# abcam

### Product datasheet

## Anti-HMGB2 antibody [EPR6302] ab133540





#### 2 References 7 Images

#### Overview

**Product name** Anti-HMGB2 antibody [EPR6302]

**Description** Rabbit monoclonal [EPR6302] to HMGB2

**Host species** Rabbit

**Tested applications** Suitable for: WB, IHC-P

Unsuitable for: ICC/IF or IP

Species reactivity Reacts with: Mouse, Rat, Human

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

Positive control WB: HEK-293T, HAP1, K562, HL-60, C6, RAW 264.7, and PC-12 cell lysates. IHC-P: Human

breast tissue and Human colon carcinoma tissue.

**General notes** 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.

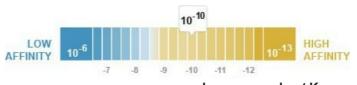
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

#### **Properties**

**Form** Liquid

Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C. Storage instructions

 $K_D = 2.97 \times 10^{-10} M$ Dissociation constant (K<sub>D</sub>)



Learn more about K<sub>D</sub>

Storage buffer pH: 7.2

Preservative: 0.05% Sodium azide

Constituents: 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue culture

supernatant

Purity Protein A purified

ClonalityMonoclonalClone numberEPR6302

**Isotype** IgG

#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab133540 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		1/1000 - 1/10000. Predicted molecular weight: 24 kDa.
IHC-P		1/250 - 1/500. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

**Application notes** Is unsuitable for ICC/IF or IP.

#### **Target**

**Function** DNA binding proteins that associates with chromatin and has the ability to bend DNA. Binds

preferentially single-stranded DNA. Involved in V(D)J recombination by acting as a cofactor of the RAG complex. Acts by stimulating cleavage and RAG protein binding at the 23 bp spacer of

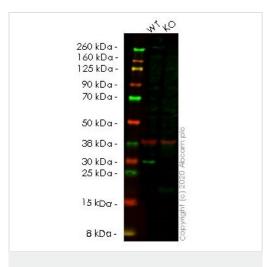
conserved recombination signal sequences (RSS).

Sequence similarities Belongs to the HMGB family.

Contains 2 HMG box DNA-binding domains.

**Cellular localization** Nucleus. Chromosome.

#### **Images**



Western blot - Anti-HMGB2 antibody [EPR6302] (ab133540)

**All lanes :** Anti-HMGB2 antibody [EPR6302] (ab133540) at 1/1000 dilution

Lane 1: Wild-type HEK-293T cell lysate

Lane 2: HMGB2 knockout HEK-293T cell lysate

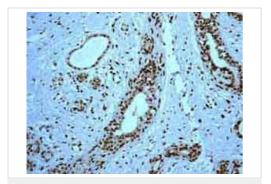
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 24 kDa **Observed band size:** 24 kDa

**Lanes 1-2:** Merged signal (red and green). Green - ab133540 observed at 24 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (ab8245) observed at 37 kDa.

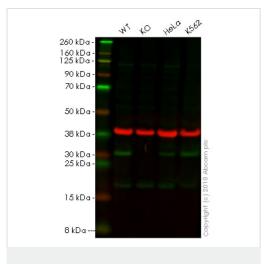
ab133540 was shown to react with HMGB2 in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line <a href="mailto:ab266358">ab266358</a> (knockout cell lysate <a href="mailto:ab257156">ab257156</a>) was used. Wild-type HEK-293T and HMGB2 knockout HEK-293T cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab133540 and Anti-GAPDH antibody [6C5] - Loading Control (<a href="mailto:ab8245">ab8245</a>) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye®800CW) preadsorbed (<a href="mailto:ab216773">ab216773</a>) and Goat anti-Mouse lgG H&L (IRDye®680RD) preadsorbed (<a href="mailto:ab216776">ab216776</a>) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-HMGB2 antibody
[EPR6302] (ab133540)

Immunohistochemistry analysis of paraffin embedded Human breast tissue labelling HMGB2 with ab133540 at 1/250 dilution.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Western blot - Anti-HMGB2 antibody [EPR6302] (ab133540)

**All lanes :** Anti-HMGB2 antibody [EPR6302] (ab133540) at 1/1000 dilution

Lane 1: Wild-type HAP1 whole cell lysate

Lane 2: HMGB2 knockout HAP1 whole cell lysate

Lane 3 : HeLa cell lysate Lane 4 : K562 cell lysate

Lysates/proteins at 20 µg per lane.

**Predicted band size:** 24 kDa **Observed band size:** 24 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - ab133540 observed at 24 kDa. Red - loading control, **ab9484**, observed at 37 kDa.

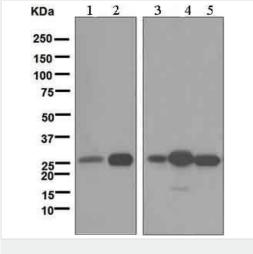
ab133540 was shown to recognize HMGB2 in wild-type HAP1 cells as signal was lost at the expected MW in HMGB2 knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and HMGB2 knockout samples were subjected to SDS-PAGE. Ab133540 and ab9484 (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-HMGB2 antibody
[EPR6302] (ab133540)

Immunohistochemistry analysis of paraffin embedded Human colon carcinoma tissue labelling HMGB2 with ab133540 at 1/250 dilution.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Western blot - Anti-HMGB2 antibody [EPR6302] (ab133540)

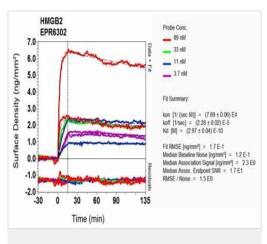
**All lanes :** Anti-HMGB2 antibody [EPR6302] (ab133540) at 1/1000 dilution

Lane 1 : K562 cell lysate
Lane 2 : HL-60 cell lysate
Lane 3 : C6 cell lysate

Lane 4: RAW 264.7 cell lysate
Lane 5: PC-12 cell lysate

Lysates/proteins at 10 µg per lane.

Predicted band size: 24 kDa



Ol-RD Scanning - Anti-HMGB2 antibody [EPR6302] (ab133540)

Equilibrium disassociation constant ( $K_D$ ) Learn more about  $K_D$ 

Click here to learn more about K<sub>D</sub>



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