# ABSENCE OF DYSTROPHIN ALTERS THE PASSIVE PROPERTIES OF THE EXTENSOR DIGITORUM LONGUS MUSCLE IN MICE

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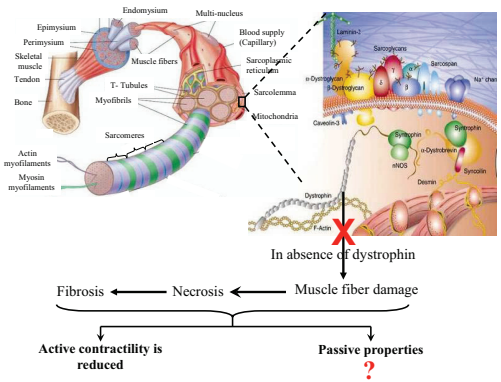
## Introduction

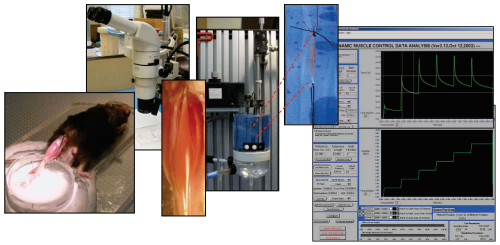
Dystrophin is a cytoskeletal protein not directly participating with the myosin-actin contractile apparatus in muscle. The loss of dystrophin leads to Duchenne muscular dystrophy. It is well-established that contractility is reduced in dystrophin-null muscle. Surprisingly, little is known about the influences of dystrophin-deficiency on the passive properties of muscle.




**Previous studies by Tidball and his colleagues$_{(1991\text{-}1995)}$ showed:**

- No difference in the passive properties between control and dystrophin deficient mice
- Ultrastructural examination of dystrophic MTJ predicted weakness in the MTJ strength due to: 1.Reduction in the junctional membrane folding 2.Absence of lateral association between the thin filaments and the sarcolemma 3. Presence of muscle degeneration.
- When dystrophin deficient muscle is strained; failure occurred within the muscle fiber not at the MTJ.

## Methods



After carefully dissecting the extensor digitorum longus (EDL) from the hind limp, the EDL muscle was vertically mounted at the tendon ends in a 30ºC jacketed organ bath containing ringer buffer bubbled with 95% O2 and 5% CO2 to stabilize the pH at 7.4. The proximal tendon was secured to a 305B dual-mode servomotor transducer (Aurora Scientific, Inc., Aurora, ON, Canada) while the distal tendon end was attached to a fixed post. The EDL muscle was subjected to a stepwise protocol where it was stretched by an increment of 10% of its optimal length (Lo) till it reached 160% Lo using a rate of 2 cm/s. Muscle stiffness were determined according to the stress-strain curve.

## Results

### Absence of dystrophin increases muscle stiffness

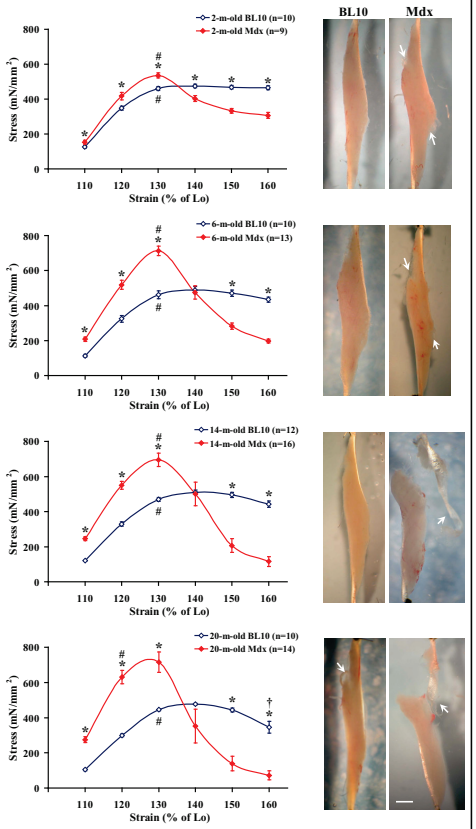



**Figure 1.** Age-matched comparison of the EDL muscle strain-stress curves in BL10 and mdx mice (Scale bar:1 mm applied to all images)

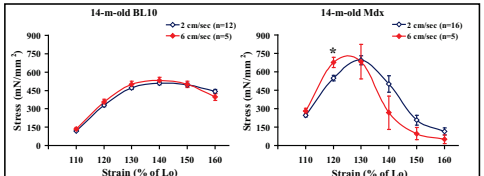



**Figure 2.** Influence of the stretch speed on the strain stress curve in 14-m-old mice

## Results

### Possibility of weakness in the MTJ strength

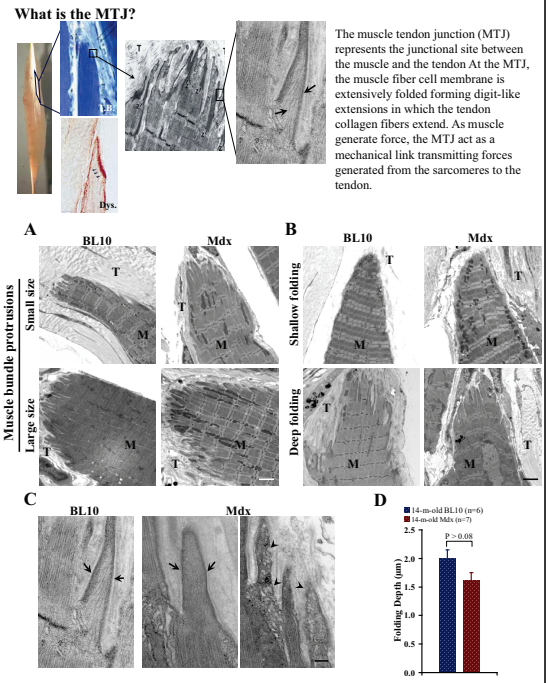
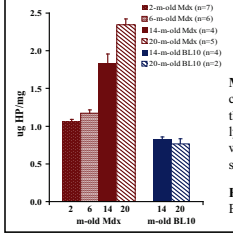



**Figure 3.** Ultrastructural changes at the MTJ. **A**, Muscle bundles that intrude into the tendon showed variable size. Scale bar, 2µm. **B**, Representative low magnificent EM photomicrographs showing variations in the depth of the digital folding of the MTJ interface. Scale bar, 2µm **C**, Representative high magnificent EM photomicrographs of the myofiber digital processing at the MTJ in 14-m-old BL10 and mdx. Arrow, lateral condensation of the myofilaments at the sarcolemma; Arrowhead, vacuolar degeneration. Scale bar, 200 nm. M, muscle; T, tendon. **D**, Quantitative analysis of the variation in the folding depth between 14-m-old BL10 and mdx .

### Increase of Collagen deposition along age in mdx




**Methods:** The EDL muscle from either BL10 or mdx was carefully dissected and both tendons were trimmed from the muscle as well as the MTJ$_{(s)}$. The muscle was lyophilized and weighted to determine the dry tissue weight. The concentration of the hydroxyproline in each sample was measured and normalized to the dry weight.

**Figure 4.** Quantification of the amount of collagen in the EDL muscle belly using the hydroxyproline assay.

## Results

### The viscoelastic property is altered in mdx

**Muscle has viscoelastic property**

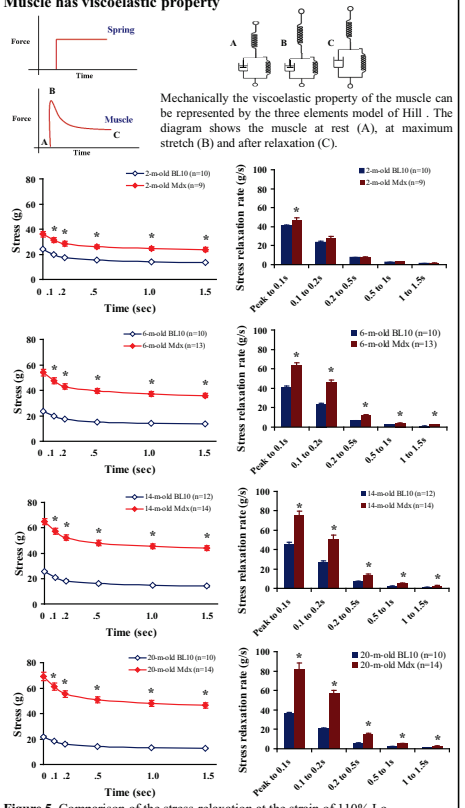



**Figure 5.** Comparison of the stress-relaxation at the strain of 110% Lo

## Conclusion

As a consequence of the dystrophic process:

- Muscle stiffens increase along age
- The viscoelastic property is altered
- In consequence of muscle stiffness and possibility of weakness in the MTJ strength, the strain failure site occurred at the MTJ in older mdx

**Can gene/pharmacological therapies halt the alternation of the passive properties?**

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