Product datasheet

Goat Anti-Rabbit IgG H&L (HRP) ab205718

Overview

Product name: Goat Anti-Rabbit IgG H&L (HRP)
Host species: Goat
Target species: Rabbit
Specificity: The antibody used for conjugation reacts with rabbit immunoglobulins of all classes. Cross-reactions as determined by ELISA for the unconjugated antibody (ab182016): Mouse IgG, rat IgG, and chicken IgY, less than 2%. Human IgG, less than 7%.

Tested applications: Suitable for: IHC-P, WB, ELISA, IP
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.
Storage buffer: pH: 7.4
Preservative: 0.1% Proclin
Constituents: PBS, 1% BSA, 30% Glycerol
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

Our Abpromise guarantee covers the use of ab205718 in the following tested applications.
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

<table>
<thead>
<tr>
<th>Application</th>
<th>Abreviews</th>
<th>Notes</th>
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<tr>
<td>IHC-P</td>
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<td>1/2000 - 1/50000</td>
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### Application

<table>
<thead>
<tr>
<th>WB</th>
<th>Abreviews</th>
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<th>ELISA</th>
<th>Notes</th>
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<td>Use at an assay dependent concentration.</td>
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<th>IP</th>
<th>Notes</th>
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### Images

**All lanes**: Anti-beta Actin antibody (ab8227) 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-Rabbit IgG H&L (HRP) (ab205718) at 1/50000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

**Observed band size**: 42 kDa

*why is the actual band size different from the predicted?*

**Exposure time**: 30 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 ab8227 overnight at
4°C. Antibody binding was detected using ab205718, and visualised using ECL development solution ab133406.

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 ab177840 at 1/1000 dilution. An HRP-conjugated secondary (Ab205718, 1/20000 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

Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab205718)

<table>
<thead>
<tr>
<th>Lanes</th>
<th>Sample Description</th>
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<tbody>
<tr>
<td>1</td>
<td>Liver (Mouse) Tissue Lysate</td>
</tr>
<tr>
<td>2</td>
<td>Liver (Rat) Tissue Lysate</td>
</tr>
</tbody>
</table>

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : ab205718 (Left Image) at 1/20,000 and a competitor secondary (Right Image) at 1/50,000. Notice the increased background of the competitor product.

Performed under reducing conditions.

Observed band size: 42 kDa why is the actual band size different from the predicted?

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 ab8227 overnight at 4°C. Antibody binding was detected using ab205718 (Left Image) and a competitor secondary (Right Image), and visualised using ECL development solution ab133406.

All lanes: No Primary Antibody

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-Rabbit IgG H&L (HRP) (ab205718) at 1/50000 dilution

Performed under reducing conditions.

Exposure time: 20 minutes

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 only the secondary antibody (ab205718), and visualised using ECL development solution ab133406.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Goat Anti-Rabbit IgG H&L (HRP) (ab205718)

IHC image of beta 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 ab6046 at 1/100 dilution. An HRP-conjugated secondary (Ab205718, 1/20000 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.

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Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab205718)

All lanes : No Primary Antibody

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

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : ab205718 (Left Image) 1/50,000 and a competitor secondary (Right Image) 1/50,000. Notice the increased background of the competitor product.

Exposure time: 20 minutes

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 ab205718 (Left Image) and a competitor secondary (Right Image), and visualised using ECL development solution ab133406.

**All lanes**: Anti-STAT3 antibody [EPR787Y] (ab68153) at 1/2000 dilution

**Lane 1**: A431 (Human epithelial carcinoma cell line) Whole Cell Lysate
**Lane 2**: Heart (Mouse) Tissue Lysate
**Lane 3**: Heart (Rat) Tissue Lysate

Lysates/proteins at 10 µg per lane.

**Secondary**

**All lanes**: Goat Anti-Rabbit IgG H&L (HRP) (ab205718) at 1/2000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

**Observed band size**: 88 kDa *why is the actual band size different from the predicted?*

**Exposure time**: 20 minutes

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 3% milk before being incubated with ab68153 overnight at 4°C. Antibody binding was detected using ab205718, and visualised using ECL development solution ab133406.
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Goat Anti-Rabbit IgG H&L (HRP) (ab205718)

IHC image of Ki67 staining in a section of formalin-fixed paraffin-embedded normal human colon tissue*. The section was pretreated using pressure cooker heat mediated antigen retrieval with sodium citrate buffer (pH6) for 30mins, and incubated overnight at +4°C with ab15580 at 1/1000 dilution. An HRP-conjugated secondary (Ab205718, 1/20000 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.

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