

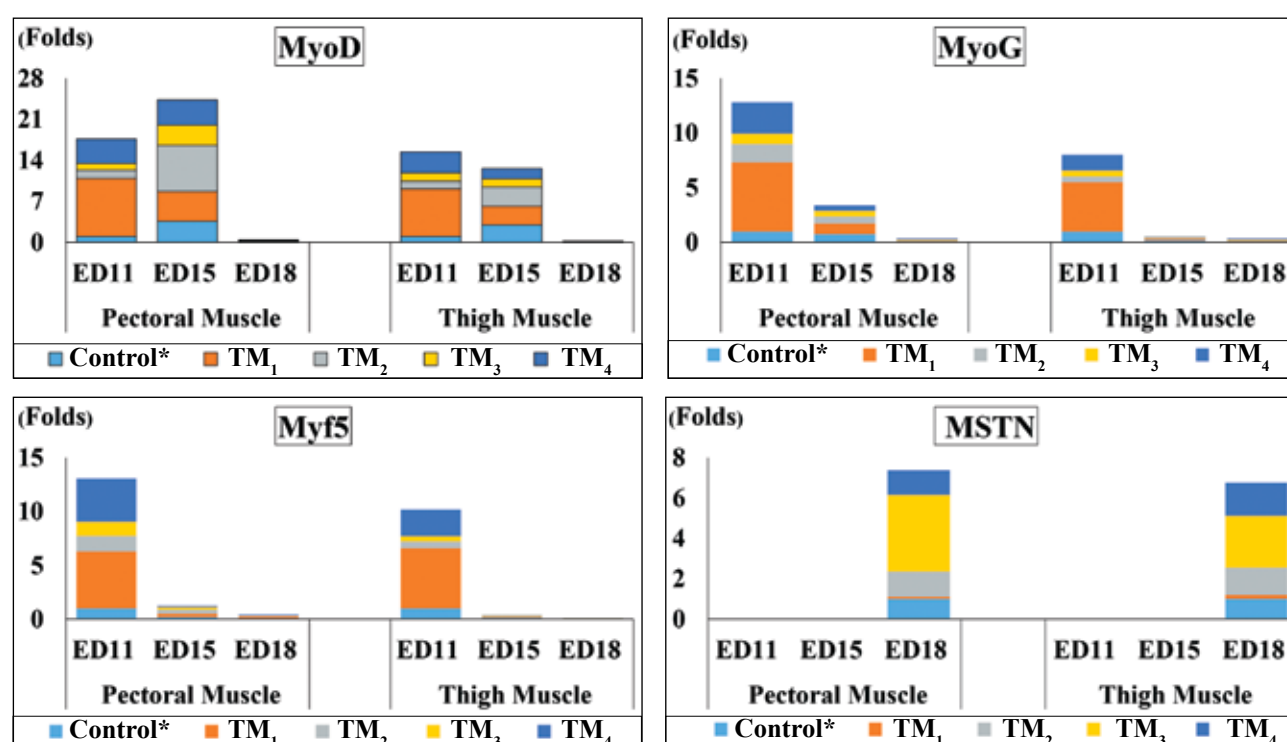


Effect of thermal manipulation during embryogenesis on gene expression of myogenic regulatory factors pre and post hatch in broilers

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Supplementary Fig. 1. Clustered Stacked Chart of relative normalized expression of Myf5, MyoD, MyoG and MSTN from ED11-ED18. Control*, 37.8°C; TM₁, Thermal manipulation from ED7-11 at 39°C for 18 h; TM₂, Thermal manipulation from ED11-15 at 39°C for 18 h; TM₃, Thermal manipulation from ED15-18 at 39°C for 18 h; TM₄, Thermal manipulation from ED7-18 at 39°C for 18 h.

Supplementary Table 1. Oligonucleotides for gene expression analysis by Real time-PCR amplification

Primer	Sequence	GenBank Reference
Myf5	F-5-CAACCCCAACCAGAGACTCC-3 R-5-TCCCGGCAGGTGATAGTAGT-3	NM_001030363.1
MyoD	F-5-TACCCAGTGCTGGAGCACTA-3 R-5-GTCTTGGAGCTTGGCTG AACT-3	NM_001004384.2
MyoG	F-5-AGCAGCCTCAACCAGCAGGA-3 R-5-TCTGCCTGGTCATCGCTCAG-3	NM_204359.2
MSTN	F-5-ATGCAAAAGCTAGCAG TC-3 R-5-TCATGAGCACCCGCAACGAT-3	NM_204170.2
GAPDH	F-5-GTGTATCATCTCAGCTCCCTCAG-3 R-5-GGTCATAAGACCCTCCACAATG-3	FJ_217667
ACTA1	F-5-CCATCGGCAATGAGCGTTTC-3 R-5-GCATGCGGTCAGCAATACCT-3	NM_001031063.1

Supplementary Table 2. Relative normalized expression of mRNA levels of MSTN in pectoral and thigh muscles at embryonic days 11, 15 and 18

MSTN	ED11		ED15			ED18		
	Expression (fold)		Expression (fold)			Expression (fold)		
	Pectoral	Thigh	Pectoral	Thigh		Pectoral	Thigh	
Control*	N/A	N/A	Control*	N/A	N/A	Control*	1.0±0.6a	1.0±0.5a
TM ₁	N/A	N/A	TM ₁	----	----	TM ₁	N/A	1.09±0.1a
TM ₂	----	----	TM ₂	N/A	N/A	TM ₂	1.16±0.7a	1.35±0.2a
TM ₃	----	----	TM ₃	----	----	TM ₃	5.80±0.4b	2.59±0.8b
TM ₄	----	----	TM ₄	----	----	TM ₄	1.23±0.3a	0.64±0.2a

Control, 37.8°C; TM₁, Thermal manipulation from ED7-11 at 39°C for 18 h; TM₂, Thermal manipulation from ED11-15 at 39°C for 18 h; TM₃, Thermal manipulation from ED15-18 at 39°C for 18 h; TM₄, Thermal manipulation from ED7-18 at 39°C for 18 h. a–c, Within the same day, means ± SD with different superscripts differ significantly (p<0.05). *, Compared to the control of each embryonic day (p<0.05).