# abcam

## Product datasheet

## Anti-ERG antibody [EPR3864] ab92513

יעלאעבע RabMAb

★★★★★ 17 Abreviews 201 References 画像数 17

#### 製品の概要

製品名 Anti-ERG antibody [EPR3864]

製品の詳細 Rabbit monoclonal [EPR3864] to ERG

由来種 Rabbit

特異性 This antibody also detects Fli-1.

アプリケーション 適用あり: WB, IHC-P, ICC/IF, Flow Cyt (Intra)

種交差性 交差種: Mouse, Rat, Human

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール WB: Jurkat, HeLa and RAW 264.7 cell lysates; Rat brain and heart lysates. IHC-P: Human kidney,

> brain and prostate adenocarcinoma tissues; Fus A5 transgenic mouse prostate tissue; Mouse brain tissue. ICC/IF: Circulating tumor cells (CTCs) from a castrate-resistant prostate cancer

(CRPC) patient; THP-1 cells. Flow Cyt (intra): THP-1 cells.

特記事項 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**.

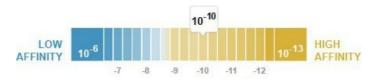
#### 製品の特性

製品の状態 Liquid

保存方法 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle.

 $K_D = 8.90 \times 10^{-10} M$ 解離定数(KD値)



### Learn more about K<sub>D</sub>

**バッファー** pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 40% Glycerol, 0.05% BSA, 59% PBS

精製度 Protein A purified

**ポリ/モノ** モノクローナル

**クローン名** EPR3864

アイソタイプ lgG

#### アプリケーション

**The Abpromise guarantee** <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab92513の使用に適用されますアプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
WB	*** <u>*</u>	1/1000 - 1/10000. Predicted molecular weight: 55 kDa.
IHC-P	****(3)	1/1000. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See IHC antigen retrieval protocols. For unpurified, use 1/100 - 1/250.
ICC/IF	★★★★★ (3)	Use a concentration of 1 µg/ml. This product gave a positive signal in THP-1 (-ve: HCT116) fixed with 4% formaldehyde (10 min).
Flow Cyt (Intra)		Use at an assay dependent concentration.

#### ターゲット情報

機能 Transcriptional regulator. May participate in transcriptional regulation through the recruitment of

SETDB1 histone methyltransferase and subsequent modification of local chromatin structure.

**関連疾患** Defects in ERG are a cause of Ewing sarcoma (ES) [MIM:612219]. A highly malignant,

metastatic, primitive small round cell tumor of bone and soft tissue that affects children and adolescents. It belongs to the Ewing sarcoma family of tumors, a group of morphologically heterogeneous neoplasms that share the same cytogenetic features. They are considered neural tumors derived from cells of the neural crest. Ewing sarcoma represents the less differentiated form of the tumors. Note=A chromosomal aberration involving ERG is found in patients with

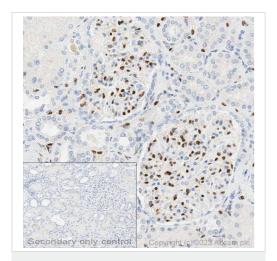
Erwing sarcoma. Translocation t(21;22)(q22;q12) with EWSR1.

Note=Chromosomal aberrations involving ERG have been found in acute myeloid leukemia (AML). Translocation t(16;21)(p11;q22) with FUS. Translocation t(X;21)(q25-26;q22) with ELF4.

**配列類似性** Belongs to the ETS family.

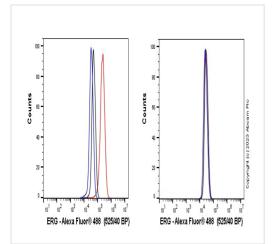
Contains 1 ETS DNA-binding domain.
Contains 1 PNT (pointed) domain.

細胞内局在 Nucleus. Cytoplasm. Localized in cytoplasmic mRNP granules containing untranslated mRNAs.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ERG antibody [EPR3864] (ab92513)

Immunohistochemical analysis of formalin-fixed paraffin-embedded human kidney labelling ERG with ab92513 at a concentration of 1µg/ml. The immunostaining was performed on a Ventana DISCOVERY ULTRA (Roche Tissue Diagnostics) instrument with an OptiView DAB IHC Detection Kit. Heat mediated antigen retrieval was conducted for 32min with ULTRA cell conditioning solution (CC1 pH8.5). ab92513 anti ERG antibody was incubated at 37°C for 16min. Sections were counterstained with Hematoxylin II. Image inset shows absence of staining in secondary antibody only control.

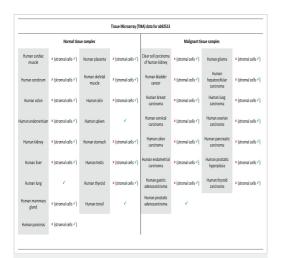


Flow Cytometry (Intracellular) - Anti-ERG antibody [EPR3864] (ab92513)

Flow cytometry overlay histogram showing left THP-1 positive cells and right negative HCT116 stained with ab92513 (red line). The cells were fixed with 4% formaldehyde (10 min) and then permeabilised with 0.1% PBS-Triton X-100 for 15 min. The cells were then incubated in 1x PBS containing 10 $\mu$ g/ml human lgG and 10% normal goat serum to block FC receptors and non-specific protein-protein interaction followed by the antibody (ab92513) (1x  $10^6$  in  $100\mu$ l at  $0.04\mu$ g/ml (1/54000)) for 30min at  $22^\circ$ C.

The secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min at 22°C Isotype control antibody (black line) was Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

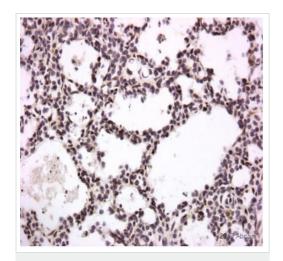
Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ERG antibody [EPR3864] (ab92513)

Tissue Microarrays stained for Anti-ERG antibody [EPR3864] using ab92513 in immunohistochemical analysis. This table provides a detailed overview of positive (tick mark) and negaive (cross mark) staining per sample type tested. The section was incubated with ab92513 for 30 mins at room temperature followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). The immunostaining was performed on a Leica Biosystems BOND® RX instrument.

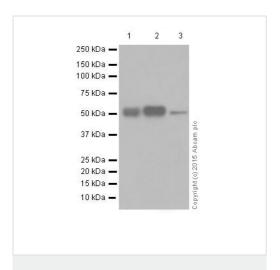
Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ERG antibody [EPR3864] (ab92513)

Formalin-fixed, paraffin-embedded mouse brain tissue stained for ERG using ab92513 at 1/200 dilution in immunohistochemical analysis. A horse radish peroxidase antibody was used as the secondary antibody.

Antigen Retrieval: 40x; Proteinase K antigen retrieval - 15 min at 37 C



Western blot - Anti-ERG antibody [EPR3864] (ab92513)

All lanes: purified

Lane 1 : rat brain lysate

Lane 2 : rat heart lysate

Lane 3: RAW 264.7 (mouse macrophage cell line transformed

with Abelson murine leukemia virus) cell lysate

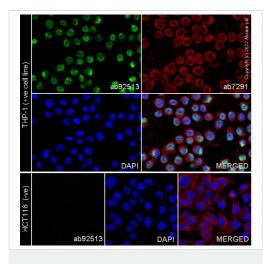
Lysates/proteins at 20 µg per lane.

## **Secondary**

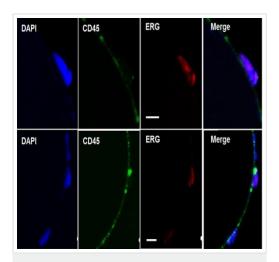
All lanes: HRP goat anti-rabbit lgG (H+L) at 1/1000 dilution

Predicted band size: 55 kDa Observed band size: 55 kDa

Blocking buffer: 5% NFDM/TBST Dilution buffer: 5% NFDM/TBST



Immunocytochemistry/ Immunofluorescence - Anti-ERG antibody [EPR3864] (ab92513) ab92513 staining ERG in THP-1 cells, with negative expression in HCT116 cells. The cells were fixed with 4% formaldehyde (10 min), permeabilised 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 ab92513 at 1 μg/ml and ab7291, Mouse monoclonal [DM1A] to alpha Tubulin at 0.5 μg/ml. Cells were then incubated with ab150081, Goat polyclonal Secondary Antibody to Rabbit lgG - H&L (Alexa Fluor<sup>®</sup> 488), pre-adsorbed at 1/1000 dilution (shown in green) and ab150119, Goat polyclonal Secondary Antibody to Mouse lgG - H&L (Alexa Fluor<sup>®</sup> 647), pre-adsorbed at 1/1000 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue). Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown.



Functional characterization and detection of genetic alterations in GEDI-captured cells. The TMPRSS2:ERG fusion protein is detected in GEDI-captured circulating tumor cells (CTCs) from a castrate-resistant prostate cancer (CRPC) patient. PSMA-captured CTCs were stained on the device with ab92513. Representative examples of PSMA+/CD45- CTCs are shown, two of which are positive for ERG. Scale bars: 10 microns.

Immunocytochemistry/ Immunofluorescence - Anti-

ERG antibody [EPR3864] (ab92513)

Image from Kirby BJ et al., PLoS One. 2012;8(12):e83903. Fig 4.; doi: 10.1371/journal.pone.0035976. Reproduced under the Creative Commons license http://creativecommons.org/licenses/by/4.0/

Tumor, ERG\*, GSTP1\*

B End GSTP1

C ERG GSTP1

D Tumor, ERG\*, GSTP1\*

GSTP1

GSTP1

GSTP1

GSTP1

GSTP1

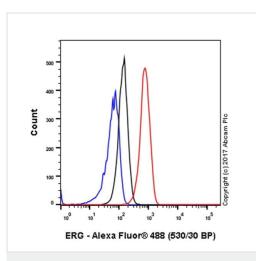
GSTP1

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ERG antibody [EPR3864] (ab92513)

Image from Litovkin K et al., PLoS One. 2015;10(6):e0130651. Fig 5.; doi: 10.1371/journal.pone.0130651. Reproduced under the Creative Commons license https://creativecommons.org/publicdomain/zero/1.0/

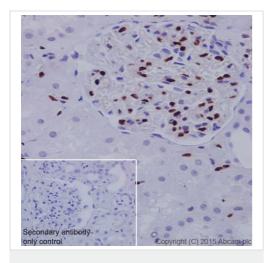
ERG and GSTP1 immunostainings of human prostate cancer samples using ab92513.

Representative immunohistochemical images of prostate cancer samples are shown that were positive for ERG and negative for GSTP1 (A), positive for both ERG and GSTP1 (B), negative for both ERG and GSTP1(C), and negative for ERG and positive for GSTP1 (D). The internal staining control for ERG is the endothelium (arrows) and for GSTP1 the stromal and/or basal cells of normal prostate glands. N, normal prostate gland; S, Stroma; T, tumor gland. Scale bars equal 100µm



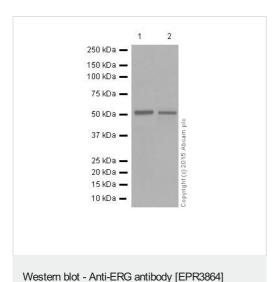
Flow Cytometry (Intracellular) - Anti-ERG antibody [EPR3864] (ab92513)

Intracellular Flow Cytometry analysis of THP-1 (human monocytic leukemia cell line) cells labeling ERG with purified ab92513 at 1/1000 dilution (1ug/ml) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit lgG (Alexa Fluorr® 488) (ab150077) (1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal lgG (Black) (ab172730) was used as the isotype control, Cell without incubation with primary antibody and secondary antibody (Blue) were used as the unlabeled control.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ERG antibody [EPR3864] (ab92513)

Immunohistochemical staining of paraffin embedded human kidney with purified ab92513 at a working dilution of 1/1000. The secondary antibody used is HRP goat anti-rabbit lgG H&L (ab97051) at 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.



(ab92513)

**All lanes :** Anti-ERG antibody [EPR3864] (ab92513) at 1/2000 dilution (purified)

Lane 1 : Jurkat (human T cell leukemia cell line from peripheral blood) cell lysate

**Lane 2 :** HeLa (human epithelial cell line from cervix adenocarcinoma) cell lysate

Lysates/proteins at 20 µg per lane.

## **Secondary**

All lanes: HRP goat anti-rabbit lgG (H+L) at 1/1000 dilution

**Predicted band size:** 55 kDa **Observed band size:** 55 kDa

Blocking buffer: 5% NFDM/TBST Dilution buffer: 5% NFDM/TBST

ab92513 MERGED

DAPI -ve control 1 -ve control 2

Immunocytochemistry/ Immunofluorescence - Anti-ERG antibody [EPR3864] (ab92513) Immunofluorescence staining of THP-1 (human monocytic leukemia cell line) cells with purified ab92513 at a working dilution of 1/100, counter-stained with DAPI. The secondary antibody was Alexa Fluor<sup>®</sup> 488 goat anti-rabbit (<u>ab150077</u>), used at a dilution of 1/1000. <u>ab7291</u>, a mouse anti-tubulin antibody (1/1000), was used to stain tubulin along with <u>ab150120</u> (Alexa Fluor<sup>®</sup> 594 goat antimouse, 1/1000), shown in the top right hand panel. The cells were fixed in 4% PFA and permeabilized using 0.1% Triton X 100. The negative controls are shown in bottom middle and right hand panels - for negative control 1, purified ab92513 was used at a dilution of 1/500 followed by an Alexa Fluor<sup>®</sup> 594 goat anti-mouse antibody (<u>ab150120</u>) at a dilution of 1/500. For negative control 2, <u>ab7291</u> (mouse anti-tubulin) was used at a dilution of 1/500 followed by an Alexa Fluor<sup>®</sup> 488 goat anti-rabbit antibody (<u>ab150077</u>) at a dilution of 1/400.

Alexa Fluor $^{\$}$  488 (<u>ab196374</u>) and Alexa Fluor $^{\$}$  647 (<u>ab196149</u>) conjugated versions are available for this clone.

kDa 250-150-100-75-50-37-25-

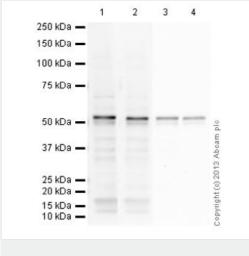
Western blot - Anti-ERG antibody [EPR3864] (ab92513)

Anti-ERG antibody [EPR3864] (ab92513) at 1/1000 dilution (unpurified) + Jurkat (human T cell leukemia cell line from peripheral blood) cell lysate at 10  $\mu g$ 

## **Secondary**

HRP labelled Goat anti-Rabbit at 1/2000 dilution

Predicted band size: 55 kDa



Western blot - Anti-ERG antibody [EPR3864] (ab92513)

Lanes 1 & 3: Anti-ERG antibody [EPR3864] (ab92513) at 1/250 dilution (unpurified)

**Lanes 2 & 4**: Anti-ERG antibody [EPR3864] (ab92513) at 1/1000 dilution (unpurified)

**Lane 1 :** Jurkat (human T cell leukemia cell line from peripheral blood) Whole Cell Lysate

Lanes 2-4: Jurkat Whole Cell Lysate

Lysates/proteins at 10 µg per lane.

#### **Secondary**

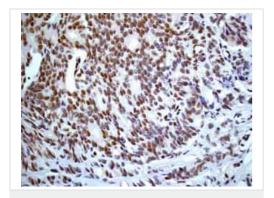
**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/10000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

**Predicted band size:** 55 kDa **Observed band size:** 55 kDa

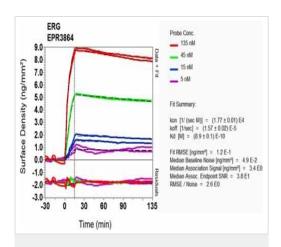
Exposure time: 12 minutes



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ERG antibody [EPR3864] (ab92513)

Immunohistochemical analysis of paraffin embedded Human
Prostatic adenocarcinoma stage 3 tissue using unpurified ab92513 showing +ve staining.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



OI-RD Scanning - Anti-ERG antibody [EPR3864] (ab92513)

Equilibrium disassociation constant ( $K_D$ ) Learn more about  $K_D$ 

Click here to learn more about KD



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