

## **Product datasheet for TA301431**

#### OriGene Technologies, Inc.

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### Carbonic Anhydrase IX (CA9) Rabbit Polyclonal Antibody

**Product data:** 

**Product Type:** Primary Antibodies

Applications: ChIP, ELISA, FC, ICC/IF, IHC, Immunoblotting, IP, Simple Western, WB

**Recommended Dilution:** Western Blot: 1 - 3 ug/ml, Proximity Ligation Assay, Gel Super Shift Assays, Simple Western:

1:50, Immunoblotting, ELISA, Immunoprecipitation: 1:10 - 1:500, Microarray,

Immunohistochemistry-Paraffin: 1:200 - 1:500, Immunocytochemistry/ Immunofluorescence: 2 - 5 ug/ml, Dual RNAscope ISH-IHC: 1:1000, Chromatin Immunoprecipitation (ChIP): 1:10-1:500, Flow Cytometry: 1:1000, Immunohistochemistry: 1:200 - 1:500, Immunohistochemistry-

Frozen: 1:200 - 1:500

Reactivity: Human, Dog

Host: Rabbit Isotype: IgG

Clonality: Polyclonal

**Immunogen:** A synthetic peptide derived from a C-terminal sequence of the human CA IX.

**Formulation:** Tris-citrate/phosphate, pH 7-8, 0.1% sodium Azide

**Concentration:** lot specific

**Purification:** Immunogen affinity purified

Conjugation: Unconjugated

**Storage:** Store at -20°C as received.

**Stability:** Stable for 12 months from date of receipt.

**Gene Name:** carbonic anhydrase 9

Database Link: NP 001207

Entrez Gene 611933 DogEntrez Gene 768 Human

Q16790





#### Background:

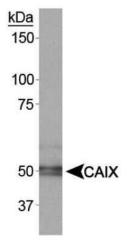
Carbonic anhydrase (CA) is an enzyme that assists rapid interconversion of carbon dioxide and water into carbonic acid, protons, and bicarbonate ions. It is abundant in all mammalian tissues. Because of its functionality, it has become an important diagnostic marker for various cancers, most notably renal cell carcinoma (RCC). There are many genes that are inducible by hypoxia, via HIF-1 alpha. CA IX is one of the most inducible genes because of its stability and the location of the expressed protein within the membrane. Carbonic anhydrases have a widespread role in regulating pH in normal tissues, by regulating hydrogen ion (H+) flux. The pH is important in cell death under hypoxia, thus a blockade of CA IX results in increased cell death under hypoxia. Therefore, CA IX has become a reliable histochemical marker of hypoxia.

Synonyms: CAIX; MN

**Protein Families:** Druggable Genome, Transmembrane

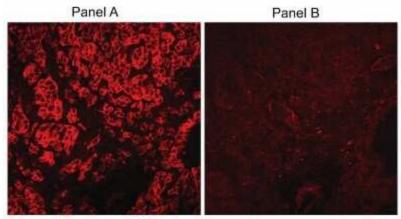
**Protein Pathways:** Nitrogen metabolism

# **Product images:**



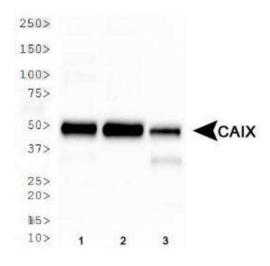
Analysis on rat renal cortex. A specific band was detected at a molecular weight of approximately 50 kDa.





SDS epitope unmasking

Immunofluorescence of human RCC tumor cryosections using TA301431 (Panel A). Panel B shows staining with normal rabbit serum.

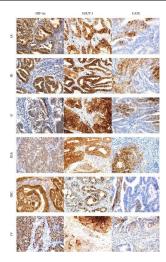


Analysis in 1) HeLa, 2) MDA-MB-231, and 3) A549 whole cell lysates. Specific bands were detected for Carbonic Anhydrase IX/CA9 at a molecular weight of 50 kDa.

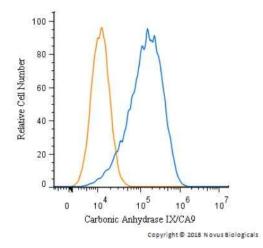


Simple Western lane view shows a specific band for CAIX in 0.1 mg/mL of HeLa lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.

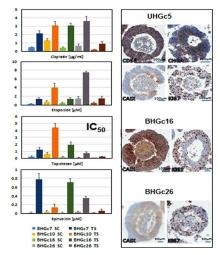




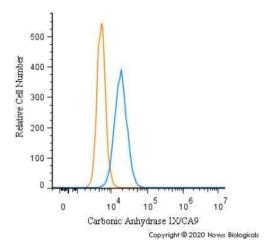
Immunohistochemical representative microphotographs representing the HIF-1 $\alpha$ , GLUT-1, and CAIX expression in endometrial cancer according to FIGO classification (IA, IB, II, IIIA, IIIC, and IV). Primary objective magnification 20x.



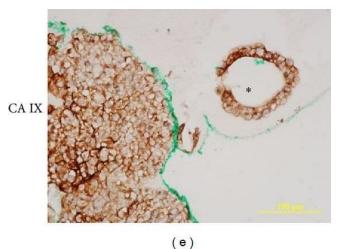
An intracellular stain was performed on U-87 MG Cells with TA301431 and a matched isotype control. Cells were fixed with 4% PFA and then permeablized with 0.1% saponin. Cells were incubated in an antibody dilution of 2.5 ug/mL for 30 minutes at room temperature, followed by Rabbit IgG APC-conjugated Secondary Antibody, (R&D Systems, F0111).



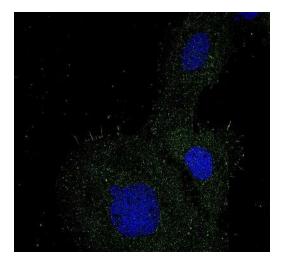
Chemosensitivity of the CTC SCLC lines (IC50, mean ± SD) and immunohistochemistry of sections of tumorospheres. All differences between single cells (SC) and tumorospheres (TS) are statistically significant. Immunohistochemical staining of sections of UHGc5, BHGc16 and BHGc26 CTCs was performed using antibodies directed to CD56, CHGA, CAIX and Ki67, respectively.



An intracellular stain was performed on A431 cells with Carbonic Anhydrase IX/CA9 Antibody TA301431 (blue) and a matched isotype control (orange). Cells were fixed with 4% PFA and then permeabilized with 0.1% saponin. Cells were incubated in an antibody dilution of 1.0 ug/mL for 30 minutes at room temperature, followed by Rabbit IgG (H+L) Cross-Adsorbed Secondary Antibody, Dylight 550 (SA5-10033, Thermo Fisher).

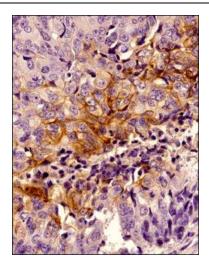


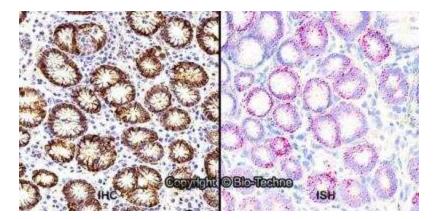
The images are representative of immunohistochemistry staining of nuclear staining of proliferation marker Ki67, membrane protein CD 44, and CA IX. The images of scaffold culture (left panel) are compared with cell pellet paraffin section (right panel). The scaffold dissolved in xylene presented as a clear region marked \*. All scale bars are 100  $\mu m$ , magnification 40x.



A431 cells were fixed in 4% paraformaldehyde for 10 minutes and permeabilized in 0.05% Triton X-100 in PBS for 5 minutes. The cells were incubated with anti-Carbonic Anhydrase IX/CA9 Antibody TA301431 at 2 ug/ml overnight at 4C and detected with an anti-rabbit Dylight 488 (Green) at a 1:1000 dilution for 60 minutes. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 100X objective and digitally deconvolved.







IHC analysis of a FFPE tissue section of human breast cancer using CAIX antibody at 1:1000 dilution. The primary antibody bound to CAIX antigens in the tissue section was detected using a HRP labeled secondary antibody and DAB reagent. Nuclei of the cells were counterstained with hematoxylin. This CAIX antibody generated an expected cytoplasmic staining of CAIX protein with an intense signal around the cellular membranes in tumor cores. The latter are more likely to be hypoxic in growing tumors which signifies that the observed CAIX staining is specific.

Formalin-fixed paraffin-embedded tissue sections of human stomach were probed for Carbonic Anhydrase IX/CA9 mRNA (ACD RNAScope Probe, catalog # 559348; Fast Red chromogen, ACD catalog # 322750). Adjacent tissue section was processed for immunohistochemistry using Rabbit Polyclonal (Novus catalog # TA301431) at 1:1000 dilution with overnight incubation at 4 degrees Celsius followed by incubation with anti-rabbit IgG VisUCyte HRP Polymer Antibody (Catalog # VC003) and DAB chromogen (yellow-brown). Tissue was counterstained with hematoxylin (blue). Specific staining was localized to glandular cells.