

Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free ab167611

Recombinant RabMAb

[1 References](#) [12 Images](#)

Overview

Product name	Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free
Description	Rabbit monoclonal [E115] to Estrogen Receptor alpha - Low endotoxin, Azide free
Host species	Rabbit
Specificity	Expression levels of ER alpha protein vary with sample type. This antibody is unsuitable to test ovary and the tissues with low expression level of Estrogen Receptor alpha, such as kidney, lung and brain, in western blot. And it failed to show good IHC signal on mouse and rat tissue sections when using our IHC testing conditions. For our in-house testing we tested the antibody on a mouse tissue array (breast, spleen, lung, stomach, muscle, pancreas, liver, colon, brain, kidney).
Tested applications	Suitable for: WB, ChIP, IHC-P, Flow Cyt (Intra), ICC/IF
Species reactivity	Reacts with: Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	ICC/IF: MCF-7 cells, 4T1 cells and GH3 cells.
General notes	<p>ab167611 is the carrier-free version of ab32063.</p> <p>Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.</p> <p>This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.</p> <p>Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.</p> <p>This product is compatible with the Maxpar[®] Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar[®] is a trademark of Fluidigm Canada Inc.</p> <p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> - 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](#).

Our **Low endotoxin, azide-free formats** have low endotoxin level (≤ 1 EU/ml, determined by the LAL assay) and are free from azide, to achieve consistent experimental results in functional assays.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	E115
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab167611 in the following tested applications.

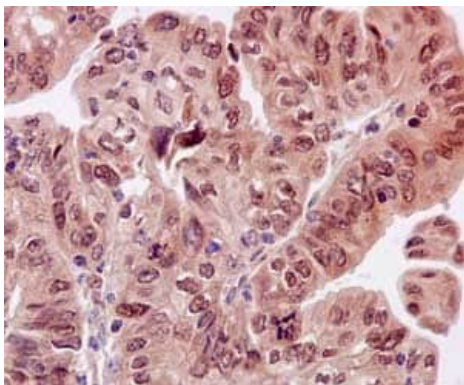
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		Use at an assay dependent concentration.
ChIP		Use at an assay dependent concentration. Please refer to the original abID, ab32063 , for information on recommended dilutions.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Please refer to the original abID, ab32063 , for information on recommended dilutions.
Flow Cyt (Intra)		Use at an assay dependent concentration. Please refer to the original abID, ab32063 , for information on recommended dilutions.
ICC/IF		Use at an assay dependent concentration. Please refer to the original abID, ab32063 , for information on recommended dilutions.

Target

Function	Nuclear hormone receptor. The steroid hormones and their receptors are involved in the regulation of eukaryotic gene expression and affect cellular proliferation and differentiation in target tissues. Can activate the transcriptional activity of TFF1.
Sequence similarities	Belongs to the nuclear hormone receptor family. NR3 subfamily. Contains 1 nuclear receptor DNA-binding domain.
Domain	Composed of three domains: a modulating N-terminal domain, a DNA-binding domain and a C-terminal ligand-binding domain.
Post-translational modifications	Phosphorylated by cyclin A/CDK2. Phosphorylation probably enhances transcriptional activity. Glycosylated; contains N-acetylglucosamine, probably O-linked. Ubiquitinated. Deubiquitinated by OTUB1. Dimethylated by PRMT1 at Arg-260. The methylation may favor cytoplasmic localization. Palmitoylated (isoform 3). Not biotinylated (isoform 3).
Cellular localization	Nucleus. Cytoplasm. Cell membrane. A minor fraction is associated with the inner membrane and Nucleus. Cytoplasm. Cell membrane. Associated with the inner membrane via palmitoylation.

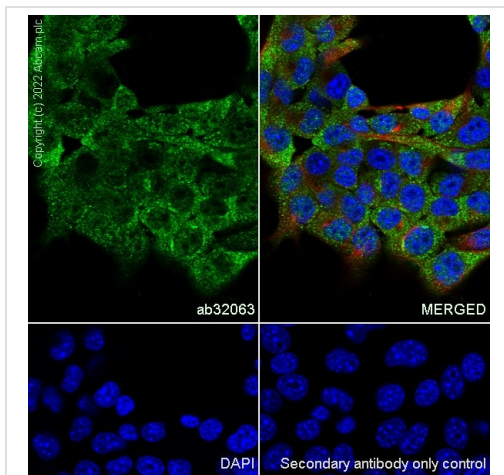
Images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free (ab167611)

Immunohistochemical staining of paraffin embedded human endometrial carcinoma with purified [ab32063](#) at a working dilution of 1 in 200. The secondary antibody used is [ab97051](#), a HRP goat anti-rabbit IgG (H+L), at 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide ([ab32063](#)).

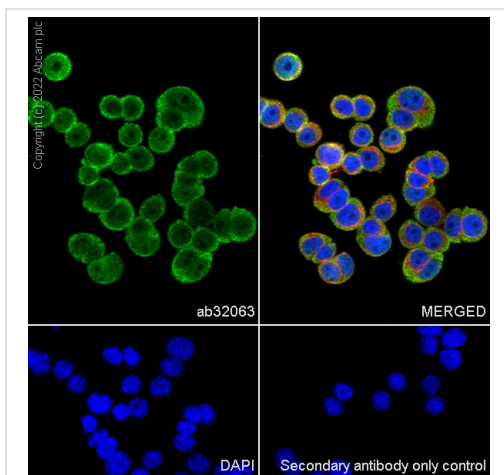


Immunocytochemistry/ Immunofluorescence - Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free (ab167611)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized 4T1 (mouse mammary gland carcinoma epithelial cell line) cells labelling Estrogen Receptor alpha with primary antibody anti-Estrogen Receptor alpha (**ab32063**) at 1/200 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150081**) secondary antibody at 1/1000 dilution (2.0 µg/mL). Confocal image showing cytoplasmic and nuclear staining in 4T1 cells. Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) (**ab195889**) was used to counterstain tubulin at 1/200 dilution (2.5 µg/mL). The nuclear counter stain is DAPI (blue).

The secondary antibody only control is : Secondary antibody is **ab150081** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution (2.0 µg/mL).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32063**).

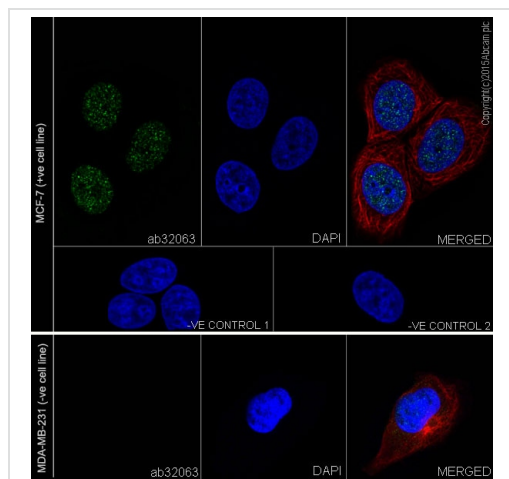


Immunocytochemistry/ Immunofluorescence - Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free (ab167611)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized GH3 (rat pituitary epithelial cell line) cells labelling Estrogen Receptor alpha with primary antibody anti-Estrogen Receptor alpha (**ab32063**) at 1/200 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150081**) secondary antibody at 1/1000 dilution (2.0 µg/mL). Confocal image showing cytoplasmic and nuclear staining in GH3 cells. Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) (**ab195889**) was used to counterstain tubulin at 1/200 dilution (2.5 µg/mL). The nuclear counter stain is DAPI (blue).

The secondary antibody only control is : Secondary antibody is **ab150081** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution (2.0 µg/mL).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32063**).



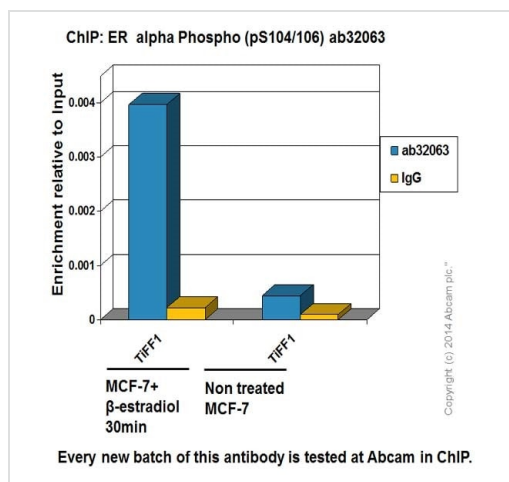
Immunocytochemistry/ Immunofluorescence - Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free (ab167611)

Immunocytochemistry/Immunofluorescence analysis of MCF-7 cells labelling Estrogen Receptor alpha with purified **ab32063** at 1/1000. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody. The cells were co-stained with **ab7291**, a mouse anti-tubulin (1/1000) using **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/1000) as the secondary antibody. Nuclei counterstained with DAPI (blue).

Control 1: primary antibody (1/1000) and secondary antibody, **ab150120**, an Alexa Fluor® 594-conjugated goat anti-mouse IgG (1/1000).

Control 2: **ab7291** (1/1000) and secondary antibody, **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32063**).



ChIP - Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free (ab167611)

Chromatin was prepared from MCF-7+ β -estradiol 30 min, and MCF-7 cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10 min. The ChIP was performed with 25 μ g of chromatin, 4 μ g of purified **ab32063** (blue), and 20 μ LI of anti-rabbit IgG sepharose beads. Rabbit normal IgG was added to the beads control (yellow). The immunoprecipitated DNA was quantified by real time PCR (SYBR approach). Primers are located in the 2nd intron of TFF1 gene.

MCF7 Cells were treated as below:

MCF-7 starved overnight, then treated with 10 nM β -Estradiol in 2% FBS media for 30 min.

Control MCF-7 was starved overnight, then in 2% FBS media for 30 mins.

Primer information:

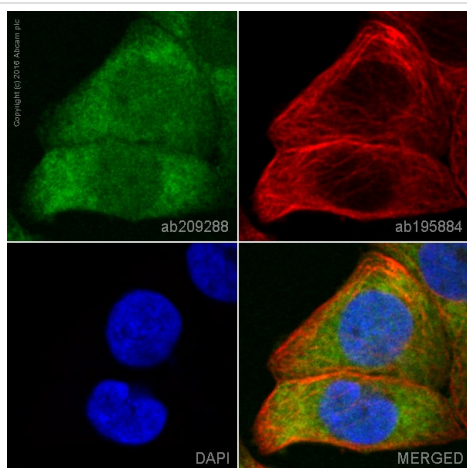
Located to the 2 intron of TFF1 gene.

Sequence:

Forward: 5' -agtctctccaacctgacctt-3'

Reverse: 5' -ttccggccatctctcactat-3'

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32063**).

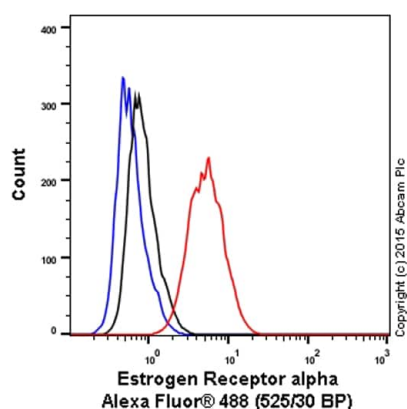


Immunocytochemistry/ Immunofluorescence - Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free (ab167611)

Clone E115 (ab167611) has been successfully conjugated by Abcam. This image was generated using Anti-Estrogen Receptor alpha antibody [E115] (PE). Please refer to **ab209288** for protocol details.

ab209288 staining Estrogen Receptor alpha in T47D cells. The cells were fixed with 4% formaldehyde (10 min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with **ab209288** at 1/500 dilution (Pseudocolored in green) and **ab195884**, Rat monoclonal to Tubulin (Alexa Fluor® 647), at 1/250 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



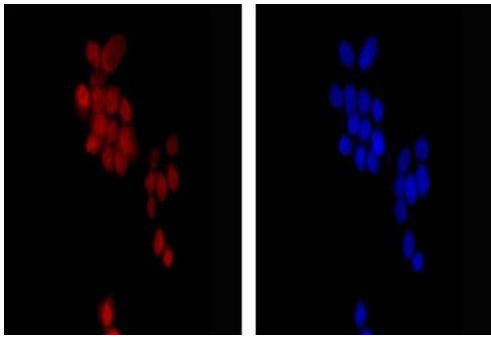
Flow Cytometry (Intracellular) - Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free (ab167611)

Clone E115 (ab167611) has been successfully conjugated by Abcam. This image was generated using Anti-Estrogen Receptor alpha antibody [E115] (Alexa Fluor® 488). Please refer to **ab194150** for protocol details.

Overlay histogram showing T47D cells stained with **ab194150** (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (**ab194150**, 1/50 dilution) for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) Alexa Fluor® 488 (**ab199091**) used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.

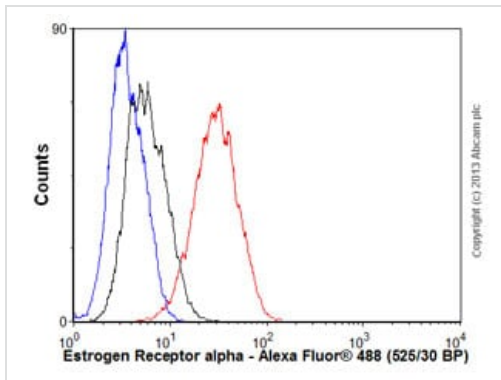
This antibody gave a positive signal in T47D cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



Immunocytochemistry/ Immunofluorescence - Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free (ab167611)

Immunofluorescent staining of MCF7 cells (fixed with 4% PFA and permeabilized with TritonX 100) with purified **ab32063** at a dilution of 1/250. An Alexa Fluor® 555 goat anti-rabbit antibody was used as the secondary at a dilution of 1/200. The panel on the right shows the DAPI counter-staining.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32063**).

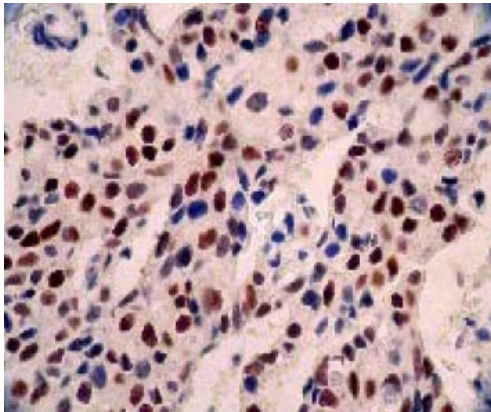


Flow Cytometry (Intracellular) - Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free (ab167611)

Overlay histogram showing MCF7 cells stained with unpurified **ab32063** (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (**ab32063**, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H+L) (**ab150077**) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10⁶ cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control.

Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in MCF7 cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

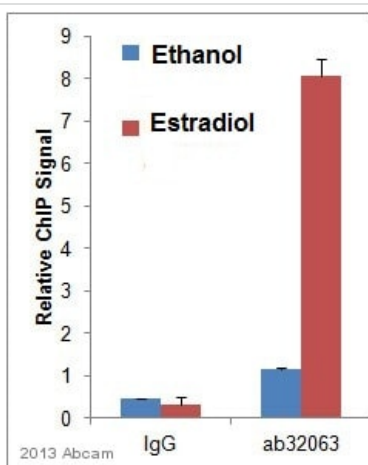
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32063**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free (ab167611)

Immunohistochemical analysis of human breast carcinoma using anti-Estrogen Receptor alpha ([ab32063](#), unpurified) diluted 1:50

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide ([ab32063](#)).



ChIP - Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free (ab167611)

This image is courtesy of an anonymous Abreview.

ChIP analysis using unpurified [ab32063](#) binding Estrogen Receptor alpha in MCF7 cells derived from Human breast carcinoma. Cells were cross-linked for 10 minutes with 1% formaldehyde. Samples were incubated with undiluted primary antibody for 16 hours at 4°C. Protein binding was detected using real-time PCR.

Positive control: Estrogen treated MCF7 cells tested at PS2 promoter.

Negative Control: IgG ChIP and ethanol-depleted cells tested at PS2 promoter.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide ([ab32063](#)).

Why choose a recombinant antibody?



Research with confidence
Consistent and reproducible results



Long-term and scalable supply
Recombinant technology



Success from the first experiment
Confirmed specificity



Ethical standards compliant
Animal-free production

Anti-Estrogen Receptor alpha antibody [E115] - Low endotoxin, Azide free (ab167611)

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