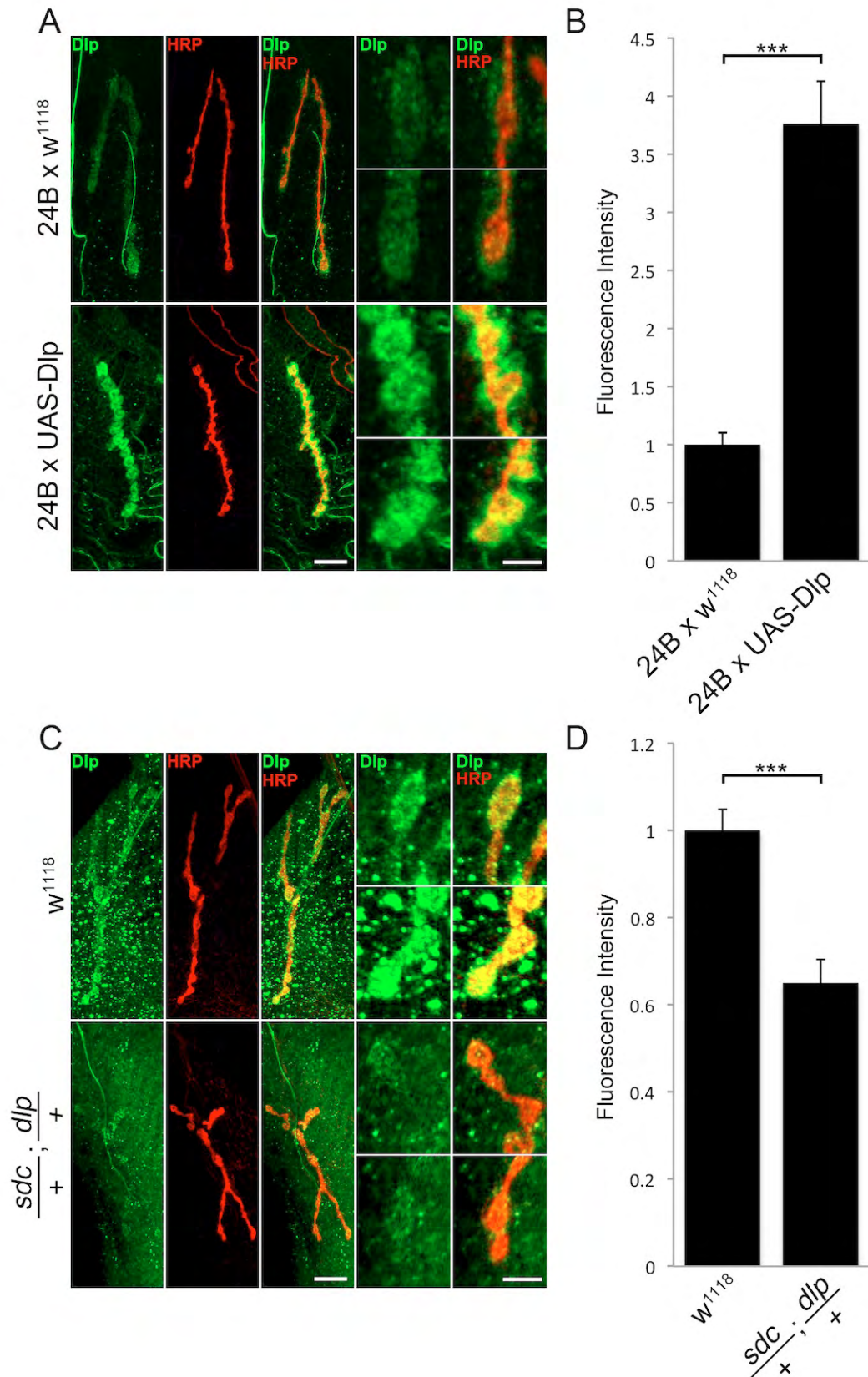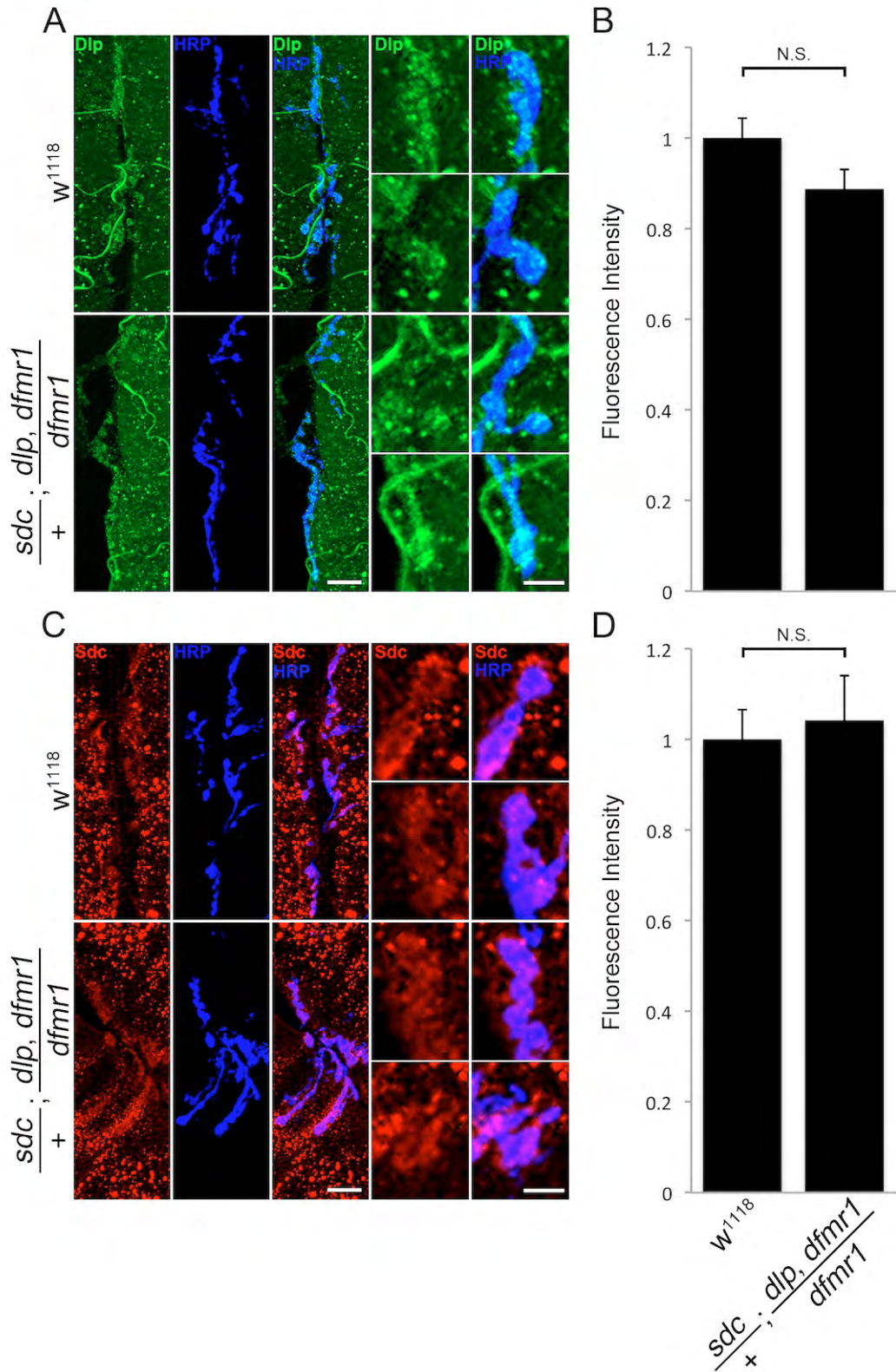**Supplementary material Fig. S1.** Reduced Jeb levels in the *dfmr1* null muscle. (A) Representative NMJ images co-labeled with presynaptic marker (HRP, red) and anti-Jelly Belly (Jeb, green) in control (*w1118*) and *dfmr1* null (*dfmr150M*) wandering third instar muscle 4. Scale bar: 15μm. (B) Quantification of Jeb muscle surface intensity away from the NMJ domain in two *dfmr1* null alleles (*dfmr150M*, *dfmr12*) normalized to the genetic control (*w1118*). Sample sizes are ≥9 animals and ≥18 muscles for each of the three genotypes. Statistical significance shown as ***P<0.001.
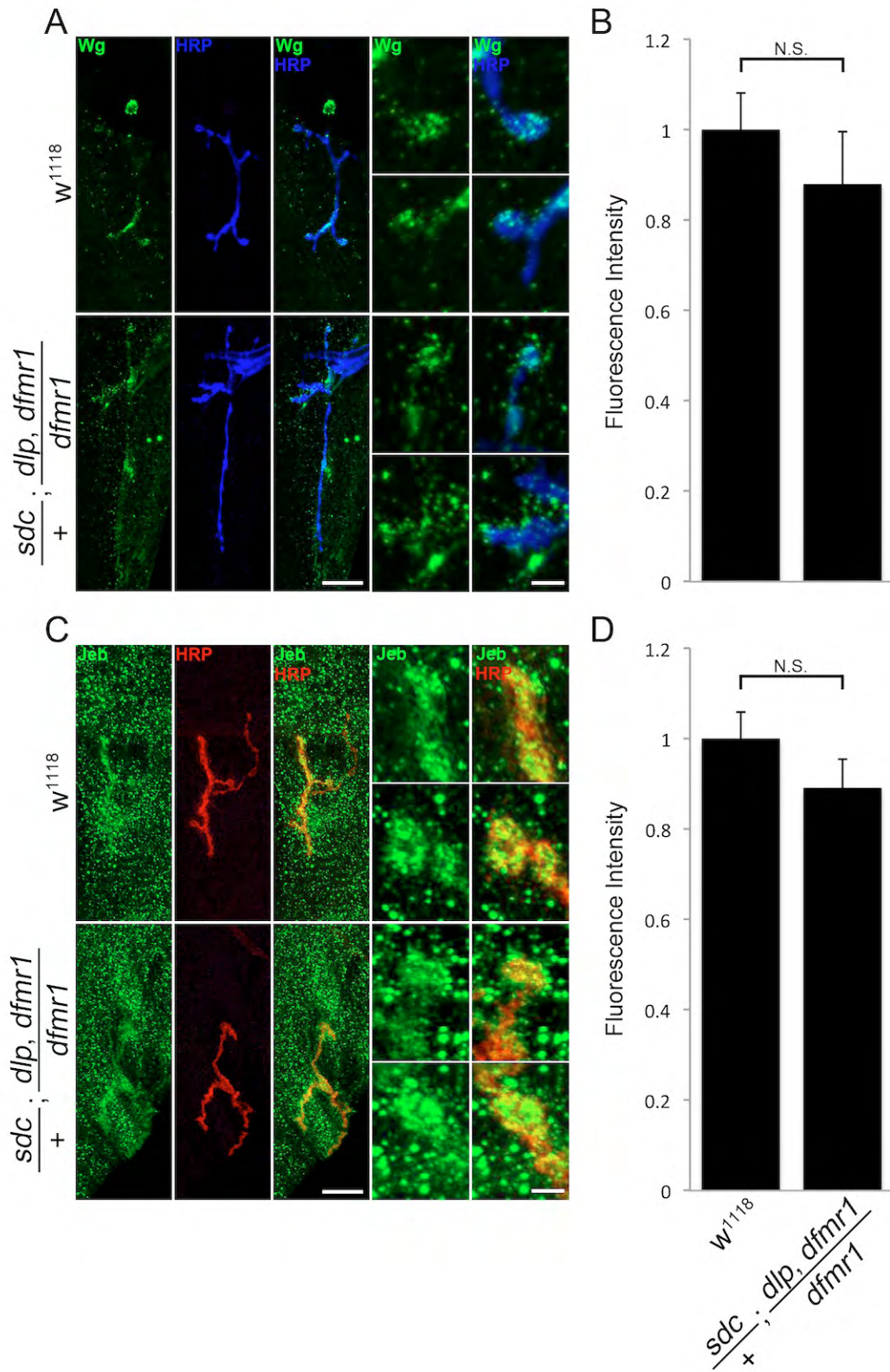
Supplementary material Fig. S2. Quantification of Dlp levels with genetic manipulations. (A) Representative NMJ images co-labeled with presynaptic marker anti-horseradish peroxidase (HRP, red) and anti-Dally-like Protein (Dlp, green) in control (w^{1118}) and postsynaptic Dlp overexpression (*24B* x UAS-*Dlp*) conditions. Scale bars: 15μm and 5 μm. (B) Quantification of Dlp intensity normalized to genetic control (w^{1118}). Sample size is ≥3 animals and ≥6 NMJs for each genotype. (C) Representative NMJ images in control and double heterozygous dlp^{A187}/+; sdc^{23}/+ conditions. Scale bars: 15μm and 5 μm. (D) Quantification of Dlp intensity normalized to genetic control (w^{1118}). Sample size is ≥8 animals and ≥16 NMJs for each genotype. Statistical significance shown as ***P<0.001.
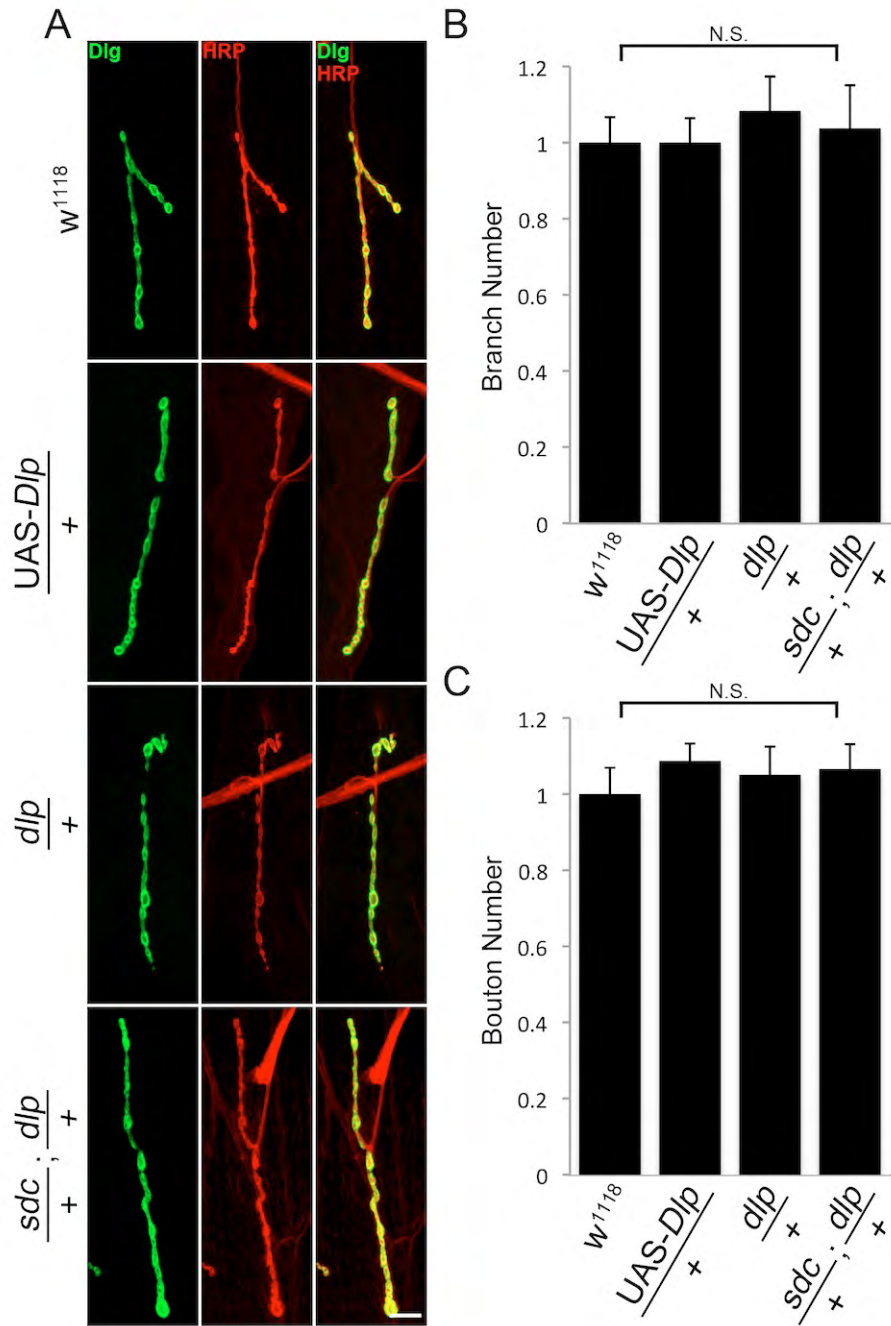
Supplementary material Fig. S3. Genetic controls for NMJ synaptic architecture studies. (A) Representative muscle 4 NMJ images co-labeled with presynaptic marker anti-horseradish peroxidase (HRP, red) and postsynaptic marker anti-Discs Large (DLG, green) in control (w^{1118}), w^{1118} crossed to UAS-Dlp, heterozygous dlp^{A187}/+, and double heterozygous dlp^{A187}/+; sdc^{23}/+. Scale bar: 15μm. Quantification of NMJ branch (B) and type 1 bouton (C) numbers. Sample size is ≥6 animals and ≥12 NMJs for each genotype. Statistical significance shown as N.S. (P>0.05).

Supplementary material Fig. S4. Synaptic HSPG levels in the triple mutant condition. (A) Representative NMJ images co-labeled with anti-HRP (red) and anti-Dlp (green) in genetic control (*w1118*) and *sdc23*/+; *dlp4187*, *dfmr150M*/*dfmr150M* triple mutant condition. Scale bars: 25μm and 5μm. (B) Quantification of Dlp intensity normalized to control. Sample size is ≥8 animals and ≥16 NMJs for genotype. (C) Representative NMJ images co-labeled with anti-HRP (red) and anti-Syndecan (Sdc, green) in control and triple mutant. Scale bars: 25μm and 5μm. (D) Quantification of Sdc intensity normalized to control. Sample sizes are ≥8 animals and ≥16 NMJs for each genotype. Statistical significance shown as N.S. (*P*>0.05).

Supplementary material Fig. S5. Wg and Jeb ligand levels in the triple mutant condition. (A) Representative NMJ images co-labeled with anti-HRP (red) and anti-Wingless (Wg, green) in control (w^{1118}) and sdc^{23}/+; dlp^{A187}, $dfmr1^{50M}$/$dfmr1^{50M}$ triple mutant conditions. Scale bars: 15μm and 5 μm. (B) Quantification of Wg intensity normalized to control. Sample size is ≥8 animals and ≥15 NMJs for each genotype. (C) Representative NMJ images co-labeled with anti- HRP (red) and anti-Jelly Belly (Jeb, green) in w^{1118} and triple mutant conditions. Scale bars: 15μm and 5μm. (D) Quantification of Jeb intensity normalized to control. Sample sizes are ≥8 animals and ≥16 NMJs for each genotype. Statistical significance shown as N.S. ($P>0.05$).
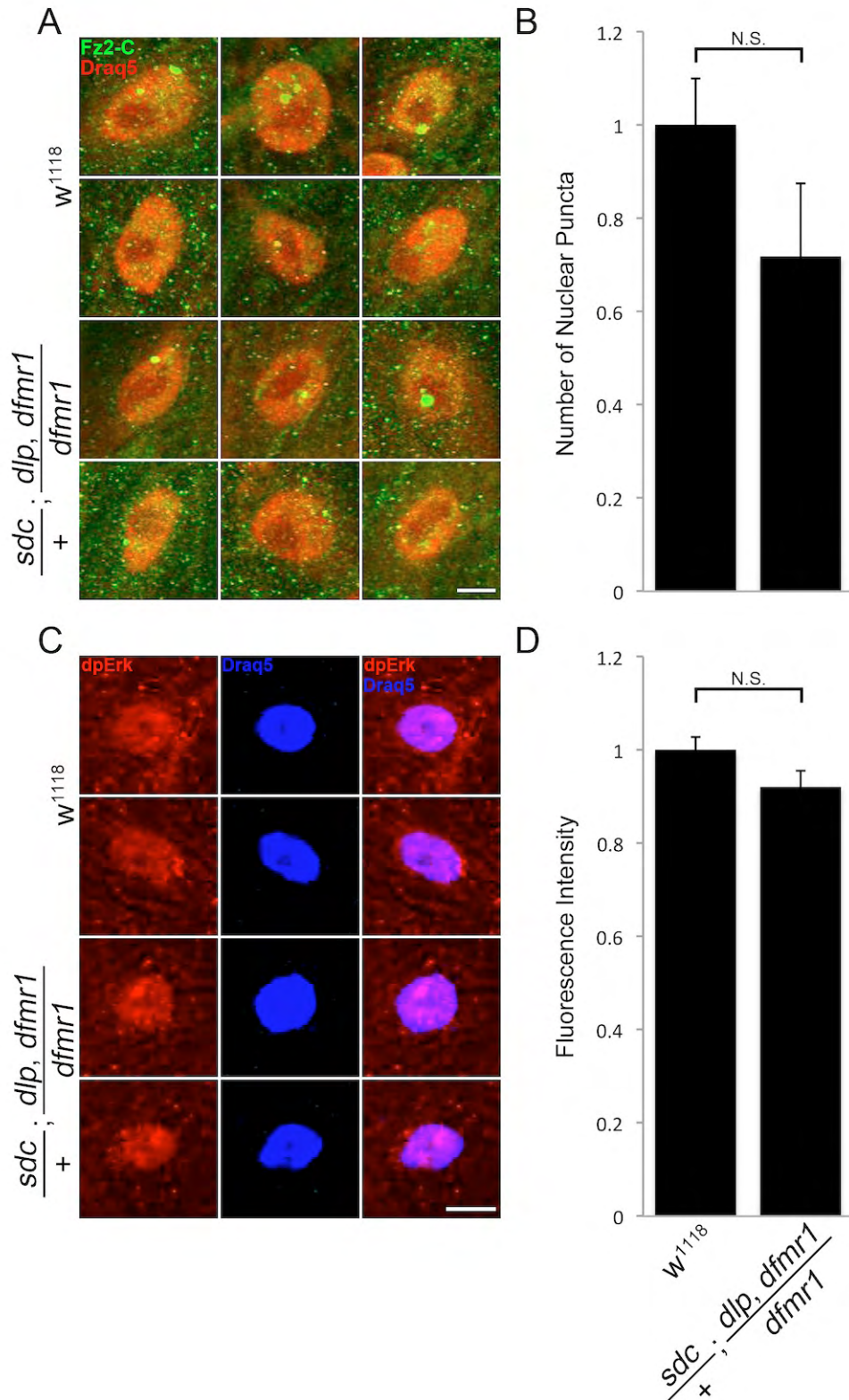
Supplementary material Fig. S6. Fz2C- and dpERK nuclear localization in the triple mutant condition. (A) Representative images of wandering third instar muscle 4 nuclei co-labeled with nuclear marker Draq5 (red) and anti-Frizzled C-terminus (Fz2-C, green) in control (w^{1118}) and sdc^{23}/+; dlp^{A187}, $dfmr1^{50M}$/$dfmr1^{50M}$ triple mutant conditions. Scale bar: 5µm. (B) Quantification of Fz2-C nuclear localization, measured as Fz2-C puncta number, normalized to genetic control. Sample sizes are ≥3 animals and ≥6 muscles for each genotype. (C) Representative images of muscle nuclei co-labeled with nuclear marker Draq5 (red) and anti-diphosphorylated extracellular signal regulated kinase (dpERK, green) in control and triple mutant. Scale bar: 5µm. (D). Quantification of dpERK nuclear intensity levels normalized to genetic control. Sample sizes are ≥8 animals and ≥15 muscles for each genotype. Statistical significance shown as N.S. (P>0.05).