

JOT0001-1: Anti-GFP VHH antibody

Clonality :	Monoclonal
Application :	IP,ELISA,WB
Gene :	GFP
Uniprot ID :	P42212
Alternative Name :	Green fluorescent protein
Isotype :	Camelid VHH

Description

Alpaca derived anti-GFP VHH single domain antibody (molecular weight:14.1 kDa) with a 6*His tag at its C-terminal, expressed in E. coli under conditions free from animal derived components.

Green fluorescent protein (GFP) is a protein composed of 238 amino acid residues(26.9kDa) derived from the Jellyfish *Aequorea victoria*, which emits green light (emission peak at 509nm) when excited by blue light (excitation peak at 395nm). GFP has become an invaluable tool in cell biology research, since its intrinsic fluorescence can be visualized in living cells. EGFP contains the double-amino-acid substitutions Phe-64 to Leu and Ser-65 to Thr (previously published as GFPmut1; PMID: 8707053). In contrast to wtGFP, EGFP has a single, strong, red-shifted excitation peak at 488 nm.

Specificity: Green fluorescent protein (GFP)

Affinity constant: KD of 1.1 nM by Biacore T200

This is a product from [Jotbody](#), Hong Kong. This antibody is made available worldwide by ABEOMICS Inc.

Product Info

Amount :	100 µg / 50 µg
Purification :	Affinity chromatography purified via Ni-charged resin
Content :	1 mg/mL by Nanodrop Buffer 25 mM TAPS pH8.5, 500 mM NaCl, 5 mM EDTA, 0.09 % NaN ₃
Storage condition :	4°C; Do not freeze.

Application Note

Positive controls : Positive ELISA detected in: recombinant full-length GFP protein Positive WB detected in: GFP-expressing 293T cells Positive IP detected in: GFP and anti-GFP VHH antibody co-expressing 293T cells Positive IF detected in: GFP-expressing Hela cells

Recommended dilutions: ELISA 1:2000-1:10000 WB 1:1000-1:5000 IP: 0.5-4.0 µg

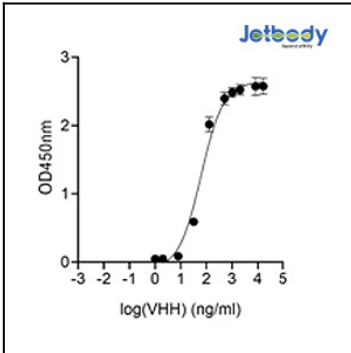


Figure 1: Indirect ELISA showing anti-GFP VHH antibody (JOT0001-1) binding to purified GFP. Plates were coated with 200ng/well purified protein and binding of JOT0001-1 assessed in serial dilution from 1ng/ml primary antibody in triplicate.

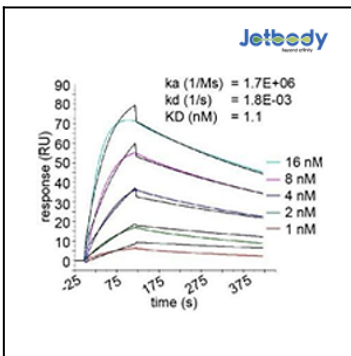


Figure 2: Captured recombinant GFP on CM5 Chip can bind anti-GFP VHH antibody (JOT0001-1) with an affinity constant of 1.1 nM as determined in SPR assay (Biacore T200).

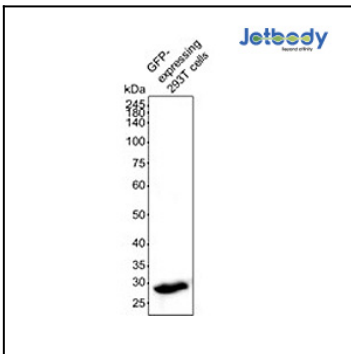


Figure 3: GFP-expressing 293T cell lysates were subjected to SDS-PAGE followed by western blot with anti-GFP VHH antibody (JOT0001-1) at dilution of 1:3000 incubated at room temperature for 1.5 hours.

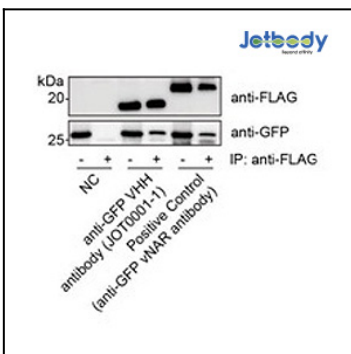


Figure 4 : 293T cells co-express GFP and single domain antibodies with a C-terminal 3xFLAG tag. VHH or VHH binding with GFP in cell lysate was captured by anti-FLAG magnetic beads and the eluate was then subjected to WB analysis. '-' represents pre-pulldown by anti-Flag magnetic beads. '+' represents post-pulldown by anti-Flag magnetic beads.

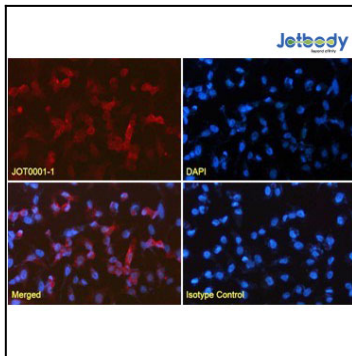


Figure 5 : Immunofluorescence analysis of paraformaldehyde fixed GFP-expressing HeLa cells stained with anti-GFP VHH antibody (JOT0001-1) at 2.5 $\mu\text{g/ml}$ followed by CoraLite® 594 secondary antibody at 1:200 dilution, showing cytoplasmic staining (under 40x lens). The nuclear stain is DAPI (blue). The isotype control was stained with anti- unknown antibody followed by CoraLite® 594 secondary antibody (under 40x lens).