

Figure S1 Representative images of mouse and rabbit IgG immunostaining in mouse testes. IgG was used as a negative control. Scale bar: 20 µm. DAPI, 4',6-diamidino-2-phenylindole.

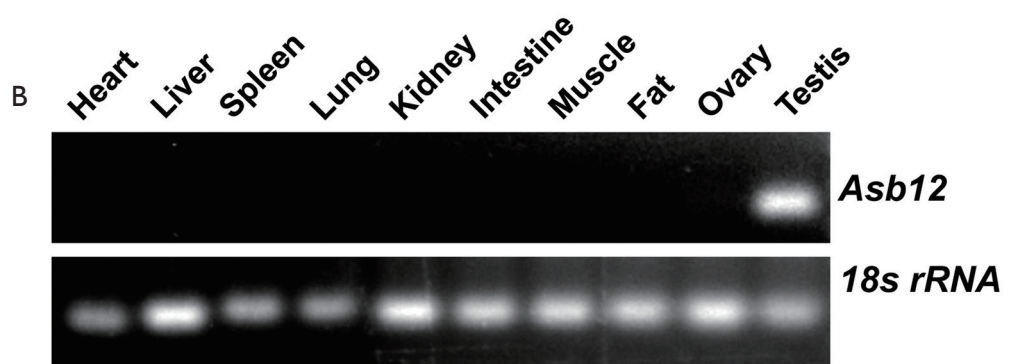
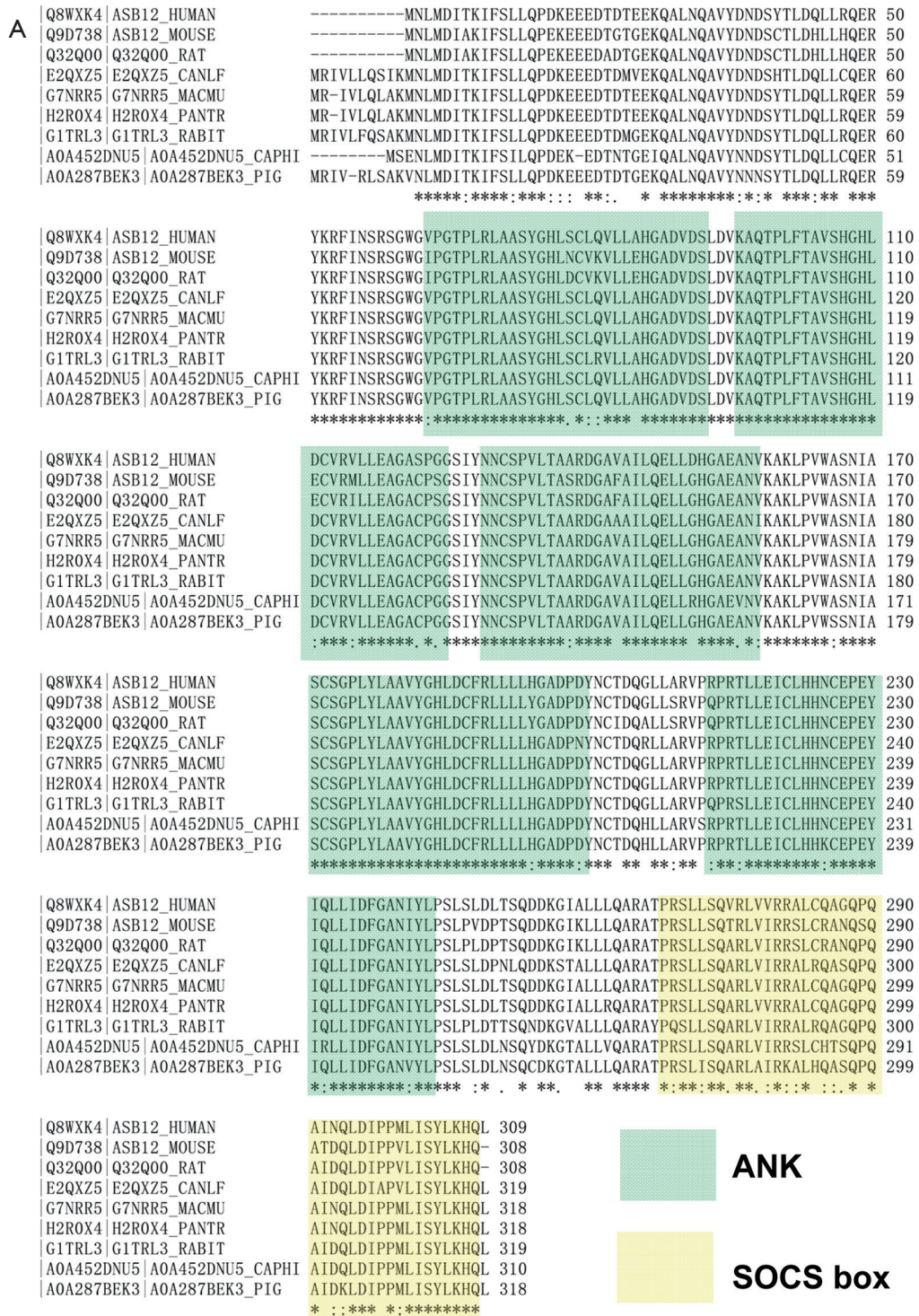


Figure S2 *Asb12* is an evolutionarily conserved and testis-enriched gene among species. (A) Sequence alignment of ASB12 proteins among species. (B) RT-qPCR of *Asb12* in multiple murine tissues. *Asb12*, ankyrin repeat and SOCS box 12.

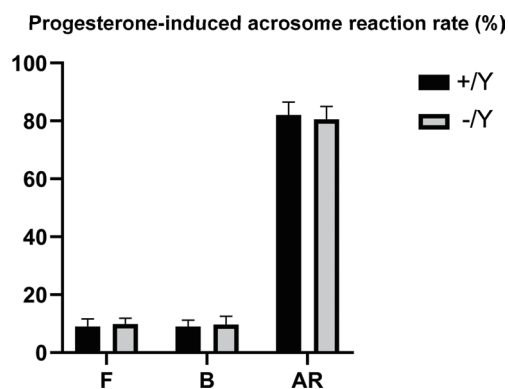


Figure S3 Chlortetracycline assay of the sperm acrosome reaction in *Asb12*-WT and *Asb12*-KO mice induced by treatment with 15 μ M progesterone for 15 min. $n=3$ for each group. $P>0.05$. F pattern: uncapacitated sperm; B pattern: capacitated sperm; AR: capacitated and acrosome-reacted sperm.

Table S1 The list of primer sequences

Gene	Forward (5'→3')	Reverse (5'→3')	Product length (bp)
<i>Asb1</i>	GCAATGTCAGTGCCCTGTTC	TGGGAAGGAAATCAGAGGGC	287
<i>Asb2</i>	CAAGGAGAAGGCAGAACCTCC	TGAAGCTCCAGCAAAGAACCA	270
<i>Asb3</i>	CAGCAGCTCTTGGGTTGATT	CTGGCCCCGAATTTCCAAACG	192
<i>Asb4</i>	CCCAGTAGTCCCGATGTAGC	CTGCCAGAATGTTCTGGTGA	281
<i>Asb5</i>	TCTTGATTTAGGGGAAAGATGGGA	AAACACAGGTAACCAATGACAGT	191
<i>Asb6</i>	ATGCAGCTGACAAGAATGGGAA	TTGGAAGCAGAAGCGGTTGA	230
<i>Asb7</i>	TATGGAGCGGACACGAACAC	TTCTGGGCTGTCTTGTGACG	124
<i>Asb8</i>	TTGGAAGTGGCCTTGCCCTAC	GCAGGCACACGTTTTTGAGT	202
<i>Asb9</i>	ACCTTATCAGCCAGGGTTGG	TCGCCAGTCTATGGTCATGC	149
<i>Asb10</i>	CTCAGTAACAGGTCCCAGGC	ACACACACCTCTGTCTTGCC	124
<i>Asb11</i>	GGTGGACTGTGACACTGCTG	GCATTGACATGTGCACCGTT	209
<i>Asb12</i>	TGGTCAGTAAGACAAGGGAAGT	CTGGCTGACTAATGCCGAGT	151
<i>Asb13</i>	CCTGCATGAGTGGGAGTTCT	GTTGTCCCGGGCATAGATGT	269
<i>Asb14</i>	ATGGCTATGACACTGAGCGA	AGAACGGGGGTTGGTCAAGA	272
<i>Asb15</i>	GATTCAAGCGTCTGCATGGC	GCTCAAAAATGCGACCTTGC	251
<i>Asb16</i>	GTCAGCGGAACAGGGATTCT	AAATGTGAGCAGGAGCCGAA	208
<i>Asb17</i>	GTAAGTGGGTTTTGCCAGGA	CACAGGTGTGAAGTTCTCCA	124
<i>Asb18</i>	ACAGGTCTCTCTCGCCTTG	TCCCTCACATAGAACCCTCTGC	238
<i>18s rRNA</i>	AAACGGCTACCACATCCAAG	CCTCCAATGGATCCTCGTTA	155

Asb, ankyrin repeat and SOCS box.

Table S2 Antibodies used in the study

Antigen	Source	Company	Application	Dilution	RRID
ASB12	Rabbit	Invitrogen	IF	1:200	AB_2817784
Lin28	Rabbit	Abcam	IF	1:200	AB_776033
SOX9	Rabbit	Millipore	IF	1:400	AB_2239761
SCP3	Rabbit	Abcam	IF	1:300	AB_301639
γ H2AX	Mouse	Abcam	IF	1:400	AB_470861

ASB12, ankyrin repeat and SOCS box 12; Lin28, lin-28 homolog; SOX9, SRY-box 9; SCP3, synaptonemal complex protein 3; γ H2AX, H2A. X variant histone.