Impaired age-associated mitochondrial translation is mitigated by exercise and PGC-1α

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Abstract

Sarcopenia, the age-related loss of skeletal muscle mass and function, can dramatically impinge on quality of life and mortality. While mitochondrial dysfunction and imbalanced proteostasis are recognized as hallmarks of sarcopenia, the regulatory and functional link between these processes is underappreciated and unresolved. We therefore investigated how mitochondrial proteostasis, a crucial process that coordinates the expression of nuclear- and mitochondrial-encoded mitochondrial proteins with supercomplex formation and respiratory activity, is affected in skeletal muscle aging. Intriguingly, a robust mitochondrial translation impairment was observed in sarcopenic muscle, which is regulated by the peroxisome proliferator-activated receptor γ coactivator 1 alpha (PGC-1α) with the estrogen-related receptor α (ERRα). Exercise, a potent inducer of PGC-1α activity, rectifies age-related reduction in mitochondrial translation, in conjunction with quality control pathways. These results highlight the importance of mitochondrial proteostasis in muscle aging, and elucidate regulatory interactions that underlie the powerful benefits of physical activity in this context.

Significance statement

Dysregulation of mitochondrial function and cellular proteostasis comprises two key mechanisms in the development of sarcopenia. In this context of pathological muscle aging, the link and dependencies between these fundamental processes are however unclear. We show here that the mitochondria in sarcopenic muscle are under proteostatic imbalance, primarily due to a selective deficiency in mitochondrial translation. PGC-1α and ERRα were identified to jointly regulate the expression of key components of the mitochondrial translation machinery. Accordingly, targeted elevation of PGC-1α, as well as induction in the context of exercise, rectify mitochondrial protein synthesis and, moreover, improve protein quality control in skeletal muscle. These results provide a mechanistic connection between proteostasis and mitochondrial function in physiological and pathological muscle aging.

Main text

Introduction

Sarcopenia, the age-related loss of skeletal muscle mass and function, poses a major societal and economic burden, and constrains the quality of life in the elderly(1). Sarcopenic individuals are at higher risk of frailty, falls, loss of independence, morbidity and mortality(2, 3). Demographic population shifts and an increasing life expectancy continuously escalates the prevalence and impact of sarcopenia in modern societies(4). The risk for and the pathoetiology of sarcopenia are multifactorial, and still only poorly understood. The loss of protein homeostasis (proteostasis) and mitochondrial dysfunction are two central aspects that are broadly associated with the aging process in most tissues. For example, sustained aberrant activation of mammalian target of rapamycin (mTOR) results in abnormal protein synthesis, as well as blunted protein degradation and clearance of defective organelles by autophagy in skeletal muscle(5). Similarly, irregular mitochondrial morphology, reduced bioenergetic efficiency, and increased production of reactive oxygen species (ROS) are hallmarks of mitochondrial dysfunction in sarcopenia(6). However, the link between cellular proteostasis and mitochondrial dysfunction in aging has been investigated only in a rudimentary manner. Importantly, as the assembly of the electron transport chain (ETC) supercomplex and the resulting mitochondrial respiratory function depend on proteins encoded both in the nuclear and the mitochondrial genomes, the synthesis and import of nucleus-encoded proteins have to be tightly matched to the availability of the corresponding mitochondria-encoded counterparts. Moreover, tightly balanced rates of mitochondrial translation, quality control and protein degradation are crucial to ensure adequate functionality(7). The formation and function of the ETC thus depends on both cytosolic and mitochondrial proteostasis. So far, most studies have primarily focused on the functional deterioration of the ETC, and its correlation with reduced mitochondrial DNA (mtDNA) abundance and general expression of ETC genes in skeletal muscle aging and sarcopenia(8, 9). In contrast, the role of mitochondrial proteostasis in this context remains underappreciated. It has been demonstrated that with advancing age, the overall capacity to counteract mitochondrial proteostatic insults declines(10). The reduced expression of the Lon Protease (LonP) in aged muscle, whose inadequate expression is associated with impaired mitochondrial function and loss of muscle mass and function such as in disuse, underlines the importance of mitochondrial proteostasis in aging(11-13).

In the present study, we used transcriptomic data of young and old mice to estimate the age-related changes in muscle mitochondrial proteostasis. Furthermore, skeletal muscle-specific gain- and loss-of-function of the peroxisome proliferator-activated receptor γ coactivator 1α (PGC-1α) were included in the analysis to assess the impact of higher and lower levels, respectively, of this key regulator of mitochondrial biogenesis and oxidative metabolism in this paradigm. Finally, based on the strong involvement of PGC-1α in controlling exercise adaptations, and the potent benefits of physical activity on sarcopenia and muscle aging, mitochondrial proteostasis was studied in untrained and trained aged muscle.

Results

Mitochondrial translation is a central transcriptional program associated with muscle aging

Transcriptionally regulated biological programs associated with the aging process were uncovered by RNA sequencing (RNA-seq) of muscle of 3- and 24-month-old mice (3mo & 24mo). These genes were furthermore stratified according to the dependency of loss- and gain-of-function of muscle PGC-1α by comparing wildtype (WT) to PGC-1α muscle-specific knockout (mKO), and PGC-1α muscle-specific transgenic (mTG) mice, respectively. Principal component (PC) analysis revealed strong impacts of relative PGC-1α expression (PC1) and age (PC2) (Fig. 1a). We found 860 genes that were significantly aligned with either PC1, 2, or both. When comparing these genes based on Z scores calculated from gene expression counts over all six conditions, a group of genes (cluster 1) emerged in which both age and PGC-1α affect transcription (Fig. 1b). The expression of most of the genes in this cluster was reduced in aging in WT mice. This pattern was largely recapitulated in young mKO animals, thus showing a 'premature aging' signature, and mirrored by the mitigation of the age-associated drop in transcript levels in mTG mice. Notably, cluster 1 largely consists of mitochondrial genes that are involved in various mitochondrial metabolic processes (Suppl. Fig. 1a, Fig. 1c). Interestingly, two of the five most enriched gene ontology (GO) terms contained genes encoding mitochondrial translation proteins and mitochondrial ribosomal RNAs (Fig. 1d-e). Adenoviral overexpression studies in C2C12 myotubes confirmed the ability of PGC-1α to increase the expression of mitochondrial translation genes, including that of mitochondrial ribosomal proteins, as seen in mTG mice *in vivo* (Fig. 1f).

PGC-1α promotes mitochondrial protein synthesis

To determine whether the transcriptional changes of mitochondrial translation genes lead to higher protein expression, we interrogated published proteomic data of PGC-1α overexpression in C2C12 myotubes(14), revealing mitochondrial translation components prominently amongst the PGC-1α-responsive proteins (Fig. 2a, genes marked in black). Translation was accordingly enriched as ontological category (three terms in the top ten) in this dataset, based on a strong contribution of mitochondrial translation protein (Fig. 2b). Next, we assessed whether these changes in gene expression and protein levels culminate in altered mitochondrial protein synthesis rates *in vitro*. Rates of mitochondrial translation were determined by analyzing ^{35}S-methionine incorporation into newly synthesized mitochondrial proteins in PGC-1α overexpressing C2C12 myotubes. Emetine was used as pharmacological inhibitor of cytosolic translation to allow the exclusive visualization of mitochondrial protein synthesis. These experiments demonstrated that PGC-1α boosts mitochondrial protein synthesis rates, notably for proteins of all sizes (Fig. 2c-d).

ERRα is required for PGC-1α-mediated upregulation of mitochondrial translation

To dissect the mechanism of PGC-1α-mediated changes in mitochondrial translation, we utilized Integrated Motif Activity Response Analysis (ISMARA) to identify motifs that are associated with gene regulation(15).

In muscles from 3- and 24-month-old mice, the activity of the Esrrb_Esrra motif closely reflects the level of PGC-1α expression across the different conditions (Fig. 3a). The transcription factor ERRα is required for expression of most PGC-1α target genes and has been implicated in PGC-1α-induced mitochondrial biogenesis(16-18). More recently, reduced Esrrb_Esrra motif activity, linked to mitochondrial function, was reported in skeletal muscle aging conserved across mouse, rat, and human(19). The Esrrb_Esrra motif activity in PGC-1α overexpression studies in myotubes(14) was fully consistent with the motif activity inferred from the *in vivo* data in mTG and mKO mice (Fig. 3a). Predicted target genes of this motif are not only linked to various mitochondrial metabolic processes, but also translation and the ribosome (Suppl. Fig. 2b-c). To assess whether these two factors directly interact at specific promoters, we compared their genomic localization using ChIP-seq data of PGC-1α and ERRα in PGC-1α-overexpressing C2C12 myotubes(18, 20). Strikingly, for the mitochondrial translation genes identified by GO analysis of in vitro data(14), all of the PGC-1α recruitment coincided with ERRα DNA binding (Fig. 3b-c). To test the functional requirement for ERRα in the PGC-1α-mediated upregulation of mitochondrial translation genes, the activity of ERRα was inhibited with the inverse agonist XCT790(16). The PGC-1α-induced upregulation of most mitochondrial translation genes was ablated when exposing C2C12 myotubes to 10μM XCT790 (Fig. 3d). Accordingly, the only significant difference in mitochondrial translation rates was found with PGC-1α + DMSO, but not upon XCT790 exposure when compared to the LacZ + DMSO control. These results indicate that the genomic co-recruitment and function interaction of PGC-1α and ERRα are centrally involved in the regulation of mitochondrial translation (Fig. 3e-f).

The aging-induced reduction of mitochondrial translation is mitigated by exercise and PGC-1α in vivo

To confirm the biological relevance of our *in vitro* findings, we studied mitochondrial translation in muscle of 28-month-old (28mo), WT mice, which classify as sarcopenic at this age(19) and showed reduced mass of the *extensor digitorum longus* muscle (Suppl. Fig. 3a). We found that transcript levels of mitochondrial translation genes were reduced in sarcopenic skeletal muscle (Fig. 4a), in parallel to mRNA expression of ERRα and PGC-1α4 (Suppl. Fig. 3b-c). Importantly, *in organello* mitochondrial translation rates, measured by incorporation of ^{35}S-methionine into newly synthesized proteins in mitochondria isolated from skeletal muscle, were also lower in sarcopenic mice (Fig. 4b). We hypothesized that reduced mitochondrial translation challenges mitochondrial proteostasis as this may cause a proportional excess of nuclear-encoded ETC subunits and therefore may engage the mitochondrial unfolded protein response (UPR^{mt}) and protein degradation to restore proteostasis. We however observed that the UPR^{mt} was not responding accordingly in old mice, at least based on gene expression of Ddit3, Atf4 and Atf5, as well as various downstream mitochondrial proteases (Suppl. Fig. 3d). Conversely, ubiquitination of mitochondrial proteins was increased in mitochondria isolated from muscle of old mice (Suppl. Fig. 3e). Lack of physical exercise in mice due to space limitations, likely worsens sarcopenic development in aged mice with regards to muscle function. Thus, we finally assessed how exercise, a stimulus with strong protective effects against muscle aging, affects mitochondrial proteostasis in young and old muscle. Indeed, proteomic data (21) from cohorts of mice that

were trained with either endurance or resistance voluntary running wheels revealed upregulated mitochondrial ribosomal and translation proteins in response to both training paradigms, without indication of differential PGC-1α isoform expression in the late post-exercise state (Fig. 4c-Suppl. Fig. 3f).Importantly, unlike muscle aging, exercise-induced changes in mitochondrial translation are linked to corresponding adaptations in UPR^{mt} markers and mitochondrial proteostasis (Suppl. Fig. 3g). This response is likely regulated by increased PGC-1α activity upon exercise, since mTG animals showed a similar upregulation on the transcriptome level in the sedentary state (Suppl. Fig. 3h). Importantly, mild treadmill training in 25-month-old mice(22) was sufficient to recapitulate these effects. The muscle of these aged, trained mice demonstrated a similar upregulation of mitochondrial translation proteins, along with elevated levels of LonP, Dnaja3 and Hspd1 (Fig. 4d, Suppl. Fig. 3i).

Discussion

Aging of skeletal muscle (and other tissues) is a complex process, involving many different internal and external factors and pathways. Nevertheless, aberrant cytosolic proteostasis, characterized by abnormally elevated muscle mTOR activity, and mitochondrial dysfunctions in respiration are two main hallmarks associated with muscle aging(23). We now have identified mitochondrial proteostasis as a crucial component linking these two events. In this manuscript, we describe a coordinated downregulation of genes and proteins important for mitochondrial translation, resulting in reduced mitochondrial protein synthesis rates. The absence of corresponding changes in mitochondrial protein quality control such as UPR^{mt} and mitochondrial proteases imply an imbalance of mitochondrial proteostasis, which is in turn potentially worsened by increased ubiquitin-dependent protein degradation in aged muscle mitochondria. As a consequence, lower abundance of mitochondrial-encoded ETC proteins is observed. Although not evaluated here, this could contribute to respiratory chain dysfunction that has been broadly reported in aged skeletal muscle(8, 9, 24, 25). Reduced ATP production could in turn impinge on translation and other energy-consuming anabolic processes, thereby leading to a vicious cycle. Pharmacological mitochondrial translation inhibitors such as chloramphenicol and doxycycline reduce ETC subunit expression and, importantly, activity of the ETC, at least *in vitro*(26-29). In addition, severe combined respiratory chain dysfunction is observed in a wide variety of diseases caused by mutations in genes that are essential for mitochondrial translation(30). Previous studies in humans and mice report varying results in protein synthesis rates in skeletal muscle mitochondria(31-34). However, in a large human sarcopenia investigation, a robust reduction of mitochondrial translation, including a cluster of mitochondrial ribosomal proteins, emerged from transcriptomic analyses(35). Importantly, a pathological contribution of impaired mitochondrial translation to muscle function is suggested by observations in muscular denervation and mechanical unloading, even in young mice(36, 37). In disuse, sex-dependent differences exist regarding consecutive mitochondrial adaptations, yet translation was reduced in both male and female mice(37). It is so far unknown if mitochondrial aberrations in sarcopenic muscle are different depending on sex.

PGC-1α and ERRα are key regulators of mitochondrial biogenesis and oxidative phosphorylation(16). Moreover, based on motif activity prediction, a functional reduction of the PGC-1α/ERRα axis was hypothesized to underlie the mitochondrial dysfunction in human sarcopenic muscle(35). We have now implicated PGC-1α and ERRα in the control of mitochondrial translation (Figure 5). PGC-1α coordinates both nuclear- and mitochondrial-encoded gene expression for mitochondrial biogenesis in a specified manner. The latter is achieved through the activation of nuclear respiratory factors 1 and 2 by PGC-1α, which leads to the induction of the mitochondrial transcription factor A (TFAM)(38). Selective activation of different transcription factors by PGC-1α for mitochondrial protein expression from both genomic sources highlights the coordinative role of PGC-1α in protecting mitochondrial proteostasis under baseline conditions and during mitochondrial biogenesis.

In light of these findings, it seems surprising that mild mitochondrial stress caused by targeted ablation of a mitochondrial ribosomal gene and the subsequent induction of UPR^{mt} induction is reported to benefit longevity in various organisms(39-41). For example, reduced expression of mitochondrial ribosomal protein S5 (Mrps5) was described to yield an increased lifespan in B6-by-D2 (BXD) recombinant inbred mice via this mechanism(40). In mammals, UPR^{mt} can inhibit mitochondrial translation, similar to the UPR in the endoplasmic reticulum on cytosolic translation(42). Given that we and others(35) did not observe any upregulation of UPR^{mt} markers in sarcopenic muscle, it is unlikely that the reduced translation rates that we observed are caused by this mechanism. Inversely, a reduced protein folding workload due to lowered translation rates may be a reason why the UPR^{mt} is not activated in aged skeletal muscle. Reduced mitochondrial translation in fibroblasts caused by pharmacological translation inhibitors does not result in UPR^{mt} activation(29). In fact, marker genes of the UPR^{mt} were downregulated, while the integrated stress response was induced(29). The lower expression of mitochondrial ribosome constituent genes that is observed in sarcopenic mice did not yield the beneficial effects described in the genetic models. It is likely that the age- and loss-of-function of PGC-1α-related changes that we observed in a large group of such proteins, as opposed to ablation of only one or very few in the genetic studies, pose a larger challenge that ultimately is detrimental rather than beneficial for a UPR^{mt}-related hormetic response for mitochondrial and cellular health in muscle. Intriguingly, the overexpression of PGC-1α not only rescues the age-associated reductions in mitochondrial translation and ETC activity, but also elevates the transcription of UPR^{mt} genes. Analogous to the coordinated control of ETC activity and ROS detoxifying genes to mitigate potential detrimental consequences arising from higher OXPHOS, the PGC-1α-mediated induction of mitochondrial protein quality control could help to deal with byproducts of enhanced mitochondrial translation, ensuring balanced mitochondrial proteostasis. Since PGC-1α is centrally involved in the response of skeletal muscle to exercise, it is not surprising that in trained animals, we found a similar boost in mitochondrial translation and UPR^{mt}. Increased expression of genes involved in mitochondrial translation in exercised murine muscle was reported previously, with some muscle and fiber type differences(37, 43). Importantly, it has also been shown that mitochondrial translation genes are inducible with exercise in humans. e.g. 12 weeks of exercise training (both resistance and high-intensity interval training (HIIT)), after which an increase in the factional synthesis rate of mitochondrial proteins and protein expression of various mitochondrial ribosomal subunits in young and old subjects was observed(33). Similarly, aerobic exercise and HIIT in old mice are associated with the activation of UPR^{mt}, further improving mitochondrial proteostasis(44, 45). Overall, this would imply that exercise is a potent protector of mitochondrial proteostasis in aging muscle, and related to engagement of PGC-1α. We observed reduced gene expression of mitochondrial translation genes already at 24 months of age, which is deemed a pre-sarcopenic state(19). In mice, resistance wheel running from middle age is effective in preventing hind limb sarcopenia in both male and female mice(46). If also true in humans, this could indicate a window of opportunity in which exercise-based interventions could be performed in a state of non-compromised muscle function to correct the dysregulation and prevent the progression of the potential futile cycle of aberrant mitochondrial translation and ATP production. Thus, until pharmacological means

are discovered, physical activity should remain the main strategy, with a proven effect to prevent and mitigate sarcopenia in the elderly population(47, 48).

Materials & Methods

Animals

C57/Bl6 mice with muscle-specific PGC-1α deletion (mKO-PGC-1α) or overexpression (mTG-PGC-1α) were previously described(49-51). C57/Bl6 wild-type (WT) mice were obtained from Janvier Labs. In this study, male mice were chosen to mitigate resource limitations and to avoid offsets in sarcopenic development caused by the differential longevity of male and female mice(52). In addition, given that in aged female mice mitochondrial content is higher and muscle tissue loss is less pronounced (46), the use of male mice was preferred due to the widened window for age-dependent detriment in mitochondria and skeletal wasting. Mice were studied at 3 and 24 months of age, as well as at 7 and 28 months of age as indicated. Mice were housed in an animal facility with a 12h night/day cycle, with free access to food and water. All experiments were approved by the veterinary office of the canton Basel-Stadt in Switzerland, following federal (Switzerland), cantonal (Basel-Stadt) and institutional (University of Basel) guidelines.

Additional descriptions and more information about materials and methods are available in the supplemental material.

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Author Contributions

L.M.d.S., A.B., M.Z., C.H. conceptualized the study. L.M.d.S., A.B.L. and J.F.G. performed experiments. L.M.d.S., A.B. and D.R. carried out bioinformatic data analysis. L.M.d.S. and C.H. wrote the manuscript. All authors read and improved the final manuscript.

Declaration of Interests

The authors declare no competing interests.

Data Availability

The transcriptomic RNA-seq data have been deposited in GEO (accession number XXX). The proteomic data have been deposited in the Proteomics Identification Database (PRIDE) under accession number XXX. All other data and code are available from the corresponding author upon reasonable request.

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Main figure titles and legends

Figure 1. Expression analysis of aging- and PGC-1α-regulated genes reveals a major impact on mitochondrial translation. (A) PC1 and 2 coordinates for gene expression data (in TPM) from *gastrocnemius* muscle of 3mo and 24mo old WT, PGC-1α mKO, and PGC-1α mTG mice. (B) Heatmap of z-scores (calculated as mean (log2 (TPM))) from gene expression data, clustering based on Euclidean distances revealed annotated cluster 1 (green). (C) Top 15 GO biological process (BP) terms, enriched in genes from cluster 1. Dashed line indicates a significance threshold of p=0.05. (D) Z-score heatmap of genes present in cluster 1, which are present in GO terms 'Translation' & 'Translational Elongation'. (E) Gene expression levels of genes (in TPM) listed in panel E. (F) Relative mRNA expression of genes involved in mitochondrial translation in C2C12 myotubes, upon infection with adenoviral vectors for LacZ or PGC-1α. Data are presented as mean ± SD. In 1A-E: n=6 for all conditions, except mKO 3mo where n=5. In 1E: *FDR<0.05. In 1F: n=4, unpaired two-tailed t-test *$p < 0.05$; **$p < 0.01$; ***$p < 0.001$ for LacZ vs. PGC-1α.

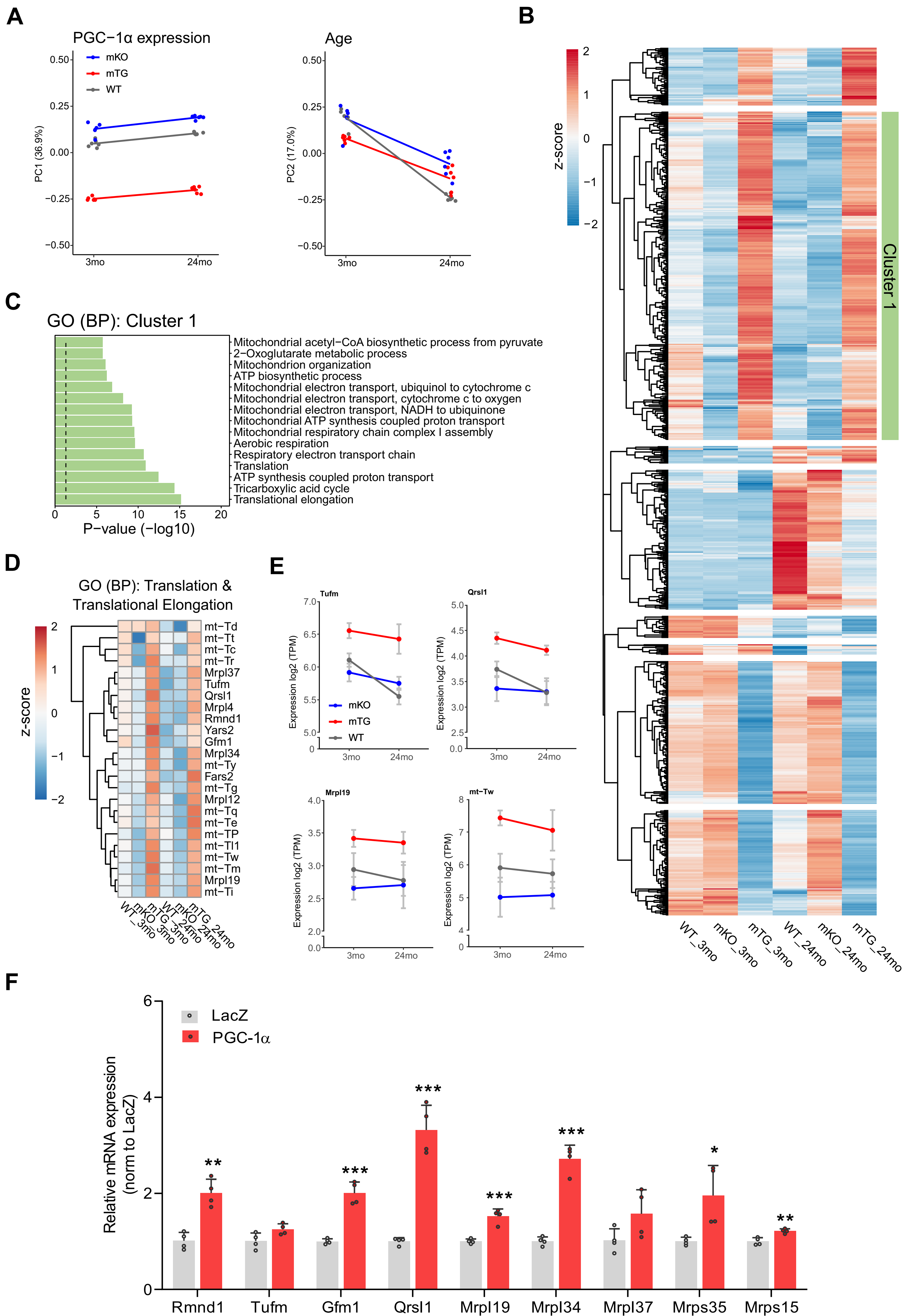
Figure 2. PGC-1α regulates mitochondrial-encoded translation genes and boosts mitochondrial translation. (A) Volcano plot of proteomic data comparing LacZ and PGC-1α overexpression in C2C12 myotubes, n=3. Black dots indicate proteins associated to GO BP terms 'Translation', 'Mitochondrial translation termination', 'Mitochondrial translation' and 'Mitochondrial translation elongation' from panel B. Horizontal dashed line indicates a significance threshold of p=0.05. (B) Top 15 enriched GO BP terms. Dashed line indicates significance threshold of p=0.05. (C) Mitochondrial protein synthesis assessed by *in vitro* ^{35}S-methionine incorporation in newly translated electron transport chain subunits in C2C12 myotubes overexpressing LacZ or PGC-1α under inhibition of cytosolic translation with 100µg/ml emetine. (D) Quantification of ^{35}S-methionine incorporation in core proteins (ND5-ATP6), relative to LacZ. Data are presented as mean ± SD. In 2D: n=3-4 in 3 independent experiments, unpaired two-tailed t-test ***$p < 0.001$ for LacZ vs. PGC-1α.

Figure 3. ERRα is the main transcription factor binding partner for PGC-1α in the regulation of mitochondrial translation genes. (A) ISMARA predicted activity of motif ERRb_ERRa from RNA-seq data depicted in Fig. 1, in 24mo WT, PGC-1α mKO, and PGC-1α mTG mice. Motif activity of ERRb_ERRa in C2C12 myotubes upon overexpression of LacZ or PGC-1α in published RNA-seq data(14). (B) Overlap of gene names associated to PGC-1α and ERRα (>300 kb upstream, <300 kb downstream) peaks from published ChIP-seq data(18, 20), and genes enriched in the GO BP term 'Translation' from RNA-seq data(14). (C) Genome browser view of PGC-1α (red) and ERRα (blue) peaks in promoters of mitochondrial translation genes. (D) Relative mRNA expression of mitochondrial translation genes in C2C12 myotubes overexpressing LacZ or PGC-1α, with 0.1% DMSO or 10uM XCT790. (E) Mitochondrial protein synthesis

assessed by *in vitro* ^{35}S-methionine incorporation in C2C12 myotubes overexpressing LacZ or PGC-1α, under inhibition of cytosolic translation with 100µg/ml emetine. (F) Quantification of ^{35}S-methionine incorporation in core proteins (ND5-ATP6), relative to LacZ + DMSO. Data are presented as mean ± SD. In 3D, F: n=3, two-way ANOVA where †p<0.05 describes the PGC-1α effect, ‡p<0.05 XCT790, #p<0.05 interaction. Sidak's multiple comparisons correction: *p<0.05, **p<0.01, ***p<0.001. In 3F: n=3 shown from 3 independent experiments.

Figure 4. Decreased mitochondrial translation in sarcopenic muscle is mitigated by exercise and PGC-1α. (A) Relative mRNA expression of mitochondrial translation genes in *quadriceps* muscle of 7mo and 28mo mice. (B) Mitochondrial protein synthesis assessed by *in organello* ^{35}S-methionine incorporation into newly translated electron transport chain subunits in mitochondria isolated from *gastrocnemius* muscles of 7mo and 28mo mice. Samples were prepared in pairs, in multiple experiments (pairs are depicted within the black lines). (C) Relative expression of proteins involved in mitochondrial translation in *plantaris* muscle of 7mo mice that underwent endurance (Endurance Run) or resistance (Resistance Run) training by voluntary wheel running. (D) Relative expression of proteins involved in mitochondrial translation in *quadriceps* muscle of sedentary and treadmill-trained 25mo mice. Data are presented as mean ± SD. In 4A: n=5, unpaired two-tailed t-test *p < 0.05; **p < 0.01; ***p < 0.001 for 7mo vs. 28mo. In 4B: n=3, samples prepared on the same day are boxed. In 4C: n=3, *p < 0.05 for Endurance or Resistance Run vs. Sedentary. In 4D: n=4, empirical Bayes moderated t-tests *p < 0.05 for Treadmill trained vs. Sedentary.

Figure 5. Exercise mitigates the decline in PGC-1α/ERRα-mediated mitochondrial translation in aged skeletal muscle. In aged muscle, decreased transcriptional activity of PGC-1α and ERRα result in an impairment of the expression of nuclear and mitochondrial encoded mitochondrial genes, mitochondrial translation, and respiratory chain activity. Exercise mitigates these deficiencies by inducing the PGC-1α/ERRα axis, thereby stimulating mitochondrial translation concomitantly with protein quality control. Consequently, dysfunctional mitochondrial activity and suboptimal ATP production in aging are rectified.


1
A
PGC−1α expression
Age
mKO
mTG
WT
PC1 (36.9%)
PC2 (17.0%)
3mo
24mo
B
z-score
Cluster 1
WT_3mo
mKO_3mo
mTG_3mo
WT_24mo
mKO_24mo
mTG_24mo
C
GO (BP): Cluster 1
Mitochondrial acetyl−CoA biosynthetic process from pyruvate
2−Oxoglutarate metabolic process
Mitochondrion organization
ATP biosynthetic process
Mitochondrial electron transport, ubiquinol to cytochrome c
Mitochondrial electron transport, cytochrome c to oxygen
Mitochondrial electron transport, NADH to ubiquinone
Mitochondrial ATP synthesis coupled proton transport
Mitochondrial respiratory chain complex I assembly
Aerobic respiration
Respiratory electron transport chain
Translation
ATP synthesis coupled proton transport
Tricarboxylic acid cycle
Translational elongation
P−value (−log10)
D
GO (BP): Translation &
Translational Elongation
z-score
mt−Td
mt−Tt
mt−Tc
mt−Tr
Mrpl37
Tufm
Qrsl1
Mrpl4
Rmnd1
Yars2
Gfm1
Mrpl34
mt−Ty
Fars2
mt−Tg
Mrpl12
mt−Tq
mt−Te
mt−TP
mt−Tl1
mt−Tw
mt−Tm
Mrpl19
mt−Ti
E
Tufm
Qrsl1
Mrpl19
mt−Tw
Expression log2 (TPM)
F
LacZ
PGC-1α
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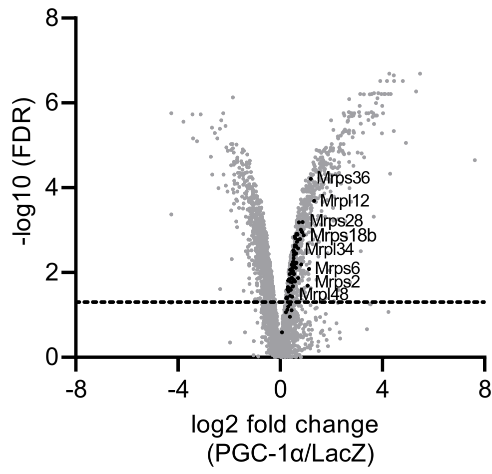
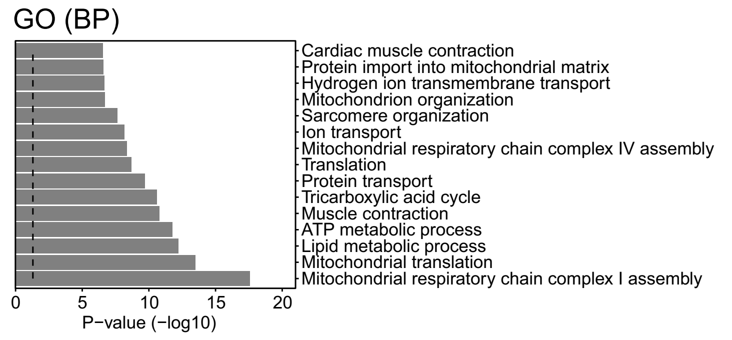
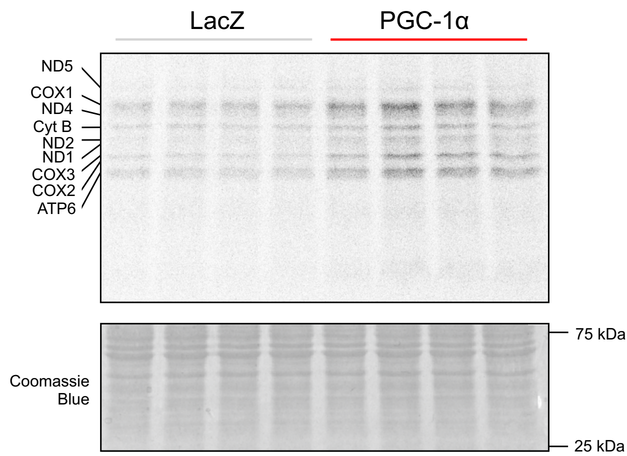
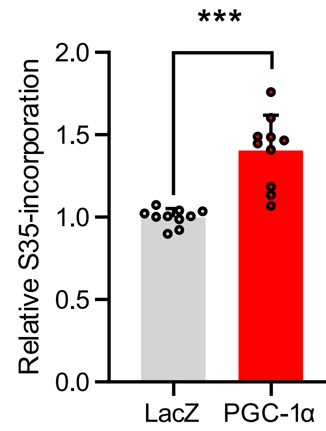
Mrpl37
Mrps35
Mrps15

2









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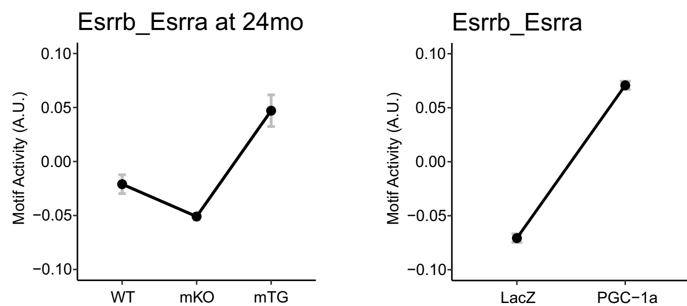
A



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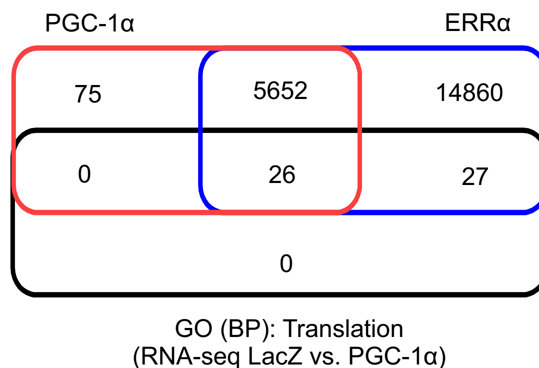

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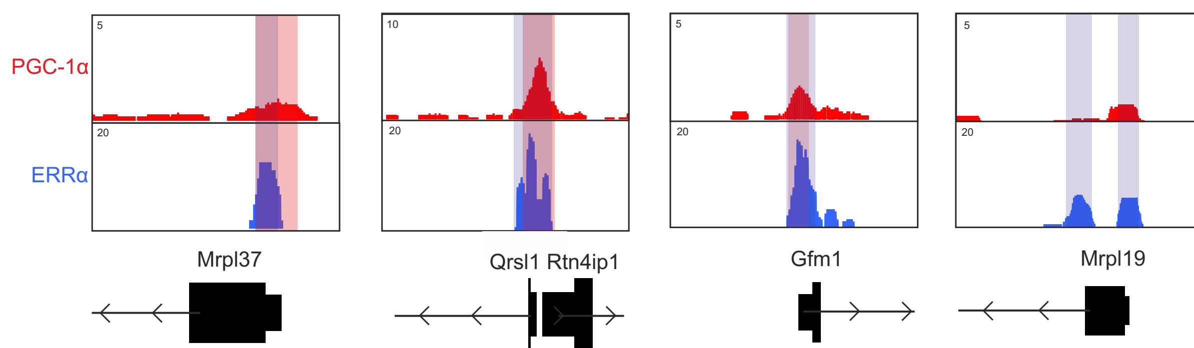

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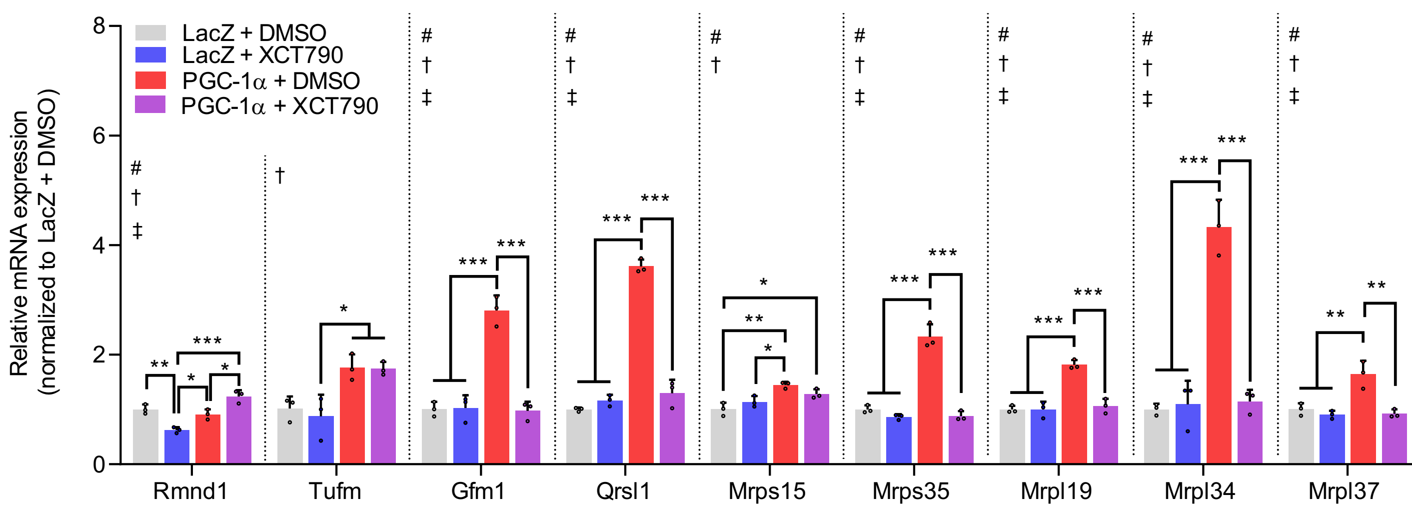

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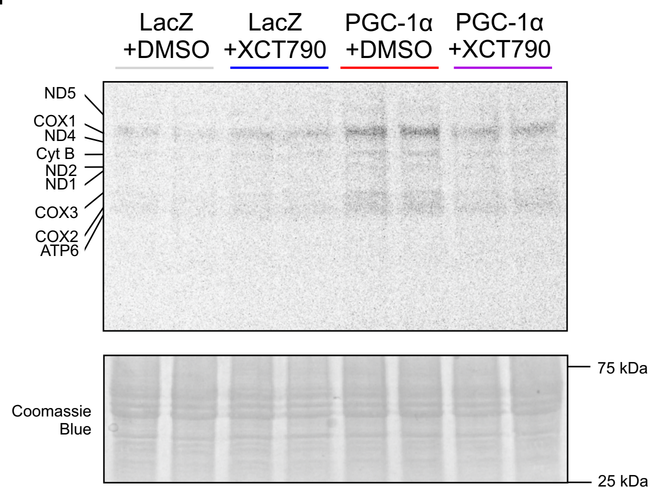

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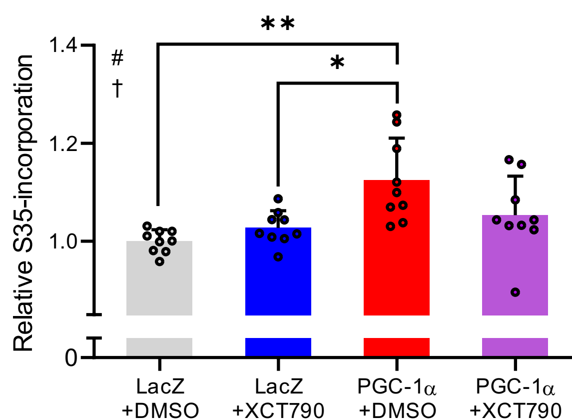

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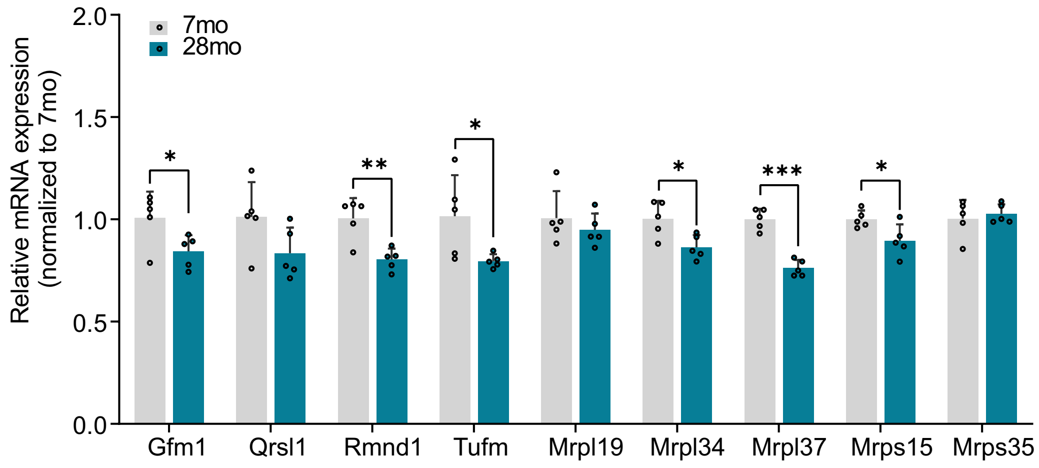

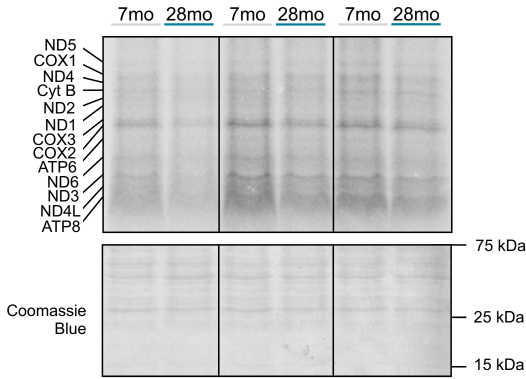

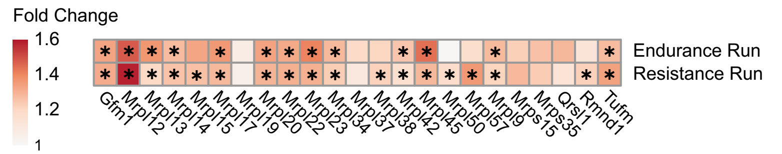

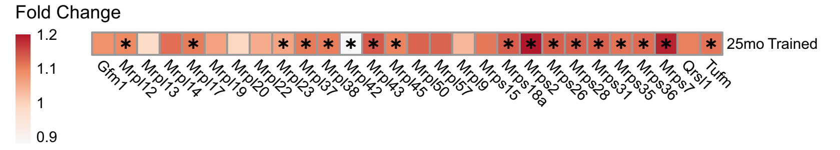

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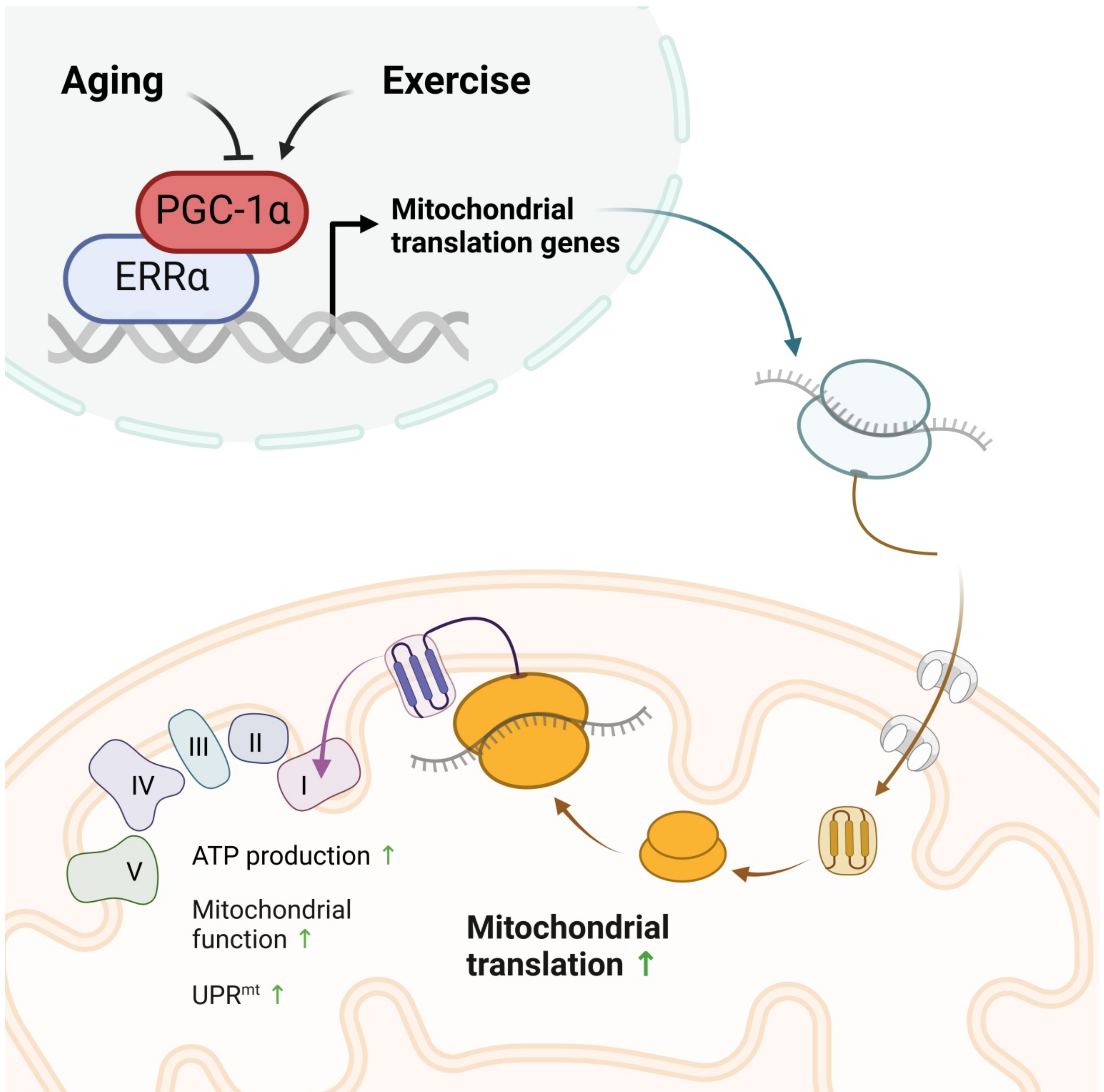
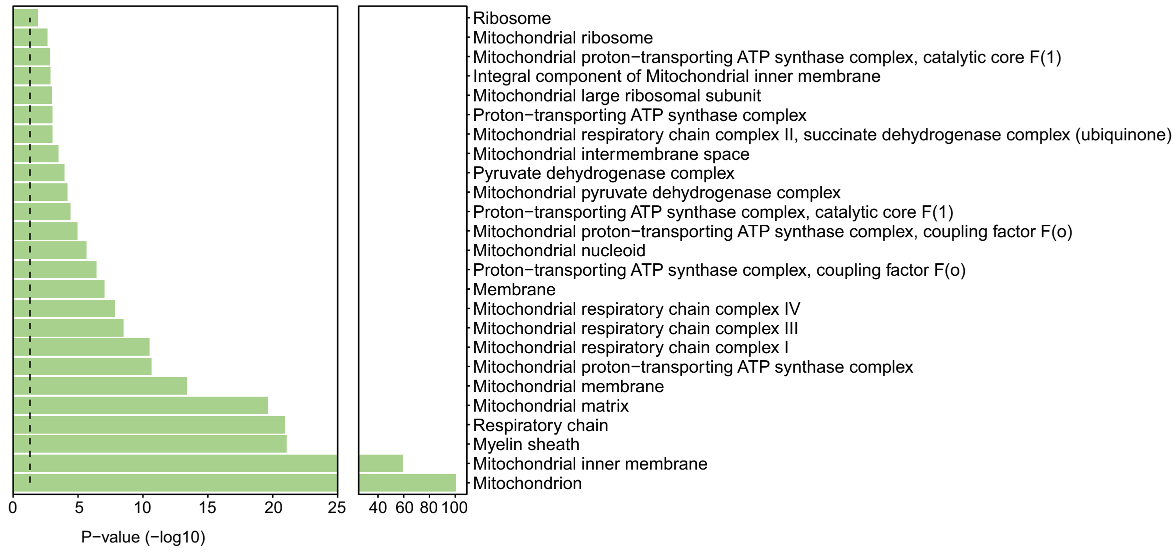


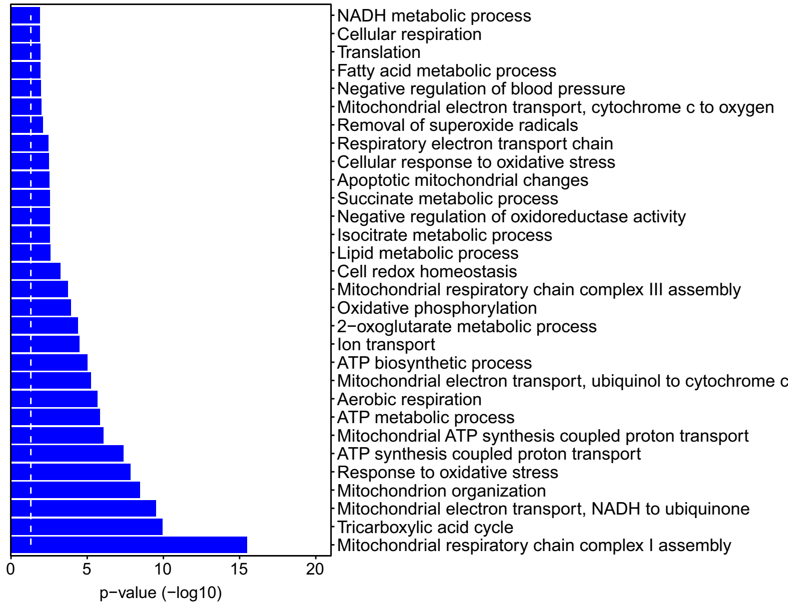
S1

A

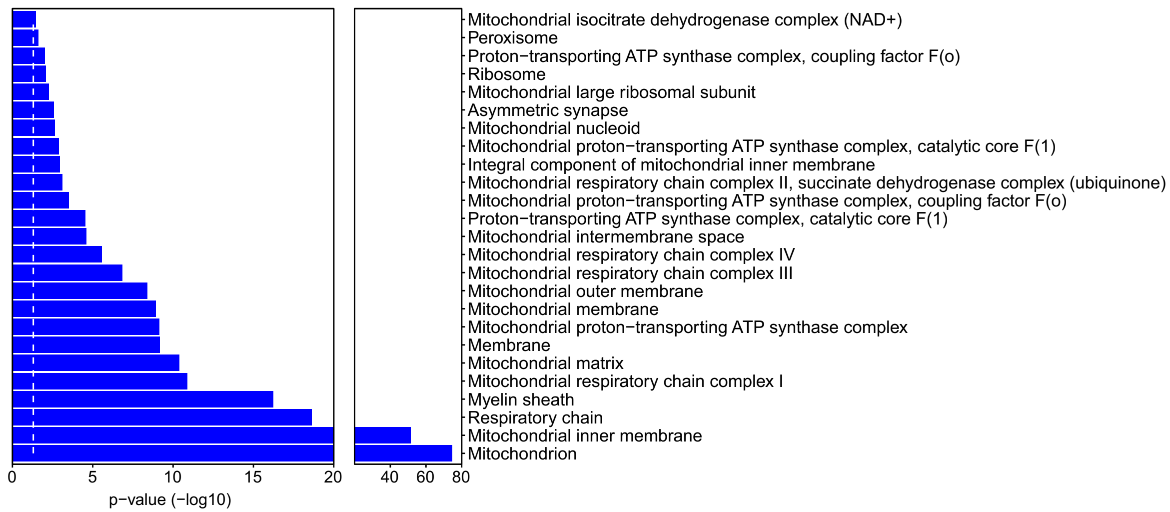


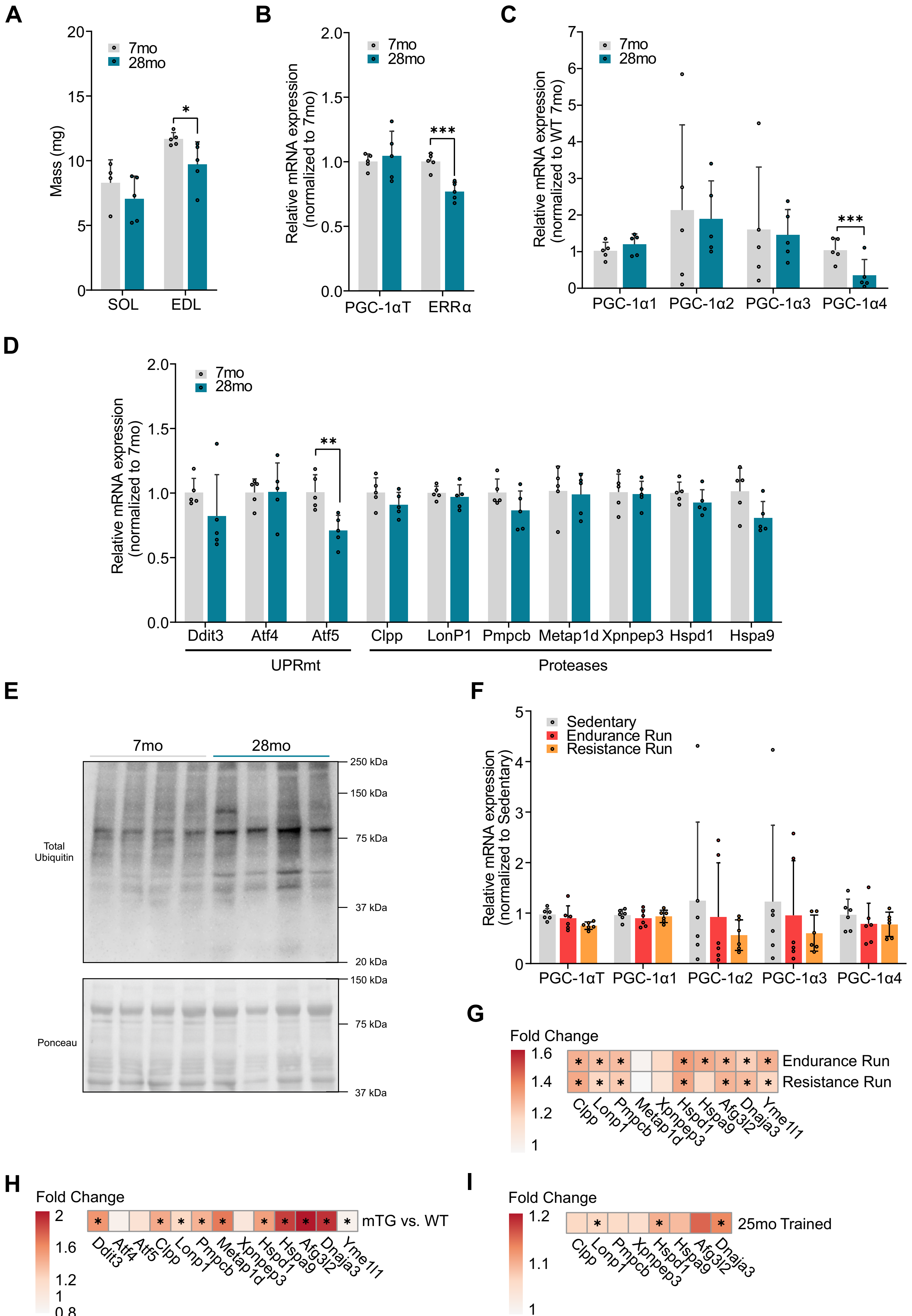
S2

A



B




S3
A
B
C
D
E
F
G
H
I
7mo
28mo
Mass (mg)
SOL
EDL
Relative mRNA expression (normalized to 7mo)
PGC-1αT
ERRα
Relative mRNA expression (normalized to WT 7mo)
PGC-1α1
PGC-1α2
PGC-1α3
PGC-1α4
Ddit3
Atf4
Atf5
Clpp
LonP1
Pmpcb
Metap1d
Xpnpep3
Hspd1
Hspa9
UPRmt
Proteases
Total Ubiquitin
Ponceau
250 kDa
150 kDa
75 kDa
37 kDa
20 kDa
Sedentary
Endurance Run
Resistance Run
Relative mRNA expression (normalized to Sedentary)
Fold Change
Endurance Run
Resistance Run
Lonp1
Afg3l2
Dnaja3
Yme1l1
mTG vs. WT
25mo Trained