abcam

Product datasheet

Anti-Progesterone Receptor antibody [Alpha PR6] - ChIP Grade ab2765

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Overview

Product name	Anti-Progesterone Receptor antibody [Alpha PR6] - ChIP Grade
Description	Mouse monoclonal [Alpha PR6] to Progesterone Receptor - ChIP Grade
Specificity	Detects the B form of the progesterone receptor (PR). This antibody does not cross-react with estrogen receptor or glucocorticoid receptor.
Tested applications	**Suitable for:** ICC, IHC-Fr, IP, WB, ChIP, Flow Cyt, IHC-P
Species reactivity	**Reacts with:** Mouse, Rat, Sheep, Rabbit, Chicken, Guinea pig, Cow, Human
Immunogen	Other Immunogen Type corresponding to Chicken Progesterone Receptor. Progesterone receptor purified from chick oviduct cytosol.

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 or -80°C. Avoid freeze / thaw cycle.
Storage buffer	Preservative: 0.05% Sodium azide Constituent: PBS
Purity	Protein G purified
Clonality	Monoclonal
Clone number	Alpha PR6
Isotype	IgG2a

Applications

Our Abpromise guarantee covers the use of **ab2765** 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
ICC		Use at an assay dependent concentration.

Application	Abreviews	Notes
IHC-Fr		Use a concentration of 20 µg/ml.
IP		Use at an assay dependent concentration.
WB		Use a concentration of 1 µg/ml. Predicted molecular weight: 99 kDa.
ChIP		Use at an assay dependent concentration.
EMSA		Use at an assay dependent concentration.
Flow Cyt		Use 0.5µg for 10^6 cells. ab170191-Mouse monoclonal IgG2a, is suitable for use as an isotype control with this antibody.
IHC-P	★★★★★	1/50.

Target

Function

The steroid hormones and their receptors are involved in the regulation of eukaryotic gene expression and affect cellular proliferation and differentiation in target tissues. Progesterone receptor isoform B (PRB) is involved activation of c-SRC/MAPK signaling on hormone stimulation.
Isoform A: inactive in stimulating c-Src/MAPK signaling on hormone stimulation.
Isoform 4: Increases mitochondrial membrane potential and cellular respiration upon stimulation by progesterone.

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

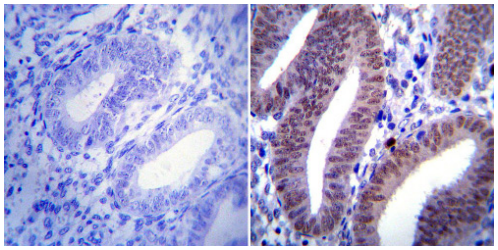
Phosphorylated on multiple serine sites. Several of these sites are hormone-dependent. Phosphorylation on Ser-294 occurs preferentially on isoform B, is highly hormone-dependent and modulates ubiquitination and sumoylation on Lys-388. Phosphorylation on Ser-102 and Ser-345 also requires induction by hormone. Basal phosphorylation on Ser-81, Ser-162, Ser-190 and Ser-400 is increased in response to progesterone and can be phosphorylated in vitro by the CDK2-A1 complex. Increased levels of phosphorylation on Ser-400 also in the presence of EGF, heregulin, IGF, PMA and FBS. Phosphorylation at this site by CDK2 is ligand-independent, and increases nuclear translocation and transcriptional activity. Phosphorylation at Ser-162 and Ser-294, but not at Ser-190, is impaired during the G(2)/M phase of the cell cycle. Phosphorylation on Ser-345 by ERK1/2 MAPK is required for interaction with SP1.
Sumoylation is hormone-dependent and represses transcriptional activity. Sumoylation on all three sites is enhanced by PIAS3. Desumoylated by SENP1. Sumoylation on Lys-388, the main site of sumoylation, is repressed by ubiquitination on the same site, and modulated by phosphorylation at Ser-294.
Ubiquitination is hormone-dependent and represses sumoylation on the same site. Promoted by MAPK-mediated phosphorylation on Ser-294.
Palmitoylated by ZDHHC7 and ZDHHC21. Palmitoylation is required for plasma membrane targeting and for rapid intracellular signaling via ERK and AKT kinases and cAMP generation.

Cellular localization

Nucleus. Cytoplasm. Nucleoplasmic shuttling is both homone- and cell cycle-dependent. On

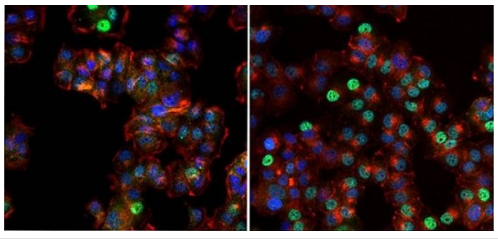
hormone stimulation, retained in the cytoplasm in the G(1) and G(2)/M phases; Mitochondrion outer membrane and Nucleus. Cytoplasm. Mainly nuclear.

Images



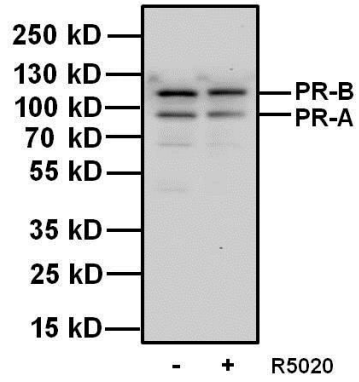
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Progesterone Receptor [Alpha PR6] antibody - ChIP Grade (ab2765)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) was performed on human uterus tissue. Antigen retrieval was performed using 10mM sodium citrate followed by microwave treatment for 8-15 minutes. Endogenous peroxidases were blocked in 3% H202-methanol for 15 minutes and tissues were blocked in 3% BSA-PBS for 30 minutes at room temperature. Cells were incubated with ab2765 (1:20) overnight in a humidified chamber. Tissues were washed in PBST and detection was performed using a secondary antibody conjugated to HRP. DAB staining buffer was applied and tissues were counterstained with hematoxylin and prepped for mounting. Images were taken at 40X magnification.



Immunocytochemistry/ Immunofluorescence - Anti-Progesterone Receptor antibody [Alpha PR6] - ChIP Grade (ab2765)

Immunocytochemistry/ Immunofluorescence analysis of T47D cells untreated (left) or stimulated with 100nm promegestone for 1 hour (right), labeling Progesterone Receptor with ab2765 (green). The cells were fixed with formalin for 15 minutes, permeabilized with 0.1% Triton X-100 in TBS for 10 minutes, and blocked with 3% Blocker BSA for 15 minutes at room temperature. Cells were stained with Anti-Progesterone Receptor antibody [Alpha PR6] - ChIP Grade (ab2765) at a dilution of 1/100 for 1 hour at 37C, and then incubated with a Alexa Fluor 488 goat anti-mouse IgG secondary antibody at a dilution of 1/1000 for 30 minutes at room temperature (both panels, green). Nuclei (both panels, blue) were stained with Hoechst 33342 dye.



Western blot - Anti-Progesterone Receptor antibody [Alpha PR6] - ChIP Grade (ab2765)

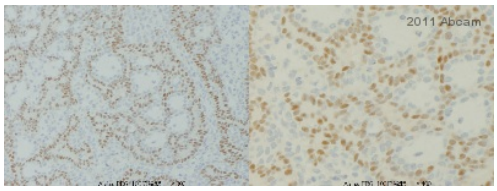
All lanes : Anti-Progesterone Receptor antibody [Alpha PR6] - ChIP Grade (ab2765) at 1 µg/ml

Lane 1 : T47D cell lysate untreated (-)
Lane 2 : T47D cell lysate stimulated (+) with 100 nm promegestone for 1 hour

Lysates/proteins at 20 µg per lane.

Secondary
Goat anti-Mouse IgG-HRP at 1/2000 dilution

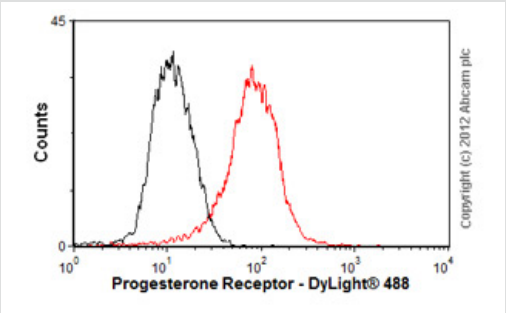
Predicted band size : 99 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Progesterone Receptor antibody [Alpha PR6] (ab2765)

This image is courtesy of Takako Akamatsu, Kansai Medical University.

ab2765 staining Progesterone Receptor in Rat mammary tissue by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded tissue sections). The sections were fixed in formaldehyde and subjected to heat-mediated antigen retrieval by pressure cooker prior to blocking with 5% BSA for 5 minutes at room temperature. The primary antibody was diluted 1/100 in TBST and incubated with the sample for 16 hours at 4°C. A biotin-conjugated goat anti-mouse polyclonal was used as the secondary antibody.



Flow Cytometry-Anti-Progesterone Receptor antibody [Alpha PR6](ab2765)

Overlay histogram showing T47D cells stained with ab2765 (red line). The cells were fixed with 80% methanol (5 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 (ab2765, 0.5µg/1x10^6 cells) for 30 min at 22ºC. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22ºC. Isotype control antibody (black line) was mouse IgG2a [ICIGG2A] (ab91361, 2µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed.

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