



**POSTER #9**

**DYNAMICS OF SKELETAL MUSCLE NUCLEI DURING  
MUSCLE CONTRACTION IN INTACT *DROSOPHILA* LARVAE**

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Mechanical stimuli, originating from the cellular and tissue micro environment, are transmitted into the nucleus, where they critically affect nuclear activity. Muscle nuclei are uniquely exposed to variable cytoplasmic mechanical stimuli emerged from the contractile nature of the muscle tissue.

Most of the current methods to study muscle mechanics involve isolation of the muscle from its intrinsic tissue environment and extensive processing of the muscle before it is analyzed, which might alter its mechanical properties and physiological activity.<sup>1</sup>

In order to study myonuclear dynamics and its mechanical characteristics, we developed a minimal constraint device for live, intact *Drosophila melanogaster* larvae, that is combined with muscle specific expression of fluorescent nuclear and Z-line markers. This setup allows to perform live imaging of intact muscles and to quantify the movement of the nuclei and Z-lines during active muscle contraction and/or passive extension. The larvae is placed in the device in a manner that minimally restricts its movement, and is awake throughout the experiment. The individual muscle fibre and its nuclei are visible during a complete event of contraction or relaxation while the contractions are feasible only along its body axis, without turning or bending. The device is mounted in a spinning disk confocal microscope, which allows rapid acquisition of the entire imaging plane and hence, tracking of the nuclei along the fiber throughout a dynamic event. Figure 1 shows two representing images of a muscle before (A) and during (B) contraction.

In order to characterize the mechanical environment of each nucleus, its displacement in time was calculated during a single active event. The x-y location of each nucleus in a time series was measured with the manual tracking plug-in from ImageJ. The displacement of a nucleus that occurred between two successive sampling time points was calculated. With the sampling times that were obtained from the sampling system, the displacement function in time was obtained. By numerically differentiating the displacement function, the velocity, and acceleration in time were determined. These parameters enabled us to extract information regarding the mechanical signals that are transmitted to the nuclei from its environment during contractile and extension events.

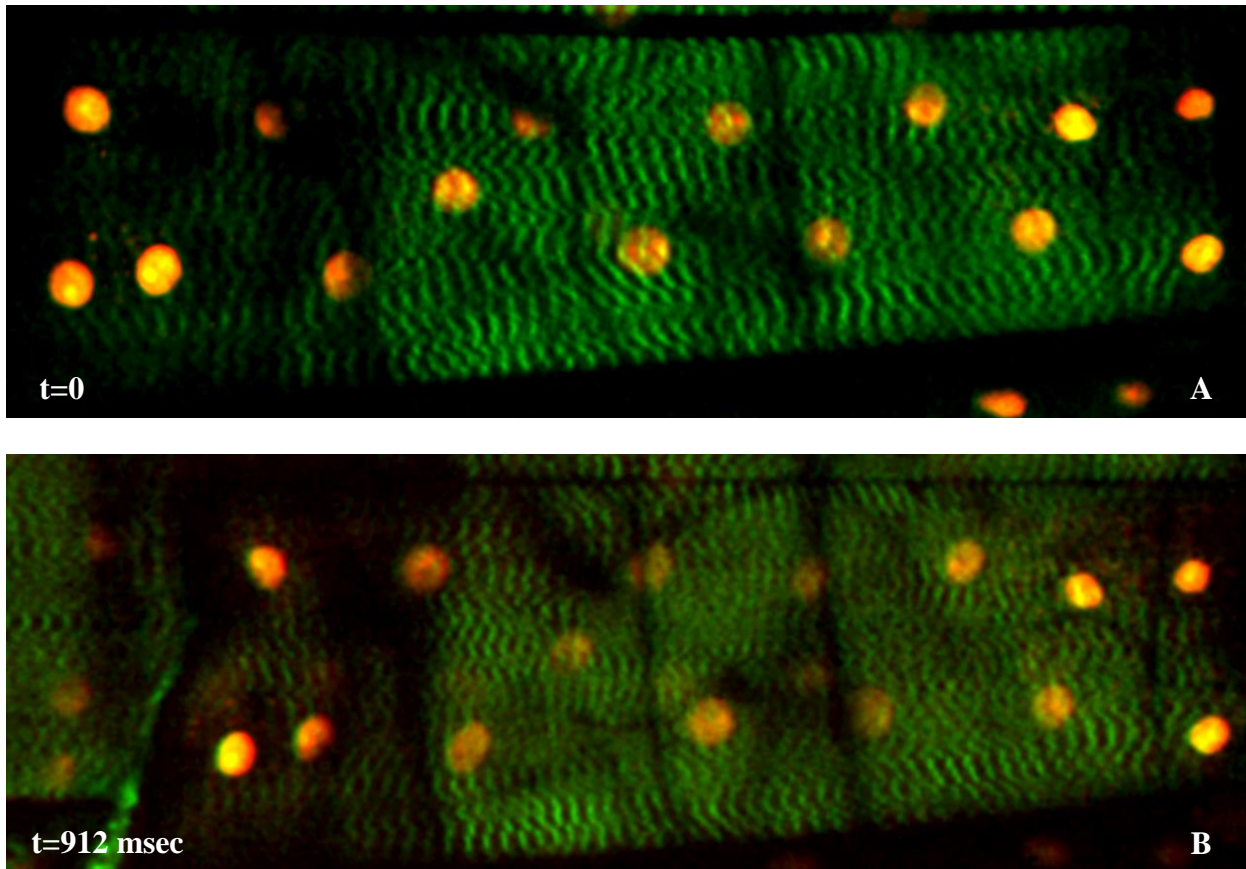


Figure 1: A whole muscle with its nuclei before (A) and during (B) contraction.

Our analysis of wild type muscles reveals that the magnitude of parameters such as nuclear displacement, average velocity, and maximal acceleration are not homogenous along the longitudinal axis of a given muscle fiber, implying that nuclei in different positions along the fiber are exposed to distinct mechanical environments. Importantly, our studies reveal that disruption of the nuclear-cytoskeleton anchoring in various mutant larvae changes the mechanical characteristic of nuclear dynamics significantly.