For Research Use Only

Nano-Secondary® anti-mouse IgG2b, recombinant VHH, Alexa Fluor® 647 [CTK0105, CTK0106]



www.ptglab.com

Catalog Number: sms2bAF647-1

4 Publications

Basic Information

Catalog Number: sms2bAF647-1 **Applications:** IF, WB Host: Alpaca Conjugate: Alexa Fluor® 647 Type:
Mixture of 2 monoclonal Nanobodies

Class: Recombinant RRID: AB_2827583

Purification Method:

Recombinant expression, affinity purification

Description

Nano-Secondary® anti-mouse IgG2b, Fc-specific recombinant VHH is an anti-mouse IgG subclass specific antibody. This secondary antibody product consists of Nanobodies that bind to mouse IgG2b with high affinity & specificity.

Species Reactivity

Mouse IgG2b Fc-fragment No cross-reactivity: goat, guinea pig, rabbit, rat, sheep, human, macaque serum proteins, mouse IgG1, IgG2a, IgG2c, IgG3

Physical State

Suggested Dilution

Immunofluorescence 1:1,000

Western blot 1:1,000

Affinity (K_D)

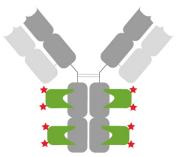
CTK0105: $K_D = 5 \text{ nM}$, CTK0106: $K_D = 0.2 \text{ nM}$

Storage

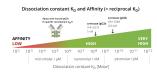
Storage: Store at -20°C short term or -80°C long term. Aliquot upon delivery. Avoid freeze-thaw cycles.

10 mM HEPES pH 7.0, 500 mM NaCl, 5 mM EDTA, Preservative: 0.09 % Sodium azide

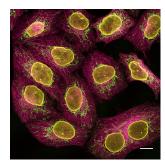
Selected Validation Data



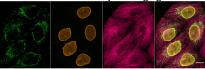
Anti-mouse IgG2b Nano-Secondary: Well-defined and characterized immunostaining. Primary antimouse IgG2b antibody (grey) with 2X2 monoclonal mouse Fc-specific Nanobodies (green) bound. In total, 8 fluorophores (red stars) label the mouse IgG2b primary antibody.



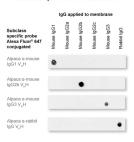
Dissociation constant Kd and affinity.



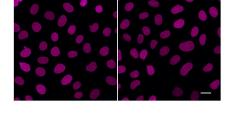
Multiplexed immunostaining of HeLa cells with two alpaca anti-mouse Nano-Secondaries and one anti-rabbit Nano-Secondary. Green: mouse IgG1 anti-COX4 + alpaca anti-mouse IgG1 VHH Alexa Fluor® 488. Magenta: mouse IgG2b anti-Tubulin + alpaca anti-mouse IgG2b VHH Alexa Fluor® 647. Yellow: rabbit anti-Lamin + alpaca anti-rabbit IgG VHH Alexa Fluor® 568. Scale bar, 10 µm. Images were recorded at the Core Facility Bioimaging at the



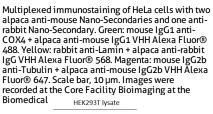
Subclass specific Nano-Secondaries

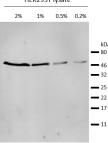


The anti-mouse IgG2b Nano-Secondary is subclass-specific and does not cross-react with IgGs from other commonly used species (here rabbit) and with mouse IgG1 and IgG3 subclasses.



One-step staining (left) vs. sequential staining (right) of HeLa cells with anti-Lamin A/C (nuclear lamina) mouse IgG2b monoclonal primary antibody + alpaca anti-mouse IgG2b VHH Alexa Fluor® 647 (magenta). Scale bar, 20 µm.

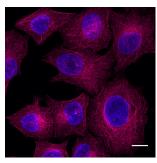




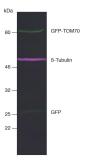
Western blot analysis of endogenous ®-Tubulin in HEK293T cell lysate. Detection with mouse anti-®-Tubulin antibody and alpaca anti-mouse IgG2b VHH Alexa Fluor® 647.



One-step immunostaining is the simultaneous incubation of mouse IgG2b primary antibody and anti-mouse IgG2b Nano-Secondary. This method reduces incubation and hands-on time. Simultaneous incubation also supports multiplexing, tissue penetration, and cell staining for flow cytometry.



HeLa cells were immunostained with mouse IgG2b anti-®-Tubulin antibody + alpaca anti-mouse IgG2b VHH Alexa Fluor® 647 (magenta). Nuclei were stained with DAPI, blue. Scale bar, 10 µm. Images were recorded at the Core Facility Bioimaging at the Biomedical Center, LMU Munich.



Multiple Nano-Secondaries can be applied for multiplex fluorescent Western blotting. This allows multiple targets to be analyzed simultaneously on the same blot at the same time. Multiplex fluorescent Western blot of GFP-TOM70, ®-Tubulin, and GFP in HEK293T cell lysate. Western blot membrane was simultaneously incubated with primary antibodies and Nano-Secondaries. Green: rabbit anti-GFP (ChromoTek PABG1) + alpaca antirabbit lgG VHH Alexa Fluor® 488. Magenta: mouse anti-®-Tubulin + alpaca anti-mouse IgG2b VHH Alexa Fluor® 647.