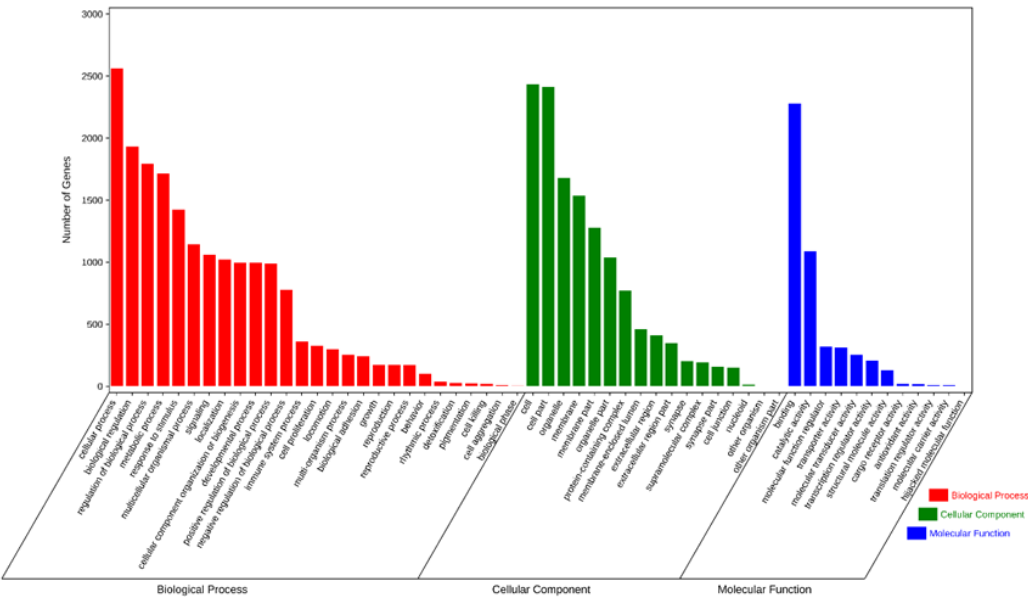
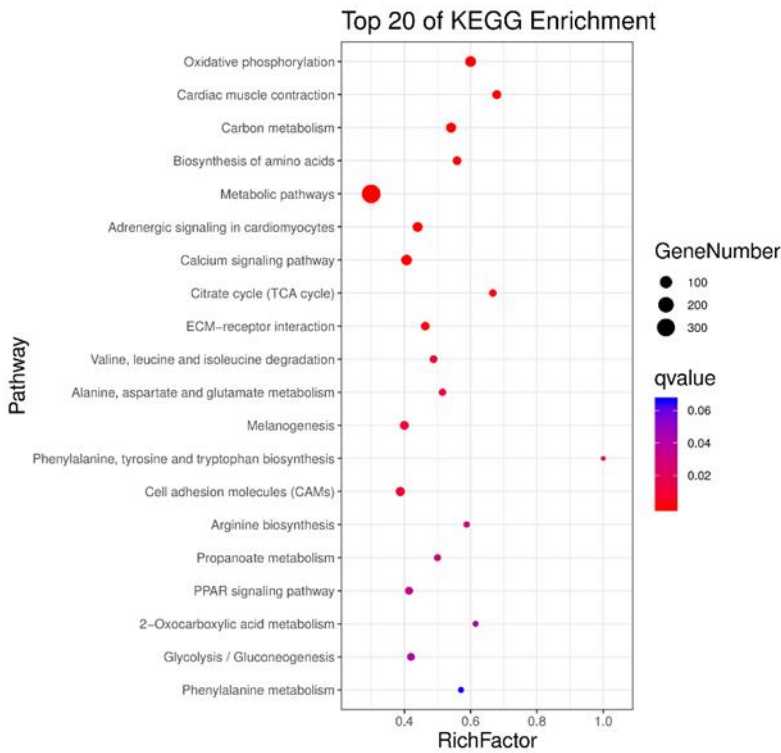


Supplementary files

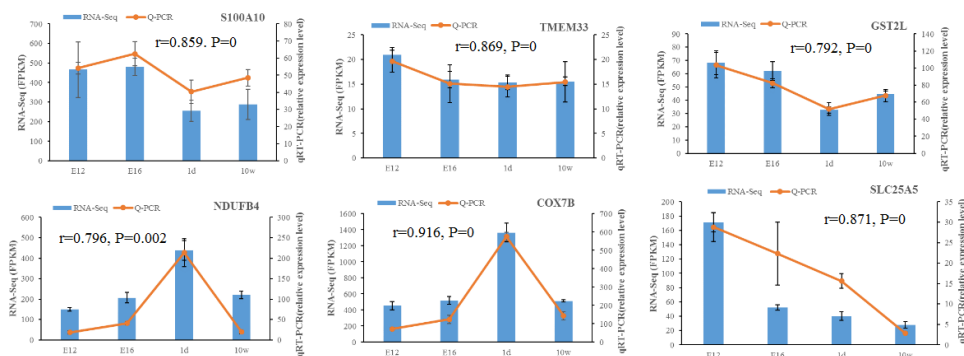


**Figure S1.** Gene Ontology enrichment analysis of all the differentially expressed genes in male chickens.



**Figure S2.** The top 20 enriched pathways of DEGs in male chicken.

The enrichment analysis of KEGG pathway of the top 20 differential genes. Rich Factor, the higher the value, the greater the enrichment degree. The color depth represents the P-value, and the darker the color, the more significant the difference. The dot size represents the number of genes enriched in the pathway.



**Figure S3.** The validation of differentially expressed genes through quantitative reverse-transcription-PCR.

R represents correlation, P represents value judgment. |r| value, the greater the correlation, the better. Present positive correlation between positive said, negative correlation between negative said. The smaller the P value, the more significant the difference.

**Table S1.** Primer Sequences for quantitative reverse-transcription PCR

Gene name	Primer sequence (5'-3')	Product (bp)	Annealing (°C)
S100A10	TTCCACAAATACGCGGGTGA	186	60
	AGCCACCAGTGAGAAGAAGC		
COX7B	TATGGAAATCTGGTGCTGATCG	155	60
	CAGTTCATTGCAAGCAAAGGC		
SLC25A5	TACCAGGGCTTCAACGTGTC	186	60
	CCGAACGGTATCGAAGGGAT		
NDUFB4	AACCGCTACGTTTCGTTGCC	218	60
	CGGAAAGTGGGGTACACGTT		
GST2L	TGCTGGAGAACCAGGTGATG	148	60
	AACCACTTTCGTGCCCCCAG		
TMEM33	GACACAGCAATGTGGATTTC	149	60
	CTTGGTGTAAGTCGGAGAGCA		
β-actin	CAGCCATCTTCTTGGGTAT	169	60
	CTGTGATCTCCTTCTGCATCC		
HSP70	TCTGCTCCTGTTGGATGTC	95	60
	TGGGAATGGTGGTGTACG		
ALOX5	GCGTGGATGCCAATAAGAC	253	60
	GAGACTAGGTGTGTCGTAAGAG		
MYOD	GCTACTACACGGAATCACCAAAT	200	60
	CTGGGCTCCACTGTCACTCA		
MEF2C	GGAGGATACCCATCAGCCAT	124	60
	AGCCAGTCACAGAACCAAGA		

MYOG	CGGAGGCTGAAGAAGGTGAA CGGTCCTCTGCCTGGTCAT	320	60
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**Table S2.** The results of RNA quality detection

Sample name	RIN	OD260/280	OD260/230	28S/18S
E12-M1	7.6	1.974	2.133	0.8
E12-M2	7.4	1.967	2.068	0.7
E12-M3	8.4	1.991	2.009	1.5
E16-M1	9.2	1.948	1.887	1.2
E16-M2	9.5	1.955	2.227	1.3
E16-M3	9.5	1.922	1.703	1.4
1d-M1	9	1.885	1.989	1.1
1d-M2	9.3	1.959	2.204	1.2
1d-M3	8.9	1.954	2.076	1.2
10W-M1	8.6	1.881	1.82	1.1
10W-M2	9.1	1.921	2.165	1.1
10W-M3	8.8	1.875	2.283	1.2

**Table S3** The results of comparison with reference genome for clean reads

Sample name	Total reads	Total mapped	Multiple mapped	Uniquely mapped
E12-M1	61790372	53387806(86.4%)	22012191(3.56%)	51186515(82.84%)
E12-M2	65909504	56686510(86.01%)	2576316(3.91%)	54110194 (82.1%)
E12-M3	64494886	54579542 (84.63%)	3880488 (6.02%)	50699054 (78.61%)
E16-M1	56999228	50121611 (87.93%)	1901087 (3.34%)	48220524 (84.6%)
E16-M2	52273528	45470227 (86.99%)	1718286 (3.29%)	43751941 (83.7%)
E16-M3	55850416	48895846 (87.55%)	1934697 (3.46%)	46961149 (84.08%)
1d-M1	61428646	53389831 (86.91%)	1904079 (3.1%)	51485752 (83.81%)
1d-M2	69820058	60119337 (86.11%)	2243311 (3.21%)	57876026 (82.89%)
1d-M3	64664226	56528888 (87.42%)	1909429 (2.95%)	54619459 (84.47%)
10W-M1	63540482	54672509 (86.04%)	2112197 (3.32%)	52560312 (82.72%)
10W-M2	54365192	47684092 (87.71%)	1541352 (2.84%)	46142740 (84.88%)
10W-M3	64191268	54433294 (84.8%)	2252817 (3.51%)	52180477 (81.29%)

Note: (1) Total reads: The number of sequences filtered by sequencing data. (2) Total mapped: Statistics on the number of sequenced sequences that can be located on the genome. (3) Multiple mapped: The number of sequencing sequences with multiple alignment locations on the reference sequence. This percentage is usually less than 10%. (4) Uniquely mapped: The number of sequencing sequences with unique alignment locations on the reference sequences is counted.