Product Datasheet

Aggrecan Antibody (BC-3) - BSA Free
NB600-504-0.1mg

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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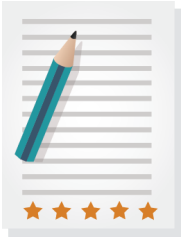
Updated 10/23/2024 v.20.1



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Aggrecan Antibody (BC-3) - BSA Free

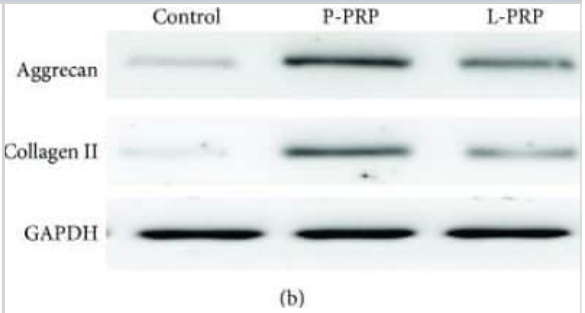
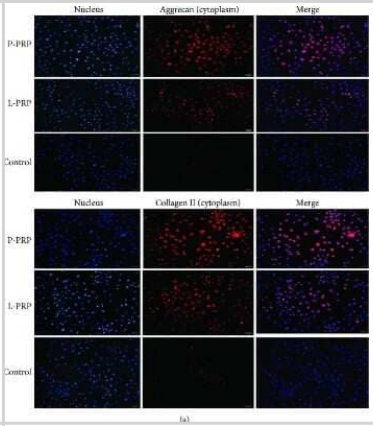
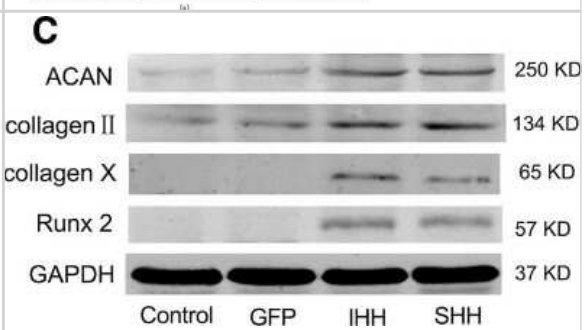
Product Information	
Unit Size	0.1 mg
Concentration	1.0 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Clone	BC-3
Preservative	0.02% Sodium Azide
Isotype	IgG1 Kappa
Purity	Protein A or G purified
Buffer	PBS
Target Molecular Weight	250 kDa

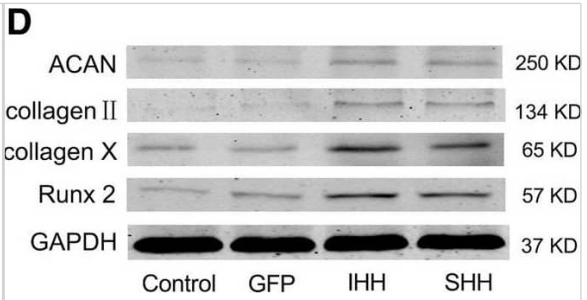
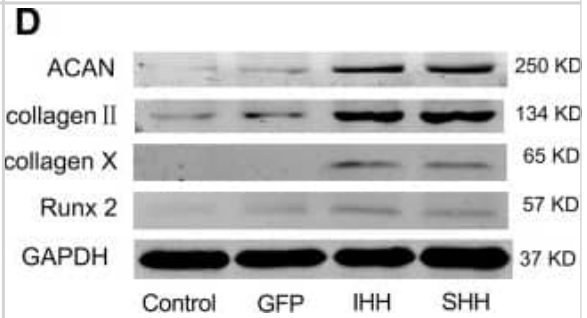
Product Description	
Host	Mouse
Gene ID	176
Gene Symbol	ACAN
Species	Human, Mouse, Rat, Porcine, Bovine, Canine, Equine, Feline, Guinea Pig, Rabbit, Sheep
Reactivity Notes	Cross-reacts with Human, Cow, Cat, Dog, Guinea pig, Horse, Pig, Rabbit, Rat and Sheep. Mouse reactivity reported in scientific literature (PMID: 27185069). Not yet tested in other species.
Marker	Chondrocyte Marker
Specificity/Sensitivity	Recognizes the aggrecanase (ADAMTS-1, -4 & -5)-generated N-terminal neoepitope ARG after cleavage between amino acids EGE and ARG within the interglobular domain of aggrecanase-catabolised aggrecan.
Immunogen	ARGSV... synthetic peptide conjugate.

Product Application Details	
Applications	Western Blot, ELISA, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin
Recommended Dilutions	Western Blot 1:100, ELISA 1:100-1:2000, Immunohistochemistry 1:10-1:500, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry-Paraffin 1:10-1:500, Immunohistochemistry-Frozen 1:10-1:500

Application Notes	This Aggrecan antibody is useful in Immunohistochemistry, ELISA and Western blot. With IHC the antibody works in formalin- or paraformaldehyde-fixed paraffin embedded sections as well as either alcohol-fixed frozen sections or un-fixed snap-frozen sections. In Western blot the antibody detects a variety of epitopes between 50 and 250 kDa. Samples must be deglycosylated using 0.01 Units Chondroitinase ABC (Sigma), 0.01 Units Keratanase (Seikagaku) and 0.0001 Units Keratanase II (Seikagaku) per 10ug S-GAG of non-deglycosylated aggrecan for optimal epitope recognition. See Little et al. and Caterson et al. 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. Use in Immunocytochemistry/immunofluorescence reported in scientific literature (PMID 27185069).

Images

Western Blot: Aggrecan Antibody (BC-3) [NB600-504] - P-PRP induces more extracellular matrix-related proteins. Production of collagen II and aggrecan, as measured by western blot. Image collected and cropped by Citeab from the following publication (The Differential Effects of Leukocyte-Containing and Pure Platelet-Rich Plasma on Nucleus Pulposus-Derived Mesenchymal Stem Cells: Implications for the Clinical Treatment of Intervertebral Disc Degeneration. *Stem Cells Int* (2018)) licensed under a CC-BY license.	
Immunocytochemistry/Immunofluorescence: Aggrecan Antibody (BC-3) [NB600-504] - P-PRP induces more extracellular matrix-related proteins. Collagen II and aggrecan in the cytoplasm of the coculture cells imaged by fluorescence microscopy. Image collected and cropped by Citeab from the following publication (The Differential Effects of Leukocyte-Containing and Pure Platelet-Rich Plasma on Nucleus Pulposus-Derived Mesenchymal Stem Cells: Implications for the Clinical Treatment of Intervertebral Disc Degeneration. Stem Cells Int (2018)) licensed under a CC-BY license.	
Western Blot: Aggrecan Antibody (BC-3) [NB600-504] - Expression levels of related genes during differentiation induction in the RCCS environment. Expression of ACAN, collagen II, collagen X and Runx2 was detected by western blotting on day 10 during differentiation induction. Image collected and cropped by Citeab from the following publication (Chondrogenic differentiation of bone marrow-derived mesenchymal stem cells following transfection with Indian hedgehog and sonic hedgehog using a rotary cell culture system. Cell Mol Biol Lett (2019)) licensed under a CC-BY license.	

Western Blot: Aggrecan Antibody (BC-3) - BSA Free [NB600-504] - Expression levels of related genes during differentiation induction in the 2D environment. (a) qRT-PCR analysis of Sox9, ACAN & collagen II on days 7, 14 & 21 during induction. (b) qRT-PCR analysis of collagen X, Runx2 & annexin V on days 7, 14 & 21 during induction. The results were normalized to B2M mRNA expression. Values are means ± SD (n = 3). (c) Expression of ACAN, collagen II, collagen X & Runx2 was detected by western blotting on day 10 during induction. (d) Expression of ACAN, collagen II, collagen X & Runx2 was detected by western blotting on day 21 during induction. Significant differences from the control group (non-transfection cells) are indicated by *p < 0.05 or **p < 0.01; differences between IHH & SHH transfection groups are indicated by #p < 0.05 or ##p < 0.01 Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/30858866), licensed under a CC-BY license. Not internally tested by Novus Biologicals.	
Western Blot: Aggrecan Antibody (BC-3) - BSA Free [NB600-504] - Expression levels of related genes during differentiation induction in the RCCS environment. (a) qPCR analysis of Sox9, ACAN & collagen II on days 7, 14 & 21 during induction. (b) qRT-PCR analysis of collagen X, Runx2, & annexin V on days 7, 14 & 21 during induction. The results were normalized to B2M mRNA expression. Values are means ± SD (n = 3). (c) Expression of ACAN, collagen II, collagen X & Runx2 was detected by western blotting on day 10 during differentiation induction. (d) Expression of ACAN, collagen II, collagen X & Runx2 was detected by western blotting on day 21 during differentiation induction. Significant differences from the control group (non-transfection cells) are indicated by *p < 0.05 or ** p < 0.01; differences between IHH & SHH transfection groups are indicated by #p < 0.05 or ##p < 0.01 Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/30858866), licensed under a CC-BY license. Not internally tested by Novus Biologicals.	

Publications

Hosseinzadeh M, Kamali A, Baghaban Eslaminejad M, Hosseini S Higher ratios of chondrocyte to mesenchymal stem cells elevate the therapeutic effects of extracellular vesicles harvested from chondrocyte/mesenchymal stem cell co-culture on osteoarthritis in a rat model Cell and tissue research 2023-08-01 [PMID: 37526734]

Qingxin S, Kai J, Dandan Z et al. Programmable DNA hydrogel provides suitable microenvironment for enhancing autophagy-based therapies in intervertebral disc degeneration treatment J Nanobiotechnology 2023-09-28 [PMID: 37759249] (Immunocytochemistry/ Immunofluorescence)

Luo Z, Liu Z, Yu H et al. Multifunctional mesoporous polydopamine near-infrared photothermal controlled release of kartogenin for cartilage repair Materials & Design 2023-07-01 (WB, Rat)

Esmaeili A, Hosseini S, Kamali A et al. Co-aggregation of MSC/chondrocyte in a dynamic 3D culture elevates the therapeutic effect of secreted extracellular vesicles on osteoarthritis in a rat model Scientific reports 2022-11-18 [PMID: 36400827] (WB, Rabbit)

Hosseinzadeh M, Kamali A, Hosseini S, Baghaban Eslaminejad M Higher Chondrogenic Potential of Extracellular Vesicles Derived from Mesenchymal Stem Cells Compared to Chondrocytes-EVs In Vitro BioMed research international 2021-12-13 [PMID: 34938811] (WB, Rabbit)

Hughes EC, Caterson B, Fosang AJ et al. Monoclonal antibodies that specifically recognize neoepitope sequences generated by 'aggrecanase' and matrix metalloproteinase cleavage of aggrecan: application to catabolism in situ and in vitro Biochem J. 1995-02-01 [PMID: 7531436]

Jung H, Mclellan P, Welter JF, Akkus O Chondrogenesis Mesenchymal Stem Cells via Local Release of TGF beta 3 from Heparinized Collagen Biofabric Tissue engineering. Part A 2021-04-07 [PMID: 33827271]

Baei P, Daemi H, Mostafaei F et al. A tough polysaccharide-based cell-laden double-network hydrogel promotes articular cartilage tissue regeneration in rabbits Chemical Engineering Journal 2021-08-01 (ICC/IF, Rabbit)

Ma S N, Xie Z G et al. Effect of Acupotomy on FAK-PI3K Signaling Pathways in KOA Rabbit Articular Cartilages. Evid Based Complement Alternat Med 2017-12-14 [PMID: 29234400] (WB, Rabbit)

He W, Wang W, Deng J et al. L-NMMA AND 1400W, INHIBITORS OF iNOS, ATTENUATE THE INDUCTION OF iNOS AND NO IN PRIMARY RABBIT COSTAL CHONDROCYTES BY FLUORIDE Fluoride 2020-01-01 (WB, Rabbit)

Hurley-Novatny AC, Arumugasaamy N, Kimicata M et al. Concurrent multi-lineage differentiation of mesenchymal stem cells through spatial presentation of growth factors Biomed Mater 2020-06-11 [PMID: 32526725]

Si HB, Yang TM, Li L et al. miR-140 Attenuates the Progression of Early-Stage Osteoarthritis by Retarding Chondrocyte Senescence Mol Ther Nucleic Acids 2019-11-09 [PMID: 31790972] (WB, IHC-P, Rat, Human)

More publications at http://www.novusbio.com/NB600-504

Procedures

Protocol specific for Aggrecan Neoepitope Antibody (NB600-504)
This protocol is from the reference Roberts S et al. Matrix metalloproteinases and aggrecanase: their role in disorders of the human intervertebral disc. Spine 25:3005-13 (2000). PubMed: 11145811

1. Immunostaining with enzyme-generated antibodies BC3, 13, 4, and 14 was carried out on 19 discs from patients 11-59 years of age (1 with prolapse, 2 with scoliosis, 2 with spondylolisthesis, and the remainder with degenerative disc disease).

2. Different fixation treatments were used to optimize antigen preservation and staining of the cryosections; for example, 70% ethanol, 100% ethanol, 10% formaldehyde, or no fixation was used.

3. After this treatment, sections to be stained for BC 3 and 14 were digested with keratanase I, II, and chondroitinase ABC for 3 hours, and those to be stained for BC4 and 13 were digested with chondroitinase ABC for only 90 minutes.

4. Labeling was as for paraffin sections with other antibodies.

5. Staining was done on 5 um paraffin sections that then were deparaffinized with xylene and rehydrated through a series of alcohols to phosphate buffered saline (PBS). Staining conditions were optimized for each individual antibody... no pretreatment was found necessary.

6. Sequential blocking of endogenous peroxidase activity was performed with 0.3% hydrogen peroxide in PBS and then 20% normal human and horse serum and 3% bovine serum albumin in PBS.

7. The primary antibody was incubated for 90 minutes at room temperature before labeling with peroxidase linked to a biotin-streptavidin complex using diaminobenzadine as the colorimetric substrate, then washing, dehydrating, and mounting in pertex.

Adjacent control sections were incubated with either a class-matched immunoglobulin raised to an irrelevant antigen or PBS in place of the primary antigen.

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Products Related to NB600-504-0.1mg

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)
NB600-504AF647-0.1ml	Aggrecan Antibody (BC-3) [Alexa Fluor® 647]

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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