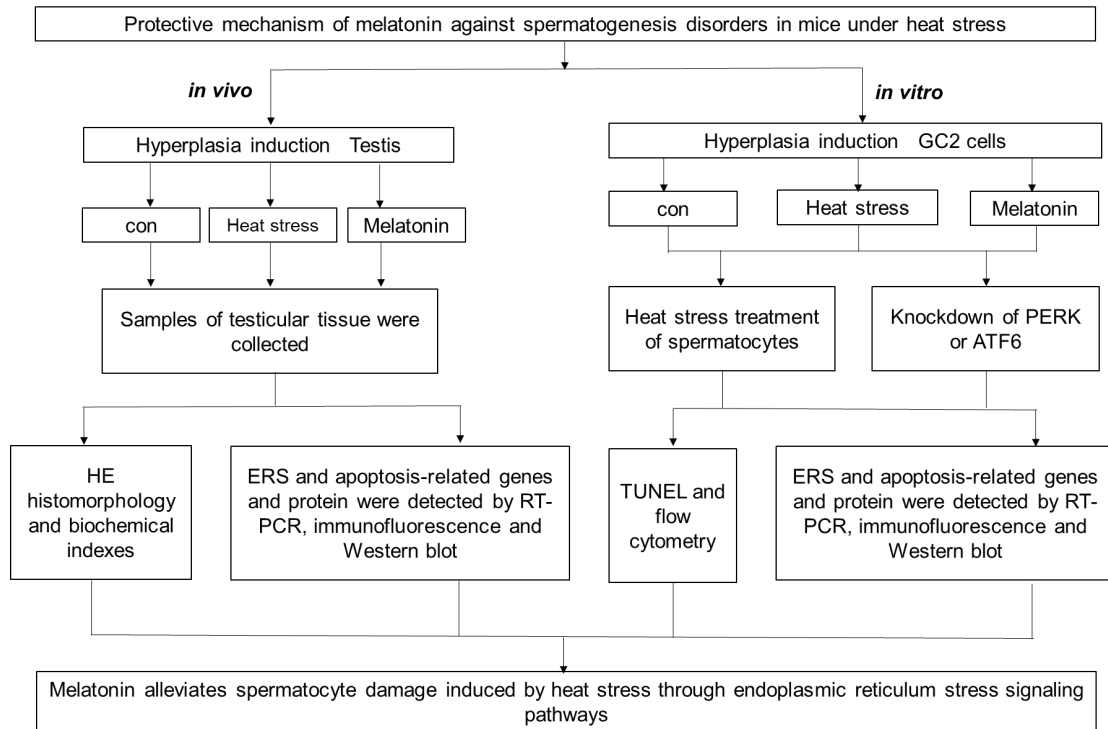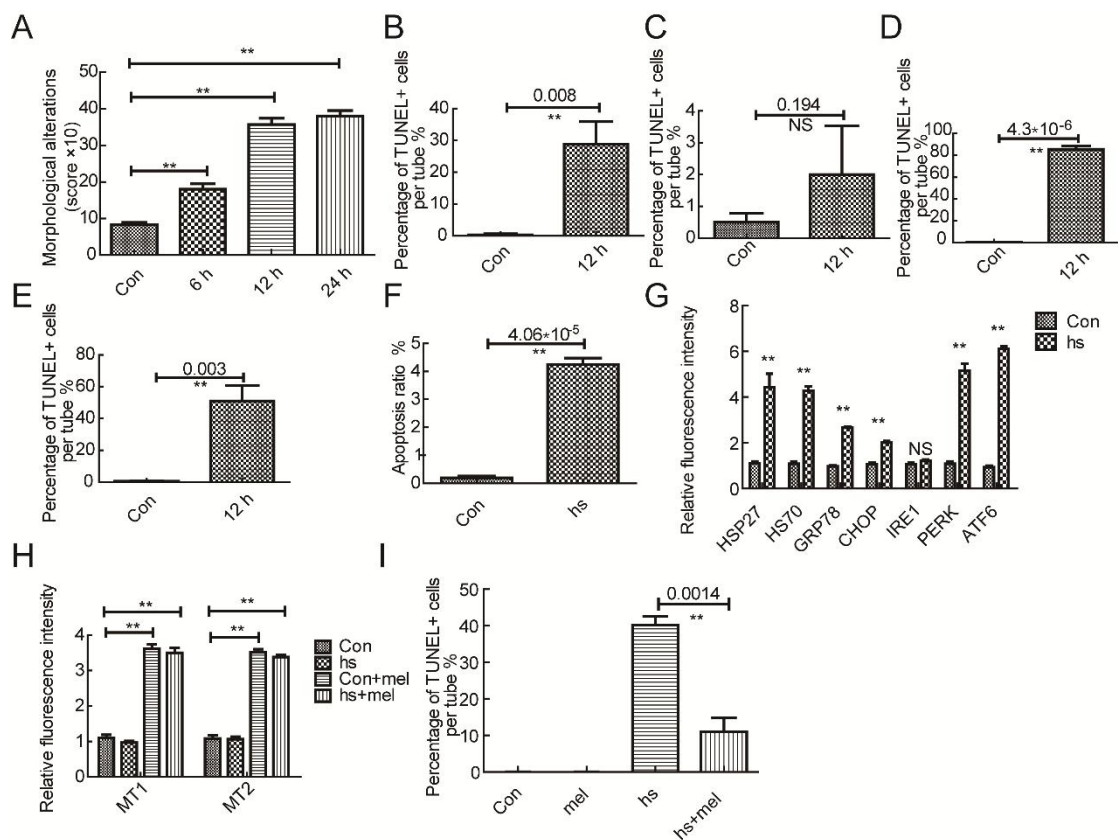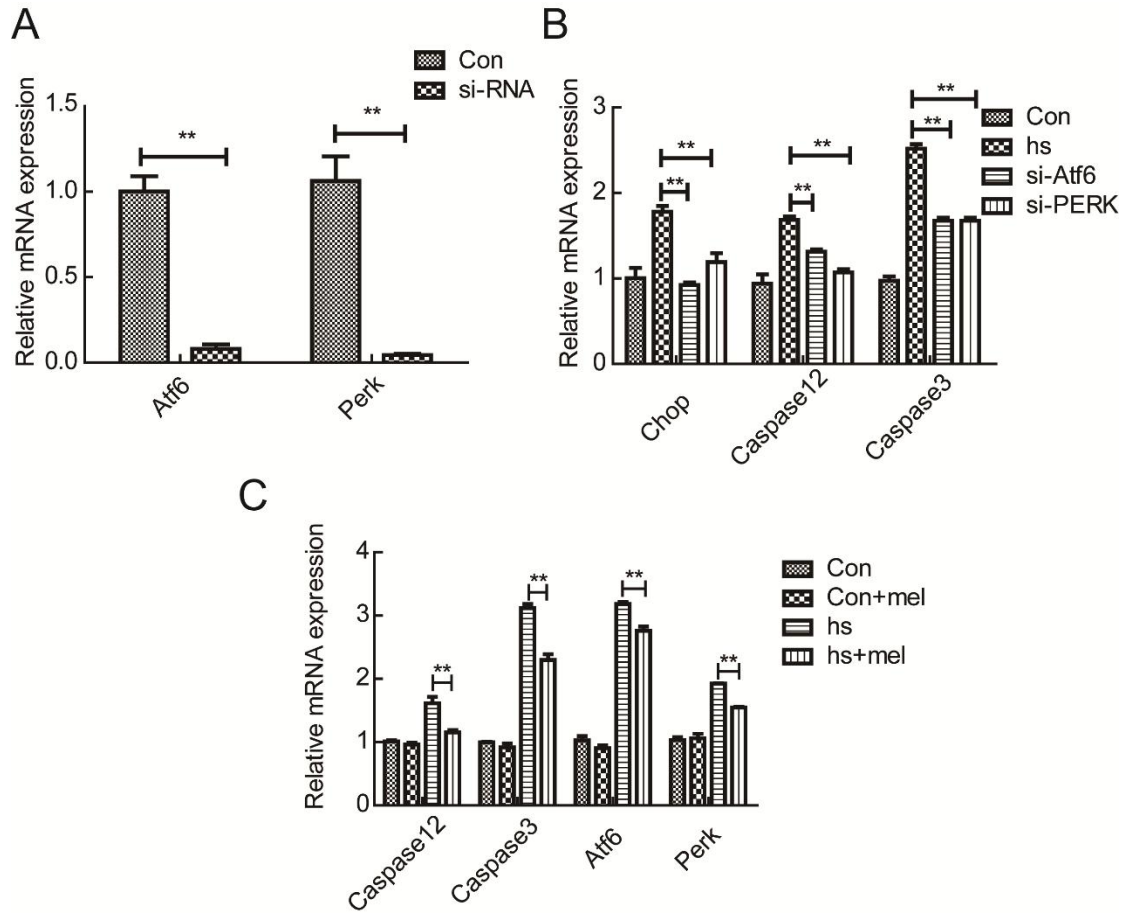# Supplementary Figures and Tables

Supplementary Figure S1. Diagram describing methodologies used in the current study.

Supplementary Figure S2. (A) Semi-quantitative analysis of seminiferous tubes in Figure 1A. (B) Quantification of percentage of TUNEL+ cells per tube. (C) Quantification of percentage of IRE1+ cells per tube. (D) Quantification of percentage of PERK+ cells per tube. (E) Quantification of percentage of ATF6+ cells per tube. (F) Quantification of percentage of apoptosis-positive cells per tube. (G) Quantification of relative fluorescence intensity of positive cells. From left to right, HSP27, HSP70, GRP78, CHOP, IRE1, PERK, and ATF6. (H) Quantification of expression of melatonin receptors. (I) Quantification of percentage of TUNEL+ cells per tube. Con: Control, hs: Heat stress, mel: melatonin. Data are mean±SD (n=3); statistical significance is expressed as: **P<0.01, NS, not significant.

Supplementary Figure S3. qRT-PCR of ERS- (*Perk* and *Atf6*) and apoptosis-related genes (*Caspase3*, *Caspase12*, and *Chop*) in 10^{-6} μM melatonin-treated spermatocytes 4 h after heat treatment. (B) qRT-PCR of apoptosis-related genes (*Caspase12*, *Caspase3*, and *Chop*) in si-Perk and si-ATF6 cell lines 4 h after heat treatment for 2 h. (C) qRT-PCR of *Perk* and *Atf6* mRNA expression levels in si-Perk and si-Atf6 cell lines. Con: Control, hs: Heat stress, mel: melatonin. Data are mean±*SD* (n=3); statistical significance is expressed as: **P<0.01.

Gene name	Forward primer (5′−3′)	Reverse primer (5′−3′)
m-*Grp78*	AGGGCAACCGCATCACG	CGCATCGCCAATCAGACG
m-*Atf6*	AACCAGGGATACGTGGGACC	CCACAGGTCCTCTTTAGGCTT
m-*Perk*	AGGCTTTAACTTCCCGCATT	AGTGCCAGACTGAAAGTAAATACG
m-*Ire1*	GCATAGTCAAAGTAGGTGGCA	GATAGTCTCTGCCCATCAACC
m-*Caspase12*	CTGGCTCTCATCATCTGCAACAA	CGGCCAGCAAACTGCATTAAC
m-*Caspase3*	GTCTGACTGGAAAGCCGAAAC	GACTGGATGAACCACGACCC
m-*Chop*	GAACAGTGGGCATCACCTC	CAGTCCCCTCCTCAGCAT
m-*β-Actin*	GTACCCAGGCATTGCTGACA	CGCAGCTCAGTAACAGTCCG
m-*Gapdh*	TGGCCTTCCGTGTTCCTAC	GAGTTGCTGTTGAAGTCGCA
m- *Hsp70*	GGCAGTACTTGGGATCAGGG	TCCACATCTTCTCCGAAGCG
m- *Hsp27*	CGAGGAGGTGGATTAGAGGC	ACAGTGCTGCTCCCAACATT

Supplementary Table S1. Primer sequences and gene numbers used in this study

Supplementary Table S2. Antibodies and dilutions used in western blotting and

Antibody	Catalog no.	Corporate brand	Dilution	Immusourcer
C-Caspase12	55238-1-AP	Proteintech Group	1:400	Rabbit IgG
C-Caspase3	bs-0081R	Beijing Biosynthesis	1:400	Rabbit IgG
CHOP	15204-1-AP	Proteintech Group	1:300	Rabbit IgG
p-PERK	bs−3330R	Beijing Biosynthesis	1:500	Rabbit IgG
p-IRE1α	bs-4308R	Beijing Biosynthesis	1:500	Rabbit IgG
ATF 6	24169-1-AP	Proteintech Group	1:500	Rabbit IgG
MT1	bs-0027R	Beijing Biosynthesis	1:200	Rabbit IgG
MT2	bs-0963R	Beijing Biosynthesis	1:200	Rabbit IgG
HSP70	bs-0244R	Beijing Biosynthesis	1:500	Rabbit IgG
HSP27	bs-0730R	Beijing Biosynthesis	1:500	Rabbit IgG
GRP78	WL00621	Wanlei Biotechnology	1:1000	Rabbit IgG
BOULE	13720-1-AP	Proteintech Group	1:500	Rabbit IgG
GAPDH	ZL9002	Beijing Jisi Jiayang	1:4000	Mouse IgG

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