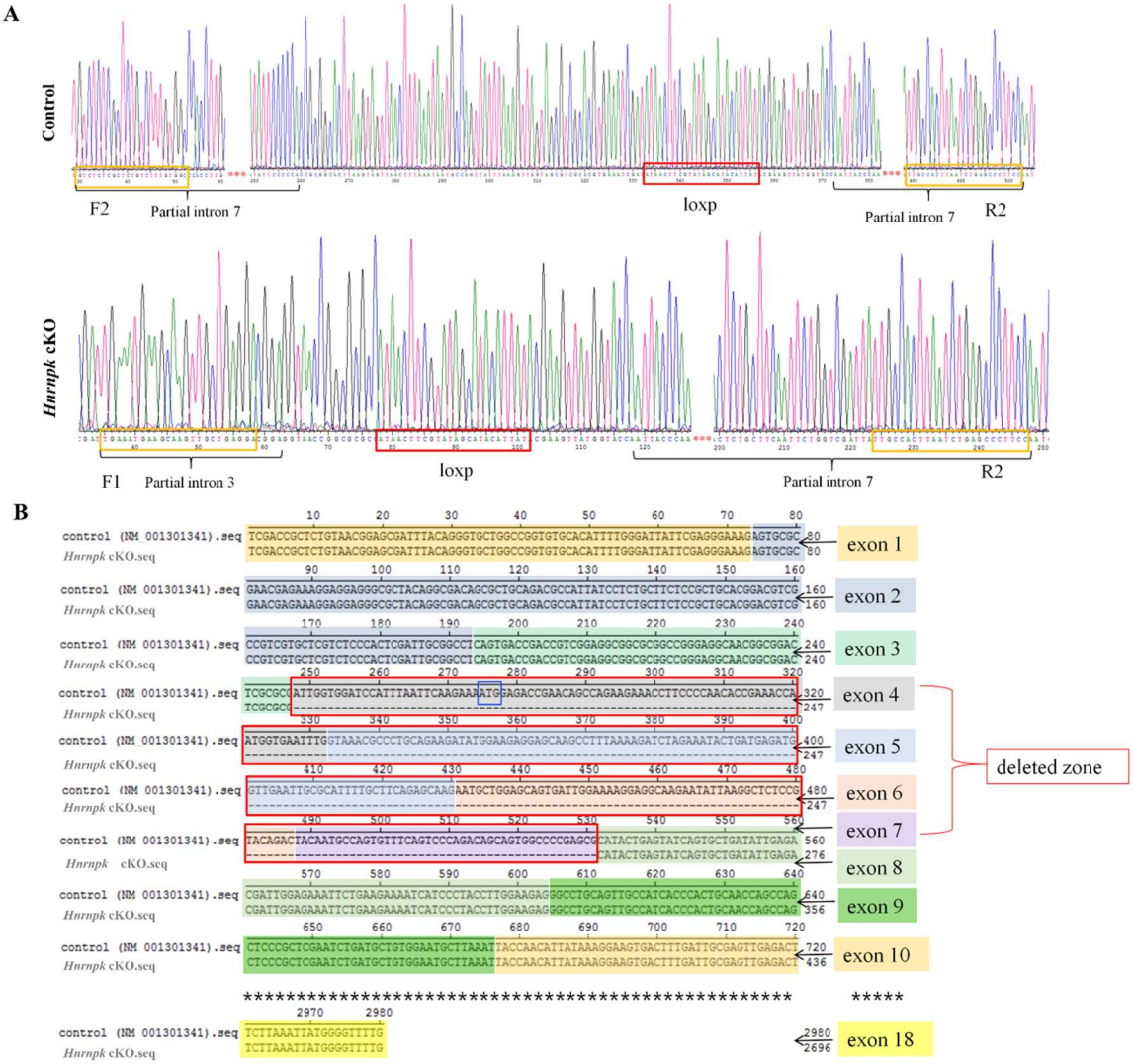
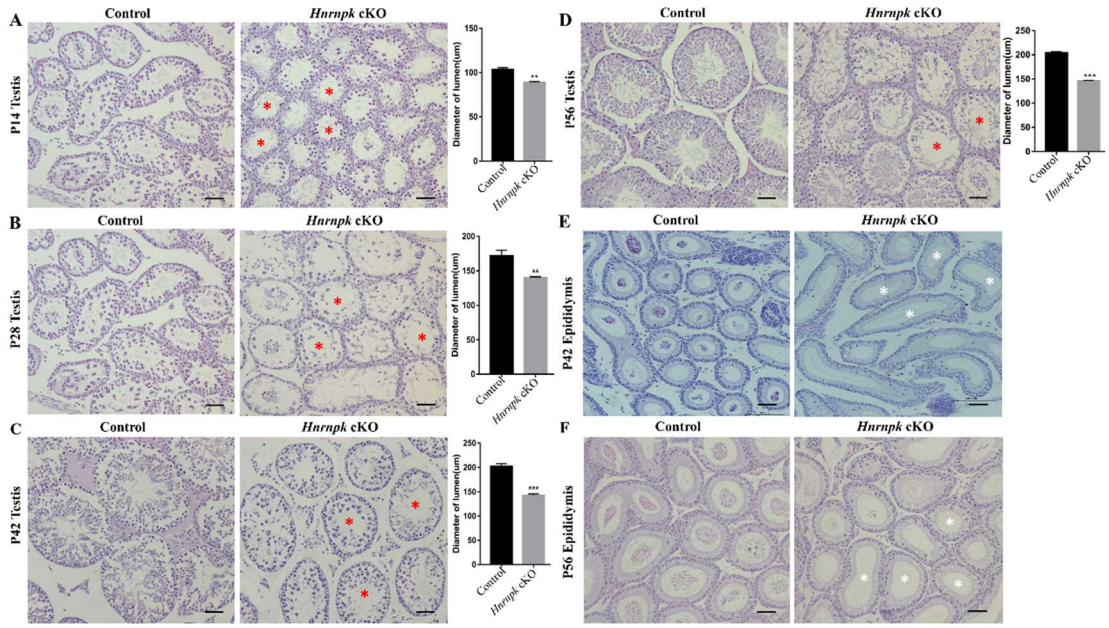
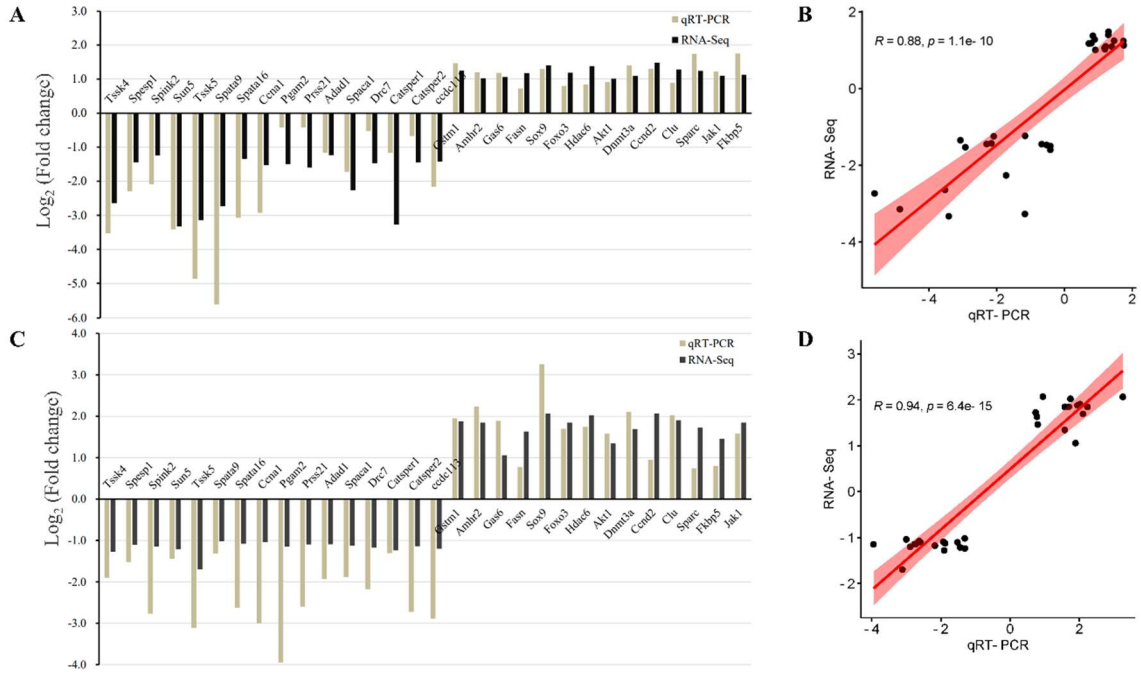


Supplementary Figure S1. DNA sequencing verified the knockout fragment of *Hnrnpk* gene. (A) Sequences of individually cloned fragments from WT control and *Hnrnpk* cKO mice genomic DNA of *Hnrnpk* gene. (B) Sequence alignment of exon 1 to exon 10 of the mouse *Hnrnpk* mRNA sequence between WT control (NM_001301341) and *Hnrnpk* cKO.



Supplementary Figure S2. HE staining of mouse testis and epididymis at different stages. (A) Typical pictures of testicular tissue morphology of P14 WT control and *Hnrnpk* cKO mice**.** (B) Typical pictures of testicular tissue morphology of P28 WT control and *Hnrnpk* cKO mice**.** (C) Typical pictures of testicular tissue morphology of P42 WT control and *Hnrnpk* cKO mice. (D)Typical pictures of testicular tissue morphology of P56 WT control and *Hnrnpk* cKO mice. (E) Typical pictures of epididymis tissue morphology of P42 WT control and *Hnrnpk* cKO mice. (F) Typical pictures of epididymis tissue morphology of P56 WT control and *Hnrnpk* cKO mice**.** The red star represents the lumen where the germ cell layer is absent, and the white asterisk indicates a noontime cluster of mature sperm in the epididymis. $P < 0.001$ (***), $P < 0.01$ (**), n = 3. Bar = 50 μm.



Supplementary Figure S3. Validation by qRT-PCR analysis of 30 differentially expressed genes from RNA-Seq approach. (A) The fold-change of differentially expressed genes obtained by RNA-Seq analysis and qRT-PCR analysis in P21 testes. (B) Correlation between the fold-change in RNA-Seq analysisand those obtained by qPCR analysis for the 30 selected genes in P21 testes. (C) The fold-change of differentially expressed genes obtained by RNA-Seq analysis and qRT-PCR analysis in P28 testes. (D) Correlation between the fold-change in RNA-Seq analysisand those obtained by qPCR analysis for the 30 selected genes in P28 testes.

Supplementary Table S1. Primer sequences for the specific genes of qRT-PCR analysis

Gene	Primer sequence（5'-3'）	Tm (℃)	Length （bp）	GenBank Accession No.
Tssk4	F:GTGAGATACAGGTAATGAAAGTCC R: GTATACTCGGGATGTGGTCTC	60	82	XM_036158850.1
Tssk5	F: TACCTGGCCTATGCAACAC R: TGATAGCCACCATAGTGTGG	60	90	NM_183099.2
Spata4	F: GCAGAGGAGTGTTTACCACTG R: GGACAGAACGACTCAGGCG	60	145	NM_133711.4
Spata9	F: TCTCTGGAAGACTTGAGGG R:CTGTGATAATCTTAAGATGGTAGGG	60	91	XM_030247441.2
Spata16	F: TCTACACAGATACCCAAGGAC R: GTCAGCAGATATTGCTGGG	60	81	NM_001177914.1
Sun5	F: ATAGACCTCCATCCTTGGC R: AACTTGCATCTCCTAGGCC	60	120	XM_030252189.2
Prss21	F:GAACTCCACGTACAAGTTTGAG R: ATGGCAGACTCTCATCTTCTC	60	82	NM_020487.4
Pgam2	F: AACATCTGGAAGGGATGTCC R: TCTGGTCCAGCTCATACAC	60	83	NM_018870.3
Adad1	F: GCAGTATACTTGTTGGTGATGG R: GTGAGTGCGTCATCAACAC	60	84	NM_009350.3
Ccna1	F: TCAAATGCTCAGCAGAGCT R: TGATTGCTGTGATCTCCTGG	60	115	XM_030252400.1
Spaca1	F: ACACTTGGATTCACCTCCA R: ACACTTGGATTCACCTCCA	60	125	NM_001290443.1
Drc7	F: GATCACTCGGTCATAAGCCA R: CTCCTCCTTCAATGAGTTGGT	60	80	NM_001042715.3
Ccdc113	F: ACTCTGCTCTCAAAACAGAAACA R: GCCTGGATCGTAACTGTGAAA	60	132	XM_006531000.3
Catsper1	F: CTGCCTCTTCCTCTTCTCTG R: GTGGTAAAGATGTTCTGGAAGC	60	80	NM_139301.3
Catsper2	F: TCCGGGAGATACTTGATCCT R: GCTTTATGGAGAAGCGCAC	60	80	XM_006531000.3
Spink2	F: CTTTGCAGCCTCTCATGAG R: CAGGGTTGAGGTTCCTAGG	60	117	XM_011249564.4
Amhr2	F: GTACTCCCTGGTCTTGCTC R: TCTTGTCCAAGAGCTCTGG	60	114	NM_001356575.1
Clu	F: AGAAAGAGGATGCTCTGGAG R: CCATCATGGTCTCGTTACAC	60	83	NM_013492.3

Sox9	F: CATGAACGCCTTCATGGTG R: TCTTCTCGCTCTCGTTCAG	60	130	NM_011448.4
Gas6	F: CCAAATGTGTTCAGAACTTGC R: TGAGGTCTTGGCAGATATGAG	60	80	NM_019521.2
Foxo3	F: CAAACGGCTCACTTTGTCC R: CCGGATGGAGTTCTTCCAG	60	108	NM_019740.3
Hdac6	F: CTGGGACAATCATCACCCT R: GGTAGGATGAGACAGCGAG	60	95	XM_017318389.3
Dnmt3a	F: AGACGTCTCCAACATGAGC R: ATCTGATCAGGACACAAGCA	60	121	XM_006514956.4
Jak1	F: CGACATTCTCCAAAGAAGCAG R: AATCATACTGTCCCTGTGCA	60	114	XM_006514956.4
Akt1	F: CTCAAGAAGGAGGTCATCGT R: GTACTTGAGGGCCGTAAGG	60	104	NM_001331107.1
Sparc	F: ATTGCAAACATGGCAAGGT R: TCATTGCTGCATACCTTCTC	60	117	NM_001290817.1
Gstm1	F: CTGTTCTCTGCCTCAGGAG R: ACATAGGTGACCTTGTCCC	60	122	NM_010358.5
Fasn	F: CTGACTCGGCTACTGACAC R: TTAGGGTAGGACCCTCAGG	60	110	NM_007988.3
Ccnd2	F: CATTGAGCACATCCTTCGC R: GCAAACTTGAAGTCGGTAGC	60	106	XM_036165787.1
Fkbp5	F: GAATATGCTTATGGCTCGGC R: TGAAATCAAGGAGCTCAATCTC	60	84	NM_010220.4
Hnrnpk	F: TTTGGCTGGATCTATTATTGGC R: CTGCTTCACACTGTTCTGC	60	181	XM_006517104.4
18S rRNA	F: ACCGCAGCTAGGAATAATGGA R: GCCTCAGTTCCGAAAACCA	60	63	NR_003278.3
Gapdh	F: AGGTCGGTGTGAACGGATTTG R: GGGGTCGTTGATGGCAACA	60	95	NM_001289726.1

Supplementary Table S2. Primary antibody list

Antibody	Resource	Usage and dilution	
Anti-γH2A.X	Beyotime, AF1201	IF	1:200
Anti-hnRNPK	Abcam, ab39975	IF	1:200
	Beyotime, AF1972	WB	1:1000
Anti-PLZF	Abcam, ab104854	IF	1:200
Anti-SYCP3	Abcam, ab97672-100	IF	1:100
Anti- Ki67	BD, 550609	IF	1:200
Anti-SOX9	Beyotime, AF2329	IF	1:50
Anti-GAPDH	Beyotime, AF0006	WB	1:1000