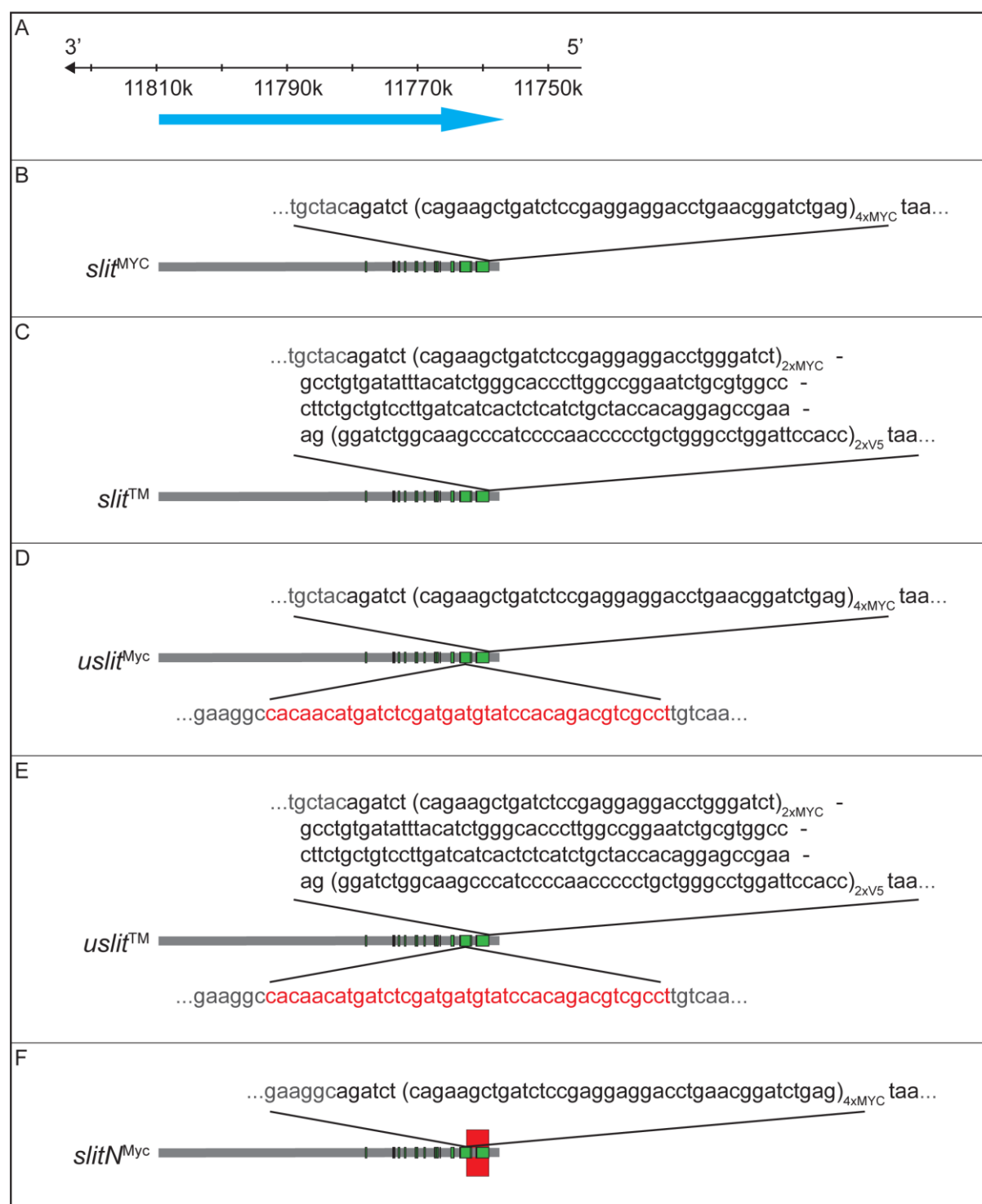


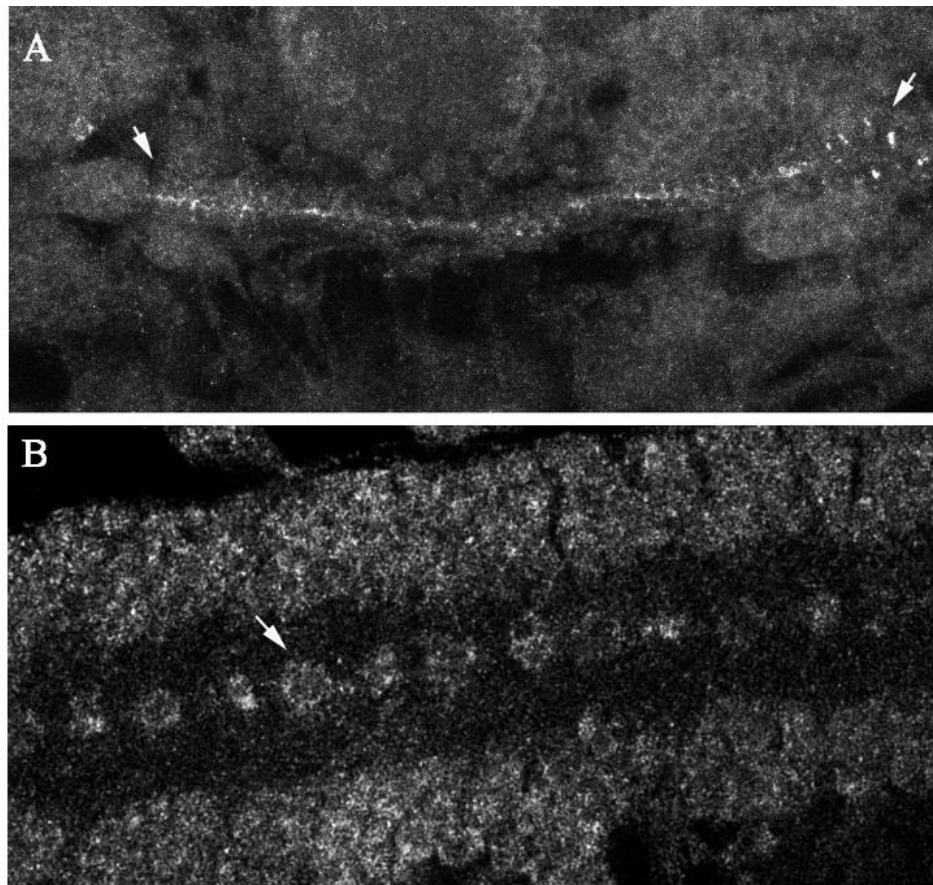
## Supplemental Figures



**Figure S1: The production of Slit knocked-in constructs (related to Figure 2)**

Five new *slit* alleles were generated using the “hands in” homologous recombination technique. Shown is the annotated position (in bp) of the endogenous *slit* locus; blue arrow indicates the *slit* reading frame (A). *slit*<sup>MYC</sup> (B), *slit*<sup>TM</sup> (C), *uslit*<sup>MYC</sup> (D), *uslit*<sup>TM</sup> (E) and

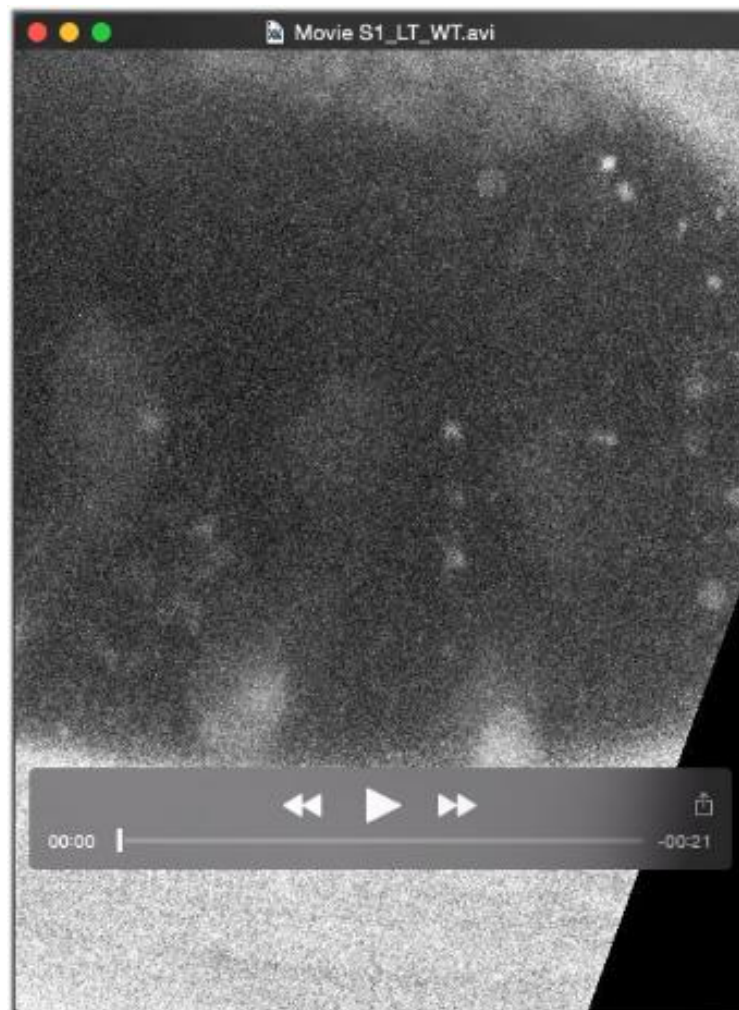
*slitN<sup>MYC</sup>* (F) loci are depicted as grey lines or grey-labeled sequence blow-ups; coding exons are marked with green bars. Black-labeled sequences indicate in-frame fusions with a MYC tag tetramer (B,D,F) or a 2xMYC-cd8TM-2xV5 membrane anchor (C,E), respectively. The red-labeled (D,E) or red-boxed sequences (F) indicate deleted endogenous *slit* sequence sections.



**Figure S2: Staining of Slit-FL-myc (related to Figure 3)**

(A) Dorsal view of an embryo carrying Slit-FL-myc and labeled with anti-myc antibody. The entire dorsal vessel is clearly labeled (arrows mark its beginning and end). (B) Ventral view of a similar embryo showing labeling of the midline glia (arrow marks a single midline glia cell).

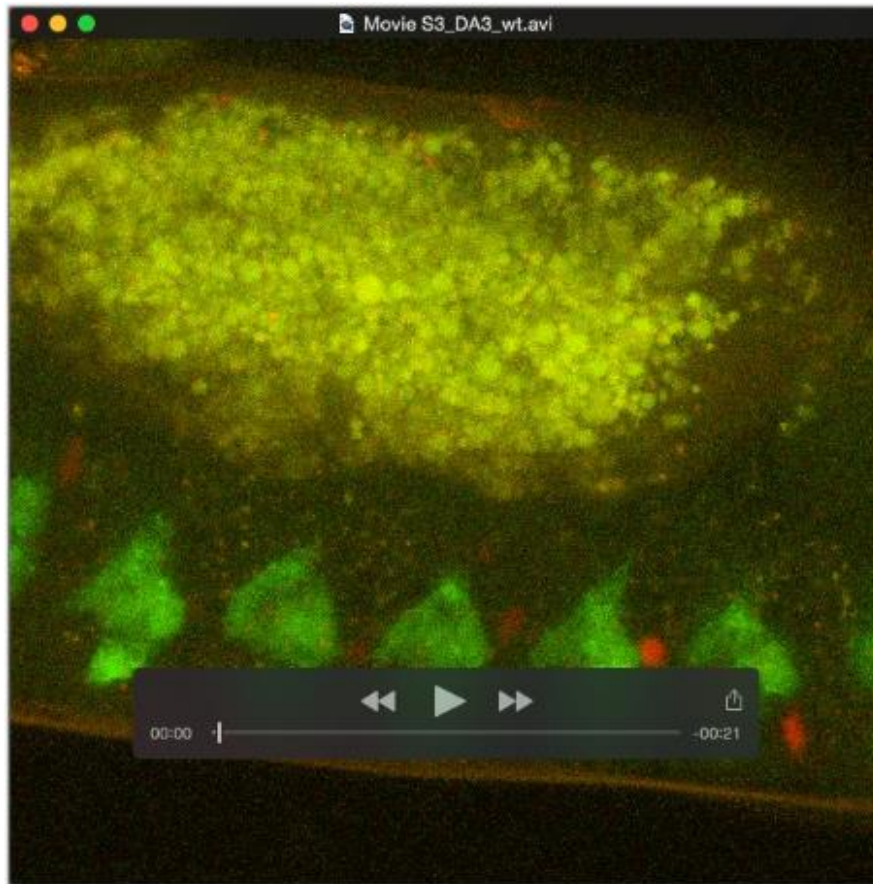
## Supplemental Movies



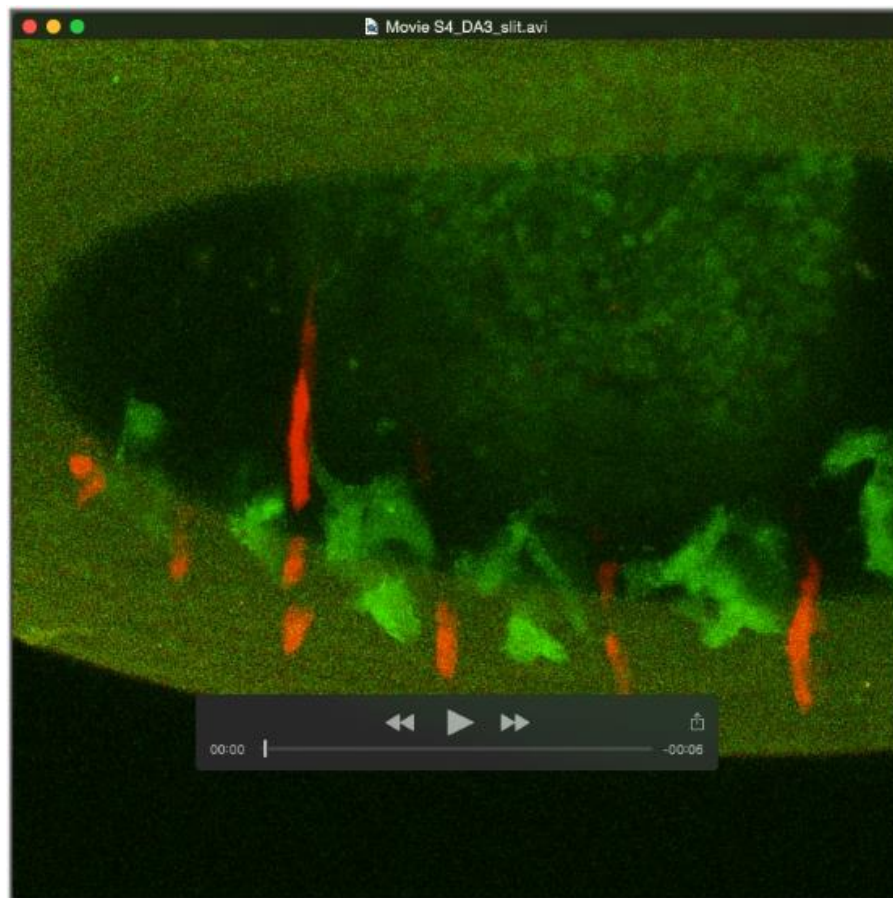
**Movie S1:** Migration of fluorescently-labeled LT muscles in WT embryo. Related to Figure 1.



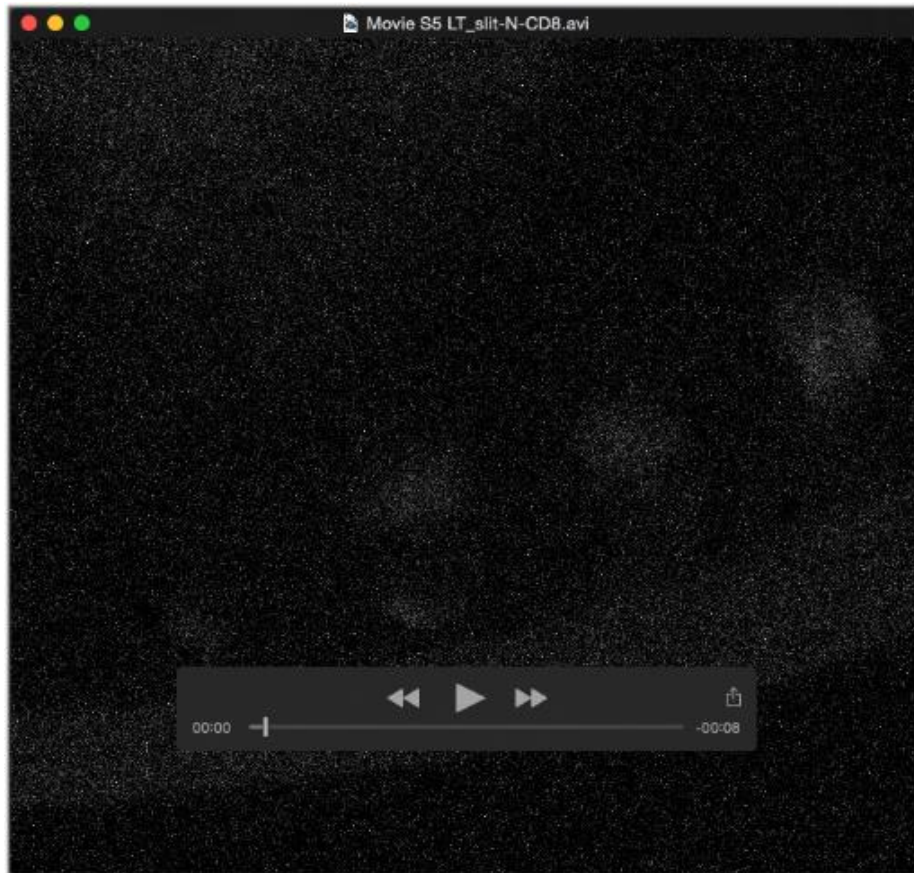
**Movie S2:** Migration of fluorescently-labeled LT muscles in *slit* mutant embryo. Related to Figure 1.



**Movie S3:** Migration of the DA3 muscle (labeled with GFP) relative to the tendon cells (labeled with RFP) in WT embryos. Related to Figure 1.



**Movie S4:** Migration of the DA3 muscle (labeled with GFP) relative to the tendon cells (labeled with RFP) in *slit* mutant embryos. Related to Figure 1.



**Movie S5:** Migration of fluorescently-labeled LT muscles in Slit-N-CD8 knock-in embryo. Related to Figure 4.