

Table S1. Mapping statistics for SPO11-oligo reads, Related to STAR Methods**Table S1A. Mapping statistics for SPO11-oligo reads in five datasets**

Dataset	Total	Nuclear genome			chrM		
		Unique	Multi ¹	Total	Unique	Multi ¹	Total
B6	69417156	46615259	22798301	69413560	1978	1618	3596
<i>Atm</i> wt	57341378	36776157	20560672	57336829	2966	1583	4549
<i>Atm</i> het	87651414	60824069	26814262	87638331	7979	5104	13083
<i>Atm</i> null 1	112091499	87894761	24191046	112085807	2917	2775	5692
<i>Atm</i> null 2	68732743	57582583	11144707	68727290	3602	1851	5453

¹ Multi counts are after imputation.

Table S1B. Mapping statistics for shortened SPO11-oligo reads in three datasets

Dataset	Total	Nuclear genome			chrM		
		Unique	Multi ¹	Total	Unique	Multi ¹	Total
<i>Atm</i> wt	2752725	1855712	896836	2752548	122	55	177
<i>Atm</i> null 1	5410887	3286639	2124064	5410703	103	81	184
<i>Atm</i> null 2	3670695	2233626	1436799	3670425	158	112	270

¹ Multi counts are after normalization.

Table S2. Crossover hotspots identified by pedigree analysis or by sperm typing, Related to Figures 1D, S1B, 6 and S6

Table S2A. Four crossover hotspots on chromosome 1, A1–A4, previously delineated by pedigree analysis

Four hotspots on chromosome 1 were previously defined by pedigree analysis in crosses involving haplotypes from C57BL/6 (“B6”), C3H/HeJ (“C3H”), SPRET/EiJ (“SPRET”) and CAST/EiJ (“CAST”) strains (Kelmenson et al., 2005).

Hotspot	Start marker ¹	End marker ¹	Coordinates in genome assembly GRCm38/mm10 (bp)					
			Crossover hotspot ¹			SPO11-oligo hotspot ²		
			Start	End	Length	Start	End	Length
A1	D1Pgn9	D1Pgn10	159980411	159984488	4078	159983887	159983964	78
A2	D1Pgn6	D1Pgn13	160000209	160020166	19958	160014582	160014614	33
A3	D1Pgn14	D1Pgn15	160025001	160026084	1084	160025419	160026167	749
A4 ³	D1Pgn11	D1Pgn12	160058414	160081429	23016	160063421	160063583	163
						160079882	160080548	667

¹ Polymorphic markers are reported in Kelmenson et al., 2005. Start marker and End marker define crossover hotspot Start position and End position, respectively. For microsatellite polymorphisms D1Pgn6, D1Pgn9, D1Pgn10, D1Pgn11 and D1Pgn12, coordinates are determined by the 3' position of the DNA segment of the marker, as compiled in <http://www.informatics.jax.org>. For SNPs D1Pgn13, D1Pgn14 and D1Pgn15, coordinates refer to the specific position of the SNP.

² SPO11-oligo hotspot coordinates reflect the Start and End in **Table S3A**.

³ A4 delineated by pedigree analysis encompasses two distinct SPO11-oligo hotspots.

Table S2 continued. Crossover hotspots identified by pedigree analysis or by sperm typing, Related to Figures 1D, S1B, 6 and S6

Table S2B. Crossover hotspots previously delineated by sperm typing

In the literature we identified 16 crossover hotspots previously defined by fine-scale analysis of crossover products in sperm isolated from F₁ hybrids of crosses between C57BL/6J (“B6”) and either A/J, C3H/HeJ (“C3H”) or DBA/2J (“DBA”) (Kauppi et al., 2007; Bois, 2007; Wu et al., 2010; Getun et al., 2010; Cole et al., 2010; Getun et al., 2012; de Boer et al., 2015). See **Figure S6** for SPO11-oligo maps, SSDS maps, crossover rates, and noncrossover conversion frequencies in 5001-bp windows around these hotspots.

Hotspot	F ₁ Hybrid	Reference	Coordinates in genome assembly GRCm38/mm10 (bp)					
			Crossover hotspot ¹			SPO11-oligo hotspot ²		
			Start	End	Length	Start	End	Length
Chromosome 1								
central	B6×A/J	de Boer et al., 2015	78589841	78591616	1776	78590834	78591048	215
<i>M1</i>	B6×C3H	Kauppi et al., 2007	143875788	143878121	2334	143877042	143877098	57
<i>A3</i>	B6×DBA	Cole et al., 2010	160024019	160027342	3324	160025419	160026167	749
distal	B6×A/J	de Boer et al., 2015	185265802	185268500	2699	185266572	185267584	1013
Chromosome 19								
<i>HS9</i>	B6×DBA	Bois 2007	10668187	10670548	2362	10669828	10670359	532
<i>HS14.9</i>	B6×DBA	Wu et al., 2010	14882384	14885235	2852	14883732	14884165	434
<i>HS18.2</i>	B6×DBA	Wu et al., 2010	18146103	18147531	1429	no hotspot by SPO11 oligos		
<i>HS22</i>	B6×DBA	Bois 2007	22960142	22962712	2571	22961596	22962032	437
<i>HS23.9</i>	B6×DBA	Wu et al., 2010	23841534	23843074	1541	23842467	23842554	88
<i>HS37</i>	B6×DBA	Bois 2007	38677802	38679593	1792	38678165	38678322	158
<i>HS44.2</i>	B6×DBA	Wu et al., 2010	44148565	44151078	2514	44148684	44150891	2208
<i>HS48.3</i>	B6×DBA	Wu et al., 2010	48213103	48216187	3085	48214759	48215364	606
<i>HS59.4</i>	B6×DBA	Getun et al., 2010	59346779	59350838	4060	59349650	59350010	361
<i>HS59.5</i>	B6×DBA	Getun et al., 2012	59442354	59443805	1452	59442410	59443524	1115
<i>HS61.1</i>	B6×DBA	Wu et al., 2010	61011233	61013223	1991	61012247	61012974	728
<i>HS61.2</i>	B6×DBA	Wu et al., 2010	61152469	61154630	2162	61152931	61153866	936

¹ Except for *HS9*, crossover hotspot coordinates are determined by SNPs that encompass all mapped crossovers (Kauppi et al., 2007; Bois, 2007; Wu et al., 2010; Getun et al., 2010; Cole et al., 2010; Getun et al., 2012; de Boer et al., 2015). For *HS9*, crossover hotspot coordinates are determined by 3' positions of internal B6-specific primers used for crossover detection (*HS9-B-2F* and *HS-9-3R* in Bois, 2007).

² SPO11-oligo hotspot coordinates reflect the Start and End in **Table S3A**.

Table S5. Data underlying chromosome-scale analyses, Related to Figure 4

Chromosome	Chromosome size (bp) ¹	SPO11 oligos (RPM) ²	SPO11-oligo density (RPM/Mb)	Crossover rate (cM) ^{3,4}
chr1	191909192	69606	363	76.2
chr2	178326651	73115	410	77.7
chr3	156398855	53878	344	63.1
chr4	152055611	60332	397	70.1
chr5	147919674	62217	421	72.0
chr6	146336543	52717	360	59.7
chr7	141855407	52687	371	68.1
chr8	125611432	50769	404	60.7
chr9	121157018	50618	418	55.7
chr10	127067662	49930	393	61.7
chr11	118745945	55509	467	69.9
chr12	116922420	45270	387	54.6
chr13	117121193	46151	394	51.4
chr14	121442110	44188	364	53.2
chr15	100653315	40639	404	49.5
chr16	95019758	37055	390	50.0
chr17	91707462	39641	432	50.9
chr18	87452634	34544	395	50.9
chr19	58205856	25629	440	50.0
non-PAR chrX	163326463	36779	225	NA
non-PAR chrY	88110223	9345	106	NA
PAR	176007	7312	41546	50.0

¹ Sequence gaps were subtracted from GRCm38/mm10 chromosome lengths.

² Counts only include SPO11 oligos mapping to each fully assembled chromosome contig (i.e., do not include SPO11 oligos mapping to unplaced or unassigned contigs).

³ Autosomal crossover rates using crossover marker MLH1 were reported in Froenicke et al., 2002.

⁴ Crossover rate in the PAR was set at 50 cM.