

Anti-SPACA1 antibody, rabbit polyclonal, KO-Validated

73-062 100 μg

Shipping and Storage: Shipped at 4° C or -20° C and store at -20° C.

Immunogen: C-terminal peptide of mouse SPACA1 protein, QSPTDIPVHEDDALSEWNE,

conjugated with KLH

Validation: Specificity validated with knock-out mice.

Form: 1.0 mg/ml IgG fraction of antiserum in PBS-, 50% glycerol, 0.05% sodium azide.

Reactivity: Mouse.

Applications:

1. Western blotting (1/1,000 dilution))

2. Immunofluorescence staining (1/100~1/500 dilution).

Background: SPACA1 is a membrane protein that localizes in the equatorial segment of spermatozoa in mammals and is reported to function in sperm-egg fusion. Disruption of Spaca1 led to the disappearance of the nuclear plate, a dense lining of the nuclear envelope facing the inner acrosomal membrane. This coincided with the failure of acrosomal expansion during spermiogenesis and resulted in the degeneration and disappearance of the acrosome in mature spermatozoa. SPACA1-deficient male mice are infertile.

Molecular mass: Mouse; 33,343 Da with 305 amino acids

Database Links: uniprot/Q9DA48 mouse SPACA1, Gene ID 67652 mouse Spaca1

Reference: This antibody was described and used in the following publication.

Fujihara Y. SPACA1-deficient male mice are infertile with abnormally shaped sperm heads reminiscent of globozoospermia. <u>Development.</u> (2012) 139:3583-9. **WB, IF.** Free access.

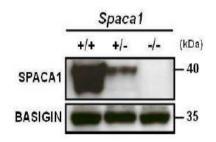


Fig.1 Western blotting of mouse testis extracts of wild-type and *Spaca1* knockout mice with anti-SPACA1 antibody.

20 μ g of Triton X-100 extracts from testes of wild type (+/+), Spaca1 single (+/-) and double (-/-)knock-out mice was reacted with anti-SPACA1 antibody at 1/1,000 dilution. Basigin was used as a loading control

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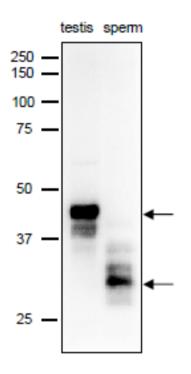


Fig. 2 Different forms of SPACA1 were identified in mouse testis and sperm extracts by western blot analysis with anti-SPACA1 antibody.

Anti-SPACA1 antibody was used at 1/1,000 dilution and as second antibody, goat anti-rabbit IgG conjugated with HRP (ab97051) was used at 10,000 dilution.

Several proteins associated with sperm undergo modification during maturation. Multiple bands are reflection of post-translational modifications by glycosylation and phosphorylation.

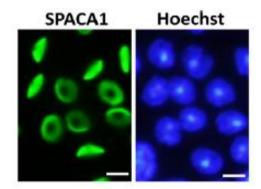


Fig.3. Immunofluorescence staining of SPACA1 in step 7 round spermatids.

Spaca1 protein in 7 round spermatids was reacted with anti-SPACA1 antibody and as a secondary antibody, Alexa-Fluor 488 conjugated anti-rabbit IgG antibody (green) was used. DNA was stained with Hoechst 33258.