### Nanobody toolbox for your research

## PRODUCT SPECIFICATION

Recombinant anti-mouse Talin nanobody 19.

Catalogue number: sdAb-Talin-Nb19



### **Background**

**Talin** is a very large (~270 kDa) mechanosensitive cytoskeletal protein that links the actin cytoskeleton to integrins. Hence, it is concentrated in regions where cells make contact with the substrate or with other cells. Talin is typically found in focal adhesions, the immune synapse (immune cells) and invadopodia (cancer cells) or podosomes (immune cells). Upon 'stretching' of the protein, vinculin binding sites are exposed.

Sources: Wikipedia, Sun et al., NCB review 2019 https://doi.org/10.1038/s41556-018-0234-9.



Model depicting Talin, a multifunctional 2541 amino acid long protein.

<u>Applications</u>: PD, IP, ELISA. This product is for R&D use only, not for drug, diagnostic,

therapeutic, household, or other uses. Not suitable for Western blot.

#### Nanobody functionality:

Immunoprecipitation of endogenous Talin from MDA-MB-231 cancer cell extracts with Talin 19 nanobody.

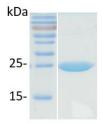


Procedure: 0.64 mg protein extract from MDA-MB-231 cells (lyzed in 20 mMTris/HCl pH 7.5, 1 % Triton X-100, inhibitor cocktail and PMSF) was incubated with 1  $\mu$ g HA-tagged Talin nanobody 19 for 1 hour at 4°C. Next, this mixture was added to 10  $\mu$ l anti-HA antibody coupled to settled sepharose beads, again for 1 hr at 4°C. Following 4 washes with 1 ml lysis buffer, Laemmli sample buffer was added to the beads and boiled for 2 minutes. The supernatant was size fractionated by SDS-PAGE (6%) and then proteins were transferred to nitrocellulose by conventional blotting. The blot was blocked with 5% milk powder in Tris buffered saline. Primary antibody was rabbit polyclonal anti Talin Ab 1/1000 dilution. A HRP-coupled antibody was used as secondary (1/2000). Finally, the blot was exposed hyperfilm for 13 minutes.

#### Source and properties

Talin nanobody 19 was raised by immunizing a llama with a fragment of mouse Talin encompassing residues 2364-2541. It binds to the talin fragment with an **affinity of 6 nM**  $(6,55 \times 10^{-9} \text{ M}, \pm 1.64 \times 10^{-10} \text{ M})$ .

Figure: purified talin fragment used for immunization. SDS-PAGE (15% gel) followed by Coomassie staining.



Availability: Nanobody 19 comes with a COOH-terminal HA or Myc epitope tag. Available in 100

 $\mu$ g, 500  $\mu$ g, 1000  $\mu$ g quantities. For bulk amounts, please inquire.

<u>Expression host</u>: VHH single domain antibody purified from *E. coli*.

<u>Cross reactivity</u>: Reactivity: mouse and human. Other species have not been tested yet.

Storage buffer: 20 mM Tris-HCl pH 8.0, 150 mM NaCl, 1mM DTT, 60 % glycerol. Store at -20°C.

The sample will not freeze. Maintain sample in cold environment during transport to

increase longevity.

Store at -20°C upon arrival. For long term storage, aliquot and store at -80°C. Avoid

repeated freeze/thaw cycles.

# **Product citations:**

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