abcam

Product datasheet

Anti-Androgen Receptor antibody [EPR1535(2)] ab133273

Recombinant RabMAb

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Overview

Product name	Anti-Androgen Receptor antibody [EPR1535(2)]
Description	Rabbit monoclonal [EPR1535(2)] to Androgen Receptor
Host species	Rabbit
Tested applications	**Suitable for:** WB, IHC-P, ICC/IF
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide within Human Androgen Receptor aa 1-100 (N terminal). The exact sequence is proprietary. Database link: **P10275** (Peptide available as **ab191380**)
Positive control	WB: T47-D, LnCaP and 22Rv1 cell lysates; Rat and mouse prostate lysates. IHC-P: Human prostate, prostatic adenocarcinoma, prostatic hyperplasia tissues; Rat and mouse testis tissues; Breast carcinoma and prostatic carcinoma T3 tissues. ICC/IF: MCF7 cells;EP156T-AR, 957E/hTERT-AR and LNCaP cells.
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
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide

	Constituents: 40% Glycerol, 0.05% BSA, 59% PBS
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR1535(2)
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab133273 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	★★★★★ (2)	1/2000. Predicted molecular weight: 98 kDa. **For unpurified use at 1:1000** For Lysate preparation protocol, please refer to the protocol book in the protocol section and/or **here (downloadable copy).**
IHC-P	★★★★☆ (7)	1/500. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**. **For unpurified use at 1/100-1/250**
ICC/IF	★★★★★ (1)	1/100 - 1/250.

Target

Function	Steroid hormone receptors are ligand-activated transcription factors that regulate eukaryotic gene expression and affect cellular proliferation and differentiation in target tissues. Transcription factor activity is modulated by bound coactivator and corepressor proteins. Transcription activation is down-regulated by NR0B2. Activated, but not phosphorylated, by HIPK3 and ZIPK/DAPK3. Isoform 3 and isoform 4 lack the C-terminal ligand-binding domain and may therefore constitutively activate the transcription of a specific set of genes independently of steroid hormones.
Tissue specificity	Isoform 2 is mainly expressed in heart and skeletal muscle (PubMed:15634333). Isoform 3 is expressed by basal and stromal cells of prostate (at protein level) (PubMed:19244107).
Involvement in disease	Androgen insensitivity syndrome Spinal and bulbar muscular atrophy X-linked 1 Defects in AR may play a role in metastatic prostate cancer. The mutated receptor stimulates prostate growth and metastases development despite of androgen ablation. This treatment can reduce primary and metastatic lesions probably by inducing apoptosis of tumor cells when they express the wild-type receptor. Androgen insensitivity, partial
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. In the presence of bound steroid the ligand-binding domain interacts with the N-terminal modulating domain, and thereby activates AR transcription factor activity. Agonist binding is required for dimerization and binding to target DNA. The transcription factor activity of the complex formed by ligand-activated AR and DNA is modulated by interactions with coactivator and corepressor proteins. Interaction with RANBP9 is mediated by both the N-terminal domain and the DNA-binding domain. Interaction with EFCAB6/DJBP is mediated by the DNA-binding domain.

Post-translational modifications

Sumoylated on Lys-388 (major) and Lys-521. Ubiquitinated. Deubiquitinated by USP26. 'Lys-6' and 'Lys-27'-linked polyubiquitination by RNF6 modulates AR transcriptional activity and specificity.
Phosphorylated in prostate cancer cells in response to several growth factors including EGF. Phosphorylation is induced by c-Src kinase (CSK). Tyr-535 is one of the major phosphorylation sites and an increase in phosphorylation and Src kinase activity is associated with prostate cancer progression. Phosphorylation by TNK2 enhances the DNA-binding and transcriptional activity and may be responsible for androgen-independent progression of prostate cancer. Phosphorylation at Ser-83 by CDK9 regulates AR promoter selectivity and cell growth. Phosphorylation by PAK6 leads to AR-mediated transcription inhibition.
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.

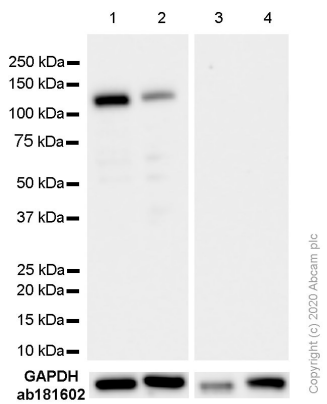
Cellular localization

Nucleus. Cytoplasm. Predominantly cytoplasmic in unligated form but translocates to the nucleus upon ligand-binding. Can also translocate to the nucleus in unligated form in the presence of RACK1.

Form

There are 2 isoforms produced by alternative splicing. Isoform 1 is also known as: AR-B; isoform 2 is known as AR-A or variant AR45.

Images



Western blot - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

All lanes : Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273) at 1/1000 dilution

Lane 1 : Mouse testis lysate
Lane 2 : Rat testis lysate
Lane 3 : Mouse liver lysate
Lane 4 : Rat liver lysate

Lysates/proteins at 20 µg per lane.

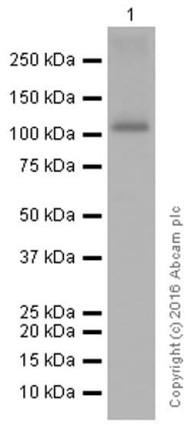
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 98 kDa
Observed band size: 110 kDa

Exposure time: 20 seconds

Blocking/Diluting buffer: 5% NFDM/TBST

Loading Control: Rabbit monoclonal [EPR16891] to GAPDH (**ab181602**)



Western blot - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

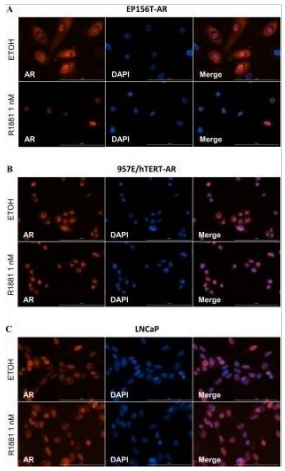
Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273) at 1/5000 dilution (purified) + Mouse prostate lysates at 15 µg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 98 kDa

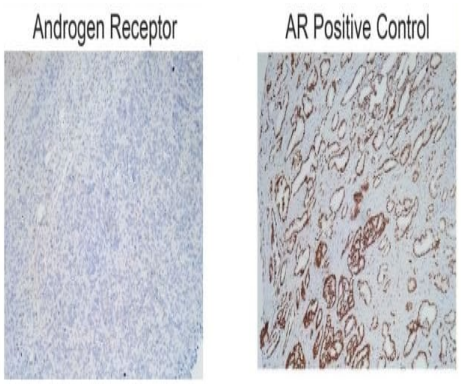
Blocking and diluting buffer: 5% NFDM/TBST



Immunocytochemistry/ Immunofluorescence - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Image from Azeem W et al., PLoS One. 2017;12(6):e0177861. Fig 6.; doi: 10.1371/journal.pone.0177861. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Texas red (Tx-Red) indirect immunofluorescent detection of Androgen Receptor using ab133273 in (A) EP156T-AR, (B) 957E/hTERT-AR and (C) LNCaP cells. The cells were treated with ± 1 nM R1881 for 24 hours.



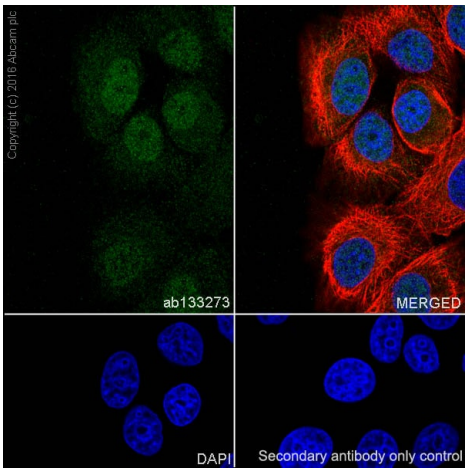
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

shao et al Oncotarget. 2018 Mar 6; 9(18): 14456–14471. Published online 2018 Feb 12. doi: 10.18632/oncotarget.24470

Formalin-fixed, paraffin-embedded tissues labeling Androgen Receptor with ab133273. Left: Tumor from orthotopic injectionof PTEN KD/TE cells. Right: Human postate cancer tissue.

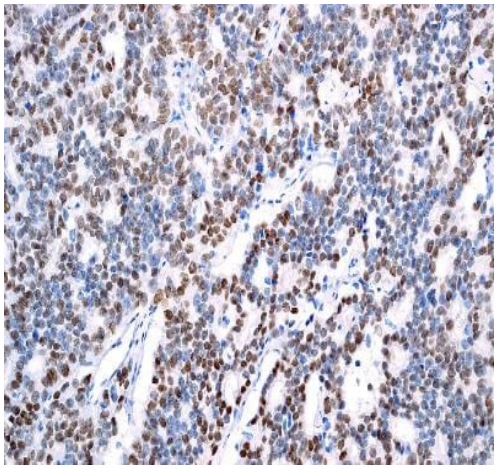
From Figure 3 of Shao et al/

Shao et al **Oncotarget**. 2018 Mar 6; 9(18): 14456–14471.Published online 2018 Feb 12. doi: **10.18632/oncotarget.24470**



Immunocytochemistry/ Immunofluorescence - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

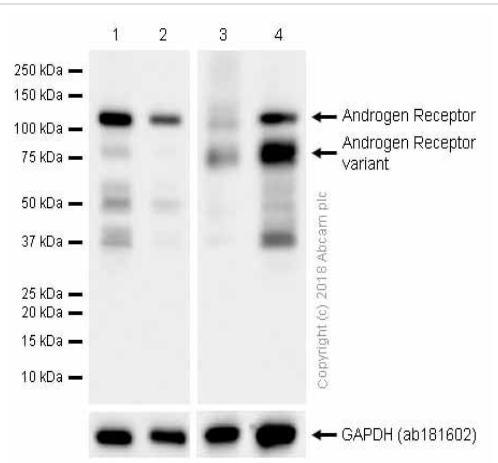
Immunocytochemistry/ Immunofluorescence analysis of MCF7 (Human breast adenocarcinoma epithelial cell) cells labeling Androgen receptor with purified ab133273 at 1:100 dilution (1.3µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 µg/ml). **ab150077** Goat anti rabbit IgG(Alexa Fluor® 488) was used as the secondary antibody at 1:1000 dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Immunohistochemical analysis of paraffin-embedded human prostatic adenocarcinoma tissue labelling Androgen Receptor using unpurified **ab133273** at 1/100 dilution.

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



Western blot - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

All lanes : Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273) at 1/20000 dilution

Lane 1 : LNCaP (Human prostate carcinoma epithelial cell) whole cell lysates prepared in RIPA lysis method
Lane 2 : LNCaP (Human prostate carcinoma epithelial cell) whole cell lysates prepared in 1%SDS Hot lysis method
Lane 3 : 22Rv1 (Human prostate carcinoma epithelial cell) whole cell lysates prepared in RIPA lysis method
Lane 4 : 22Rv1 (Human prostate carcinoma epithelial cell) whole cell lysates prepared in 1%SDS Hot lysis method

Lysates/proteins at 1/15 dilution per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution (Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated)

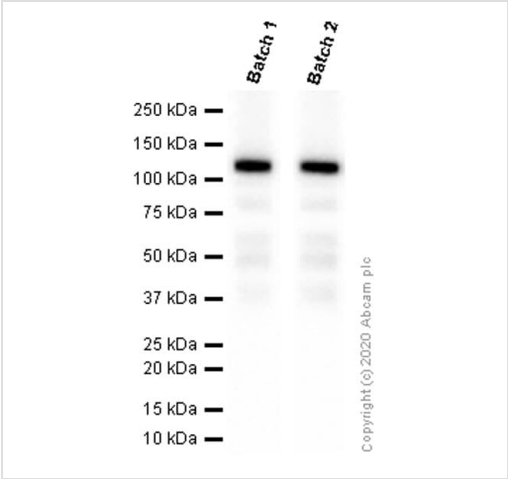
Predicted band size: 98 kDa
Observed band size: 120 kDa

Exposure time: 10 seconds

Blocking/Diluting buffer - 5% NFDM/TBST

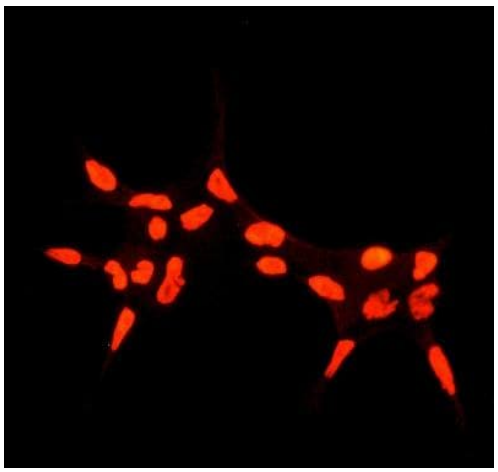
The androgen receptor variant band detected in 22RV1 cells is reported by PMID: 22315407.

We recommend you to try both RIPA and 1%SDS Hot lysis preparation methods to get desired bands.



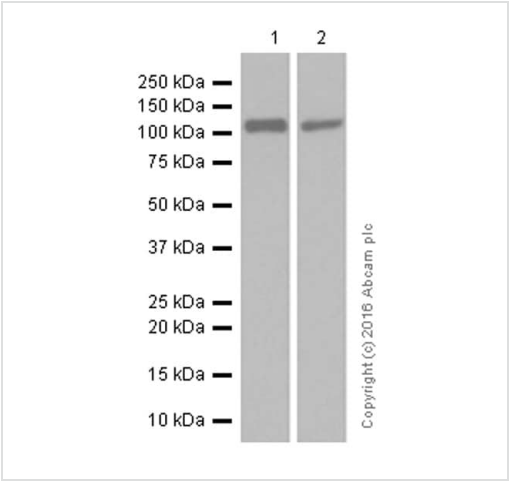
Western blot - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Different batches of ab133273 were tested on LNCaP (Human prostate carcinoma epithelial cell) lysate at 1.0 µg/ml. 15 µg of lysate was loaded in each lane. Bands observed at 120 kDa.



Immunocytochemistry/ Immunofluorescence - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Immunofluorescent staining of LnCAP cells labelling Androgen Receptor using unpurified **ab133273**, at 1/100 dilution



Western blot - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

All lanes : Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273) at 1/5000 dilution (purified)

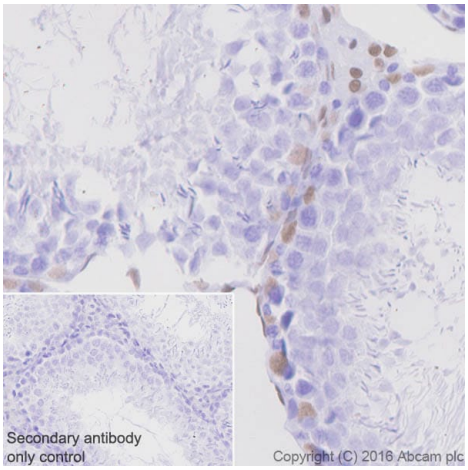
Lane 1 : LNCaP (Human prostate carcinoma epithelial cell) whole cell lysates
Lane 2 : Rat prostate lysates

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/2000 dilution

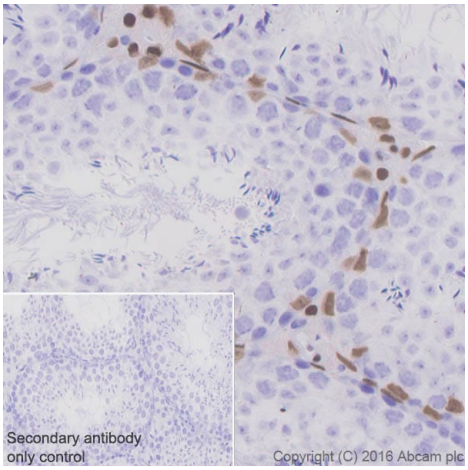
Predicted band size: 98 kDa

Blocking and diluting buffer: 5% NFDM/TBST



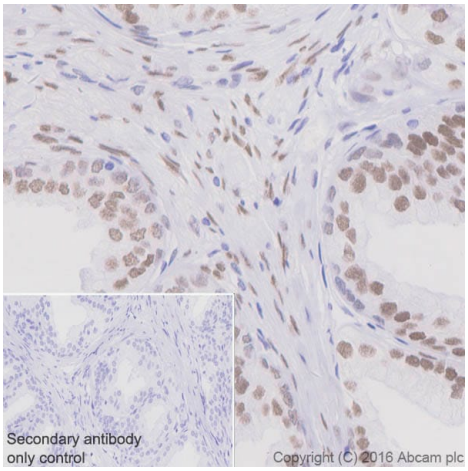
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat testis tissue sections labeling Androgen Receptor with Purified ab133273 at 1:500 dilution (0.25 µg/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control.



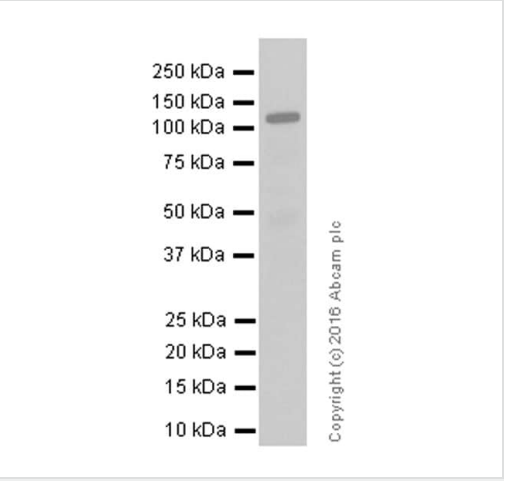
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse testis tissue sections labeling Androgen Receptor with Purified ab133273 at 1:500 dilution (0.25 µg/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human prostatic hyperplasia tissue sections labeling Androgen Receptor with Purified ab133273 at 1:500 dilution (0.25 µg/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control.



Western blot - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273) at 1/1000 dilution (unpurified) + LNCaP (Human prostate carcinoma epithelial cell) at 10 µg

Secondary

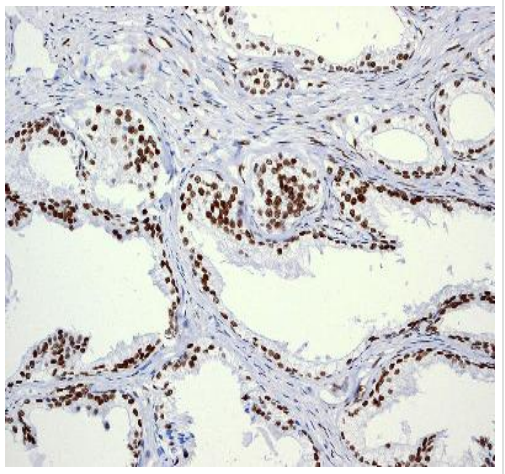
Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 98 kDa

Observed band size: 110 kDa

Exposure time: 3 seconds

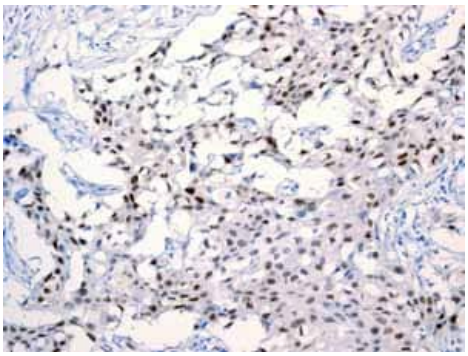
Blocking/Diluting buffer 5% NFDM/TBST



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Immunohistochemical analysis of paraffin-embedded human prostate tissue labelling Androgen Receptor using unpurified **ab133273**, at1/100 dilution.

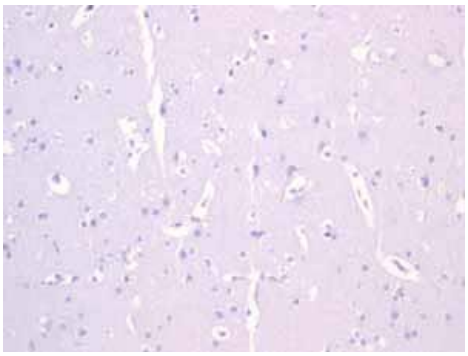
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Unpurified **ab133273** showing positive staining in Breast carcinoma tissue.

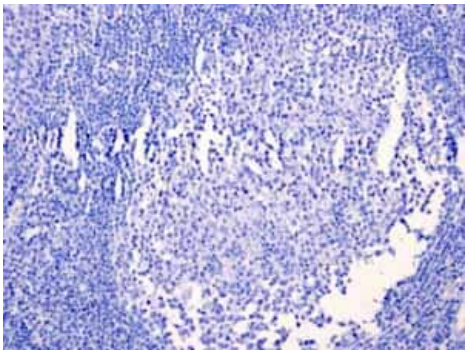
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Unpurified ab133273 showing negative staining in Normal brain tissue.

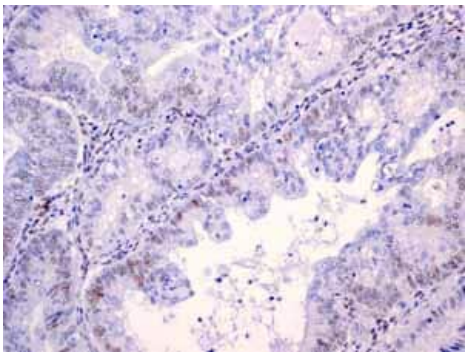
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Unpurified ab133273 showing negative staining in Normal tonsil tissue.

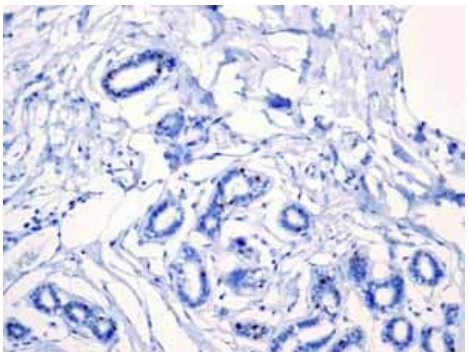
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Unpurified **ab133273** showing positive staining in Endometrial carcinoma tissue.

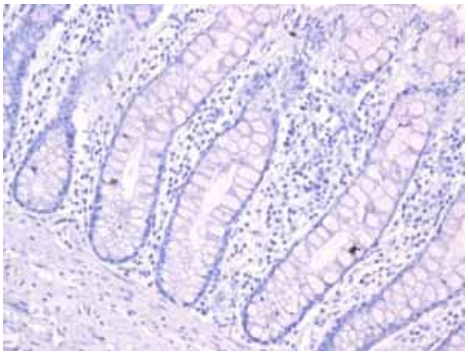
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Unpurified **ab133273** showing negative staining in Normal breast tissue.

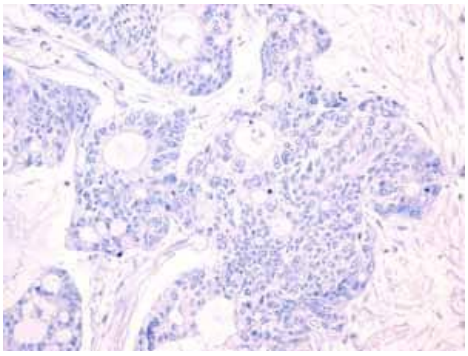
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Unpurified **ab133273** showing negative staining in Normal colon tissue.

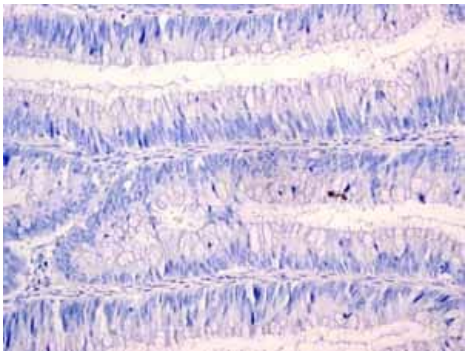
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Unpurified ab133273 showing negative staining in Ovarian carcinoma tissue.

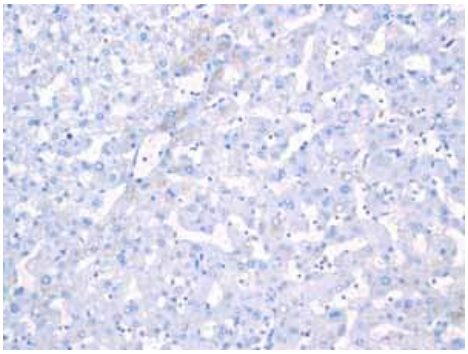
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Unpurified **ab133273** showing negative staining in Colonic adenocarcinoma tissue.

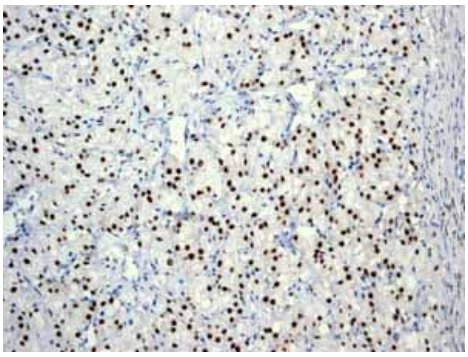
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Unpurified **ab133273** showing negative staining in Normal liver tissue.

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



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

Unpurified **ab133273** showing positive staining in Prostatic carcinoma T3 tissue.

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

Anti-Androgen Receptor antibody [EPR1535(2)] (ab133273)

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