Anti-V5 Rabbit Monoclonal Antibody

Product Datasheet

Catalog# PAR04-100 Clone#RR697

Predicted Molecular Wt: Depending on customers' target of interest **Purity:** ProA affinity purified IgG

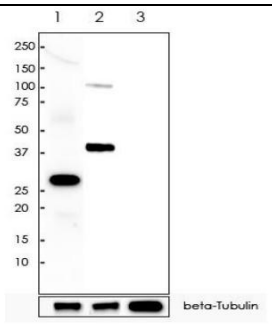
Species Cross-reactivity: Species independent **Form:** Liquid

Species cross-reactivity determined by WB **Swissprot ID:** N/A

Applications: WB IF/ICC FC IP

Background:

The V5 epitope tag is derived from a small epitope (Pk) present on the P and V proteins of the paramyxovirus of simian virus 5 (SV5). The V5 tag is usually used with all 14 amino acids (GKPIPNPLLGLDST), and useful for western blotting, immunofluorescence and immunoprecipitation experiments, although they also find use in antibody purification.



Predicted MW: Depend on fusion protein with V5 tag
Lane 1: 293 cell lysates transfected with N-terminal V5 tagged gene
(RR700 at 1:2,000 dilution).
Lane 2: 293 cell lysates transfected with C-terminal V5 tagged gene
(RR700 at 1:2,000 dilution).
Lane 3: Mock 293 cell lysates (RR700 at 1:2,000 dilution)
All lanes : 2 µg per lane
2nd Ab:
GAR HRP(H+L) 1:5,000
Exposure: 60s

Immunogen:

GKPIPNPLLGLDST (V5 epitope) conjugated to KLH

Storage Buffer:

PBS 59%, Sodium azide 0.01%, Glycerol 40%, BSA 0.05%.

Storage conditions:

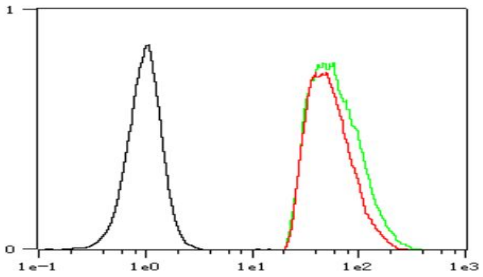
-20° C.

Storage instructions:

Shipped on blue ice. Upon delivery, aliquot, and store at -20°C. Avoid freeze / thaw cycles.

Recommended Dilutions:

WB:	1:1000 - 1:2000
IF/ICC:	1:2000- 1:10000
FC:	1:2000 - 1:10000
IP:	1:50



Overlay histogram showing 293 cells transfected with N - terminal (Red) and C-terminal (Green) V5 tagged gene stainedwith RR700. The cells were then incubated in the antibody(RR700 , 1:10,000 dilution) in 1x PBS/1% BSA for 30 min at room temperature. The secondary antibody used was a Goat Anti-Rabbit Alexa Fluor® 488 (IgG H+L) at 1:2,000 dilution for 20 min at room temperature. Unlabelled sample (Black) was used as a control.

Background References:

1. Kim D et al. Nucleic Acids Res 45:5112-5125 (2017).

2. Varney SD et al. J Cell Sci 129:774-87 (2016)

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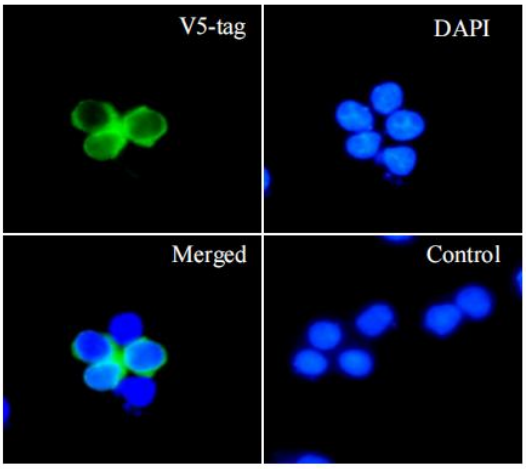
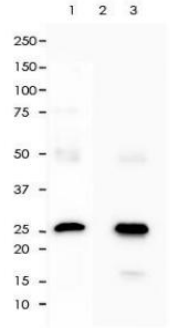
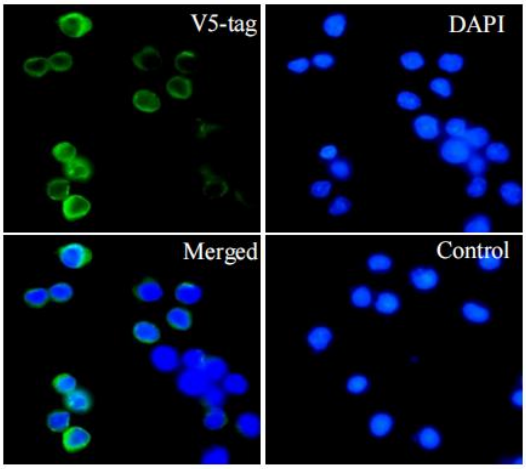


RR700 staining V5 tag in 293 cells transfected with N-terminal V5 tagged gene by IF/ICC (immunofluorescence/immunocytochemistry). Cells were fixed with paraformaldehyde, permeabilized with 0.1% Triton X-100 and blocked with 10% goat serum for half an hour at room temperature. Samples were incubated with primary antibody (1:10,000) at 4°C. An Alexa Fluor® 488-conjugated Goat AntiRabbit IgG polyclonal was used as the secondary antibody (1:500). DAPI (blue) was used as the nuclear counter stain.

Control: PBS and secondary antibody, An Alexa Fluor® 488- conjugated Goat Anti-Rabbit IgG (1:500)

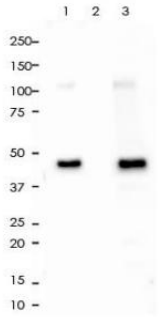


V5 tag was immunoprecipitated from 0.1mg of 293 whole cell lysates transfected with N-terminal V5 tagged gene with RR700 at 1:50 dilution.
2nd Ab:
GAR HRP for IP 1:500
Lane 1: RR700 IP in 293 whole cell lysates transfected with N-terminal V5 tagged gene
Lane 2: PBS instead of RR700 in 293 whole cell lysates transfected with N-terminal V5 tagged gene
Lane 3: 293 whole cell lysate transfected with N-terminal V5 tagged gene, 4 µg (input)
Exposure: 60s



RR700 staining V5 tag in 293 cells transfected with C-terminal V5 tagged gene by IF/ICC (immunofluorescence/immunocytochemistry). Cells were fixed with paraformaldehyde, permeabilized with 0.1% Triton X-100 and blocked with 10% goat serum for half an hour at room temperature. Samples were incubated with primary antibody (1:10,000) at 4°C. An Alexa Fluor® 488-conjugated Goat AntiRabbit IgG polyclonal was used as the secondary antibody (1:500). DAPI (blue) was used as the nuclear counter stain.

Control: PBS and secondary antibody, An Alexa Fluor® 488- conjugated Goat Anti-Rabbit IgG (1:500)



V5 tag was immunoprecipitated from 0.1mg of 293 whole cell lysates transfected with C-terminal V5 tagged gene with RR700 at 1:50 dilution.
2nd Ab:
GAR HRP for IP 1:500
Lane 1: RR700 IP in 293 whole cell lysates transfected with Cterminal V5 tagged gene
Lane 2: PBS instead of RR700 in 293 whole cell lysates transfected with C-terminal V5 tagged gene
Lane 3: 293 whole cell lysate transfected with C-terminal V5
tagged gene, 4 µg (input)
Exposure: 60s

Product QC’dby:

For research use only. Not for use in diagnostic or therapeutic applications.

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