# anti-VASP

# rabbit antiserum M4

Lot: A02

data sheet - catalog #: 0010-10



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#### Background information

VASP (vasodilator stimulated phosphoprotein) is a proline-rich<sup>21</sup> protein substrate of cAMP- and kinases<sup>18,20,26-43</sup> protein cGMP-dependent Phosphorylation of VASP at Ser-157 causes a mobility shift in SDS gel electrophoresis from 46 to 50 kDa<sup>25</sup>, which has been used as a convenient marker to monitor cyclic nucleotidedependent protein kinase activity 16,18,20,26-40 VASP is the founding member of the Ena-VASP protein family, comprising the Drosophila protein Enabled (Ena)<sup>19</sup>, its mouse homologue Mena (mammalian Enabled)9, and mouse EVL (Ena-VASP-like protein)9. With these proteins VASP shares a conserved overall domain organization 9,21: a) the conserved N-terminal Ena-VASP homology domain 1 (EVH1), which mediates binding to a novel proline-rich motif<sup>1,6,10</sup>, b) a more divergent proline-rich central domain (which is responsible for profilin binding)<sup>4,5,24</sup>, and c) a conserved C-terminal EVH2 domain. Particularly high VASP levels are present in platelets 33,36,41, although VASP is expressed in a variety of mammalian cell types and tissues 11,32,33,36,41. In cultured cells, VASP is associated with focal adhesions, cell-cell contacts, microfilaments, and highly dynamic membrane regions<sup>2,24,36</sup>. From *in vitro* binding data VASP has been suggested to link profilin to zyxin<sup>25</sup>, vinculin<sup>6,10</sup>, and the *Listeria spp.* surface protein ActA<sup>8,17,22</sup>, respectively. Functional evidence indicates that VASP is a crucial factor involved in the enhancement of actin filament formation and the actin-dependent motility of intracellular bacterial pathogens 1-3,10,12-14,23.

#### Serum preparation and storage

100 µl of crude rabbit antiserum M4 (Ref. 40) containing 0.01% (w/v) NaN<sub>3</sub> (vials have been overfilled by 10% to ensure complete recovery of the specified amount). For repeated use store at 4°C (short term), stable for one year from date of shipment when stored at -20°C.

#### Antigen

VASP purified<sup>42</sup> from human platelets.

## Species cross-reactivity 21,36,41

human, mouse, rat, canine, porcine, chicken, marsupial

#### Specificity

The serum recognizes both the 46 kDa (Ser-157 dephospho) and 50 kDa (Ser-157 phospho) form of VASP. Especially with rodent cells and tissues there may be a reactivity with a 80 kDa band of yet unknown identity<sup>2</sup>.

## **Applications**

Western (immuno) blotting (dilution 1:1500 - 1:3000), immunofluorescence of acetone or formaldehyde fixed cells (dilution 1:500), immunoprecipitation (dilution 1:125).

All dilutions refer to the analysis of cells and tissues with intermediate to high levels of VASP expression and must be viewed as approximate. The serum should be titrated for each individual application.

#### Positive control

Human platelet protein (500  $\mu$ g), supplied at 5 mg/ml in SDS-stop solution (100 mM NaCl, 73 mM Tris/HCl pH 6.7, 10 mM DTT, 8 mM EDTA, 5 % [v/v] glycerol, 2% [w/v] SDS, 10  $\mu$ g/ml Bromophenol Blue sodium salt). Use 5  $\mu$ l (25  $\mu$ g) per lane.

#### Related products

- Pre-immune serum to M4, 25 μl (catalog # 0013-02)
- monoclonal antibody IE273 to human VASP, 50 μg (catalog # 0016-05)
- positive control: human platelet protein in SDSstop solution, 500 μg (catalog # 8010-50)

#### References

## (\*: papers referencing the VASP antiserum M4)

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