

# **Product Datasheet**

# Anti-Anterior Gradient 2/AGR2 Antibody (orb389446)

Description	Anti-Anterior Gradient 2/AGR2 Antibody
Species/Host	Rabbit
Reactivity	Human, Mouse, Rat
Conjugation	Unconjugated
Tested Applications	FC, IHC, WB
Immunogen	E.coli-derived human Anterior Gradient 2 recombinant protein (Position: R21-L175). Human Anterior Gradient 2 shares 93.5% amino acid (aa) sequence identity with mouse Anterior Gradient 2.
Form/Appearance	Lyophilized
Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 $\mu$ g/ml.
Storage	Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at - 20°C in small aliquots to prevent freeze-thaw cycles.
Note	For research use only
Application notes	Western blot, 0.1-0.5µg/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin- embedded Section), 2-5µg/ml, Human Flow Cytometry(Fixed), 1-3 µg/1x106 cells, Human. Add 0.2ml of distilled water will yield a concentration of 500ug/ml
lsotype	Rabbit IgG
Clonality	Polyclonal
Antibody Type	Primary Antibody
MW	20 kDa

## **Biorbyt Ltd.**

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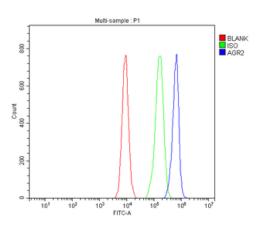
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# **Uniprot ID**

# 095994

## **Expiration Date**

12 months from date of receipt.

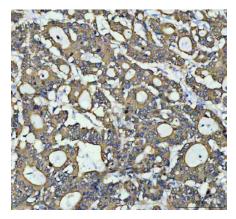


The cells were fixed with 4% paraformaldehyde and blocked with 10% normal goat serum. And then incubated with rabbit anti-AGR2 Antibody (1  $\mu$ g/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (5-10  $\mu$ g/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1  $\mu$ g/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Flow Cytometry analysis of CACO-2 cells using anti-AGR2

antibody. Overlay histogram showing CACO-2 cells (Blue line).

IHC analysis of AGR2 using anti-AGR2 antibody. AGR2 was detected in a paraffin-embedded section of human bladder infiltrating urothelial carcinoma with squamous differentiation tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 μg/ml rabbit anti-AGR2 Antibody overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.



IHC analysis of AGR2 using anti-AGR2 antibody. AGR2 was detected in a paraffin-embedded section of human colorectal adenocarcinoma tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2  $\mu$ g/ml rabbit anti-AGR2 Antibody overnight at 4°C. Peroxidase Conjugated Goat Antirabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.

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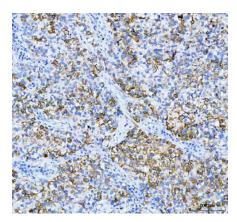
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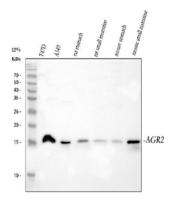
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IHC analysis of AGR2 using anti-AGR2 antibody. AGR2 was detected in a paraffin-embedded section of human non-small cell lung cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2  $\mu$ g/ml rabbit anti-AGR2 Antibody overnight at 4°C. Peroxidase Conjugated Goat Antirabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit with DAB as the chromogen.



Western blot analysis of AGR2 using anti-AGR2 antibody. Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. Lane 1: human T47D whole cell lysates, Lane 2: human A549 whole cell lysates, Lane 3: rat stomach tissue lysates, Lane 4: rat small intestine tissue lysates, Lane 5: mouse stomach tissue lysates, Lane 6: mouse small intestine tissue lysates. After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-AGR2 antigen affinity purified polyclonal antibody at  $0.5 \mu g/mL$  overnight at 4°C, then washed with TBS-0.1% Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit with Tanon 5200 system. A specific band was detected for AGR2 at approximately 17 kDa. The expected band size for AGR2 is at 20 kDa.

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