abcam

ab322963

# Anti-COL1A2 antibody [RM1295]

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** | RM1295 |
| **Purification technique** | Affinity purification Protein A |
| **Specificity** | Unsuitable for mouse IHC-P, FC and IP, rat IHC-P and IP. |
| **Concentration** | 0.481 - 0.496 mg/mL The concentration of this product may be batch-dependent<br>Batch concentration finder → |

## Reactivity data

### WB

#### Tested

| | |
|---|---|
| **Species** | Transfected cell lysate |
| **Dilution info** | 1/1000 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/1000 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Rat |
| **Dilution info** | 1/1000 |
| **Notes** | - |

### Expected

| | |
|---|---|
| **Species** | Mouse |
| **Dilution info** | Use at an assay dependent concentration. |
| **Notes** | - |

### Not recommended

| | |
|---|---|
| **Species** | Synthetic peptide - Human |
| **Dilution info** | - |
| **Notes** | - |

## Dot

### Tested

| | |
|---|---|
| **Species** | Synthetic peptide - Human |
| **Dilution info** | 1/1000 |
| **Notes** | - |

### Expected

| | |
|---|---|
| **Species** | Human, Rat, Mouse |
| **Dilution info** | Use at an assay dependent concentration. |
| **Notes** | - |

### Not recommended

| | |
|---|---|
| **Species** | Transfected cell lysate |
| **Dilution info** | - |
| **Notes** | - |

## IHC-P

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/10000 |
| **Notes** | Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |

### Not recommended

| | |
|---|---|
| **Species** | Mouse, Rat, Transfected cell lysate, Synthetic peptide - Human |
| **Dilution info** | - |
| **Notes** | - |

## Flow Cyt (Intra)

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/5000 |
| **Notes** | - |

### Expected

| | |
|---|---|
| **Species** | Rat |
| **Dilution info** | Use at an assay dependent concentration. |
| **Notes** | - |

### Not recommended

| | |
|---|---|
| **Species** | Mouse, Transfected cell lysate, Synthetic peptide - Human |

| | |
|---|---|
| **Dilution info** | - |
| **Notes** | - |

## IP

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/30 |
| **Notes** | - |

### Not recommended

| | |
|---|---|
| **Species** | Mouse, Rat, Transfected cell lysate, Synthetic peptide - Human |
| **Dilution info** | - |
| **Notes** | - |

## ICC/IF

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/100 |
| **Notes** | - |

| | |
|---|---|
| **Species** | Mouse |
| **Dilution info** | 1/100 |
| **Notes** | - |

### Expected

| | |
|---|---|
| **Species** | Rat |
| **Dilution info** | Use at an assay dependent concentration. |
| **Notes** | - |

### Not recommended

| | |
|---|---|
| **Species** | Transfected cell lysate, Synthetic peptide - Human |
| **Dilution info** | - |
| **Notes** | - |

## Target data

See full target information COL1A2

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

## 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

**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.

**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.

## 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.

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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.

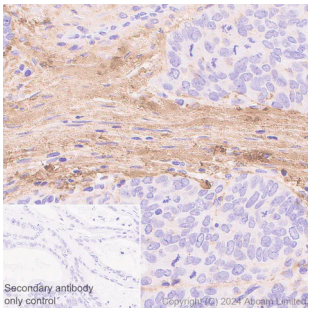
In the unlikely event of one of our products not working as expected, you are covered by our product promise.

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

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## 10 product images

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### Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-COL1A2 antibody [RM1295] (ab322963)

Immunohistochemical analysis of paraffin-embedded Human colon cancer tissue labeling COL1A2 with ab322963 at 1/10000 (0.05 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

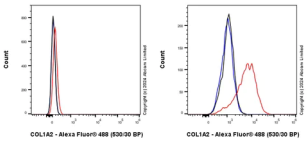
Positive staining on stromal cells of human colon cancer.
The section was incubated with ab322963 for 30 mins at room temperature.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument

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

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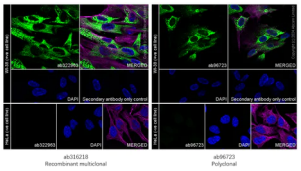


## Flow Cytometry (Intracellular) - Anti-COL1A2 antibody [RM1295] (ab322963)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized HeLa (human cervical adenocarcinoma epithelial cell, Left) / WI-38 (human fetal lung fibroblast, Right) cells labellng COL1A2 with ab322963 at 1/5000 dilution (0.01 ug) (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (Blue).

Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/5000 dilution was used as the secondary antibody.

Negative control: HeLa.



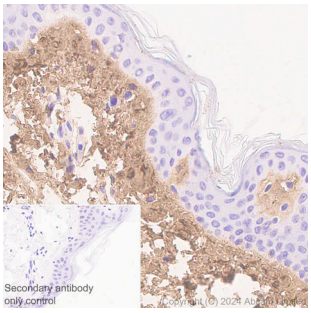
## Immunocytochemistry/ Immunofluorescence - Anti-COL1A2 antibody [RM1295] (ab322963)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized WI-38 (human fetal lung fibroblast) cells labelling COL1A2 with ab322963 at 1/100 (5.11 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 2ug/ml dilution (Green).

Confocal image showing cytoplasmic staining in WI-38 cell line (shown in green). The counterstain was observed in magenta. Nuclear DNA was labelled with DAPI (shown in blue).
Negative control: HeLa.
Left Panel is applied with Anti-COL1A2 antibody [RM1295] (ab322963) at 1/100 dilution (5.11 ug/ml);
Right panel is applied with Anti-COL1A2 antibody (ab96723) at 1/100 dilution (13.3 ug/ml).
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 2.5ug/ml dilution (Magenta). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 2ug/ml dilution.




## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-COL1A2 antibody [RM1295] (ab322963)

Immunohistochemical analysis of paraffin-embedded Human skin tissue labeling COL1A2 with ab322963 at 1/10000 (0.05 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

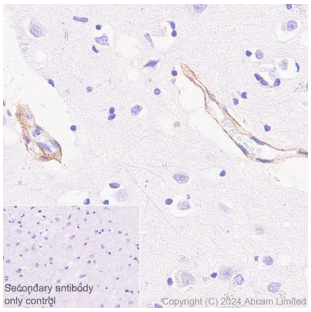
Positive staining on stromal cells of human skin.
The section was incubated with ab322963 for 30 mins at room temperature.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument

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

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## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-COL1A2 antibody [RM1295] (ab322963)

Immunohistochemical analysis of paraffin-embedded Human cerebrum tissue labeling COL1A2 with ab322963 at 1/10000 (0.05 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

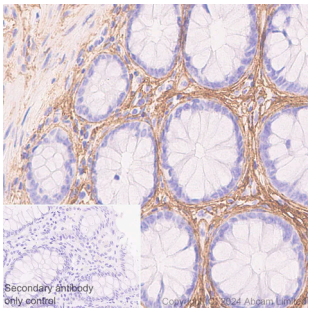
Low expression: positive staining on blood vessles of human cerebrum.
The section was incubated with ab322963 for 30 mins at room temperature.
The immunostaining was performed on a Leica Biosystems BOND® RX instrument

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

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## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-COL1A2 antibody [RM1295] (ab322963)

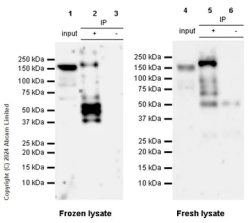
Immunohistochemical analysis of paraffin-embedded Human colon tissue labeling COL1A2 with ab322963 at 1/10000 (0.05 ug/ml) dilution, followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Positive staining on stromal cells of human colon.
The section was incubated with ab322963 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-COL1A2 antibody [RM1295] (ab322963)

COL1A2 was immunoprecipitated from 0.35 mg WI-38 (human fetal lung fibroblast) whole cell lysate with ab322963 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab322963 at 1/1000 dilution.

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

To minimize protein degradation, cells were lysed immediately after harvest and then applied for Immunoprecipitation as soon as possible in lanes 4-6.

Lanes 1-3: 180 seconds, Lanes 4-6: 58 seconds.

All lanes:
Immunoprecipitation - Anti-COL1A2 antibody [RM1295] (ab322963) at 1/1000 dilution

Lane 1:
(Input) WI-38 (human fetal lung fibroblast) whole cell lysate (Frozen) at 10 µg

Lanes 2 and 5:
ab322963 at 1/30 IP in WI-38 (human fetal lung fibroblast) whole cell lysate (Frozen) at 10 µg

Lane 3:
Rabbit monoclonal IgG (ab172730) instead of ab322963 in WI-38 whole cell lysate (Frozen) at 10 µg

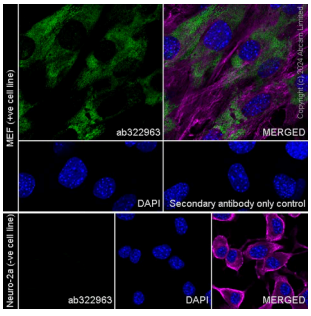
Lane 4:
(Input) WI-38 (human fetal lung fibroblast) whole cell lysate (Fresh) at 10 µg

Lane 6:
Rabbit monoclonal IgG (ab172730) instead of ab322963 in WI-38 whole cell lysate (Fresh) at 10 µg

Secondary

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

Observed band size: 160 kDa



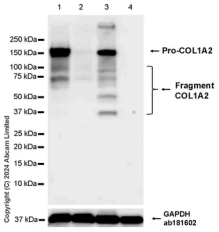

## Immunocytochemistry/ Immunofluorescence - Anti-COL1A2 antibody [RM1295] (ab322963)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized MEF (mouse embryo fibroblast) cells labelling COL1A2 with ab322963 at 1/100 (5.11 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 2ug/ml dilution (Green).

Confocal image showing cytoplasmic staining in MEF cell line (shown in green). The counterstain was observed in magenta. Nuclear DNA was labelled with DAPI (shown in blue).
Negative control: Neuro-2a.
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 2.5ug/ml dilution (Magenta). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 2ug/ml dilution.




## Western blot - Anti-COL1A2 antibody [RM1295] (ab322963)

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

Negative control: HeLa.

The expression profile/ molecular weight observed is consistent with what has been described in the literature (PMID: 23311625, PMID: 24599274 and PMID: 29162919).

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

All lanes:
Western blot - Anti-COL1A2 antibody [RM1295] (ab322963) at 1/1000 dilution

Lane 1:
WI-38 (human fetal lung fibroblast) transfected with scrambled siRNA control whole cell lysate at 20 µg

Lane 2:
WI-38 transfected with siRNA specifically targeting COL1A2 whole cell lysate at 20 µg

Lane 3:
IMR-90 (human lung fibroblast) whole cell lysate at 20 µg

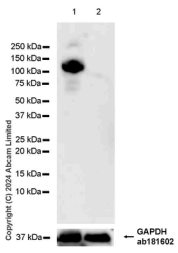
Lane 4:
HeLa (human cervical adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Observed band size: 30-160 kDa, 36 kDa

Exposure time: 26s




## Western blot - Anti-COL1A2 antibody [RM1295] (ab322963)

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

Low expression: brain.

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

All lanes:
Western blot - Anti-COL1A2 antibody [RM1295] (ab322963) at 1/1000 dilution

Lane 1:
Rat skin tissue lysate at 40 µg

Lane 2:
Rat brain tissue lysate at 40 µg

Secondary

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

Observed band size: 129 kDa, 36 kDa

Exposure time: 103s

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