Abstract: 18O

Fibre type-specific abundance of dysferlin in rodent skeletal muscle

Stefan G Wette and Robyn M Murphy

Department of Biochemistry and Chemistry, School of Agriculture Biomedicine and Environment La Trobe University, VIC 3086, Australia

Dysferlin is an important protein, playing a role in maintaining skeletal muscle function. Mutations in the dysferlin gene result in one of two diverse dysferlinopathies, miyoshi myopathy and limb-girdle muscular dystrophy 2B. Both of these present with muscle weakness and atrophy and are distinguished by the site of muscle weakness being distal limb-girdle and proximal lower limb girdle musculature, respectively. For most patients with dysferlinopathies, loss of lower leg muscle mass is evident in the gastrocnemius and soleus and muscle loss to all lower leg muscles is evident with the disease.

Skeletal muscle is heterogenous in nature, being comprised of slow, oxidative through to fast, glycolytic fibres, typically distinguished by the presence of specific isoforms of myosin heavy chain (MHC), MHCI (type I), MHCIIa (type IIa) and MHCIIb/x (type IIb/x). A proteomic study showed the abundance of dysferlin is higher in type II compared to type I fibres in human *vastus lateralis* muscle (4). In contrast, mouse proteomic data suggested that dysferlin abundance was similar in type I and type IIb fibres (3). There have been no studies comparing the abundance of dysferlin in rat muscle fibres. This study aimed to determine the fibre type-specific abundance of dysferlin in extensor digitorum longus (EDL) and soleus muscle fibres from C57BL/6J mice and Sprague-Dawley rats that were 2-3 months old. Animals were sacrificed using a lethal overdose of fluothane in accordance with the La Trobe University Ethics Committee. As a first step, we compared the levels of dysferlin in homogenate samples from rat and mice soleus and EDL muscles. These muscles were chosen for their distinctly different muscle fibre types, including different proportions type I, IIa, and IIx that are present in human soleus and gastrocnemius muscles (Table 1). The abundance of dysferlin in mouse EDL muscle was ~120% higher than in the soleus (n = 7, P = 0.016, paired t-test). In contrast to the mouse muscles, the abundance of dysferlin in EDL was ~56% lower than in soleus in rats (n = 11, P = 0.019, paired t-test). The next step in this study is to determine the fibre type-specific abundance of dysferlin in pooled type I and II fibres from mouse and rat EDL and soleus muscles.

Table 1. Proportions (%) of muscle fibre types in rat and mouse soleus, EDL compared to human soleus and gastrocnemius muscles. Data obtained from (1, 2, 5).

Species	Strain	Muscle	I (slow oxidative)	IIa (fast oxidative)	IIx (fast glycolytic)	IIb (fast glycolytic)
Rat	Sprague-Dawley	Soleus	86	6	8	0
		EDL	3	11	29	58
Mouse	C57Bl/6J	Soleus	38	52	8	2
		EDL	0	0	12	88
Human		Soleus*	88	9	0	Not present in humans
		Gastrocnemius#	70	10	3	

*Human soleus mixed fibres = 3%, #gastrocnemius mixed fibres = 16%

1.**Delp MD, and Duan C**. Composition and size of type I, IIA, IID/X, and IIB fibers and citrate synthase activity of rat muscle. *J Appl Physiol* 80: 261-270, 1996. 2.**Lloyd EM, Xu H, Murphy RM, Grounds MD, and Pinniger GJ**. Dysferlin-deficiency has greater impact on function of slow muscles, compared with fast, in aged BLAJ mice. *PLoS One* 14: e0214908, 2019. 3.**Murgia M, Nagaraj N, Deshmukh AS, Zeiler M, Cancellara P, Moretti I, Reggiani C, Schiaffino S, and Mann M**. Single muscle fiber proteomics reveals unexpected mitochondrial specialization. *EMBO Rep* 16: 387-395, 2015. 4.**Murgia M, Nogara L, Baraldo M, Reggiani C, Mann M, and Schiaffino S**. Protein profile of fiber types in human skeletal muscle: a single-fiber proteomics study. *Skelet Muscle* 11: 24, 2021. 5.**Trappe S, Costill D, Gallagher P, Creer A, Peters JR, Evans H, Riley DA, and Fitts RH**. Exercise in space: human skeletal muscle after 6 months aboard the International Space Station. *J Appl Physiol (1985)* 106: 1159-1168, 2009.