DR5 Antibody  
Cat. No.: 2019

Western blot analysis of DR5 in HeLa (H) and KS62 (K) cell lysates with DR5 antibody at 2 μg/mL.

**Figure 2 Western Blot Validation in Mouse Cell Lines**  
Loading: 15 μg of lysates per lane. Antibodies: DR5 2019, (1 μg/mL), 1h incubation at RT in 5% NFDM/TBST.  
Secondary: Goat anti-rabbit IgG HRP conjugate at 1:10000 dilution.

**Figure 3 Immunofluorescence Validation of DR5 in Mouse Kidney tissue**  
Immunofluorescent analysis of 4% paraformaldehyde-fixed mouse kidney tissue labeling DR5 with 2019 at 20 μg/mL, followed by goat anti-rabbit IgG secondary antibody at 1/500 dilution (red).

**Figure 4 Immunocytochemistry Validation of DR5 in HeLa Cells**  
Immunocytochemical analysis of HeLa cells using anti-DR5 antibody (2019) at 5 μg/ml. Cells was fixed with formaldehyde and blocked with 10% serum for 1 h at RT; antigen retrieval was by heat mediation with a citrate buffer (pH6). Samples were incubated with primary antibody overnight at 4˚C. A goat anti-rabbit IgG H&L (HRP) at 1/250 was used as secondary. Counter stained with Hematoxylin.

**Figure 5 Immunofluorescence Validation of DR5 in HeLa Cells**  
Immunofluorescent analysis of 4% paraformaldehyde-fixed HeLa cells labeling DR5 with 2019 at 20 μg/mL, followed by goat anti-rabbit IgG secondary antibody at 1/500 dilution (green).
Figure 6 Immunohistochemistry Validation of DR5 in Mouse kidney tissue
Immunohistochemical analysis of paraffin-embedded mouse kidney tissue using anti-DR5 antibody (2019) at 5μg/ml. Tissue was fixed with formaldehyde and blocked with 10% serum for 1 h at RT; antigen retrieval was by heat mediation with a citrate buffer (pH6). Samples were incubated with primary antibody overnight at 4˚C. A goat anti-rabbit IgG H&L (HRP) at 1/250 was used as secondary. Counter stained with Hematoxylin.

Figure 7 KO Validation of DR5 in HCT116 Cells (Han et al., 2015)
Anti-cancer drug, Carfilzomib (CFZ), induced up-regulation of DR5 and the expression of DR5 was not detected in DR5-KO HCT 116 cell line with anti-DR5 antibodies (2019).

Figure 8 KD Validation of DR5 in MB231 Cells (Rahman et al., 2009)
Western blot analysis with anti-DR5 antibodies was performed for DR5 in MB231 cells transfected with control siRNA or DR5 siRNA. DR5 expression was disrupted after DR5 siRNA knockdown.

Figure 9 Immunohistochemistry Validation of BIM in Human Colon Tumors (Devetzi et al., 2016)
Protein analysis for DR5 by immunohistochemistry with anti-DR5 antibodies in human colon tumors. Strong immunoreactivity is shown for DR5 in T167 patient with colorectal cancer.

Figure 10 Regulated Expression Validation of DR5 in Thyroid Epithelial Cells (Bretz et al., 2002)
Immunostaining with anti-DR5 antibodies shows high levels of DR5 expression in untreated cells and cells treated with each of the three cytokines alone or TNFalpha combined with IL-1β. In contrast, treatment with both IFNg and TNFalpha or all three cytokines greatly reduces DR5 staining. The reduction in staining appears most significant in cytoplasmic regions while some staining is maintained in or around the nucleus.

Figure 11 Immunofluorescence Validation of DR5 in Rat Brain Tissue with Tumors (Candolfi et al., 2009)
Rats were implanted in the striatum with CNS-1 tumors and 9 days later brains were processed for immunocytochemistry. Confocal microphotographs show detection of therapeutic targets (green) using specific antibodies against the death receptors TNFR1 and TRAILR2. Tumor cells were labeled with anti-vimentin antibodies (red), neurons were stained with anti-NeuN (red) and astrocytes with anti-GFAP antibodies (red). Nuclei were stained with DAPI (blue). T: tumor area. N: necrotic patch.

Specifications

<table>
<thead>
<tr>
<th>HOST SPECIES:</th>
<th>Rabbit</th>
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<td>SPECIES REACTIVITY:</td>
<td>Human, Mouse, Rat</td>
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### IMMUNOGEN:
DR5 antibody was raised against a peptide corresponding to 20 amino acids near the carboxy terminus of human DR5 precursor. The immunogen is located within the last 50 amino acids of DR5.

### TESTED APPLICATIONS:
ELISA, ICC, IF, IHC-P, WB

### APPLICATIONS:
- **WB:** 0.5-1 μg/mL; ICC: 5 μg/mL; IHC-P: 5 μg/mL; IF: 20 μg/mL.
- Antibody validated: Western Blot in human and mouse samples; Immunohistochemistry in mouse samples; Immunocytochemistry in human samples and Immunofluorescence in human and mouse samples. All other applications and species not yet tested.

### SPECIFICITY:
Antibody has no cross reaction to DR4.

### POSITIVE CONTROL:
1) Cat. No. 1201 - HeLa Cell Lysate
2) Cat. No. 1204 - K562 Cell Lysate
3) Cat. No. 17-001 - HeLa Cell Slide
4) Cat. No. 1282 - 3T3/NIH Cell Lysate
5) Cat. No. 1285 - C2C12 Cell Lysate

### PREDICTED MOLECULAR WEIGHT:
- Predicted: 48kD and 45kD
- Observed: 48kD and 45kD

### Advanced Validation

#### VALIDATION:
- **KO Validation** (Figure 7) shows DR5 expression detected by anti-DR5 antibodies (2019) was disrupted in DR5 KO HCT116 cells.
- **KD validation** (Figure 8): Anti-DR5 antibody (2019) specificity was further verified by DR5 specific knockdown. DR5 signal in MB231 cells transfected with DR5 siRNAs was disrupted in comparison with that in cells transfected with control siRNAs.
- **Regulated expression validation** (Figure 7,10): DR5 expression detected by anit-DR5 antibodies (2019) was up-regulated by carfilzomib treatment and was down-regulated by treatment with two or more cytokines.

### ISOFORMS:
- Human DR5 has 3 isoforms, including isoform L (440aa, 48kD), isoform S (411aa, 45kD) and isoform 3 (118aa, 13kD). Mouse DR5 has 1 isoform (381aa, 42kD), and Rat DR5 also has 1 isoform (336aa, 37kD). 2019 can detect two human isoforms and can also detect mouse and rat isoforms.

### Properties

#### PURIFICATION:
DR5 Antibody is affinity chromatography purified via peptide column.

#### CLONALITY:
Polyclonal

#### ISOTYPE:
IgG

#### CONJUGATE:
Unconjugated

#### PHYSICAL STATE:
Liquid
BUFFER: DR5 Antibody is supplied in PBS containing 0.02% sodium azide.

CONCENTRATION: 1 mg/ml

STORAGE CONDITIONS: DR5 antibody can be stored at 4°C for three months and -20°C, stable for up to one year. As with all antibodies care should be taken to avoid repeated freeze thaw cycles. Antibodies should not be exposed to prolonged high temperatures.

Additional Info

OFFICIAL SYMBOL: TNFRSF10B

ALTERNATE NAMES: DR5 Antibody: DR5, CD262, KILLER, TRICK2, TRICKB, ZTNFR9, TRAILR2, TRICK2A, TRICK2B, TRAIL-R2, KILLER/DR5, DR5, UNQ160/PRO186, Tumor necrosis factor receptor superfamily member 10B, Death receptor 5, TRAIL receptor 2

ACCESSION NO.: AF012535

PROTEIN GI NO.: 2338419

GENE ID: 8795

USER NOTE: Optimal dilutions for each application to be determined by the researcher.

Background and References

BACKGROUND: DR5 Antibody: Apoptosis is induced by certain cytokines including TNF and Fas ligand in the TNF family through their death domain containing receptors. TRAIL/Apo2L is a new member of the TNF family. DR4 was recently identified as the receptor for TRAIL. A novel death domain containing receptor for TRAIL was more recently identified and designated DR5, Apo2, TRAIL-R2, TRICK2, or KILLER by several groups independently. Like DR4, DR5 transcript is widely expressed in normal tissues and in many types of tumor cells. DR5 binds to TRAIL and mediates TRAIL induced cell death. Overexpression of DR5 induces apoptosis and activates NF-κB.

REFERENCES:

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