abcam

ab322927

Anti-Nectin-4 antibody [RM1257]

Recombinant | RabMAb | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Multiclonal
Immunogens	The exact immunogen used to generate this antibody is proprietary information.
Clone number	RM1257
Purification technique	Affinity purification Protein A
Concentration	0.488 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

Flow Cyt

Tested

Species	Human
Dilution info	1/50
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	1/1000
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	1/2000
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

IP

Tested

Species	Human
Dilution info	1/30
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

ICC/IF

Not recommended

Species	Human, Mouse, Rat
Dilution info	-
Notes	-

Target data

See full target information NECTIN4

Function	Seems to be involved in cell adhesion through trans-homophilic and -heterophilic interactions, the latter including specifically interactions with NECTIN1. Does not act as receptor for alpha-herpesvirus entry into cells. (Microbial infection) Acts as a receptor for measles virus.

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C

Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

What are recombinant multiclonals?
Recombinant multiclonals are a mixture of recombinant antibodies co-expressed from a library of heavy and light chains. They offer several advantages including:

- The sensitivity of polyclonal antibodies by recognising multiple epitopes
- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free batch production

View our range of recombinant multiclonal antibodies.

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

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

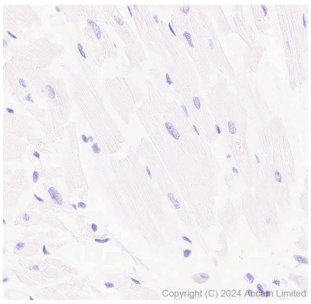
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

10 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Nectin-4 antibody [RM1257] (ab322927)

Nectin-4 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human cardiac muscle using rabbit Anti-Nectin-4 antibody

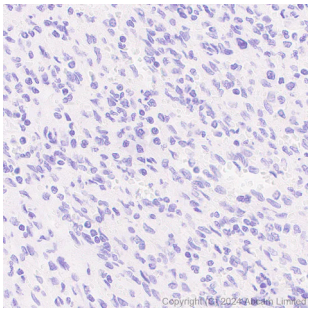
Immunohistochemical analysis of paraffin-embedded Human cardiac muscle tissue labeling Nectin-4 with ab322927 at 1/2000 (0.265 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Negative control: no staining on human cardiac muscle.
The section was incubated with ab322927 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).

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Nectin-4 antibody [RM1257] (ab322927)

Nectin-4 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human spleen using rabbit Anti-Nectin-4 antibody

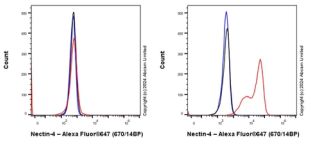
Immunohistochemical analysis of paraffin-embedded Human spleen tissue labeling Nectin-4 with ab322927 at 1/2000 (0.265 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Negative control: no staining on human spleen.
The section was incubated with ab322927 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).

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins

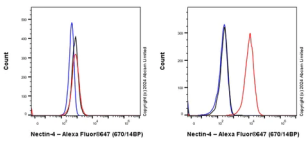


Flow Cytometry - Anti-Nectin-4 antibody [RM1257] (ab322927)

Flow cytometric analysis of T-47D (human ductal breast epithelial tumor epithelial cell) (Right) / A549 (human lung carcinoma epithelial cell) (Left) cells labelling Nectin-4 with ab322927 at 1/50 dilution (1ug) / Right compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody).

Goat anti-Rabbit IgG (Alexa Fluor® 647, ab150083) at 1/5000 dilution was used as the secondary antibody.

Gated on viable cells.
Low expression: A549 (PMID:21901103).

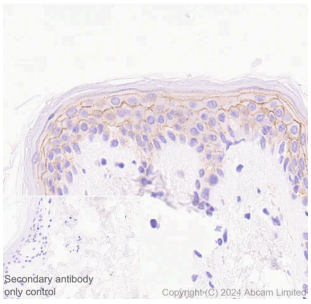


Flow Cytometry - Anti-Nectin-4 antibody [RM1257] (ab322927)

Flow cytometric analysis of DLD-1 (human colorectal adenocarcinoma epithelial cell) (Right) / HeLa (human cervical adenocarcinoma epithelial cell) (Left) cells labelling Nectin-4 with ab322927 at 1/50 dilution (1ug) / Right compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody).

Goat anti-Rabbit IgG (Alexa Fluor® 647, ab150083) at 1/5000 dilution was used as the secondary antibody.

Gated on viable cells.
Low expression: HeLa (PMID:21901103).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Nectin-4 antibody [RM1257] (ab322927)

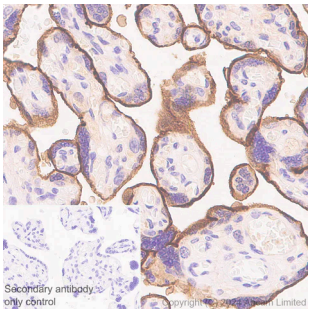
Immunohistochemical analysis of paraffin-embedded Human skin tissue labeling Nectin-4 with ab322927 at 1/2000 (0.265 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Positive staining on human skin.
The section was incubated with ab322927 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).

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Nectin-4 antibody [RM1257] (ab322927)

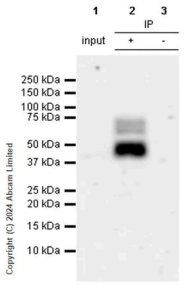
Immunohistochemical analysis of paraffin-embedded Human placenta tissue labeling Nectin-4 with ab322927 at 1/2000 (0.265 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Positive staining on human placenta.
The section was incubated with ab322927 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).

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins



Immunoprecipitation - Anti-Nectin-4 antibody [RM1257] (ab322927)

Nectin-4 was immunoprecipitated from 0.35 mg DLD-1 (human colorectal adenocarcinoma epithelial cell) whole cell lysate with ab322927 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab322927 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(ab131366) was used at 1/5000 dilution.

Lane 1: DLD-1 (human colorectal adenocarcinoma epithelial cell) whole cell lysate
Lane 2: ab322927 IP in DLD-1 (human colorectal adenocarcinoma epithelial cell) whole cell lysate
Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab322927 in DLD-1 whole cell lysate

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

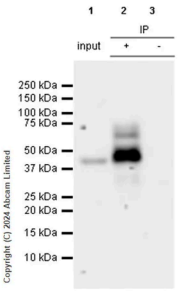
All lanes:
Immunoprecipitation - Anti-Nectin-4 antibody [RM1257] (ab322927) at 1/30 dilution

All lanes:
DLD-1 (human colorectal adenocarcinoma epithelial cell) whole cell lysate

Secondary

All lanes:
Immunoprecipitation - VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/5000 dilution

Exposure time: 50s



Immunoprecipitation - Anti-Nectin-4 antibody [RM1257] (ab322927)

Nectin-4 was immunoprecipitated from 0.35 mg T-47D whole cell lysate with ab322927 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab322927 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(ab131366) was used at 1/5000 dilution.

Lane 1: T-47D (human ductal breast epithelial tumor epithelial cell) whole cell lysate
Lane 2: ab322927 IP in T-47D whole cell lysate
Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab322927 in T-47D whole cell lysate

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

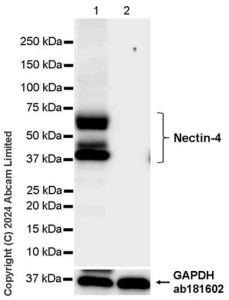
All lanes:
Immunoprecipitation - Anti-Nectin-4 antibody [RM1257] (ab322927) at 1/30 dilution

All lanes:
T-47D (human ductal breast epithelial tumor epithelial cell) whole cell lysate

Secondary

All lanes:
Immunoprecipitation - VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/5000 dilution

Exposure time: 15s



Western blot - Anti-Nectin-4 antibody [RM1257] (ab322927)

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

Low expression: Spleen (PMID: 11544254).

In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) staining at 1/200000 dilution.

All lanes:
Western blot - Anti-Nectin-4 antibody [RM1257] (ab322927) at 1/1000 dilution

Lane 1:
Human placenta tissue lysate at 20 µg

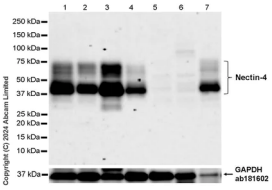
Lane 2:
Human spleen tissue lysate at 20 µg

Secondary

All lanes:
Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Observed band size: 40-70 kDa, 36 kDa

Exposure time: 180s



Western blot - Anti-Nectin-4 antibody [RM1257] (ab322927)

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

Low expression: HeLa and A549 (PMID: 21901103).

The expression profile/ molecular weight observed is consistent with what has been described in the literature (PMID: 15784625).

In Western blot, Anti-GAPDH antibody [EPR16891] - Loading Control (ab181602) staining at 1/200000 dilution.

All lanes:
Western blot - Anti-Nectin-4 antibody [RM1257] (ab322927) at 1/1000 dilution

Lane 1:
HT-29 (human colorectal adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Lane 2:
DLD-1 (human colorectal adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Lane 3:
T-47D (human ductal breast epithelial tumor epithelial cell) whole cell lysate at 20 µg

Lane 4:
MCF7 (human breast adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Lane 5:
HeLa (human cervical adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Lane 6:
A549 (human lung carcinoma epithelial cell) whole cell lysate at 20 µg

Lane 7:

A431 (human epidermoid carcinoma epithelial cell) whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Developed using the ECL technique.

Observed band size: 40-70 kDa, 36 kDa

Exposure time: 147s

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.