

Garcinol Promotes the Formation of Slow-Twitch Muscle Fibers by Inhibiting p300-Dependent Acetylation of PGC-1 α

Online Supplementary Material

Supplemental Table S1

Ingredient and chemical composition of the diets

Item	
Ingredient, %	
Corn	67.0
Soybean meal, dehulled	14.0
Wheat bran	15.6
Fish meal	0.5
Soybean oil	-
Dicalcium phosphate	1.2
Limestone	0.8
L-Lysine HCl (78%)	-
Salt	0.38
Premix ¹	0.52
Total	100
Calculated composition	
Gross energy, GE, MJ/kg	14.21
Crude protein, CP, %	21.7
Fat, %	9.4
Ca, %	0.95
Total P, %	0.61
Analyzed composition	
Metabolizable energy, ME, MJ/kg	12.04
Crude protein, CP, %	20.5
Fat, %	7.6
Ash, %	5.4
Ether extract, EE, %	1.7

¹Premix supplied the following per kilogram of diet: 1) cobalt (as cyanocobalamin), 0.6; 2) copper (as CuSO₄ · 5H₂O), 5.0; 3) iodine (as CaI₂), 0.48; 4) iron (as FeSO₄), 75.0; 5) manganese (as MnO), 20.0; 6) selenium (as Na₂SeO₃), 0.40; and 7) zinc (as ZnO), 10.0; 8) all-rac- α -tocopheryl acetate, 64.0; 9) D-biotin, 0.2; 10) calcium

d-pantothenate, 24.0; 11) cholecalciferol, 5.5; 12) folic acid, 6.0; 13) menadione sodium bisulfite, 2.2; 14) nicotinic acid, 30.3; 15) pyridoxine-HCl, 12.0; 16) retinyl acetate, 1.9; 17) riboflavin, 5.5; 18) thiamin-HCl, 13.0; and 19) vitamin B-12, 0.022.

Supplemental Table S2 Name, type, dilution, and source of primary antibodies¹

Antibody	Isotype	Dilution	Source
p300	Rabbit	1:1000	Cell Signaling Technology (Boston, MA, USA)
PGC-1 α	Rabbit	1:2000	Cell Signaling Technology (Boston, MA, USA)
Sirt1	Rabbit	1:2000	Cell Signaling Technology (Boston, MA, USA)
Slow MyHC	Rabbit	1:1000	Abcam (Cambridge, MA, USA)
Fast MyHC	Rabbit	1:1000	Abcam (Cambridge, MA, USA)
ACTB	Rabbit	1:2000	Cell Signaling Technology (Boston, MA, USA)
Secondary Antibody			
HRP labelled Antibody	Goat anti rabbit	1:10,000	Santa Cruz (Santa Cruz, CA, USA)

¹PGC-1 α , peroxisome proliferator-activated receptor- γ coactivator-1 α ; Sirt1, Silent information regulator 1; Slow MyHC, slow myosin heavy chain; Fast MyHC, fast myosin heavy chain; ACTB, β -Actin.

Supplemental Table S3 Primers used for real-time quantitative PCR¹

Gene	Primer sequence (5'–3')	Length, bp
MyHCI	F:AAGGGCTTGAACGAGGAGTAGA R:TTATTCTGCTTCCTCCAAAGGG	115
MyHCIIa	F: GCTGAGCGAGCTGAAATCC R: ACTGAGACACCAGAGCTTCT	137
MyHCIIx	F: AGAAGATCAACTGAGTGAAC R: AGAGCTGAGAACTAACGTG	149
MyHCIIb	F: ATGAAGAGGAACCACATTA R: TTATTGCCTCAGTAGCTTG	166
Myoglobin	F: CACCATGGGGCTCAGTGATG R:CTCAGCCCTGGAAGCCTAGC	143
TnI slow	F: AGTTGAGAGGAAATCCAAGAT R:CTTCAGCTTCAGGTCCTTGAT	125
TnI fast	F:TCAAGGATAAGGGCGACAGC R:TGTCCATGGACGCAGCTCTT	126
18srRNA	F:CCCACGGAATCGAGAAAGAG R:TTGACGGAAGGGCACCA	122

¹MyHCI, Myosin heavy chain I; MyHCIIa, Myosin heavy chain IIa; MyHCIIx, Myosin heavy chain IIx; MyHCIIb, Myosin heavy chain IIb; TnI slow, Troponin I slow; TnI fast, Troponin I fast.

F: forward primer; R: reverse primer

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Figure S1. Dose-response curves for C2C12 myotubes to garcinol

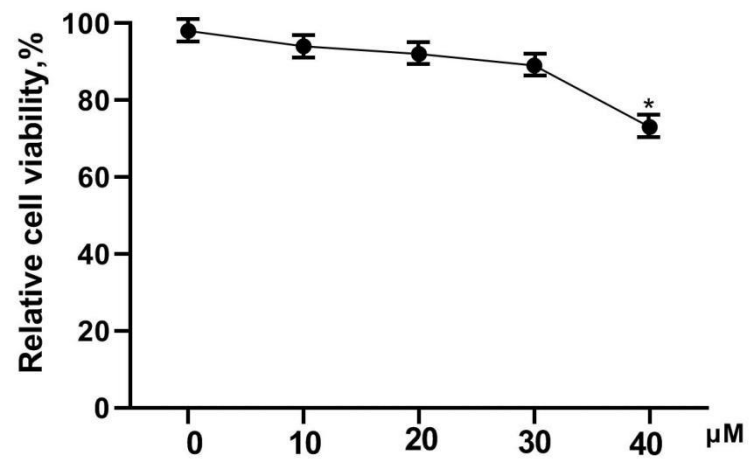


Figure S2. Time-response curve of C2C12 myotubes to garcinol

