A severe contractile function impairment of the hindlimb and diaphragm muscles in the dystrophin-deficient *Dmd*^{mdx} rat model

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Introduction

Duchenne Muscular Dystrophy (DMD) is a severe muscle-wasting disorder caused by mutations in the *Dmd* gene encoding dystrophin. Using TALENs, we generated a DMD^{mdx} rat model carrying an out-of-frame mutation in exon 23 of the *Dmd* gene and exhibiting muscular and cardiac phenotypes similar to those observed in DMD patients. In order to better characterize the impact of dystrophin loss on the contractile muscular phenotype of this animal model, we explored *in vitro* contractile parameters of various muscles as soleus, EDL and diaphragm.

Methods

To analyze muscle function, *in vitro* experiments were performed on isolated fast-(EDL), slow-(soleus) twitch muscles and diaphragm bundles. Muscle preparations were mounted in an *in vitro* muscle test system (model 1205, Aurora Scientific In., Aurora, Canada) containing Ringer physiological salt solution at 25° C and continuously bubbled.

Animals : Dmd^{mdx} male rats ages 3 months

Shorter life span and reduced body weight for Dmd^{mdx} rats

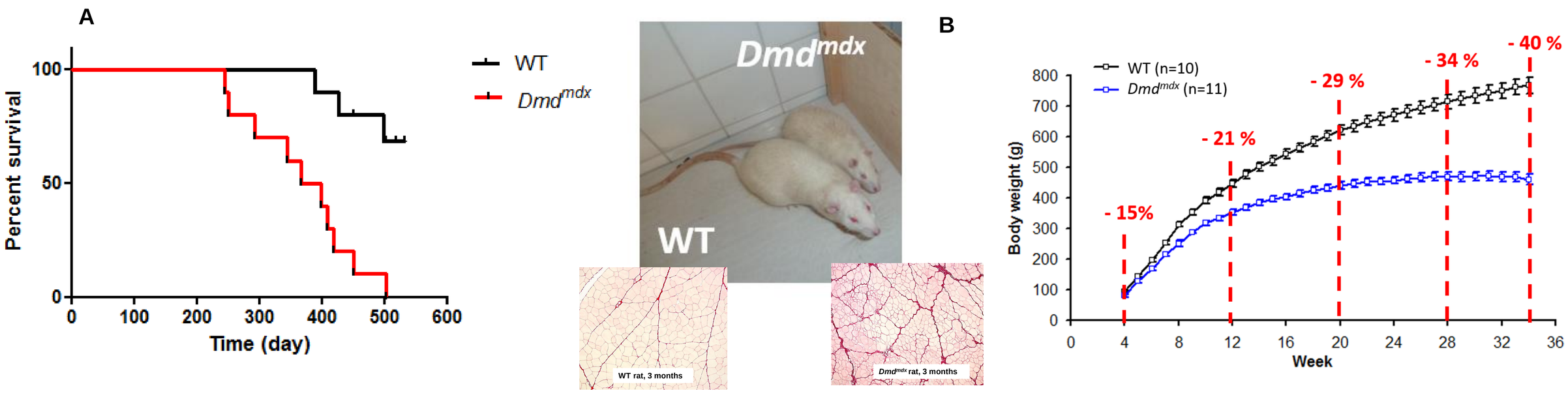
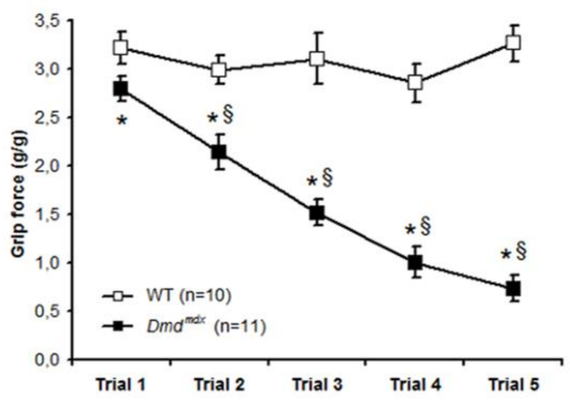


Figure 1 : Life span (**A**) and body weight (**B**) curves in WT and Dmd^{mdx} rats. **A**) Shorter life span in Dmd^{mdx} rats : 382 days 50% of death in DMD^{mdx} with no death in WT rats. **B**)Decrease of body weight from 4 weeks in Dmd^{mdx} rats.

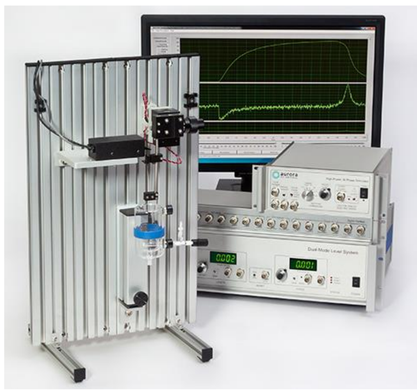
Reduced force in Edl, Soleus and diaphragm muscle from Dmd^{mdx} rats

Grip force and isolated muscle tension



	WT	*Dmd*^{mdx}	
Force max (g/g)	3,66 +/- 0,13	2,83 +/- 0,12	(-23 %)
Decrease (%)	1,90 +/- 4,27	-71,87 +/- 6,97	
n	10	11	

Figure 2 : Forelimb force measurements at 3 months in WT and Dmd^{mdx} rats using the grip test (5 trials). Weaker globale and relative force produced by forelimbs in Dmd^{mdx} rats and strong muscular fatigue.



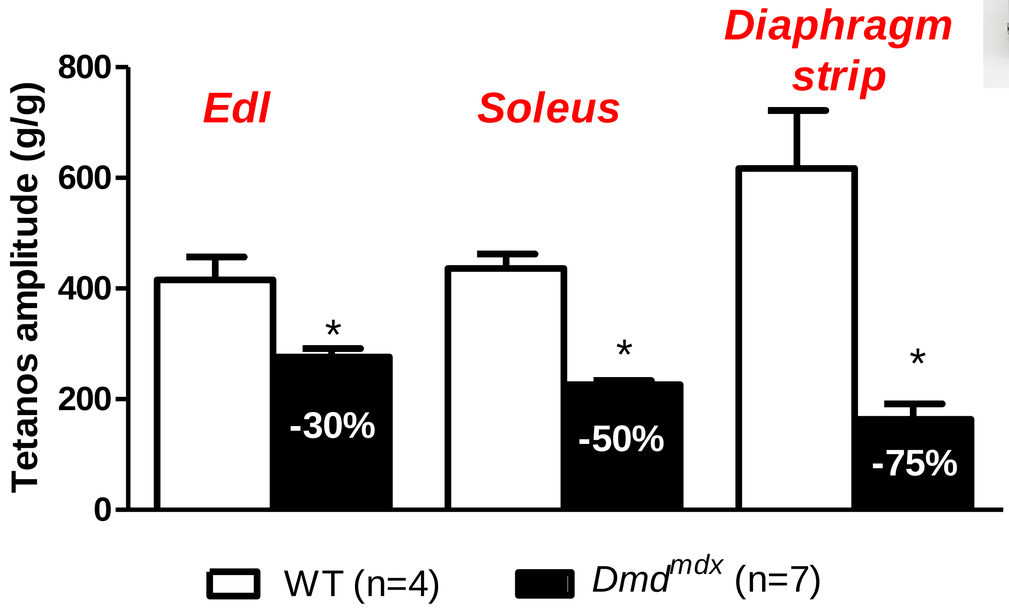


Figure 3 : Amplitude of tetanos developed by Edl and Soleus muscle, and diaphragm strip.

Conclusion

In conclusion, these results are a new demonstration of the robustness of the DMD^{mdx} rat as a model for DMD. They also showed that skeletal muscles are clearly altered in this model, and that the diaphragm force is severely affected as soon as 3 months of age. Knowing that most of the DMD patients die from respiratory dysfunction, related to the impairment of respiratory muscles (including the diaphragm), our data reinforce the relevance of using the DMD^{mdx} rat model for the preclinical evaluations of new treatments for DMD at a global level, including those that target the respiratory muscular function

References

Characterization of dystrophin deficient rats: a new model for Duchenne muscular dystrophy. Larcher T, Lafoux A, Tesson L, Remy S, Thepenier V, François V, Le Guiner C, Goubin H, Dutilleul M, Guigand L, Toumaniantz G, De Cian A, Boix C, Renaud JB, Cherel Y, Giovannangeli C, Concordet JP, Anegon I, Huchet C. PLoS One. 2014 Oct 13;9(10):e110371.