

# Product Datasheet

## SCP3/SYCP3 Antibody - BSA Free NB300-231

Unit Size: 0.1 ml

Store at 4C. Do not freeze.

[www.novusbio.com](http://www.novusbio.com)



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**NB300-231**

SCP3/SYCP3 Antibody - BSA Free

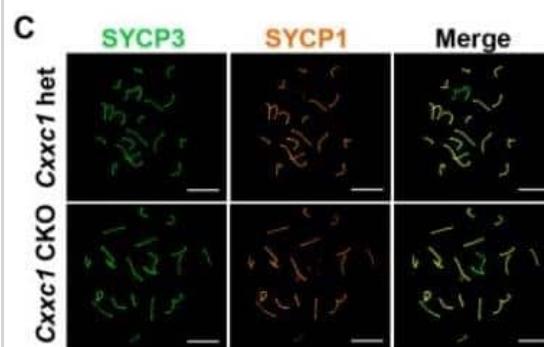
Product Information	
Unit Size	0.1 ml
Concentration	1 mg/ml
Storage	Store at 4C. Do not freeze.
Clonality	Polyclonal
Preservative	0.09% Sodium Azide
Isotype	IgG
Purity	Immunogen affinity purified
Buffer	Tris-Citrate/Phosphate (pH 7.0 - 8.0)

Product Description	
Host	Rabbit
Gene ID	50511
Gene Symbol	SYCP3
Species	Human, Mouse, Chicken
Reactivity Notes	Human reactivity reported in scientific literature (PMID: 21079786). Chicken reactivity reported in scientific literature (PMID: 23840850).
Immunogen	A synthetic peptide made to the C-terminal region of the mouse SCP3 protein. [UniProt# P70281]

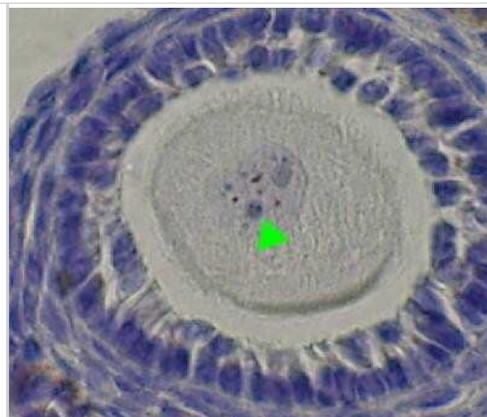
Product Application Details	
Applications	Western Blot, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, Immunoprecipitation
Recommended Dilutions	Western Blot, Immunohistochemistry 1:750, Immunocytochemistry/ Immunofluorescence 1:100 - 1:750, Immunoprecipitation reported in scientific literature (PMID 27932493), Immunohistochemistry-Paraffin 1:750, Immunohistochemistry-Frozen

**Images**

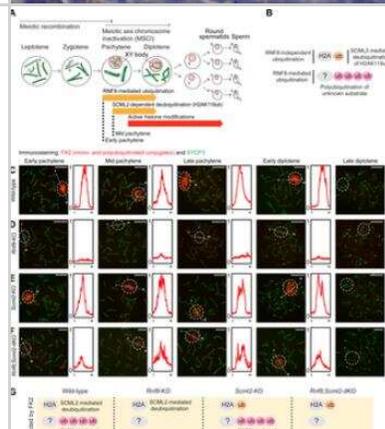
Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - Immunostaining of SCP3/SYCP3 (NB300-231) and SCP1 (NB300-229) on adult Cxxc1 het and CKO chromosome spreads. Green, SCP3/SYCP3; orange, SCP1. Scale bar, 10 um. Image collected and cropped by CiteAb from the following publication ([//pubmed.ncbi.nlm.nih.gov/30365547/](https://pubmed.ncbi.nlm.nih.gov/30365547/)) licensed under a CC-BY license.



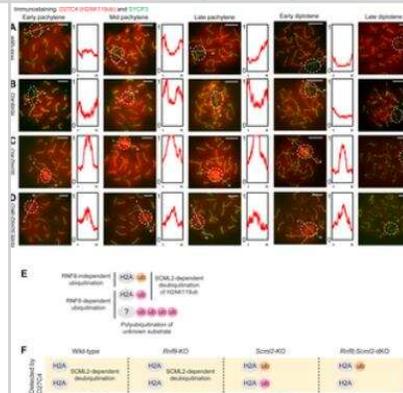
Immunohistochemistry: SCP3/SYCP3 Antibody [NB300-231] - Punctate staining of murine SCP3 in mouse ovary using NB300-231.



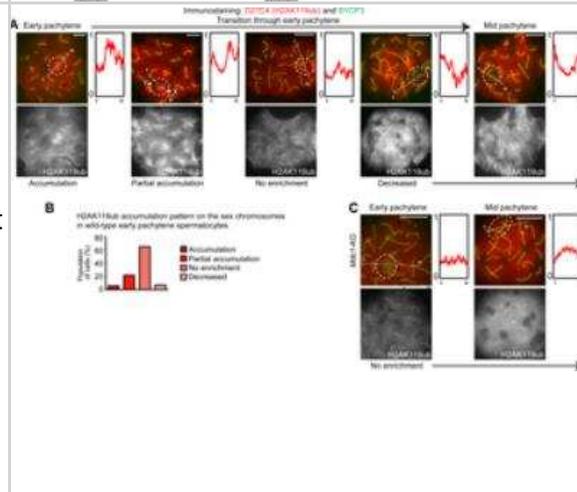
Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - Distinct forms of ubiquitin regulation by RNF8 and SCML2. (C-F) Immunostaining on meiotic chromosome spreads of FK2 and SYCP3 antibodies. Dotted circles are sex chromosomes and Scale bar are 10 micrometers. At least 30 spermatocytes from each substage are shown from at least 3 independent mice per mouse model. Immunostaining intensity is qualified by densitometry across the indicated path and plotted in a relative intensity of 0-1 (normalized among stages). Citation: Adams SR, Maezawa S, Alavattam KG, Abe H, Sakashita A, Shroder M, et al. (2018) RNF8 and SCML2 cooperate to regulate ubiquitination and H3K27 acetylation for escape gene activation on the sex chromosomes. PLoS Genet 14(2): e1007233. <https://doi.org/10.1371/journal.pgen.1007233>



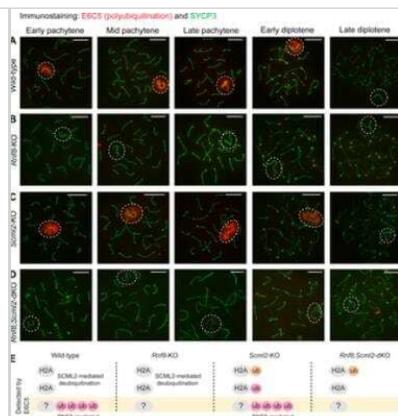
Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - (A-D) Immunostaining of D27C4 and SYCP3 on meiotic chromosome spreads. Dotted circles indicate sex chromosomes and scale bar is 10 micrometers. At least 30 spermatocytes from each substage are shown from at least 3 independent mice per mouse model. Immunostaining intensity is qualified by densitometry across the indicated path and plotted in a relative intensity of 0-1 (normalized among stages). Citation: Adams SR, Maezawa S, Alavattam KG, Abe H, Sakashita A, Shroder M, et al. (2018) RNF8 and SCML2 cooperate to regulate ubiquitination and H3K27 acetylation for escape gene activation on the sex chromosomes. PLoS Genet 14(2): e1007233. <https://doi.org/10.1371/journal.pgen.1007233>



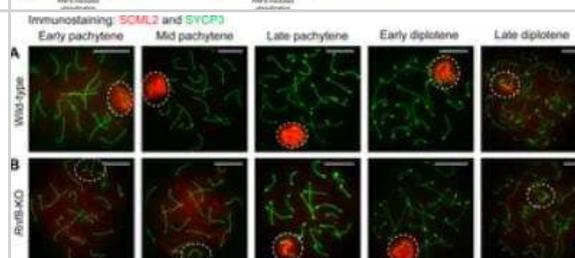
Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - (A) Immunostaining of D27C4 and SYCP3 on wild-type meiotic chromosome spreads. Representative images are shown for each category through the early pachytene stage for spermatocytes from 9 independent mice. (C) Immunostaining of SYCP3 and D27C4 on Mdc1-KO meiotic chromosome spreads. Representative images are shown through the early pachytene stage for spermatocytes from 3 independent Mdc1-KO mice. (A and C) Dotted circles indicate sex chromosomes and scale bar is 10 micrometers. Immunostaining intensity is qualified by densitometry across the indicated path and plotted in a relative intensity of 0-1 (normalized among stages). Citation: Adams SR, Maezawa S, Alavattam KG, Abe H, Sakashita A, Shroder M, et al. (2018) RNF8 and SCML2 cooperate to regulate ubiquitination and H3K27 acetylation for escape gene activation on the sex chromosomes. PLoS Genet 14(2): e1007233. <https://doi.org/10.1371/journal.pgen.1007233>



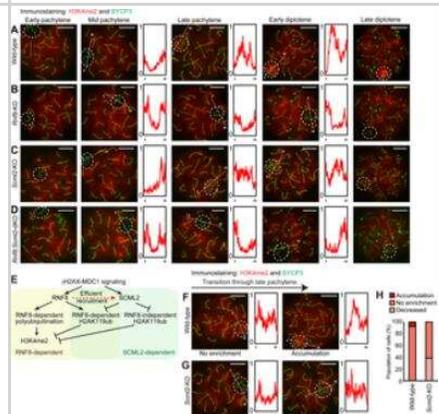
Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - (A-D) Immunostaining of E6C5 and SYCP3 on meiotic chromosome spreads. Dotted circles indicate sex chromosomes and scale bar is 10 micrometers. At least 30 spermatocytes from each substage are shown from at least 3 independent mice per mouse model. Citation: Adams SR, Maezawa S, Alavattam KG, Abe H, Sakashita A, Shroder M, et al. (2018) RNF8 and SCML2 cooperate to regulate ubiquitination and H3K27 acetylation for escape gene activation on the sex chromosomes. *PLoS Genet* 14(2): e1007233. <https://doi.org/10.1371/journal.pgen.1007233>



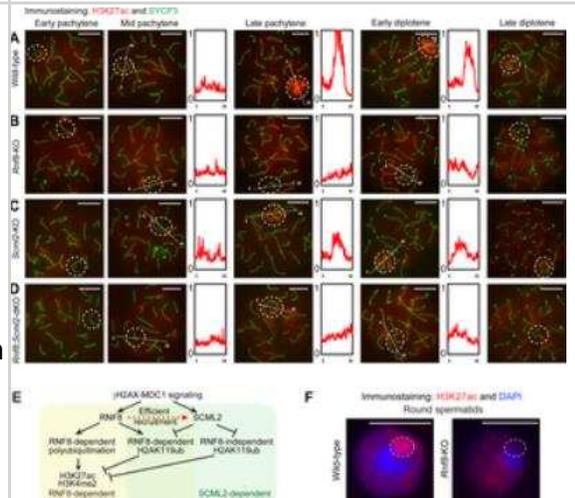
Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - (A, B) Immunostaining of SCML2 and SYCP3 on meiotic chromosome spreads. Dotted circles indicate sex chromosomes and scale bar is 10 micrometers. At least 30 spermatocytes from each substage are shown from at least 3 independent mice per mouse model. Citation: Adams SR, Maezawa S, Alavattam KG, Abe H, Sakashita A, Shroder M, et al. (2018) RNF8 and SCML2 cooperate to regulate ubiquitination and H3K27 acetylation for escape gene activation on the sex chromosomes. *PLoS Genet* 14(2): e1007233. <https://doi.org/10.1371/journal.pgen.1007233>



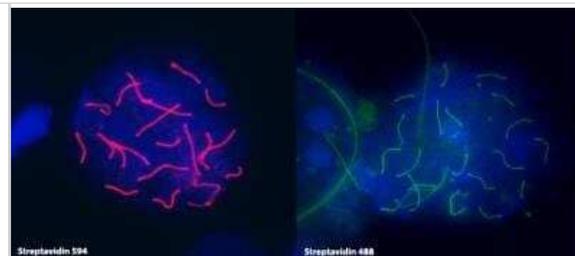
Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - (A-D) Immunostaining of H3K4me2 and SYCP3 on meiotic chromosome spreads. Dotted circles: sex chromosomes. (E) Regulation of H3K4me2. (F, G) Immunostaining of H3K4me2 and SYCP3 on wild-type and Scml2-KO meiotic chromosome spreads. All: Dotted circles indicate sex chromosomes and scale bar is 10 micrometers. Immunostaining intensity is qualified by densitometry across the indicated path and plotted in a relative intensity of 0-1 (normalized among stages). Citation: Adams SR, Maezawa S, Alavattam KG, Abe H, Sakashita A, Shroder M, et al. (2018) RNF8 and SCML2 cooperate to regulate ubiquitination and H3K27 acetylation for escape gene activation on the sex chromosomes. *PLoS Genet* 14(2): e1007233. <https://doi.org/10.1371/journal.pgen.1007233>



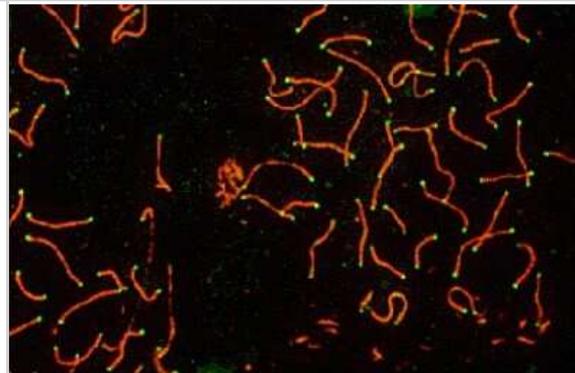
Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - (A-D) Immunostaining of H3K27ac and SYCP3 on meiotic chromosome spreads. At least 30 spermatocytes from each substage are shown from at least 3 independent mice per mouse model. Immunostaining intensity is qualified by densitometry across the indicated path and plotted in a relative intensity of 0-1 (normalized among stages). (E) Model of the regulation of H3K27ac. (F) Immunostaining of SYCP3 and H3K27ac on wild-type and Rnf8-KO round spermatids. At least 30 round spermatids shown from at least 3 independent mice. All: Dotted circles indicate sex chromosomes and scale bar is 10 micrometers. Citation: Adams SR, Maezawa S, Alavattam KG, Abe H, Sakashita A, Shroder M, et al. (2018) RNF8 and SCML2 cooperate to regulate ubiquitination and H3K27 acetylation for escape gene activation on the sex chromosomes. *PLoS Genet* 14(2): e1007233. <https://doi.org/10.1371/journal.pgen.1007233>



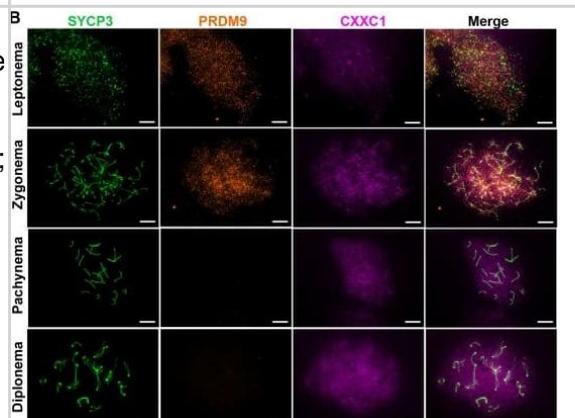
Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - Mouse spermatocytes stained with biotin-conjugated anti-SYCP3 detected with streptavidin-Alexa Fluor 594 (left panel), streptavidin Alexa Fluor 488 (right panel). Image from verified customer review.



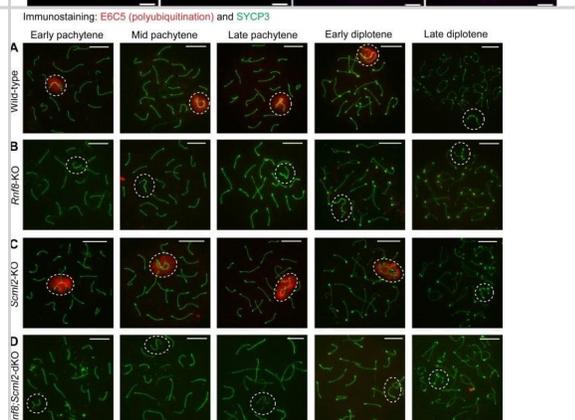
Immunocytochemistry/Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - SCP3 labeled in mouse pachytene preparation (red), using NB 300-231. CDK2 staining, near telomeres, is also present (green).



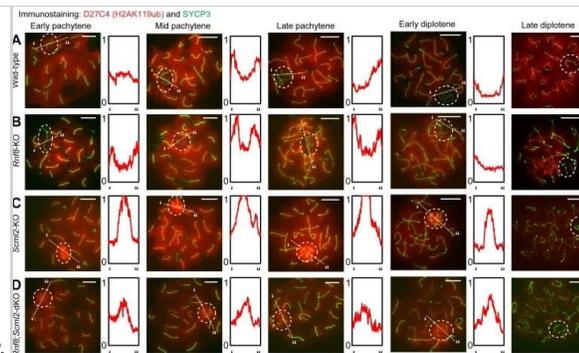
Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - CXXC1 is expressed in the spermatocytes in the presence or absence of PRDM9. (A) Immunofluorescence staining for CXXC1 & PRDM9 in adult B6, 14-dpp B6 & 14-dpp Prdm9<sup>-/-</sup> seminiferous tubule cross sections. Green, CXXC1; red, PRDM9; blue, DAPI. SC, Sertoli cell; Z, zygonema; P, pachynema; RS, round spermatid. Scale bars, first 4 columns: 50  $\mu$ m, last column: 10  $\mu$ m. (B) Immunofluorescence staining for CXXC1 & PRDM9 on chromosome spreads from adult B6. Green, SYCP3; orange, PRDM9; magenta, CXXC1. Scale bars, 10  $\mu$ m. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/30365547>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



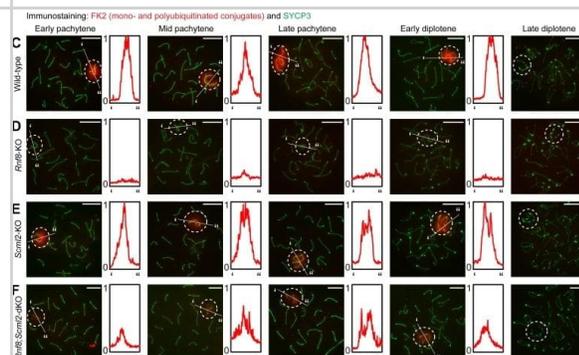
Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - RNF8 is required for polyubiquitination of the sex chromosomes: Immunostaining with E6C5 antibody, which recognizes polyubiquitination. (A-D) Immunostaining of SYCP3 & E6C5 on meiotic chromosome spreads. Dotted circles: sex chromosomes. Scale bar: 10  $\mu$ m. Representative images are shown for at least 30 spermatocytes from each substage, from at least 3 independent mice per mouse model. (E) Schematic of ubiquitin targets recognized by the E6C5 antibody in each mouse model. Image collected & cropped by CiteAb from the following publication (<https://dx.plos.org/10.1371/journal.pgen.1007233>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



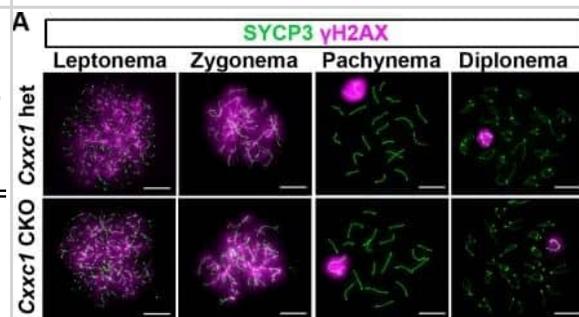
**Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - RNF8-dependent H2AK119ub is removed by SCML2:** Immunostaining with D27C4 antibody, which recognizes H2AK119ub. (A-D) Immunostaining of SYCP3 & D27C4 (H2AK119ub) on meiotic chromosome spreads. Dotted circles: sex chromosomes. Scale bar: 10  $\mu$ m. Representative images are shown for at least 30 spermatocytes from each substage, from at least 3 independent mice per mouse model. The intensity of immunostaining is quantified by densitometry across the indicated path (' to') & plotted in a relative intensity range of 0–1, which is normalized among all images in this figure, Fig 3 & S2 Fig. (E) Updated model of distinct forms of regulation of ubiquitination by RNF8 & SCML2, including RNF8-dependent H2AK119ub, which is removed by SCML2 based on data in this figure. (F) Schematic of ubiquitin targets recognized by the D27C4 (H2AK119ub) antibody in each mouse model. Image collected & cropped by CiteAb from the following publication (<https://dx.plos.org/10.1371/journal.pgen.1007233>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



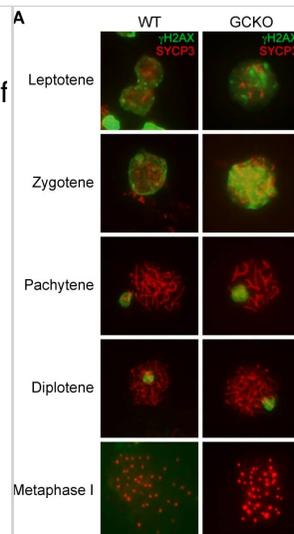
**Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - Distinct forms of ubiquitin regulation by RNF8 & SCML2:** Immunostaining with FK2 antibody, which recognizes both mono- & polyubiquitinated conjugates. (A) Schematic of spermatogenesis. (B) Model of distinct forms of regulation of ubiquitination by RNF8 & SCML2. (C-F) Immunostaining of SYCP3 & FK2 on meiotic chromosome spreads. Dotted circles: sex chromosomes. Scale bar: 10  $\mu$ m. Representative images are shown for at least 30 spermatocytes from each substage, from at least 3 independent mice per mouse model. The intensity of immunostaining is quantified by densitometry across the indicated path (' to') & plotted in a relative intensity range of 0–1, which is normalized among the samples at the same stage. (G) Schematic of ubiquitin targets recognized by the FK2 antibody in each mouse model. A pink circle denotes RNF8-dependent ubiquitination & an orange circle denotes events mediated by a different E3 ubiquitin ligase. Image collected & cropped by CiteAb from the following publication (<https://dx.plos.org/10.1371/journal.pgen.1007233>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



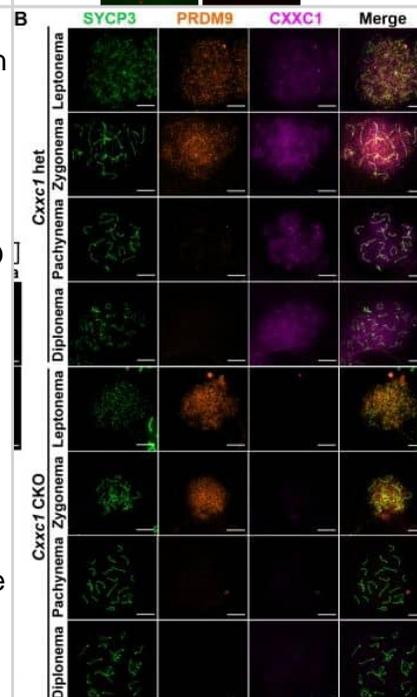
**Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - No major meiotic DSB repair or chromosome synapsis defects are observed in Cxxc1 CKO testis.** (A) Immunostaining of SYCP3 &  $\gamma$ H2AX on adult Cxxc1 het & CKO chromosome spreads. Green, SYCP3; magenta,  $\gamma$ H2AX. Scale bar, 10  $\mu$ m. (B) Spermatocyte stage proportion in adult Cxxc1 het (n = 1,062 from two individuals) & CKO (n = 1,105 from two individuals) spermatocytes based on SYCP3/SYCP1/ $\gamma$ H2AX staining. p = 0.7 by Chi-square test. (C) Immunostaining of SYCP3 & SYCP1 on adult Cxxc1 het & CKO chromosome spreads. Green, SYCP3; orange, SYCP1. Scale bar, 10  $\mu$ m. (D) Crossover number measured by MLH1 staining on chromosome spreads of adult Cxxc1 het & CKO spermatocytes. Left, magenta, SYCP3; green, MLH1. Scale bar, 10  $\mu$ m. Right, number of MLH1 foci per late pachynema in Cxxc1 het (n = 32 from two individuals) & CKO (n = 33 from two individuals). Bars represent mean  $\pm$  SD. p = 0.4 by Student's t-test. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/30365547>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



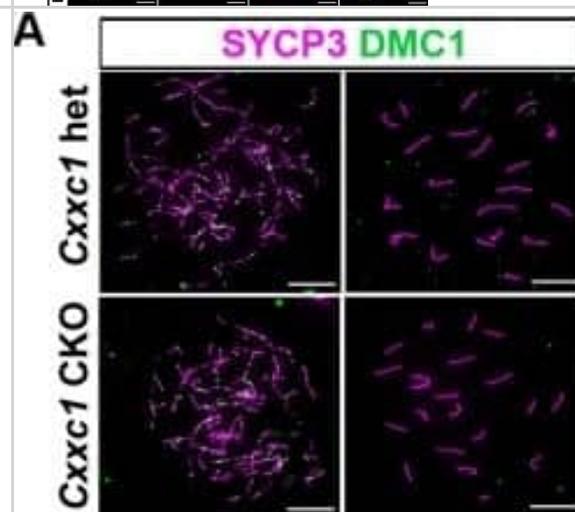
Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - Meiotic spreads show deletion of Dicer1 disrupts progression of meiosis I. (A) It was possible to identify the 5 sub-stages of meiosis I by combining the reactivity patterns of two antibodies against synaptonemal complex protein 3 (SYCP3) &  $\gamma$ H2AX. SYCP3 is a protein essential for synapsis of homologous chromosomes &  $\gamma$ H2AX localizes to double-strand breaks & XY bodies during meiosis. A total of 200 spreads were counted in testis cell preparations from WT & GCKO mice at P22. (B) Chi square analysis shows GCKO testes contained significantly higher numbers of germ cell spreads at the leptotene & zygotene stages of meiosis I & fewer spreads at pachytene, diplotene & metaphase I stages ( $P < 0.001$ ), suggesting that the loss of Dicer1 lead to disruptions in progression through meiosis I. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/23056286>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - PRDM9 expression & catalytic function are not impaired in Cxxc1 CKO. (A) Immunostaining of PRDM9 shows unchanged pattern in adult CKO seminiferous tubules compared to the heterozygous control. Red, PRDM9; grey, DAPI. Scale bar, 20  $\mu$ m. (B) PRDM9 & SYCP3 expression patterns are not changed in CXXC1 CKO chromosome spreads compared to the heterozygous control. Co-immunostaining of CXXC1 & PRDM9 on chromosome spreads from adult Cxxc1 het & CKO mice. Green, SYCP3; orange, PRDM9; magenta, CXXC1. First 4 rows, het control; last 4 rows, Cxxc1 CKO. Scale bar, 10  $\mu$ m. (C) Meiosis progression occurs normally in Cxxc1 CKO testes. Immunostaining of H3K4me3 in adult Cxxc1 het & CKO chromosome spreads. Green, SYCP3; magenta, H3K4me3. Scale bar, 10  $\mu$ m. (D) Testis-specific gene expression is not changed in Cxxc1 CKO testes. H3K4me3 ChIP-qPCR with chromatin isolated from Cxxc1 het & CKO mice. Promoter regions from Actinb & Sycp3, Dom2 hotspots Pbx1 & Fcgr4 were amplified. Cst hotspot Hlx1 was used as a negative control. Bars present mean  $\pm$  SD of three biological replicates. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/30365547>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Immunocytochemistry/ Immunofluorescence: SCP3/SYCP3 Antibody [NB300-231] - DSB number is not affected in Cxxc1 CKO. The DSB number was determined by three markers reflecting different stages of their processing. (A) DMC1 staining on Cxxc1 control & CKO chromosome spread. Bottom panels, distribution plot of DMC1 foci in early zygotene ( $n = 22$  in het,  $n = 34$  in CKO), late zygotene ( $n = 29$  in het,  $n = 36$  in CKO) & pachytene ( $n = 36$  in het,  $n = 26$  in CKO) spermatocytes. (B) RAD51 staining on Cxxc1 control & CKO chromosome spread. Bottom panels, distribution plot of RAD51 foci in early zygotene ( $n = 22$  in het,  $n = 13$  in CKO), late zygotene ( $n = 24$  in het,  $n = 11$  in CKO) & pachytene ( $n = 52$  in het,  $n = 36$  in CKO) spermatocytes. (C) RPA staining on Cxxc1 control & CKO chromosome spread. Bottom panels, distribution plot of RPA foci in early zygotene ( $n = 35$  in het,  $n = 43$  in CKO), late zygotene ( $n = 33$  in het,  $n = 28$  in CKO) & pachytene ( $n = 89$  in het,  $n = 92$  in CKO) spermatocytes. For A-C, two individuals per genotype were measured. Bars represent mean  $\pm$  SD. Scale bar, 10  $\mu$ m. Image collected & cropped by CiteAb from the following publication (<https://pubmed.ncbi.nlm.nih.gov/30365547>), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



## Publications

Jessica Hopkins, Grace Hwang, Justin Jacob, Nicklas Sapp, Rick Bedigian, Kazuhiro Oka, Paul Overbeek, Steve Murray, Philip W. Jordan, Neil Hunter Meiosis-Specific Cohesin Component, Stag3 Is Essential for Maintaining Centromere Chromatid Cohesion, and Required for DNA Repair and Synapsis between Homologous Chromosomes PLoS Genetics 2014-07-01 [PMID: 24992337]

Beth L Dumont, Daniel M Gatti, Mallory A Ballinger, Dana Lin, Megan Phifer-Rixey, Michael J Sheehan, Taichi A Suzuki, Lydia K Wooldridge, Hilda Opoku Frempong, Raman Akinyanju Lawal, Gary A Churchill, Cathleen Lutz, Nadia Rosenthal, Jacqueline K White, Michael W Nachman Into the Wild: A novel wild-derived inbred strain resource expands the genomic and phenotypic diversity of laboratory mouse models. PLoS genetics 2024-04-23 [PMID: 38598567]

Hui Ma, Tao Li, Xuefeng Xie, Long Jiang, Jingwei Ye, Chenjia Gong, Hanwei Jiang, Suixing Fan, Huan Zhang, Baolu Shi, Beibei Zhang, Xiaohua Jiang, Yang Li, Jianteng Zhou, Jianze Xu, Xingxia Zhang, Xiaoning Hou, Hao Yin, Yuanwei Zhang, Qinghua Shi RAD51AP2 is required for efficient meiotic recombination between X and Y chromosomes Science Advances 2022-01-01 [PMID: 35020426]

Tian H, Billings T, Walker M et al. EWSR1 affects PRDM9-dependent histone 3 methylation and provides a link between recombination hotspots and the chromosome axis Mol Biol Cell 2020-11-11 [PMID: 33175657]

Li, L;Krasnykov, K;Homolka, D;Gos, P;Mendel, M;Fish, RJ;Pandey, RR;Pillai, RS; The XRN1-regulated RNA helicase activity of YTHDC2 ensures mouse fertility independently of m6A recognition Molecular cell [PMID: 35305312]

Lei WL, Li YY, Meng TG Et al. Specific deletion of protein phosphatase 6 catalytic subunit in Sertoli cells leads to disruption of spermatogenesis Cell death & disease 2021-09-27 [PMID: 34580275]

Tian H, Petkov PM Mouse EWSR1 is critical for spermatid post-meiotic transcription and spermiogenesis Development (Cambridge, England) 2021-05-21 [PMID: 34100066]

Zhang M, Liu L, Cao X et al. Efficiently accumulating germ-like stem cells from mouse postnatal ovary by in situ tissue culture Dev. Growth Differ. 2020-03-18 [PMID: 32189336] (IF/IHC, Mouse)

Zhang R, Ma J, Avery JT et al. GLI1 Inhibitor SRI-38832 Attenuates Chemotherapeutic Resistance by Downregulating NBS1 Transcription in BRAFV600E Colorectal Cancer Front Oncol 2020-02-28 [PMID: 32185127] (ICC/IF, Human)

Lei WL, Han F, Hu MW, et al. Protein phosphatase 6 is a key factor regulating spermatogenesis Cell Death Differ. 2019-12-09 [PMID: 31819157] (ICC/IF, Mouse)

Tu Z, Mu X, Chen X, et al. Dibutyl phthalate exposure disrupts the progression of meiotic prophase I by interfering with homologous recombination in fetal mouse oocytes Environ. Pollut. 2019-05-24 [PMID: 31158667] (ICC/IF, Mouse)

Bhattacharyya T, Walker M, Powers NR et al. Prdm9 and Meiotic Cohesin Proteins Cooperatively Promote DNA Double-Strand Break Formation in Mammalian Spermatocytes Curr. Biol. 2019-03-18 [PMID: 30853435] (ICC/IF, Mouse)

More publications at <http://www.novusbio.com/NB300-231>



## Procedures

### Protocol specific for SCP3 Antibody (NB300-231)

#### 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 humid chamber (37 degrees Celsius).
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 Celsius).
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.





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### **Products Related to NB300-231**

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NB300-231PEP	SCP3/SYCP3 Antibody Blocking Peptide
HAF008	Goat anti-Rabbit IgG Secondary Antibody [HRP]
NB7160	Goat anti-Rabbit IgG (H+L) Secondary Antibody [HRP]
NBP2-24891	Rabbit IgG Isotype Control

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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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