

## Product datasheet

# Human ENG (CD105) knockout HeLa cell lysate ab256906

5 Images

### Overview

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<b>Product name</b>	Human ENG (CD105) knockout HeLa cell lysate
<b>Product overview</b>	Knockout cell lysate achieved by CRISPR/Cas9.
<b>Parental Cell Line</b>	HeLa
<b>Organism</b>	Human
<b>Mutation description</b>	Knockout achieved by using CRISPR/Cas9, 11 bp deletion in exon2 and 19 bp deletion in exon2 and 1 bp insertion in exon2.
<b>Passage number</b>	<20
<b>Knockout validation</b>	Sanger Sequencing, Western Blot (WB)
<b>Reconstitution notes</b>	To use as WB control, resuspend the lyophilizate in 50 µL of LDS* Sample Buffer to have a final concentration of 2 mg/ml. For reducing conditions, we recommend a final concentration of 0.1 M DTT. <i>*Usage of SDS sample buffer is not recommended with these lyophilized lysates.</i>

### Notes

**Lysate preparation:** Our lysates are made using RIPA buffer to which we add a protease inhibitor cocktail and phosphatase inhibitor cocktail (ratio: 300:100:10). *This means that the protein of interest is denatured.* If you require a native form of the protein please use the live cell version - found [here](#). Please refer to our lysis protocol for further details on how our lysates are prepared.

**User storage instructions:** Lyophilizate may be stored at 4°C. After reconstitution, store at -20°C for short-term storage or -80°C for long-term storage.

Access thousands of knockout cell lysates, generated from commonly used cancer cell lines. [See here for more information on knockout cell lysates.](#)

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Tested applications

Suitable for: WB

## Properties

**Storage instructions** Store at -80°C. Please refer to protocols.

Components	1 kit
ab261947 - Human ENG knockout HeLa cell lysate	1 x 100µg
ab255929 - Human wild-type HeLa cell lysate	1 x 100µg

**Cell type** epithelial  
**Disease** Adenocarcinoma  
**Gender** Female  
**STR Analysis** Amelogenin X D5S818: 11, 12 D13S317: 12, 13.3 D7S820: 8, 12 D16S539: 9, 10 vWA: 16, 18 TH01: 7 TPOX: 8,12 CSF1PO: 9, 10

## Target

**Function** Major glycoprotein of vascular endothelium. May play a critical role in the binding of endothelial cells to integrins and/or other RGD receptors.

**Tissue specificity** Endoglin is restricted to endothelial cells in all tissues except bone marrow.

**Involvement in disease** Defects in ENG are the cause of hereditary hemorrhagic telangiectasia type 1 (HHT1) [MIM:187300, 108010]; also known as Osler-Rendu-Weber syndrome 1 (ORW1). HHT1 is an autosomal dominant multisystemic vascular dysplasia, characterized by recurrent epistaxis, muco-cutaneous telangiectases, gastro-intestinal hemorrhage, and pulmonary (PAVM), cerebral (CAVM) and hepatic arteriovenous malformations; all secondary manifestations of the underlying vascular dysplasia. Although the first symptom of HHT1 in children is generally nose bleed, there is an important clinical heterogeneity.

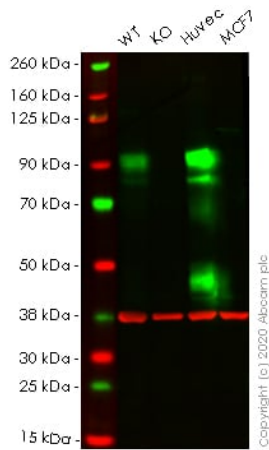
**Cellular localization** Membrane.

## Applications

**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab256906 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		Use at an assay dependent concentration.

## Images



Western blot - Human ENG knockout HeLa cell lysate (ab256906)

**Lane 1:** Wild-type HeLa cell lysate (20 µg)

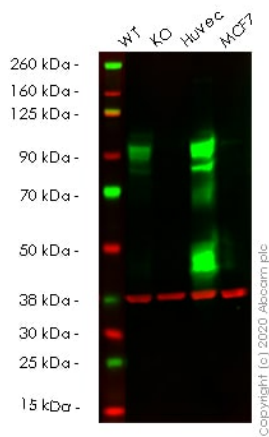
**Lane 2:** ENG knockout HeLa cell lysate (20 µg)

**Lane 3:** HUVEC cell lysate (20 µg)

**Lane 4:** MCF7 cell lysate (20 µg)

**Lanes 1-4:** Merged signal (red and green). Green - **ab170943** observed at 70-120 kDa. Red - loading control **ab8245** observed at 37 kDa.

**ab170943** Anti-CD105 antibody [EPR10145-10] was shown to specifically react with CD105 in wild-type HeLa cells. Loss of signal was observed when knockout cell line **ab265178** (knockout cell lysate ab256906) was used. Wild-type and CD105 knockout samples were subjected to SDS-PAGE. **ab170943** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Human ENG knockout HeLa cell lysate (ab256906)

**Lane 1:** Wild-type HeLa cell lysate (20 µg)

**Lane 2:** ENG knockout HeLa cell lysate (20 µg)

**Lane 3:** HUVEC cell lysate (20 µg)

**Lane 4:** MCF7 cell lysate (20 µg)

**Lanes 1-4:** Merged signal (red and green). Green - **ab169545** observed at 70-120 kDa. Red - loading control **ab8245** observed at 37 kDa.

**ab169545** Anti-CD105 antibody [EPR10145-12] was shown to specifically react with CD105 in wild-type HeLa cells. Loss of signal was observed when knockout cell line **ab265178** (knockout cell lysate ab256906) was used. Wild-type and CD105 knockout samples were subjected to SDS-PAGE. **ab169545** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

```

Mut  CCAACAGGTCTTGCAGAAACAGTCCATTGT-----CCGAGAGGGGC
      |||
WT   CCAACAGGTCTTGCAGAAACAGTCCATTGTGACCTTCAGCCTGTGGGCCCCGAGAGGGGC

```

Allele-1: 19 bp deletion in exon2

Sanger Sequencing - Human ENG knockout HeLa  
cell lysate (ab256906)

```

Mut  CCAACAGGTCTTGCAGAAACAGTCCATTGT-----TGTGGGCCCCGAGAGGGGC
      |||
WT   CCAACAGGTCTTGCAGAAACAGTCCATTGTGACCTTCAGCCTGTGGGCCCCGAGAGGGGC

```

Allele-2: 11 bp deletion in exon2

Sanger Sequencing - Human ENG knockout HeLa  
cell lysate (ab256906)

```

Mut  CCAACAGGTCTTGCAGAAACAGTCCATTGTGACCTTCAGCCTGTGGGCCCCGAGAGGGG
      |||
WT   CCAACAGGTCTTGCAGAAACAGTCCATTGTGACCTTCAGCCTGTGGGCCCCGAGAGGGG

```

Allele-3: 1 bp insertion in exon2

Sanger Sequencing - Human ENG knockout HeLa  
cell lysate (ab256906)

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