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Supplemental Information

Molecular and Functional Networks Linked to Sarcopenia Prevention by Caloric Restriction in Rhesus Monkeys

Timothy W. Rhoads, Josef P. Clark, Grace E. Gustafson, Karl N. Miller, Matthew W. Conklin, Tyler M. DeMuth, Mark E. Berres, Kevin W. Eliceiri, Laura K. Vaughan, Christine W. Lary, T. Mark Beasley, Ricki J. Colman, and Rozalyn M. Anderson

Supplemental Figure Legends.

Fig. S1, related to Table 1. Longitudinal upper leg lean mass for study animals. The upper leg lean mass as a fraction of body weight (*left*) or total upper leg lean mass in grams (*right*), from DEXA measurements is presented as a function of either time on study (*top*) or animal age (*bottom*). A polynomial regression is fit to each diet group, with 95% confidence intervals denoted by the shaded regions. N = 5 for control and 8 for CR.

Figure S2, related to Figure 1. Chr7 miRNAs and putative gene targets statistically different between control and CR. Network diagram depicting miR members of the Chr7 locus and putative targets identified by TargetScan that were also statistically significant between control and CR.

Fig. S3, related to Fig. 2. Relative expression of lipid metabolism genes. Box plots of delta Ct values for lipid metabolism genes involved in: Fatty acid biosynthesis (*ACACA*, *FASN*, *SCD*), beta-oxidation (*CPT1A*, *ACADL*), lipid droplet metabolism (*PLIN2*, *PLIN3*, *PLIN4*, *PLIN5*), and lipid biosynthesis (*DGAT2*, *GPAT4*). 18S rRNA is used as the reference gene. N = 5 for control and 8 for CR.

Figure S4, related to Figure 4. Activity counts. Accelerometer counts for morning (*left*), afternoon (*middle*), and night (*right*) time periods. N = 5 for control and 8 for CR (box – Inter-quartile range, whiskers – minimum and maximum values, bar – median, small box – mean).

Figure S5, related to Figure 4. Regression analysis of skeletal muscle mass. Linear regressions of skeletal muscle mass measures with age and activity counts; ESM % of peak vs. age (*top*), ESM % of peak vs. 24 hour accelerometer counts (*middle*), and UL Lean % of peak vs. night accelerometer counts (*bottom*).

Fig. S1, related to Table 1

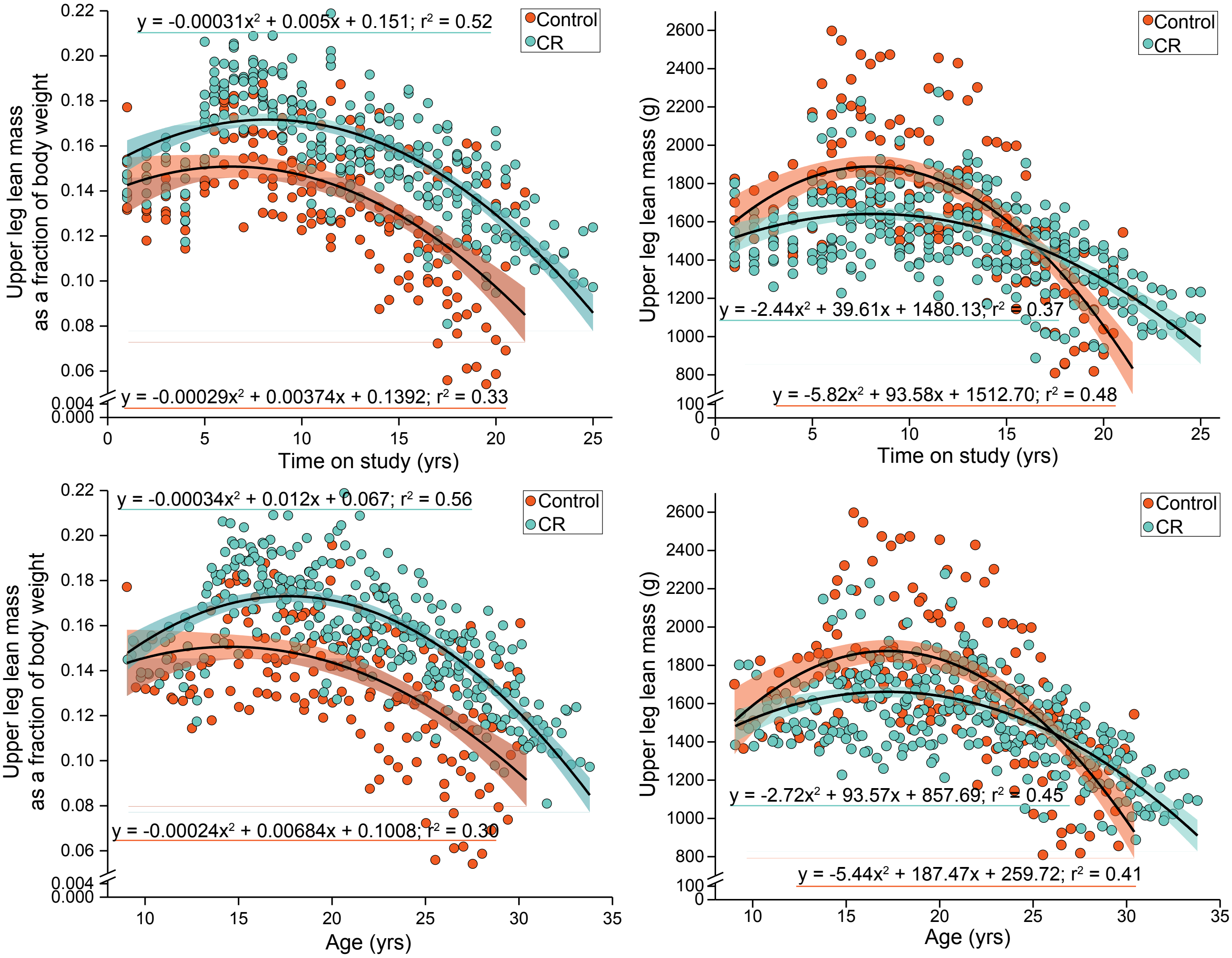


Fig. S2, related to Fig. 1

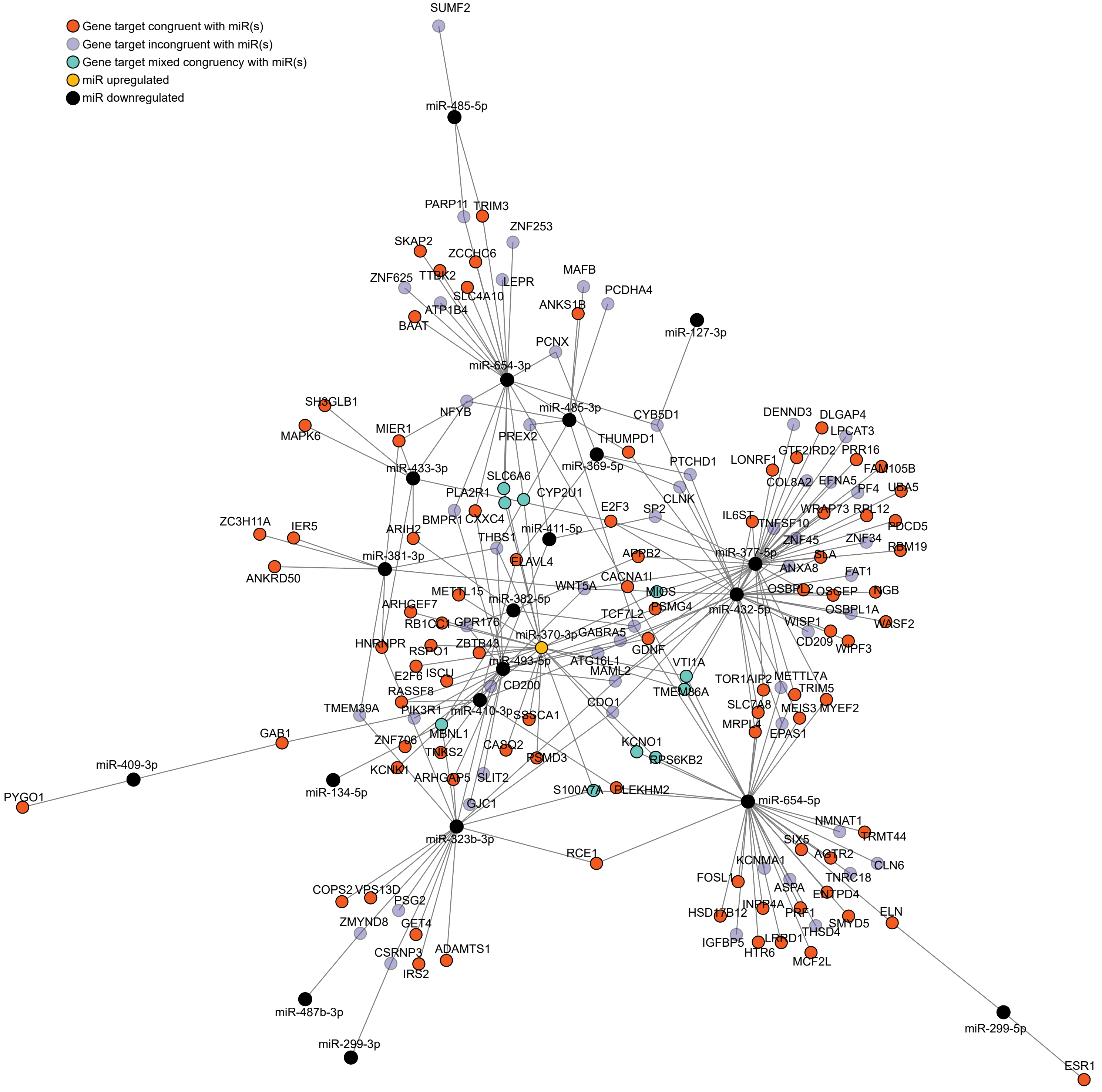
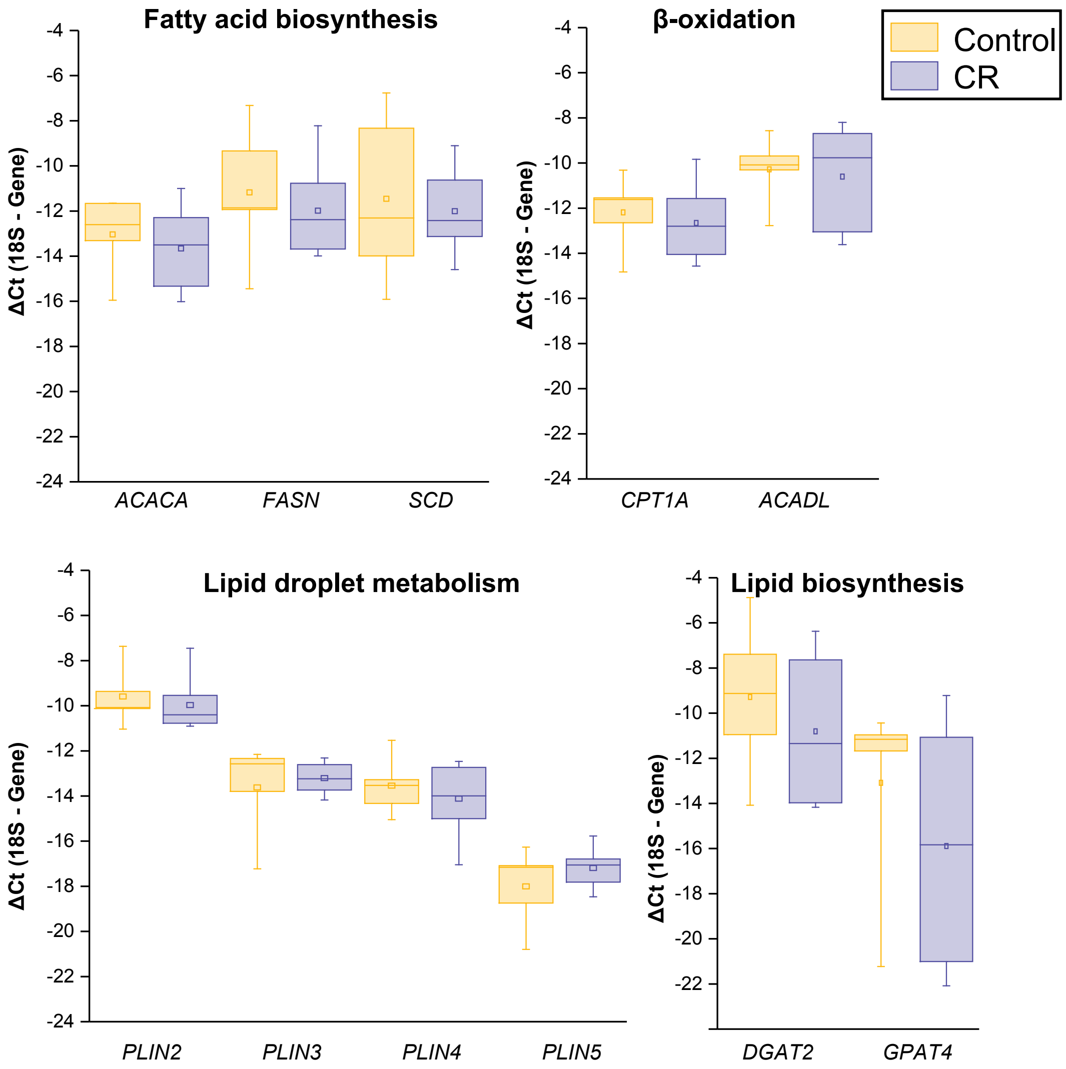


Fig. S3, related to Fig.2



Fatty acid biosynthesis

β-oxidation

Lipid droplet metabolism

Lipid biosynthesis

Fig. S4, related to Fig. 4

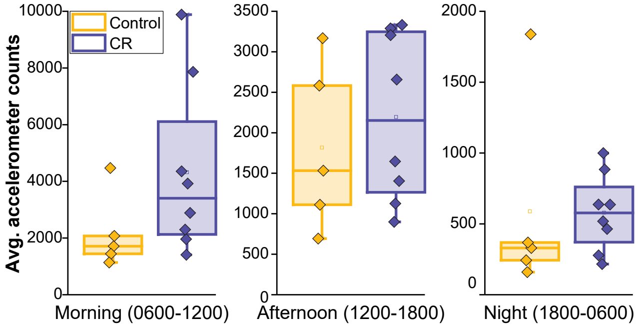


Figure S5, related to Figure 4

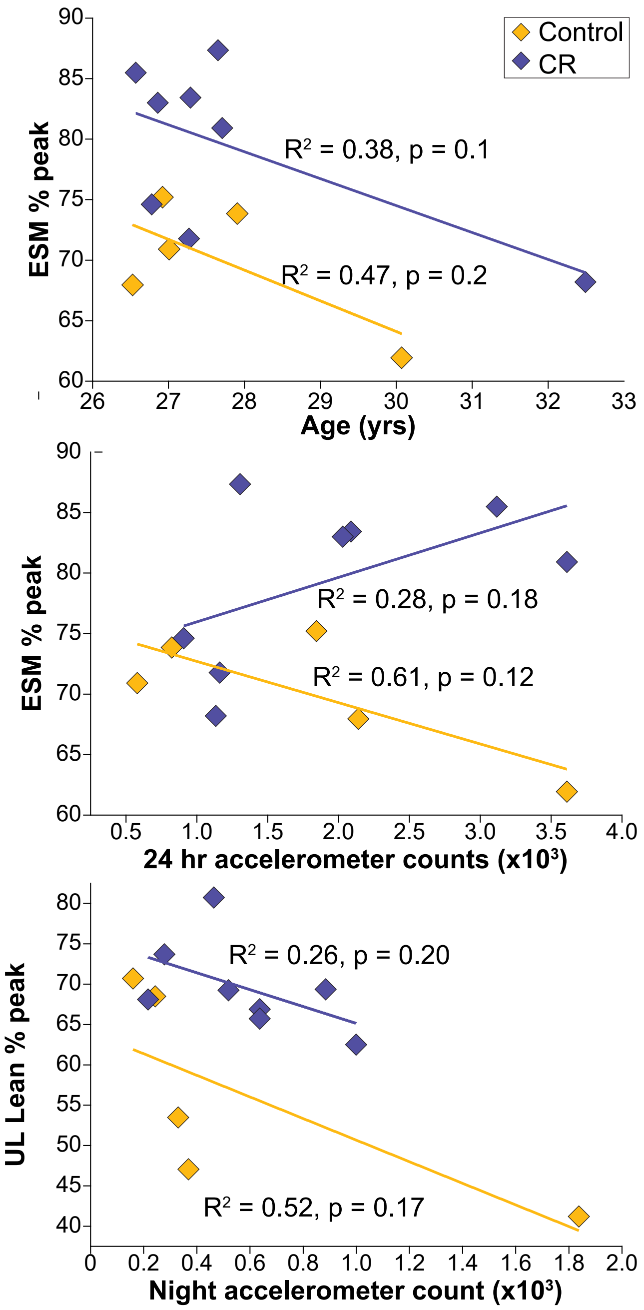


Table S1, related to Figure 1; Microarray analysis

Formatted as excel sheet with 3 tabs:

- BAM genes
- PAGE pathways
- IPA details

Table S2, related to Figure 1; MicroRNA analysis

Formatted as excel sheet with 2 tabs:

- All miRs
- Families/loci of interest

Table S3, related to Figure 2; 2-photon microscopy lifetime components

Lifetime component	Control mean	CR mean	F-stat	p-value
τ_M (ps)	1024.603 ± 86.650	924.025 ± 41.280	305.210	< 0.0001*****
τ_1 (ps)	406.735 ± 32.389	383.676 ± 30.844	36.26	< 0.0001*****
τ_2 (ps)	2328.184 ± 67.003	2220.502 ± 93.971	40.31	< 0.0001*****
a_1 (%)	67.856 ± 2.934	70.681 ± 0.622	187.62	< 0.0001*****

All data is presented as sample mean ± standard deviation

*****$p < 0.05$

Table S4, related to Figure 3; Fiber histology parameters

Measure	Control	CR	Fstat	p-value
Fiber size				
Type I (pixel^2)	48369.551 ± 24057.213	46543.754 ± 16048.230	.02	0.882
Type II (pixel^2)	45612.918 ± 23128.762	68979.970 ± 34874.971	1.77	0.186
MMF (pixel^2)	58394.446 ± 20459.225	95402.143 ± 46398.013	3.41	0.067
Cyt c Ox intensity				
Type I	**42.757 ± 13.602**	**70.353 ± 17.075***	**5.39**	**0.022**
Type II	48.697 ± 19.863	45.000 ± 15.864	0.08	0.780
MMF	18.778 ± 9.593	15.927 ± 7.529	0.84	0.361
Intensity localization				
Mid	118.149 ± 17.010	138.272 ± 18.216		0.173
Perimeter	**152.509 ± 24.023** **p-value < 0.0001*** **Control mid vs peri**	**187.790 ± 27.730*** **p-value < 0.0001*** **CR mid vs peri**		**0.021**
% non-contractile area	19.806 ± 12.292	12.214 ± 3.592	13.26	0.005

Table S5, related to Figure 4. P values for differences in variance between diet groups across biometric measures

Biometric	*p-value*
Body weight (g)	**4.39×10^{-3}**
Total lean mass (g)	0.75
% fat	0.05
ESM as % of peak	0.34
UL lean as % of peak	**0.01**
Basal glucose (mg/dL)	0.12
Basal insulin (μU/mL)	0.09
Insulin sensitivity index; Si ($x10^{-4}$)	0.23
Metabolic cost of movement ($x10^{-4}$)	**0.03**