

## anti-Sds22 (S. pombe) antibody, rabbit serum

63-141 100 μl

Shipping and Storage: Shipped at 4°C or -20°C and store at -20°C.
Immunogen: Recombinant C-terminal region (1.8kb) of S. pombe Sds22 (1)
Form: Rabbit antiserum added with 0.05 % sodium azide
Reactivity: Specific to S. pombe

## Applications:

- 1. Immunoblotting (dilution: 1/200~1/500) (Ref 1,2,3)
- 2. Immunoprecipitation (Ref 2)
- 3. Immunofluorescence microscopy

**Background**: *Schizosaccharomyces pombe* **Sds22** protein contains leucine-rich repeats and physically interacts with the catalytic subunits of two type 1 protein phosphatases (Dis2 and Sds21). **Sds22** is a regulatory subunit of these phosphatases and the **Sds22**-bound phosphatases carry a key phosphatase activity essential for the progression from metaphase to anaphase. **Sds22** is essential for cell viability and in its absence, cells were blocked in metaphase. **Sds22** protein is predicted to form a repeating helical rod that is capable of enhancing a PP1-dependent dephosphorylation activity.

Data Link: Swiss-Prot P22194

**References:** This antibody has been used in Ref. 1, 2 and 3.

- Ohkura H and Yanagida M "S.pombe gene sds22+ essential for a midmitotic transition encodes a leucine-rich repeat protein that positively modulates protein phosphatase-1." *Cell* 64: 149-157 (1991) PMID: <u>1846086</u>
- Stone EM *et al* "Mitotic regulation of protein phosphatases by the fission yeast sds22 protein." *Curr Biol* 3: 13-26 (1993) PMID: <u>15335873</u>
- Ishii K et al "Requirement for PP1 phosphatase and 20S cyclosome/APC for the onset of anaphase is lessened by the dosage increase of a novel gene sds23<sup>+</sup>." EMBO J. 15:6629-6640 (1996) PMID: <u>8978689</u>

to be continued



- 40kD

140-2

wt

Fig.1 Immunoblot with anti-Sds22 antiserum of yeast extracts from (1) wild type strain HM123, (2) *sds::ura4+* deletion mutant carrying pHR140-2 (ref.2).

The 40kD protein band was identified by immunoblot analysis of wild-type strain using anti-Sds22 antisera (lane1). The 40 kD band is enhanced in the *sds22::ura4+* disruptrion mutant strain that is rescued by the multicopy *sds22+* plasmid pHR140-2 (lane2).

## Fig.2 Sds22 coprecipitates with Dis2 and Sds21 (ref.2).

Yeast extracts of wild type (wt) strain HM123 (lane 1 and 2), dis2::ura4+ deletion mutant ( $\Delta$  d2, lane 3), sds21::ura4+ deletion mutant ( $\Delta$  s21, lane 4) were immunoprecipitated followed by immunoblotting with the indicated antiserum, to detect the Sds22 or Dis2/Sds21 proteins.

Lane 1 was immunoprecipitated with the appropriate preimmune serum, lane 2-4 with the anti-Sds22 serum.

(a) denotes anti-Sds22 immunoblot; (b) denotes anti-D2C immunoblot. Anti-D2C crossreacts with both Sds21 and Dis2.

Anti-Sds22 antiserum coprecipitates both Dis2 and Sds21 proteins in the wild type strain (lane 2b). Consistently, Sds21 alone is precipitated in the *dis2* deletion mutant (lane 3b), and Dis2 alone is precipitated in the *sds21* deletion mutant (lane 4b).

## Fig.3 Sds22 subcellular localization

Indirect immunofluorescence microscopy was performed by staining methanol fixed cells with (first column) anti-Sds22 antiserum, or (second column) DAPI to visualize chromosomal DNA.

(a) wild type HM123; (b) HM123 carrying multicopy *sds22+* plasmid pHR140-2. Anti-Sds22 antibody stains the cytoplasm as well as the non-chromosomal domain of the nucleus of a wild type strain, as shown in (a). Nuclear staining increases in strains carrying a multicopy *sds22+* plasmid (b).





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