

Product datasheet

Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free ab212175

Recombinant RabMAb

14 Images

| Overview | |
|---------------------|---|
| Product name | Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free |
| Description | Rabbit monoclonal [ER179(2)] to Androgen Receptor - BSA and Azide free |
| Host species | Rabbit |
| Tested applications | Suitable for: WB, IHC-P, ChIP, ChIC/CUT&RUN-seq, ICC/IF Unsuitable for: Flow Cyt or IP |
| Species reactivity | Reacts with: Mouse, Rat, Human |
| Immunogen | Synthetic peptide. This information is proprietary to Abcam and/or its suppliers. |
| Positive control | T47-D and LnCaP cell lysates. Paraffin-embedded human prostatic adenocarcinoma tissue. Paraffin-embedded human prostate tissue. ChlC/CUT&RUN seq: LNCaP cell. |
| General notes | ab212175 is the carrier-free version of <u>ab108341</u> . |
| | Our <u>carrier-free</u> 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. |
| | 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. |
| | 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. |
| | This product is compatible with the Maxpar [®] Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. $Maxpar^{\mathbb{R}}$ is a trademark of Fluidigm Canada Inc. |
| | 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 <u>see here</u> . 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. Do Not Freeze. |
| Storage buffer | pH: 7.20 Constituent: PBS |
| Carrier free | Yes |
| Purity | Protein A purified |
| Clonality | Monoclonal |
| Clone number | ER179(2) |
| lsotype | lgG |

Applications

The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab212175 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. Predicted molecular weight: 99 kDa. |
| IHC-P | | Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. Perform heat mediated antigen retrieval with citrate buffer (pH 6.0) or Tris-EDTA buffer (pH9.0) before commencing with IHC staining protocol. |
| ChIP | | Use at an assay dependent concentration. PubMed: 23817021 |
| ChIC/CUT&RUN-seq | | Use at an assay dependent concentration. |
| ICC/IF | | Use at an assay dependent concentration. |

Application notes

Is unsuitable for Flow Cyt or IP.

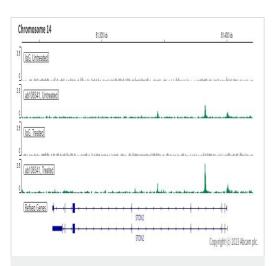
Target

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.

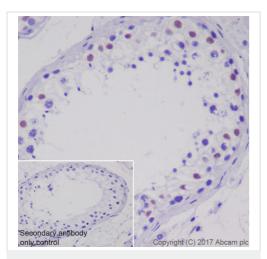
| Tissue specificity | lsoform 2 is mainly expressed in heart and skeletal muscle (PubMed:15634333). lsoform 3 is expressed by basal and stromal cells of prostate (at protein level) (PubMed:19244107). |
|-------------------------------------|---|
| Involvement in disease | Androgen insensitivity syndrome 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 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



ChIC/CUT&RUN sequencing - Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free (ab212175) This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108341**).

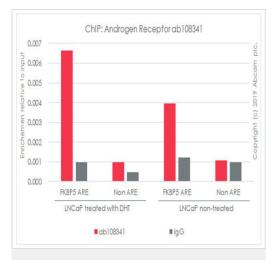
ChIC/CUT&RUN was performed using a pAG-MNAse at a final concentration of 700 ng/ μ L, 2.5 x 10^5 LNCaP (Human prostate carcinoma epithelial cell) cells cultured in phenol red free medium and 5% charcoal stripped FBS for 3 days then treated with DHT (10 nM 4h), and 5 μ g of **ab108341** [ER179(2)]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control **ab172730** is also shown. Additional screenshots of mapped reads can be downloaded **here**. The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free (ab212175)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human testis tissue sections labeling Androgen receptor with Purified **ab108341** at 1:100 dilution. 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.

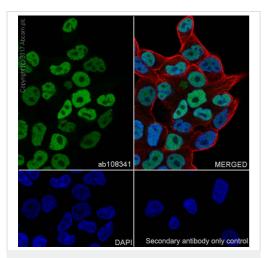
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab108341</u>).



ChIP - Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free (ab212175) Chromatin was prepared from LNCaP cells according to the Abcam Dual-X-ChIP protocol*. Cells were fixed with 1.5 mM EGS for 30mins and then formaldehyde for 10min. The ChIP was performed with 25 µg of chromatin, 5 µg of **ab108341** (red), or 5 µg of rabbit normal IgG **ab172730** (gray) and 20 µl of Protein A/G sepharose beads. The immunoprecipitated DNA was quantified by real time PCR (Taqman approach for active and inactive loci, Sybr green approach for heterochromatic loci). Primers and probes are commercial primers from Paper (PMID: 25802280)

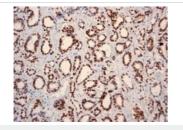
*http://www.abcam.com/resources? keywords=X%20ChIP%20protocol

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide <u>ab108341</u>).

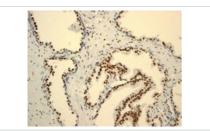


Immunocytochemistry/ Immunofluorescence - Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free (ab212175) Immunocytochemistry/ Immunofluorescence analysis of LNCaP (Human prostate carcinoma epithelial cell) cells labeling Androgen receptor with Purified <u>ab108341</u> at 1:500 dilution. Cells 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). <u>ab150077</u> Goat anti rabbit lgG(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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab108341</u>).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free (ab212175)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free (ab212175) Unpurified <u>ab108341</u>, at 1/250, staining Androgen Receptor in paraffin-embedded Human prostatic adenocarcinoma tissue by Immunohistochemistry.

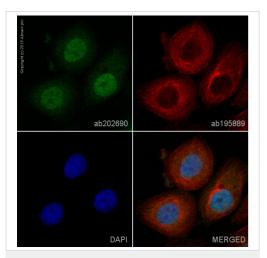
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab108341</u>).

Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.

Unpurified <u>ab108341</u>, at 1/250, staining Androgen Receptor in paraffin-embedded Human prostate tissue by Immunohistochemistry.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab108341**).

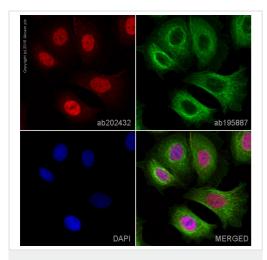
Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



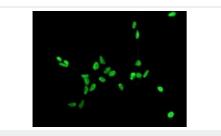
Immunocytochemistry/ Immunofluorescence - Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free (ab212175) Clone ER179(2) (ab212175) has been successfully conjugated by Abcam. This image was generated using Anti-Androgen Receptor antibody [ER179(2)] (Alexa Fluor® 488). Please refer to <u>ab202690</u> for protocol details.

ab202690 staining Androgen Receptor in MCF7 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 **ab202690** at 1/100 dilution (shown in green) and **ab195889**, Mouse monoclonal to alpha Tubulin (Alexa Fluor[®] 594), 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).



Immunocytochemistry/ Immunofluorescence - Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free (ab212175)



Immunocytochemistry/ Immunofluorescence - Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free (ab212175)

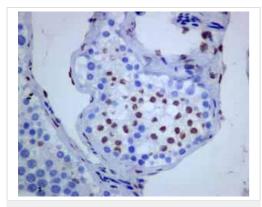
Clone ER179(2) (ab212175) has been successfully conjugated by Abcam. This image was generated using Anti-Androgen Receptor antibody [ER179(2)] (Alexa Fluor® 647). Please refer to **ab202432** for protocol details.

ab202432 staining Androgen Receptor in MCF7 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 **ab202432** at 1/200 dilution (shown in red) and **ab195887**, Mouse monoclonal to alpha Tubulin (Alexa Fluor[®] 488), at 1/250 dilution (shown in green). Nuclear DNA was labelled with DAPI (shown in blue).

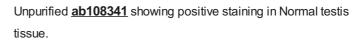
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

Unpurified <u>ab108341</u>, at 1/100, staining Androgen Receptor in LnCaP cells by Immunofluorescence.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab108341</u>).

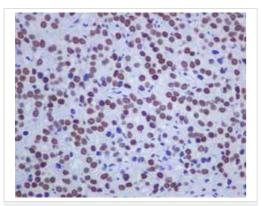


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free (ab212175)



This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab108341</u>).

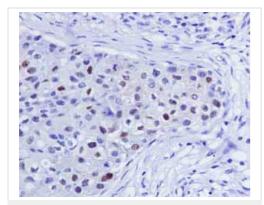
Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free (ab212175) Unpurified <u>ab108341</u> showing positive staining in Prostatic carcinoma T3 tissue.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab108341</u>).

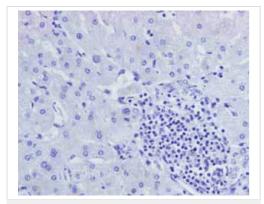
Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free (ab212175) Unpurified <u>ab108341</u> showing positive staining in Breast carcinoma tissue.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab108341</u>).

Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Androgen Receptor antibody [ER179(2)] - BSA and Azide free (ab212175)



and Azide free (ab212175)

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Unpurified <u>ab108341</u> showing negative staining in Normal liver tissue.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab108341</u>).

Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.

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