Systemic Down-Regulation of Delta-9 Desaturase Promotes Muscle Oxidative Metabolism and Accelerates Muscle Function Recovery following Nerve Injury

Ghulam Hussain1,2,*, Florent Schmitt1,2, Alexandre Henriques1,2, Thiebault Lequeu1,2, Frederique Rene1,2, Françoise Bindler3, Sylvie Dirrig-Grosch1,2, Hugues Oudart4, Lavinia Palamiu4,5, Marie-Helene Metz-Boutigue6, Luc Dupuis1,2, Eric Marchioni3, Jose-Luis Gonzalez De Aguilar1,2, Jean-Philippe Loeffler1,2,4,5

1 INSERM, U1118, Mécanismes Centraux et Périphériques de la Neurodégénérescence, Strasbourg, France, 2 Université de Strasbourg, UMR5118, Strasbourg, France, 3 CNRS, UMR7178, Institut Pluridisciplinaire Hubert Curien, Équipe de Chimie Analytique des Molécules Bioactives, Illkirch, France, 4 CNRS, UPR9010, Institut Pluridisciplinaire Hubert Curien, Département d’Ecologie, Physiologie et Ethologie, Strasbourg, France, 5 INSERM, U977, Laboratoire des Biomatériaux et Ingénierie Tissulaire, Strasbourg, France

Abstract

The progressive deterioration of the neuromuscular axis is typically observed in degenerative conditions of the lower motor neurons, such as amyotrophic lateral sclerosis (ALS). Neurodegeneration in this disease is associated with systemic metabolic perturbations, including hypermetabolism and dyslipidemia. Our previous gene profiling studies on ALS muscle revealed down-regulation of delta-9 desaturase, or SCD1, which is the rate-limiting enzyme in the synthesis of monounsaturated fatty acids. Interestingly, knocking out SCD1 gene is known to induce hypermetabolism and stimulate fatty acid beta-oxidation. Here we investigated whether SCD1 deficiency can affect muscle function and its restoration in response to injury. The genetic ablation of SCD1 was not detrimental per se to muscle function. On the contrary, muscles in SCD1 knockout mice shifted toward a more oxidative metabolism, and enhanced the expression of synaptic genes. Repressing SCD1 expression or reducing SCD-dependent enzymatic activity accelerated the recovery of muscle function after inducing sciatic nerve crush. Overall, these findings provide evidence for a new role of SCD1 in modulating the restorative potential of skeletal muscles.

Introduction

In mammals, the contraction of voluntary skeletal muscles is under the control of motor neurons whose cell bodies are located in the spinal cord and the brainstem. These so-called lower motor neurons directly communicate with muscle fibers through the neurotransmitter acetylcholine at the neuromuscular junctions. The progressive functional deterioration of this neuromuscular axis is typically found in neurodegenerative conditions such as amyotrophic lateral sclerosis (ALS). This disease, which is the most frequent adult-onset form of motor neuron disease, is characterised by motor neuron death, skeletal muscle atrophy and paralysis [1]. Studies conducted on genetic animal models of ALS showed that the process leading to motor neuron degeneration is not cell-autonomous but involves defects in other cell types than neurons [2–5]. Consistent with this notion, ALS neurodegeneration is also associated with systemic defects, including hypermetabolism and dyslipidemia, which are observed in both patients and animal models [6–8]. Particularly, transgenic mice overexpressing a mutated form of Cu/Zn superoxide dismutase (SOD1), linked to familial ALS, are in energy deficit and have decreased adipose tissue stores. These deficiencies appear to be elicited by increased energy expenditure (that is to say, hypermetabolism), and due to an increased consumption of nutrients by skeletal muscles [9,10].

To gain insight into the relationships between neurodegeneration and metabolic dysfunction, we recently analyzed the gene expression profiles of skeletal muscles from mutant SOD1 mice...
and patients with sporadic ALS [11,12]. We found a decrease in the expression of stearoyl-CoA desaturase-1 (SCD1), an enzyme that introduces the first \( \text{cis} \) double bond in the \( \delta-9 \) position of saturated fatty acyl-CoA substrates. The preferred substrates are palmitoyl-CoA (C16:0) and stearoyl-CoA (C18:0), which are converted to palmitoleoyl-CoA (C16:1) and oleoyl-CoA (C18:1), respectively [13]. These monounsaturated fatty acids are the major constituents of complex lipids such as diacylglycerols, phospholipids, triglycerides, wax esters and cholesterol esters. Interestingly, the targeted disruption of the mouse SCD1 gene triggers an increase in the expression of genes involved in the \( \beta \)-oxidation of fatty acids and a decrease in the expression of genes involved in lipogenesis [14]. Therefore, these SCD1 knockout mice exhibit augmented energy expenditure and reduced body adiposity [14], a situation that is reminiscent of the metabolic phenotype of mutant SOD1 mice [9]. Despite sharing common features at the metabolic level, it is not known whether a decrease in SCD1 expression could be implicated by any means in the maintenance of muscle function and, perhaps, in motor neuron degeneration. In this work, first we characterized the expression of SCD1 in muscles from mutant SOD1 mice as well as in experimentally denervated muscles. Second, we studied the muscle phenotype of mice deficient in SCD1. Third, we analyzed the impact of the absence of SCD1, as obtained by both genetic and pharmacological means, on the recovery of muscle function in response to transient nerve lesion. We conclude that the systemic down-regulation of SCD1 promotes muscle oxidative metabolism and accelerates muscle function recovery after nerve injury, thus providing evidence for a new role of this enzyme in modulating the restorative potential of skeletal muscles. These findings therefore

**Figure 1. SCD1 expression and activity in ALS mouse muscle.**

(A) Time course of SCD1 expression in gastrocnemius (GT, brown columns) and tibialis anterior (TA, orange columns) from SOD1(G86R) mice at indicated ages. Wild-type expression is represented by 100% baseline. ***P<0.001 (One sample t-test, n = 5–11). (B) C18:1/C18:0 fatty acid ratio in gastrocnemius and tibialis anterior from SOD1(G86R) mice (brown columns) and wild-type littermates (white columns) at indicated ages. *P<0.05 (1-way ANOVA followed by Bonferroni’s multiple comparison test for gastrocnemius, and unpaired t-test for tibialis anterior, n = 3–10).
doi:10.1371/journal.pone.0064525.g001

**Figure 2. SCD1 expression in ALS patient muscle and after nerve injury.**

(A) Expression of SCD1 and SCD5 in deltoid muscle biopsies from ALS patients and healthy subjects (CT, white columns), as identified by microarray analysis of the database deposited at http://www.ebi.ac.uk/arrayexpress/ (accession number E-MEXP-3260) [12]. ALS samples were obtained from muscle not clinically or electromyography affected (Unaff, orange columns) and from muscle with advanced pathology, characterized by reduced strength and neurogenic electromyography pattern (Aff, brown columns). *P<0.05 (1-way ANOVA followed by Tukey’s multiple comparison test, n = 4–10). (B) Expression of SCD1 in gastrocnemius following sciatic nerve axotomy (Axo) or crush at indicated post-operation days. Contralateral muscle expression is represented by 100% baseline. **P<0.01, ***P<0.001 (One sample t-test, n = 4–10).
doi:10.1371/journal.pone.0064525.g002
Materials and Methods

Animals

FVB/N males overexpressing the murine G86R SOD1 mutation [15], and C57BL/6 males knockout for the SCD1 gene (The Jackson Laboratory, Bar Harbor, ME) were maintained in our animal facility at 23°C with a 12 hours light/dark cycle. They had water and regular A04 rodent chow ad libitum. SOD1(G86R) mice were 60-, 75-, 90- and 105 days of age. SCD1 knockout mice were 4–6 months old. The corresponding non-transgenic male littermates served as controls. To induce peripheral nerve injury, mice were anesthetized with 100 mg/kg body mass ketamine chlorhydrate and 5 mg/kg body mass xylazine. The sciatic nerve was exposed at the midthigh level, and crushed with a fine forceps contralateral to the lesion served as controls [16]. To induce SCD was sutured, and mice were allowed to recover. Hind limbs were 60-, 75-, 90- and 105 days of age. SCD1 knockout mice were 60- , 75-, 90- and 105 days of age. SCD1 knockout mice were 4–6 months old. The corresponding non-transgenic male littermates served as controls. To induce peripheral nerve injury, mice were anesthetized with 100 mg/kg body mass ketamine chlorhydrate and 5 mg/kg body mass xylazine. The sciatic nerve was exposed at the midthigh level, and crushed with a fine forceps contralateral to the lesion served as controls [16]. To induce SCD was sutured, and mice were allowed to recover. Hind limbs were crushed with a fine forceps contralateral to the lesion served as controls [16]. To induce SCD was sutured, and mice were allowed to recover. Hind limbs were crushed with a fine forceps contralateral to the lesion served as controls [16]. To induce SCD was sutured, and mice were allowed to recover. Hind limbs were crushed with a fine forceps contralateral to the lesion served as controls [16]. To induce SCD was sutured, and mice were allowed to recover.
oxidative muscle fibers. Fourteen-μm serial sections were obtained by cutting isopentane fresh-frozen tibialis anterior muscles (brown columns) and wild-type littersmates (white columns). *P<0.05, **P<0.01 (Unpaired t-test, n = 3–11).

Indirect calorimetry

O2 consumption and CO2 production were measured using an open-circuit indirect calorimetry system (Klogor, Lannion, France), as previously described [9]. Concentrations of O2 and CO2 in the outgoing air were successively measured in five different cages. The system was rinsed for 90 s between each measurement. Final values of gas concentrations were the mean of 10 measures obtained during 40 s. Each cage was sampled every 11 min, and one cage was left vacant as reference of ambient gas concentrations. Measurements were performed continuously over 23 hours and a half, a 30-min period being required for calibration of the O2 and CO2 analyzers. In total, 127 measures...
Results

Statistical analysis

Electromyography

were collected per day and mouse. The respiratory quotient was the ratio of CO₂ production over O₂ consumption.

Electromyography

Recordings were obtained with a standard electromyography apparatus (Dantec, Les Ulis, France), in accordance with the guidelines of the American Association of Electrodiagnostic Medicine. Mice were anesthetized as indicated above and kept under a heating lamp to maintain a physiological muscle temperature (at about 31°C). A concentric needle electrode (no. 9013S0011, diameter 0.3 mm; Medtronic, Minneapolis, MN) was inserted in the gastrocnemius, and a monopolar needle electrode (no. 9013R0312, diameter 0.3 mm; Medtronic) was inserted into the tail of the mouse to ground the system. Each muscle was monitored in four different regions, and the degree of denervation was scored as the number of regions with spontaneous activity expressed in percentage. Spontaneous activity was differentiated from voluntary activity by visual and auditory inspection. Only spontaneous activity with a peak-to-peak amplitude of at least 50 μV was considered to be significant.

Statistical analysis

Unless otherwise indicated, data are expressed as the mean ± SEM. PRISM version 5.0a software (GraphPad, San Diego, CA) was used for statistical analysis. Tests are indicated in the legends under the figures. Differences with P-values of at least less than 0.05 were considered significant.

Results

SCD1 expression is altered in ALS muscle

On the basis of our previous microarray data, obtained from a transgenic mouse model of mutant SOD1-linked familial ALS [11], in this study we investigated the significance of the down-regulation of SCD1 for the metabolic capacity of muscles and their response to injury. The expression of SCD1 in the gastrocnemius of SOD1(G86R) mice, which are affected by a progressive denervation atrophy [19], was already diminished at 60 days of age. In this respect, it is noteworthy to mention that our previous electromyography studies on this mouse line revealed that the amplitudes of the compound muscle action potentials, a reduction of which typically reflects a decrease in the number of functional motor units, were normal at the age of 75 days. In addition, mice did not present at this age any abnormal spontaneous electrical activity, which would have reflected the common response of muscle to loss of innervation [16]. According to these findings, we can conclude that SCD1 down-regulation occurred precociously in our SOD1(G86R) mouse model. We then showed here that the decrease in SCD1 expression also persisted during the course of the disease, at 90 days of age, when muscle denervation becomes detectable and motor deficits usually arise, and at about 105 days of age, when hind legs start to be paralysed. At that moment, the decrease in SCD1 expression was also noticeable in the tibialis anterior, which is another muscle in the mouse hind leg displaying less oxidative metabolism than the gastrocnemius (Figure 1A). As a consequence of the repression of muscle SCD1 expression, we observed that the C18:1/C18:0 fatty acid ratio, an index of the desaturation activity of the enzyme [20], was slightly reduced in a presymptomatic muscle extracts but significantly diminished at the end stage in both gastrocnemius and tibialis anterior (Figure 1B). It is noteworthy to mention that our previous studies had shown that SOD1(G86R) mice typically exhibit decreased postprandial lipidemia and increased peripheral clearance of lipids, both of which can be ascribed to muscle hypermetabolism [10]. Therefore, an excess of uptake of exogenous lipids in this tissue could mask otherwise earlier and more robust differences in the index of SCD activity.

To obtain independent evidence that SCD1 down-regulation is a typical feature of ALS, we took advantage of our transcriptome database composed of deltoid biopsies from patients with the sporadic form of the disease [12]. The expression of not only SCD1 but also SCD5, a primate-specific enzyme variant with identical function [21], was lower in ALS patients, as compared to normal control subjects. Furthermore, the repression of SCD1 expression was much more remarkable in a muscle not clinically or electromyography affected than in a muscle at an advanced stage of pathology, characterized at the clinical level by reduced strength and neurogenic electromyography pattern (Figure 2A). That SCD1 down-regulation could be observed both in presymptomatic SOD1(G86R) mouse muscle and in relatively healthy human ALS muscle prompted us to speculate that such a pattern of expression might not be solely related to the loss of muscle innervation characteristic of the disease. To address this question, we compared SCD1 expression in gastrocnemius submitted to acute denervation, as obtained by cutting and removing several millimeters of the sciatic nerve, or transient denervation followed by re-innervation, as obtained by crushing the sciatic nerve for several seconds. Under these conditions, the expression of SCD1 was increased after axotomy but significantly reduced after crush (Figure 2B). Overall, these findings provide evidence for the implication of SCD1 in the pathological process triggering ALS.
and suggest that SCD1 down-regulation could be involved in the restoration of muscle function in response to injury.

**SCD1 knockout mice do not manifest motor impairment but display exacerbated muscle metabolic oxidative capacity**

To gain insight into the way in which the lack of SCD1 expression impacts on muscle function, we investigated several characteristics of muscles in these SCD1 knockout mice reflecting their metabolic status, and also evaluated their motor behavior. At the molecular level, we measured the expression of PGC1-α, PPARγ and PDK4, of which an increase is involved in stimulating mitochondrial biogenesis and in switching the energy source from glucose to fatty acids [22]. The expression of these genes was significantly higher in the gastrocnemius of SCD1 knockout mice, as compared to wild-type littermates; in the tibialis anterior, there was also a trend toward an increased expression (Figure 3A). Despite this latter attenuated response, the tibialis anterior represents, better than the gastrocnemius, a typical example of glycolytic muscle in which to evaluate changes in the relative density of the various fiber types. We therefore used this muscle to determine potential morphological and biochemical changes triggered by the absence of SCD1. The number of fibers per muscle section was higher in SCD1 knockout mice than in wild-type mice (Figure 3B). Accordingly, the distribution of fiber calibers showed an increase in the amount of fibers of small caliber in SCD1 knockout mice (Figure 3C). We extended these findings by performing SDH histochemistry, and found that the average cross-sectional area of both SDH-positive and SDH-negative fibers was smaller in SCD1 knockout mice than in wild-type mice (Figure 3D). These differences were associated in SCD1 knockout mice with a significant predominance of SDH-positive fibers, which are characterized by a higher metabolic oxidative capacity (Figure 3E).

Evaluation of muscle function using the grip strength test revealed no changes in the force developed by hind limbs between SCD1 knockout mice and their wild-type littermates (0.37 ± 0.024 N in SCD1 knockout mice versus 0.37 ± 0.021 N in wild-type mice, n = 7). Along with this, no abnormal spontaneous electrical activity, which would have reflected the typical response of muscle to loss of innervation, was found in the gastrocnemius of SCD1 knockout mice (data not shown). In contrast, we also measured the expression of a series of genes specific to the motor end plate, including the acetylcholine receptor subunits α, γ and ε (AChR-α, AChR-γ and AChR-ε, respectively), and muscle-specific receptor tyrosine kinase (MunSK). Except for AChR-γ, of which an increase would have been considered a sign of muscle denervation [23], the expression of these genes was significantly increased in the gastrocnemius of SCD1 knockout mice as compared to their wild-type littermates, although the changes were less pronounced in the tibialis anterior (Figure 4). In all, these results indicate that the genetic ablation of SCD1 is not detrimental per se to muscle function but promotes a metabolic shift toward a more oxidative capacity, and stimulates the neuromuscular junction gene expression program.

**SCD deficiency accelerates muscle function recovery after nerve injury**

As shown above, the lack of SCD1 expression does not represent a handicap for muscle function. Therefore, its down-regulation, as observed in ALS or after nerve crush, prompts us to hypothesize that the enzyme may participate in the restorative efforts that muscles experience at the early stages of disease, when neuromuscular deterioration is not generalized yet, or during the process of recovery following a brief disruption of the neuromuscular communication. To address this question, we took advantage of such a model of transient denervation and re- innervation as a means to evaluate, by performing relatively manageable short-term experiments [3], the importance of SCD1 for the restoration of muscle function in response to nerve damage. We performed these experiments using not only SCD1 knockout mice but also mice deficient in SCD enzymatic activity, as obtained by feeding them with MF-438, which is an orally bioavailable pharmacological agent inhibiting SCD-dependent desaturation of fatty acids [17]. To verify if our treatment was biologically active in vivo, we measured several parameters that should reflect the deficiency in SCD enzymatic activity. First, MF-438 significantly reduced both C16:1/C16:0 and C18:1/C18:0 fatty acid ratios in circulating lipids (Figure 5A). Second, MF-438 also induced a decrease in the respiratory quotient as determined by indirect calorimetry, which indicated a switch of the energy source from glucose to fatty acids (Figure 5B). Finally, the drug triggered concomitantly a small but significant decrease in body mass of mice treated for two weeks (Figure 5C). SCD deficiency did not alter hind limb grip strength during the 2-week treatment, as determined either by a percentage of peak force at day 0 (Figure 5D) or by normalizing peak force to body mass (Figure 5E). Also, there were no detectable electromyography abnormalities suggestive of denervation (data not shown). Furthermore, MF-438 stimulated the expression of several genes specific to the motor end plate (Figure 5F). Overall, these findings strongly support that the effects of the SCD inhibitor are very similar to those observed in SCD1 knockout mice.

To monitor the recovery of muscle function after crushing the sciatic nerve, we measured hind limb grip strength during a post-lesion period of two weeks, and found that the force in SCD1 knockout mice was restored to its initial level more rapidly than in their wild-type littermates. Accordingly, the proportion of abnormal electromyography episodes reflecting neurogenic muscle denervation was lower in SCD1 knockout mice at 14 days post-lesion (Figure 6A). We also measured at that time the relative density of muscle fiber types as a witness to the restorative process. In the denervated tibialis anterior of wild-type mice, the distribution of fiber types in the denervated gastrocnemius of SCD1 knockout mice (data not shown).

**Discussion**

SCD1 is an essential lipogenic enzyme thought to be implicated in the development of obesity and associated metabolic disorders [24]. In this study, we have shown that its expression is down-regulated in skeletal muscles suffering from slowly progressing...
motor neuron degeneration, as seen in ALS, and from transitory denervation/re-innervation, as obtained experimentally by sciatic nerve crush. Based on these findings, we have also shown here that the systemic down-regulation of SCD1, as generated by both genetic and pharmacological means, enhances the oxidative metabolism of muscles, stimulates the expression of synaptic genes, and ameliorates the restoration of muscle function following transient denervation and subsequent re-innervation.

SCD1 knockout mice are mainly characterized by decreased adiposity and increased metabolic rate [14], two characteristics of the mutant SOD1 mouse model of ALS, even observed before the onset of any motor neuropathy [9]. The lack of SCD1 expression has been associated with inhibition of lipogenesis and stimulation of mitochondrial β-oxidation of fatty acids [25]. Notably, we had previously observed that the expression of genes involved in the uptake of fatty acids, such as FAT/CD36 and other related genes, was presymptomatically increased in skeletal muscle of SOD1(G86R) mice [10]. In addition, it is known that the muscle-specific overexpression of FAT/CD36 is sufficient to trigger fatty acid oxidation [26]. It seems therefore not very surprising that, in accordance with an enhanced uptake of fatty acids, the muscles of SOD1(G86R) mice exhibit concomitantly a decrease in the expression of SCD1. The situation appears to be less straightforward in the case of ALS patients. Although most of them are markedly hypermetabolic [6], as in the animal model, they usually present with increased levels of circulating lipids [7], which would not intuitively substantiate the hypothesis of the muscle down-regulation of SCD1. It is noteworthy, however, that the expression of SCD1 in patients was much more repressed in muscles not clinically or electromyography affected, which lets envisage that SCD1 down-regulation might occur as a result of whatever mechanism preceding overt disease. The fact that SCD1 down-regulation was observed early in presymptomatic SOD1(G86R) mouse gastrocnemius and in relatively healthy

Figure 5. Effects of MF-438 on metabolism and muscle function. (A) C16:1/C16:0 and C18:1/C18:0 fatty acid ratio in plasma from MF-438 treated mice (brown columns) and control littermates (CT, white columns). ***P<0.001 (Unpaired t-test, n = 4–6). (B) Time course of respiratory quotient (RQ) before and after treatment with MF-438 at a dose of 10 mg/kg body mass/day (indicated by the black bar) (n = 4). Time course of body mass (C), muscle grip strength expressed as a percentage of day 0 (D), and specific grip strength, as determined by normalizing peak force to body mass (E), in mice fed regular chow (white circles) and mice fed regular chow supplemented with MF-438 (black circles). **P<0.01 (2-way ANOVA followed by Bonferroni test, n = 5–6). (F) Expression of PGC1-α, AChR-α, and MuSK in gastrocnemius from MF-438 treated mice (brown columns) and control littermates (white columns). *P<0.05 (Unpaired t-test, n = 3–9).

doi:10.1371/journal.pone.0064525.g005
human ALS deltoid suggests that the transcriptional regulation of SCD1 might not be (solely) related to the loss of innervation of muscles characteristic of these conditions. In support of this notion, such a down-regulation of SCD1 expression was not found after severe denervation, at least at a time when re-innervation was not present according to our protocol of axotomy. In contrast, a more subtle denervation, as that obtained by crushing the sciatic nerve for only several seconds, simulated the inhibitory effect of ALS on SCD1 expression. Because sciatic nerve crush allows the rapid recovery (in around two weeks) of muscle function, we speculate that SCD1 down-regulation may be somehow related to the restorative potential of skeletal muscles.

As a first step toward the understanding of the importance of SCD1 for muscle function, we found that the genetic ablation of SCD1 promoted a decrease in the cross-sectional area of muscle fibers, and an increase in the amount of those enriched in the mitochondrial enzyme SDH. In parallel, we also observed increased expression of PGC1-α, PPARα and PDK4. Overall, these observations provide persuasive evidence for a higher metabolic oxidative capacity, in accordance with several previous studies. For instance, overexpressing PGC1-α has been reported to trigger a decrease in the size of muscle fibers and a concomitant fast to slow fiber type shift [27]. In contrast, knocking out PGC1-α specifically in muscle has been shown to induce a shift from oxidative to glycolytic muscle fibers [28]. Moreover, PGC1-α not only triggers metabolic changes in muscle but also activates the expression of genes specific to the motor end plate [29]. Therefore, the increased expression of the signaling kinase MuSK and several acetylcholine receptor subunits that we observed in our SCD1 knockout mice could be the consequence of a higher transcriptional activity involving, at least in part, PGC1-α. Despite these modifications, the lack of SCD1 did not cause any alteration of muscle function as determined by behavioral and electrophysiological means. Furthermore, inhibiting global SCD enzymatic activity with a diet containing MF-438 induced very similar changes as those found in SCD1 knockout mice.

Subsequent efforts were concentrated in determining if SCD1 down-regulation might affect muscle function when challenged by sciatic nerve crush. Both SCD1 knockout mice and MF-438 treated mice recovered their force more rapidly than their corresponding control groups. Two weeks after lesion, the degree of denervation was more important in these control groups than in SCD-deficient mice. In support of these findings, very early studies had already reported that oxidative muscles can recuperate better
than glycolytic ones after nerve crush [30]. Also, PGC1-α has been shown to protect muscles from denervation atrophy [31]. It can be therefore postulated that the pro-restorative power of SCD deficiency is the result of an enhancement of oxidative metabolism, as that shown in this work. A fast to slow fiber type shift has been shown to occur progressively in ALS, and it has been proposed that this metabolic modification would render muscle fibers with higher oxidative capacity more resistant to disease [32]. Recent studies have further reported that a modest increase in the expression of MuSK, as that observed here, can maintain neuromuscular junctions in mutant SOD1 mice, hence retarding denervation and ameliorating muscle function [33]. Taking our present findings as a whole, we can put forward that SCD1 stands at a regulatory crossroad shaping muscle function in health and disease. In this respect, the beneficial effects of inhibitors of SCD enzymatic activity could pave the way for developing novel therapeutic strategies to palliate motor neuron injury.

Acknowledgments

We thank Annie Piccininna and Marie Jose Ruivo for excellent technical assistance. We are also indebted to Dr. Benoit Halter for his aid with part of the initial SOD1(G93R) mouse experiments.

Author Contributions

Conceived and designed the experiments: GH FS JLGDA JPL. Performed the experiments: GH FS AH TL FR SDG LP. Analyzed the data: GH FS AH TL FB SDG HO LD EM JLGDA. Contributed reagents/materials/analysis tools: FB HO MHMB EM. Wrote the paper: JLGDA.

References

1. Kierman MC, Vuic S, Cheal BC, Turner MR, Eisen A, et al. (2011) Amyotrophic lateral sclerosis. Lancet 377: 942–953.
2. Boillère S, Yannanaka K, Lobisier CS, Copeland NG, Jenkins NA, et al. (2006) Onset and progression in inherited ALS determined by motor neurons and microglia. Science 312: 1389–1392.
3. Dupuis L, Gonzalez De Aguilar JL, Echaniz-Laguna A, Escobach J, Rene F, et al. (2009) Muscle mitochondrial uncoupling dismantles neuromuscular junction and triggers distal degeneration of motor neurons. PLoS One 4: e5390. doi:10.1371/journal.pone.0005390.
4. Wong M, Martin LJ (2010) Skeletal muscle-restricted expression of human PGC1α protects mice against adiposity. Proc Natl Acad Sci U S A 107: 18116–181111.
5. Routeloup C, Desport JC, Clavelou P, Guy N, Derumeaux-Burel H, et al. (2009) Hypermetabolism in ALS patients: an early and persistent phenomenon. J Neurol 256: 1236–1242.
6. Dupuis L, Corcia P, Fergani A, Gonzalez De Aguilar JL, Bonnefont-Rousselot D, et al. (2008) Dyslipidemia is a protective factor in amyotrophic lateral sclerosis. Neurology 70: 1004–1009.
7. Dupuis L, Pradat PF, Ludolph AC, Loeffler JP (2011) Energy metabolism in amyotrophic lateral sclerosis. Lancet Neurol 10: 75–82.
8. Dupuis L, Oudart H, Rene F, Gonzalez De Aguilar JL, Loeffler JP (2004) Evidence for defective energy homeostasis in amyotrophic lateral sclerosis: benefits of a high-energy diet in a transgenic mouse model. Proc Natl Acad Sci U S A 101: 11159–11164.
9. Fergani A, Oudart H, Gonzalez De Aguilar JL, Fricker B, Rene F, et al. (2007) Increased peripheral lipid clearance in an animal model of amyotrophic lateral sclerosis. J Lipid Res 48: 1571–1580.
10. Gonzalez De Aguilar JL, Niederhauser-Wiederkehr C, Halter B, Gonzalez De Aguilar JL, Bonnefont-Rousselot D, et al. (2009) Gene expression of skeletal muscle in an amyotrophic lateral sclerosis mouse model. Physiol Genomics 32: 207–218.
11. Pradat PF, Dubourg O, de Tapia M, di Scala F, Dupuis L, et al. (2012) Muscle gene expression is a marker of amyotrophic lateral sclerosis severity. Neurodegener Dis 9: 38–52.
12. Enoch HG, Català A, Strittmatter P (1976) Mechanism of rat liver microsomal stearoyl-CoA desaturase. Studies of the substrate specificity, enzyme-substrate interactions, and the function of lipid. J Biol Chem 251: 5093–5103.
13. Ntambi JM, Miyazaki M, Steoch JP, Luan H, Krendzicki CM, et al. (2002) Loss of stearoyl-CoA desaturase-1 function protects mice against adiposity. Proc Natl Acad Sci U S A 99: 11462–11466.
14. Rippke ME, Huntley GW, Hof PR, Morrison JH, Gordon JW (1995) Transgenic mice expressing an altered murine superoxide dismutase gene provide an animal model of amyotrophic lateral sclerosis. Proc Natl Acad Sci U S A 92: 689–693.
15. Halter B, Gonzalez De Aguilar JL, Rene F, Petri S, Fricker B, et al. (2010) Oxidative stress in skeletal muscle stimulates early expression of Rand in a mouse model of amyotrophic lateral sclerosis. Free Radic Biol Med 48: 915–923.
16. Halter B, Gonzalez De Aguilar JL, Rene F, Petri S, Fricker B, et al. (2010) Phenotypically aberrant astrocytes that promote motoneuron damage in a mouse model of amyotrophic lateral sclerosis. J Neurol 256: 1236–1242.
17. Leser S, Black WC, Deschenes D, Dolman S, Falgueyret JP, et al. (2010) Synthesis and biological activity of a potent and orally bioavailable SCD inhibitor (MF-438). Bioorg Med Chem Lett 20: 499–502.
18. Bligh EG, Dyer WJ (1959) A rapid method of total lipid extraction and purification. Can J Biochem Physiol 37: 911–917.
19. Roua C, Pasieleva I, Rene F, Gonzalez De Aguilar JL, Echaniz-Laguna A, et al. (2007) Sodium valproate exerts neuroprotective effects in vivo through CREB-binding protein-dependent mechanisms but does not improve survival in an amyotrophic lateral sclerosis mouse model. J Neurosci 27: 5535–5545.
20. Attie AD, Krauss RM, Gray-Keller MP, Brown D, Miyazaki M, et al. (2008) Stearoyl-CoA desaturase-1 deficiency reduces ceramide synthesis by downregulating serine palmitoyltransferase and increasing beta-oxidation in skeletal muscle. Am J Physiol Endocrinol Metab 298: E599–E607.
21. Blachet E, Annicotte JS, Lagarrigue S, Aguilar V, Clapi C, et al. (2011) E2F transcription factor-1 regulates oxidative metabolism. Nat Cell Biol 15: 1146–1152.
22. Martinou JC, Merlie JP (1991) Nerve-dependent modulation of acetylcholine receptor epsilon-subunit gene expression. J Neurosci 11: 1291–1299.
23. Sampath H, Ntambi JM (2011) The role of stearoyl-CoA desaturase in obesity, insulin resistance, and inflammation. Ann N Y Acad Sci 1243: 47–53.
24. Dobrzan A, Dobrzan P, Lee SH, Miyazaki M, Cohen P, et al. (2005) Stearoyl-CoA desaturase-1 deficiency reduces ceramide synthesis by downregulating serine palmitoyltransferase and increasing beta-oxidation in skeletal muscle. Am J Physiol Endocrinol Metab 298: E599–E607.
25. Ibrahim A, Bosen A, Blinn WD, Hajri T, Li X, et al. (1999) Muscle-specific overexpression of FAT/CID3 enhances fatty acid oxidation by contracting muscle, reduces plasma triglycerides and fatty acids, and increases plasma glucose and insulin. J Biol Chem 274: 26761–26766.
26. Selby JT, Morine KJ, Penduk K, Barton ER, Sweeney HL (2012) Rescue of dystrophic skeletal muscle by PGC-1α involves a fast to slow fiber type shift in the mdx mouse. PLoS One 7: e30063. doi:10.1371/journal.pone.0030063.
27. Handschin C, Chen S, Li F, Liu F, Marazzato E, et al. (2007) Skeletal muscle fiber-type switching, exercise intolerance, and myopathy in PGC-1α/l-function specific knock-out animals. J Biol Chem 282: 30014–30012.
28. Handschin C, Kobayashi YM, Chin S, Scale P, Campbell KP, et al. (2007) PGC-1α regulates the neuromuscular junction program and ameliorates Duchenne muscular dystrophy. Genes Dev 21: 770–783.
29. Lowrie MB, Krishnan S, Vrbova G (1982) Recovery of slow and fast muscles following nerve injury during early post-natal development in the rat. J Physiol 331: 51–66.
30. Sandri M, Lin J, Handschin C, Yang W, Arany ZP, et al. (2006) PGC-1alpha regulates the neuromuscular junction program and ameliorates dystrophic skeletal muscle. Neuron 331: 51–66.
31. Leger S, Black WC, Deschenes D, Dolman S, Falgueyret JP, et al. (2010) Synthesis and biological activity of a potent and orally bioavailable SCD inhibitor (MF-438). Bioorg Med Chem Lett 20: 499–502.