abcam

Product datasheet

Anti-Collagen I antibody [EPR24331-53] - BSA and Azide free ab279711





* ★ ★ ★ ★ ★ 1 Abreviews 1 References 11 Images

Overview

Product name Anti-Collagen I antibody [EPR24331-53] - BSA and Azide free

Description Rabbit monoclonal [EPR24331-53] to Collagen I-BSA and Azide free

Host species Rabbit

Tested applications Suitable for: IHC-P, IHC-Fr, ICC/IF, Flow Cyt (Intra), IP, WB

Species reactivity Reacts with: Mouse. Rat

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: NIH/3T3 whole cell lysate; Mouse skin and Rat skin tissue lysates. IHC-P: Mouse skin,

stomach and pancreatic cancer tissue; Rat skin tissue. IHC-Fr: Rat skin; Mouse skin tissue. Flow

Cyt (intra): NIH/3T3 cells. ICC/IF: NIH/3T3 cells. IP: Mouse skin tissue lysate.

General notes ab279711 is the carrier-free version of ab270993.

> Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cellbased assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar[®] is a trademark of Fluidigm Canada Inc.

This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

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Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C.

Storage buffer Constituent: 100% PBS

Carrier free Yes

Purity Protein A purified

ClonalityMonoclonalClone numberEPR24331-53

Isotype IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab279711 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IHC-Fr	★★★☆☆ (1)	Use at an assay dependent concentration. Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20)
ICC/IF		Use at an assay dependent concentration.
Flow Cyt (Intra)		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Predicted molecular weight: 139 kDa.

Target

Function Type I collagen is a member of group I collagen (fibrillar forming collagen).

Tissue specificity Forms the fibrils of tendon, ligaments and bones. In bones the fibrils are mineralized with calcium

hydroxyapatite.

Involvement in disease

Defects in COL1A1 are the cause of Caffey disease (CAFFD) [MIM:114000]; also known as infantile cortical hyperostosis. Caffey disease is characterized by an infantile episode of massive subperiosteal new bone formation that typically involves the diaphyses of the long bones, mandible, and clavicles. The involved bones may also appear inflamed, with painful swelling and systemic fever often accompanying the illness. The bone changes usually begin before 5 months

of age and resolve before 2 years of age.

Defects in COL1A1 are a cause of Ehlers-Danlos syndrome type 1 (EDS1) [MIM:130000]; also known as Ehlers-Danlos syndrome gravis. EDS is a connective tissue disorder characterized by hyperextensible skin, atrophic cutaneous scars due to tissue fragility and joint hyperlaxity. EDS1 is the severe form of classic Ehlers-Danlos syndrome.

Defects in COL1A1 are the cause of Ehlers-Danlos syndrome type 7A (EDS7A) [MIM:130060]; also known as autosomal dominant Ehlers-Danlos syndrome type VII. EDS is a connective tissue disorder characterized by hyperextensible skin, atrophic cutaneous scars due to tissue fragility and joint hyperlaxity. EDS7A is marked by bilateral congenital hip dislocation, hyperlaxity of the joints, and recurrent partial dislocations.

Defects in COL1A1 are a cause of osteogenesis imperfecta type 1 (OI1) [MIM:166200]. A dominantly inherited connective tissue disorder characterized by bone fragility and blue sclerae. Osteogenesis imperfecta type 1 is non-deforming with normal height or mild short stature, and no dentinogenesis imperfecta.

Defects in COL1A1 are a cause of osteogenesis imperfecta type 2A (Ol2A) [MIM:166210]; also known as osteogenesis imperfecta congenita. A connective tissue disorder characterized by bone fragility, with many perinatal fractures, severe bowing of long bones, undermineralization, and death in the perinatal period due to respiratory insufficiency.

Defects in COL1A1 are a cause of osteogenesis imperfecta type 3 (Ol3) [MIM:259420]. A connective tissue disorder characterized by progressively deforming bones, very short stature, a triangular face, severe scoliosis, grayish sclera, and dentinogenesis imperfecta.

Defects in COL1A1 are a cause of osteogenesis imperfecta type 4 (OI4) [MIM:166220]; also known as osteogenesis imperfecta with normal sclerae. A connective tissue disorder characterized by moderately short stature, mild to moderate scoliosis, grayish or white sclera and dentinogenesis imperfecta.

Genetic variations in COL1A1 are a cause of susceptibility to osteoporosis (OSTEOP) [MIM:166710]; also known as involutional or senile osteoporosis or postmenopausal osteoporosis. Osteoporosis is characterized by reduced bone mass, disruption of bone microarchitecture without alteration in the composition of bone. Osteoporotic bones are more at risk of fracture.

Note=A chromosomal aberration involving COL1A1 is found in dermatofibrosarcoma protuberans. Translocation t(17;22)(q22;q13) with PDGF.

Sequence similarities

Belongs to the fibrillar collagen family.

Contains 1 fibrillar collagen NC1 domain.

Contains 1 VWFC domain.

Post-translational modifications

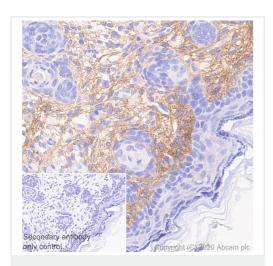
Proline residues at the third position of the tripeptide repeating unit (G-X-Y) are hydroxylated in some or all of the chains. Proline residues at the second position of the tripeptide repeating unit (G-X-Y) are hydroxylated in some of the chains.

O-linked glycan consists of a Glc-Gal disaccharide bound to the oxygen atom of a post-translationally added hydroxyl group.

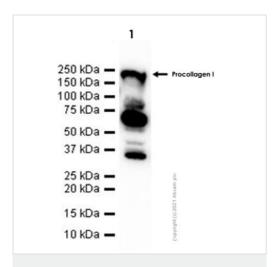
Cellular localization

Secreted > extracellular space > extracellular matrix.

Images



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - (ab279711)



Western blot - Anti-Collagen I antibody [EPR24331-53] - BSA and Azide free (ab279711)

This data was developed using <u>ab270993</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Mouse skin tissue labeling Collagen I with <u>ab270993</u> at 1/500 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining in connective tissues of mouse skin. The section was incubated with <u>ab270993</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) was used.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

Anti-Collagen I antibody [EPR24331-53] (ab270993) at 1/1000 dilution + NIH/3T3 (mouse embryonic fibroblast), whole cell lysate at 20 μg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 139 kDa **Observed band size:** 220 kDa

Exposure time: 20 seconds

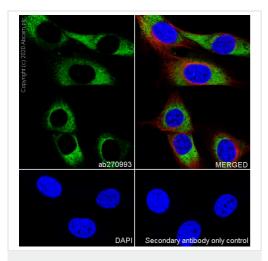
This data was developed using <u>ab270993</u>, the same antibody clone in a different buffer formulation.

Blocking buffer and concentration: 5% NFDM/TBST

Diluting buffer and concentration: 5% NFDM/TBST

We are unsure how to define these extra bands below 100kDa.

The molecular weight observed is consistent with what has been described in the literature (PMID:23940311;PMID:29853175).

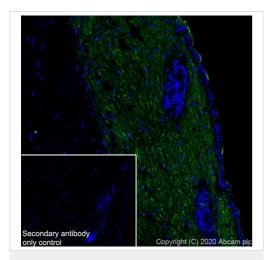


Immunocytochemistry/ Immunofluorescence - Anti-Collagen I antibody [EPR24331-53] - BSA and Azide free (ab279711)

This data was developed using <u>ab270993</u>, the same antibody clone in a different buffer formulation.

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized NIH/3T3 cells labelling Collagen I with ab270993 at 1/2000 dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) antibody at 1/1000 dilution (Green). Confocal image showing cytoplasmic staining in NIH/3T3 cells is observed. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is <u>ab150077</u> Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) at 1/1000 dilution.



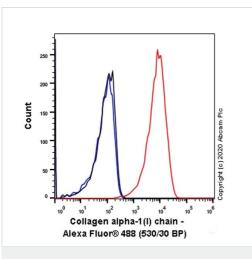
Immunohistochemistry (Frozen sections) - (ab279711)

This data was developed using <u>ab270993</u>, the same antibody clone in a different buffer formulation.

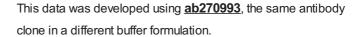
Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen Mouse skin tissue labeling Collagen I with ab270993 at 1/100 dilution followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution (Green). Positive staining on mouse skin is observed. The nuclear counterstain was DAPI (Blue).

Secondary antibody control: Secondary antibody is <u>ab150077</u> Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488)at 1/1000 dilution.

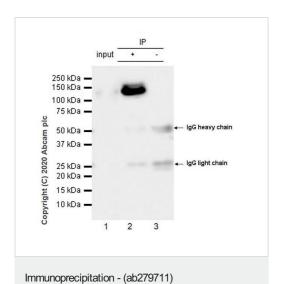
Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20).



Flow Cytometry (Intracellular) - Anti-Collagen I antibody [EPR24331-53] - BSA and Azide free (ab279711)



Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized NIH/3T3 (Mouse embryonic fibroblast) cells labelling Collagen I with ab270993 at 1/500 dilution (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.



This data was developed using <u>ab270993</u>, the same antibody clone in a different buffer formulation.

Collagen I was immunoprecipitated from 0.35 mg Mouse skin tissue lysate 10 ug with <u>ab270993</u> at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using <u>ab270993</u> at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP)(<u>ab131366</u>) was used at 1/5000 dilution.

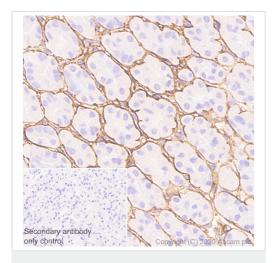
Lane 1: Mouse skin tissue lysate 10 ug

Lane 2: ab270993 IP in Mouse skin tissue lysate

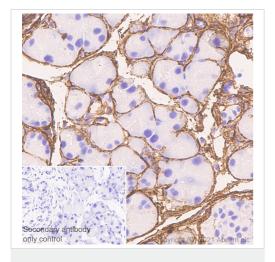
 $\mbox{\bf Lane 3: Rabbit monoclonal lgG } (\mbox{\bf \underline{ab172730}}) \mbox{ instead of } \mbox{\bf \underline{ab270993}} \\ \mbox{in Mouse skin tissue lysate} \\$

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 32 seconds



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - (ab279711)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - (ab279711)

This data was developed using <u>ab270993</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Mouse stomach tissue labeling Collagen I with <u>ab270993</u> at 1/500 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining in connective tissues of mouse stomach. The section was incubated with <u>ab270993</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) was used.

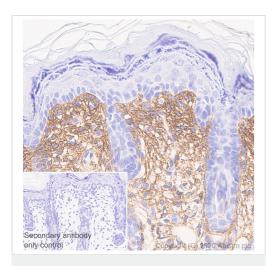
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

This data was developed using <u>ab270993</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Mouse pancreatic cancer tissue labeling Collagen I with <u>ab270993</u> at 1/500 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining in connective tissues of mouse pancreatic cancer. The section was incubated with <u>ab270993</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) was used.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - (ab279711)

1 2 250 kDa-250 kDa-150 kDa-150 kDa-100 kDa-100 kDa-75 kDa-75 kDa = Copyright (C) 2020 Abcam plc 50 kDa-50 kDa-37 kDa-37 kDa-25 kDa-25 kDa = 20 kDa = 20 kDa-15 kDa= 15 kDa-10 kDa-10 kDa-

Western blot - Anti-Collagen I antibody [EPR24331-53] - BSA and Azide free (ab279711)

This data was developed using <u>ab270993</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Rat skin tissue labeling Collagen I with <u>ab270993</u> at 1/500 dilution followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining in connective tissues of rat skin. The section was incubated with <u>ab270993</u> for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) was used.

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.

All lanes : Anti-Collagen I antibody [EPR24331-53] (**ab270993**) at 1/1000 dilution

Lane 1 : Mouse skin tissue lysate

Lane 2 : Rat skin tissue lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution (Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated)

Predicted band size: 139 kDa **Observed band size:** 138 kDa

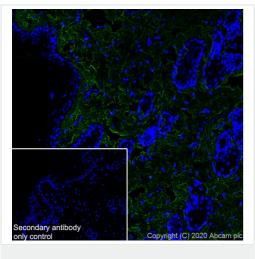
This data was developed using <u>ab270993</u>, the same antibody clone in a different buffer formulation.

Blocking and diluting buffer and concentration: 5% NFDM/TBST

The observed MW is consistent with what has been described in the literature (PMID: 27740527;PMID: 22278938; PMID: 26973392).

Exposure time: Lane 1: 3.25 seconds

Lane 2: 5.5 seconds



Immunohistochemistry (Frozen sections) - (ab279711)

This data was developed using <u>ab270993</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen Rat skin tissue labeling Collagen I with ab270993 at 1/500 dilution followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution (Green). Positive staining on rat skin is observed. The nuclear counterstain was DAPI (Blue).

Secondary antibody control: Secondary antibody is <u>ab150077</u> Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488)at 1/1000 dilution.

Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20).

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- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

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Product datasheet

Anti-Collagen III antibody [FH-7A] ab6310

★★★★★ 13 Abreviews 132 References 3 Images

Overview

Product name Anti-Collagen III antibody [FH-7A]

Description Mouse monoclonal [FH-7A] to Collagen III

Host species Mouse

Specificity ab6310 specifically recognizes collagen type III from human and rat origin. It does not recognize

collagen types I, II, IV, V, VI and X.

Tested applications Suitable for: IHC-Fr, IHC-P, WB, ELISA, Dot blot, Indirect ELISA, ICC/IF

Species reactivity Reacts with: Rat, Human

Immunogen Full length native protein (purified) (Human).

Positive control IHC-P: Rat skin sections. IHC-Fr: Rat skin sections.

General notes

Type III collagen, [a1(III)]3 ,is an approx. 300 kDa molecule, found predominantly in skin, blood

vessels, liver, placenta, tongue, and thymus. Collagen type III forms cofibrils with type I and/or V collagens in a number of tissues of mesenchymal origin, such as skin, tendon, ligaments, and bone. This collagen type is involved, directly or indirectly in several genetic diseases, including

Ehlers-Danlos type IV disease.

This product was changed from ascites to tissue culture supernatant on 17 May 2019. Please note that the dilutions may need to be adjusted accordingly. If you have any questions, please do

not hesitate to contact our scientific support team.

The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

1

Storage buffer Preservative: 0.097% Sodium azide

Constituent: Whole serum

Purity Tissue culture supernatant

Clonality Monoclonal

Clone number FH-7A

Isotype IgG1

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab6310 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-Fr		Use at an assay dependent concentration.
IHC-P	★★★★★ (10)	Use at an assay dependent concentration.
WB	★★★★★ (1)	Use at an assay dependent concentration.
ELISA	★★★★☆ (2)	Use at an assay dependent concentration.
Dot blot		Use at an assay dependent concentration.
Indirect ELISA		Use at an assay dependent concentration.
ICC/IF		Use at an assay dependent concentration. PubMed: 25136258

Target

Function

Involvement in disease

Collagen type III occurs in most soft connective tissues along with type I collagen.

Defects in COL3A1 are a cause of Ehlers-Danlos syndrome type 3 (EDS3) [MIM:130020]; also known as benign hypermobility syndrome. EDS is a connective tissue disorder characterized by hyperextensible skin, atrophic cutaneous scars due to tissue fragility and joint hyperlaxity. EDS3 is a form of Ehlers-Danlos syndrome characterized by marked joint hyperextensibility without skeletal deformity.

Defects in COL3A1 are the cause of Ehlers-Danlos syndrome type 4 (EDS4) [MIM:130050]. EDS is a connective tissue disorder characterized by hyperextensible skin, atrophic cutaneous scars due to tissue fragility and joint hyperlaxity. EDS4 is the most severe form of the disease. It is characterized by the joint and dermal manifestations as in other forms of the syndrome, characteristic facial features (acrogeria) in most patients, and by proneness to spontaneous rupture of bowel and large arteries. The vascular complications may affect all anatomical areas. Defects in COL3A1 are a cause of susceptibility to aortic aneurysm abdominal (AAA) [MIM:100070]. AAA is a common multifactorial disorder characterized by permanent dilation of the abdominal aorta, usually due to degenerative changes in the aortic wall. Histologically, AAA is characterized by signs of chronic inflammation, destructive remodeling of the extracellular matrix, and depletion of vascular smooth muscle cells.

Sequence similaritiesBelongs to the fibrillar collagen family.

Contains 1 fibrillar collagen NC1 domain.

Contains 1 VWFC domain.

Post-translational

modifications

Proline residues at the third position of the tripeptide repeating unit (G-X-Y) are hydroxylated in

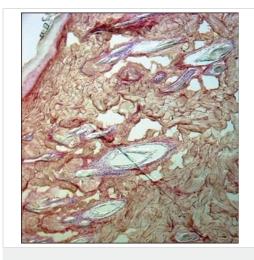
some or all of the chains.

O-linked glycan consists of a Glc-Gal disaccharide bound to the oxygen atom of a post-

translationally added hydroxyl group.

Cellular localization Secreted > extracellular space > extracellular matrix.

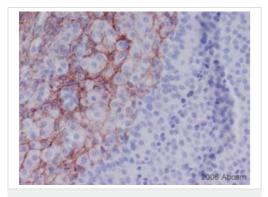
Images



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Collagen III antibody [FH-7A] (ab6310)

Staining of formalin-fixed, paraffin-embedded rat skin with 1:4,000 ab6310 using biotin/ExtrAvidin®-Peroxidase.

This image was generated using the ascites version of the product.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Collagen III antibody [FH-7A] (ab6310)

This image is courtesy of an Abreview submitted by Birgitta Weijdegard

ab6310 at 1/600 diltuion staining preovulatory follicle and whole ovary tissue sections by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections). Antigens were retrieved by boiling with an antigen unmasking solution for 20 min at 120°C in an autoclave and then cooled down in water for 5 minutes. The tissue sections were formaldehyde fixed and incubated with the antibody for 1 hour. An alkaline phosphatase conjugated antibody was used as the secondary. The image shows a section of whole preovulatory follicle. Staining for collagen type III is seen in the theca interna cell layer. No staining in the granulosa cells.

This image was generated using the ascites version of the product.



Immunohistochemistry (Frozen sections) - Anti-Collagen III antibody [FH-7A] (ab6310)

Staining of frozen rat skin sections with 1:8,000 ab6310 using biotin/ExtrAvidin[®]-Peroxidase.

This image was generated using the ascites version of the product.

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Product datasheet

Anti-Collagen IV antibody ab6586

Overview

Product name Anti-Collagen IV antibody

Description Rabbit polyclonal to Collagen IV

Host species Rabbit

Specificity ab6586 is designed to bind specifically to NATIVE collagen epitopes composed of multiple

subunit strands. Negligible cross-reactivity with Type I, II, III, V or VI collagens. Non-specific cross reaction of anti-collagen antibodies with other human serum proteins or non-collagen extracellular

matrix proteins is negligible.

Tested applications Suitable for: ELISA, IHC-Fr, WB, IHC-P, IP, ICC/IF, IHC-FrFI, IHC-FoFr

Species reactivity Reacts with: Mouse, Rat, Hamster, Cow, Dog, Human, Pig, Zebrafish, African green monkey,

Chinese hamster, Syrian hamster

Predicted to work with: Mammals 4

Immunogen Full length native protein (purified) corresponding to Collagen V. Collagen Type IV from human

and bovine placenta. The immunogen maintains the native conformation of the protein.

Positive control IHC-P: Human kidney and liver tissue.

General notes

There are other recombinant monoclonal options, such as **Recombinant Anti-Collagen IV**

antibody.

Abcam recommended secondaries - Goat Anti-Rabbit HRP (<u>ab205718</u>) and Goat Anti-Rabbit

Alexa Fluor[®] 488 (<u>ab150077</u>).

See other anti-rabbit secondary antibodies that can be used with this antibody.

The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

Properties

Form Liquid

1

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

Storage buffer Preservative: 0.01% Sodium azide

Constituents: 0.8766% Sodium chloride, 0.424% Potassium phosphate

Purity Immunogen affinity purified

Purification notes Immunoaffinity chromatography using immobilized antigens followed by extensive cross-

adsorption against other collagens, human serum proteins and non-collagen extracellular matrix

proteins to remove any unwanted specificities.

Primary antibody notes This antibody is well suited to detect extracellular matrix proteins in normal as well as disease

state tissues. Disruption of tissue organization is the hallmark of neoplasia. Malignant lesions can be distinguished from benign by examining the breakdown of basement membranes and loss of 3-dimensional architecture. Malignant cells are presumed to use matrix metalloproteases to degrade barriers created by the extracellular matrix which then allows metastasis to occur. Collagenases, stomelysins and gelatinases can collectively degrade all of the various

components of the extracellular matrix, including fibrillar and non-fibrillar collagens and basement

membrane glycoproteins.

Clonality Polyclonal

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab6586 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
ELISA		Use at an assay dependent concentration.
IHC-Fr	★★★★★ (21)	Use at an assay dependent concentration.
WB	**** (26)	Use at an assay dependent concentration. Predicted molecular weight: 161 kDa. This product is not recommended for use under denaturing conditions in WB, IP, and ELISA. We would suggest testing it under native conditions.
IHC-P	**** (35)	1/500. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
IP	★★★★★ (3)	Use at an assay dependent concentration.
ICC/IF	★★★★★ (23)	Use at an assay dependent concentration. PubMed: 19933193
IF		Use at an assay dependent concentration.
IHC-FrFI	**** (1)	Use at an assay dependent concentration.
IHC-FoFr	****(8)	Use at an assay dependent concentration.

Target

Function

Type IV collagen is the major structural component of glomerular basement membranes (GBM), forming a 'chicken-wire' meshwork together with laminins, proteoglycans and entactin/nidogen. Arresten, comprising the C-terminal NC1 domain, inhibits angiogenesis and tumor formation. The C-terminal half is found to possess the anti-angiogenic activity. Specifically inhibits endothelial cell proliferation, migration and tube formation. Inhibits expression of hypoxia-inducible factor 1alpha and ERK1/2 and p38 MAPK activation. Ligand for alpha1/beta1 integrin.

Tissue specificity

Highly expressed in placenta.

Involvement in disease

Defects in COL4A1 are a cause of brain small vessel disease with hemorrhage (BSVDH) [MIM:607595]. Brain small vessel diseases underlie 20 to 30 percent of ischemic strokes and a larger proportion of intracerebral hemorrhages. Inheritance is autosomal dominant. Defects in COL4A1 are the cause of hereditary angiopathy with nephropathy aneurysms and muscle cramps (HANAC) [MIM:611773]. The clinical renal manifestations include hematuria and bilateral large cysts. Histologic analysis revealed complex basement membrane defects in kidney and skin. The systemic angiopathy appears to affect both small vessels and large arteries. Defects in COL4A1 are a cause of porencephaly familial (PCEPH) [MIM:175780]. Porencephaly is a term used for any cavitation or cerebrospinal fluid-filled cyst in the brain. Porencephaly type 1 is usually unilateral and results from focal destructive lesions such as fetal vascular occlusion or birth trauma. Type 2, or schizencephalic porencephaly, is usually symmetric and represents a primary defect or arrest in the development of the cerebral ventricles.

Sequence similarities

Belongs to the type IV collagen family.

Contains 1 collagen IV NC1 (C-terminal non-collagenous) domain.

Domain

Alpha chains of type IV collagen have a non-collagenous domain (NC1) at their C-terminus, frequent interruptions of the G-X-Y repeats in the long central triple-helical domain (which may cause flexibility in the triple helix), and a short N-terminal triple-helical 7S domain.

Post-translational modifications

Lysines at the third position of the tripeptide repeating unit (G-X-Y) are hydroxylated in all cases and bind carbohydrates.

Prolines at the third position of the tripeptide repeating unit (G-X-Y) are hydroxylated in some or all of the chains.

Type IV collagens contain numerous cysteine residues which are involved in inter- and intramolecular disulfide bonding. 12 of these, located in the NC1 domain, are conserved in all known type IV collagens.

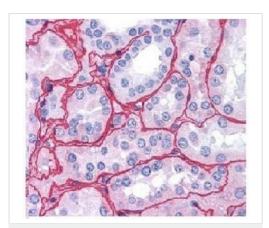
The trimeric structure of the NC1 domains is stabilized by covalent bonds between Lys and Met residues.

Proteolytic processing produces the C-terminal NC1 peptide, arresten.

Cellular localization

Secreted > extracellular space > extracellular matrix > basement membrane.

Images



Paraffin-embedded human kidney tissue stained for Collagen IV using ab6586 at 1/400 dilution in immunohistochemical analysis with strong staining observed in glomeruli.

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Collagen IV antibody (ab6586)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Collagen IV antibody (ab6586)

Paraffin-embedded human liver tissue stained for Collagen IV using ab6586 at 1/400 dilution in immunohistochemical analysis, strong staining was observed in the sinusoids.

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Product datasheet

Anti-Collagen V antibody ab7046

★★★★★ 10 Abreviews 50 References 3 Images

Overview

Product name Anti-Collagen V antibody

Description Rabbit polyclonal to Collagen V

Host species Rabbit

SpecificityNegligible cross-reactivity with Type I, II, III, IV or VI collagens. Non-specific cross reaction of anti-

collagen antibodies with other human serum proteins or non-collagen extracellular matrix proteins

is negligible.

Tested applications Suitable for: WB, Sandwich ELISA, IHC-P, IP, IHC-Fr, ICC

Species reactivity Reacts with: Mouse, Cow, Human

Immunogen Full length native protein (purified) corresponding to Collagen V aa 1-1745.

General notes Some class specific anti-collagens may be specific for three-dimensional epitopes which may

result in diminished reactivity with denatured collagen or formalin-fixed, paraffin embedded

tissues.

The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

Storage buffer Preservative: 0.01% Sodium azide

Constituents: 4.8% Sodium borate, 0.15% EDTA, 0.44% Sodium chloride

Purity Immunogen affinity purified

Purification notes Immunoaffinity chromatography using immobilized antigens followed by extensive cross-

adsorption against other collagens, human serum proteins and non-collagen extracellular matrix

proteins to remove any unwanted specificities.

1

Clonality Polyclonal

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab7046 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		1/5000 - 1/10000. Detects a band of approximately 180 kDa (predicted molecular weight: 180 kDa). Native conditions are recommended.
Sandwich ELISA		Use at an assay dependent concentration. Can be used with a suitable biotinylated detection antibody such as Rabbit polyclonal to Collagen V (Biotin) (ab6582).
IHC-P	**** <u>(6)</u>	1/50 - 1/200.
IP		Use at an assay dependent concentration.
IHC-Fr	★★★★ <u>(2)</u>	Use at an assay dependent concentration.
ICC		Use at an assay dependent concentration. PubMed: 18385800

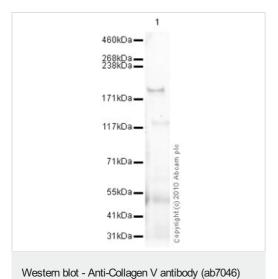
Function	Type V collagen is a member of group I collagen (fibrillar forming collagen). It is a minor connective tissue component of nearly ubiquitous distribution. Type V collagen binds to DNA, heparan sulfate, thrombospondin, heparin, and insulin.
Involvement in disease	Ehlers-Danlos syndrome 1 Ehlers-Danlos syndrome 2
Sequence similarities	Belongs to the fibrillar collagen family. Contains 1 fibrillar collagen NC1 domain. Contains 1 laminin G-like domain.
Domain	The C-terminal propeptide, also known as COLFI domain, have crucial roles in tissue growth and repair by controlling both the intracellular assembly of procollagen molecules and the extracellular assembly of collagen fibrils. It binds a calcium ion which is essential for its function.
Post-translational modifications	Prolines at the third position of the tripeptide repeating unit (G-X-Y) are hydroxylated in some or all of the chains. Sulfated on 40% of tyrosines.

Secreted > extracellular space > extracellular matrix.

Images

Cellular localization

Target



Secondary

Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (ab97080) at 1/5000 dilution

tissue lysate - total protein (ab29816) at 10 µg

Anti-Collagen V antibody (ab7046) at 1 µg/ml + Human pancreas

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 180 kDa **Observed band size:** 180 kDa

Additional bands at: 122 kDa, 54 kDa. We are unsure as to the

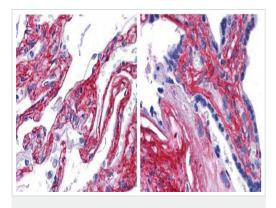
identity of these extra bands.

Exposure time: 8 minutes

ab7046 staining Collagen V in Human pancreas tissue by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded tissue sections). The sections were fixed in formaldehyde and subjected to heat-mediated antigen retrieval in citrate pH 6 prior to blocking with 0.25% casein for 5 minutes at 25°C. The primary antibody was diluted 1/75 in Tris-HCL and incubated with the sample for 30 minutes at 25°C. An HRP polymer-conjugated goat anti-rabbit antibody was used as the secondary antibody.

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Collagen V antibody (ab7046)

This image is courtesy of an anonymous Abreview.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Collagen V antibody (ab7046)

Immunohistochemical analysis of formalin-fixed paraffinembedded human lung (left) and placenta (right) sections labelling collagen V with ab7046 at a dilution of 1/200 for 45 minutes at room temperature. An antigen retrival step was performed with 0.01 M sodium citrate buffer pH 6.0 at 100°C for 20 mins.

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Product datasheet

Goat Anti-Rabbit IgG H&L (HRP) ab6721

**** 12 Abreviews 4973 References 6 Images

Overview

Product name Goat Anti-Rabbit lgG H&L (HRP)

Host species Goat
Target species Rabbit

Tested applications Suitable for: IHC-P, WB, ELISA, Immunomicroscopy, Dot blot, ICC, IHC-Fr

Immunogen Rabbit IgG, whole molecule

Conjugation HRP

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle. Please see notes section. Store In the Dark.

Storage buffer Preservative: 0.01% Gentamicin sulphate

Constituents: 0.42% Potassium phosphate, 0.88% Sodium chloride

Purity Immunogen affinity purified

Purification notes This product was prepared from monospecific antiserum by immunoaffinity chromatography using

Rabbit IgG coupled to agarose beads.

Conjugation notes Horseradish Peroxidase (HRP)

Clonality Polyclonal

Isotype IgG

General notes HRP conjugated anti-rabbit secondary antibody optimized for western blot and

immunohistochemistry. Some customers reported seeing brown precipitates in the vials. The brown precipitates are very common with HRP conjugated antibodies; we suggest vortexing the vial and using this antibody as normal. Our customer's feedback says the antibody worked great. If in case the antibody fails to give results then please contact our Scientific Support team for

assistance.

Centrifuge product if not completely clear after standing at room temperature. Dilute only prior to

immediate use.

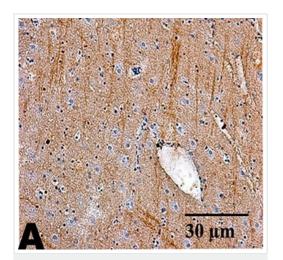
The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab6721 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-P	★★★★★ (2)	1/1000.
WB	★★★★★ (10)	1/2000 - 1/20000. Suggested working dilution of 1/3000 (see PMID: 17222046). In addition, found to work at 1/20000 (see PMID: 16936283). Working dilutions are highlighted in the table below. Please note that the antibody can be diluted to 1:48,000 to 1:207,000 in many instances.
ELISA		1/120000.
Immunomicroscopy		Use at an assay dependent concentration.
Dot blot		Use at an assay dependent concentration.
ICC		1/1000 - 1/5000.
IHC-Fr		1/1000.

Images



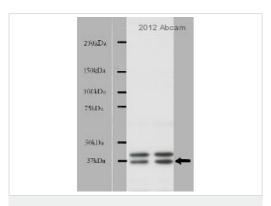
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Goat Anti-Rabbit IgG H&L (HRP) (ab6721)

Farah et al PLoS One. 2018 Feb 2;13(2):e0191526. doi: 10.1371/journal.pone.0191526. eCollection 2018. Fig 3. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Anti-phospho-tau immunostaining in Dryocopus lineatus.

Tau-positivity in the midbrain (**Panel A (shown)** and B) and the corpus callosum (C) of the *Dryocopus lineatus* brain. The axonal tract staining demonstrates a thread-like pattern, similar to that seen with Gallyas sliver staining. Occasional intracellular tau-accumulations were identified within neurons (D).

For full method, please see paper.



Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab6721)

This image is courtesy of an Abreview submitted by Maryna Polyakova.

All lanes: Anti-NEK7 antibody (ab80948) at 1/2000 dilution

All lanes: Mouse brain tissue cytoplasmic lysate

Lysates/proteins at 10 µg per lane.

Secondary

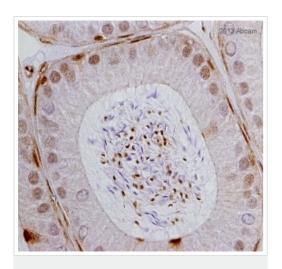
All lanes : Goat Anti-Rabbit lgG H&L (HRP) (ab6721) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Exposure time: 10 seconds

Blocked with 5% non-fat milk for 1 hour at 18°C

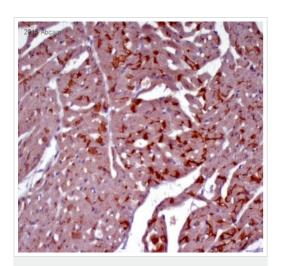


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Goat Anti-Rabbit IgG H&L (HRP) (ab6721)

This image is courtesy of an anonymous Abreview.

<u>ab3580</u> staining glucocorticoid receptor in mouse epididymis tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections).

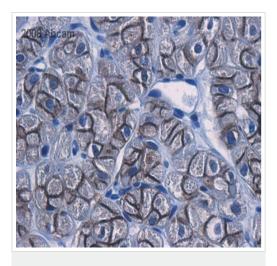
Tissue was fixed with Bouin's solution and blocked with 1.5% serum for 30 minutes at 25°C; antigen retrieval was by heat mediation in a citrate buffer. Samples were incubated with primary antibody (1/1000) for 14 hours at 4°C. An HRP-conjugated goat anti-rabbit IgG H&L (ab6721) (1/200) was used as the secondary antibody.



Immunohistochemical analysis of PFA-fixed paraffin-embedded rat cardiac tissue sections, labeling Conexin 43 with <u>ab117843</u> at a dilution of 1/500 incubated for 12 hours at 4°C in 1% BSA in TBS. Antigen retrival was via Tris-EDTA pH 9.0 (heat mediated). Blocking was 3% BSA incubated for 1 hour at 37°C. The secondary was ab6721 at 1/500.

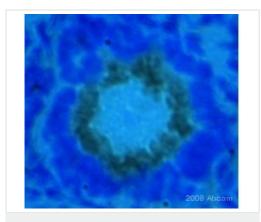
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Goat Anti-Rabbit IgG H&L (HRP) (ab6721)

Image is courtesy of an anonymous AbReview.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Goat Anti-Rabbit IgG H&L (HRP) (ab6721)

ab6721 was used at dilution 1/100 with the primary antibody **ab11370** in IHC-P. See the review on **ab11370**.



ab6721 was used at dilution 1/100 with the primary antibody ab35604 in IHC-Fr. See the review on ab35604.

Immunohistochemistry (Frozen sections) - Goat Anti-Rabbit IgG H&L (HRP) (ab6721)

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Product datasheet

Goat Anti-Mouse IgG H&L (HRP) ab205719

*** * * 4 Abreviews 1026 References 8 Images

Overview

Product nameGoat Anti-Mouse IgG H&L (HRP)

Host species Goat

Target species Mouse

Specificity The antibody used for conjugation reacts with mouse immunoglobulins of all classes. Cross-

reactions as determined by ELISA for the unconjugated antibody (ab182017): Chicken lgY, less

than 2%. Human lgG, less than 6%. Rabbit lgG, less than 7%. Rat lgG, less than 47%.

Tested applications Suitable for: WB, IP, ELISA, IHC-P

Immunogen The details of the immunogen for this antibody are not available.

Conjugation HRP

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle. Store In the Dark.

Storage buffer pH: 7.40

Preservative: 0.1% Proclin 300 Solution

Constituents: PBS, 1% BSA, 30% Glycerol (glycerin, glycerine)

Purity Immunogen affinity purified

Purification notes This antibody was isolated by affinity chromatography using antigen coupled to agarose beads

and conjugated to Horse Radish Peroxidase (HRP).

Clonality Polyclonal

Isotype IgG

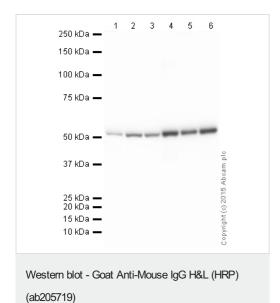
Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab205719 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB	★★★★★ (4)	1/2000 - 1/20000.
IP		Use at an assay dependent concentration.
ELISA		Use at an assay dependent concentration.
IHC-P		1/2000 - 1/20000.

Images



All lanes : Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) at 1 μ g/ml

Lane 1 : Liver (Human) Tissue Lysate
Lane 2 : Liver (Mouse) Tissue Lysate

Lane 3: Liver (Rat) Tissue Lysate

Lane 4 : HeLa (Human epithelial carcinoma cell line) Whole Cell

Lysate

Lane 5: NIH 3T3 (Mouse embryonic fibroblast cell line) Whole Cell

Lysate

Lane 6: PC12 (Rat adrenal pheochromocytoma cell line) Whole

Cell Lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Mouse IgG H&L (HRP) (ab205719) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Observed band size: 52 kDa

Exposure time: 5 seconds

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes.

The membrane was then blocked for an hour using 2% Bovine Serum Albumin before being incubated with <u>ab7291</u> overnight at 4°C. Antibody binding was detected using ab205719, and visualised using ECL development solution <u>ab133406</u>.

Secondary only control

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Goat Anti-Mouse IgG H&L (HRP) (ab205719)

IHC image of alpha tubulin staining in a section of formalin-fixed paraffin-embedded normal human colon tissue*. The section was pre-treated using pressure cooker heat mediated antigen retrieval with sodium citrate buffer (pH6) for 30mins, and incubated overnight at +4°C with ab7291 at 1/1000 dilution. An HRP-conjugated secondary (Ab205719, 1/10000 dilution) was used to detect the primary for 1hr at room temperature. DAB was used as the chromogen (ab103723), diluted 1/100 and incubated for 10min at room temperature. The section was counterstained with haematoxylin and mounted with DPX. The inset negative control image is taken from an identical assay without primary antibody.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre

ab205719 Competitor

250 kDa — 1 2 3 250 kDa — 1 2 3 150 kDa — 150

Western blot - Goat Anti-Mouse IgG H&L (HRP) (ab205719)

All lanes : Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) at 1 µg/ml

Lane 1: Liver (Human) Tissue Lysate

Lane 2 : Liver (Mouse) Tissue Lysate

Lane 3: Liver (Rat) Tissue Lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : ab205719 (Left Image) at 1/5000 and a competitor secondary (Right Image) at 1/5000. Notice the decreased signal of the competitor product.

Performed under reducing conditions.

Observed band size: 52 kDa

Exposure time: 5 seconds

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 2% Bovine Serum Albumin before being incubated with ab7291 overnight at 4°C. Antibody binding was detected using ab205719 (Left Image) and a competitor secondary (Right Image), and visualised using ECL development solution ab133406.

Secondary-only control — Gobyright (s) 2016; Ussait Sic

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Goat Anti-Mouse IgG H&L (HRP) (ab205719)

IHC image of histone H4 staining in a section of formalin-fixed paraffin-embedded normal human colon tissue*. The section was pre-treated using pressure cooker heat mediated antigen retrieval with sodium citrate buffer (pH6) for 30mins, and incubated overnight at +4°C with ab31830 at 1/1000 dilution. An HRP-conjugated secondary (Ab205719, 1/10000 dilution) was used to detect the primary for 1hr at room temperature. DAB was used as the chromogen (ab103723), diluted 1/100 and incubated for 10min at room temperature. The section was counterstained with haematoxylin and mounted with DPX. The inset negative control image is taken from an identical assay without primary antibody.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre

ab205719 Competitor 1 2 250 kDa -250 kDa = 150 kDa -150 kDa -100 kDa -100 kDa --75 kDa -75 kDa 🕳 50 kDa 🕳 50 kDa -37 kDa -37 kDa -25 kDa — 20 kDa — 20 kDa -15 kDa 🕳 15 kDa --10 kDa -10 kDa -

Western blot - Goat Anti-Mouse IgG H&L (HRP) (ab205719)

All lanes: No Primary Antibody

Lane 1 : Liver (Human) Tissue Lysate
Lane 2 : Liver (Mouse) Tissue Lysate

Lysates/proteins at 10 µg per lane.

Lane 3: Liver (Rat) Tissue Lysate

Secondary

All lanes : ab205719 (Left Image) 1/2000 and a competitor secondary (Right Image) 1/2000. Notice the increased background of the competitor product.

Performed under reducing conditions.

Exposure time: 10 seconds

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was incubated overnight with 2% Bovine Serum Albumin at 4°C. Any non-specific background binding was assessed by incubating the membrane with ab205719 (Left Image) and a competitor secondary (Right Image), and visualised using ECL development solution **ab133406**.

1 2 3 4 5 6
250 kDa —
150 kDa —
100 kDa —
75 kDa —
37 kDa —
25 kDa —
20 kDa —
15 kDa —
15 kDa —
10 kDa —
15 kDa —
10 kDa —

Western blot - Goat Anti-Mouse IgG H&L (HRP) (ab205719)

All lanes : Anti-beta Actin antibody [mAbcam 8226] - Loading Control (ab8226) at 1 µg/ml

Lane 1: Liver (Human) Tissue Lysate

Lane 2: Liver (Mouse) Tissue Lysate

Lane 3: Liver (Rat) Tissue Lysate

Lane 4 : HeLa (Human epithelial carcinoma cell line) Whole Cell

Lysate

Lane 5: NIH 3T3 (Mouse embryonic fibroblast cell line) Whole Cell

Lysate

Lane 6: PC12 (Rat adrenal pheochromocytoma cell line) Whole

Cell Lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: Goat Anti-Mouse IgG H&L (HRP) (ab205719) at 1/5000

dilution

Developed using the ECL technique.

Performed under reducing conditions.

Observed band size: 42 kDa

Exposure time: 10 seconds

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes.

The membrane was then blocked for an hour using 2% Bovine Serum Albumin before being incubated with **ab8226** overnight at 4°C. Antibody binding was detected using ab205719, and visualised using ECL development solution **ab133406**.

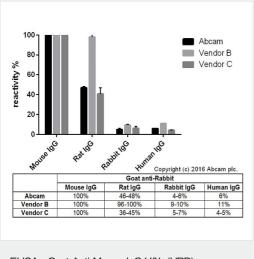
100 80 60 40 20 60 Rathing Rathing Rathing Chicken Let Chicken Let

ELISA - Goat Anti-Mouse IgG H&L (HRP) (ab205719)

Cross-reactivity of the polyclonal secondary antibody <u>ab182017</u> was tested using a sandwich ELISA approach. The wells were coated with the indicated IgG standards at 1 μ g/ml (50 μ l/well) and incubated overnight at 4°C, followed by a 5% BSA blocking step for 2h at RT. <u>ab182017</u> was then added starting at 1 μ g/ml and gradually diluted 1/4 (50 μ l/well), followed by incubation for 2h. For the detection Donkey anti-Goat IgG H&L (HRP) (<u>ab6885</u>) was used at 1/10,000 dilution (50 μ l/well), followed by incubation for 1h at RT.

Fot the batch tested, <u>ab182017</u> showed a cross-reactivity below 2% towards Chicken IgY, 6% towards Human IgG, 7% towards Rabbit IgG and 47% towards Rat IgG.

This data was developed using the unconjugated antibody (ab182017).



ELISA - Goat Anti-Mouse IgG H&L (HRP) (ab205719)

Cross-reactivity of Goat anti-Mouse IgG H&L (ab182017) and Goat anti-Mouse IgG H&L obtained from two different vendors was tested using a sandwich ELISA approach. The wells were coated with the indicated IgG standards (Rabbit, Human, Mouse and Rat) at 1 μ g/ml (50 μ l/well) and incubated overnight at 4°C, followed by a 5% BSA blocking step for 2h at RT. Secondary antibodies were then added starting at 1 μ g/ml and gradually diluted 1/4 (50 μ l/well), followed by incubation for 2h. For the detection Donkey anti-Goat IgG H&L (HRP) (ab6885) was used at 1/10,000 dilution (50 μ l/well), followed by incubation for 1h at RT. This data is from a representative dilution.

This data was developed using the unconjugated antibody (ab182017).

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Product datasheet

Goat Anti-Rabbit IgG H&L (FITC) ab6717

**** 2 Abreviews 501 References 4 Images

Overview

Product nameGoat Anti-Rabbit IgG H&L (FITC)

Host species Goat

Target species Rabbit

Tested applications Suitable for: IHC-FoFr, Immunomicroscopy, Flow Cyt, IHC-P, IHC-Fr, ICC/IF, ELISA

Immunogen Rabbit IgG whole molecule

Conjugation FITC. Ex: 493nm, Em: 528nm

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C. Store In the Dark.

Storage buffer Preservative: 0.01% Sodium azide

Constituents: 0.42% Potassium phosphate, 0.87% Sodium chloride, 1% BSA

Purity Affinity purified

Purification notes This product was prepared from monospecific antiserum by immunoaffinity chromatography using

Rabbit IgG coupled to agarose beads followed by solid phase adsorption(s) to remove any

unwanted reactivities.

Conjugation notes Fluorescein isothiocyanate (FΠC) (Molecular Weight 390 daltons) Absorption Wavelength: 495

 $nm\ Emission\ Wavelength:\ 528\ nm\ Fluorochrome/Protein\ Ratio:\ 2.7\ moles\ FITC\ per\ mole\ of\ Goat$

ΙgG

Clonality Polyclonal

Isotype IgG

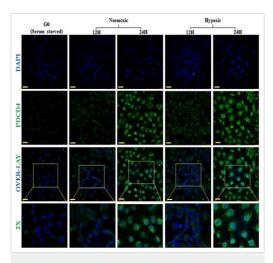
Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab6717 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-FoFr		Use at an assay dependent concentration. PubMed: 24216136
Immunomicroscopy		Use at an assay dependent concentration.
Flow Cyt		1/500 - 1/2500.
IHC-P		Use at an assay dependent concentration.
IHC-Fr		Use at an assay dependent concentration.
ICC/IF	★★★★ (1)	1/1000 - 1/5000.
ELISA		1/10000 - 1/50000.

Images



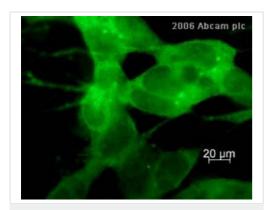
Immunocytochemistry/ Immunofluorescence - Goat Anti-Rabbit IgG H&L (FITC) (ab6717)

Kumar S et al. The role of tumour suppressor PDCD4 in beta cell death in hypoxia. PLoS One 12:e0181235 (2017). Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Subcellular localisation of PDCD4 was assessed by immunocytochemical staining. ARIP cells were seeded at 3×10^5 cells/well on sterile cover slips in six well plates and incubated overnight under standard culture conditions.

Following specific experimental conditions, media was removed and cells were washed with PBS and fixed with 3.7% formalin. Cells were then permeabilised with 0.1% triton X-100 in PBS followed by blocking in blocking buffer (10% Goat serum; 2% BSA; 0.2% Tween 20; 0.7% Glycerol in PBS) for 1 hour. Cells were incubated overnight with <u>ab51495</u> at 1/100 dilution followed by ab6717 (green) at 1/80 dilution. The nuclear counterstain is DAPI (Blue).

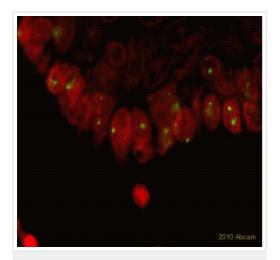
Results are representative of three separate experiments and images were representative of six separate fields. PDCD4 localized and expressing At G0: Cytoplasmic and very low expression; At N12: Cytoplasmic and low expression; At N24: very high cytoplasmic and low nuclear expression; At H12: Cytoplasmic expression and At H24 very high cytoplasmic and low nuclear expression. Over-all PDCD4 was highly expressed in the cytoplasm, with very low expression under serum starved conditions



Immunocytochemistry/ Immunofluorescence - Goat
Anti-Rabbit IgG H&L (FITC) (ab6717)

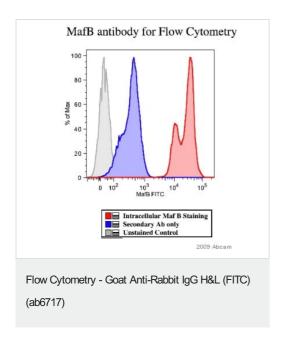
This image is courtesy of an anonymous Abreview

ab11320 (Rabbit polyclonal to gamma Tubulin - Centrosome Marker) at 1/400 dilution staining human neuroblastoma cells by ICC/IF. The cells were fixed with paraformaldehyde and blocked with serum and then incubated with the primary antibody for 16 hours. ab6717 was used as the secondary.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Goat Anti-Rabbit IgG H&L (FITC) (ab6717)

Ab6717 was used at dilution 1/200 with the primary antibody **ab36823** in IHC-P. See the review on **ab36823**.



ab6717 was used with the primary antibody <u>ab66506</u> in Flow Cyt. See Abreview on <u>ab66506</u>.

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Product datasheet

Goat Anti-Mouse IgG H&L (FITC) ab6785

* ★ ★ ★ ★ 1 Abreviews 473 References 4 Images

Overview

Product name Goat Anti-Mouse IgG H&L (FITC)

Host species Goat

Target species Mouse

Tested applications Suitable for: IHC (PFA fixed), Immunomicroscopy, Flow Cyt, IHC-P, IHC-Fr, ICC/IF, ELISA, WB

Immunogen mouse whole molecule

Conjugation FITC. Ex: 493nm, Em: 528nm

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C.

Storage buffer Preservative: 0.01% Sodium azide

Constituents: 0.42% Potassium phosphate, 0.87% Sodium chloride, 1% BSA

Purity Affinity purified

Purification notes This product was prepared from monospecific antiserum by immunoaffinity chromatography using

mouse IgG coupled to agarose beads followed by solid phase adsorption(s) to remove any

unwanted reactivities.

Conjugation notes Diaminotriazinylaminofluorescein (DTAF) (Molecular Weight 530 daltons) Absorption

Wavelength: 495 nm Emission Wavelength: 528 nm Fluorochrome/Protein Ratio: 3.3 moles FITC

per mole of Goat lgG

Clonality Polyclonal

Isotype IgG

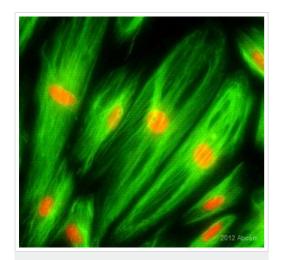
Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab6785 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes	
IHC (PFA fixed)		1/1000.	
Immunomicroscopy		Use at an assay dependent concentration.	
Flow Cyt		1/500 - 1/2500. PubMed: 17989178	
IHC-P		Use at an assay dependent concentration.	
IHC-Fr		Use at an assay dependent concentration. PubMed: 15716365For PFA fixed tissue see PMID 15716365.	
ICC/IF	★★★★★ (1)	1/1000 - 1/5000. (PMID16006199)	
ELISA		1/10000 - 1/50000.	
WB		1/10000. PubMed: 17217624	

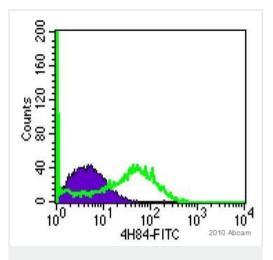
Images



Immunocytochemistry/ Immunofluorescence - Goat Anti-Mouse IgG H&L (FITC) (ab6785)

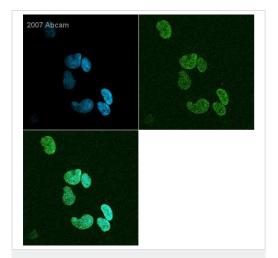
This image is courtesy of an Abreview submitted by J $\mbox{\sc Chai.}$

ab8978 staining vimentin in human colon fibroblasts by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with methanol, permeabilized with 0.1% Triton X-100 and blocked with Dako serum free protein blocker for 20 minutes at 28°C. Samples were incubated with primary antibody (1/100 in Dako antibody diluent) for 2 hours at 28°C. A FITC-conjugated goat antimouse IgG H&L (ab6785) (1/800) was used as the secondary antibody.



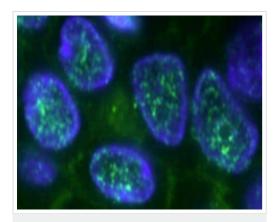
Ab6785 was used at dilution 1/100 with the primary antibody **ab52455** in Flow Cyt. See the review on **ab52455**.

Flow Cytometry - Goat Anti-Mouse IgG H&L (FITC) (ab6785)



Immunocytochemistry/ Immunofluorescence - Goat Anti-Mouse IgG H&L (FITC) (ab6785)

Ab6785 was used at dilution 1/2000 with the primary antibody **ab1220** in ICC/IF. See the review on **ab1220**.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Goat Anti-Mouse IgG H&L (FITC) (ab6785)

Ab6785 was used at dilution 1/300 with the primary antibody **ab6002** in IHC-P. See the review on **ab6002**.

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Product datasheet

Goat Anti-Mouse IgG H&L (TRITC) ab6786

50 References 1 Image

Overview

Product name Goat Anti-Mouse IgG H&L (TRITC)

Host species Goat
Target species Mouse

Tested applications Suitable for: Immunomicroscopy, Flow Cyt, IHC-P, IHC-Fr, ICC/IF, ELISA

Immunogen Mouse IgG whole molecule

Conjugation TRITC. Ex: 547nm, Em: 572nm

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -

80°C. Avoid freeze / thaw cycle.

Storage buffer Preservative: 0.01% Sodium azide

Constituents: 0.878% Sodium chloride, 1% BSA, 0.424% Potassium phosphate

Purity Affinity purified

Purification notes This product was prepared from monospecific antiserum by immunoaffinity chromatography using

mouse IgG coupled to agarose beads followed by solid phase adsorption(s) to remove any

unwanted reactivities.

Conjugation notes Tetramethylrhodamine isothiocyanate (TRITC) (Molecular Weight 444 daltons) Absorption

Wavelength: 550 nm Emission Wavelength: 570 nm Fluorochrome/Protein Ratio: 2.8 moles

TRITC per mole of Rabbit IgG

Clonality Polyclonal

Isotype IgG

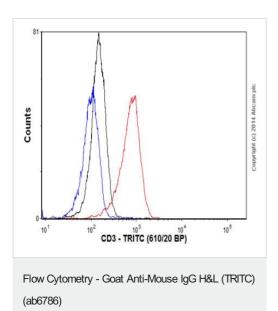
Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab6786 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Immunomicroscopy		Use at an assay dependent concentration.
Flow Cyt		1/2000 - 1/4000.
IHC-P		Use at an assay dependent concentration.
IHC-Fr		Use at an assay dependent concentration.
ICC/IF		1/1000 - 1/5000. (PubMed:15659422)
ELISA		1/10000 - 1/50000.

Images



Overlay histogram showing Jurkat cells stained with <u>ab8090</u> (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (<u>ab8090</u>, 0.1 μ g/1x10⁶ cells) for 30 min at 22°C. The secondary antibody Goat anti-mouse lgG H&L (TRITC) (ab6786) was used at 1/4000 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse lgG2a [ICIGG2A] (<u>ab91361</u>, 0.1 μ g/1x10⁶ cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 561nm laser and 610/20 bandpass filter.

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Product datasheet

Ethidium homodimer, Fluorimetric detection reagent ab145323

3 References 1 Image

Overview

Product name Ethidium homodimer, Fluorimetric detection reagent

Description Reagent for the fluorimetric detection of nucleic acids.

Purity = 90%

CAS Number 61926-22-5

Chemical structure

Properties

Excitation 528nm **Emission** 617nm

Molecular weight 856.75

Molecular formula $C_{46}H_{50}CI_4N_8$

Storage instructions Store at -20°C. Store under desiccating conditions. The product can be stored for up to 12

months.

Solubility overview Soluble in water and in DMSO

Handling Wherever possible, you should prepare and use solutions on the same day. However, if you need

1

to make up stock solutions in advance, we recommend that you store the solution as aliquots in tightly sealed vials at -20°C. Generally, these will be useable for up to one month. Before use, and prior to opening the vial we recommend that you allow your product to equilibrate to room temperature for at least 1 hour.

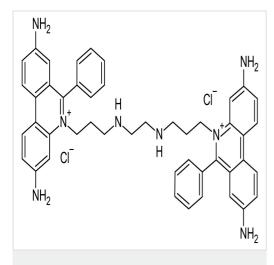
Refer to SDS for further information.

Need more advice on solubility, usage and handling? Please visit our <u>frequently asked</u> <u>questions (FAQ) page</u> for more details.

Source

Synthetic

Images



2D chemical structure image of ab145323, Ethidium homodimer, Fluorimetric detection reagent

Chemical Structure - Ethidium homodimer,

Fluorimetric detection reagent (ab145323)

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•	Abcam biochemicals are novel compounds and we have not tested their biological activity in house. Please use the literature to identify how to use these products effectively. If you require further assistance please contact the scientific support team

Product datasheet

Calcein AM, fluorescent dye for cell viability ab141420

**** 1 Abreviews 11 References 1 Image

Overview

Product nameCalcein AM, fluorescent dye for cell viability

DescriptionCell-permeable fluorescent dye for determining cell viability

Purity > 96%

CAS Number 148504-34-1

Chemical structure

Properties

 Excitation
 495nm

 Emission
 515nm

Chemical name N,N'-[[3',6'-Bis(acetyloxy)-3-oxospiro[isobenzofuran-1(3H),9'-

[9*H*]xanthene]-2',7'-diyl]bis(methylene)]bis[*N*-[2-[(acetyloxy)methoxy]-2-oxoethyl]-glycine 1,1'-

bis[(acetyloxy)methyl] ester

Molecular weight 994.87

Molecular formula $C_{46}H_{46}N_2O_{23}$

PubChem identifier 390986

Storage instructions Store at -20°C. It is important to note that this product is reported to be light sensitive. Store In the

Dark. Store under desiccating conditions.

Solubility overview Soluble in DMSO

Handling Wherever possible, you should prepare and use solutions on the same day. However, if you need

to make up stock solutions in advance, we recommend that you store the solution as aliquots in tightly sealed vials at -20°C. Generally, these will be useable for up to one month. Before use, and

prior to opening the vial we recommend that you allow your product to equilibrate to room

temperature for at least 1 hour.

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For more information on AM esters please visit our AM esters FAQ page.

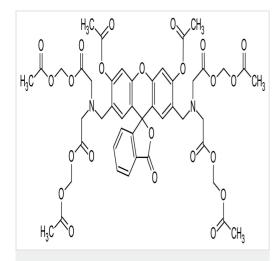
Need more advice on solubility, usage and handling? Please visit our <u>frequently asked</u> <u>questions (FAQ) page</u> for more details.

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2) OC (= O) C) CN (CC (= O) OCOC (= O) C) CC (= O) OCOC (= O) C) OC (= O) C) CC (= O) COC (= O) C

Source Synthetic

Images



Chemical Structure - Calcein AM, fluorescent dye for cell viability (ab141420)

2D chemical structure image of ab141420, Calcein AM, fluorescent dye for cell viability

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Product datasheet

ECL Substrate Kit (High Sensitivity) ab133406

★★★★★ <u>5 Abreviews</u> <u>46 References</u> 2 Images

Overview

Product name

ECL Substrate Kit (High Sensitivity)

Product overview

High Sensitivity ECL Substrate Kit ab133406 is designed for the detection of proteins with 23pg-187ng of protein per band. It uses an enhanced chemiluminescent substrate for western blotting that was developed for film imaging and is also compatible with CCD imaging. The high sensitivity ECL substrate produces a strong signal with very low background. Additionally, the ECL signal is long lasting, allowing repeated exposures without fear of losing data.

Our ECL kits include our popular <u>ECL Substrate Kit ab65623</u> and our high sensitivity ECL substrate kits:

- this kit (High Sensitivity ECL Substrate Kit ab133406) to detect 23pg-187ng of protein per band
- Very High Sensitivity ECL Substrate Kit ab133408 to detect 4.6pg-4.7ng of protein per band
- Ultra High Sensitivity ECL Substrate Kit ab133409 to detect 1.2pg-2ng of protein per band

Detection ranges in pg and ng stated above should be used for guidance only as detection range is dependent on the molecular weight of a protein.

This product was previously called Optiblot ECL Detect Kit (23pg-187ng).

Notes

Use:

- 200ml kit for 2000cm² membrane
- 500ml kit for 5000cm² membrane

The primary antibody can often be diluted 5 to 10 fold more than usual when using this ECL substrate. A typical primary antibody dilution range using the substrate is 1/5000 - 1/20,000, with a typical secondary antibody dilution range of 1/20,000 - 1/100,000. Some optimisation may be required.

Tested applications

Suitable for: WB

Properties

Storage instructions

Store at +4°C. Please refer to protocols.

Components	200 ml	20 ml	500 ml
Luminol/Enhancer Solution	1 x 100ml	1 x 10ml	1 x 250ml
Peroxide Chemiluminescent Detection Reagent	1 x 100ml	1 x 10ml	1 x 250ml

Applications

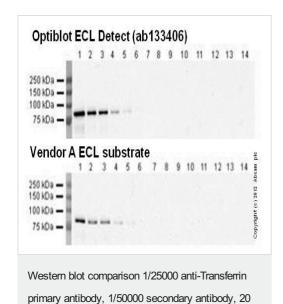
The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab133406 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		Use at an assay dependent concentration.

Images

second exposure

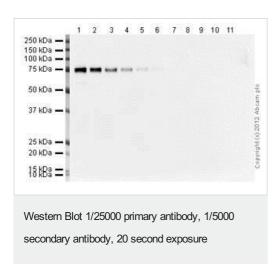


Anti-Transferrin antibody (**ab1223**) at **1/25000** dilution. Lanes 1-14 Transferrin protein (**ab91435**), Loading dilution (Lanes 1-8): 12.5, 6.2, 3.1, 1.6, 0.8, 0.3, 0.19 μ g protein.

Secondary

HRP conjugated polyclonal to Rabbit lgG (<u>ab97080</u>) at 1/50000 developed using Optiblot ECL Detect and Vendor A ECL subtrate.

Exposure time: 20 seconds



Each lane contains the following amount of COX2 recombinant protein (ab58868): 1) 100ng 2) 50ng 3) 25ng 4) 12.5ng 5) 6.25ng 6) 3.13ng 7) 1.56ng 8) 780pg 9) 390pg 10) 195pg 11) 98pg. The blot was probed with rabbit anti-COX2 antibody (ab15191) at 1/25000 dilution and with a rabbit secondary (ab97080) at 1/5000 dilution. The blot was developed with 20ml of Optiblot ECL Detect.

Exposure time: 20 seconds

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Product datasheet

Superoxide Dismutase Activity Assay Kit (Colorimetric) ab65354

*** * * * 2 Abreviews 218 References 4 Images

Overview

Notes

Product name Superoxide Dismutase Activity Assay Kit (Colorimetric)

Detection methodColorimetric

Sample type Urine, Serum, Plasma, Other biological fluids, Tissue Extracts, Cell Lysate, Cell culture media

Assay type Enzyme activity

Assay time 0h 30m

Species reactivity Reacts with: Mammals, Other species

Product overview Superoxide Dismutase Activity Assay Kit (Colorimetric) ab65354 is a simple and rapid assay for

superoxide dismutase (SOD) activity.

In the SOD assay protocol:

- superoxide anions are produced by the action of xanthine oxidase

- SOD catalyzes the dismutation of the superoxide anion into hydrogen peroxide and $\mbox{\rm O}_2$

- superoxide anions act on WST-1 to produce a water-soluble formazan dye which can be

detected by the increase in absorbance at 450 nm

The greater the activity of SOD in the sample, the less formazan dye is produced.

Superoxide dismutase assay protocol summary:

- add samples to wells

- add WST-1 working solution and enzyme working solution and incubate for 20 min at 37°C

- analyze with microplate reader

This product is manufactured by BioVision, an Abcam company and was previously called K335

Superoxide Dismutase (SOD) Activity Colorimetric Assay Kit. K335-100 is the same size as the

100 test size of ab65354.

Superoxide dismutase (SOD) is one of the most important antioxidative enzymes. It catalyzes the

dismutation of the superoxide anion into hydrogen peroxide and molecular oxygen.

Related products

Review the oxidative stress marker and assay guide to learn about more assays for oxidative

stress.

Platform Microplate reader

1

Properties

Storage instructions

Store at +4°C. Please refer to protocols.

Components	Identifier	100 tests	2000 tests
SOD Assay Buffer	WM	1 x 20ml	20 x 20ml
SOD Dilution Buffer	NM	1 x 10ml	20 x 10ml
SOD Enzyme Solution	Green	1 x 20µl	20 x 20µl
WST Solution	Red	1 x 1ml	20 x 1ml

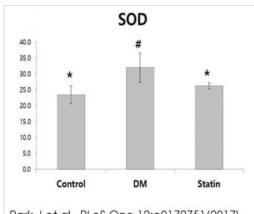
Relevance

Superoxide dismutase (SOD) is an antioxidant enzyme involved in the defense system against reactive oxygen species (ROS). SOD catalyzes the dismutation reaction of superoxide radical anion (O_2 -) to hydrogen peroxide, which is then catalyzed to innocuous O_2 and H_2O by glutathione peroxidase and catalase. Several classes of SOD have been identified. These include intracellular copper, zinc SOD (Cu, Zn SOD/SOD1), mitochondrial manganese SOD (Mn SOD/SOD2) and extracellular Cu, Zn SOD (EC SOD/SOD3).

Cellular localization

Cytoplasmic

Images



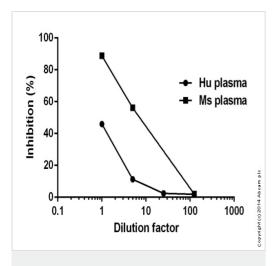
Park J et al., PLoS One 12:e0172751 (2017)

Functional studies - ab65354

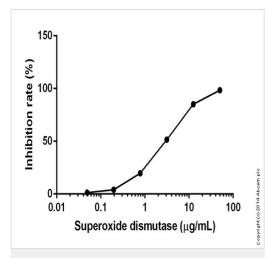
Image from Park J et al., PLoS One 12(2), fig 4b. Doi: 10.1371/journal.pone.0172751 Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Park J et al investigates the recovery in erectile function after administration of chronic statin alone in DM (streptozotocin (STZ)-induced diabetes mellitus) rats. SOD activity was determined using Superoxide Dismutase activity assay kit (ab65354).

* Indicates statistical significance in comparison with DM group (P < 0.05). # Indicates statistical significance in comparison with the statin group (P<0.05).

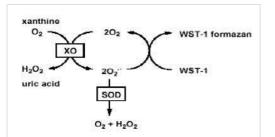


Functional Studies- Superoxide Dismutase Activity Assay Kit (ab65354)



Functional Studies - Superoxide Dismutase Activity Assay Kit (Colorimetric) (ab65354) Superoxide dismutase measured in biofluids at various dilutions

Superoxidase dismutase (<u>ab90040</u>) measured showing inhibition rate (%) per concentration (microgram per mL)



Functional Studies - Superoxide Dismutase Activity
Assay Kit (Colorimetric) (ab65354)

Principle of Superoxide Dismutase Assay.

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Product datasheet

Protein Carbonyl Content Assay Kit ab126287

* ★ ★ ★ ★ 1 Abreviews 48 References 2 Images

Overview

Product name Protein Carbonyl Content Assay Kit

 Detection method
 Colorimetric

 Assay type
 Quantitative

 Sensitivity
 0.015 μM

 Assay time
 1h 30m

Product overview Protein Carbonyl Content Assay Kit ab126287 is designed to provide a simple and accurate

method of quantifying carbonyls in protein samples.

The protein carbonl assay protocol is based on the reaction of DNPH with protein carbonyls. This is the most popular assay type for measuring protein carbonyl content. DNP hydrazones formed in this reaction are easily quantifiable at 375 nm absorbance.

When used with, for example a 1 mg (~15 nmol) sample, this protein carbonyl assay protocol has a detection limit of about 0.15 nmol carbonyl. For context, BSA typically contains approximately 1-3 nmol carbonyl/mg.

Protein carbonyl assay protocol summary:

- add DNPH to samples and incubate for 10 min
- add TCA to samples, incubate for 5 min, spin for 2 min, and discard supernatant
- wash pellet by sonicating in acetone, spin and remove acetone, repeat acetone wash step
- add Guanidine solution and resolubilize pellet, and transfer to plate
- analyze with a microplate reader to measure protein carbonyl content, also perform a protein assay (eg. BCA assay) on samples to establish protein concentration

This product is manufactured by BioVision, an Abcam company and was previously called K830 Protein Carbonyl Content Assay Kit. K830-100 is the same size as the 100 test size of ab126287.

Related products

This is the most popular Protein Carbonyl Content assay kit. Other assays include:

- Protein Carbonyl Content Assay (Fluorometric) ab235631
- Protein Carbonyl Content ELISA (DNPH) ab238536
- Protein Carbonyl Content Western Blot Assay ab178020

Review the oxidative stress marker and assay guide, or the full metabolism assay guide to

Notes

learn about more assays for metabolites, metabolic enzymes, mitochondrial function, and oxidative stress, and also how to assay metabolic function in live cells using your plate reader.

Platform

Microplate reader

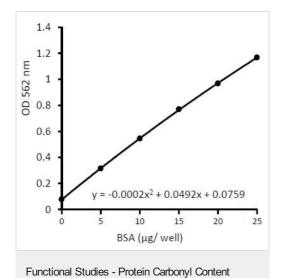
Properties

Storage instructions

Store at +4°C. Please refer to protocols.

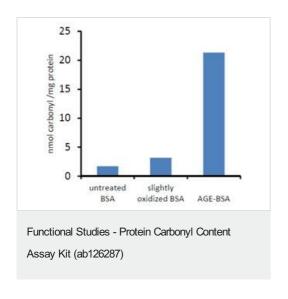
Components	Identifier	100 tests
10% Streptozocin Solution	Blue	1 x 1ml
87% TCA Solution	NM	1 x 3ml
6 M Guanidine Solution	WM	1 x 20ml
96-Well Clear Plate		1 unit
DNPH Solution	Amber	1 x 11ml

Images



Assay Kit (ab126287)

Typical Standard Curve using BCA Protein Quantification Assay (ab102536)



Representative Data Obtained Using the Protein Carbonyl Content Assay Kit

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Product datasheet

Glutathione Assay Kit (Fluorometric) ab65322

★★★★★ 1 Abreviews 33 References 2 Images

Overview

Product name Glutathione Assay Kit (Fluorometric)

Detection method Fluorescent

Sample type Urine, Serum, Plasma, Other biological fluids, Tissue Extracts, Cell Lysate, Cell culture media

Assay type Quantitative Assay time 2h 00m

Species reactivity Reacts with: Mammals, Other species

Product overview

Glutathione Assay Kit (Fluorometric) (ab65322) provides a simple in vitro assay for detection of total glutathione changes during cellular response to toxicity, apoptosis and other conditions. The assay uses the dye monochlorobimane (MCB), which forms an adduct with glutathione in a reaction catalyzed by glutathione-S-Transferase (GST). The unbound MCB is almost nonfluorescent, whereas it emits a fluorescent blue light (Ex/Em = 380nm/461nm) when bound to reduced or oxidized glutathione. Thus, the amount of glutathione can be easily detected using a

fluorometer or a 96-well fluorometric plate reader.

Visit our **FAQs page** for tips and troubleshooting.

Notes This product is manufactured by BioVision, an Abcam company and was previously called K251

Glutathione Fluorometric Assay Kit. K251-100 is the same size as the 100 test size of ab65322.

Glutathione (GSH) is a tripeptide that contains L-cysteine, L-glutamic acid and glycine. It is the smallest intracellular protein thiol molecule in the cells, which prevents cell damage caused by reactive oxygen species such as free radicals and peroxides. Glutathione exists in reduced (GSH) and oxidized (GSSG) states. Reduced glutathione (GSH) is a major tissue antioxidant that provides reducing equivalents for the glutathione peroxidase (GPx) catalyzed reduction of lipid hydroperoxides to their corresponding alcohols and hydrogen peroxide to water. In the GPx catalyzed reaction, the formation of a disulfide bond between two GSH molecules generates oxidized glutathione (GSSG). The enzyme glutathione reductase (GR) recycles GSSG to GSH with the simultaneous oxidation of β -nicotinamide adenine dinucleotide phosphate (β -NADPH2). In healthy cells, more than 90% of the total glutathione pool is in the reduced form (GSH). When cells are exposed to increased levels of oxidative stress, GSSG accumulates and the ratio of GSSG to GSH increases. An increased ratio of GSSG-to-GSH is an indication of oxidative stress. The monitoring of reduced and oxidized GSH in biological samples is essential for evaluating the redox and detoxification status of the cells and tissues against oxidative and free radicals mediated cell injury.

Platform Microplate reader

Properties

Storage instructions

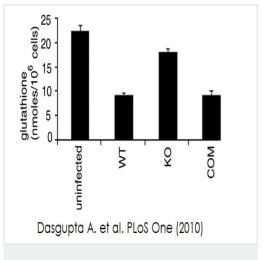
Store at -20°C. Please refer to protocols.

Components	Identifier	100 tests
Cell Lysis Buffer		1 x 25ml
GSH Standard	Yellow	1 vial
GST Reagent	Green	1 x 200µl
Monochlorobimane Substrate	Red	1 x 200µl

Relevance

Glutathione is a small peptide composed of three amino acids: cysteine, glutamic acid, and glycine and is present in tissues in concentrations as high as one millimolar. Glutathione is the principal intracellular low-molecular-weight thiol that plays a critical role in the cellular defense against oxidative and nitrosative stress in mammalian cells. Diminished glutathione levels have been observed in the early stages of apoptosis.

Images

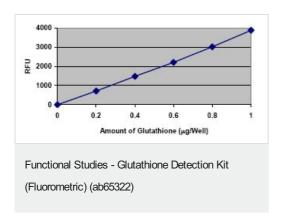


Glutathione Detection Kit (Fluorometric) (ab65322)

Dasgupta A et al., PLoS One, 5, , 2010 Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Glutathione pool measured in THP-1 macrophages: uninfected cells; WT: infected with M.tuberculosis wild type; KO: infected with M.tuberculosis OppD knock-out; COM: infected with M.tuberculosis OppD knock-out complemented with OppDA gene. 10^6 cells were infected and lysed by treating them with 100μ I of ice cold lysis buffer. Cell lysate was diluted and mixed as described in the kit protocol. After 30 min incubation at 37C, fluorescence was measured at Ex=380nm/ Em=460nm. Results represent the means of \pm S.D. of three determinations.

Image obtained from Dasgupta A. et al; PLoS One; 2010 Aug 17; 5(8): e12225.



Glutathione assays were performed using various amounts of Glutathione as indicated. Results were analyzed according to the kit instructions.

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Product datasheet

Human CRP ELISA Kit (C-Reactive protein) ab260058

Recombinant SimpleStepELISA

3 References 11 Images

Overview

Recovery

Product name Human CRP ELISA Kit (C-Reactive protein)

Detection method Colorimetric

Precision Intra-assav

Sample	n	Mean	SD	CV%
Serum	8			1.4%

Inter-assay

Sample	n	Mean	SD	CV%	
Serum	3			4%	

Sample type Cell culture supernatant, Serum, Hep Plasma, EDTA Plasma, Cit plasma

Sandwich (quantitative) Assay type

Sensitivity 5.36 pg/ml

18.75 pg/ml - 1200 pg/ml Range

	Sample specific recovery

Sample type	Average %	Range
Cell culture supernatant	100	99% - 100%
Serum	109	105% - 112%
Hep Plasma	114	112% - 119%
EDTA Plasma	110	107% - 113%
Cit plasma	107	105% - 109%

Assay time 1h 30m

Assay duration One step assay

Species reactivity

Product overview

Reacts with: Human

Human CRP ELISA Kit (C-Reactive protein) (ab260058) is a single-wash 90 min sandwich ELISA designed for the quantitative measurement of CRP (C-Reactive protein) protein in cell culture supernatant, cit plasma, edta plasma, hep plasma, and serum. It uses our proprietary SimpleStep ELISA® technology. Quantitate Human CRP (C-Reactive protein) with 5.36 pg/ml sensitivity.

SimpleStep ELISA® technology employs capture antibodies conjugated to an affinity tag that is recognized by the monoclonal antibody used to coat our SimpleStep ELISA® plates. This approach to sandwich ELISA allows the formation of the antibody-analyte sandwich complex in a single step, significantly reducing assay time. See the SimpleStep ELISA® protocol summary in the image section for further details. Our SimpleStep ELISA® technology provides several benefits:

- Single-wash protocol reduces assay time to 90 minutes or less
- High sensitivity, specificity and reproducibility from superior antibodies
- Fully validated in biological samples
- 96-wells plate breakable into 12 x 8 wells strips

A 384-well SimpleStep ELISA® microplate (<u>ab203359</u>) is available to use as an alternative to the 96-well microplate provided with SimpleStep ELISA® kits.

ASSAY SPECIFICITY: This kit recognizes both native and recombinant human and monkey CRP protein in serum, plasma, and cell culture supernatant.

Cell and tissue extract, urine, milk, and saliva samples have not been tested with this kit.

CROSS REACTIVITY: Recombinant mouse CRP and rat CRP were both prepared at 50 ng/mL respectively and assayed for cross reactivity. No cross-reactivity was observed.

INTERFERENCE: Recombinant mouse CRP and rat CRP were both prepared at 50 ng/mL respectively and tested for interference. No interference with was observed.

SPECIES REACTIVITY: This kit recognizes human and monkey CRP protein.

Other species reactivity was determined by measuring 1:1000 serum samples of various species, interpolating the protein concentrations from the human standard curve, and expressing the interpolated concentrations as a percentage of the protein concentration in human serum assayed at the same dilution.

Reactivity < 3% was determined for the following species: Mouse, Rat, Cow.

Other species reactivity not determined.

C-reactive protein (CRP) displays several functions associated with host defense: it promotes agglutination, bacterial capsular swelling, phagocytosis and complement fixation through its calcium-dependent binding to phosphorylcholine. CRP can interact with DNA and histones and it may scavenge nuclear material released from damaged circulating cells. CRP is secreted; it

Notes

forms a homopentamer pentaxin (or pentraxin) which have a discoid arrangement of 5 non-covalently bound subunits. CRP binds 2 calcium ions per subunit. The concentration of CRP in plasma increases greatly during acute phase response to tissue injury, infection or other inflammatory stimuli. It is induced by IL1/interleukin-1 and IL6//interleukin-6. This kit requires the use of acid treatment for the disassociation of CRP.

Platform

Pre-coated microplate (12 x 8 well strips)

Properties

Storage instructions

Store at +4°C. Please refer to protocols.

Components	1 x 96 tests
10X Human CRP (C-Reactive Protein) Capture Antibody	1 x 600µl
10X Human CRP (C-Reactive Protein) Detector Antibody	1 x 600µl
10X Wash Buffer PT (ab206977)	1 x 20ml
Antibody Diluent 4BI	1 x 6ml
Human CRP (C-Reactive Protein) Lyophilized Recombinant Protein	2 vials
Plate Seals	1 unit
Sample Diluent NS (ab193972)	1 x 50ml
SimpleStep Pre-Coated 96-Well Microplate (ab206978)	1 unit
Stop Solution	1 x 12ml
TMB Development Solution	1 x 12ml

Function Displays several functions associated with host defense: it promotes agglutination, bacterial

capsular swelling, phagocytosis and complement fixation through its calcium-dependent binding to phosphorylcholine. Can interact with DNA and histones and may scavenge nuclear material

released from damaged circulating cells.

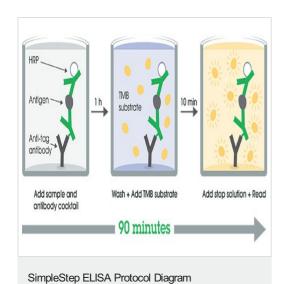
Tissue specificity Found in plasma.

Sequence similarities Belongs to the pentaxin family.

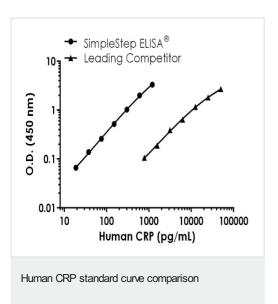
Contains 1 pentaxin domain.

Cellular localization Secreted.

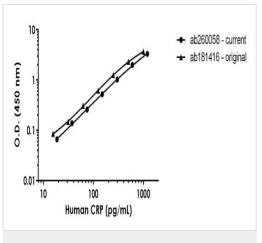
Images



SimpleStep ELISA technology allows the formation of the antibodyantigen complex in one single step, reducing assay time to 90 minutes. Add samples or standards and antibody mix to wells all at once, incubate, wash, and add your final substrate. See protocol for a detailed step-by-step guide.

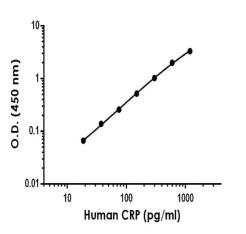


Standard Curve comparison between human CRP SimpleStep ELISA kit and traditional ELISA kit from leading competitor. SimpleStep ELISA kit shows increased sensitivity.



Human CRP ELISA kit comparison comparison

Standard Curve comparison between ab260058 and <u>ab181416</u> SimpleStep ELISA kits. ab260058 SimpleStep ELISA kit shows comparable sensitivity.



Example of human CRP standard curve in Sample

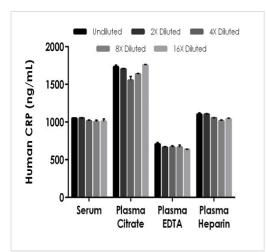
The CRP standard curve was prepared as described in Section 10. Raw data values are shown in the table. Background-subtracted data values (mean +/- SD) are graphed.

Concentration	O.D 450 nm		Mean
(pg/ml)	1	2	O.D
0	0.064	0.058	0.061
18.75	0.125	0.130	0.127
37.5	0.195	0.205	0.200
75	0.311	0.329	0.320
150	0.578	0.586	0.582
300	1.076	1.103	1.090
600	2.052	2.070	2.061
1200	3.395	3.383	3.389

Standard curve

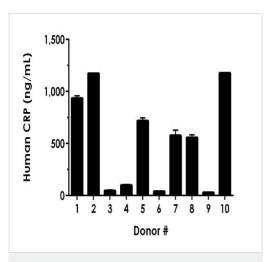
Diluent NS.

Example of human CRP standard curve in Sample Diluent NS. Raw $$
data values are shown in the table. Background-subtracted data
values (mean +/- SD) are graphed.



Interpolated concentrations of native CRP in human serum and plasma samples.

The concentrations of CRP were measured in duplicates, interpolated from the CRP standard curves and corrected for sample dilution. Undiluted samples are as follows: serum 1:1000, plasma (citrate) 1:2000, plasma (EDTA) 1:1000, plasma (heparin) 1:1000. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean CRP concentration was determined to be 1025.5 ng/mL in serum, 1676.5 ng/mL in plasma (citrate) and 668.3 ng/mL in plasma (EDTA), and, 1062.8 ng/mL in plasma (heparin).



Interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean CRP concentration was determined to be 535.7 ng/mL with a range of 28.1 - 1180.4 ng/mL.

Serum from ten individual healthy human male donors was measured in duplicate.

Dilution Factor	Interpolated value	1:1000 Human Serum	1:2000 Human Plasma (Citrate)	1:1000 Human Plasma (EDTA)	1:1000 Human Plasma (Heparin)
11. on a o	pg/mL	1049.0	865.9	707.4	1102.8
Undiluted	% Expected value	100	100	100	100
	pg/mL	526.4	426.1	331.9	551.1
2	% Expected value	100	98	94	100
	pg/mL	253.4	194.2	166.6	263.3
4	% Expected value	97	90	94	96
	pg/mL	125.4	102.3	83.2	126.8
8	% Expected value	96	95	94	92
17	pg/mL	63.0	54.9	39.9	65.1
16	% Expected value	96	101	90	94

Linearity of dilution is determined based on interpolated values from the standard curve. Linearity of dilution defines a sample concentration interval in which interpolated target concentrations are directly proportional to sample dilution.

Native CRP was measured in the following biological samples in a 2-fold dilution series. Sample dilutions are made in Sample Diluent NS.

Linearity of dilution.

Dilution Factor	Interpolated value	25% PBMC supernatant
na alticata al	pg/mL	587.8
Undiluted	% Expected value	100
2	pg/mL	286.6
2	% Expected value	98
4	pg/mL	139.9
4	% Expected value	95
8	pg/mL	74.2
0	% Expected value	101
14	pg/mL	39.9
16	% Expected value	109

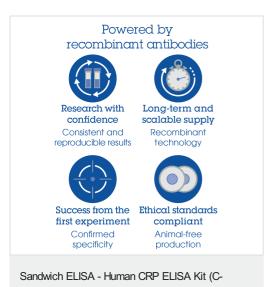
Recombinant CRP was spiked into the following biological samples and diluted in a 2-fold dilution series in Sample Diluent NS.

Linearity of dilution.

Sample Type	Average % Recovery	Range (%)
1:4000 Serum	109	105 - 112
1:4000 Plasma - Citrate	107	105 - 109
1:4000 Plasma - EDTA	110	107 - 113
1:4000 Plasma - Heparin	114	112 - 119
25% PBMC Cell culture supernatant	100	99 - 100

Three concentrations of CRP recombinant protein were spiked in duplicate to the indicated biological matrix to evaluate signal recovery in the working range of the assay.

Recovery



Reactive protein) (ab260058)

To learn more about the advantages of recombinant antibodies see **here**.

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Product datasheet

Human Procalcitonin ELISA Kit ab221828

Recombinant SimpleStep ELISA

★★★★★ 1 Abreviews 3 References 7 Images

Overview

Product name

Human Procalcitonin ELISA Kit

Detection method

Colorimetric

Precision

Sample n Mean SD CV%

Urine 8 2.9%

Inter-assay

Intra-assay

Sample	n	Mean	SD	CV%	
Urine	3			4.4%	

Sample type

Cell culture supernatant, Saliva, Milk, Urine, Serum, Hep Plasma, EDTA Plasma, Cit plasma

Assay type

Sandwich (quantitative)

Sensitivity

1.51 pg/ml

Range

6.25 pg/ml - 400 pg/ml

Recovery

Sample specific recovery

Sample type	Average %	Range
Saliva	94	84% - 104%
Milk	79	75% - 88%
Urine	90	88% - 92%
Serum	79	74% - 82%
Cell culture media	100	99% - 101%
Hep Plasma	93	90% - 100%

1

Sample type	Average %	Range
EDTA Plasma	78	75% - 82%
Cit plasma	83	79% - 88%

Assay time

1h 30m

Assay duration

One step assay

Species reactivity

Reacts with: Human

Does not react with: Cow

Product overview

Human Procalcitonin ELISA Kit (ab221828) is a single-wash 90 min sandwich ELISA designed for the quantitative measurement of Procalcitonin protein in cell culture supernatant, cit plasma, edta plasma, hep plasma, milk, saliva, serum, and urine. It uses our proprietary SimpleStep ELISA® technology. Quantitate Human Procalcitonin with 1.51 pg/ml sensitivity.

SimpleStep ELISA® technology employs capture antibodies conjugated to an affinity tag that is recognized by the monoclonal antibody used to coat our SimpleStep ELISA® plates. This approach to sandwich ELISA allows the formation of the antibody-analyte sandwich complex in a single step, significantly reducing assay time. See the SimpleStep ELISA® protocol summary in the image section for further details. Our SimpleStep ELISA® technology provides several benefits:

- Single-wash protocol reduces assay time to 90 minutes or less
- High sensitivity, specificity and reproducibility from superior antibodies
- Fully validated in biological samples
- 96-wells plate breakable into 12 x 8 wells strips

A 384-well SimpleStep ELISA® microplate (<u>ab203359</u>) is available to use as an alternative to the 96-well microplate provided with SimpleStep ELISA® kits.

Notes

Human Procalcitonin is a 116-amino acid precursor protein of the peptide hormone called Calcitonin coded by the gene CALCA. Procalcitonin is mainly produced by the C-cells of the thyroid and certain endocrine cells of the lung. Under normal expression conditions, Procalcitonin is cleaved into three specific fragments: an N-terminal residue, Calcitonin, and Katacalcin. Levels of unprocessed Procalcitonin have been shown to rise significantly after bacterial infection, trauma, and shock. Calcitonin regulates the blood phosphate and calcium levels by promoting incorporation into bones. Populations with chronic kidney disease have been shown to have higher procalcitonin levels. The antibodies used in this product were raised to the propeptide sequence residues 26-82. The standard protein in this product has been mass calibrated to a full-length procalcitonin (residues 26-141).

Platform

Pre-coated microplate (12 x 8 well strips)

Properties

Storage instructions

Store at +4°C. Please refer to protocols.

Components	1 x 96 tests
10X Human Procalcitonin Capture Antibody	1 x 600µl
10X Human Procalcitonin Detector Antibody	1 x 600µl
Human Procalcitonin Lyophilized Recombinant Protein	2 vials
Antibody Diluent 4BI	1 x 6ml
10X Wash Buffer PT (ab206977)	1 x 20ml
TMB Development Solution	1 x 12ml
Stop Solution	1 x 12ml
Sample Diluent NS (ab193972)	1 x 50ml
SimpleStep Pre-Coated 96-Well Microplate (ab206978)	1 unit
Plate Seals	1 unit

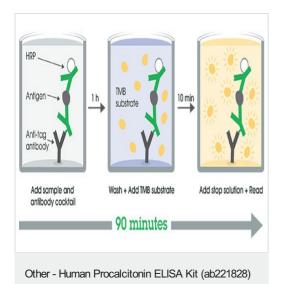
Relevance

Procalcitonin (PCT) is a 116 amino acid residue peptide with molecular weight of about 13 kDa. PCT itself has no known hormonal activity. PCT belongs to a group of related proteins including calcitonin gene-related peptides I and II, amylin, adrenomodulin and calcitonin (CAPA peptide family). PCT, like other peptides of CAPA family, appears from the common precursor preprocalcitonin consisting of 141 amino acids by removal of 25 amino acids from the N-terminus. PCT undergoes successive cleavages to form three molecules: N-terminal fragment (55 a.a.), calcitonin (32 a.a.) and katacalcin (21 a.a.). Under normal metabolic conditions, PCT is only present in the C cells of the thyroid gland. In bacterial infection and sepsis, however, intact PCT is found in the blood and, more importantly, its level is related to the severity of bacterial sepsis. Today, PCT is considered to be one of the earliest and most specific markers of sepsis.

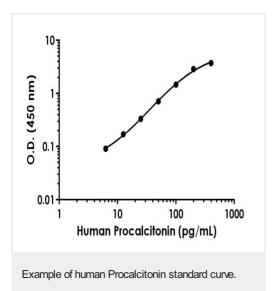
Cellular localization

Secreted

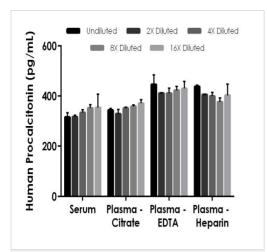
Images



SimpleStep ELISA technology allows the formation of the antibodyantigen complex in one single step, reducing assay time to 90 minutes. Add samples or standards and antibody mix to wells all at once, incubate, wash, and add your final substrate. See protocol for a detailed step-by-step guide.

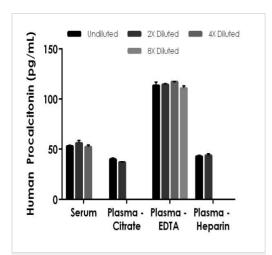


Background-subtracted data values (mean +/- SD) are graphed.

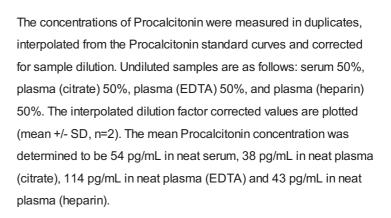


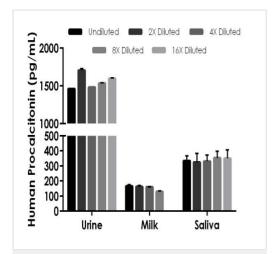
Interpolated concentrations of spiked Procalcitonin in human serum and plasma samples.

The concentrations of Procalcitonin were measured in duplicates, interpolated from the Procalcitonin standard curves and corrected for sample dilution. Undiluted samples are as follows: serum 50%, plasma (citrate) 50%, plasma (EDTA) 50%, and plasma (heparin) 50%. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean Procalcitonin concentration was determined to be 335 pg/mL in neat serum, 351 pg/mL in neat plasma (citrate), 424 pg/mL in neat plasma (EDTA) and 405 pg/mL in neat plasma (heparin).



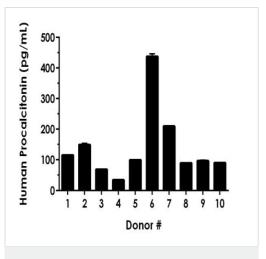
Interpolated concentrations of native Procalcitonin in human serum and plasma samples.





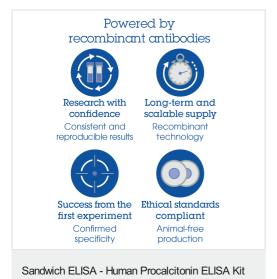
Interpolated concentrations of native Procalcitonin in human urine, milk, and spiked Procalcitonin in human saliva samples.

The concentrations of Procalcitonin were measured in duplicates, interpolated from the Procalcitonin standard curves and corrected for sample dilution. Undiluted samples are as follows: urine 25%, milk 50%, and saliva 50%. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean Procalcitonin concentration was determined to be 1,555 pg/mL in neat urine, 154 pg/mL in neat milk, and 338 pg/mL in neat saliva.



Interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean Procalcitonin concentration in neat serum was determined to be 139 pg/mL with a range of 33-438 pg/mL.

Serum from ten individual healthy human male donors was measured in duplicate.



(ab221828)

To learn more about the advantages of recombinant antibodies see **here**.

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Product datasheet

Superoxide Dismutase Activity Assay Kit (Colorimetric) ab65354

*** * * * 2 Abreviews 218 References 4 Images

Overview

Notes

Product name Superoxide Dismutase Activity Assay Kit (Colorimetric)

Detection methodColorimetric

Sample type Urine, Serum, Plasma, Other biological fluids, Tissue Extracts, Cell Lysate, Cell culture media

Assay type Enzyme activity

Assay time 0h 30m

Species reactivity Reacts with: Mammals, Other species

Product overview Superoxide Dismutase Activity Assay Kit (Colorimetric) ab65354 is a simple and rapid assay for

superoxide dismutase (SOD) activity.

In the SOD assay protocol:

- superoxide anions are produced by the action of xanthine oxidase

- SOD catalyzes the dismutation of the superoxide anion into hydrogen peroxide and $\mbox{\rm O}_2$

- superoxide anions act on WST-1 to produce a water-soluble formazan dye which can be

detected by the increase in absorbance at 450 nm

The greater the activity of SOD in the sample, the less formazan dye is produced.

Superoxide dismutase assay protocol summary:

- add samples to wells

- add WST-1 working solution and enzyme working solution and incubate for 20 min at 37°C

- analyze with microplate reader

This product is manufactured by BioVision, an Abcam company and was previously called K335

Superoxide Dismutase (SOD) Activity Colorimetric Assay Kit. K335-100 is the same size as the

100 test size of ab65354.

Superoxide dismutase (SOD) is one of the most important antioxidative enzymes. It catalyzes the

dismutation of the superoxide anion into hydrogen peroxide and molecular oxygen.

Related products

Review the oxidative stress marker and assay guide to learn about more assays for oxidative

stress.

Platform Microplate reader

1

Properties

Storage instructions

Store at +4°C. Please refer to protocols.

Components	Identifier	100 tests	2000 tests
SOD Assay Buffer	WM	1 x 20ml	20 x 20ml
SOD Dilution Buffer	NM	1 x 10ml	20 x 10ml
SOD Enzyme Solution	Green	1 x 20µl	20 x 20µl
WST Solution	Red	1 x 1ml	20 x 1ml

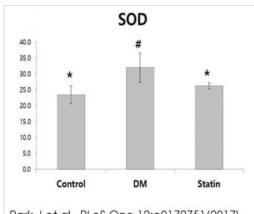
Relevance

Superoxide dismutase (SOD) is an antioxidant enzyme involved in the defense system against reactive oxygen species (ROS). SOD catalyzes the dismutation reaction of superoxide radical anion (O_2 -) to hydrogen peroxide, which is then catalyzed to innocuous O_2 and H_2O by glutathione peroxidase and catalase. Several classes of SOD have been identified. These include intracellular copper, zinc SOD (Cu, Zn SOD/SOD1), mitochondrial manganese SOD (Mn SOD/SOD2) and extracellular Cu, Zn SOD (EC SOD/SOD3).

Cellular localization

Cytoplasmic

Images



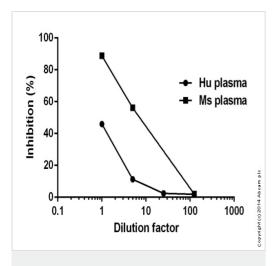
Park J et al., PLoS One 12:e0172751 (2017)

Functional studies - ab65354

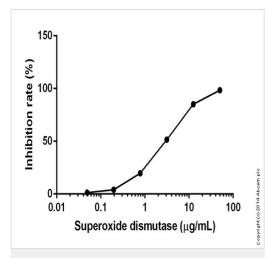
Image from Park J et al., PLoS One 12(2), fig 4b. Doi: 10.1371/journal.pone.0172751 Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Park J et al investigates the recovery in erectile function after administration of chronic statin alone in DM (streptozotocin (STZ)-induced diabetes mellitus) rats. SOD activity was determined using Superoxide Dismutase activity assay kit (ab65354).

* Indicates statistical significance in comparison with DM group (P < 0.05). # Indicates statistical significance in comparison with the statin group (P<0.05).

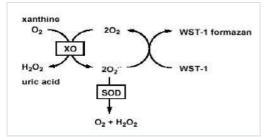


Functional Studies- Superoxide Dismutase Activity Assay Kit (ab65354)



Functional Studies - Superoxide Dismutase Activity Assay Kit (Colorimetric) (ab65354) Superoxide dismutase measured in biofluids at various dilutions

Superoxidase dismutase (<u>ab90040</u>) measured showing inhibition rate (%) per concentration (microgram per mL)



Functional Studies - Superoxide Dismutase Activity Assay Kit (Colorimetric) (ab65354) Principle of Superoxide Dismutase Assay.

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Product datasheet

Glutathione Peroxidase Assay Kit (Colorimetric) ab102530

★★★★★ 1 Abreviews 103 References 6 Images

Overview

Product name Glutathione Peroxidase Assay Kit (Colorimetric)

Detection methodColorimetric

Sample type Cell culture supernatant, Urine, Serum, Plasma, Platelets, Other biological fluids, Tissue Extracts

Assay type Enzyme activity

Sensitivity 0.5 mU/ml
Assay time 0h 40m

Species reactivity Reacts with: Mammals, Other species

Product overview Glutathione Peroxidase Assay Kit (Colorimetric) ab102530 can be used to quantitate the activity

of all of the glutathione dependent peroxidases in plasma, erythrocyte lysates, tissue

homogenates, and cell lysates.

In the glutathione peroxidase assay protocol, glutathione peroxidase (GPx) oxidizes GSH to produce GSSG as part of the reaction in which it reduces cumene hydroperoxide. Glutathione reductase (GR) then reduces the GSSG to produce GSH, and in the same reaction consumes NADPH. The decrease of NADPH (measured at OD=340 nm) is proportional to GPx activity.

The assay has a detection sensitivity of ~ 0.5 mU/ml of GPx in samples.

Glutathione peroxidase assay protocol summary:

- add samples and standards to wells
- add reaction mix and incubate for 15 min at room temp to deplete all GSSG in sample
- add cumene hydroperoxide
- analyze with microplate reader immediately, and after at least 5 min

This product is manufactured by BioVision, an Abcam company and was previously called K762 Glutathione Peroxidase Activity Colorimetric Assay Kit. K762-100 is the same size as the 100

test size of ab102530.

The Glutathione Peroxidase (GPx, EC 1.11.1.9) family of enzymes plays and important role in the protection of organisms from oxidative damage. GPx converts reduced glutathione (GSH) to oxidized glutathione (GSSG) while reducing lipid hydroperoxides to their corresponding alcohols or free hydrogen peroxide to water.

Notes

Properties

Storage instructions

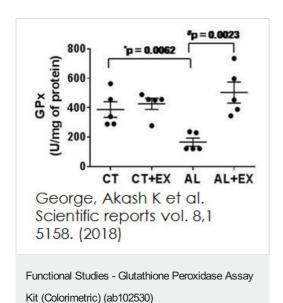
Store at -20°C. Please refer to protocols.

Components	Identifier	100 tests	2000 tests
Cumene Hydroperoxide	Yellow	1 vial	20 vials
Glutathione	Brown	1 vial	20 vials
Glutathione Reductase	Green	1 vial	20 vials
GPx Assay Buffer	WM	1 x 50ml	20 x 50ml
GPx Positive Control	Red	1 vial	20 vials
NADPH	Blue	1 vial	20 vials

Relevance

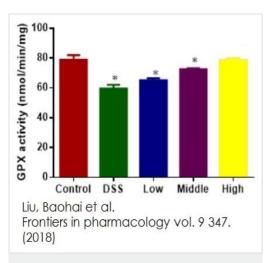
Glutathione Peroxidase (GPx, EC 1.11.1.9) is an enzyme family with peroxidase activity, and plays important role in protecting of organisms from oxidative damage. It converts reduced glutathione (GSH) to oxidized glutathione (GSSG), to reduce lipid hydroperoxides to their corresponding alcohols, or reduce free hydrogen peroxide to water. Several isozymes have been found in different cellular locations and with different substrate specificity. Low levels of GPx have been correlated with free radical related disorders.

Images



George, Akash K et al., Scientific reports?vol. 8,1 5158., Fig 4, doi:10.1038/s41598-018-23568-z

Scatter dot plots representing the levels of glutathione peroxidase (GPx) in brain tissue in different mice groups.

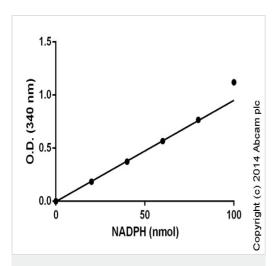


The effects of PCSE on the activity of GPX in a mouse UC model.

Functional Studies - Glutathione Peroxidase Assay

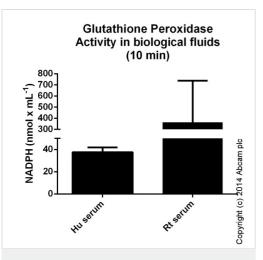
Kit (Colorimetric) (ab102530)

Liu, Baohai et al., Frontiers in pharmacology?vol. 9 347., Fig 8, doi:10.3389/fphar.2018.00347



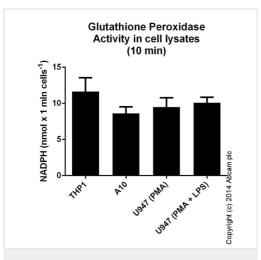
Functional Studies - Glutathione Peroxidase Assay Kit (ab102530)

Standard curve: mean of duplicates (+/- SD) with background reads subtracted



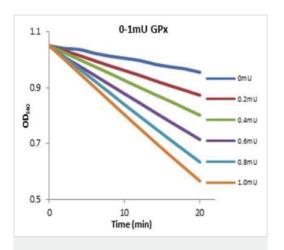
Functional Studies - Glutathione Peroxidase Assay Kit (ab102530)

Glutathione Peroxidase Activity measured in biological fluids showing quantity (nmol) per 1 mL after 10 min of incubation. Samples diluted 1-27 fold.



Functional Studies - Glutathione Peroxidase Assay Kit (ab102530)

Glutathione Peroxidase Activity measured in cell lysates showing quantity (nmol) per 1 mln cells after 10 min of incubation. Undiluted sample contained 2.5e6 cells per well. Samples were diluted 1-9 fold.



Functional Studies - Glutathione Peroxidase Assay Kit (ab102530)

Tests example obtained using ab102530

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Product datasheet

Glutathione Reductase (GR) Assay Kit ab83461

19 References 6 Images

Overview

Product name Glutathione Reductase (GR) Assay Kit

Detection methodColorimetric

Sample type Urine, Serum, Plasma, Other biological fluids, Tissue Extracts, Cell Lysate

Assay type Enzyme activity
Sensitivity > 0.1 mU/ml

Range 0.1 mU/ml - 40 mU/ml

Assay time 0h 40m

Product overview Glutathione Reductase Assay Kit (ab83461) is a highly sensitive, simple, direct and HTS-ready

colorimetric assay for measuring GR activity in biological samples. In the assay, GR reduces GSSG to GSH, which reacts with 5, 5'-Dithiobis (2-nitrobenzoic acid) (DTNB) to generate TNB2-

(yellow color, λ max = 405 nm). The assay can detect 0.1-40 mU/ml GR in various samples.

Since Glutathione Reductase has significantly higher concentrations in cells (mM range) compared to Thioredoxin Reductase (µM range), we predict that ab83461 will detect

mostly GR activity in samples.

Notes This product is manufactured by BioVision, an Abcam company and was previously called K761

Glutathione Reductase Activity Colorimetric Assay Kit. K761-200 is the same size as the 200 test

size of ab83461.

Glutathione Reductase (GR, EC 1.8.1.7) catalyzes the NADPH-dependent reduction of oxidized glutathione (GSSG) to reduced glutathione (GSH), which plays an important role in the GSH redox cycle that maintains adequate levels of reduced GSH. A high GSH/GSSG ratio is essential for

protection against oxidative stress.

Related products

Review the **oxidative stress marker and assay guide** to learn about more assays for oxidative

stress.

Platform Microplate reader

Properties

Storage instructions Store at -20°C. Please refer to protocols.

1

Components	Identifier	200 tests
Catalase (lyophilized)	Clear	1 x 1ng
DTNB (lyophilized)	Red	1 vial
GR Assay Buffer	NM	1 x 100ml
GR Positive Control (10 mU; lyophilized)	Green	1 vial
GSSG (lyophilized)	Yellow	1 vial
Hydrogen peroxide	Orange	1 x 1ml
NADPH-GNERAT™ (lyophilized)	Blue	2 vials
TNB Standard (2.5 μmol)	Brown	1 x 1ng

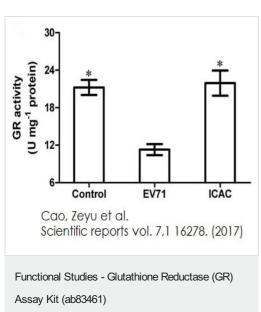
Relevance

Glutathione Reductase (GR, EC 1.8.1.7) catalyzes the NADPH-dependent reduction of oxidized glutathione (GSSG) to reduced glutathione (GSH), which plays an important role in the GSH redox cycle that maintains adequate levels of reduced GSH. A high GSH/GSSG ratio is essential for protection against oxidative stress.

Cellular localization

Cytoplasmic and Mitochondrial

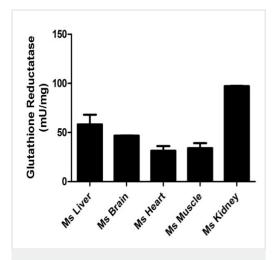
Images



ICAC decreased the ROS level induced by EV71 infection via modulating the antioxidant enzymes involved in GSH metabolism. Infected (100 TCID50 EV71) Vero cells were treated with medium or 100 μ M ICAC for 12 h. Uninfected cells were used as the control group. The antioxidant enzymes activities of the Vero cells were detected (n = 3). All results were expressed as the means \pm SEs. Asterisks indicate that the data significantly differ from the EV71 group at the P < 0.05 level according to one-way analysis of variance.

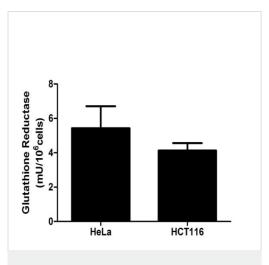
Cao, Zeyu et al., Scientific reports?vol. 7,1 16278., Fig 5, doi:10.1038/s41598-017-16446-7

2



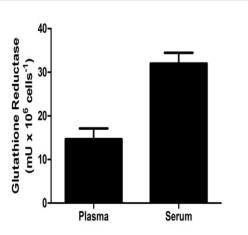
Glutathione reductase measured in mouse tissue lysates showing activity (mU) per mg of extracted protein (T_1 =2 min; T_2 =30 min).





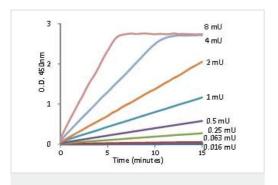
Functional Studies - Glutathione Reductase Assay Kit (ab83461)

Glutathione reductase measured in cell lysates (mU) per 10^6 cells (T₁=2 min; T₂=30 min)



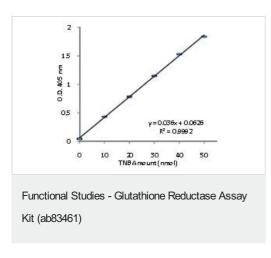
Glutathione reductase measured in human plasma and serum (T_1 =2 min; T_2 =30 min) showing activity (mU) per ml of tested sample. No activity was detected in saliva or urine.





Functional Studies - Glutathione Reductase Assay Kit (ab83461)

Glutathione Reductase assay time line using ab83461.



TNB Standard Curve using ab83461.

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NovocastraTM Anticorp monoclonal lichid de șoarece CD3

Cod produs: NCL-L-CD3-565

Utilizare prevăzută

Pentru diagnosticare in vitro.

NCL-L-CD3-565 este destinat identificării calitative, prin intermediul microscopiei optice, a antigenului CD3 uman în secțiunile de parafină. Interpretarea clinică a oricărei colorări sau a absenței acesteia trebuie completată cu studii morfologice utilizând controale adecvate si trebuie evaluată în contextul antecedentelor clinice ale pacientului, precum si al altor teste de diagnosticare efectuate de către un patolog calificat.

Principiul de procedură

Tehnicile de colorare imunohistochimică (IHC) permit vizualizarea antigenilor prin aplicarea secventială a unui anumit anticorp pe antigen (anticorp primar), a unui anticorp secundar pe anticorpul primar și a unui complex enzimatic cu un substrat cromogen, cu etape de spălare intercalate. Activarea enzimatică a cromogenului duce la un produs de reacție vizibil la locul aplicării antigenului. Specimenul poate fi apoi contracolorat și acoperit cu lamelă. Rezultatele sunt interpretate folosind un microscop optic și ajută la diagnosticul diferențial al proceselor patofiziologice, care pot sau nu să fie asociate cu un anumit antigen.

Clonă

LN10

Imunogen

Proteină recombinantă procariotică corespunzând regiunii C-terminale a moleculei CD3 umane.

Specificitate

Antigen CD3 uman.

Compoziția reactivului

NCL-L-CD3-565 este un supernatant de cultură tisulară lichid care conține azidă de sodiu drept conservant.

Clasa Ig

IgG1

Concentrație proteină totală Total Protein

Consultați eticheta flaconului pentru concentrația proteinelor totale specifică lotului.

Concentrație anticorpi

Mai mare sau egală cu 32 mg/L, așa cum este determinată prin ELISA. Consultați eticheta flaconului pentru concentrația Ig specifică lotului.

Recomandări privind utilizarea

Imunohistochimie pe sectiuni de parafină.

Recuperarea indusă de căldură a epitopilor (HIER): Urmați instrucțiunile de utilizare din Novocastra Epitope Retrieval Solution pH 6. Diluție sugerată: 1:500 timp de 30 de minute la 25 °C. Aceste informații sunt furnizate cu rol de îndrumare, iar utilizatorii trebuie să-și stabilească singuri propriile diluții de lucru optime.

Vizualizare: Respectați instrucțiunile de utilizare din Novolink™ Polymer Detection Systems. Pentru informații suplimentare despre produs sau asistență, luați legătura cu distribuitorul dvs. local sau cu biroul regional al Leica Biosystems sau, ca alternativă, vizitați siteul web al Leica Biosystems, www.LeicaBiosystems.com

Performanta acestui anticorp trebuie validată atunci când este utilizat cu alte sisteme de colorare manuală sau alte platforme automatizate.

Depozitare și stabilitate

A se depozita la 2-8 °C. A nu se congela. A se returna la 2-8 °C imediat după utilizare. A nu se utiliza după data expirării indicată pe eticheta flaconului. Alte condiții de depozitare decât cele specificate mai sus trebuie verificate de către utilizator.

Pregătirea specimenului

Mediul de fixare recomandat este formalină tamponată neutru 10% pentru secțiunile de tesut încorporate în parafină.

Avertismente și precauții

Acest reactiv a fost pregătit din supernatantul culturii celulare. Întrucât este un produs biologic, trebuie să se actioneze cu prudență rezonabilă la manipularea sa.

Acest reactiv conține azidă de sodiu. O Fișă tehnică de securitate a materialului este disponibilă la cerere sau pe site-ul www. LeicaBiosystems.com

Consultati reglementările nationale sau locale pentru informatii privind eliminarea tuturor componentelor potential toxice.

Specimenele, înainte și după fixare, precum și toate materialele expuse la acestea, trebuie manevrate ca și când ar avea potențialul de a transmite infectii si trebuie eliminate luând măsurile de precautie adecvate. Nu pipetati niciodată reactivii pe gură si evitati contactul reactivilor și specimenelor cu pielea și mucoasele. Dacă reactivii sau probele vin în contact cu suprafețele sensibile, spălați cu apă din abundentă. Solicitati asistentă medicală.

Reduceti la minimum contaminarea microbiană a reactivilor, în caz contrar poate apărea o crestere a colorării nespecifice.

Timpii sau temperaturile de incubație care diferă de valorile specificate pot genera rezultate eronate. Orice astfel de modificări trebuie validate de către utilizator.

Controlul calității

Diferențele în ceea ce privește procesarea țesutului și procedurile tehnice în laboratorul utilizatorului pot cauza o variabilitate semnificativă a rezultatelor, necesitând efectuarea cu regularitate de controale interne, în plus față de următoarele proceduri. Probele de control trebuie să fie probe proaspete de autopsie/biopsie/chirurgicale, fixate în formalină, procesate și încorporate în ceară de parafină cât mai curând posibil și în aceeași manieră ca și probele pacientului.

Tesutul de control pozitiv

Folosit pentru a indica țesuturile pregătite corect și tehnicile de colorare adecvate.

O probă de țesut de control pozitiv trebuie să fie inclusă pentru fiecare set de condiții de testare în fiecare etapă de colorare. Un țesut cu colorare pozitivă slabă este mai adecvat decât un țesut cu colorare pozitivă puternică în vederea unui control optim al calității și pentru a detecta nivelurile minore de degradare a reactivului.²

Tesutul de control pozitiv recomandat este de amigdale.

Dacă țesutul de control pozitiv nu demonstrează colorația pozitivă, rezultatele obținute cu acele probe de testare trebuie considerate nevalide.

Tesutul de control negativ

Trebuie examinat după țesutul de control pozitiv pentru a verifica specificitatea informațiilor de etichetare ale antigenului țintă în funcție de anticorpul primar.

Țesutul de control negativ recomandat este mușchiul scheletic.

Ca alternativă, varietatea de tipuri diferite de celule prezente în majoritatea secțiunilor tisulare oferă frecvent locuri de control negativ, dar acest lucru trebuie verificat de către utilizator.

Colorația nespecifică, dacă este prezentă, are, de obicei, un aspect difuz. Colorația sporadică a țesutului conjunctiv poate fi observată, de asemenea, în secțiuni de țesuturi fixate în mod excesiv în formalină. Folosiți celule intacte pentru interpretarea rezultatelor de colorare. Celulele necrotice sau degenerate se colorează deseori într-un mod nespecific. Se pot observa rezultate fals pozitive ca urmare a legării non-imunologice a proteinelor sau produșilor de reacție ai substratului. Acestea pot fi cauzate, de asemenea, de enzimele endogene precum pseudoperoxidaza (eritrocite), peroxidaza endogenă (citocromul C) sau biotina endogenă (de exemplu, ficat, sân, creier, rinichi), în funcție de tipul de imunocolorație folosit. Pentru a diferenția activitatea enzimelor endogene sau legarea nespecifică a enzimelor de imunoreactivitatea specifică, pot fi colorate țesuturi suplimentare de la pacient numai cu substrat-cromogen sau, respectiv, complexe enzimatice (avidină-biotină, streptavidină, polimer etichetat) și substrat-cromogen. În cazul în care colorația specifică are loc în țesutul de control negativ, rezultatele obținute pe probele pacientului trebuie să fie considerate nevalide.

Reactivul de control negativ

Folosiți un reactiv de control negativ non-specific în locul anticorpului primar cu o secțiune din fiecare specimen al pacientului pentru a evalua colorația nespecifică și a permite o mai bună interpretare a colorării specifice la situl antigenului.

Ţesutul pacientului

Examinați specimenele pacientului colorate cu NCL-L-CD3-565 ultimele. Intensitatea colorației pozitive trebuie evaluată în contextul oricărei colorații de fond nespecifice a reactivului de control negativ. La fel ca în cazul oricărui test imunohistochimic, un rezultat negativ înseamnă că antigenul nu a fost detectat, și nu că antigenul a fost absent în celulele/țesuturile analizate. Dacă este necesar, folosiți un panel pentru anticorpi pentru identificarea reacțiilor fals negative.

Rezultate așteptate

Tesuturi normale

Clona LN10 a detectat antigenul CD3 în celule T din splină, nod limfatic și amigdală și în celulele T infiltrate din diverse alte țesuturi. (Numărul total al cazurilor normale evaluate = 55).

Tesuturi anormale

Clona LN10 a colorat 16/280 țesuturi anormale evaluate, incluzând malignități hematologice (16/130, incluzând 4/4 limfoame limfoblastice T, 4/5 limfoame periferice cu celule T, 2/8 limfoame anaplastice cu celule mari, 2/2 limfoame cu celule NK/T, 1/1 limfom malign cu celule T, 1/1 limfom difuz cu celule T mari, 1/1 limfom angioimunoblastic cu celule T, 1/1 timom, 0/50 limfoame difuze cu celule B, 0/15 limfoame Hodgkin, 0/11 limfoame foliculare, 0/9 limfoame difuze cu celule B mari, 0/5 limfoame MALT, 0/3 limfoame cu celule de manta, 0/3 limfoame difuze cu celule mari, 0/2 limfoame de zonă marginală, 0/2 limfoame Burkitt, 0/2 limfoame limfocitare plasmacitoide, 0/2 limfoame nespecificate cu celule T, 0/1 limfom difuz cu celule T, 0/1 limfom non-Hodgkin și 0/1 limfom similar Burkitt), tumori ale pielii (0/81, incluzând 0/18 melanoame, 0/15 carcinoame cu celule scuamoase, 0/15 carcinoame cu celule bazale, 0/10 carcinoame ale glandei sudoripare, 0/9 dermatofibrosarcoame, 0/3 adenocarcinoame metastatice, 0/3 schwannoame maligne, 0/2 carcinoame chistice adenoide, 0/1 adenocarcinom sebaceu, 0/1 fibrosarcom, 0/1 sarcom pleomorfic nediferențiat, 0/1 leiomiosarcoma, 0/1 fibroxantom atipic și 0/1 tumoare cu celule Merkel), tumori ale țesuturilor moi (0/8), carcinoame mamare (0/7), tumori ovariene (0/7), tumori pulmonare (0/7), tumori hepatice (0/5), tumori neuroendocrine (0/4), carcinoame renale (0/4), tumori gastrice (0/3), carcinoame ale vezicii urinare (0/3), tumori cu celule germinale (0/3), tumori suprarenale (0/3), tumori endometriale (0/3), tumori pancreatice (0/2), tumori tiroidiene (0/2), adenocarcinoame de prostată (0/2), adenocarcinoame ale colon (0/1), carcinom al intestinului subţire (0/1), carcinoame cu celule scuamoase ale penisului (0/1), carcinom cu celule scuamoase al coului uterin (0/1), ganglioneurom (0/1) și hiperplazie de prostată (0/1). (Numărul total al cazurilor anormale evaluate = 280).

NCL-L-CD3-565 este recomandat pentru a fi utilizat ca parte a unui panel de anticorpi pentru a indica fenotipul celulelor T în tulburări limfoprofilerative.

Limitări generale

Imunohistochimia este un proces de diagnostic cu mai multe etape, care constă din instruirea specializată în ceea ce privește alegerea reactivilor adecvați; alegerea, fixarea și procesarea țesutului; prepararea lamei IHC; și interpretarea rezultatelor de colorare. Colorarea tisulară depinde de manipularea și procesarea țesutului înainte de colorare. Fixarea, congelarea, dezghețarea, spălarea, uscarea, încălzirea, secționarea necorespunzătoare sau contaminarea cu alte țesuturi ori fluide pot cauza artefacte, captura anticorpilor sau rezultate fals negative. Rezultatele inconsecvente pot fi atribuite diferențelor în ceea ce privește metodele de fixare și încorporare, ori neregularităților inerente ale țesutului.⁴

Contracolorația excesivă sau incompletă poate compromite interpretarea adecvată a rezultatelor.

Interpretarea clinică a oricărei colorări sau a absenței acesteia trebuie completată cu studii morfologice utilizând controale adecvate și trebuie evaluată în contextul antecedentelor clinice ale pacientului, precum și al altor teste de diagnosticare efectuate de către un patolog calificat.

Anticorpii de la Leica Biosystems Newcastle Ltd sunt destinați utilizării, conform indicațiilor, fie pe secțiuni congelate, fie pe secțiuni încorporate în parafină cu cerințe de fixare specifice. Poate apărea exprimarea neașteptată a antigenului, în special în neoplasme. Interpretarea clinică a oricărei secțiuni tisulare colorate trebuie să includă analiza morfologică și evaluarea probelor de control adecvate.

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Amendamente la ediția anterioară

Compoziția reactivilor, Concentrația totală a proteinelor, Recomandări de utilizare, Avertizări și măsuri de precauție, Rezultate preconizate.

Data publicării

01 noiembrie 2018

Anticorpul primar gata de utilizare BOND™ CD4 (4B12)

Nr. catalog: PA0427

Utilizare prevăzută

Acest reactiv este destinat utilizării pentru diagnosticare in vitro.

Anticorpul monoclonal CD4 (4B12) este destinat utilizării pentru identificarea calitativă, prin intermediul microscopiei optice, a antigenului CD4 uman din țesut fixat în formalină, încorporat în parafină, prin colorare imunohistochimică utilizând sistemul automat BOND (care include sistemul Leica BOND-MAX și sistemul Leica BOND-III).

Interpretarea clinică a oricărei colorații sau a absenței acesteia trebuie verificată prin studii morfologice, folosind proceduri de control adecvate, și trebuie evaluată în contextul antecedentelor clinice ale pacientului, precum și al altor teste de diagnosticare efectuate de către un patolog calificat.

Rezumat și explicație

Pot fi utilizate tehnici imunohistochimice pentru a demonstra prezența antigenilor în țesut și celule (a se vedea "Utilizarea reactivilor BOND" din documentația de utilizare BOND). Anticorpul primar CD4 (4B12) este un produs gata de utilizare care a fost optimizat în mod specific pentru utilizarea cu BOND Polymer Refine Detection. Demonstrarea prezenței antigenului CD4 uman este realizată mai întâi prin permiterea legării CD4 (4B12) la secțiune și apoi prin vizualizarea acestei legări utilizând reactivii furnizați în sistemul de detecție. Utilizarea acestor produse, în combinație cu sistemul automat BOND (care include sistemul Leica BOND-MAX și sistemul Leica BOND-III), reduce posibilitatea producerii erorii umane și variabilitatea inerentă care rezultă din diluția individuală a reactivului, pipetarea manuală și aplicarea reactivului.

Reactivi furnizați

CD4 (4B12) este un anticorp monoclonal anti-uman de șoarece produs ca supernatant de cultură tisulară și furnizat în soluție salină tamponată cu trometamină cu proteină purtătoare, care conține 0,35 % ProClin™ 950 drept conservant.

Volum total = 7 ml.

Clonă

4B12

Imunogen

Proteină procariotă recombinantă corespunzând domeniului extern al moleculei CD4.

Specificitate

Antigen CD4 uman.

Clasa Ig

lgG1

Concentratie proteină totală

Aproximativ 10 mg/ml.

Concentrație anticorpi

Mai mare sau egală cu 6,6 mg/L, așa cum este determinată prin ELISA.

Diluare și amestecare

Anticorpul primar CD4 (4B12) este diluat în mod optim pentru utilizare pe sistemul BOND (care include sistemul Leica BOND-MAX și sistemul Leica BOND-III). Reconstituirea, amestecarea, diluarea sau titrarea acestui reactiv nu sunt necesare.

Materiale necesare, dar care nu sunt furnizate

Consultați "Utilizarea reactivilor BOND" din documentația dumneavoastră de utilizare a sistemului BOND pentru o listă completă a materialelor necesare pentru tratarea probelor și colorația imunohistochimică utilizând sistemul BOND (care include sistemul Leica BOND-MAX și sistemul Leica BOND-III).

Depozitare și stabilitate

A se depozita la 2 $-8\,^{\circ}$ C. A nu se utiliza după data expirării indicată pe eticheta recipientului.

Semnele care indică contaminarea şi/sau instabilitatea CD4 (4B12) sunt: turbiditatea soluției, formarea de mirosuri și prezența precipitatului.

A se returna la 2-8 °C imediat după utilizare.

Alte condiții de depozitare decât cele specificate mai sus trebuie verificate de către utilizator1.

Precauții

- · Acest produs este destinat utilizării pentru diagnosticare in vitro.
- Concentraţia de ProClin™ 950 este 0,35 %. Acesta conţine ingredientul activ 2-metil-4-izotiazolin-3-ona şi poate cauza iritarea pielii, ochilor, membranelor mucoase şi tractului respirator superior. Purtaţi mănuşi de unică folosinţă atunci când manipulaţi reactivii.
- Pentru a obține o copie a fișei tehnice de securitate pentru material, luați legătura cu distribuitorul dvs. local sau cu biroul regional al Leica Biosystems sau, ca alternativă, vizitați site-ul web al Leica Biosystems, www.LeicaBiosystems.com

- Specimenele, înainte și după fixare, precum și toate materialele expuse la acestea, trebuie manipulate ca și când ar avea potențialul
 de a transmite infecții și trebuie eliminate luând măsurile de precauție adecvate². Nu pipetați niciodată reactivii cu gura și evitați
 contactul reactivilor și probelor cu pielea și membranele mucoase. Dacă reactivii sau probele vin în contact cu suprafețele sensibile,
 spălați cu apă din abundență. Solicitați asistență medicală.
- Consultaţi reglementările naţionale, judeţene sau locale pentru informaţii privind eliminarea oricăror componente cu potenţial toxic.
- · Reduceți la minimum contaminarea microbiană a reactivilor, în caz contrar poate apărea o crestere a colorării nespecifice.
- Timpii sau temperaturile de recuperare, incubare care diferă de valorile specificate pot genera rezultate eronate. Orice astfel de modificare trebuie validată de către utilizator.

Instrucțiuni de utilizare

Anticorpul primar CD4 (4B12) a fost dezvoltat pentru utilizarea pe sistemul automat BOND (care include sistemul Leica BOND-MAX și sistemul Leica BOND-III) în combinație cu BOND Polymer Refine Detection. Protocolul de colorare recomandat pentru anticorpul primar CD4 (4B12) este IHC Protocol F. Se recomandă recuperarea indusă de căldură a epitopilor utilizând BOND Epitope Retrieval Solution 2 timp de 20 de minute.

Rezultate așteptate

Tesuturi normale

Clona 4B12 a detectat antigenul CD4 pe suprafața celulară a sub-populației ajutătoare/inductoare de celule T normale și în endoteliul sinusoidal al ficatului. (Numărul total al cazurilor normale = 97).

Tesuturi tumorale

Clona 4B12 a colorat 4/7 limfoame anaplastice cu celule T mari, 2/4 limfoame angioimunoblastice cu celule T, 1/3 limfoame cu celule NK/T, 1/1 limfom periferic cu celule T și 1/1 limfom cu celule T. Cu excepția celulelor T infiltrate, nu s-a observat vreo colorare în limfoame difuze cu celule B mari (0/107), limfoame limfocitare cronice (0/11), limfoame foliculare (0/11), boala lui Hodgkin (0/11), limfoame cu celule de manta (0/7), un limfom limfoblastic acut cu celule B (0/1), un limfom limfoblastic acut cu celule B/T primitive (0/1), un limfom de zonă marginală (0/1), tumori ale tiroidei (0/4), tumori pulmonare (0/4), tumori hepatice (0/4), tumori ovariene (0/4), tumori cerebrale (0/2), tumori ale esofagului (0/2), tumori mamare (0/2), tumori gastrice (0/2), tumori ale țesuturilor moi (0/2), tumori ale limbii (0/2), tumori metastatice de origine necunoscută (0/2), tumori renale (0/2) tumori ale colului uterin (0/2), tumori testiculare (0/2), tumori ale colonului (0/2), tumori ale rectului (0/2), tumori ale pielii (0/2), o tumoare a laringelui (0/1) sau o tumoare a timusului (0/1). (Numărul total al cazurilor anormale evaluate = 210).

CD4 (4B12) este recomandat pentru utilizare ca parte a unui panel pentru anticorpi pentru a ajuta la caracterizarea tulburărilor celulelor T.

Restrictii specifice produsului

CD4 (4B12) a fost optimizat la Leica Biosystems pentru utilizarea cu BOND Polymer Refine Detection și cu reactivii auxiliari BOND. Utilizatorii care se abat de la procedurile de testare recomandate trebuie să accepte responsabilitatea pentru interpretarea rezultatelor pacientului în aceste circumstanțe. Timpii protocolului pot varia, datorită variației în fixarea țesutului și eficacității intensificării antigenului, și trebuie să fie determinați empiric. Atunci când se optimizează condițiile de recuperare și timpii protocolului, trebuie să fie utilizați reactivi de control negativ.

Rezolvarea problemelor

Consultați referința 3 pentru acțiuni de remediere.

Contactați distribuitorul dumneavoastră local sau biroul regional al Leica Biosystems pentru raportarea colorării neobișnuite.

Informații suplimentare

Informații suplimentare referitoare la imunocolorarea cu reactivii BOND, sub titlurile Principiul procedurii, Materiale necesare, Pregătirea specimenului, Controlul calității, Verificarea analizei, Interpretarea colorării, Codul simbolurilor de pe etichete și Limitări generale pot fi găsite în "Utilizarea reactivilor BOND" din documentația dumneavoastră de utilizare a sistemului BOND.

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Data publicării

05 decembrie 2018

Novocastra[™] Anticorp monoclonal lichid de șoarece CD8

Cod produs: NCL-L-CD8-4B11

Utilizare prevăzută

Pentru diagnosticare in vitro.

NCL-L-CD8-4B11 este destinat identificării calitative, prin intermediul microscopiei optice, a antigenului CD8 uman în secțiunile de parafină. Interpretarea clinică a oricărei colorări sau a absenței acesteia trebuie completată cu studii morfologice utilizând controale adecvate și trebuie evaluată în contextul antecedentelor clinice ale pacientului, precum și al altor teste de diagnosticare efectuate de către un patolog calificat.

Principiul de procedură

Tehnicile de colorare imunohistochimică (IHC) permit vizualizarea antigenilor prin aplicarea secvențială a unui anumit anticorp pe antigen (anticorp primar), a unui anticorp secundar pe anticorpul primar și a unui complex enzimatic cu un substrat cromogen, cu etape de spălare intercalate. Activarea enzimatică a cromogenului duce la un produs de reacție vizibil la locul aplicării antigenului. Specimenul poate fi apoi contracolorat și acoperit cu lamelă. Rezultatele sunt interpretate folosind un microscop optic și ajută la diagnosticul diferențial al proceselor patofiziologice, care pot sau nu să fie asociate cu un anumit antigen.

Clonă

4B11

Imunogen

Peptidă corespunzând porțiunii citoplasmice a lanțului alfa al moleculei CD8 umane.

Specificitate

Antigen CD8 uman.

Compoziția reactivului

NCL-L-CD8-4B11 este un supernatant de cultură tisulară lichid care conține azidă de sodiu drept conservant.

Clasa Ig

IgG2b

Concentrație proteină totală

Total Protein

Consultați eticheta flaconului pentru concentrația proteinelor totale specifică lotului.

Concentrație anticorpi

Mai mare sau egală cu 28,5 mg/L, așa cum este determinată prin ELISA. Consultați eticheta flaconului pentru concentrația Ig specifică lotului.

Recomandări privind utilizarea

Imunohistochimie pe secțiuni de parafină.

Recuperarea indusă de căldură a epitopilor (HIER): Urmați instrucțiunile de utilizare din Novocastra Epitope Retrieval Solution pH 9. Diluție sugerată: 1:50 timp de 30 minute la 25 °C. Aceste informații sunt furnizate cu rol de îndrumare, iar utilizatorii trebuie să-și stabilească singuri propriile diluții de lucru optime.

Vizualizare: Respectați instrucțiunile de utilizare din Novolink™ Polymer Detection Systems. Pentru informații suplimentare despre produs sau asistență, luați legătura cu distribuitorul dvs. local sau cu biroul regional al Leica Biosystems sau, ca alternativă, vizitați site-ul web al Leica Biosystems, www.LeicaBiosystems.com

Performanța acestui anticorp trebuie validată atunci când este utilizat cu alte sisteme de colorare manuală sau alte platforme automatizate.

Depozitare și stabilitate

A se depozita la 2–8 °C. A nu se congela. A se returna la 2–8 °C imediat după utilizare. A nu se utiliza după data expirării indicată pe eticheta flaconului. Alte condiții de depozitare decât cele specificate mai sus trebuie verificate de către utilizator.

Pregătirea specimenului

Mediul de fixare recomandat este formalină tamponată neutru 10% pentru secțiunile de țesut încorporate în parafină.

Avertismente și precauții

Acest reactiv a fost pregătit din supernatantul culturii celulare. Întrucât este un produs biologic, trebuie să se acționeze cu prudență rezonabilă la manipularea sa.

Acest reactiv conține azidă de sodiu. Fișa cu informații de siguranță despre material este disponibilă la cerere sau poate fi obținută de pe site-ul www,LeicaBiosystems,com

Consultați reglementările naționale, județene sau locale pentru informații privind eliminarea tuturor componentelor potențial toxice. Specimenele, înainte și după fixare, precum și toate materialele expuse la acestea, trebuie manipulate ca și când ar avea potențialul de a transmite infecții și trebuie eliminate la deșeuri luând măsurile de precauție adecvate.¹ Nu pipetați niciodată reactivii pe gură și evitați contactul reactivilor și specimenelor cu pielea și membranele mucoase. Dacă reactivii sau probele vin în contact cu suprafețele sensibile, spălati cu apă din abundentă. Solicitati asistentă medicală.

Reduceti la minimum contaminarea microbiană a reactivilor, în caz contrar poate apărea o crestere a colorării nespecifice.

Timpii sau temperaturile de incubație care diferă de valorile specificate pot genera rezultate eronate. Orice astfel de modificări trebuie validate de către utilizator.

Controlul calității

Diferențele în ceea ce privește procesarea țesutului și procedurile tehnice în laboratorul utilizatorului pot cauza o variabilitate semnificativă a rezultatelor, necesitând efectuarea cu regularitate de controale interne, în plus față de următoarele proceduri. Probele de control trebuie să fie probe proaspete de autopsie/biopsie/chirurgicale, fixate în formalină, procesate și încorporate în ceară de parafină cât mai curând posibil și în aceeași manieră ca și probele pacientului.

Tesutul de control pozitiv

Folosit pentru a indica tesuturile pregătite corect și tehnicile de colorare adecvate.

O probă de țesut de control pozitiv trebuie să fie inclusă pentru fiecare set de condiții de testare în fiecare etapă de colorare. Un țesut cu colorare pozitivă slabă este mai adecvat decât un țesut cu colorare pozitivă puternică în vederea unui control optim al calității și pentru a detecta nivelurile minore de degradare a reactivului.²

Țesutul de control pozitiv recomandat este de amigdale.

Dacă țesutul de control pozitiv nu demonstrează colorația pozitivă, rezultatele obținute cu acele probe de testare trebuie considerate nevalide.

Tesutul de control negativ

Trebuie examinat după țesutul de control pozitiv pentru a verifica specificitatea informațiilor de etichetare ale antigenului țintă în funcție de anticorpul primar.

Țesutul de control negativ recomandat este mușchiul scheletic.

Ca alternativă, varietatea de tipuri diferite de celule prezente în majoritatea secțiunilor tisulare oferă frecvent locuri de control negativ, dar acest lucru trebuie verificat de către utilizator.

Colorația nespecifică, dacă este prezentă, are, de obicei, un aspect difuz. Colorația sporadică a țesutului conjunctiv poate fi observată, de asemenea, în secțiuni de țesuturi fixate în mod excesiv în formalină. Folosiți celule intacte pentru interpretarea rezultatelor de colorare. Celulele necrotice sau degenerate se colorează deseori într-un mod nespecific. Se pot observa rezultate fals pozitive ca urmare a legării non-imunologice a proteinelor sau produșilor de reacție ai substratului. Acestea pot fi cauzate, de asemenea, de enzimele endogene precum pseudoperoxidaza (eritrocite), peroxidaza endogenă (citocromul C) sau biotina endogenă (de exemplu, ficat, sân, creier, rinichi), în funcție de tipul de imunocolorație folosit. Pentru a diferenția activitatea enzimelor endogene sau legarea nespecifică a enzimelor de imunoreactivitatea specifică, pot fi colorate țesuturi suplimentare de la pacient numai cu substrat-cromogen sau, respectiv, complexe enzimatice (avidină-biotină, streptavidină, polimer etichetat) și substrat-cromogen. În cazul în care colorația specifică are loc în țesutul de control negativ, rezultatele obținute pe probele pacientului trebuie să fie considerate nevalide.

Reactivul de control negativ

Folosiți un reactiv de control negativ non-specific în locul anticorpului primar cu o secțiune din fiecare specimen al pacientului pentru a evalua colorația nespecifică și a permite o mai bună interpretare a colorării specifice la situl antigenului.

Ţesutul pacientului

Examinați specimenele pacientului colorate cu NCL-L-CD8-4B11 ultimele. Intensitatea colorației pozitive trebuie evaluată în contextul oricărei colorații de fond nespecifice a reactivului de control negativ. La fel ca în cazul oricărui test imunohistochimic, un rezultat negativ înseamnă că antigenul nu a fost detectat, și nu că antigenul a fost absent în celulele/țesuturile analizate. Dacă este necesar, folosiți un panel pentru anticorpi pentru identificarea reacțiilor fals negative.

Rezultate așteptate

Tesuturi normale

Clona 4B11 detectează antigenul CD8 pe suprafața celulară a sub-populației citotoxice de celule T în timus, splină, ganglioni limfatici și amigdală. (Numărul total al cazurilor normale evaluate = 44).

Tesuturi anormale

Clona 4B11 a colorat 3/4 limfoame angioimunoblastice cu celule T. Cu excepția celulelor T reactive, nu a fost detectată vreo colorare în limfoame difuze cu celule B mari (0/108), limfoame limfocitare cronice (0/12), limfoame foliculare (0/11), boala lui Hodgkin (0/11), limfoame cu celule B mari (0/7), limfoame anaplastice cu celule T cu celule mari (0/7), limfoame cu celule T/NK (0/3), un limfom limfoblastic acut cu celule B/T (0/1), un limfom cu celule T periferice (0/1), un limfom cu celule T (0/1), un limfom al zonei marginale (0/1), tumori tiroidiene (0/4), tumori pulmonare (0/4), tumori ovariene (0/4), tumori hepatice (0/4), tumori cerebrale (0/2), tumori esofagiene (0/2), tumori mamare (0/2), tumori gastrice (0/2), tumori ale țesuturilor moi (0/2), tumori ale limbii (0/2), tumori metastatice de origine necunoscută (0/2), tumori renale (0/2), tumori cervicale (0/2), tumori testiculare (0/2), tumori ale colonului (0/2), tumori rectale (0/2), tumori ale pielii (0/2), o tumoră a laringelui (0/1) și o tumoră a timusului (0/1). (Numărul total al cazurilor tumorale evaluate = 212).

NCL-L-CD8-4B11 este recomandat pentru utilizare ca parte a unui panel de anticorpi pentru a ajuta la caracterizarea tulburărilor celulelor T.

Limitări generale

Imunohistochimia este un proces de diagnostic cu mai multe etape, care constă din instruirea specializată în ceea ce privește alegerea reactivilor adecvați; alegerea, fixarea și procesarea țesutului; prepararea lamei IHC; și interpretarea rezultatelor de colorare. Colorarea tisulară depinde de manipularea și procesarea țesutului înainte de colorare. Fixarea, congelarea, dezghețarea, spălarea, uscarea, încălzirea, secționarea necorespunzătoare sau contaminarea cu alte țesuturi ori fluide pot cauza artefacte, captura anticorpilor sau rezultate fals negative. Rezultatele inconsecvente pot fi atribuite diferențelor în ceea ce privește metodele de fixare și încorporare, ori neregularităților inerente ale țesutului.⁴

Contracolorația excesivă sau incompletă poate compromite interpretarea adecvată a rezultatelor.

Interpretarea clinică a oricărei colorări sau a absenței acesteia trebuie completată cu studii morfologice utilizând controale adecvate și trebuie evaluată în contextul antecedentelor clinice ale pacientului, precum și al altor teste de diagnosticare efectuate de către un patolog calificat.

Anticorpii de la Leica Biosystems Newcastle Ltd sunt destinați utilizării, conform indicațiilor, fie pe secțiuni congelate, fie pe secțiuni încorporate în parafină cu cerințe de fixare specifice. Poate apărea exprimarea neașteptată a antigenului, în special în neoplasme. Interpretarea clinică a oricărei secțiuni tisulare colorate trebuie să includă analiza morfologică și evaluarea probelor de control adecvate.

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Amendamente la ediția anterioară

Compoziția reactivilor, Concentrația totală a proteinelor, Recomandări de utilizare, Avertizări și măsuri de precauție, Rezultate preconizate.

Data publicării

05 octombrie 2018

Novocastra[™] Anticorp lichid monoclonal de șoarece CD20

Cod produs: NCL-L-CD20-L26

Utilizare prevăzută

Pentru diagnosticare in vitro.

NCL-L-CD20-L26 este destinat identificării calitative, prin intermediul microscopiei optice, a moleculelor de CD20 în secțiunile de parafină. Interpretarea clinică a oricărei colorări sau a absenței acesteia trebuie completată cu studii morfologice utilizând controale adecvate și trebuie evaluată în contextul antecedentelor clinice ale pacientului, precum și al altor teste de diagnosticare efectuate de către un patolog calificat.

Principiul de procedură

Tehnicile de colorare imunohistochimică (IHC) permit vizualizarea antigenilor prin aplicarea secvențială a unui anumit anticorp pe antigen (anticorp primar), a unui anticorp secundar pe anticorpul primar și a unui complex enzimatic cu un substrat cromogen, cu etape de spălare intercalate. Activarea enzimatică a cromogenului duce la un produs de reacție vizibil la locul aplicării antigenului. Specimenul poate fi apoi contracolorat și acoperit cu lamelă. Rezultatele sunt interpretate folosind un microscop optic și ajută la diagnosticul diferențial al proceselor patofiziologice, care pot sau nu să fie asociate cu un anumit antigen.

Clonă

L26

Imunogen

Celule B de amigdale umane.

Specificitate

Un epitop intracitoplasmatic localizat pe molecula CD20 umană. Reacționează predominant cu o polipeptidă 33 kD, dar și cu o componentă minoră a 30 kD.

Compoziția reactivului

NCL-L-CD20-L26 este un supernatant de cultură tisulară lichid care conține azidă de sodiu drept conservant.

Clasa Iq

IgG2a, kappa

Concentrație proteină totală Total Protein

Consultați eticheta flaconului pentru concentrația proteinelor totale specifică lotului.

Concentratie anticorpi

Mai mare sau egală cu 95 mg/L, așa cum este determinată prin ELISA. Consultați eticheta flaconului pentru concentrația Ig specifică lotului.

Recomandări privind utilizarea

Imunohistochimie pe secțiuni de parafină.

Recuperarea indusă de căldură a epitopilor (HIER): Urmați instrucțiunile de utilizare din Novocastra Epitope Retrieval Solution pH 6. Diluție sugerată: 1:100-1:200 timp de 30 minute la 25 °C. Aceste informații sunt furnizate cu rol de îndrumare, iar utilizatorii trebuie săși stabilească singuri propriile diluții de lucru optime.

Vizualizare: Respectați instrucțiunile de utilizare din Novolink™ Polymer Detection Systems. Pentru asistență sau informații suplimentare cu privire la produs, luați legătura cu distribuitorul dvs. local sau cu biroul regional al Leica Biosystems sau, ca alternativă, vizitați site-ul web al Leica Biosystems, www.LeicaBiosystems.com.

Eficiența acestui anticorp trebuie validată atunci când este utilizat cu alte sisteme de colorare manuală sau alte platforme automatizate.

Depozitare și stabilitate

A se depozita la 2–8 °C. A nu se congela. A se returna la 2–8 °C imediat după utilizare. A nu se utiliza după data expirării indicată pe eticheta flaconului. Alte condiții de depozitare decât cele specificate mai sus trebuie verificate de către utilizator.

Pregătirea specimenului

Mediul de fixare recomandat este formalină tamponată neutru 10% pentru secțiunile de țesut încorporate în parafină.

Avertismente și precauții

Acest reactiv a fost pregătit din supernatantul culturii celulare. Întrucât este un produs biologic, trebuie să se acționeze cu prudență rezonabilă la manipularea sa.

Acest reactiv conține azidă de sodiu. O Fișă tehnică de securitate a materialului este disponibilă la cerere sau pe site-ul www. LeicaBiosystems.com

Consultați reglementările naționale sau locale pentru informații privind eliminarea la deșeuri a tuturor componentelor potențial toxice. Probele, înainte și după fixare, precum și toate materialele expuse la acestea, trebuie manipulate ca și când ar avea potențialul de a transmite infecții și trebuie eliminate la deșeuri luând măsurile de precauție adecvate.¹ Nu pipetați niciodată reactivii cu gura și evitați contactul reactivilor și probelor cu pielea și membranele mucoase. Dacă reactivii sau probele vin în contact cu suprafețele sensibile, spălați cu apă din abundență. Solicitați asistență medicală.

Reduceți la minimum contaminarea microbiană a reactivilor, în caz contrar poate apărea o creștere a colorării nespecifice.

Timpii sau temperaturile de incubație care diferă de valorile specificate pot genera rezultate eronate. Orice astfel de modificări trebuie validate de către utilizator.

Controlul calității

Diferențele în ceea ce privește procesarea țesutului și procedurile tehnice în laboratorul utilizatorului pot cauza o variabilitate semnificativă a rezultatelor, necesitând efectuarea cu regularitate de controale interne, în plus față de următoarele proceduri. Probele de control trebuie să fie probe proaspete de autopsie/biopsie/chirurgicale, fixate în formalină, procesate și încorporate în ceară de parafină cât mai curând posibil și în aceeași manieră ca și probele pacientului.

Țesutul de control pozitiv

Folosit pentru a indica tesuturile pregătite corect și tehnicile de colorare adecvate.

O probă de țesut de control pozitiv trebuie să fie inclusă pentru fiecare set de condiții de testare în fiecare etapă de colorare. Un țesut cu colorare pozitivă slabă este mai adecvat decât un țesut cu colorare pozitivă puternică în vederea unui control optim al calității și pentru a detecta nivelurile minore de degradare a reactivului.²

Tesutul de control pozitiv recomandat este de amigdale.

Dacă țesutul de control pozitiv nu demonstrează colorația pozitivă, rezultatele obținute cu acele probe de testare trebuie considerate nevalide.

Tesutul de control negativ

Trebuie examinat după țesutul de control pozitiv pentru a verifica specificitatea informațiilor de etichetare ale antigenului țintă în funcție de anticorpul primar.

Tesutul de control negativ recomandat este cerebelul.

Ca alternativă, varietatea de tipuri diferite de celule prezente în majoritatea secțiunilor tisulare oferă frecvent locuri de control negativ, dar acest lucru trebuie verificat de către utilizator.

Colorația nespecifică, dacă este prezentă, are, de obicei, un aspect difuz. Colorația sporadică a țesutului conjunctiv poate fi observată, de asemenea, în secțiuni de țesuturi fixate în mod excesiv în formalină. Folosiți celule intacte pentru interpretarea rezultatelor de colorare. Celulele necrotice sau degenerate se colorează deseori într-un mod nespecific. Se pot observa rezultate fals pozitive ca urmare a legării non-imunologice a proteinelor sau produșilor de reacție ai substratului. Acestea pot fi cauzate, de asemenea, de enzimele endogene precum pseudoperoxidaza (eritrocite), peroxidaza endogenă (citocromul C) sau biotina endogenă (de exemplu, ficat, sân, encefal, rinichi), în funcție de tipul de imunocolorație folosit. Pentru a diferenția activitatea enzimelor endogene sau legarea nespecifică a enzimelor de imunoreactivitatea specifică, pot fi colorate țesuturi suplimentare de la pacient numai cu substrat-cromogen sau, respectiv, complexe enzimatice (avidină-biotină, streptavidină, polimer etichetat) și substrat-cromogen. În cazul în care colorația specifică are loc în țesutul de control negativ, rezultatele obținute pe probele pacientului trebuie să fie considerate nevalide.

Reactivul de control negativ

Folosiți un reactiv de control negativ non-specific în locul anticorpului primar cu o secțiune din fiecare specimen al pacientului pentru a evalua colorația nespecifică și a permite o mai bună interpretare a colorării specifice la situl antigenului.

Ţesutul pacientului

Examinați specimenele pacientului colorate cu NCL-L-CD20-L26 ultimele. Intensitatea colorației pozitive trebuie evaluată în contextul oricărei colorații de fond nespecifice a reactivului de control negativ. La fel ca în cazul oricărui test imunohistochimic, un rezultat negativ înseamnă că antigenul nu a fost detectat, și nu că antigenul a fost absent în celulele/țesuturile analizate. Dacă este necesar, folosiți un panel pentru anticorpi pentru identificarea reacțiilor fals negative.

Rezultate așteptate

Tesuturi normale

Clona L26 detectează antigenul CD20 pe suprafața celulară a celulelor din linia de celule B, cu excepția celulelor plasmatice. (Numărul total al cazurilor normale evaluate = 96).

Tesuturi anormale

Clona L26 a colorat 105/106 limfoame difuze cu celule B mari, 11/11 limfoame foliculare, 10/11 limfoame limfocitare cronice, 2/11 boala lui Hodgkin, 7/7 limfoame cu celule de manta, 1/1 limfom limfoblastic acut cu celule B și 1/1 limfom de zonă marginală. Cu excepția celulelor B reactive, nu a fost observată vreo colorare în limfoame anaplastice cu celule T mari (0/7), limfoame angioimunoblastice cu celule T (0/4), limfoame cu celule T/NK (0/3), un limfom periferic cu celule T (0/1), un limfom cu celule T (0/1), un limfom limfoblastic acut cu celule B/T primitive (0/1), tumori cerebrale (0/2), tumori ale esofagului (0/2), tumori ale laringelui (0/1), tumori ale timusului (0/1), tumori tiroidiene (0/4), tumori mamare (0/2), tumori gastrice (0/2), tumori ale țesuturilor moi (0/2), tumori ale limbii (0/2), tumori pulmonare (0/4), tumori metastatice de origine necunoscută (0/2), tumori hepatice (0/4), tumori renale (0/2), tumori ovariene (0/4), tumori ale colului uterin (0/2), tumori testiculare (0/2), tumori ale colonului (0/2), tumori ale rectului (0/2) or tumori ale pielii (0/2). (Numărul total al cazurilor tumorale evaluate = 209).

NCL-L-CD20-L26 este recomandat pentru utilizare ca parte a unui panel de anticorpi pentru a ajuta în caracterizarea tulburărilor cu celule B.

Limitări generale

Imunohistochimia este un proces de diagnostic cu mai multe etape, care constă din instruirea specializată în ceea ce privește alegerea reactivilor adecvați; alegerea, fixarea și procesarea țesutului; prepararea lamei IHC; și interpretarea rezultatelor de colorare. Colorarea tisulară depinde de manipularea și procesarea țesutului înainte de colorare. Fixarea, congelarea, dezghețarea, spălarea, uscarea, încălzirea, secționarea necorespunzătoare sau contaminarea cu alte țesuturi ori fluide pot cauza artefacte, captura anticorpilor sau rezultate fals negative. Rezultatele inconsecvente pot fi atribuite diferențelor în ceea ce privește metodele de fixare și încorporare, ori neregularităților inerente ale țesutului.⁴

 $Contracolorația\ excesiv\\ {\tt incomplet}\\ {\tt incomplet}\\ {\tt incomplet}\\ {\tt interpretarea}\ {\tt adecvat}\\ {\tt incomplet}\\ {\tt interpretarea}\ {\tt adecvat}\\ {\tt incomplet}\\ {\tt interpretarea}\ {\tt adecvat}\\ {\tt int}\\ {\tt interpretarea}\ {\tt adecvat}\\ {\tt interpretarea}\ {\tt interpretarea}\\ {$

Interpretarea clinică a oricărei colorări sau a absenței acesteia trebuie completată cu studii morfologice utilizând controale adecvate și trebuie evaluată în contextul antecedentelor clinice ale pacientului, precum și al altor teste de diagnosticare efectuate de către un patolog calificat.

Anticorpii de la Leica Biosystems Newcastle Ltd sunt destinați utilizării, conform indicațiilor, fie pe secțiuni congelate, fie pe secțiuni încorporate în parafină cu cerințe de fixare specifice. Poate apărea exprimarea neașteptată a antigenului, în special în neoplasme. Interpretarea clinică a oricărei secțiuni tisulare colorate trebuie să includă analiza morfologică și evaluarea probelor de control adecvate.

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Amendamente la ediția anterioară

Compoziția reactivului, Recomandări de utilizare, Rezultate așteptate.

Data publicării

09 noiembrie 2018

Novocastra™ Liquid Mouse Monoclonal Antibody CD56 (NCAM)

Product Code: NCL-L-CD56-504

Intended Use

For in vitro diagnostic use.

NCL-L-CD56-504 is intended for the qualitative identification by light microscopy of human CD56 antigen (NCAM) in paraffin sections. The clinical interpretation of any staining or its absence should be complemented by morphological studies using proper controls and should be evaluated within the context of the patient's clinical history and other diagnostic tests by a qualified pathologist.

Principle of Procedure

Immunohistochemical (IHC) staining techniques allow for the visualization of antigens via the sequential application of a specific antibody to the antigen (primary antibody), a secondary antibody to the primary antibody and an enzyme complex with a chromogenic substrate with interposed washing steps. The enzymatic activation of the chromogen results in a visible reaction product at the antigen site. The specimen may then be counterstained and coverslipped. Results are interpreted using a light microscope and aid in the differential diagnosis of pathophysiological processes, which may or may not be associated with a particular antigen.

Clone

CD564

Immunogen

Prokaryotic recombinant protein corresponding to a region of the extracellular domain of the human CD56 molecule.

Specificity

Human CD56 antigen (NCAM).

Reagent Composition

NCL-L-CD56-504 is a liquid tissue culture supernatant containing sodium azide as a preservative.

Ig Class

lgG2b

Total Protein Concentration Total Protein



Refer to vial label for lot specific total protein concentration.

Antibody Concentration

Greater than or equal to 18 mg/L as determined by ELISA. Refer to vial label for lot specific lg concentration.

Recommendations On Use

Immunohistochemistry on paraffin sections.

Heat Induced Epitope Retrieval (HIER): Please follow the instructions for use in Novocastra Epitope Retrieval Solution pH 6. Suggested dilution: 1:100 for 30 minutes at 25 °C. This is provided as a guide and users should determine their own optimal working

Visualization: Please follow the instructions for use in the Novolink™ Polymer Detection Systems. For further product information or support, contact your local distributor or regional office of Leica Biosystems, or alternatively, visit the Leica Biosystems Web site, www.LeicaBiosystems.com

The performance of this antibody should be validated when utilized with other manual staining systems or automated platforms.

Storage and Stability

Store at 2-8 °C. Do not freeze. Return to 2-8 °C immediately after use. Do not use after expiration date indicated on the vial label. Storage conditions other than those specified above must be verified by the user.

Specimen Preparation

The recommended fixative is 10% neutral-buffered formalin for paraffin-embedded tissue sections.

Warnings and Precautions

This reagent has been prepared from the supernatant of cell culture. As it is a biological product, reasonable care should be taken when handling it.

This reagent contains sodium azide. A Material Safety Data Sheet is available upon request or available from www.LeicaBiosystems.com

Consult federal, state or local regulations for disposal of any potentially toxic components.

Specimens, before and after fixation, and all materials exposed to them, should be handled as if capable of transmitting infection and disposed of with proper precautions. 1 Never pipette reagents by mouth and avoid contacting the skin and mucous membranes with reagents and specimens. If reagents or specimens come in contact with sensitive areas, wash with copious amounts of water. Seek

Minimize microbial contamination of reagents or an increase in non-specific staining may occur.

Incubation times or temperatures, other than those specified, may give erroneous results. Any such changes must be validated by the

Quality Control

Differences in tissue processing and technical procedures in the user's laboratory may produce significant variability in results, necessitating regular performance of in-house controls in addition to the following procedures.

Controls should be fresh autopsy/biopsy/surgical specimens, formalin-fixed, processed and paraffin wax-embedded as soon as possible in the same manner as the patient sample(s).

Positive Tissue Control

Used to indicate correctly prepared tissues and proper staining techniques.

One positive tissue control should be included for each set of test conditions in each staining run.

A tissue with weak positive staining is more suitable than a tissue with strong positive staining for optimal quality control and to detect minor levels of reagent degradation.²

Recommended positive control tissue is cerebellum.

If the positive tissue control fails to demonstrate positive staining, results with the test specimens should be considered invalid.

Negative Tissue Control

Should be examined after the positive tissue control to verify the specificity of the labeling of the target antigen by the primary antibody. Recommended negative control tissue is placenta.

Alternatively, the variety of different cell types present in most tissue sections frequently offers negative control sites, but this should be verified by the user.

Non-specific staining, if present, usually has a diffuse appearance. Sporadic staining of connective tissue may also be observed in sections from excessively formalin-fixed tissues. Use intact cells for interpretation of staining results. Necrotic or degenerated cells often stain non-specifically.³ False-positive results may be seen due to non-immunological binding of proteins or substrate reaction products. They may also be caused by endogenous enzymes such as pseudoperoxidase (crythrocytes), endogenous peroxidase (cytochrome C), or endogenous biotin (eg. liver, breast, brain, kidney) depending on the type of immunostain used. To differentiate endogenous enzyme activity or non-specific binding of enzymes from specific immunoreactivity, additional patient tissues may be stained exclusively with substrate chromogen or enzyme complexes (avidin-biotin, streptavidin, labeled polymer) and substrate-chromogen, respectively. If specific staining occurs in the negative tissue control, results with the patient specimens should be considered invalid.

Negative Reagent Control

Use a non-specific negative reagent control in place of the primary antibody with a section of each patient specimen to evaluate non-specific staining and allow better interpretation of specific staining at the antigen site.

Patient Tissue

Examine patient specimens stained with NCL-L-CD56-504 last. Positive staining intensity should be assessed within the context of any non-specific background staining of the negative reagent control. As with any immunohistochemical test, a negative result means that the antigen was not detected, not that the antigen was absent in the cells/tissue assayed. If necessary, use a panel of antibodies to identify false-negative reactions.

Results Expected

Normal Tissues

Clone CD564 detected the CD56 (NCAM) antigen on the membrane of NK cells, a subset of activated T cells and neuroectodermal cells. (Total number of normal cases evaluated = 44).

Abnormal Tissues

Clone CD564 stained 5/10 plasma cell myelomas, 4/118 diffuse large B-cell lymphomas, 3/9 T-cell lymphomas, 3/3 T/NK cell lymphomas, 1/3 Burkitt's lymphomas, 1/1 primitive B/T cell acute lymphoblastic lymphoma, 1/1 neuroblastoma, 1/1 anaplastic astrocytoma of the brain, 1/1 choroid plexus papilloma of the brain, 1/1 atypical carcinoid of the thymus, 1/1 soft tissue ganglioneuroma, 1/2 kidney renal cell carcinomas, 1/2 seminomas, 1/4 lung tumors (including 1/1 non-small cell carcinoma, 0/1 adenocarcinoma, 0/1 squamous cell carcinoma and 0/1 large cell carcinoma), 1/4 liver carcinomas (including 1/1 liver metastatic carcinoma, 0/1 cholangiocarcinoma and 0/2 hepatocellular carcinomas) and 1/4 ovarian carcinomas (including 1/1 serous cystadenocarcinoma, 0/1 malignant germ cell tumor, 0/1 clear cell carcinoma and 0/1 mucinous cystadenocarcinoma). No staining was observed in Hodgkin's disease (0/21), chronic lymphocytic lymphomas (0/12), follicular lymphomas (0/11), mantle cell lymphomas (0/7), T-cell anaplastic large cell lymphomas (0/7), angioimmunoblastic T-cell lymphomas (0/4), follicular B-cell lymphomas (0/3), a B-cell acute lymphoblastic lymphoma (0/1), a peripheral T-cell lymphoma (0/1), a marginal zone lymphoma (0/1), thyroid tumors (0/3), esophageal tumors (0/2), breast tumors (0/2), metastatic tumors of unknown origin (0/2), tumors of the tongue (0/2), tumors of the stomach (0/2), tumors of the stomach (0/2), tumor of the larynx (0/1), (Total number of tumor cases evaluated = 256).

NCL-L-CD56-504 is recommended for use as part of an antibody panel to aid in the determination of neuroectodermal tumor origin.

General Limitations

Immunohistochemistry is a multistep diagnostic process that consists of specialized training in the selection of the appropriate reagents; tissue selection, fixation, and processing; preparation of the IHC slide; and interpretation of the staining results.

Tissue staining is dependent on the handling and processing of the tissue prior to staining. Improper fixation, freezing, thawing, washing, drying, heating, sectioning or contamination with other tissues or fluids may produce artifacts, antibody trapping, or false negative results. Inconsistent results may be due to variations in fixation and embedding methods, or to inherent irregularities within the tissue.⁴ Excessive or incomplete counterstaining may compromise proper interpretation of results.

The clinical interpretation of any staining or its absence should be complemented by morphological studies using proper controls and should be evaluated within the context of the patient's clinical history and other diagnostic tests by a qualified pathologist. Antibodies from Leica Biosystems Newcastle Ltd are for use, as indicated, on either frozen or paraffin-embedded sections with specific fixation requirements. Unexpected antigen expression may occur, especially in neoplasms. The clinical interpretation of any stained tissue section must include morphological analysis and the evaluation of appropriate controls.

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 activity of immune cells in human ovarian and abdominal wall endometriomas. Reproductive Biology and Endocrinology. 2006; 4:41.
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Amendments to Previous Issue

Reagent Composition, Total Protein Concentration, Recommendations On Use, Warnings and Precautions, Results Expected.

Date of Issue

12 June 2013

Novocastra[™] Anticorp lichid monoclonal de șoarece CD68

Cod produs: NCL-L-CD68

Utilizare prevăzută

Pentru diagnosticare in vitro.

NCL-L-CD68 este destinat identificării calitative, prin intermediul microscopiei optice, a antigenului CD68 uman în secțiunile de parafină. Interpretarea clinică a oricărei colorări sau a absenței acesteia trebuie completată cu studii morfologice utilizând controale adecvate și trebuie evaluată în contextul antecedentelor clinice ale pacientului, precum și al altor teste de diagnosticare efectuate de către un patolog calificat.

Principiul de procedură

Tehnicile de colorare imunohistochimică (IHC) permit vizualizarea antigenilor prin aplicarea secvențială a unui anumit anticorp pe antigen (anticorp primar), a unui anticorp secundar pe anticorpul primar și a unui complex enzimatic cu un substrat cromogen, cu etape de spălare intercalate. Activarea enzimatică a cromogenului duce la un produs de reacție vizibil la locul aplicării antigenului. Specimenul poate fi apoi contracolorat și acoperit cu lamelă. Rezultatele sunt interpretate folosind un microscop optic și ajută la diagnosticul diferențial al proceselor patofiziologice, care pot sau nu să fie asociate cu un anumit antigen.

Clonă

514H12

Imunogen

Proteină procariotă recombinantă corespunzând jumătății carboxi-terminale a domeniului extern al moleculei CD68 umane.

Specificitate

Antigen CD68 uman.

Compoziția reactivului

NCL-L-CD68 este un supernatant de cultură tisulară lichid care conține azidă de sodiu drept conservant.

Clasa Ig

IgG2a, kappa

Concentrație proteină totală

Total Protein

Consultați eticheta flaconului pentru concentrația proteinelor totale specifică lotului.

Concentrație anticorpi

Mai mare sau egală cu 37 mg/L, așa cum este determinată prin ELISA. Consultați eticheta flaconului pentru concentrația Ig specifică lotului.

Recomandări privind utilizarea

Imunohistochimie pe sectiuni de parafină.

Recuperarea indusă de căldură a epitopilor (HIER): Urmați instrucțiunile de utilizare din Novocastra Epitope Retrieval Solution pH 9. Diluție sugerată: 1:100 timp de 30 de minute la 25 °C. Aceste informații sunt furnizate cu rol de îndrumare, iar utilizatorii trebuie să-și stabilească singuri propriile diluții de lucru optime.

Vizualizare: Respectați instrucțiunile de utilizare din Novolink™ Polymer Detection Systems. Pentru asistență sau informații suplimentare cu privire la produs, luați legătura cu distribuitorul dvs. local sau cu biroul regional al Leica Biosystems sau, ca alternativă, vizitați site-ul web al Leica Biosystems, www.LeicaBiosystems.com.

Notă în unele cazuri, etapa de bloc de peroxidază imediat după HIER poate afecta colorarea obținută cu acest anticorp. Performanța acestui anticorp trebuie validată atunci când este utilizat cu alte sisteme de colorare manuală sau alte platforme automatizate.

Depozitare și stabilitate

A se depozita la 2–8 °C. A nu se congela. A se returna la 2–8 °C imediat după utilizare. A nu se utiliza după data expirării indicată pe eticheta flaconului. Alte condiții de depozitare decât cele specificate mai sus trebuie verificate de către utilizator.

Pregătirea specimenului

Mediul de fixare recomandat este formalină tamponată neutru 10% pentru secțiunile de tesut încorporate în parafină.

Avertismente și precauții

Acest reactiv a fost pregătit din supernatantul culturii celulare. Întrucât este un produs biologic, trebuie să se acționeze cu prudență rezonabilă la manipularea sa.

Acest reactiv conține azidă de sodiu. O Fișă tehnică de securitate a materialului este disponibilă la cerere sau pe site-ul www.LeicaBiosystems.com

Consultați reglementările naționale sau locale pentru informații privind eliminarea la deșeuri a tuturor componentelor potențial toxice. Probele, înainte și după fixare, precum și toate materialele expuse la acestea, trebuie manipulate ca și când ar avea potențialul de a transmite infecții și trebuie eliminate la deșeuri luând măsurile de precauție adecvate.¹ Nu pipetați niciodată reactivii cu gura și evitați contactul reactivilor și probelor cu pielea și membranele mucoase. Dacă reactivii sau probele vin în contact cu suprafețele sensibile, spălați cu apă din abundență. Solicitați asistență medicală.

Reduceți la minimum contaminarea microbiană a reactivilor, în caz contrar poate apărea o creștere a colorării nespecifice. Timpii sau temperaturile de incubație care diferă de valorile specificate pot genera rezultate eronate. Orice astfel de modificări trebuie validate de către utilizator.

Controlul calității

Diferențele în ceea ce privește procesarea țesutului și procedurile tehnice în laboratorul utilizatorului pot cauza o variabilitate semnificativă a rezultatelor, necesitând efectuarea cu regularitate de controale interne, în plus față de următoarele proceduri. Probele de control trebuie să fie probe proaspete de autopsie/biopsie/chirurgicale, fixate în formalină, procesate și încorporate în ceară de parafină cât mai curând posibil și în aceeași manieră ca și probele pacientului.

Tesutul de control pozitiv

Folosit pentru a indica țesuturile pregătite corect și tehnicile de colorare adecvate.

O probă de țesut de control pozitiv trebuie să fie inclusă pentru fiecare set de condiții de testare în fiecare etapă de colorare. Un țesut cu colorare pozitivă slabă este mai adecvat decât un țesut cu colorare pozitivă puternică în vederea unui control optim al calității și pentru a detecta nivelurile minore de degradare a reactivului.²

Tesutul de control pozitiv recomandat este amigdală sau intestin (macrofage).

Dacă țesutul de control pozitiv nu demonstrează colorația pozitivă, rezultatele obținute cu acele probe de testare trebuie considerate nevalide.

Tesutul de control negativ

Trebuie examinat după țesutul de control pozitiv pentru a verifica specificitatea informațiilor de etichetare ale antigenului țintă în funcție de anticorpul primar.

Țesutul de control negativ recomandat este amigdală (celule endoteliale).

Ca alternativă, varietatea de tipuri diferite de celule prezente în majoritatea secțiunilor tisulare oferă frecvent locuri de control negativ, dar acest lucru trebuie verificat de către utilizator.

Colorația nespecifică, dacă este prezentă, are, de obicei, un aspect difuz. Colorația sporadică a țesutului conjunctiv poate fi observată, de asemenea, în secțiuni de țesuturi fixate în mod excesiv în formalină. Folosiți celule intacte pentru interpretarea rezultatelor de colorare. Celulele necrotice sau degenerate se colorează deseori într-un mod nespecific. Se pot observa rezultate fals pozitive ca urmare a legării non-imunologice a proteinelor sau produșilor de reacție ai substratului. Acestea pot fi cauzate, de asemenea, de enzimele endogene precum pseudoperoxidaza (eritrocite), peroxidaza endogenă (citocromul C) sau biotina endogenă (de exemplu, ficat, sân, encefal, rinichi), în funcție de tipul de imunocolorație folosit. Pentru a diferenția activitatea enzimelor endogene sau legarea nespecifică a enzimelor de imunoreactivitatea specifică, pot fi colorate țesuturi suplimentare de la pacient numai cu substrat-cromogen sau, respectiv, complexe enzimatice (avidină-biotină, streptavidină, polimer etichetat) și substrat-cromogen. În cazul în care colorația specifică are loc în tesutul de control negativ, rezultatele obținute pe probele pacientului trebuie să fie considerate nevalide.

Reactivul de control negativ

Folosiți un reactiv de control negativ non-specific în locul anticorpului primar cu o secțiune din fiecare specimen al pacientului pentru a evalua coloratia nespecifică si a permite o mai bună interpretare a colorării specifice la situl antigenului.

Ţesutul pacientului

Examinați specimenele pacientului colorate cu NCL-L-CD68 ultimele. Intensitatea colorației pozitive trebuie evaluată în contextul oricărei colorații de fond nespecifice a reactivului de control negativ. La fel ca în cazul oricărui test imunohistochimic, un rezultat negativ înseamnă că antigenul nu a fost detectat, și nu că antigenul a fost absent în celulele/țesuturile analizate. Dacă este necesar, folosiți un panel pentru anticorpi pentru identificarea reacțiilor fals negative.

Rezultate așteptate

<u>Tesuturi normale</u>

Clona 514H12 a detectat glicoproteina CD68 atât în citoplasma cât şi în membrana celulară a unei varietăți de tipuri de celule din linia celulară mielomonocitică incluzând monocite, macrofage, granulocite, celule microgliale, celule Kupffer hepatice, celule Hofbauer placentare şi o proporție de celule dendritice. (Numărul total al cazurilor normale evaluate = 47).

Tesuturi anormale

Clona 514H12 a colorat 1/201 tumori evaluate, incluzând seminoame testiculare (1/2), limfoame (0/80, incluzând 0/44 limfom difuz cu celule B, 0/6 limfom nodular difuz cu celule B, 0/6 limfom Hodgkin cu celularitate mixtă, 0/5 limfom anaplastic cu celule mari, 0/4 limfom folicular non-Hodgkin, 0/3 limfom asociat mucoasei celulelor B, 0/3 limfom Hodgkin cu tip predominant de limfocite, 0/2 limfom cu celule T, 0/2 limfom plasmacitoid limfocitic, 0/1 limfom similar cu Burkitt. 0/1 limfom cu celule de manta, 0/1 limfom cu celule clare cu celule T, 0/1 limfom Lennert și 0/1 limfom Hodgkin cu scleroză nodulară), tumori ale pielii (0/77, incluzând 0/16 carcinom cu celule scuamoase, 0/15 melanom malign, 0/14 carcinom cu celule bazale, 0/10 carcinom al glandelor sudoripare, 0/10 dermatofibrosarcom, 0/3 adenocarcinom metastatic, 0/3 schwannom malign, 0/2 carcinom cistic adenoid, 0/1 adenocarcinom sebaceu, 0/1 fibrosarcom, 0/1 leiomiosarcom și 0/1 sarcom pleomorfic nediferențiat), carcinoame hepatice (0/6), carcinoame papilare tiroidiene (0/4), carcinoame pulmonare (0/4), tumori ovariene (0/4), tumori cerebrale (0/2), tumori ale țesuturilor moi (0/2), metastaze de carcinom nespecificat (0/2), carcinoame cu celule scuamoase ale esofagului (0/2), carcinoame ductale mamare (0/2), carcinoame cu celule renale (0/2), adenocarcinoame gastrice (0/2), adenocarcinoame ale colonului (0/2), carcinoame rectale (0/2), carcinoame cu celule scuamoase ale limbii (0/2), carcinoame cu celule scuamoase ale colului uterin (0/2), carcinom cu celule scuamoase ale laringelui (0/1) și tumoare carcinoidă atipică a timusului (0/1). (Numărul total al cazurilor tumorale evaluate = 201).

NCL-L-CD68 este recomandat pentru identificarea antigenului CD68 într-o varietate de tesuturi normale si neoplazice.

Limitări generale

Imunohistochimia este un proces de diagnostic cu mai multe etape, care constă din instruirea specializată în ceea ce privește alegerea reactivilor adecvați; alegerea, fixarea și procesarea țesutului; prepararea lamei IHC; și interpretarea rezultatelor de colorare. Colorarea tisulară depinde de manipularea și procesarea țesutului înainte de colorare. Fixarea, congelarea, dezghețarea, spălarea, uscarea, încălzirea, secționarea necorespunzătoare sau contaminarea cu alte țesuturi ori fluide pot cauza artefacte, captura anticorpilor

sau rezultate fals negative. Rezultatele inconsecvente pot fi atribuite diferențelor în ceea ce privește metodele de fixare și încorporare, ori neregularitătilor inerente ale tesutului.

Contracoloratia excesivă sau incompletă poate compromite interpretarea adecvată a rezultatelor.

Interpretarea clinică a oricărei colorări sau a absenței acesteia trebuie completată cu studii morfologice utilizând controale adecvate și trebuie evaluată în contextul antecedentelor clinice ale pacientului, precum și al altor teste de diagnosticare efectuate de către un patolog calificat.

Anticorpii de la Leica Biosystems Newcastle Ltd sunt destinați utilizării, conform indicațiilor, fie pe secțiuni congelate, fie pe secțiuni încorporate în parafină cu cerințe de fixare specifice. Poate apărea exprimarea neașteptată a antigenului, în special în neoplasme. Interpretarea clinică a oricărei secțiuni tisulare colorate trebuie să includă analiza morfologică și evaluarea probelor de control adecvate.

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Amendamente la ediția anterioară

Compoziția reactivilor, Concentrația totală a proteinelor, Recomandări de utilizare, Avertizări și măsuri de precauție, Rezultate preconizate.

Data publicării

07 noiembrie 2018

abcam

Product datasheet

Anti-Plasma Cell antibody [LIV3G11] ab270730

1 Image

Overview

Product name Anti-Plasma Cell antibody [LIV3G11]

Description Mouse monoclonal [LIV3G11] to Plasma Cell

Host species Mouse

Tested applications Suitable for: ℍC-P

Species reactivity Reacts with: Human

Does not react with: Rat

Immunogen Tissue, cells or virus corresponding to Human Plasma Cell. Pancreatic cancer-related mucin.

Positive control IHC-P: Human tonsil tissue.

General notes

The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle.

Storage buffer pH: 7.2

Preservative: 0.05% Sodium azide Constituents: PBS, 0.05% BSA

Purity Protein A/G purified

Purification notes Purified from Bioreactor Concentrate.

Clonality Monoclonal
Clone number LIV3G11
Isotype IgG2a
Light chain type kappa

1

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab270730 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IHC-P		Use a concentration of 0.1 - 0.2 µg/ml. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

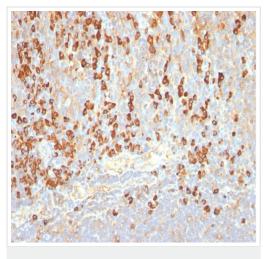
Target

Relevance Plasma cells (also called plasma B cells or plasmocytes) are cells of the immune system that

secrete antibodies.

Cellular localization Cytoplasmic

Images



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Plasma Cell antibody
[LIV3G11] (ab270730)

Formalin-fixed, paraffin-embedded human tonsil tissue stained for Plasma Cell using ab270730 at 0.2 μ g/ml in immunohistochemical analysis.

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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Novocastra

Novolink Max Polymer Detection System (1250 tests)

Nr. produs RE7280-K

Novolink Polymer Detection System (500 tests)

Nr. produs RE7150-K

Novolink Polymer Detection System (250 tests)

Nr. produs RE7140-K

Novolink Min Polymer Detection System (50 tests)

Nr. produs RE7290-K

Novolink Max Polymer (1250 tests)

Nr. produs RE7260-K

Novolink Polymer (250 tests)

Nr. produs RE7200-K

Novolink Max DAB (Polymer) 1250 tests

Nr. produs RE7270-K

Novolink DAB (Polymer) 250 tests

Nr. produs RE7230-K

Utilizare prevăzută

Pentru diagnosticare in vitro.

Novolink Polymer Detection Systems sunt utilizate pentru vizualizarea anticorpilor primari IgG de șoarece, IgM de șoarece și IgG de iepure. Novolink Polymer și Novolink DAB (Polymer) conțin reactivi componenți ai acestor sisteme și sunt prevăzute pentru utilizare în procedura descrisă mai jos. Interpretarea clinică a oricărei colorări sau a absenței acesteia trebuie completată cu studii morfologice utilizând controale adecvate și trebuie evaluată în contextul istoricului clinic al pacientului, precum și al altor teste de diagnosticare efectuate de către un patolog calificat.

Principiul procedurii

Prima tehnică cu imunoperoxidază a fost raportată de Nakane și Pierce¹. De atunci au avut loc multe evoluții care au dus la creșterea sensibilității față de tehnicile mai vechi. O evoluție recentă a fost utilizarea etichetării polimerice. Această tehnologie a fost aplicată atât la anticorpii primari² cât și la sistemele de detecție. Novolink Polymer Detection Systems utilizează o tehnologie nouă de polimerizare controlată pentru prepararea conjugatelor de anticorp de legătură HRP. Prin urmare, nu apare problema colorației nespecifice care se poate produce cu sistemele de detecție cu Streptavidină/Biotină din cauza biotinei endogene.

Aceste produse sunt utilizate într-o procedură imunohistochimică (IHC), care permite identificarea calitativă prin microscopie optică a antigenilor în secțiuni de țesut fixat cu formalină, încorporat în parafină, prin etape secvențiale cu etape de spălare intercalate. Dacă este necesar pentru anticorpul primar, secțiunile sunt supuse recuperării epitopilor înainte de colorație. Activitatea peroxidazei endogene este neutralizată utilizând Peroxidase Block. Acesta este urmat de aplicarea Novocastra Protein Block pentru a reduce legarea nespecifică a anticorpului primar și a polimerului Secțiunea este apoi incubată cu anticorp primar în diluție optimă. Apoi se utilizează Post Primary (IgG de iepure anti șoarece) pentru detectarea anticorpilor de șoarece. Novolink Polymer recunoaște imunoglobulinele de iepure, detectează Post Primary și orice anticorpi primari de iepure legați de țesut. Secțiunile sunt apoi incubate în continuare cu substratul/cromogen, 3,3'-diaminobenzidină (DAB), preparat din DAB Chromogen și Novolink DAB Substrate Buffer (Polymer), după cum se descrie mai jos. Reacția cu peroxidaza produce un precipitat cafeniu vizibil la situl antigenului. Secțiunile sunt contracolorate cu Hematoxylin și acoperite cu lamele. Rezultatele sunt interpretate folosind un microscop optic și ajută la diagnosticul diferențial al proceselor patofiziologice, care pot sau nu să fie asociate cu un anumit antigen.

Reactivi furnizați

Detalii reactivilor din lista următoare care sunt furnizați în fiecare produs sunt date în tabelul de mai jos.

- 1. Peroxidase Block, apă oxigenată 3-4% (v/v).
- 2. Protein Block. 0,4% cazeină în soluție salină tamponată cu fosfat, cu stabilizatori, surfactant și 0,2% Bronidox L drept conservant.
- 3. Post Primary. IgG de iepure anti șoarece (<10 µg/ml) în ser animal 10% (v/v) în soluție salină tamponată cu trometamină/0,1% ProClin™ 950.
- 4. Novolink Polymer. Poly-HRP-IgG anti-iepure (<25μg/ml) conţinând 10% (v/v) ser animal în soluţie salină tamponată cu trometamină/0,1% ProClin™ 950.
- 5. DAB Chromogen. 1,74% w/v 3,3' diaminobenzidină, în soluție stabilizatoare.
- 6. Novolink DAB Substrate Buffer (Polymer). Soluție tamponată conținând apă oxigenată ≤ 0,1% și conservant.
- 7. Hematoxylin. Hematoxylin <0,1%.

Reactiv	Număr produs	Novolink Max Polymer Detection System (1250 tests) RE7280-K	Novolink Polymer Detection System (500 tests) RE7150-K	Novolink Polymer Detection System (250 tests) RE7140-K	Novolink Min Polymer Detection System (50 tests) RE7290-K
Peroxidase Block	RE7101		2 x 25 ml	1 x 25 ml	
Protein Block	RE7102		2 x 25 ml	1 x 25 ml	
Post Primary	RE7111		2 x 25 ml	1 x 25 ml	
Novolink Polymer	RE7112		2 x 25 ml	1 x 25 ml	
DAB Chromogen	RE7105		1 x 3ml	1 x 3ml	
Novolink DAB Substrate Buffer (Polymer)	RE7143		2 x 30ml	1 x 30ml	
Hematoxylin	RE7107		2 x 25 ml	1 x 25 ml	
Peroxidase B l ock	RE7157	1 x 125ml			
Protein Block	RE7158	1 x 125ml			
Post Primary	RE7159	1 x 125ml			
Novolink Polymer	RE7161	1 x 125ml			
DAB Chromogen	RE7162	1 x 8ml			
Novolink DAB Substrate Buffer (Polymer)	RE7163	1 x 150ml			
Hematoxylin	RE7164	1 x 125ml			
Peroxidase Block	RE7165				1 x 5ml
Protein Block	RE7166				1 x 5ml
Post Primary	RE7167				1 x 5ml
Novolink Polymer	RE7168				1 x 5ml
DAB Chromogen	RE7169				1 x 1ml
Novolink DAB Substrate Buffer (Polymer)	RE7171				1 x 5ml
Hematoxylin	RE7172				1 x 5ml

Reactiv	Număr produs	Novolink Max Polymer (1250 tests) RE7260-K	Novolink Polymer (250 tests) RE7200-K	Novolink Max DAB (Polymer) 1250 tests RE7270-K	Novolink DAB (Polymer) 250 tests RE7230-K
Post Primary	RE7111		1 x 25 ml		
Novolink Polymer	RE7112		1 x 25 ml		
Post Primary	RE7159	1 x 125ml			
Novolink Polymer	RE7161	1 x 125ml			
DAB Chromogen	RE7105				1 x 3ml
Novolink DAB Substrate Buffer (Polymer)	RE7143				1 x 30ml
DAB Chromogen	RE7162			1 x 8ml	
Novolink DAB Substrate Buffer (Polymer)	RE7163			1 x 150ml	

Reconstituire, amestecare, diluare, titrare

Peroxidase Block, Protein Block, Post Primary, Novolink Polymer şi Hematoxylin sunt prediluate. Reconstituirea, amestecarea, diluarea sau titrarea acestor reactivi nu sunt recomandate. Diluarea poate duce la pierderea colorării antigenilor. Utilizatorul trebuie să valideze orice astfel de schimbare.

DAB Chromogen necesită diluare la 1/20 în Novolink DAB Substrate Buffer (Polymer) înainte de utilizare. Diluarea poate duce la pierderea colorării antigenilor. Utilizatorul trebuie să valideze orice astfel de schimbare.

Depozitare și stabilitate

A se depozita la 2–8 °C. A nu se congela. A se returna la 2–8 °C imediat după utilizare. A nu se utiliza după data expirării indicată pe eticheta produsului. Alte condiții de depozitare decât cele specificate trebuie verificate de către utilizator. Nu există semne evidente care să indice instabilitatea acestui produs, astfel că trebuie rulate controale pozitive și negative simultan cu eșantioanele pacientului.

Pregătirea specimenului

Fixativul recomandat este formalină tamponată neutru 10% pentru secțiunile de țesut încorporate în parafină.

Avertismente și precauții

O Fisă tehnică de securitate a materialului este disponibilă la cerere sau poate fi obtinută de pe site-ul www.LeicaBiosystems.com

Pentru utilizatori profesioniști.

Nu amestecați reactivi din sisteme de detecție diferite.

Specimenele, înainte și după fixare, precum și toate materialele expuse la acestea, trebuie manipulate ca și când ar avea potențialul de a transmite infecții si trebuie eliminate luând măsurile de precauție adecvate.⁴

Nu pipetați niciodată reactivii cu gura și evitați contactul reactivilor și specimenelor cu pielea și membranele mucoase. Dacă reactivii sau probele vin în contact cu suprafețele sensibile, spălați cu apă din abundență.

Consultați reglementările naționale, județene sau locale pentru informații privind eliminarea la deșeuri a oricăror componente cu potential toxic.

Reduceți la minimum contaminarea microbiană a reactivilor, în caz contrar poate apărea o creștere a colorației nespecifice. Timpii sau temperaturile de incubație care diferă de valorile specificate pot genera rezultate eronate. Orice astfel de modificare trebuie validată de către utilizator.

Procedură

A. Reactivi necesari care nu sunt însă furnizați

- 1. Solvenți standard folosiți în imunohistochimie.
- 2. Soluție salină tamponată cu trometamină 50 mM (SSTT) pH 7,6.
- 3. Soluție(i) de recuperare cu antigen.
- 4. Soluție(i) de recuperare cu enzime.
- 5. Diluant pentru anticorpi.
- 6. Anticorp primar.
- 7. Mediu de montare.

B. Echipamente necesare care nu sunt însă furnizate

- 1. Echipament necesar pentru recuperarea cu antigen, dacă este recomandată pentru anticorpul primar.
- 2. Echipament de laborator general pentru imunohistochimie.

C. Metodologie

Înainte de a aplica această metodologie, utilizatorii trebuie să fie instruiti în ceea ce priveste tehnicile imunohistochimice.

Toti pasii trebuie urmati conform instructiunilor, în caz contrar putând fi afectată performanta.

Combinația între anticorpul primar, diluția acestuia, împreună cu sistemul de detecție, trebuie validată de utilizator pe o serie de controale pozitive și negative cunoscute.

Dacă nu se indică altfel, toate etapele se efectuează la temperatura camerei (25 °C).

Pentru utilizare pe țesut congelat, tăiați secțiuni și fixați conform recomandărilor pentru anticorp primar, începeți la pasul 11.

- 1. Tăiați și montați secțiunile pe lame acoperite cu un adeziv tisular adecvat.
- 2. Deparafinizati sectiunile în xilen sau substitute de xilen.
- 3. Rehidratați cu ajutorul alcoolilor cu gradație descrescătoare.
- 4. Spălați lamele cu apă de la robinet.
- 5. Realizați recuperarea antigenilor după cum este necesar(a se vedea Recomandări de utilizare pentru anticorpul primar).
- 6. Spălați lamelele în apă deionizată.
- 7. Neutralizați peroxidaza endogenă utilizând Peroxidase Block timp de 5 minute.
- 8. Spălați în SSTT timp de 2 x 5 minute.
- 9. Incubați cu Protein Block timp de 5 minute.
- 10. Spălați în SSTT timp de 2 x 5 minute.
- 11. Incubați cu anticorp primar diluat optim (a se vedea Recomandări de utilizare pentru anticorpul primar).
- 12. Spălați în SSTT timp de 2 x 5 minute.
- 13. Incubați cu Post Primary timp de 30 de minute.
- 14. Spălați în SSTT timp de 2 x 5 minute.

- 15. Incubați cu Novolink Polymer timp de 30 de minute.
- 16. Spălați în soluție tampon SSTT timp de 2 x 5 minute, legănând usor.
- 17.Dezvoltați activitatea peroxidazei cu soluție de lucru DAB (a se vedea Soluție de lucru DAB) timp de 5 minute.
- 18. Clătiți lamele în apă.
- 19. Contracolorati cu Hematoxylin.
- 20. Clătiți lamelele în apă timp de 5 minute.
- 21. Deshidratați, curățați și montați secțiunile.

Solutie de lucru DAB

Adăugați 50µl de DAB Chromogen la 1ml de Novolink DAB Substrate Buffer (Polymer). Utilizați în maxim șase ore de la preparare.

Controlul calității

Diferențele în ceea ce privește procesarea țesutului și procedurile tehnice în laboratorul utilizatorului pot cauza o variabilitate semnificativă a rezultatelor, necesitând efectuarea cu regularitate de controale interne, în plus față de următoarele proceduri. Probele de control trebuie să fie probe proaspete de autopsie/biopsie/chirurgicale, fixate în formalină, procesate și încorporate în ceară de parafină cât mai curând posibil si în aceeasi manieră ca si esantioanele pacientului.

Tesutul de control pozitiv

Folosit pentru a indica țesuturile pregătite corect și tehnicile de colorare adecvate. O probă de țesut de control pozitiv trebuie să fie inclusă pentru fiecare set de condiții de testare/anticorp primar în fiecare etapă de colorație. Un țesut cu colorație pozitivă slabă este mai adecvat decât un țesut cu colorație pozitivă puternică pentru controlul optim al calității și pentru a detecta nivele minore de degradare a reactivilor.⁵ Pentru țesutul de control pozitiv recomandat a se vedea Recomandări de utilizare. Dacă țesutul de control pozitiv nu demonstrează colorația pozitivă, rezultatele obținute cu acele probe de testare trebuie considerate nevalide.

Țesutul de control negativ

Trebuie examinat după țesutul de control pozitiv pentru a verifica specificitatea informațiilor de etichetare ale antigenului țintă în funcție de anticorpul primar. Pentru țesutul de control negativ recomandat, a se vedea Instrucțiunile de utilizare pentru anticorpul primar Ca alternativă, varietatea de tipuri diferite de celule prezente în majoritatea secțiunilor tisulare oferă frecvent locuri de control negativ, dar acest lucru trebuie verificat de către utilizator. Colorația nespecifică, dacă este prezentă, are, de obicei, un aspect difuz. Colorația sporadică a țesutului conjunctiv poate fi observată, de asemenea, în secțiuni de țesuturi fixate în mod excesiv în formalină. Folosiți celule intacte pentru interpretarea rezultatelor de colorație. Celulele necrotice sau degenerate se colorează deseori într-un mod nespecific.⁶ Se pot observa rezultate fals pozitive ca urmare a legării non-imunologice a proteinelor sau produșilor de reacție ai substratului. Acestea pot fi cauzate, de asemenea, de enzimele endogene precum pseudoperoxidaza (eritrocite), peroxidaza endogenă (citocromul C) sau biotina endogenă⁷ (de exemplu, ficat, sân, creier, rinichi), Pentru a diferenția activitatea enzimelor endogene sau legarea nespecifică de imunoreactivitatea specifică, pot fi colorate țesuturi suplimentare de la pacient numai cu substrat-cromogen, cu polimer etichetat și cu substrat-cromogen sau cu Post Primary, polimer etichetat și substrat-cromogen. În cazul în care colorația specifică are loc în țesutul de control negativ, rezultatele obținute pe eșantioanele pacientului trebuie să fie considerate nevalide.

Reactivul de control negativ

Folosiți un reactiv de control negativ nespecific în locul anticorpului primar cu o secțiune din fiecare specimen al pacientului pentru a evalua colorația nespecifică și a permite o mai bună interpretare a colorării specifice la situl antigenului.

Ţesutul pacientului

Examinați specimenele pacientului ultimele. Intensitatea colorării pozitive trebuie evaluată în contextul oricărei colorații de fundal nespecifice a reactivului de control negativ. La fel ca în cazul oricărui test imunohistochimic, un rezultat negativ înseamnă că antigenul nu a fost detectat, și nu că antigenul a fost absent în celulele/țesuturile analizate. Dacă este necesar, folosiți un panel de anticorpi pentru identificarea reacțiilor fals negative.

Limitări

Imunohistochimia este un proces de diagnostic cu mai multe etape, care constă din instruirea specializată în ceea ce privește alegerea reactivilor adecvați; alegerea, fixarea și procesarea țesutului; prepararea lamei IHC; și interpretarea rezultatelor de colorație.

Colorația tisulară depinde de manipularea și procesarea țesutului înainte de colorație. Fixarea, congelarea, dezghețarea, spălarea, uscarea, încălzirea, secționarea necorespunzătoare sau contaminarea cu alte țesuturi ori fluide pot cauza artefacte, captura anticorpilor sau rezultate fals negative. Rezultatele inconsecvente pot fi atribuite diferențelor în ceea ce privește metodele de fixare și încorporare, ori neregularităților inerente ale țesutului.8

Contracolorația excesivă sau incompletă poate compromite interpretarea adecvată a rezultatelor.

Interpretarea clinică a oricărei colorări sau a absenței acesteia trebuie completată cu studii morfologice utilizând controale adecvate și trebuie evaluată în contextul istoricului clinic al pacientului, precum și al altor teste de diagnosticare efectuate de către un patolog calificat. Novolink Polymer Detection Systems și componentele acestora sunt pentru utilizare pe secțiuni încorporate în parafină cu cerințe specifice de fixare. Poate apărea expresia neașteptată a antigenului, în special în neoplasme. Interpretarea clinică a oricărei secțiuni tisulare colorate trebuie să includă analiza morfologică și evaluarea probelor de control adecvate.

Caracteristici de performanță

Performanța Novolink Polymer Detection Systems, Novolink Polymer și Novolink DAB (Polymer) a fost validată utilizând o varietate de anticorpi primari Novocastra IgG de soarece, IgM* de soarece IgG de iepure.

*Se poate observa o colorare slabă cu unii anticorpi din izotipul IgM.

Aceste produse sunt stabile până la data expirării indicată pe eticheta produsului.

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Amendamente la ediția anterioară

Nu este cazul.

Data publicării

11 martie 2020

BOND Wash Solution 10X Concentrate

Nr. catalog: AR9590

Utilizare prevăzută

Acest reactiv este destinat utilizării pentru diagnosticare in vitro.

BOND Wash Solution 10X Concentrate este o soluție tampon concentrată, care necesită diluare înainte de utilizare. Soluția diluată este destinată spălării secțiunilo de țesut fixat cu formalină, încorporat în parafină în sistemul automat BOND (include sistemul Leica BOND-MAX și sistemul Leica BOND-III).

Interpretarea clinică a oricărei colorații sau a absenței acesteia trebuie verificată prin studii morfologice, folosind proceduri de control adecvate, și trebuie evaluată în contextul antecedentelor clinice ale pacientului, precum și al altor teste de diagnosticare efectuate de către un patolog calificat.

Rezumat și explicație

Pot fi utilizate tehnici imunohistochimice pentru a demonstra prezența antigenilor în țesut și celule (a se vedea "Utilizarea reactivilor BOND" din documentația de utilizare BOND).

Sistemul automat BOND necesită utilizarea unei soluții tampon de spălare specifice pentru eliminarea materialului neatașat la sfârșitul fiecărei etape de incubație. Această soluție tampon este preparată prin diluare, conform instrucțiunilor de mai jos, utilizând BOND Wash Solution 10X Concentrate. Apoi este umplut recipientul de vrac corespunzător și introdus în BOND Processing Module.

Reactivi furnizați

BOND Wash Solution 10X Concentrate conține soluție salină tamponată cu trometamină, surfactant și 3,5% ProClin™ 950. Volum total = 1 I. pH 7,5–7,7 la 25 °C, așa cum a fost măsurat la momentul formulării și punerii pe piață a concentratului și nereprezentativ pentru solutia de lucru. Suficient pentru a prepara 10 litri de BOND Wash Solution.

Diluare și amestecare

Diluați înainte de utilizare. Pentru a prepara 1 I de Soluție de spălare BOND amestecați 100 ml de BOND Wash Solution 10X Concentrate cu 900 ml de apă deionizată. BOND Wash Solution trebuie turnată în recipientul de vrac marcat "Wash Buffer" situat în BOND Processing Module. Acest recipient poate conține până la 2 I.

Materiale necesare, dar care nu sunt furnizate

Consultați "Utilizarea reactivilor BOND" din documentația dumneavoastră de utilizare a sistemului BOND pentru o listă completă a materialelor necesare pentru tratarea specimenelor si colorația imunohistochimică utilizând sistemul BOND.

Depozitare și stabilitate

A se depozita BOND Wash Solution 10X Concentrate la 2–8 °C ferită de radiație solară directă. Ocazional poate fi observată o cantitate mică de precipitat care se dizolvă la diluare. A nu se utiliza după data expirării indicată pe eticheta flaconului.

BOND Wash Solution diluată poate fi depozitată la 2-26 °C și poate fi utilizată timp de 4 luni.

Semnele care indică contaminarea și/sau instabilitatea Soluției de spălare diluate sunt: turbiditatea soluției, formarea de mirosuri și prezenta precipitatului.

Alte condiții de depozitare decât cele specificate mai sus trebuie verificate de către utilizator1.

Precautii

- Numai pentru utilizatori profesioniști.
- · Acest produs este destinat utilizării pentru diagnosticare in vitro.
- Concentrația de ProClin⁻ 950 este 3,5%. Acesta conține ingredientul activ 2-methilizotiazol-3(2H)-onă și poate cauza iritarea pielii, ochilor, membranelor mucoase și tractului respirator superior. Purtați mănuși de unică folosință atunci când manipulați reactivii.
- Pentru a obține o copie a fișei tehnice de securitate a materialului, luați legătura cu distribuitorul dvs. local sau cu biroul regional al Leica Biosystems sau, ca alternativă, vizitați site-ul web al Leica Biosystems, www.LeicaBiosystems.com
- Specimenele, înainte și după fixare, precum și toate materialele expuse la acestea, trebuie manipulate ca și când ar avea potențialul
 de a transmite infecții și trebuie eliminate luând măsurile de precauție adecvate². Nu pipetați niciodată reactivii cu gura și evitați
 contactul reactivilor și specimenelor cu pielea și membranele mucoase. Dacă reactivii sau probele vin în contact cu suprafețele
 sensibile, spălați cu apă din abundență. Solicitați asistență medicală.
- Consultați reglementările naționale, județene sau locale pentru informații privind eliminarea la deșeuri a oricăror componente cu
 potențial toxic.
- · Reduceți la minimum contaminarea microbiană a reactivilor, în caz contrar poate apărea o creștere a colorării nespecifice.
- Acest reactiv a fost formulat optim pentru o diluţie de 1:9. O diluţie mai mare poate duce la o performanţă necorespunzătoare a sistemului BOND şi la pierderea colorării.
- Nu trebuie utilizate alte soluții tampon în locul BOND Wash Solution 10X Concentrate cu sistemul BOND.

Instrucțiuni de utilizare

Pentru utilizarea BOND Wash Solution 10X Concentrate consultați "Diluare și amestecare".

Rezolvarea problemelor

Consultați referința 3 pentru acțiuni de remediere.

Contactati distribuitorul dumneavoastră local sau biroul regional al Leica Biosystems pentru raportarea colorării neobisnuite.

Informații suplimentare

Informații suplimentare referitoare la imunocolorația cu reactivii BOND, sub titlurile Principiul procedurii, Materiale necesare, Pregătirea specimenului, Controlul calității, Verificarea analizei, Interpretarea colorării, Codul simbolurilor de pe etichete și Limitări generale pot fi găsite în "Utilizarea reactivilor BOND" din documentația dumneavoastră de utilizare a sistemului BOND.

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