

## PRODUCT SPECIFICATION

Recombinant anti-human Son-of-Sevenless1 nanobody 25.

Catalogue number: sdAb-SOS1-Nb25



### Background

SOS1 is a large multidomain protein and guanine nucleotide exchange factor for Ras small GTPases. In view of its multiple functional domains, prominent role in cell signaling and close association with the plasma membrane, SOS1 participates in regulation of many biological events including, but not limited to, G-protein coupled receptor signaling, blood vessel morphogenesis, axon guidance, pericardium morphogenesis, insulin receptor signaling, regulation of Rho signal transduction, leukocyte migration, ... Sources: <http://www.uniprot.org>, <http://smart.embl-heidelberg.de>, Rojas JM, Oliva JL, Santos E (2011) Mammalian son of sevenless Guanine nucleotide exchange factors: old concepts and new perspectives. *Genes Cancer* 2: 298-305.



Schematic, depicting SOS1 domain organization. PR = proline-rich.

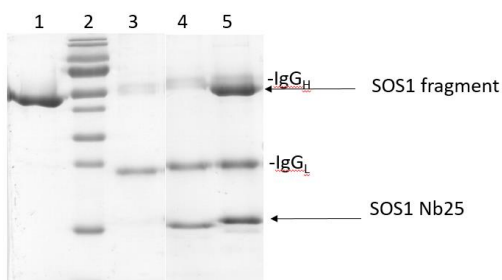
### Applications:

Currently: pull down of recombinant SOS1. This nanobody is not suitable for Western blot and is apparently not able to immunoprecipitate SOS1 from resting mammalian cells, possibly because the epitope of the nanobody is cryptic in full length SOS1, but this has not yet been further studied. Indeed, literature indicates that the protein adopts an auto-inhibited conformation that arises through inter-domain interactions that block access of the SOS1 catalytic core to its Ras-GTPase targets (*Sondermann et al., 2004, Cell. 119, 393–405*).

Other applications have not yet been tested. This product is for R&D use only, not for drug, diagnostic, therapeutic, household, or other uses.

### Source and properties

SOS1 nanobody 25 was raised by immunizing a llama with a recombinant SOS1 protein fragment encompassing the first ~400 amino acids of SOS1, up to the RhoGEF domain. Nanobody 25 binds to the SOS1 fragment with a  $K_d$  around  $1 \times 10^{-9}$  M.



#### Legend:

1. Purified recombinant SOS1 fragment
2. Protein marker
3. Negative control: anti-HA agarose + SOS1 fragment
4. Pull down with an irrelevant nanobody
5. Pull down with SOS1 Nb25.

### Pull down experiment:

Purified SOS1 (lane 1) was incubated with HA-tagged SOS1 Nb25 and the complex was immobilized on anti-HA agarose. Following boiling in Laemmli sample buffer, the immunoprecipitate was analyzed by SDS-PAGE (15%) and stained with Coomassie brilliant blue. Note that nanobody and SOS1 fragment stain virtually equally intense on the gel, indicative of a strong binder. This is confirmed in the  $K_d$  (~1nM).

The recombinant SOS1 fragment used for immunization is shown below:

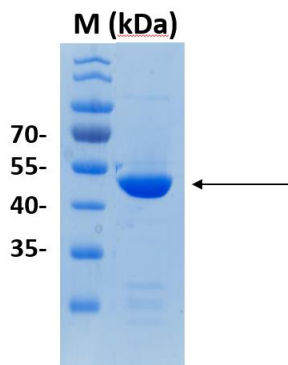


Figure: purified SOS1 fragment encompassing the Histone and RhoGEF domains, used for immunization. SDS-PAGE (10% gel) followed by Coomassie staining. M = protein standards (kDa).

Use: This nanobody can be useful in biochemical experiments and structural studies, i.e. X-ray crystallography or SAXS.

Availability: SOS1 Nanobody 25 comes with a COOH-terminal HA epitope tag. Available in 100 µg, 500 µg, 1000 µg quantities. For bulk amounts, please inquire.

Expression host: VHH single domain antibody purified from *E. coli*.

Cross reactivity: Reactivity of this nanobody with SOS1 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.

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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