# **Product Datasheet**

# SCP3/SYCP3 Antibody - BSA Free NB300-232SS

Unit Size: 0.025 ml

Store at 4C. Do not freeze.

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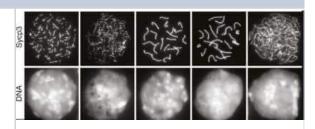
# NB300-232SS

SCP3/SYCP3 Antibody - BSA Free

Product Information	
Unit Size	0.025 ml
Concentration	1 mg/ml
Storage	Store at 4C. Do not freeze.
Clonality	Polyclonal
Preservative	0.02% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	PBS
Product Description	
Host	Rabbit
Gene ID	50511
Gene Symbol	SYCP3
Species	Human, Mouse, Rat, Porcine, Bovine, Chicken, Feline, Parasite
Reactivity Notes	Chicken reactivity reported in scientific literature (PMID: 26096940). Parasite, and Feline reactivity reported in scientific literature (PMID: 27084479). Human reactivity has been observed in IHC. Human reactivity reported in scientific literature (PMID: 29231814).
Immunogen	A synthetic peptide made to the C-terminal region of the human SCP3 protein. [UniProt# Q8IZU3]
Product Application Details	
Applications	Western Blot, Immunoblotting, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry- Paraffin, Immunoprecipitation
Recommended Dilutions	Western Blot 0.5 ug/ml, Immunohistochemistry 1:200-1:500, Immunocytochemistry/ Immunofluorescence 1:100-1:500, Immunoprecipitation reported in scientific literature (PMID 18694876), Immunohistochemistry-Paraffin 1:100 - 1:500, Immunohistochemistry-Frozen 1:200-1:500, Immunoblotting reported in scientific literature (PMID 27612028)
Application Notes	In Western blot, a band is seen at ~28 kDa.

### Images

Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-232] - Chromosome spread for sycp3. ICC/IF image submitted by a verified customer review.



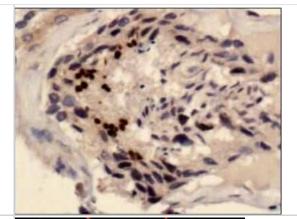


Immunohistochemistry-Paraffin: SCP3/SYCP3 Antibody [NB300-232] -IHC analysis of a formalin fixed and paraffin embedded tissue section of Mouse testis using SCP3 antibody at 1:1000 dilution. The signal was detected using HRP-DAB detection method and the nuclei were counterstained with hematoxylin. The antibody generated a specific signal of SCP3 in the spermatogonial cells and the spermatocytes. The signal was strongest in the nuclei of spermatogonial cells. Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody Leptotene Zygotene [NB300-232] - Exposure to DEHP impairs meiotic progression of oocytes from pachytene to diplotene. Immunolabeling of the oocyte chromosomes with anti-SYCP3 antibody (red) and Hoechst 33342 (blue). All experiments were repeated at least three times independently. (\* P<0.05; \*\* P<0.01) Image collected and cropped by CiteAb from the following publication Diplotene Pachytene (https://www.nature.com/doifinder/10.1038/cddis.2017.350), licensed under a CC-BY license. Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-232] - Loss of lamin C2 has no effect on meiotic telomere attachment. Chromosome spread preparations of pachytene-like lamin C2-/- spermatocytes showing that all telomeres are associated with SUN1. Anti-SUN1 staining in co-localisation with SYCP3. Image collected and cropped by CiteAb from the following publication (https://dx.plos.org/10.1371/journal.pgen.1003261), licensed under a CC-BY license. Western Blot: SCP3/SYCP3 Antibody [NB300-232] - SCP3 Antibody kDa [NB300-232] - Analysis of SCP3 in mouse testis protein. 191 97. 64 51 39 SCP3 28 19





Immunohistochemistry-Paraffin: SCP3/SYCP3 Antibody [NB300-232] - Analysis of SCP3 in human testis using DAB with hematoxylin counterstain.

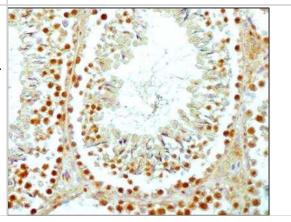


Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-232] - SCP3 Antibody [NB300-232] - SCP3 labeled in mouse pachytene preparation (red), using NB300-232 SCP3 antibody. CDK2 staining, near teleomeres, is also present (green).

Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-232] - Loss of lamin C2 has no effect on meiotic telomere attachment.. Chromosome spread preparations of pachytene-like lamin C2-/- spermatocytes showing that all telomeres are associated with SUN1. TeloFISH in co-localisation with SYCP3. Image collected and cropped by CiteAb from the following publication (https://dx.plos.org/10.1371/journal.pgen.1003261), licensed under a CC-BY license.



Immunohistochemistry-Paraffin: SCP3/SYCP3 Antibody [NB300-232] -IHC-P analysis of formalin fixed paraffin embedded tissue section of mouse testes using SCP3 antibody at 1:200 dilution. Specific staining may be seen in the spermatogonial cells and the primary spermatocytes.





Uhrf111: Stra8-cre

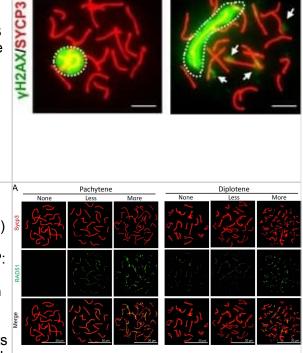
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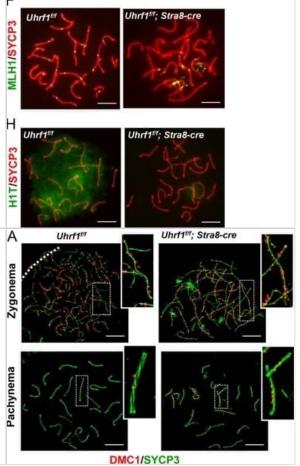
UHRF1 deficiency resulted in impaired meiotic recombination & defective pachynema.a Double immunofluorescence of SYCP3 (green) & DMC1 (red) in testicular spread preparations. b, c The number of DMC1 foci in zygotene stage (b) & pachytene stage (c). d Immunostaining for SYCP3 (red) &  $\gamma$ H2AX (green). e The percentage of abnormal  $\gamma$ H2AX foci in the pachytene stage. f Immunostaining for SYCP3 (red) & MLH1 (green). g The number of MLH1 foci in pachynema. h Immunostaining for SYCP3 (red) & H1t (green). i The percentage of spermatocytes with H1T staining. \*\*\*p ≤ 0.001; \*p ≤ 0.05. Scale bar, 5 µm in a, d, f, h. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/32081844), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - Effects of MLT on HR in DEHP-exposed fetal oocytes. (A)The immunofluorescence with Sycp3 (red) & RAD51 (green) in fetal oocytes. (B) The percentages of more, less & none staining of RAD51 in all MPI stages (control:  $32.48 \pm 2.54\%$ ,  $67.52 \pm 2.54\%$ ; DEHP:  $59.83 \pm 7.44\%$ ,  $40.17 \pm 7.44\%$ ; DEHP+MLT:  $33.13 \pm 2.79\%$ ,  $66.87 \pm$ 2.79%). (C) The percentages of more, less & none staining of RAD51 in pachytene & diplotene oocytes (control:  $36.27 \pm 8.02\%$ ,  $19.09 \pm 1.03\%$ ,  $44.64 \pm 8.08\%$ ; DEHP:  $44.24 \pm 2.98\%$ ,  $22.50 \pm 4.28\%$ ,  $33.26 \pm 1.30\%$ ; DEHP+MLT:  $32.97 \pm 4.27\%$ ,  $22.00 \pm 1.03\%$ ,  $45.03 \pm 4.76\%$ ). The results were presented as mean  $\pm$  SEM. \*P < 0.05, \*\* P < 0.01. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/30591620), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - UHRF1 deficiency resulted in impaired meiotic recombination & defective pachynema.a Double immunofluorescence of SYCP3 (green) & DMC1 (red) in testicular spread preparations. b, c The number of DMC1 foci in zygotene stage (b) & pachytene stage (c). d Immunostaining for SYCP3 (red) &  $\gamma$ H2AX (green). e The percentage of abnormal  $\gamma$ H2AX foci in the pachytene stage. f Immunostaining for SYCP3 (red) & MLH1 (green). g The number of MLH1 foci in pachynema. h Immunostaining for SYCP3 (red) & H1t (green). i The percentage of spermatocytes with H1T staining. \*\*\*p ≤ 0.001; \*p ≤ 0.05. Scale bar, 5 µm in a, d, f, h. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/32081844), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - UHRF1 deficiency resulted in impaired meiotic recombination & defective pachynema.a Double immunofluorescence of SYCP3 (green) & DMC1 (red) in testicular spread preparations. b, c The number of DMC1 foci in zygotene stage (b) & pachytene stage (c). d Immunostaining for SYCP3 (red) &  $\gamma$ H2AX (green). e The percentage of abnormal  $\gamma$ H2AX foci in the pachytene stage. f Immunostaining for SYCP3 (red) & MLH1 (green). g The number of MLH1 foci in pachynema. h Immunostaining for SYCP3 (red) & H1t (green). i The percentage of spermatocytes with H1T staining. \*\*\*p < 0.001; \*p < 0.05. Scale bar, 5 µm in a, d, f, h. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/32081844), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



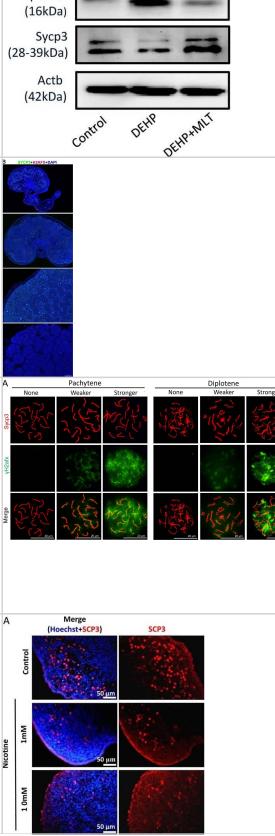




Western Blot: SCP3/SYCP3 Antibody - BSA Free [NB300-232] - Effects B (n=3) of MLT on meiotic progression & DSBs in DEHP-exposed fetal ovaries. (A) Morphology of 12.5 dpc ovaries cultured for 6 days in control, DEHP yH2afx & DEHP+MLT group in vitro. (B) Western blot analyses of the expression (16kDa) of Sycp3 & yH2afx protein in control, DEHP, & DEHP+MLT groups. (C) Relative expression level of genes Sycp3 & Trp53 in control, DEHP & Sycp3 DEHP+MLT groups. The results were presented as mean ± SEM. \*P < (28-39kDa) 0.05, \*\* P < 0.01. Image collected & cropped by CiteAb from the Actb following publication (https://pubmed.ncbi.nlm.nih.gov/30591620), (42kDa) licensed under a CC-BY license. Not internally tested by Novus control Biologicals. Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - Dynamic expression of germ cell markers in human fetal testes. (A) Histological sections of human testes at W10, W12, W17 & W22, immunostained for the early germ cell marker (nuclear) POU5F1 (red) & late germ cell marker (cytoplasmic) DDX4 (green). Inserts are magnifications of the dotted boxes. (B) Histological sections of human ovaries at W10, W12, W17 & W22, immunostained for the meiotic markers H2AFX (red) & SYCP3 (green). Note the presence of autofluorescent red blood cells. Scale bars are 200 µm & in the magnified inserts 20 µm. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/26834021), licensed under a CC-BY license. Not internally tested by Novus Biologicals. Pachytene Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - The effect of MLT on the formation of DSBs in fetal oocytes. (A)The immunofluorescence with Sycp3 (red) & yH2afx (green) in fetal oocytes. (B) The percentages of stronger, none & weaker yH2afx signal in oocyte in all MPI stages (control: 77.60 ± 5.04%, 22.40 ± 5.04%; DEHP: 87.24 ± 4.02%, 12.46 ± 4.02%; DEHP+MLT: 56.03 ± 9.52%, 43.97 ± 9.52%). (C) The percentages of stronger, weaker & none staining of yH2afx in pachytene & diplotene oocytes (control: 62.13 ±

3.37%,  $35.92 \pm 2.99\%$ ,  $1.95 \pm 0.78\%$ ; DEHP:  $80.48 \pm 1.53\%$ ,  $17.41 \pm 1.94\%$ ,  $2.11 \pm 0.78\%$ ; DEHP+MLT:  $64.36 \pm 2.39\%$ ,  $35.64 \pm 2.39\%$ ,  $0.00 \pm 0.00\%$ ). The results were presented as mean  $\pm$  SEM. \*P < 0.05, \*\* P < 0.01. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/30591620), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - Dose-dependent decrease or increase of the number of meiotic germ cells (SCP3, MLH1) & of  $\gamma$ H2AX positive cell, respectively, in nicotine treated fetal ovaries cultured for 4 days. (A) Representative IF images of ovarian tissue sections for SCP3; (B) representative IF images of ovarian tissue sections for MLH1 &  $\gamma$ H2AX; (C) Relative percentage of SCP3 & MLH1 positive cells of ovaries cultured without (control) & with 1mM or 10mM nicotine. All experiments were repeated at least three times. (\*) & (\*\*) indicate significant (P < 0.05) & highly significant (P < 0.01) difference, respectively. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/30001218), licensed under a CC-BY license. Not internally tested by Novus Biologicals.





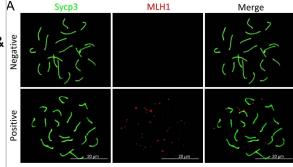
Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - Effects of MLT on mismatch repair in DEHPexposed fetal oocytes. (A)The immunofluorescence with Sycp3 (green) & MLH1 (red) in fetal oocytes. (B) The percentages of positive & negative MLH1 signal in oocytes (control: 23.63  $\pm$  0.55%, 76.37  $\pm$  0.55%; DEHP: 77.01  $\pm$  4.41%, 22.99  $\pm$  4.41%; DEHP+MLT: 55.04  $\pm$  17.04%, 43.87  $\pm$ 17.08%). (C & D) The amounts of the MLH1 positive foci in pachytene (control: 7.91  $\pm$  1.33; DEHP: 13.97  $\pm$  0.95; DEHP+MLT: 6.74  $\pm$  0.58) & diplotene (control: 12.11  $\pm$  1.74; DEHP: 13.18  $\pm$  1.16; DEHP+MLT: 7.58  $\pm$  1.07) oocytes, respectively. The results were presented as mean  $\pm$ SEM. \*P < 0.05, \*\* P < 0.01. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/30591620), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - Hyper-5hmC resulted from UHRF1 deletion.a Venn diagram depicting reduced transcripts associated with 5hmC downregulation. b 5hmC level of meiosis prophase I spermatocytes (16 dpp). c 5hmC densities of all chromosomes. d The distribution of 5hmC density on the genome of spermatocytes. e Venn diagram depicting 5hmC peaks in Uhrf1f/f;Stra8-cre & Uhrf1f/f spermatocytes. f 5hmC densities was shown in the proximal promoter, TSS, & gene body regions of the DEGs. g 5hmC densities in TSSs of the total refgenes with different RPKMs. h The percentage of RNA polymerase II staining in pachynema. i Double immunofluorescence of testicular spread preparations, SYCP3 (red) & RNA polymerase II (green). Scale bar, 5 µm in i. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/32081844), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

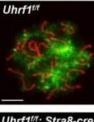
Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - UHRF1 deletion disrupted the meiotic progression & synaptonemal complex assembly.a Relative amounts of four spermatocyte populations (leptotene stage, zygotene stage, pachytene stage, & diplotene stage) during the prophase I in testes based on analyzing >600 spermatocytes in each stage. b, c The immunostaining of SYCP3 in the testicular sections (b) & surface-spread chromatin preparations of Uhrf1 deletion & control mice (c); d the percentage of spermatocytes with abnormal SYCP3 location. e Double immunofluorescence of testicular spread preparations of the adult mice, SYCP3 (green) & SYCP1 (red). f The percentage of spermatocytes with abnormal SYCP1 location. Lep leptotene, Zyg zygotene, Pac pachytene, Dip diplotene. Data are presented as mean  $\pm$  SEM of three mice. \*\*\*p  $\leq$ 0.001. Scale bar, 25 µm in b, 5 µm in c, e. Image collected & cropped by CiteAb from the following publication

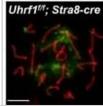
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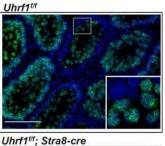


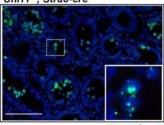






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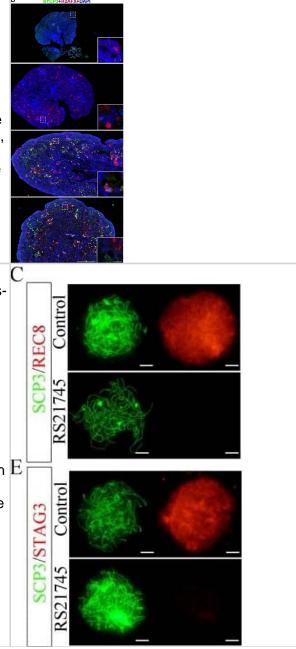


SYCP3/DAPI



Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - Dynamic expression of germ cell markers in human fetal ovaries. (A) Histological sections of human ovaries at W10.5, W14, W17 & W21.5, immunostained for the early germ cell marker (nuclear) POU5F1 (red) & late germ cell marker (cytoplasmic) DDX4 (green). In zone 1, most germ cells are POU5F1+DDX4-/low; in zone 2 & 3, most germ cells are DDX4+. Several germ cells in zone 3 have developed into primordial follicles. Inserts are magnifications of the dotted boxes. (B) Histological sections of human ovaries at W10.5, W14, W17 & W21.5, immunostained for the meiotic markers H2AFX (red) & SYCP3 (green). Inserts are magnifications of the dotted boxes. Note the presence of autofluorescent red blood cells. Scale bars are 200 µm & in the magnified inserts 20 µm. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/26834021), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - CYP51 regulates the expression of the meiosisspecific cohesin subunits REC8 & STAG3. (A,B) RS21745 treatment decreased REC8 & STAG3 expression. Ovaries at 14.5 dpc were cultured with 10 µM RS21745 for 3 days in vitro. (A) Expression of the cohesin subunits following RS21745 treatment by gRT-PCR. All gRT-PCR values were normalized to β-actin & were expressed as a relative ratio to the control; the means±s.e.m. of 3 values are shown. (B) Expression of REC8 & STAG3 following RS21745 treatment by western blotting GAPDH was used as internal reference. (C-F) Inhibition of CYP51 disturbed the distribution of both REC8 & STAG3 on the chromosomes. (C,E) In the control groups, REC8 & STAG3 expression was observed at high levels in zygotene oocytes. In the CYP51 inhibition  ${f E}$ groups, the cohesin signals were absent in zygotene cells. (D,F) The percentage of REC8-positive or STAG3-positive oocytes at the zygotene stage was recorded. Unidentified germ cells were not included in the analyses. Scale bars: 10 µm. The data are presented as the means ±s.e.m. of 3–9 ovaries per group. Asterisk (\*) denotes a statistically significant difference between the control & the treatment groups. \*\*\*P<0.001 (t-test). Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/30420384), licensed under a CC-BY license. Not internally tested by Novus Biologicals.





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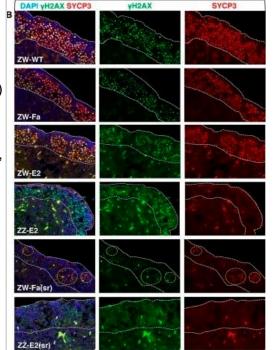
Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - Induction of meiosis is compromised in the cortical germ cells of D17 embryos subject to oestrogen level alterations. (A) D17 (HH43) left gonad sections immunostained for vH2AX (green) & P63 (red); nuclei are counterstained with DAPI (blue). (B) Sections from the D17 (HH43) left gonad shown in A immunostained for yH2AX (green) & SYCP3 (red). ZW-WT, ZW wild type; ZW-Fa, ZW treated with fadrozole from D7-7.5 (HH31); ZW-E2, ZW treated with β-oestradiol at D7-7.5; ZZ-E2, ZZ treated with  $\beta$ -oestradiol at D7-7.5; ZW-Fa(sr), ZW, partially sex-reversed gonad, treated with fadrozole at D4; ZZ-E2(sr), ZZ, partially sex-reversed gonad, treated with  $\beta$ -oestradiol at D4. All gonadal models have a cortical domain containing germ cells. In ZW-Fa ovary & ZW-E2 ovary, most cortical germ cells express SYCP3 like the ZW-WT control. In ZW-Fa(sr) ovotestis, very few germ cells express yH2AX & SYCP3 (orange dotted circled areas). In ZZ-E2(sr) ovotestis & ZZ-E2 testis overlain by a cortex, some cortical germ cells express vH2AX but none expresses SYCP3. White dotted line highlights the cortical domain borders. See Fig. S4 for the medullary structure of the ZZ-E2, ZW-Fa(sr) & ZZ-E2(sr) models. Image collected & cropped by CiteAb from the following publication (https://pubmed.ncbi.nlm.nih.gov/32001442), licensed under a CC-BY license. Not internally tested by Novus Biologicals.

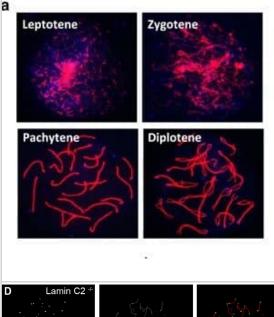
Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - Exposure to DEHP impairs meiotic progression of oocytes from pachytene to diplotene. (a) Immunolabeling of the oocyte chromosomes with anti-SYCP3 antibody (red) & Hoechst 33342 (blue). (b) Effect of DEHP on meiotic progression of oocytes throughout prophase I stages; percentage of each group is presented as mean±SD. Control: 36.27±0.80% pachytene & 54.99±0.66% diplotene; 10 µM & 100 µM DEHP 57.79±4.22% & 56.62±6.62% pachytene & 39.79±4.22% & 36.62±6.62% diplotene, respectively. (c) Representative WB showing the effect of DEHP on the expression of germ cell (DAZL) & meiotic (STRA8 & SCP3) specific proteins. (d) Effect of DEHP on the levels of mRNA in the ovarian tissues of germ cell (Mvh & Dazl), meiotic (Stra8, Rec8, Scp1 & Scp3). All experiments were repeated at least three times independently. (\* P<0.05; \*\* P<0.01) Image collected & cropped by CiteAb from the following publication

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Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody -BSA Free [NB300-232] - Loss of lamin C2 has no effect on meiotic telomere attachment.(A) 3D-preserved swab preparations showing wildtype (A) & knockout (A') spermatocytes simultaneously labelled with anti-TRF1 & SUN1 antibodies. As in the wildtype, in lamin C2-/spermatocytes virtual all telomeres appear to be attached to the NE as indicated by co-localisation of TRF1 & SUN1 signals. Scale bars 5 µm. (B) Quantifications of co-localised & non-co-localised TRF1/SUN1 signals (see A) revealed that ratios of co-localised to non-co-localised spots comparing wildtype & knockout spermatocytes show no significant difference (wildtype n=33; lamin C2-/- n=45; Pearson's Chi2 test pvalue: 0.799). (C,D) Chromosome spread preparations of pachytene-like lamin C2-/- spermatocytes showing that all telomeres are associated with SUN1. In (C) TeloFISH & in (D) anti-SUN1 staining in co-localisation with SYCP3. Scale bars 10 µm. Image collected & cropped by CiteAb from the following publication

(https://dx.plos.org/10.1371/journal.pgen.1003261), licensed under a CC-BY license. Not internally tested by Novus Biologicals.





100µm

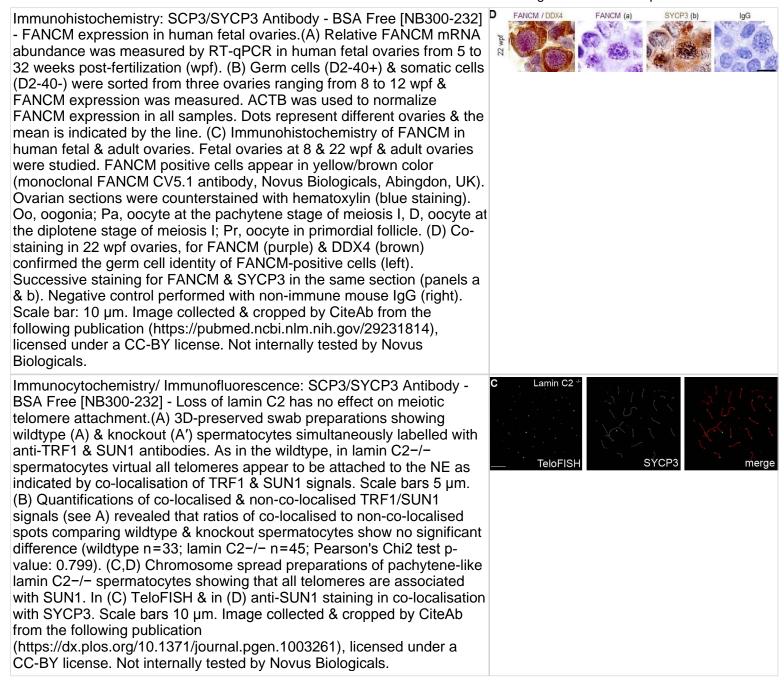


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#### **Publications**

Xiaowei Yan, Yanni Feng, Yanan Hao, Ruqing Zhong, Yue Jiang, Xiangfang Tang, Dongxin Lu, Hanhan Fang, Manjree Agarwal, Liang Chen, Yong Zhao, Hongfu Zhang, Henning Seedorf Gut-Testis Axis: Microbiota Prime Metabolome To Increase Sperm Quality in Young Type 2 Diabetes Microbiology Spectrum 2022-10-10 [PMID: 36214691]

Yong Zhao, Pengfei Zhang, Wei Ge, Yanni Feng, Lan Li, Zhongyi Sun, Hongfu Zhang, Wei Shen Alginate oligosaccharides improve germ cell development and testicular microenvironment to rescue busulfan disrupted spermatogenesis Theranostics 2020-01-01 [PMID: 32194870]

Hinch AG, Becker PW, Li T et al. The Configuration of RPA, RAD51, and DMC1 Binding in Meiosis Reveals the Nature of Critical Recombination Intermediates Mol. Cell 2020-06-23 [PMID: 32610038]

Yanan Hao, Yanni Feng, Xiaowei Yan, Liang Chen, Xiangping Ma, Xiangfang Tang, Ruqing Zhong, Zhongyi Sun, Manjree Agarwal, Hongfu Zhang, Yong Zhao, Jasna Kovac Gut Microbiota-Testis Axis: FMT Mitigates High-Fat Diet-Diminished Male Fertility via Improving Systemic and Testicular Metabolome Microbiology Spectrum 2022-04-21 [PMID: 35446112]

He J, Yan A, Chen B et al. 3D genome remodeling and homologous pairing during meiotic prophase of mouse oogenesis and spermatogenesis Developmental cell 2023-11-13 [PMID: 37963468]

Davies B, Zhang G, Moralli D et al. Characterization of meiotic recombination intermediates through gene knockouts in founder hybrid mice Genome research 2023-11-17 [PMID: 37977820] (IHC, Mouse)

Details:

Sample type: Testis

Wu D, Huang H, Chen T et al. The BRCA1/BARD1 complex recognizes pre-ribosomal RNA to facilitate homologous recombination Cell discovery 2023-10-03 [PMID: 37789001]

Larose H, Kent T, Ma Q et al. Regulation of meiotic progression by Sertoli-cell androgen signaling Molecular Biology of the Cell 2020-12-01 [PMID: 33026960] (Immunocytochemistry/ Immunofluorescence)

Younis N, Caldeira-Brant AL, Chu T et al. Human immature testicular tissue organ culture: a step towards fertility preservation and restoration Front Endocrinol (Lausanne) 2023-08-28 [PMID: 37701899] (Immunocytochemistry/ Immunofluorescence)

Islam KN, Ajao A, Venkataramani K et al. The RNA-binding protein Adad1 is necessary for germ cell maintenance and meiosis in zebrafish PLOS Genetics 2023-08-08 [PMID: 37552671]

Bailey A, Fuller M YTHDC2 serves a distinct late role in spermatocytes during germ cell differentiation bioRxiv 2023-01-23

Dil S, Ye J, Ma H et al. Cornichon protein CNIH4 is not essential for mice gametogenesis and fertility Developmental biology 2023-01-16 [PMID: 36657507] (ICC/IF, Mouse)

Details: Dilution used in ICC 1:100

More publications at http://www.novusbio.com/NB300-232

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#### **Procedures**

#### Immunocytochemistry/ Immunofluorescence Protocol for SCP3/SYCP3 Antibody (NB300-232)

Immunofluorescence Procedure

- 1. Freshly prepared slides are soaked in 1X ADB for 75 minutes.
- 2. Primary antibodies are added concurrently (SCP3 and CDK2).
- 3. The primary antibodies are incubated overnight in a hudid chamber (37 degrees Celcius).
- 4. The slides are washed for 40 minutes in 1X ADB.
- 5. The slides are detected with the appropriate secondary antibodies (RDAR for SCP1 and FDAM for CDK2).
- 6. The slides are incubated for 4 hours in a humid chamber (37 degrees Celcius).
- 7. The slides are washed for 20 minutes in 1X ADB, followed by 3 washes, 10 minutes each, in 1X PBS.
- 8. The slides are counterstained with DAPI.
- 9. Images are captured after allowing the slides to remain in the dark overnight at RT.
- 14. Dehydrate sections.
- 15. Mount coverslips.

\*The above information is only intended as a guide. The researcher should determine what protocol best meets their needs. Please follow safe laboratory procedures.

Immunohistochemistry-Paraffin Protocol for SCP3/SYCP3 Antibody (NB300-232) Immunohistochemistry-Paraffin Embedded Sections

Antigen Unmasking:

Bring slides to a boil in 10 mM sodium citrate buffer (pH 6.0) then maintain at a sub-boiling temperature for 10 minutes. Cool slides on bench-top for 30 minutes (keep slides in the sodium citrate buffer all the time).

Staining:

- 1. Wash sections in deionized water three times for 5 minutes each.
- 2. Wash sections in PBS for 5 minutes.
- 3. Block each section with 100-400 ul blocking solution (1% BSA in PBS) for 1 hour at room temperature.
- 4. Remove blocking solution and add 100-400 ul diluted primary antibody. Incubate overnight at 4 C.
- 5. Remove antibody solution and wash sections in wash buffer three times for 5 minutes each.
- 6. Add 100-400 ul HRP polymer conjugated secondary antibody. Incubate 30 minutes at room temperature.
- 7. Wash sections three times in wash buffer for 5 minutes each.
- 8. Add 100-400 ul DAB substrate to each section and monitor staining closely.
- 9. As soon as the sections develop, immerse slides in deionized water.
- 10. Counterstain sections in hematoxylin.
- 11. Wash sections in deionized water two times for 5 minutes each.
- 12. Dehydrate sections.
- 13. Mount coverslips.



# Western Blot Protocol for SCP3/SYCP3 Antibody (NB300-232)

Western Blot Protocol

1. Perform SDS-PAGE on samples to be analyzed, loading 10-25 ug of total protein per lane.

2. Transfer proteins to PVDF membrane according to the instructions provided by the manufacturer of the membrane and transfer apparatus.

3. Stain the membrane with Ponceau S (or similar product) to assess transfer success, and mark molecular weight standards where appropriate.

4. Rinse the blot TBS -0.05% Tween 20 (TBST).

5. Block the membrane in 5% Non-fat milk in TBST (blocking buffer) for at least 1 hour.

6. Wash the membrane in TBST three times for 10 minutes each.

7. Dilute primary antibody in blocking buffer and incubate overnight at 4C with gentle rocking.

8. Wash the membrane in TBST three times for 10 minutes each.

9. Incubate the membrane in diluted HRP conjugated secondary antibody in blocking buffer (as per manufacturer's instructions) for 1 hour at room temperature.

10. Wash the blot in TBST three times for 10 minutes each (this step can be repeated as required to reduce background).

11. Apply the detection reagent of choice in accordance with the manufacturers instructions.

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## Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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