

PRODUCT SPECIFICATION

Recombinant Manduhai alfa-actinin 4 nanobody 64.

Catalogue number: sdAb-ACT4-Nb64/Man.



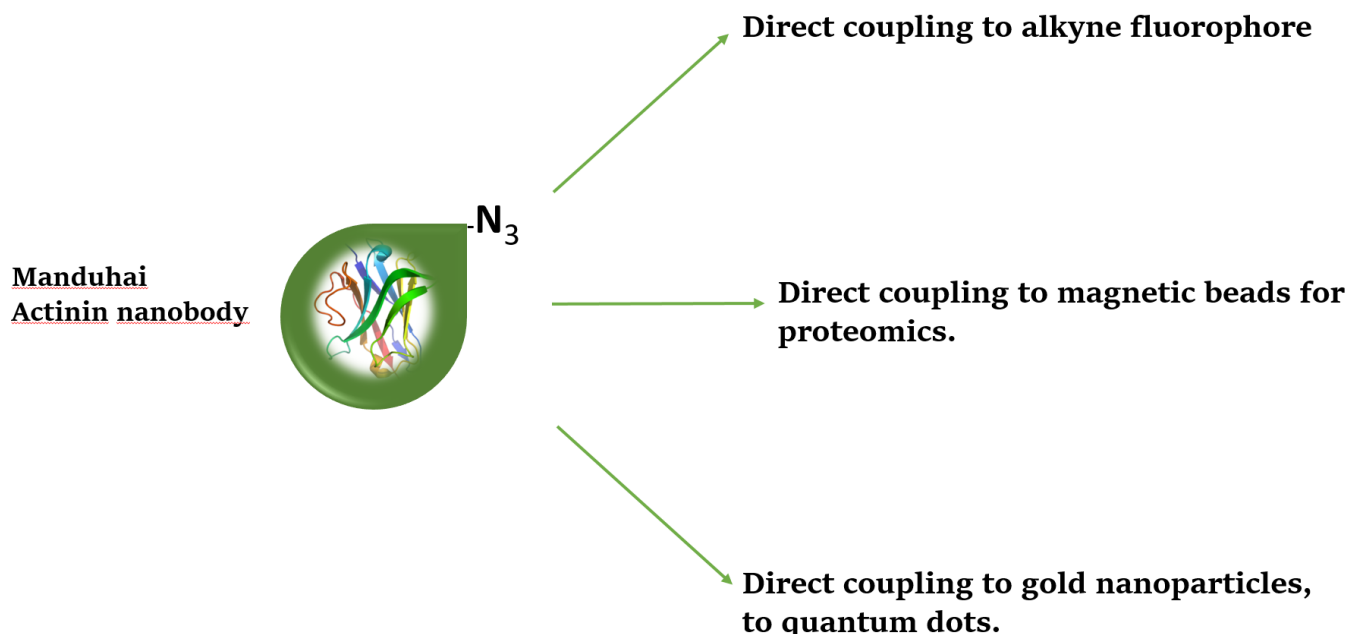
Background

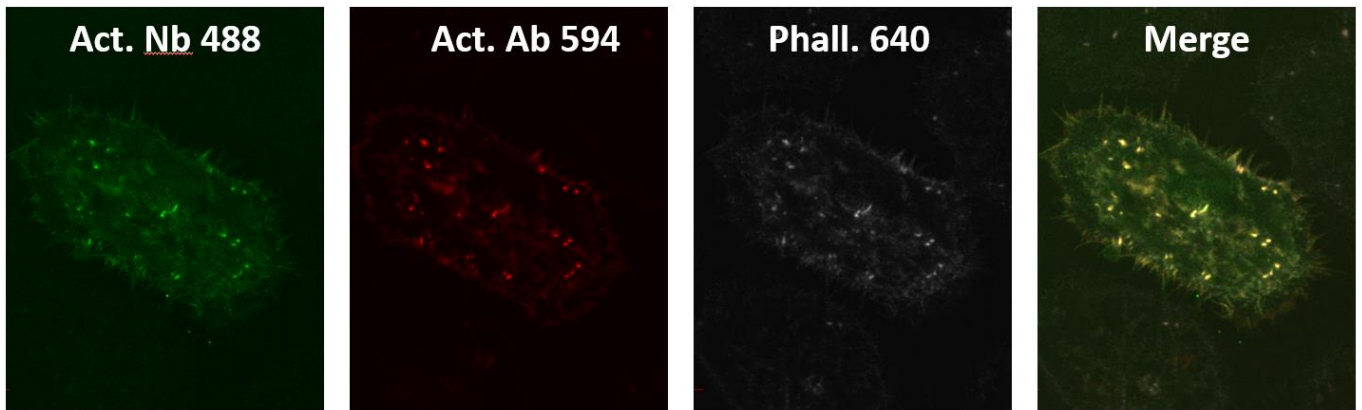
Alfa-actinin4 is a cytoskeletal protein and actin cross-linker encoded by the *ACTN4* gene, and is a member of the spectrin superfamily of cytoskeletal proteins. Together with isoform 1, actinin-4 is found in nearly every non-muscle cell but also in cancer cells. The N-terminal pair of Calponin-homology domains constitute an actin binding site whereas the spectrin repeats likely form a rod-like domain. It forms homodimers and has been observed to shuttle between nucleus and cytoplasm under certain conditions. Alfa-actinin function is not restricted to cell motility and cytoskeletal organization, but extends to modulation of gene expression, apoptosis and endocytosis. Actinin-4 deficient mice develop proteinurea and glomerular disease.

Derivatized Actinin nanobody for click chemistry

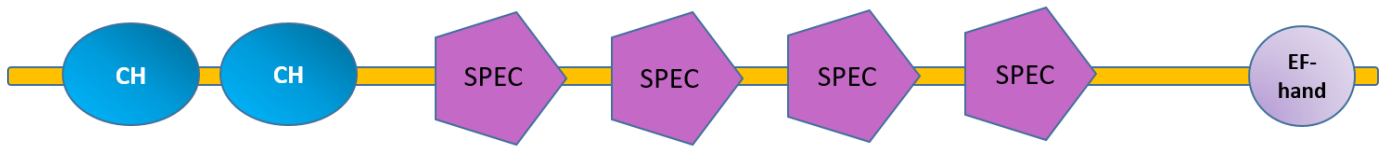
The Manduhai Actinin 4 nanobody 64 carries at its carboxy-terminus a *para*-azido-Phe residue, enzymatically inserted. This residue is the same as natural Phe, except that it carries an azido group in its aromatic ring (-N₃). As a result, the nanobody is endowed with a singular reactive group, allowing down stream *click chemistry*. *Reproducible and site-specific labeling becomes standard in this way.* Through this modification the antigen binding properties of the nanobody remain unchanged because the carboxy-terminal region of a nanobody is generally not involved in antigen binding.

New possibilities arise for research:





Images show a HNSCC-61 head and neck squamous carcinoma cell that was *photoporated* with Manduhai Actinin Nb-488 (green) according to Liu et al. (2020, <https://doi.org/10.1007/s12274-020-2633-z>). The cell was stained with anti-actinin antibody 594 for comparison (red) and with phalloidin 640 (grey) for actin. The cells were coated on gelatin. Actinin 4 localizes in invadopodia, dot-like structures involved in focal degradation of the underlying extracellular matrix, facilitating escape of cancer cells from the primary tumor.



Model depicting alfa-actinin4 with its 2 NH₂-terminal calponin homology domains, 4 central spectrin repeats, and COOH-terminal EF-hand.

Source and properties

Alfa-actinin 4 nanobody 64 was raised by immunizing a llama with a protein fragment encompassing actinin 4 amino acids 404-765. Nanobody 64 binds to the actinin fragment with a K_d of 1.2×10^{-8} M.

The actinin4 fragment used for immunization is shown below:

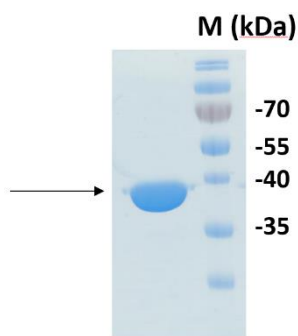


Figure: purified actinin 4 fragment used for immunization. SDS-PAGE (10% gel) followed by Coomassie staining. M = protein standards (kDa).

Availability: The Manduhai Alfa-actinin 4 nanobody 64 comes with a COOH-terminal para-Azido-Phe residue. Available in 10 µg, 50 µg, 100µg quantities. For bulk amounts, please inquire.

Cross reactivity: Reactivity of this nanobody with alfa-actinin 4 from species other than human has not been tested.

Storage buffer: 20 mM Tris-HCl pH 8.0, 150 mM NaCl, 1mM DTT.

Stability: 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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Please enquire about other actinin 4 nanobodies at info@gulliverbiomed.com