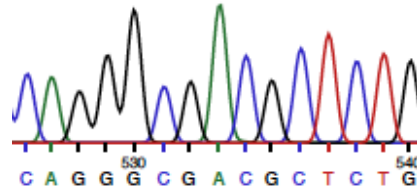
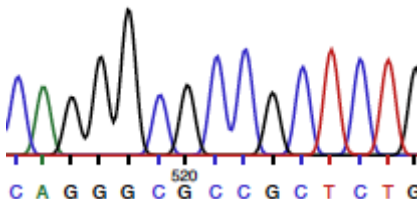


A**GDF11PRO-Fc** 5'-

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 A-3'

**B****GDF11PRO-Fc D122A** 5'-

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 A-3'

**C****GDF11** 5'-

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Figure S1. Entire DNA sequences of transgenes. (A) Sequence of GDF11PRO-Fc with D122 position in blue with the Sanger sequencing output of the highlighted region. **(B)** Sequence of GDF11PRO-Fc D122A with mutated site in red with the Sanger sequencing output of the highlighted region. **(C)** Sequence of GDF11.

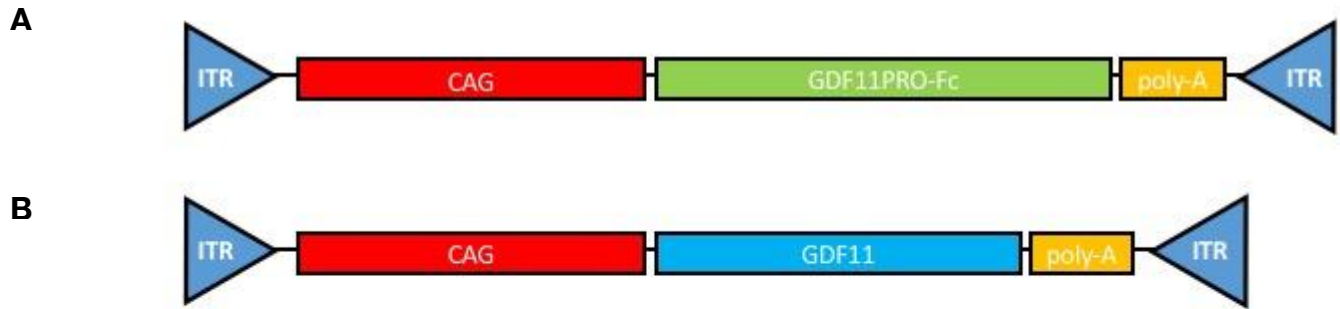


Figure S2. Vector construction. (A) GDF11PRO-Fc transgene cassette. **(B)** GDF11 transgene cassette. ITR: AAV2 inverted terminal repeat; CAG: promoter, containing the cytomegalovirus (CMV) early enhancer, first exon and first intron of chicken β -actin gene and splice acceptor of rabbit β -globin; poly-A: bGH (bovine growth hormone) polyadenylation signal

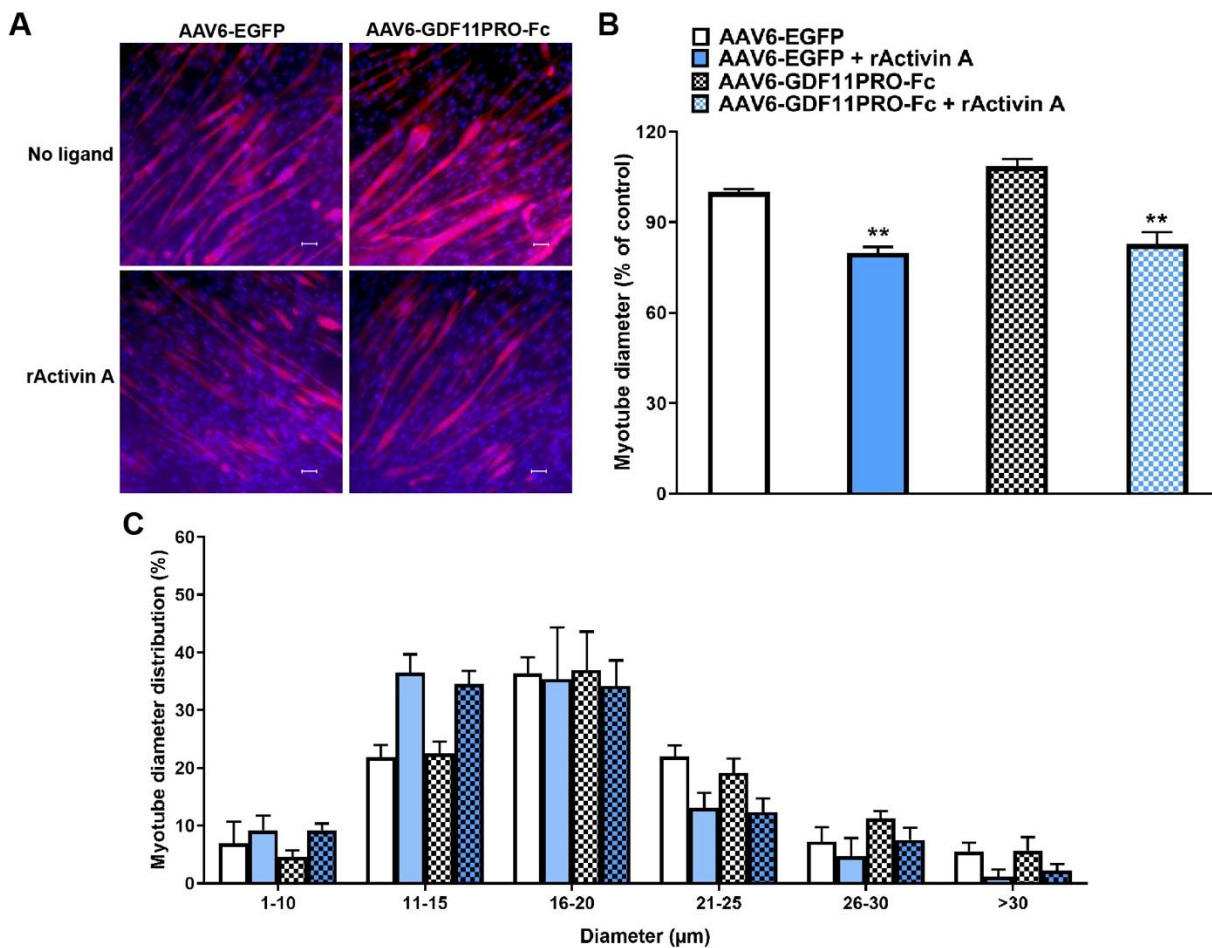


Figure S3. GDF11PRO-Fc does not prevent rActivin A-induced myotube atrophy in C2C12 cells. Differentiated C2C12 myotubes were infected with AAV6-EGFP (control) or AAV6-GDF11PRO-Fc. **(A)** Representative immunofluorescence images of dystrophin-stained C2C12 myotubes 72 hours after treatment with rActivin A or nothing. Scale bar represents 50 μ m. **(B)** Myotube diameter relative to control. **(C)** Distribution of myotube diameter. ** $p < 0.01$

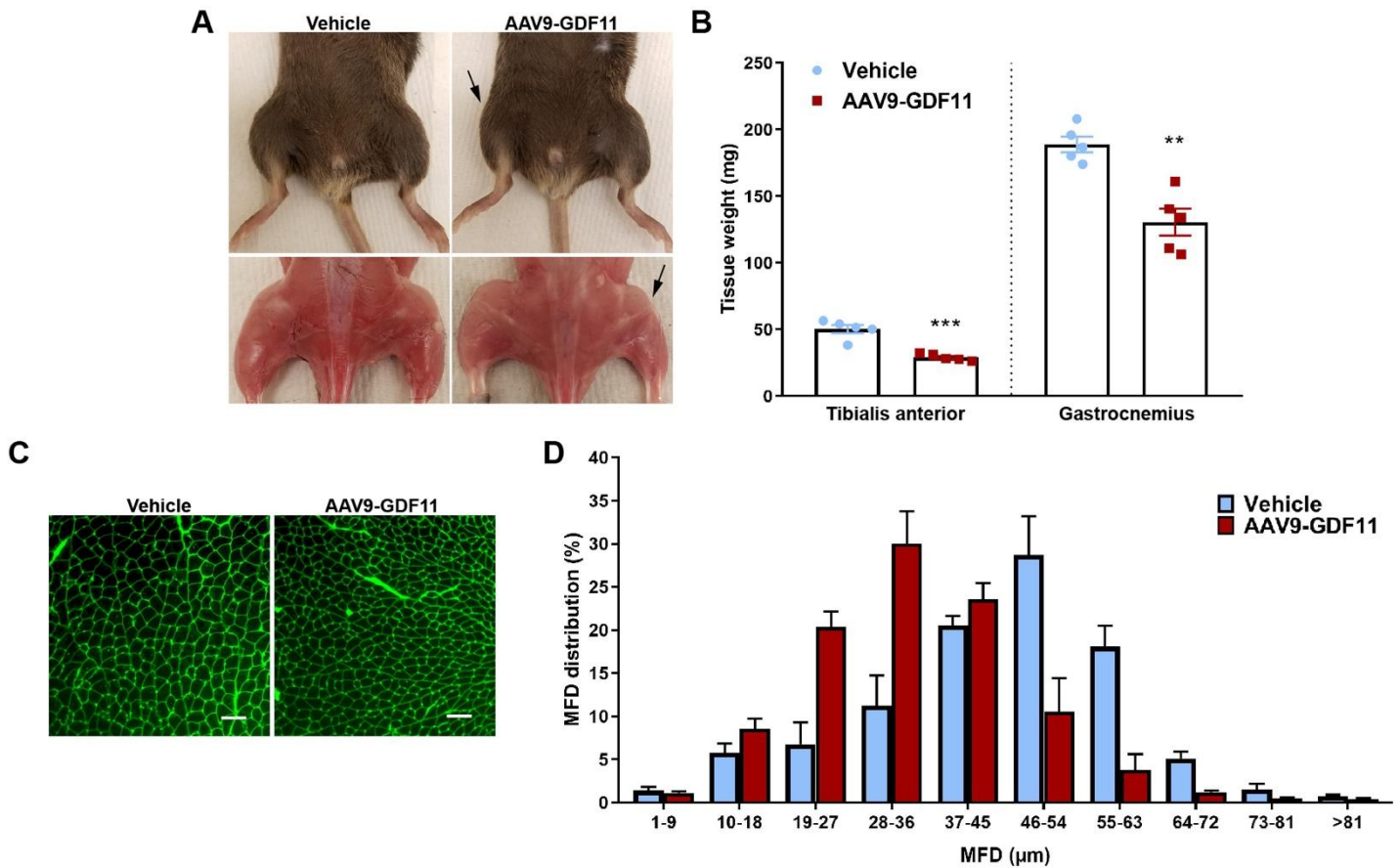


Figure S4. GDF11 overexpression induces muscle atrophy after localized intramuscular injection. 8-week-old male C57BL/6J mice were administered a unilateral intramuscular injection of AAV9-GDF11 or vehicle at a dose of 1×10^{12} vg/kg (2.5×10^{10} vg/mouse) into the right-side hindlimb. Mice were sacrificed 10 weeks post-injection. **(A)** Gross musculature of mice hindlimbs. Muscle atrophy in the AAV9-GDF11-injected limb was evident (indicated by black arrow). **(B)** Wet tissue mass of injected right-side tibialis anterior and gastrocnemius. **(C)** Representative immunofluorescence images of AlexaFluor-488-conjugated wheat germ agglutinin-stained sections of the injected right-side gastrocnemius. Scale bar represents 100 μ m. **(D)** Minimum Feret diameter (MFD) distribution of muscle fibers in the injected-right side gastrocnemius. *** $p < 0.001$; ** $p < 0.01$

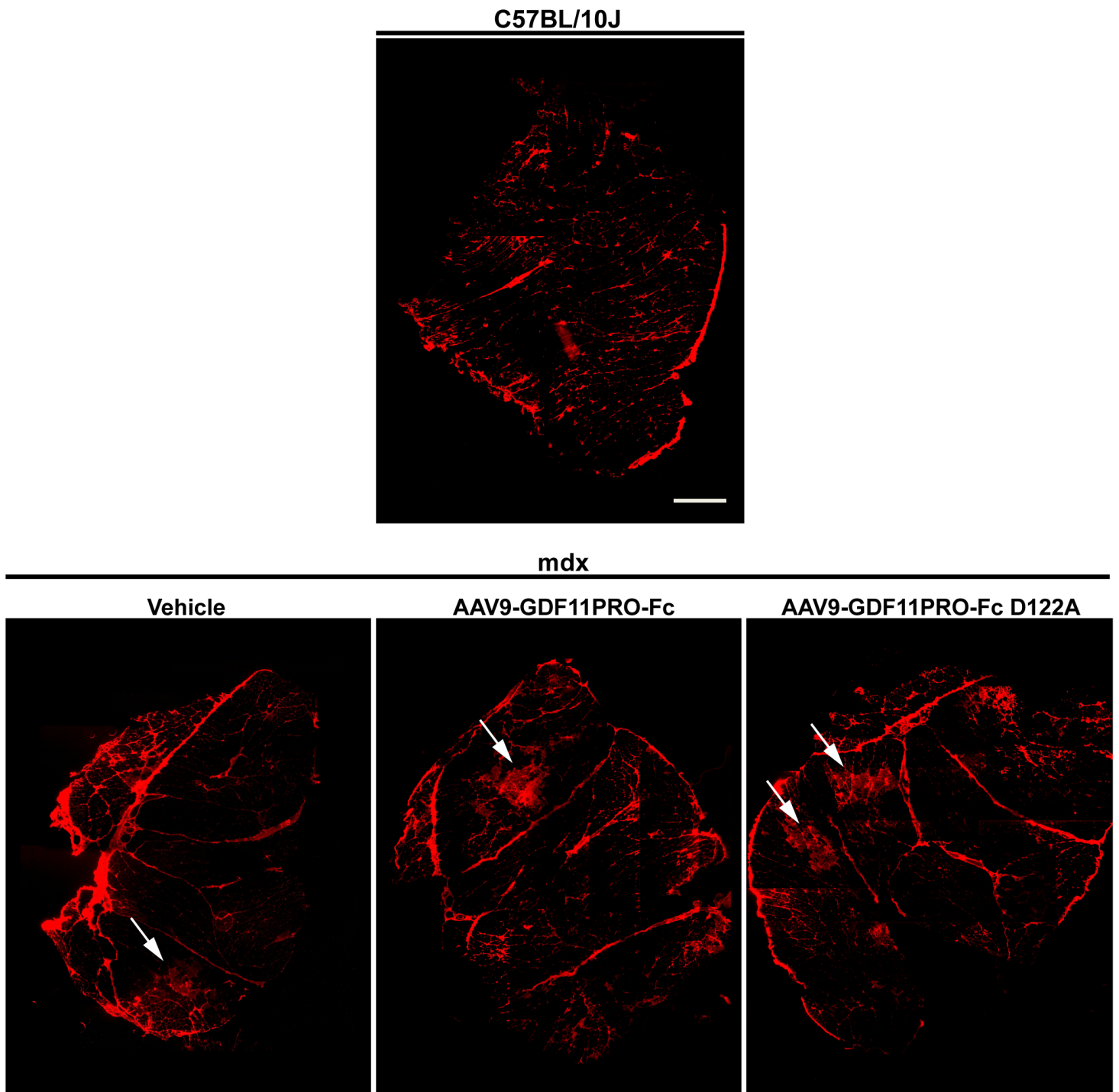


Figure S5. Serum IgG permeability in mdx gastrocnemius muscle. 6-week old male mdx mice were treated with a single intravenous injection of AAV9-GDF11PRO-Fc, AAV9-GDF11PRO-Fc D122A or vehicle at a dose of 3×10^{13} vg/kg (1×10^{12} vg/mouse). Age-matched male C57BL/10J mice were used as a wild type control. Mice were sacrificed 12 weeks post-injection. Transverse gastrocnemius muscle sections were collected and stained with a Cy3-conjugated anti-mouse IgG antibody for immunofluorescence analysis. Up to 30 separate images per sample were stitched together using a semi-automated approach to generate the final composite. Regions of positive myofiber IgG staining are marked with white arrows. Scale bar represents 300 μ m.