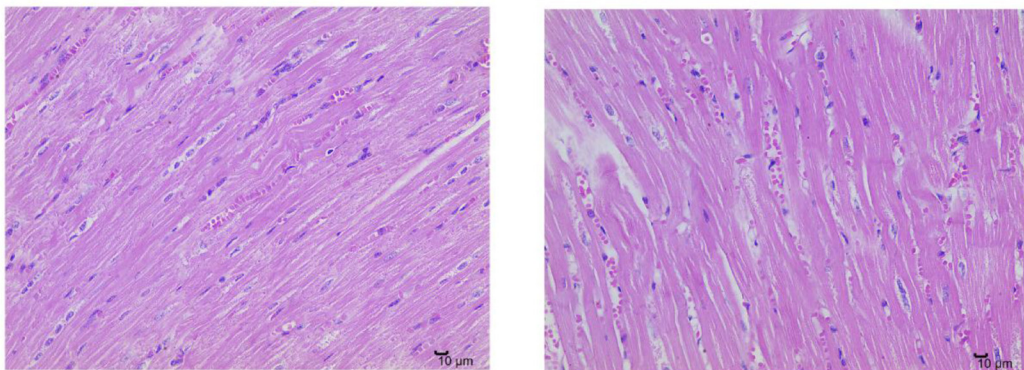


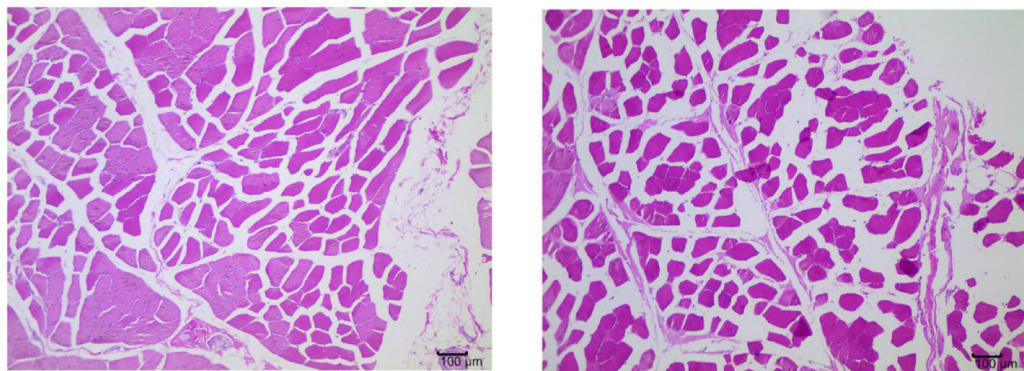
## Comprehensive transcriptional landscape of porcine cardiac and skeletal muscles reveals differences of aging

### SUPPLEMENTARY MATERIALS

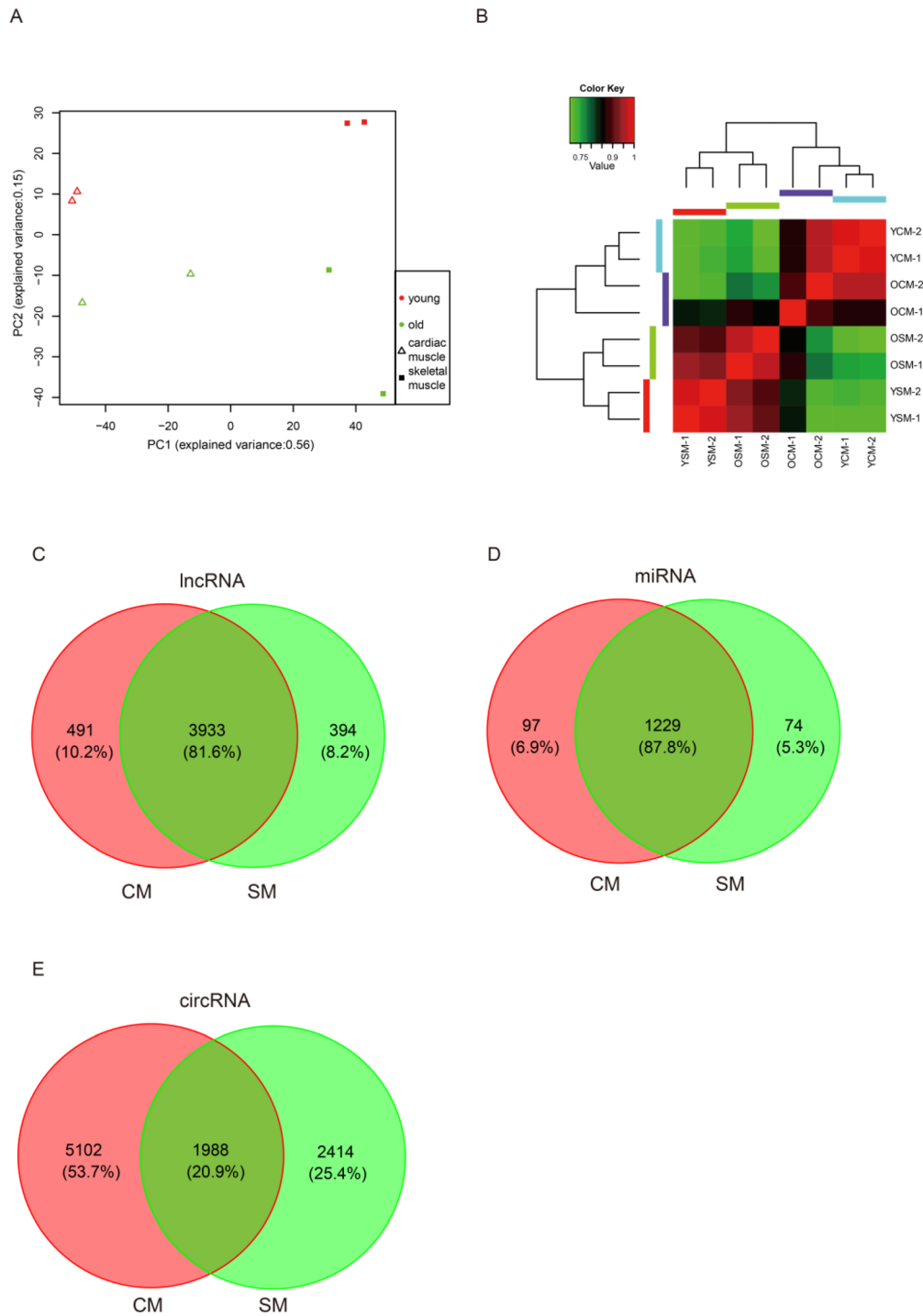
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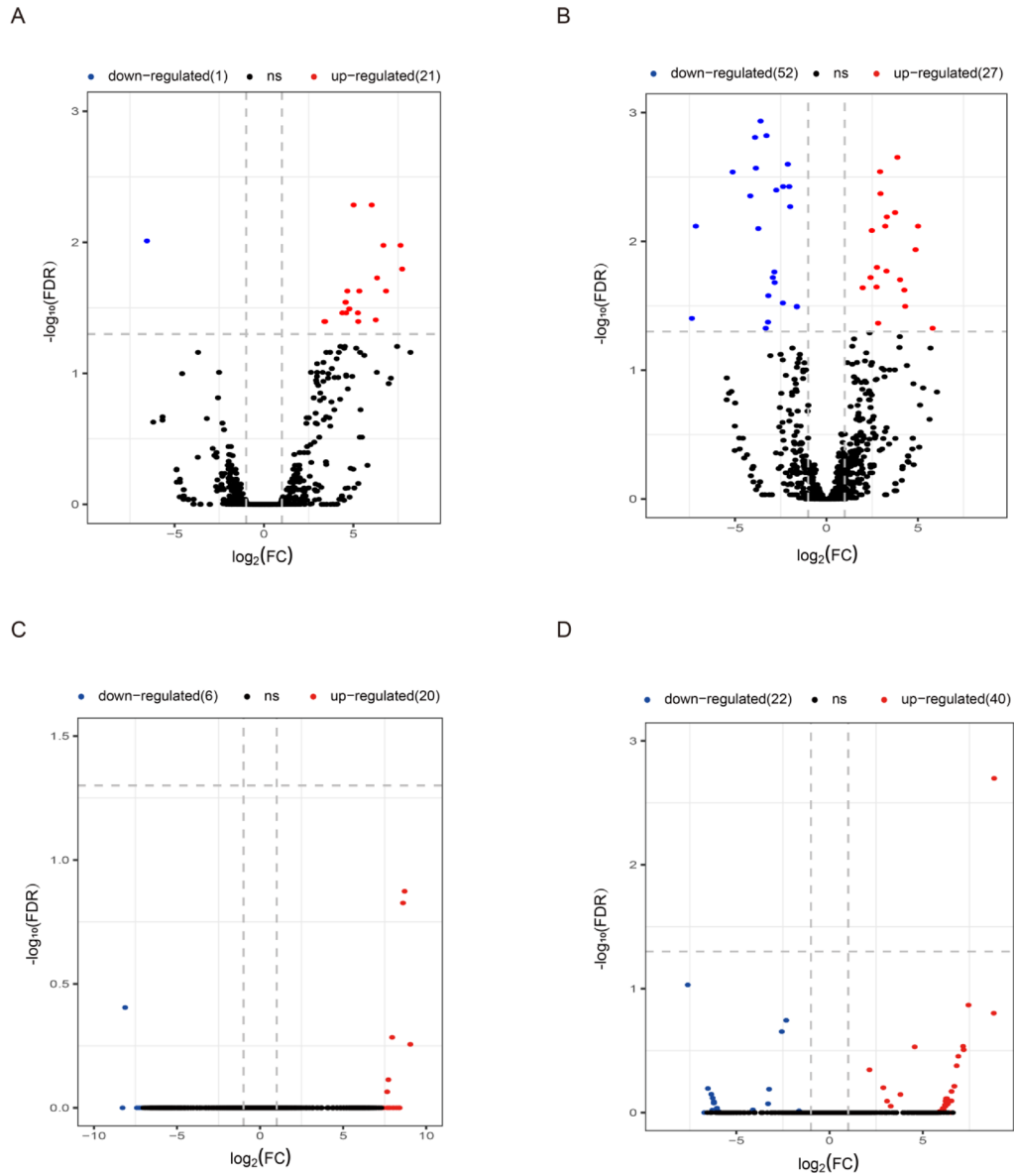
B



**Supplementary Figure 1: Hematoxylin and eosin staining of two muscle tissues at two different age stages.** (A) Left figure: longitudinal section of OCM (10 µm). Right figure: longitudinal section of OCM (B) Left figure: cross section of YSM (100 µm). Right figure: cross section of YSM (100 µm). “YCM” is short for “young cardiac muscle”, “OCM” is short for “old cardiac muscle”; “YSM” is short for “young skeletal muscle”, “OSM” is short for “old skeletal muscle”.

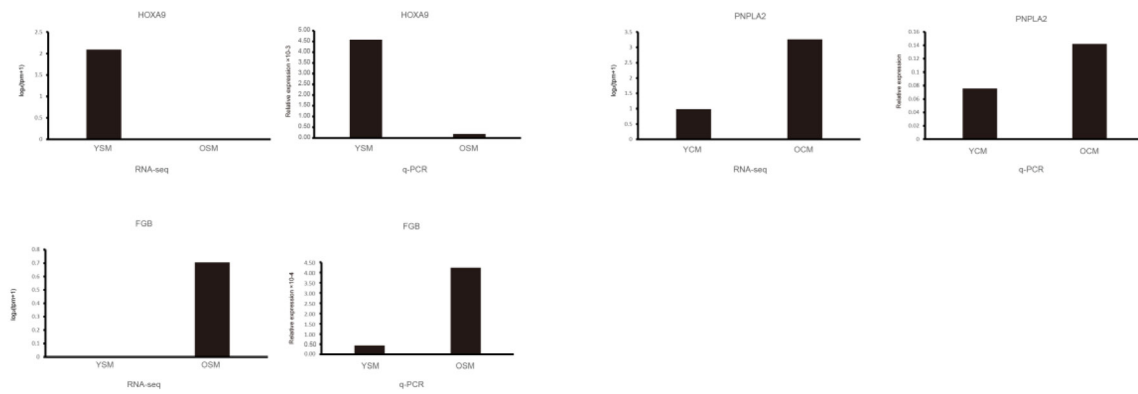


**Supplementary Figure 2: Global lncRNA, miRNA, and circRNA expression pattern across samples.** (A) Principal Component Analysis (PCA) plot based on normalized expression level ( $\log_2$  (TPM)) of identified miRNAs. (B) Heatmap of miRNA showing matrix of Pearson correlation coefficient between young and old replicate samples calculated from the variance in count values where values closer to 1 are less variable. Common expressed lncRNA (C), miRNA (D) and circRNA (E) in CM and SM. “YCM” is short for “young cardiac muscle”, “OCM” is short for “old cardiac muscle”; “YSM” is short for “young skeletal muscle”, “OSM” is short for “old skeletal muscle”; “CM” is short for “cardiac muscle”; SM” is short for “skeletal muscle”.

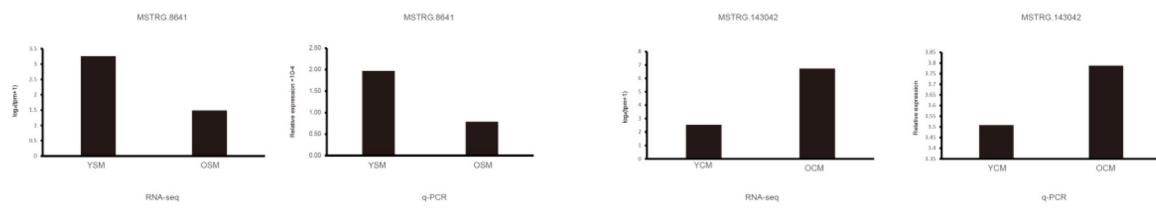


**Supplementary Figure 3: Differentially expressed miRNA and circRNA during aging.** (A) Differentially expressed miRNA in OYCM (old cardiac muscle VS young cardiac muscle), the red dots and blue dots respectively represent up-regulated and down-regulated mRNAs during aging. (B) Differentially expressed miRNA in OYSM (old skeletal muscle VS young skeletal muscle). (C) Differentially expressed circRNA in OYCM. (D) Differentially expressed circRNA in OYSM.

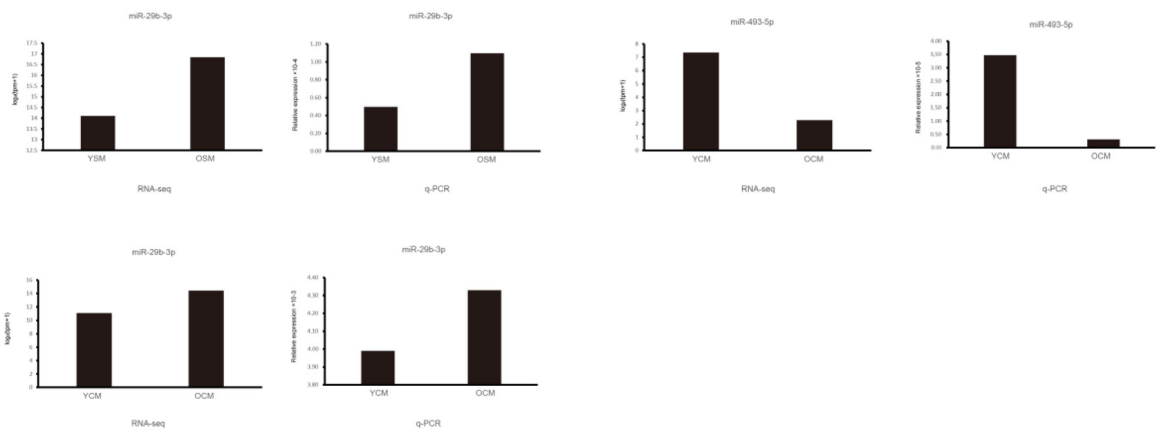
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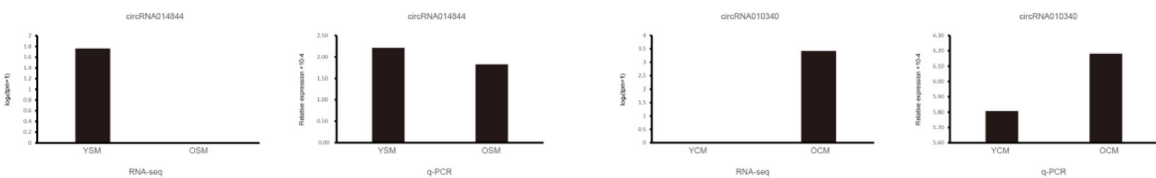
B



C



D



**Supplementary Figure 4: Validation of differentially expressed genes.** (A) Validation of selected differentially expressed mRNAs. (B) Validation of selected differentially expressed lncRNAs. (C) Validation of selected differentially expressed miRNAs. (D) Validation of selected differentially expressed circRNAs. “YCM” is short for “young cardiac muscle”, “OCM” is short for “old cardiac muscle”; “YSM” is short for “young skeletal muscle”, “OSM” is short for “old skeletal muscle”.

**Supplementary Table 1: Obtained clean data for RNA sequencing**

Sample ID	Reads number (M)	Data size (G)	Mapping rate (%)	GC (%)	Q30 (%)
YCM-1	62.86	9.43	65.5%	45	92.79
YCM-2	68.24	10.24	69.7%	46	92.79
OCM-1	75.58	11.34	67.2%	43	93.48
OCM-2	69.19	10.38	71.8%	49	95.68
YSM-1	83.96	12.59	68.5%	53	95.3
YSM-2	74.15	11.12	69.5%	53	95.31
OSM-1	80.19	12.03	70.6%	51	93.46
OSM-2	62.98	9.45	67.7%	54	92.71

**Supplementary Table 2: Obtained clean data for small RNA sequencing**

Sample ID	Reads number	Mapped reads	Mapping rate (%)	Q30 (%)
YCM-1	10,953,027	8,149,216	74.40	97.92
YCM-2	9,734,686	7,554,101	77.60	97.86
OCM-1	11,407,462	8,295,659	72.72	97.00
OCM-2	11,035,054	9,485,281	85.96	97.82
YSM-1	9,248,915	8,349,084	90.27	98.26
YSM-2	15,202,241	13,293,605	87.45	98.19
OSM-1	12,138,912	10,228,412	84.26	97.18
OSM-2	17,005,570	15,121,275	88.92	97.30

**Supplementary Table 3: Primer sequences for qPCR**

Genes	Forward	Reverse	Tm (°C)
HOXA9	5'-GCATTAACCTGAACCGCTGTC-3'	5'- AACTGGAGGAGAACCACAAGCA-3'	63
PNPLA2	5'- ATGTCATCATAACCCGCTTCG-3'	5'- GTTGTCTGAGATGCCACCGTC-3'	63
FGB	5'- GATCTCGCTGAGCAAATACAAGG-3'	5'- TCGTAGGTGCTGAAGAACATGCT-3'	63
GAPDH	5'- TTCCAGTATGATTCCACCCACG-3'	5'- CCATTTGATGTTGGCGGGAT-3'	63
ACTB	5'- CTGCGGCATCCACGAAACT-3'	5'- GTGATCTCCTTCTGCATCCTGTC-3'	63
MSTTRG.8641	5'- GCGCAGACAGAGCTATCCAG-3'	5'- AGGGTGTTCAAAGTAGTCAAGGGA-3'	63
MSTTRG.143042	5'- TGTCGGTTCTTCCCGCACT-3'	5'- CGTCCATCTTCCACCCCTCAT -3'	63
miR-29b-3p	5'- GGTAGCACCATTTGAAATCAGTGTT -3'	Universal primer supplied by kit	63
miR-493-5p	5'- GGCTTGACATGGTAGGCTTTCAT -3'	Universal primer supplied by kit	63
U6	5'-GGAACGATACAGAGAAGATTAGC-3'	5'-TGGAACGCTTCACGAATTTGCG-3'	63
circRNA014844	5'-AAACAAGCCATGGAGAGGTTACTAG-3'	5'-TCCGAGTGTCTTCCACAGAGT-3'	60
circRNA010340	5'-GTCTCTTGCAGCAACTCAG-3'	5'-AAGAAACGGAACAGTCACTCAAGA-3'	60

**Suppelemntary Dataset 1: Identified genes in our data.** See\_ [Suppelemntary\\_Dataset 1](#)

**Suppelemntary Dataset 2: Identified tissue-specific expressed genes in our data.** See\_ [Suppelemntary\\_Dataset 2](#)

**Suppelemntary Dataset 3: Clustering of special expressed genes in cardiac muscle and skeletal muscle.** See\_ [Suppelemntary\\_Dataset 3](#)

**Suppelemntary Dataset 4: Differentially expressed genes during cardiac muscle and skeletal muscle aging.** See\_ [Suppelemntary\\_Dataset 4](#)

**Suppelemntary Dataset 5: Gene Ontology analysis for clustering of special expressed genes.** See\_ [Suppelemntary\\_Dataset 5](#)

**Suppelemntary Dataset 6: Gene Ontology analysis for differential expression genes.** See\_ [Suppelemntary\\_Dataset 6](#)

**Suppelemntary Dataset 7: Gene Ontology analysis for differentially expressed lncRNAs.** See\_ [Suppelemntary\\_Dataset 7](#)

**Suppelemntary Dataset 8: Co-expression of differentially expressed miRNAs and differentially expressed circRNAs.** See\_ [Suppelemntary\\_Dataset 8](#)