PRODUCT SPECIFICATION

Recombinant anti-human alfa-tubulin nanobody 14/17.

Catalogue number: sdAb-tubulin-Nb14/17



Background

Tubulin is a major component of the cytoskeleton, together with intermediate filaments and actin filaments. It has a molecular weight of 55 kDa and different isoforms of tubulin exist: alfa, beta, gamma, ... The scientific literature on tubulin is vast and not surprisingly, this protein is a key component in many cellular phenomena.

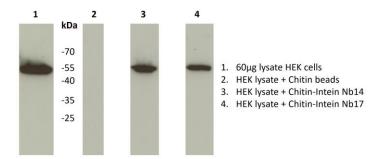
Tubulin is the building block of microtubules. These are tube-like structures and arise by a process known as polymerization. Tubulin is also known to bind and hydrolyse GTP (for comparison, actin binds and hydrolyzes ATP). Tubulin is a target for different cytostatics. These are usually small pharmacological compounds like paclitaxel (taxol) that bind to tubulin and block its biological functions. As a result, microtubules cannot be formed or cannot disassemble by depolymerization. This prevents cancer cells (but also normal cells) to undergo normal cell division. Since cancer cells divide more rapidly than normal cells, they are primarily targeted.

Applications:

PD, IP, ELISA. Not suitable for Western blot. This product is for R&D use only, not for drug, diagnostic, therapeutic, household, or other uses.

Nanobody functionality:

Immunoprecipitation of endogenous alfa-tubulin from HEK 293T cell extracts with tubulin nanobody 14 and 77.



Procedure: 1 mg protein extract from HEK 293T cells (lyzed in 20 mMTris/HCl pH 7.5, 1 % Triton X-100, inhibitor cocktail and PMSF) was incubated with 1 μ g intein-tagged tubulin nanobody 14/17 for 1 hour at 4°C. The nanobodies were coupled onto chitin beads. 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 (8%) 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 mouse monoclonal against alfa-tubulin at 1:1000 dilution. A HRP-coupled antibody was used as secondary. Finally, the blot was exposed to hyperfilm.

Source and properties

Tubulin nanobodies were raised by immunizing a llama with the full length protein.

Availability: Tubulin nanobodies 14/17 come 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 of this nanobody with tubulin from species other than human has not been

tested.

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.

<u>Product citations</u>: /