

Supplementary Information

Genome-wide identification of long noncoding RNA and their potential interactors in ISWI mutants

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Table S1 Primer used in TSA-FISH

Transcript type	Transcript name	5' sequence	3' sequence
lncRNA	CR33938	CCTTGTTGGCTCATTCAAGTG	GCTCTTAGTTCTTGGACGGA
	Hsromea	CAAACCCCCTGGAAAAGAT G	GTATGGGGAAGCAAAACTGG
	CR44097	CACTGTATGATGGCCTTGAG	GAGTAATCGGGTTGTTTGGG
mRNA	brat	TCTATCAGACTCTCCGCTCA	GGCGTACTTGAATCATTCTG
	bt	GCCTGGGGGATAAGTGTAATG	GTAAGACCTGTAACATCGGC
	ap	CAGTCACCTCCGACATCATA	GGGGAGTCAGGCTAATATGG
	me31B	ACGAGATCAACCTGATGGA G	CTCACCCAAATGACCGAAAC
	AGO2	GTGACATACAGACCGTTCAC	GATAGGCGTTACGGTACTCT
Transposon	297 6	CCTCCTTTGAACATCACTCC	GTTGGAGTCTGGGTAAGGAA
	Max 5659	TACGAGTGAAGAGATCCGA G	GGATAGCCTTCGTGGAAAAG

The sequences of promoters connected to the flanks of the primers are not listed in the table. The forward primers connect T3 promoter: TGTTGGGAAATCACTCCCAATTAA. The reverse primers connect T7 promoter: GTAATACGACTCACTATAGGGAGACCAC.

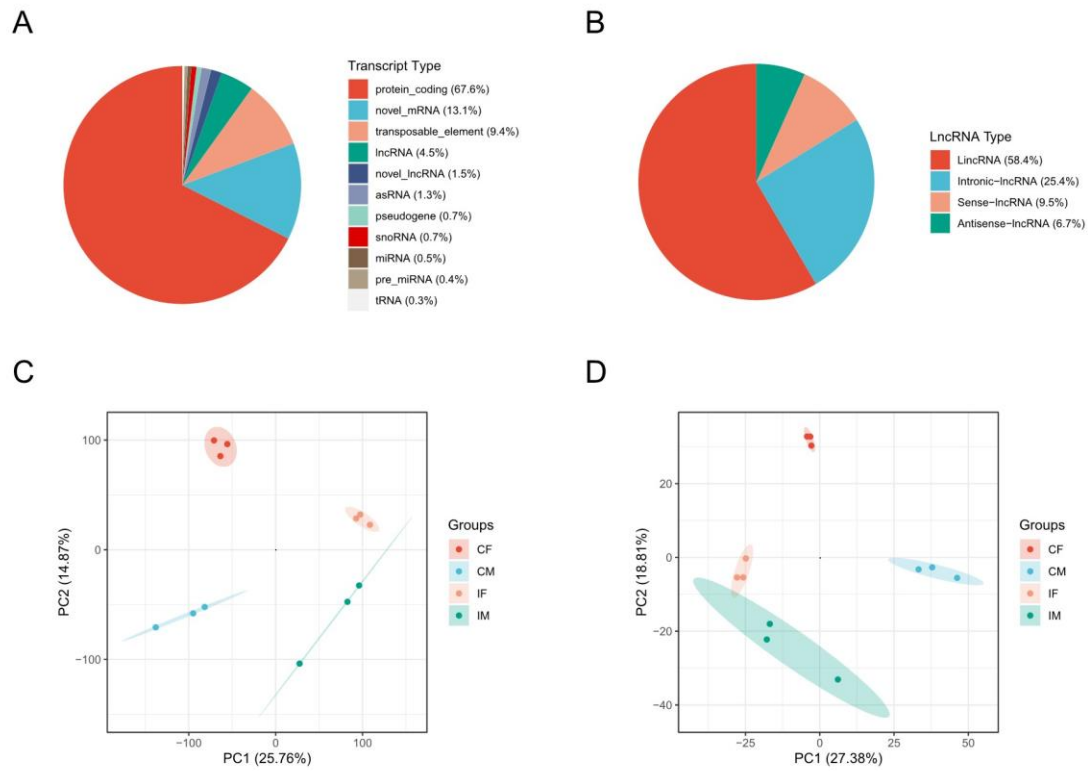


Figure S1 Overview of all transcripts of four kinds of samples. (A) Transcript type of all samples. (B) LncRNA types of green part of Figure S1(A). (C, D) Principal component analysis (PCA) plots show mRNA (C) and LncRNA (D) in mutant samples and controls. The circle represents a 95% confidence interval. CF, wildtype female control; CM, wildtype male control; IF, ISWI mutant female; IM, ISWI mutant male.

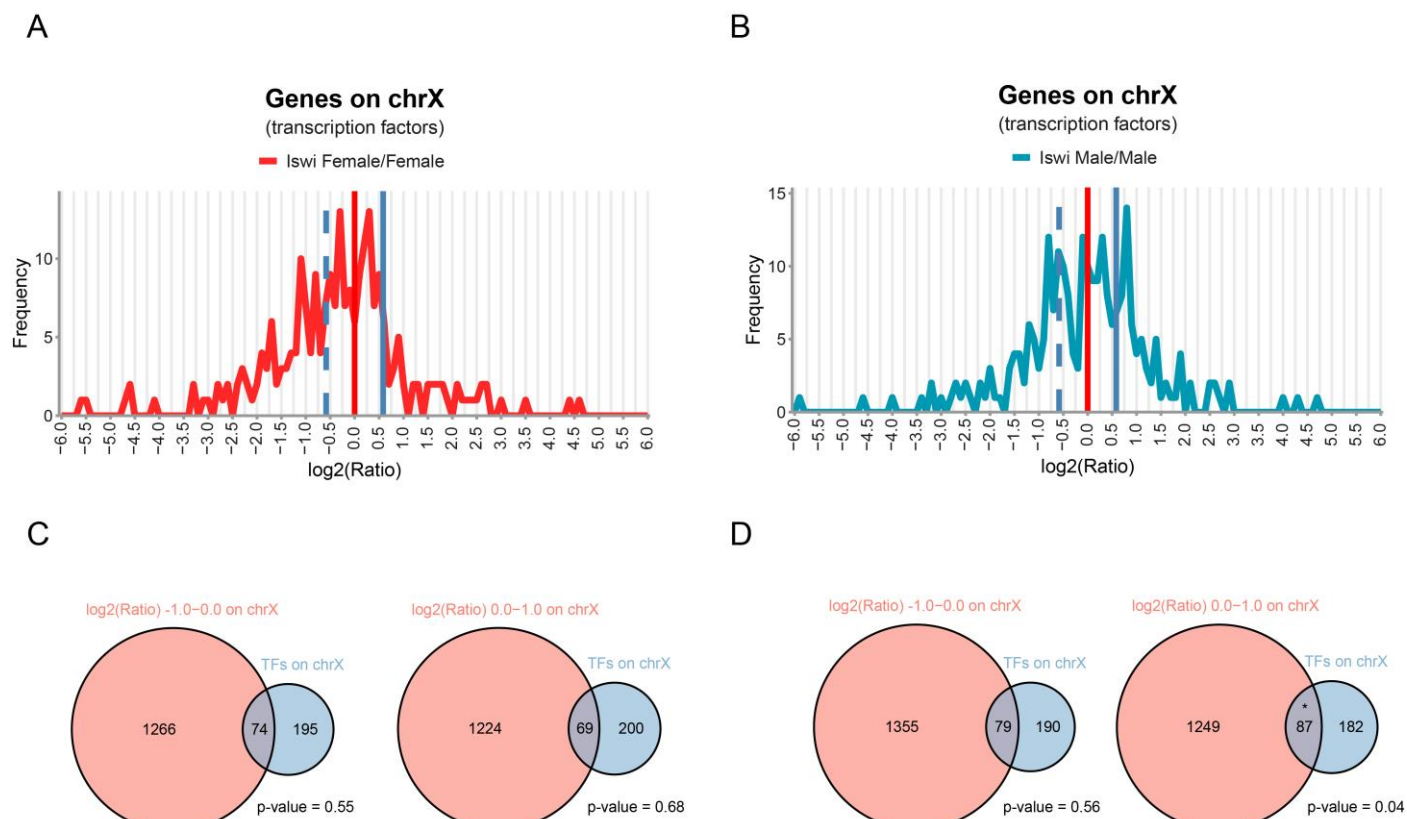


Figure S2 transcription factors expression in ISWI mutant compared to wild-type *Drosophila*. (A-B). Ratio distributions of transcription factors expression of female(A) and male(B) *Drosophila*. (C-D) The Venn diagrams of transcripts with $\log_2(\text{Ratio})$ values of -1.0-0.0 or 0.0-1.0 and transcription factors on X chromosome of female(C) and male(D) *Drosophila*. Fisher's exact test p values are shown beside the Venn diagrams.

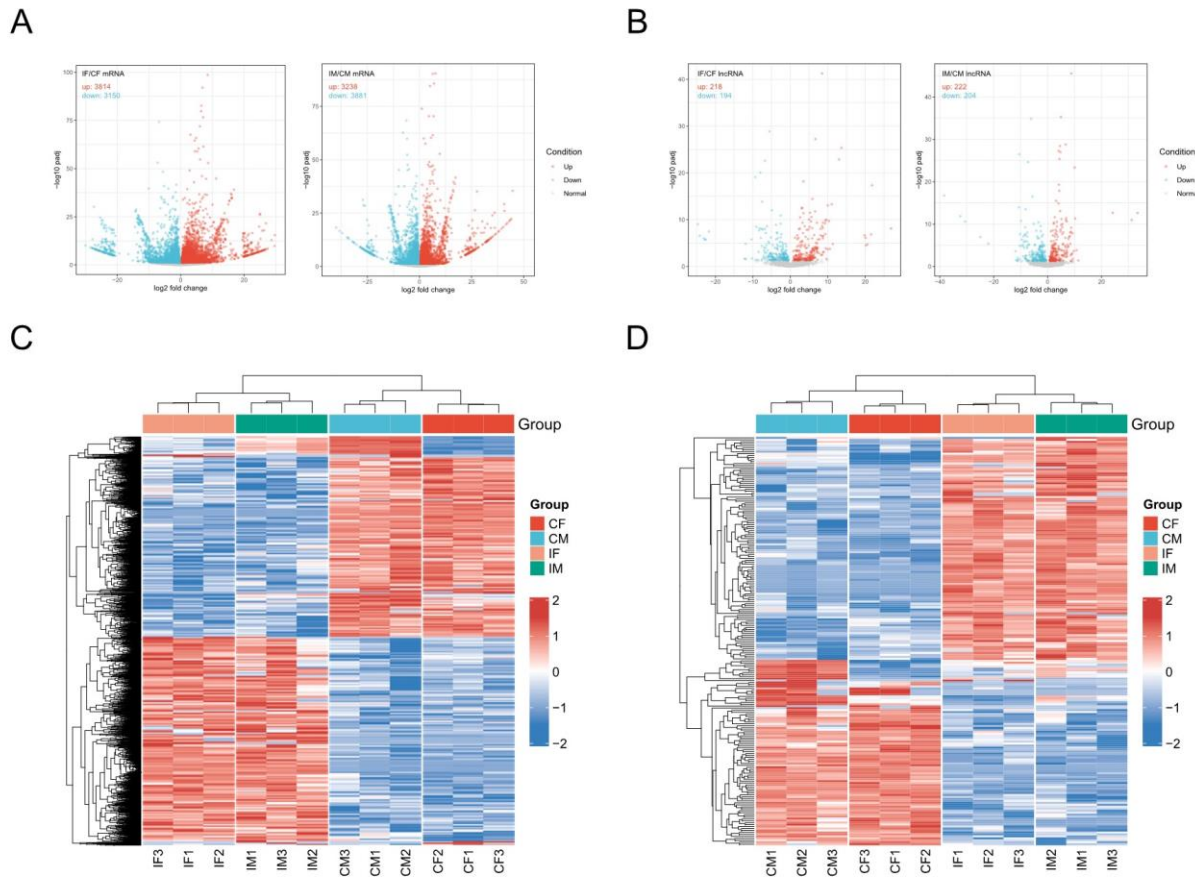


Figure S3 Differentially expressed transcripts of ISWI mutant and wild type. (A,B) Volcano plots of differentially expressed mRNA (A) and lncRNA (B) in ISWI mutation compared to the corresponding sex wild-type drosophila. Differential expressed transcripts are defined as adjusted p-value < 0.05 and fold change >1.25 or <0.8. (C,D) Heatmaps of mRNA (C) and lncRNA (D) of all mutant samples and control groups.

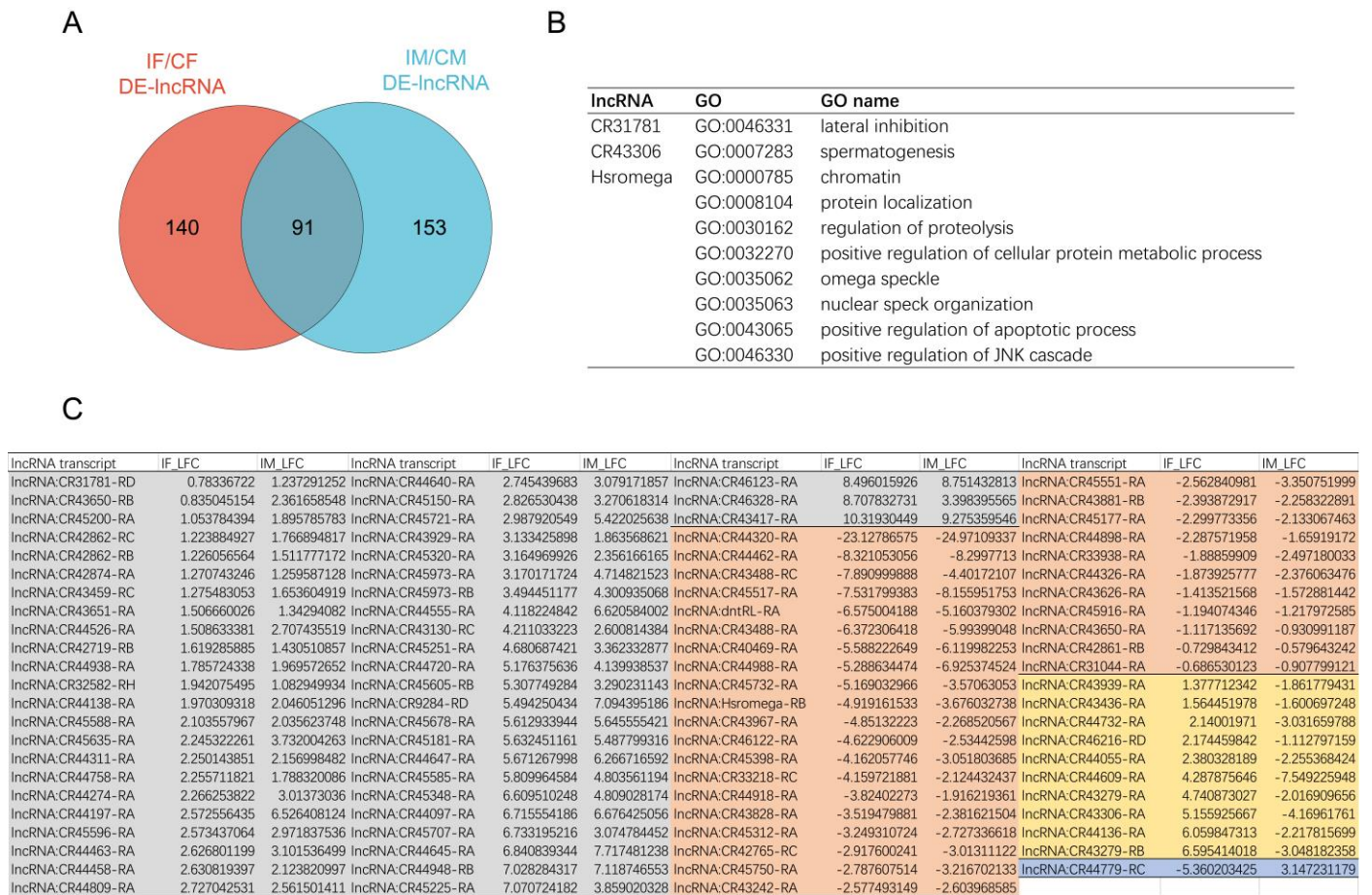
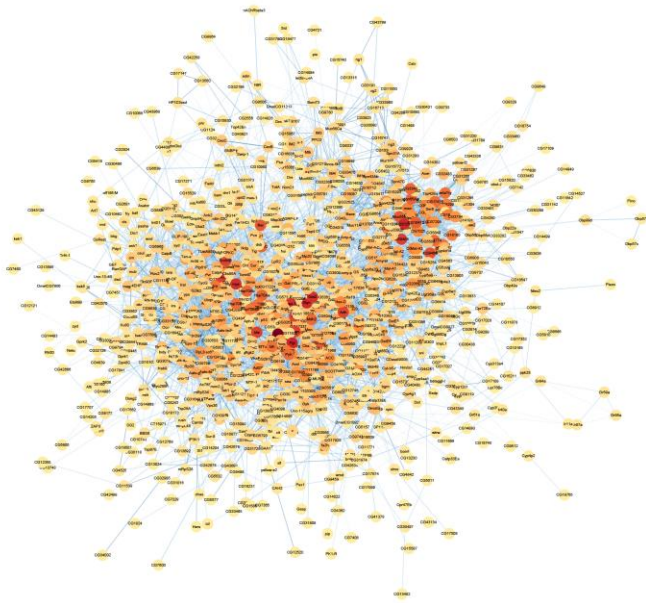
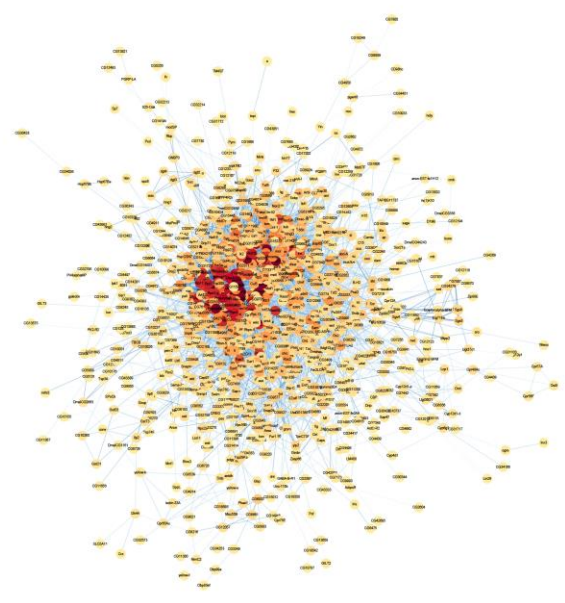


Figure S4 Differentially expressed known lncRNAs. (A) Differentially expressed known lncRNAs in both sexes; (B) Functions of differentially expressed lncRNAs in both sexes; (C) 91 differentially expressed lncRNAs in both sexes (gray: up-regulated lncRNAs in both sexes; orange: down-regulated lncRNAs in both sexes; yellow: up-regulated in female flies and down-regulated in male flies; blue: down-regulated in female and up-regulated in male flies).

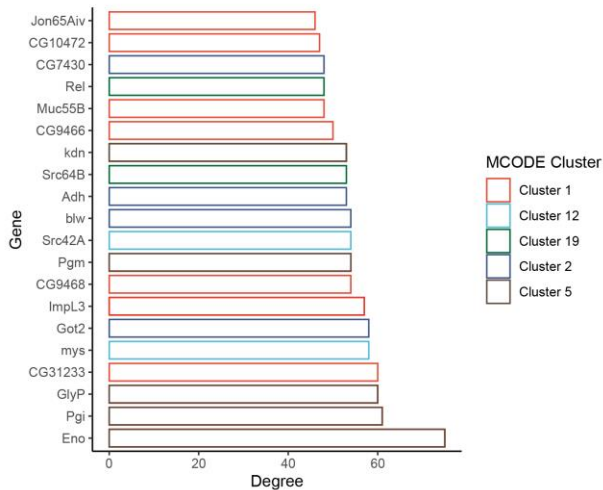
A



B



C



D

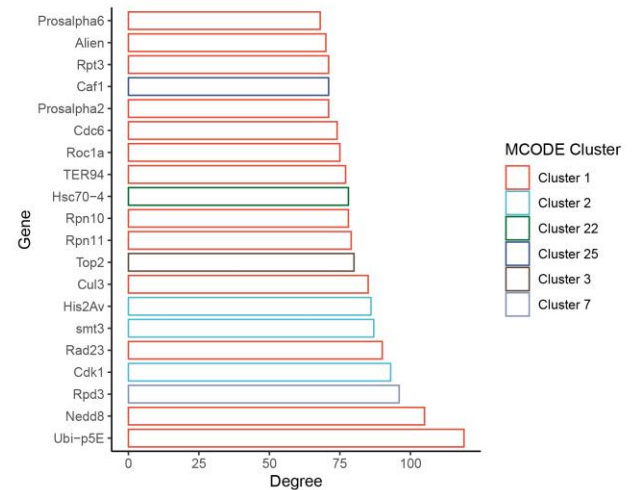
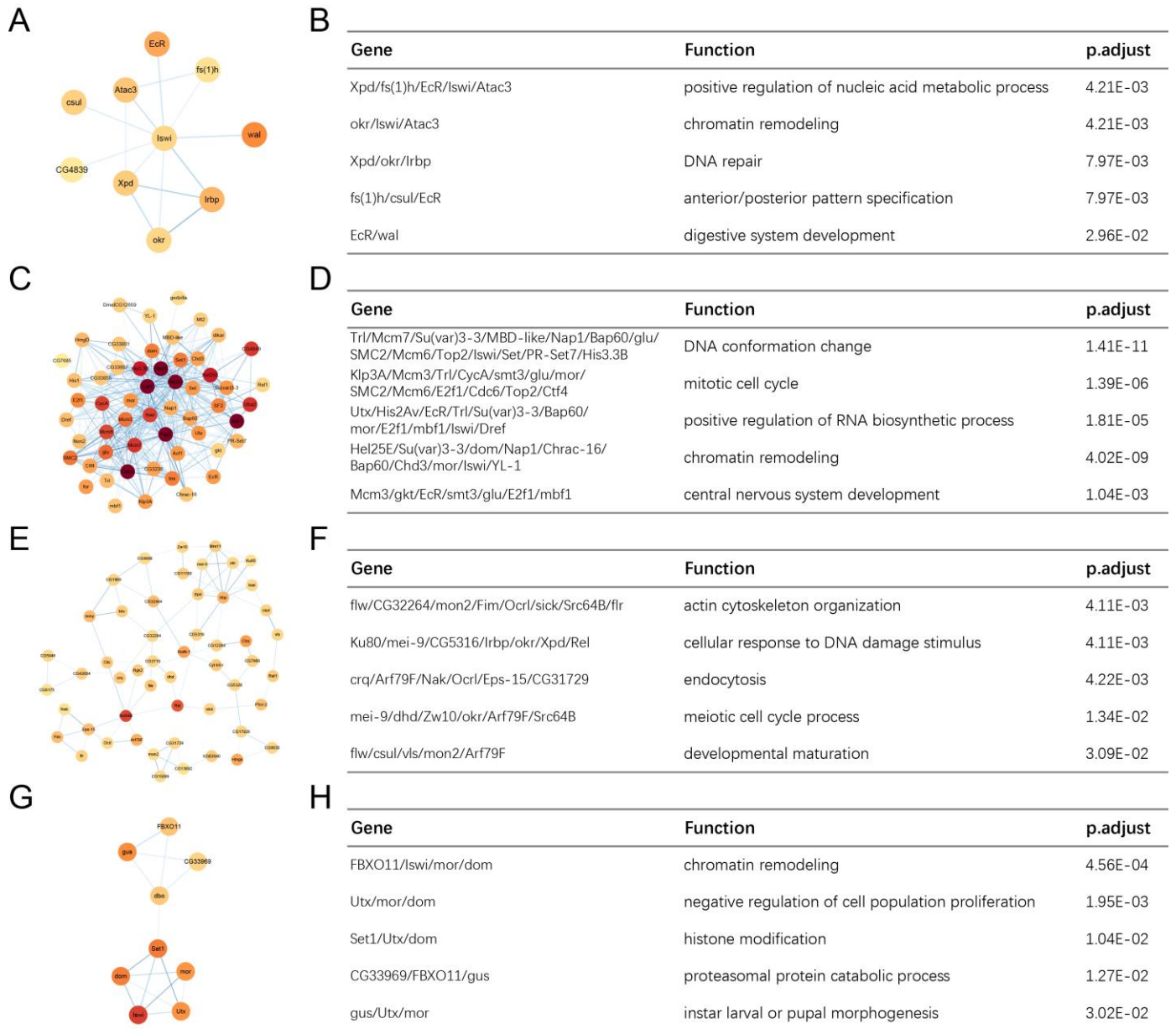


Figure S5 Protein-protein interaction (PPI) network of all DEGs. (A, B) PPI network of all up-(A) or down-(B) regulated DEGs. (C,D) The MCODE-modules of protein-protein interaction (PPI) networks of up- or down-regulates differential expressed genes. The top 20 connectedness genes and their Mcode modules are listed.



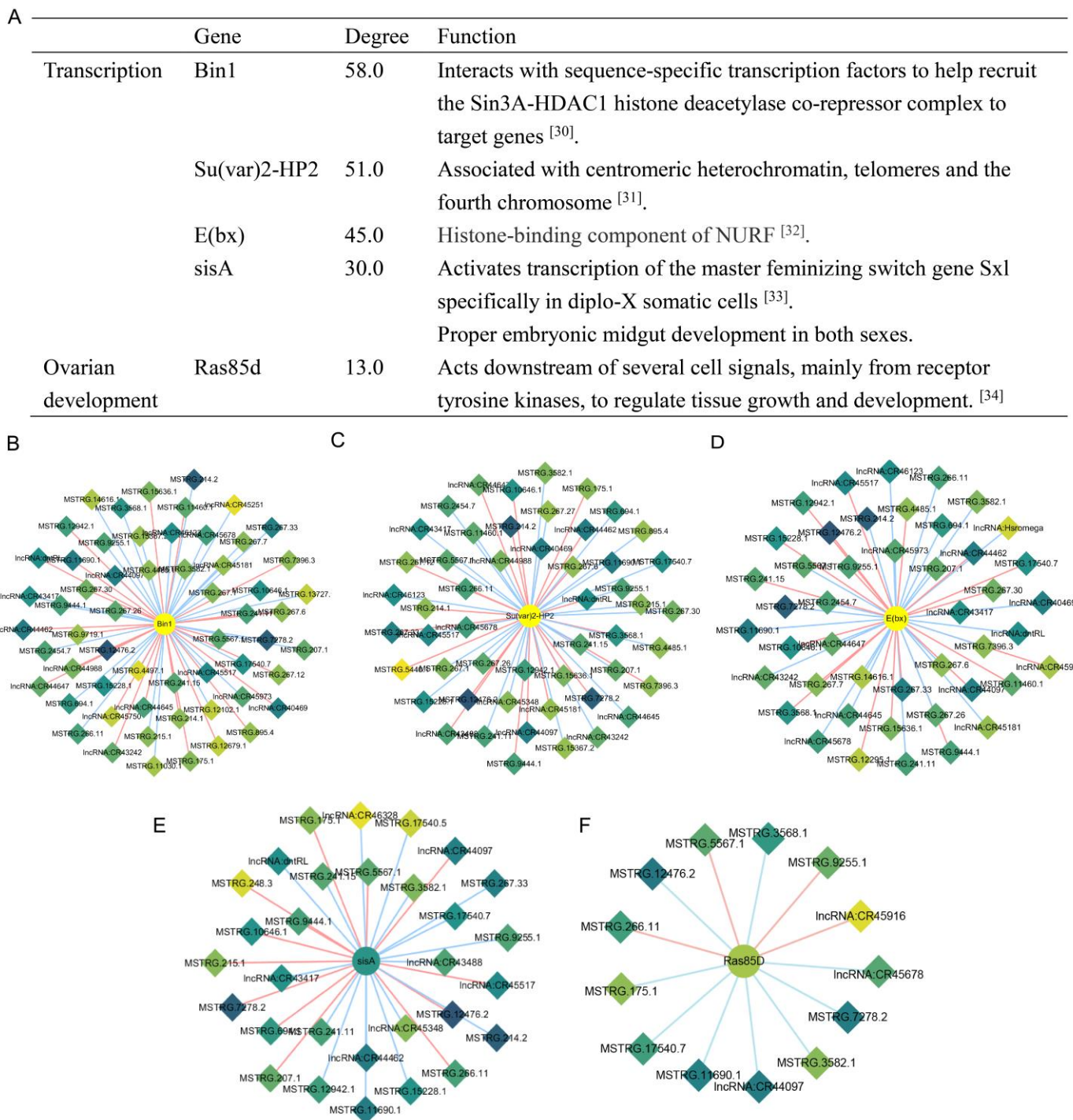


Figure S7 Possible interactors for DE-lncRNAs. (A) Five possible target genes and their connection degree (the number of co-expressed lncRNAs or mRNAs with it) and main functions. (B-F) The five genes in Figure A and their co-expressed lncRNAs: Bin (B), Su(VAR) 2-HP2 (C), E(bx)(D), sisA (E) and Ras85D(F). Network diagram of me31B and co-expressed lncRNAs. The diamond nodes represent lncRNAs, and the circle nodes represent mRNAs. The darker the color of node, the higher the degree of connection (the more co-expressed interactions). The diamond nodes represent lncRNAs, and the circle nodes represent mRNAs. The darker the color of node, the higher the degree of connection (the more co-expressed interactions). The red edge represents a positive correlation and the blue edge represents a negative correlation.

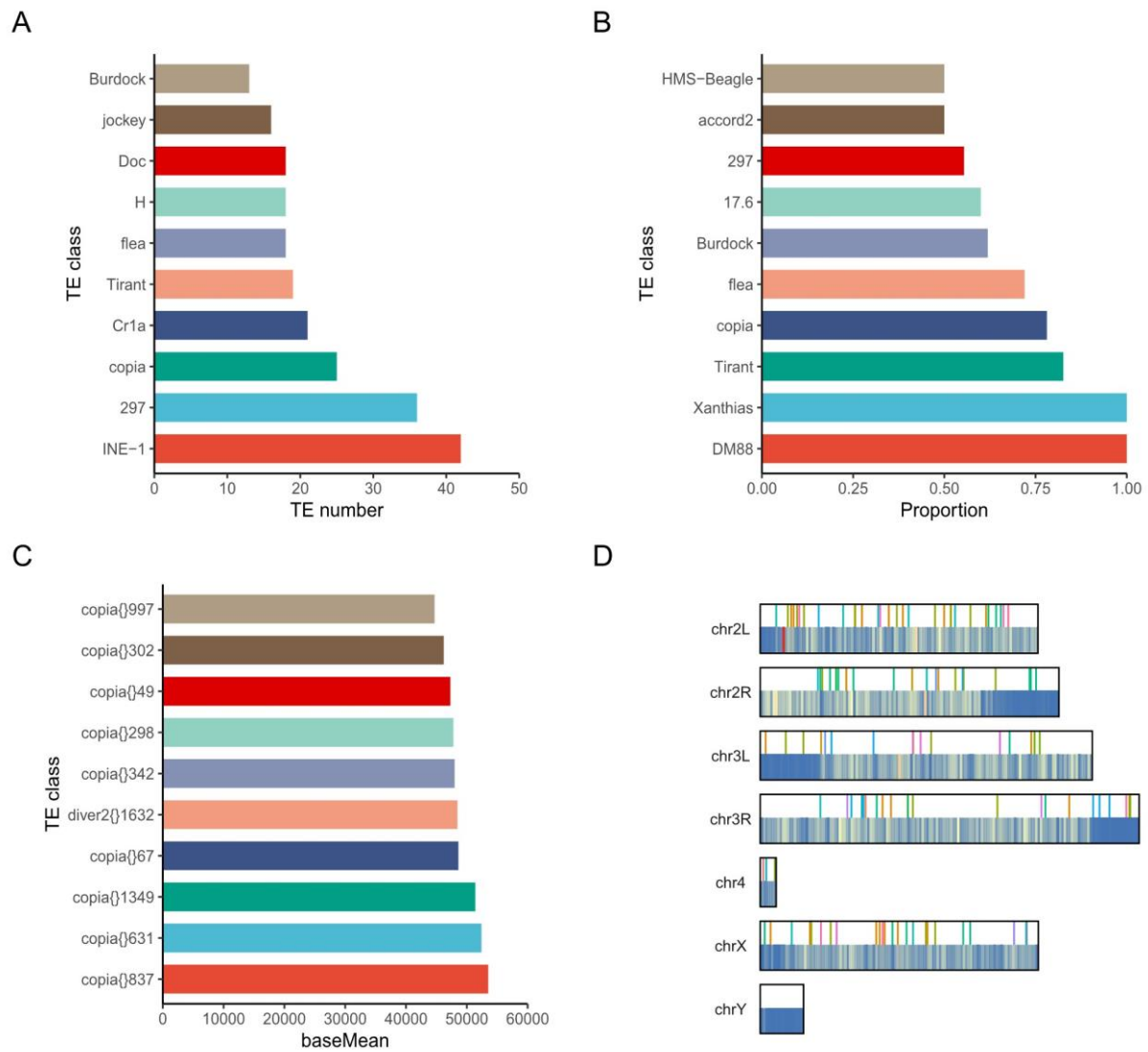


Figure S8 Characteristics and chromosomal localization of transposons with differential expression. (A) Top 10 TE class by number of transposons. (B) Top 10 TE class by the proportion of DE-TEs in the TE class. (C) Top 10 transposons in terms of expression in all samples. The horizontal coordinate baseMean is the amount of expression processed and standardized by DEseq2. (D) The location of TE class on chromosomes. The upper part of the white is the TEs on the chromosome, and the lower part of the blue is the gene on the chromosome.

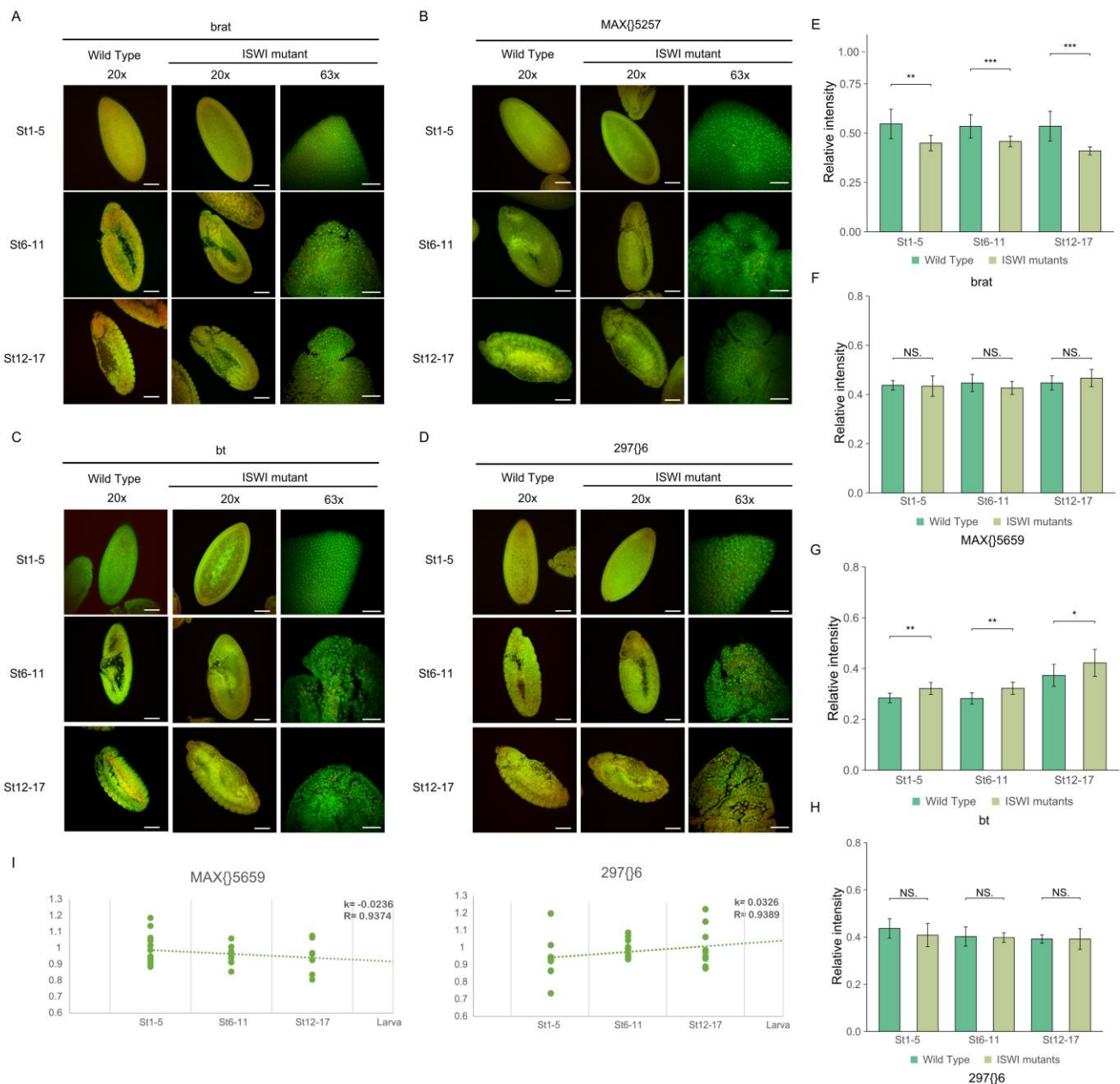


Figure S9 Embryo-FISH of two genes and co-expressed transposons. (A-D) *brat*(A), *MAX{5659*(B), *bt*(C) and *297{6* RNA expression patterns in total embryo preparations. The genotype of the sample is shown in the horizontal axis above, and the development stage of the sample is shown in the left vertical axis. The red pseudo-color is the signal from Probe and the green pseudo-color is the signal from nucleus. Scale bars, 80 μ m. RNA subcellular Location Patterns in mutant *Drosophila*. Scale bars, 30 μ m. (E-H) The RNA *brat*(E), *MAX{5659*(F), *bt*(G), *297{6*(H) relative fluorescence intensity of ISWI mutants and wild-type RNA signals was determined by comparing them with DAPI signals. The asterisk indicates $p < 0.05$ for the two-tailed student t test. (I) The ratio of transposon *MAX{5659* and *297{6* expression in ISWI mutants to that in wild-type at three stages of embryonic development.