

## DESCRIPTION

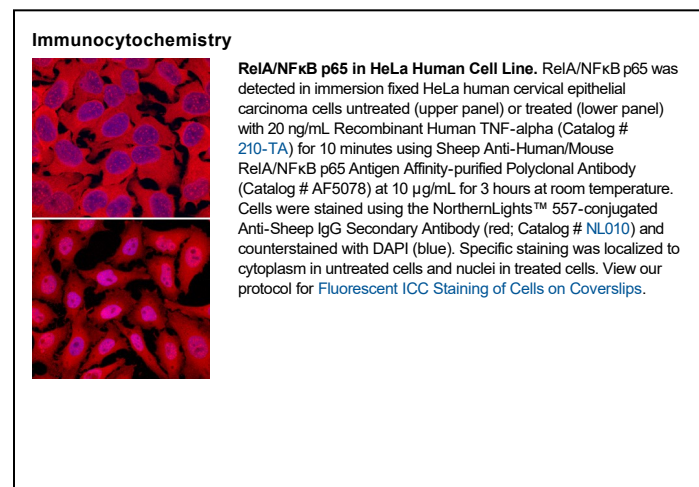
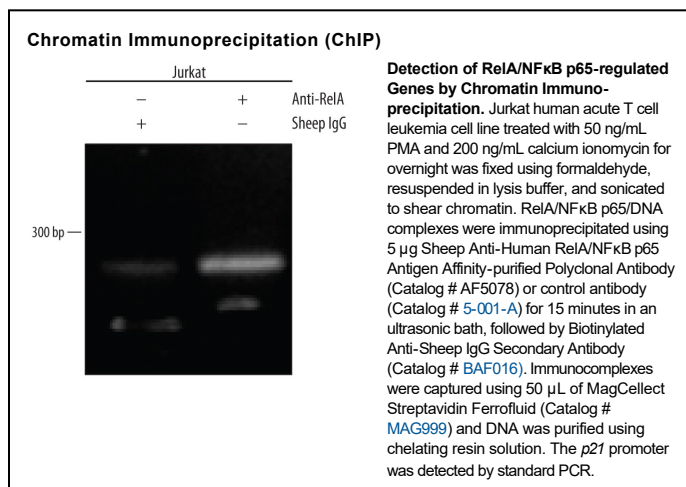
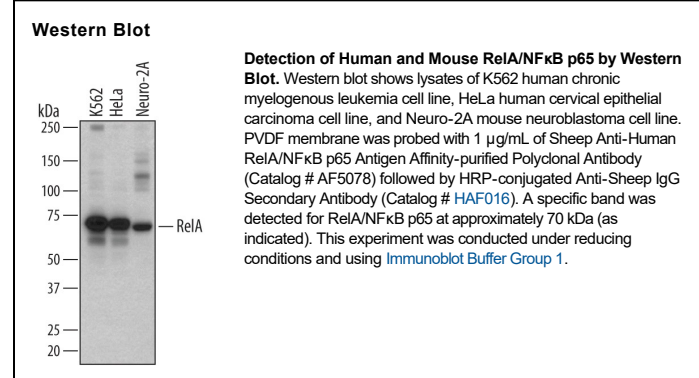
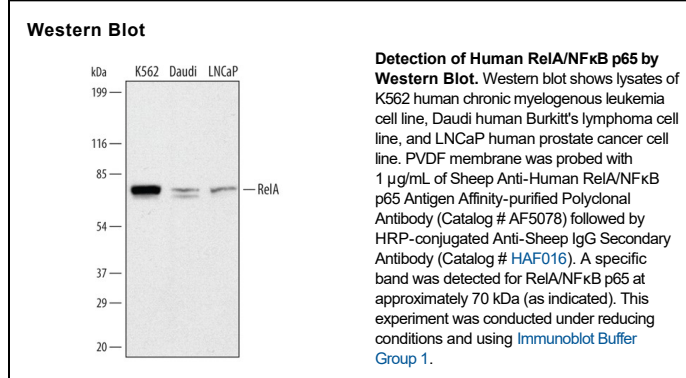
<b>Species Reactivity</b>	Human/Mouse
<b>Specificity</b>	Detects human and mouse RelA/NFκB p65 in Western blots.
<b>Source</b>	Polyclonal Sheep IgG
<b>Purification</b>	Antigen Affinity-purified
<b>Immunogen</b>	<i>E. coli</i> -derived recombinant human RelA/NFκB p65 isoform 1 Asn456-Ser551 Accession # Q04206
<b>Formulation</b>	Lyophilized from a 0.2 μm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 μm filtered solution in PBS.

## APPLICATIONS

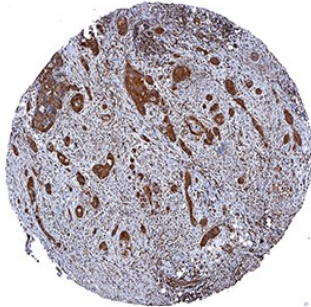
**Please Note:** Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

	Recommended Concentration	Sample
<b>Western Blot</b>	1 μg/mL	See Below
<b>Chromatin Immunoprecipitation (ChIP)</b>	5 μg/5 x 10 <sup>6</sup> cells	See Below
<b>Immunocytochemistry</b>	5-15 μg/mL	See Below
<b>Immunohistochemistry</b>	5-15 μg/mL	See Below
<b>Simple Western</b>	10 μg/mL	See Below
<b>Knockout Validated</b>	RelA/NFκB p65 is specifically detected in HeLa human cervical epithelial carcinoma parental cell line but is not detectable in RelA/NFκB p65 knockout HeLa cell line.	

## DATA

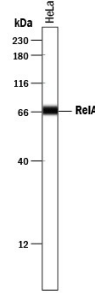


## Immunohistochemistry



**RelA/NFκB p65 in Human Squamous Cell Carcinoma.** RelA/NFκB p65 was detected in immersion fixed paraffin-embedded sections of human squamous cell carcinoma using Sheep Anti-Human/Mouse RelA/NFκB p65 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF5078) at 3 μg/mL overnight at 4 °C. Tissue was stained using the Anti-Sheep HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS019) and counterstained with hematoxylin (blue). Specific staining was localized to cytoplasm in cancer cells. View our protocol for [Chromogenic IHC Staining of Paraffin-embedded Tissue Sections](#).

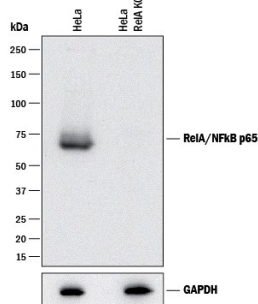
## Simple Western



**Detection of Human RelA/NFκB p65 by Simple Western™.** Simple Western lane view shows lysates of HeLa human cervical epithelial carcinoma cell line, loaded at 0.2 mg/mL. A specific band was detected for RelA/NFκB p65 at approximately 68 kDa (as indicated) using 10 μg/mL of Sheep Anti-Human/Mouse RelA/NFκB p65 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF5078) followed by 1:50 dilution of HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # HAF016). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.



## Knockout Validated



**Western Blot Shows Human RelA/NFκB p65 Specificity by Using Knockout Cell Line.** Western blot shows lysates of HeLa human cervical epithelial carcinoma parental cell line and RelA/NFκB p65 knockout HeLa cell line (KO). PVDF membrane was probed with 1 μg/mL of Sheep Anti-Human/Mouse RelA/NFκB p65 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF5078) followed by HRP-conjugated Anti-Sheep IgG Secondary Antibody (Catalog # HAF016). A specific band was detected for RelA/NFκB p65 at approximately 65 kDa (as indicated) in the parental HeLa cell line, but is not detectable in knockout HeLa cell line. GAPDH (Catalog # AF5718) is shown as a loading control. This experiment was conducted under reducing conditions and using [Immunoblot Buffer Group 1](#).

## PREPARATION AND STORAGE

<b>Reconstitution</b>	Reconstitute at 0.2 mg/mL in sterile PBS.
<b>Shipping</b>	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
<b>Stability &amp; Storage</b>	<p><b>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</b></p> <ul style="list-style-type: none"> <li>● 12 months from date of receipt, -20 to -70 °C as supplied.</li> <li>● 1 month, 2 to 8 °C under sterile conditions after reconstitution.</li> <li>● 6 months, -20 to -70 °C under sterile conditions after reconstitution.</li> </ul>

## BACKGROUND

RelA belongs to a family of transcription factors (NFκB (nuclear factor kappa from B cells) complex) that play a fundamental role in inflammatory and immune responses. The NFκB complex is composed of a heterodimer of a Rel family member (RelA, c-Rel, RelB) and either NFκB1 or NFκB2 subunits. RelA and NFκB1 are the most common heterodimeric pair. The NFκB complex is sequestered in the cytoplasm by inhibitory IκB proteins. Upon cellular activation, the ubiquitin-proteasome pathway degrades the IκB proteins allowing the NFκB complex to translocate to the nucleus and activate gene transcription.