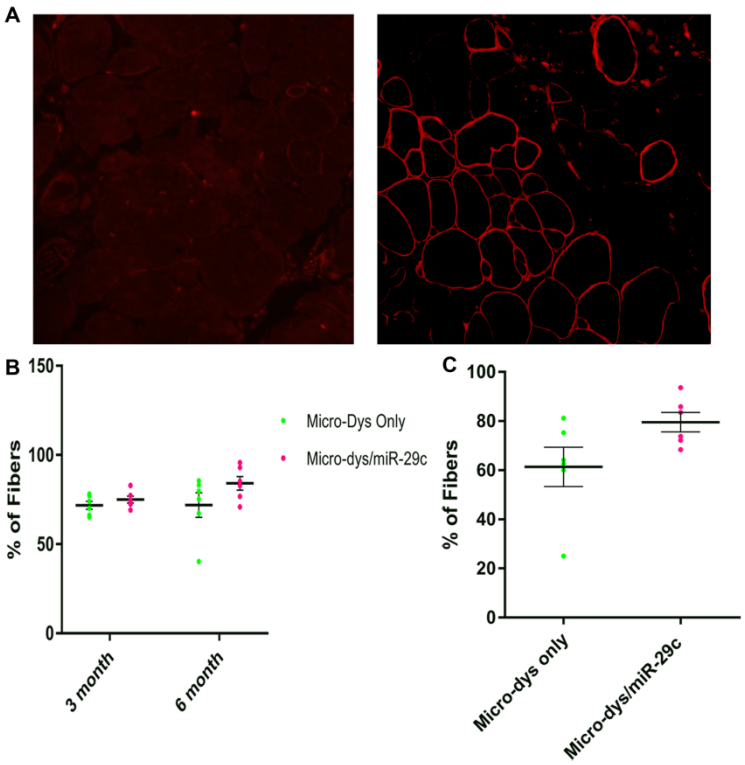
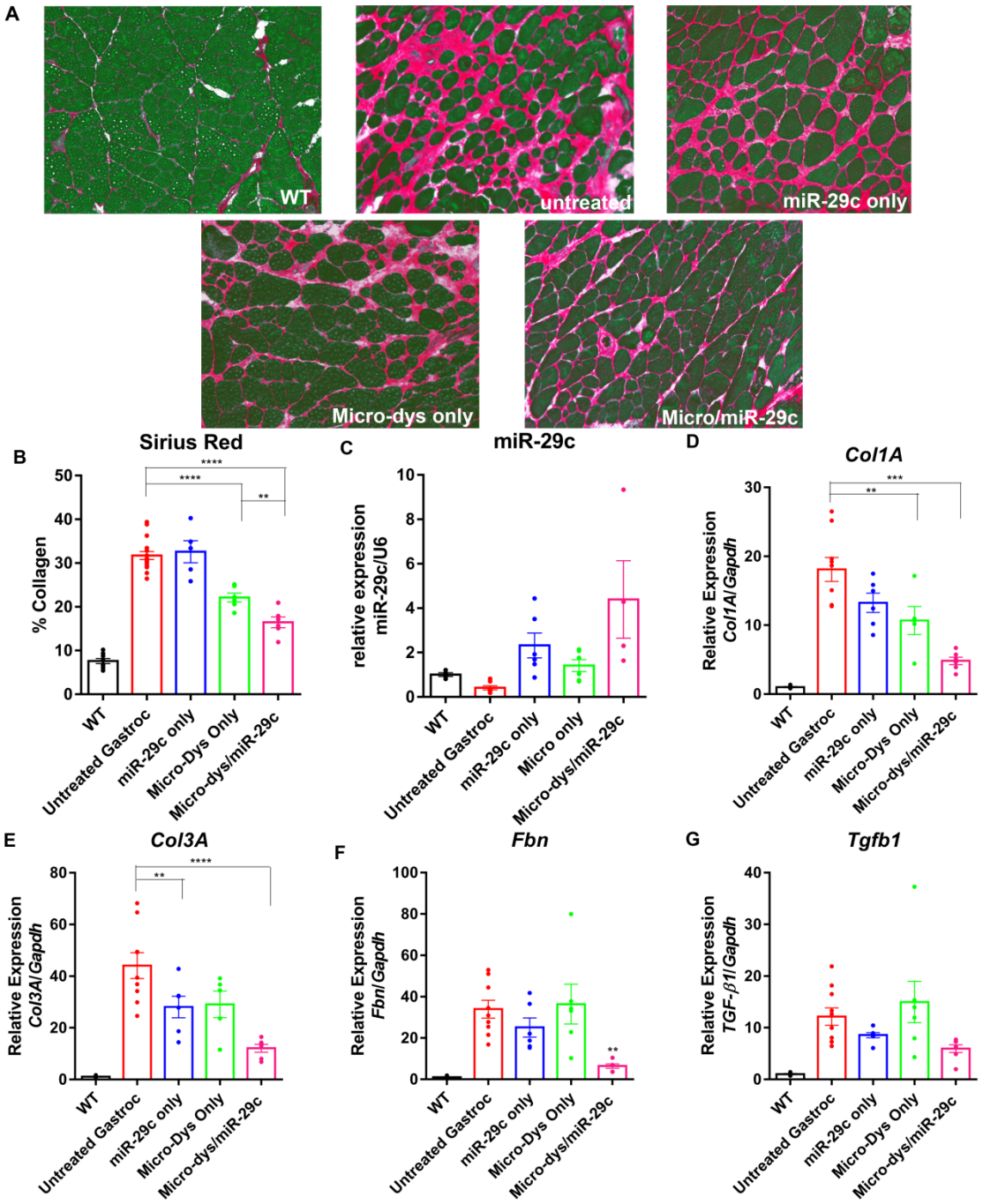
Supplemental Figures

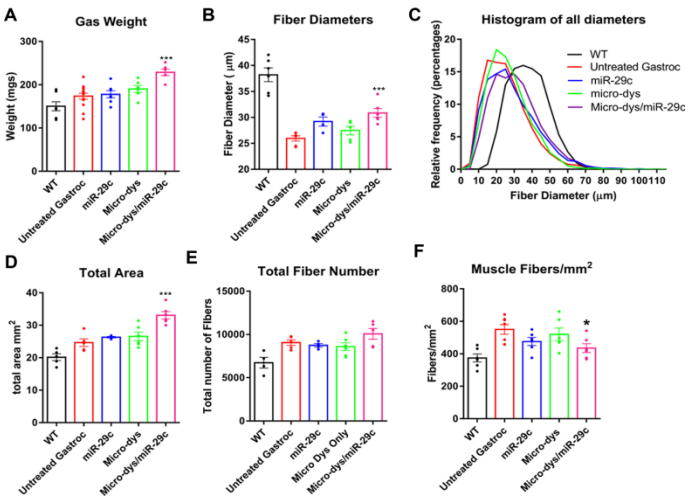


Supplemental Figure 1. (A) Flag staining on untreated muscle (B) micro-dystrophin expression in an injected GAS muscle with AAV.MCK.micro-dystrophin. Original magnification, x20. (C) Quantification of the amount of fibers expressing micro-dystrophin in the late treatment mice (D) quantification of the amount of fibers expressing micro-dystrophin in the early treatment mice. All timepoints showed no difference between micro-dystrophin expression in each of the groups receiving AAV.micro-dystrophin (n=6 for all groups).

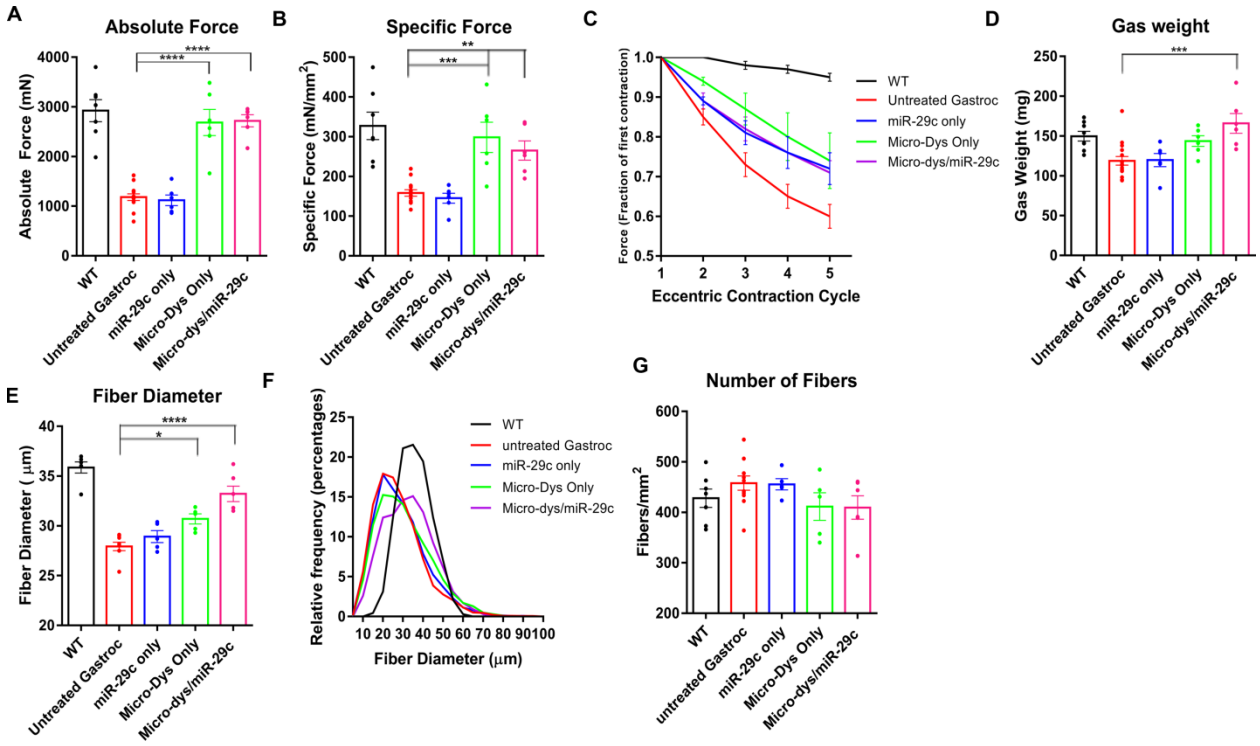


Supplemental Figure 2. (A) Picrosirius red staining of wild-type, untreated, AAV.miR-29c, AAV.micro-dystrophin, and AAV.miR-29c/AAV.micro-dystrophin twenty-four weeks post-injection. Original magnification, x20. (B) Quantification of picrosirius red staining shows co-treated muscle had a 46.9% reduction in collagen compared to untreated GAS muscle. (n=5-8 for all groups except n=15 for untreated), One-way ANOVA (C) qRT-PCR confirms an increase in miR-29c transcript levels in the treated cohorts. (n=4-7 for all groups).Semi-quantitative qRT-PCR shows a significant reduction in *Col1A1* and *Col3A1* (D, E), *Fbn* (F) and *Tgfb1* (G) levels in the AAV.miR-29c/AAV.micro-

dystrophin treated muscle compared to the contralateral limb and each of the single therapies (n=6). (D-G n= 5-6 for all groups except n=9 for untreated), One-way ANOVA. All data represent mean ± SEM. (**p<0.01, ***p<0.001, ****p<0.0001)



Supplemental Figure 3. Combination treatment increases muscle hypertrophy 3 months post injection. (A) miR-29c co-delivered with micro-dystrophin increased the overall weight of the injected GAS compared to either one injected alone (n=6-7 per group except n=14 for untreated), One-way ANOVA. (B) miR-29c/micro-dystrophin combination treatment demonstrated an increase in average fiber size. Comparing *mdx*/utrn$^{+/-}$ controls with miR-29c/micro-dystrophin treated *mdx*/utrn$^{+/-}$, the average diameter increased from 25.96 to 30.97µm (n=4-6 per group), One-way ANOVA. (C) The co-delivery produced a shift towards wild-type fiber size distribution. (D) We then measured total cross-sectional area of the muscle. GAS muscles from all groups were full slide scanned and the total area was measured. Muscles co-treated with micro-dystrophin/miR-29c had a significant increase in cross sectional area compared to untreated and either treatment alone (untreated: 24.6 vs. miR-29c: 26.3 vs. micro-dystrophin: 26.6 vs. micro-dystrophin/miR-29c: 33.1). (E) Total number of muscle fibers following miR-29c/micro-dystrophin combination treatment showed no difference from untreated limb. (F) We found the number of muscle fibers per mm^2 in the miR-29c/micro-dystrophin combination treatment was significantly less than untreated mice, which was no different than wild-type. C-F (n=4-7 per group), One-way ANOVA. All data represent mean ± SEM. (*p<0.05, ***p<0.001)



Supplemental Figure 4. Measurement of absolute (A) and normalized specific force (b) following tetanic contraction in all three treatment injected GAS muscles 6 months post injection were significantly increased compared to untreated *mdx*/utrn$^{+/-}$ muscle (*p<0.05,**p<0.01,****p<0.0001). (A, B n=6-7 per group except n=13 untreated), One-way ANOVA). (C) Muscles were then assessed for loss of force following repetitive eccentric contractions. Only mice co-treated with miR-29c/micro-dystrophin and micro-dystrophin alone showed a protection from loss of force compared with untreated mdx/utrn $^{+\backslash-}$ muscles (blue) (n=3-6 per group except n=12 untreated), Two-way ANOVA. (D) miR-29c co-delivered with micro-dystrophin increased the overall weight of the injected GAS compared to either one injected alone. (n=6-8 per group except n=16 untreated), One-way ANOVA. (E) miR-29c/micro-dystrophin combination treatment demonstrated an increase in average fiber size. Comparing *mdx*/utrn$^{+/-}$ controls with miR-29c/micro-dystrophin treated *mdx*/utrn$^{+/-}$, the average diameter increased from 27 to 33.2μm (n=5-8 per group) One-way ANOVA. (F) The co-delivery produced a shift towards wild-type fiber size distribution. Given that muscle fiber diameters in *mdx*/utrn$^{+/-}$ mice is heterogeneous with many small fibers and some hypertrophic fibers, we asked whether the number of fibers per unit area (cells/mm^2) was affected with treatment. (G) We found there was no difference in muscle numbers in any of the groups (n=5-7 per group except n=11 untreated), One-way ANOVA. All data represent mean ± SEM. (*p<0.05,**p<0.01, *** P<0.001, ****p<0.0001).