iWAT, eWAT, BAT, muscle, and liver from C57BL/6 J mice fed with CD (chow diet, n = 5) or HFD (high-fat diet, n = 7) for 16 weeks. b Immunoblots (top) and quantification of protein levels (bottom) of TonEBP and Hsc70 in iWAT and eWAT (CD, n = 4; HFD, n = 6). c Correlation of TONEBP mRNA levels in human subcutaneous adipocytes and BMI (n = 15). TonEBP mRNA (d) and representative immunoblots (e, top) and quantification of protein levels (e, bottom) in 3T3-L1 adipocytes transfected with miR-negative control (NC), miR-30b (30b), or miR-30c (30c) (n = 4). f–iTonEBP+/Δ mice (+ /Δ) and their TonEBP+/+ littermates (+/+) were fed with CD or HFD as indicated. f Changes in body weight after a switch to HFD (CD+/+, n = 7; CD + /Δ, n = 7; HFD+/+, n = 13; HFD+/Δ, n = 11). g Representative images of animals fed with HFD. h Body weight, height, fat mass, and lean mass (n = 8). i Representative images (top) and weight (bottom) of fat pads from HFD-fed animals (n = 4). n represents number of biologically independent samples (a–c, i) or animals (f–h) or independent experiments with triplicate (d, e). All data are presented as mean + s.e.m. (a, b, f, h, i) or + s.d. (d, e). AU, arbitrary unit. The p-values are determined by unpaired t-test (a,b, h, i) or one-way ANOVA (d–f). *p < 0.05 vs. CD (a), NC (d), or +/+ (f, h, i). Source data are provided as a Source Data file Figure provided by CiteAb. Source: Nat Commun, PMID: 31387996.



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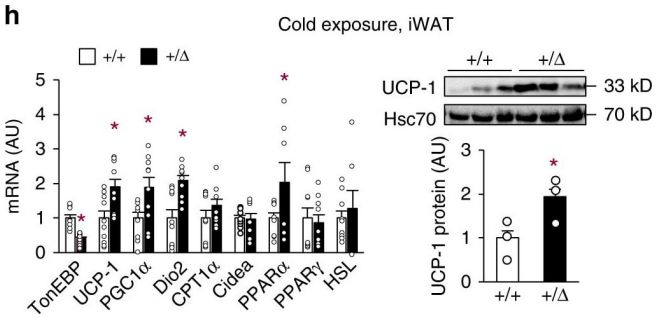
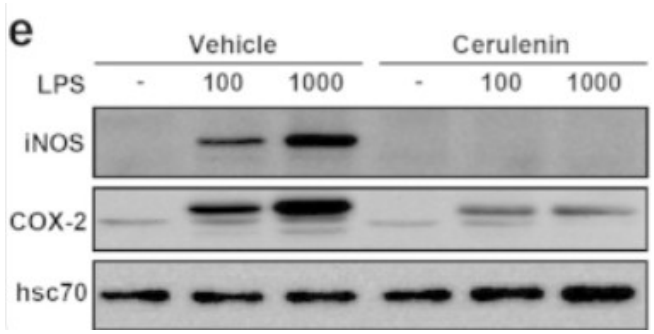


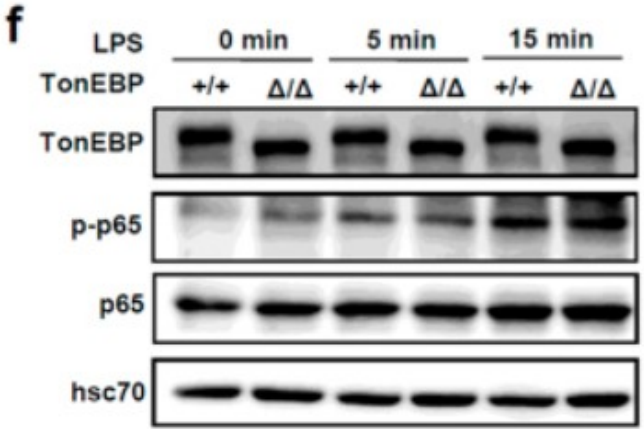
Western Blot

TonEBP deficiency promotes energy expenditure and beiging of WAT. HFD-fed animals were analyzed by indirect calorimetry to obtain VO2 (a), VCO2 (b), and heat production (c) (n = 4). Rectal temperature (temp.) measured in CD-fed animals at room temperature (n = 8) (d) and after exposure to cold up to 6 h as indicated (4 °C) (n = 6) (e). f, g mRNA abundance of thermogenic genes (f) and beiging marker genes (g) in iWAT of HFD-fed animals (n = 5). h mRNA abundance of thermogenic genes (left, n = 10) and immunoblots of UCP-1 and Hsc70 (right, n = 3) in iWAT of CD-fed animals exposed to cold (4 °C). i Representative images of iWAT sections stained with H&E and UCP-1 antibody from CD-fed animals exposed to cold (4 °C). Scale bars, 100 µm. j, k Thermogenic gene (j) and beige marker (k) mRNA abundance in beige adipocytes differentiated from the stromal vascular cells of iWAT (n = 4). n represents number of biologically independent animals (a–e) or samples (f–k). a–h, j, k All data are presented as mean + s.e.m. AU arbitrary unit. The p-values are determined by unpaired t-test (d, f–k) or one-way ANOVA (a–c, e). *p < 0.05 vs. +/+. Source data are provided as a Source Data file Figure provided by CiteAb. Source: Nat Commun, PMID: 31387996.

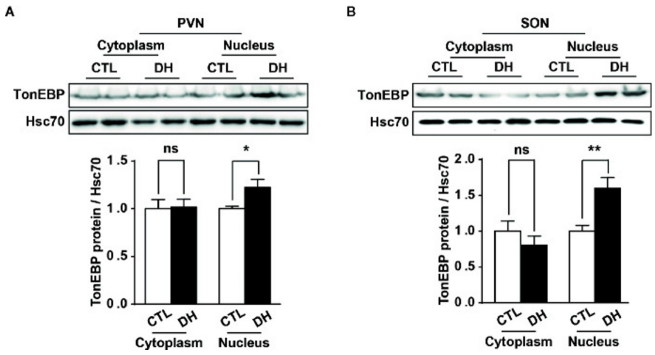


Western Blot

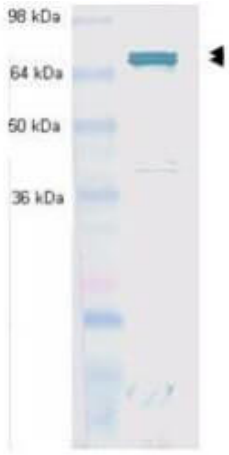
(e) PECs pre-treated for 1 h with vehicle or cerulenin (10 µM) were stimulated for 24 h with 0, 100, and 1,000 ng/ml of LPS. Expression of iNOS and COX-2 was visualized by immunoblotting.
Fig 8
PMID: 27118681



Western Blot
(f) Cells were treated with LPS for 0, 5, and 15 min as indicated, and immunoblotted for TonEBP, serine 276 phosphorylated p65 (p-p65), p65, and hsc70.
Fig 6
PMID: 27118681



Western Blot
Dehydration-induced nuclear translocation of TonEBP in the PVN and SON. Western blotting analysis was used to quantify cytoplasmic or nucleus TonEBP in the PVN and SON in mice deprived of water for 2 days and euhydrated control mice. Representative images show intracellular region-specific TonEBP proteins in the PVN (A) and SON (B), and quantificational analyses demonstrate that dehydration resulted in an increased TonEBP nuclear translocation in both regions. CTL, euhydrated control; DH, 2 days of dehydration; N= 5 mice per group. Data are presented as mean ± SEM. *p < 0.05; **p < 0.01; ns, not significant.
Fig 3 PMID: 33796071



Western Blot
Western blot using Rockland's Protein G purified anti-Hsc70 (Hsp73) antibody shows detection of Hsc70 (Hsp73) in whole cell lysates from heat shocked HeLa cells. The band marked by the double arrowheads corresponds to Hsc70 (Hsp73) at an approximate molecular weight of 72/73 kDa. The primary antibody was used at a 1:1,000 dilution.

References

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- Y BJ et al. TonEBP in dendritic cells mediates pro-inflammatory maturation and Th1/Th17 responses. *Cell Death Dis.* (2020)
- Lee HH et al. TonEBP/NFAT5 promotes obesity and insulin resistance by epigenetic suppression of white adipose tissue beiging. *Nat Commun.* (2019)
- Lee et al. LPS-induced NFκB enhanceosome requires TonEBP/NFAT5 without DNA binding. *Scientific Reports* (2016)

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