

Table S1A. Antibodies used for WB, IF, IP and ChIP: application, source and working dilution

Antibodies	Application	Species	Source
H3K4me3	ChIP	Rabbit	Merck Millipore 07473
FUS (4H11)	IP WB 1:1000 IF 1:300	Mouse	Santa Cruz sc-47711
SYCP3	IF 1:300	Rabbit	Novus Biologicals NB 300-231
SYCP3	WB 1:1000	Guinea Pig	Bernard de Massy ¹
SYCP3	IF 1:300	Rat	Mary Ann Handel ²
SPO11	IP WB 1:1000	Mouse	Scott Keeney ³
IGg2A	IP	Mouse	Sigma-Aldrich M5409
TUBULIN	WB 1:1000	Mouse	Sigma-Aldrich T4026
PRDM9	WB 1:500 IF 1:100	Rabbit	Corinne Grey ¹
REC114	WB 1:1000	Rabbit	Attila Toth ⁴
CLATHRIN	WB 1:1000	Mouse	BD Bioscience 610500

WB: western blotting; IF: immunofluorescence; IP immunoprecipitation;

ChIP: Chromatin immunoprecipitation

1. Diagouraga B, Clement JAJ, Duret L, Kadlec J, de Massy B, Baudat F. PRDM9 Methyltransferase Activity Is Essential for Meiotic DNA Double-Strand Break Formation at Its Binding Sites. *Mol Cell*. 2018;69(5):853-65 e6. Epub 2018/02/27. doi: 10.1016/j.molcel.2018.01.033. PubMed PMID: 29478809.
2. Eaker S, Pyle A, Cobb J, Handel MA. Evidence for meiotic spindle checkpoint from analysis of spermatocytes from Robertsonian-chromosome heterozygous mice. *J Cell Sci*. 2001;114(Pt 16):2953-65. Epub 2001/11/01. doi: 10.1242/jcs.114.16.2953. PubMed PMID: 11686299.
3. Neale MJ, Pan J, Keeney S. Endonucleolytic processing of covalent protein-linked DNA double-strand breaks. *Nature*. 2005;436(7053):1053-7. Epub 2005/08/19. doi: 10.1038/nature03872. PubMed PMID: 16107854; PubMed Central PMCID: PMC1262668.
4. Stanzione M, Baumann M, Papanikos F, Dereli I, Lange J, Ramlal A, et al. Meiotic DNA break formation requires the unsynapsed chromosome axis-binding protein IHO1 (CCDC36) in mice. *Nat Cell Biol*. 2016;18(11):1208-20. Epub 2016/10/28. doi: 10.1038/ncb3417. PubMed PMID: 27723721; PubMed Central PMCID: PMC5089853.

Table S1B. Secondary antibodies used for WB, IF, IP: application, source, and working dilution

Secondary antibodies	Application	Source
Protein A (HRP)	WB 1:10.000	BioLegend 689202
Goat α Guinea Pig (IgG H+L) (HRP)	WB 1:10.000	Novus Biologicals NB 7398
Mouse (m-IgGk BP-HRP)	WB 1:10.000	Santa Cruz sc-516102
Goat α Rabbit (IgG-HRP)	WB 1:10.000	Santa Cruz sc-2054
Alexa Fluor 568 Mouse	IF 1:400	Life Technologies A11004
Alexa Fluor 647 Rabbit	IF 1:400	Life Technologies A21244
Alexa Fluor 488 Rabbit	IF 1:400	Life Technologies A11008
Goat Anti-Mouse IgG 488	IF 1:400	SouthernBiotech 1031-30
Alexa Fluor 594 Rat	IF 1:400	Life Technologies A48264

WB: western blotting; IF: immunofluorescence

Table S2. Primers used for ChIP-qPCR

Locus	Name	Sequence_F	Sequence_R
Chromosome 14	14a	TCCTGGTAATTTTTCTTGCTTT	CCAATCCAATGTCCCTTACA
Chromosome 17	17b	CATGGACATGGAGACCTAACTG	TCAGTGGAAAGCTCAGAAAATGA
Chromosome 10	10qC2	GCCAAGTAAGCTCTTCCTCC	TGACTTCTGCCTACCACTCA
Chromosome 12	12qA1.1	AACGTCCAGCCTAATTGTCC	GAATCAGGACCTCAGGCAAG
Intragenic region Polr2a	Pol II-Intra	CTGTCTCCAGGTCATTGAGAAGG	GTTGATCTTGGAACCCTTGGCTCC
Chromosome X	PAR1	CTCCTGGTGATCGAGTCAGG	GGAAGAAAGTCTCAGGCCAT
Chromosome X	PAR2	ACAAGAGTGTCTATACTGCTCAGG	GATCTCCACACTAGCTGGCG
Chromosome X	X-Hs	GTTTCATGCACTATATTGTGGTTATA	GACCTAAACACAAATACATGCACAC
Chromosome X	X-Cs	TGTTGACTTTGGCTGTGCAA	AGGACAACATGGCAGGAGTG