# abcam

#### Product datasheet

## Donkey Anti-Goat IgG H&L (HRP) ab205723

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Overview

Product name Donkey Anti-Goat IgG H&L (HRP)

Host species Donkey

Target species Goat

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

reactions as determined by ELISA for the unconjugated antibody (ab182021): Human IgG, mouse

lgG, rat lgG, rabbit lgG and chicken lgY, less than 2%.

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

**Immunogen** Full length protein corresponding to Goat lgG.

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

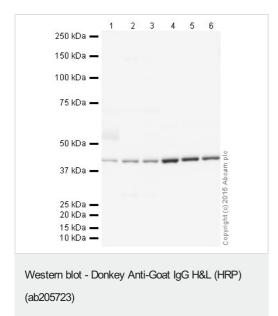
**Applications** 

The Abpromise guarantee Our Abpromise guarantee covers the use of ab205723 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
IP		Use at an assay dependent concentration.
WB	**** <u>(1)</u>	1/2000 - 1/20000.
IHC-P		1/2000 - 1/20000.
ELISA		Use at an assay dependent concentration.

#### **Images**



**All lanes :** Anti-beta Actin antibody - Loading Control (ab8229) at 1  $\mu g/ml$ 

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 :** Donkey Anti-Goat lgG H&L (HRP) (ab205723) at 1/10000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Observed band size: 42 kDa

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 <u>ab8229</u> overnight at 4°C. Antibody binding was detected using ab205723, and visualised using ECL development solution <u>ab133406</u>.

1 2 3 4 5 6

250 kDa —

150 kDa —

100 kDa —

75 kDa —

37 kDa —

25 kDa —

25 kDa —

15 kDa —

10 kDa —

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

(ab205723)

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 :** Donkey Anti-Goat lgG H&L (HRP) (ab205723) at 1/2000 dilution

Performed under reducing conditions.

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 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 (ab205723), and visualised using ECL development solution ab133406.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Donkey Anti-Goat IgG H&L (HRP) (ab205723)

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 1ug/ml. Ab98800, goat anti-mouse lgG, was then added as a secondary bridging antibody, at 1/250 dilution for 1h.

An HRP-conjugated secondary (Ab205723, 1/2000 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

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