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Alteration of mitochondrial function in the livers of mice with glycogen branching enzyme deficiency

Dominika Malinska [a, *], Giorgia Testoni [b], Malgorzata Bejtka [a], Jordi Duran [b, c], Joan J. Guinovart [b, c, d], Jerzy Duszynski [a]

[a] Nencki Institute of Experimental Biology, Pasteur Street 3, 02-093, Warsaw, Poland
[b] Institute for Research in Biomedicine (IRB Barcelona), Barcelona Institute of Science and Technology (BIST), Barcelona, 08028, Spain
[c] Centro de Investigación Biomédica en Red de Diabetes y Enfermedades Metabólicas Asociadas (CIBERDEM), Madrid, 28029, Spain
[d] Department of Biochemistry and Molecular Biomedicine, University of Barcelona, Barcelona, 08028, Spain

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ABSTRACT

Glycogen storage disease type IV (GSD IV) is caused by mutations in the glycogen branching enzyme gene (*GBE1*) that lead to the accumulation of aberrant glycogen in affected tissues, mostly in the liver. To determine whether dysfunctional glycogen metabolism in GSD IV affects other components of cellular bioenergetics, we studied mitochondrial function in heterozygous *Gbe1* knockout (*Gbe1*$^{+/-}$) mice. Mitochondria isolated from the livers of *Gbe1*$^{+/-}$ mice showed elevated respiratory complex I activity and increased reactive oxygen species production, particularly by respiratory chain complex III. These observations indicate that GBE1 deficiency leads to broader rearrangements in energy metabolism and that the mechanisms underlying GSD IV pathogenesis may include more than merely mechanical cell damage caused by the presence of glycogen aggregates.

1. Introduction

Glycogen branching enzyme (GBE1, EC 2.4.1.18) introduces regular branching points in the glycogen structure during its synthesis. The branched structure of glycogen improves its solubility in the intracellular milieu and provides an essential amount of free ends for glycogen synthesis and degradation [1]. Proper glycogen structure may also be important for the interaction of glycogen with proteins regulating glycogen turnover as well as other metabolic pathways [2].

The importance of GBE1 activity to cell function becomes apparent when we look at the consequences of pathogenic mutations in the *GBE1* gene, causing glycogen storage disease type IV (GSD IV) [3]. This disease is characterized by the intracellular accumulation of structurally abnormal, poorly branched glycogen molecules, accompanied by dysfunctions in the liver, cardiac and skeletal muscle, and neural tissue [4]. The intracellular mechanisms responsible for those dysfunctions remain poorly studied, and it is not clear to what extent the secondary metabolic disturbances may contribute to cell damage.

Several mouse models of GBE1 deficiency with different extents of decreased GBE1 activity have been described [5–8]. Homozygous *Gbe1* knockout in mice was lethal during fetal development or in the perinatal period [5,6], while heterozygous *Gbe1* knockout was accompanied by altered liver glycogen structure, but there was no accumulation of excessive glycogen in this tissue [8]. In the livers of aged *Gbe1*$^{+/-}$ mice, we found increased oxidative stress and decreased activity of mitochondrial respiratory complex I, indicating that the impaired glycogen structure accompanying GSD IV can lead to cellular stress even in the absence of glycogen overaccumulation [8].

Oxidative stress occurs when reactive oxygen species (ROS) synthesis overwhelms intracellular antioxidant mechanisms. One of the intracellular ROS sources is the mitochondrial respiratory chain [9]. Moderate ROS release from mitochondria participates in

Abbreviations: BSA, bovine serum albumine; CCCP, carbonyl cyanide m-chlorophenylhydrazone; CI, CII, CIII, CIV, respiratory complexes I, II, III and IV; CoQ, coenzyme Q; GBE1, glycogen branching enzyme; GSD IV, glycogen storage disease type IV; HSP60, 60 kDa heat shock protein; ROS, reactive oxygen species; TIM23, mitochondrial import inner membrane translocase subunit; TOM20, translocase of outer membrane 20 kDa subunit; WT, wild-type.

* Corresponding author.
E-mail address: d.malinska@nencki.edu.pl (D. Malinska).

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intracellular signaling, such as in the feedback regulation of mitochondrial function [10]. However, when produced in excess, mitochondrial ROS are potentially harmful to the cell. There is increasing evidence that mitochondrial dysfunction is involved in the pathogenesis of various metabolic or neurodegenerative diseases [11], and thus, interest in investigating mitochondrial function under pathological conditions has emerged.

In previous work, we presented evidence for increased oxidative stress and impairment within the mitochondrial respiratory chain in the livers of aged *Gbe1*$^{+/-}$ mice [8]. In the current study, we performed a more detailed investigation of mitochondrial function and mitochondrial ROS production in GBE1-deficient mouse liver tissue.

2. Materials and methods

The *Gbe1* heterozygous knockout mouse on a C57BL/6 mouse background was obtained as described previously [8]. The animals analyzed were adult males (4 months old), and experiments were conducted using littermates. All procedures were approved by the PCB Animal Experimentation Committee and were in accordance with the National Institutes of Health and the European Community Council Directive guidelines for the care and use of laboratory animals.

Mouse liver mitochondria were isolated by differential centrifugation [12]. Livers were homogenized in cold isolation medium (225 mM mannitol, 75 mM sucrose, 0.1 mM EGTA, 5 mM Tris, 5 mg/ml bovine serum albumin (BSA), pH 7.4) with a motor-driven Potter-Elvehjem homogenizer (600 rpm). Debris was removed by two centrifugations at 740×*g* for 5 min. Mitochondria were then sedimented by means of a 10-min centrifugation at 9000×*g* and subsequently washed in isolation medium without BSA. The obtained mitochondrial pellet was suspended in isolation medium without BSA, supplemented with 1 mM $MgCl_2$. All procedures were carried out on ice, centrifugations were performed at 4 °C.

Oxygen consumption rates were measured with an O2k oxygraph (Oroboros Instruments, Austria) at 30 °C in measurement medium (110 mM mannitol, 60 mM Tris, 60 mM KCl, 10 mM KH_2PO_4, 0.5 mM EDTA, 5 mM $MgCl_2$, pH 7.4) with 10 mM glutamate and 10 mM malate as respiratory substrates. Respiratory state 3 was achieved by the addition of 0.4 mM ADP.

The release of reactive oxygen species (ROS) from isolated mitochondria was measured as described previously [13] at 30 °C in the same measurement medium as in the respiratory measurements, using an Amplex red fluorescent probe (1 μM) in the presence of horseradish peroxidase (20 U/ml) and superoxide dismutase (35 U/ml). Amplex red oxidation was followed at $\lambda_{ex} = 560$ nm and $\lambda_{em} = 590$ nm. At the end of each measurement, 13 800 U/ml catalase was added, and the catalase-insensitive slope was subtracted from the measurement results. In each experiment, liver mitochondria were isolated in parallel from one mouse with wild-type *Gbe1* (WT) and one *Gbe1*$^{+/-}$ mouse, and the measurements from both preparations were taken simultaneously.

The activities of respiratory complexes I (CI), II (CII) and IV (CIV) were measured with microplate ELISA kits (Abcam, UK) following the manufacturer's instructions. Citrate synthase (CS) activity was determined as described previously [14]. The levels of respiratory proteins were checked by Western blot with the use of Total OXPHOS Rodent WB Antibody Cocktail (Abcam). The mitochondrial content of the homogenates was checked by Western blot with the antibodies directed against 60 kDa heat shock protein, HSP60 (Abcam), mitochondrial import inner membrane translocase subunit TIM23 (Abcam) and translocase of outer membrane 20 kDa subunit, TOM20 (Santa Cruz Biotechnology, USA).

3. Results and discussion

In our previous study, we detected increased oxidative stress and impaired mitochondrial respiratory chain function in the livers of aged *Gbe1*$^{+/-}$ mice [8]. Those animals had 50% decrease in Gbe1 levels [8] and activities (unpublished data) in the liver, resulting in mild decrease in glycogen branching degree but not leading to excessive glycogen accumulation [8]. Since mitochondria are important intracellular ROS sources, we took a closer look at mitochondrial function and ROS production in the livers of young *Gbe1*$^{+/-}$ animals.

The respiration rates were similar in mitochondrial preparations from the livers of WT and *Gbe1*$^{+/-}$ mice (Fig. 1A). In contrast, ROS production was significantly higher in *Gbe1*$^{+/-}$ mouse mitochondria, irrespective of which substrates were used to fuel the mitochondrial respiratory chain: glutamate and malate, which drive CI-dependent respiration, or succinate, which is a CII substrate (Fig. 1C). This reveals, that in Gbe1-deficient mouse liver tissue alterations of intracellular ROS synthesis can be observed much earlier than the symptoms of oxidative tissue damage [8].

In mitochondrial respiratory chain the main superoxide-producing sites are located at CI and CIII [9,15]. Those two sites differ with respect to the intracellular compartment to which the superoxide is released: CIII-derived superoxide is released mostly into the intermembrane space, while CI is responsible for ROS release into the matrix [15]. As a result, superoxide produced at these two sites has different potential targets and physiological effects [16]. In isolated mitochondria fueled with either glutamate + malate, or with succinate, both CI and CIII can contribute to the superoxide synthesis (Fig. 1B): when succinate is used, ROS synthesis by CI depends on the extent of reversed electron transfer from CII to CI, while forward electron transfer drives the CIII-dependent ROS synthesis [17]. To assess the contribution of CI and CIII to the observed increase in ROS synthesis in the liver mitochondria of *Gbe1*$^{+/-}$ mice, we checked the effect of respiratory chain modulators on mitochondrial ROS release. Since ROS synthesis by the respiratory chain strongly depends on the redox status of the superoxide-releasing sites [9], we modulated respiratory chain redox state with the following chemicals: protonophore carbonyl cyanide m-chlorophenylhydrazone (CCCP, 1 μM), which dissipates mitochondrial membrane potential, promoting oxidation of respiratory chain redox centers, and the respiratory chain inhibitors: CI inhibitor rotenone (1 μM) and CIII inhibitor antimycin A (2 μg/ml), which lead to a reduction in reaction centers upstream of the inhibitor target site. Out of the conditions applied, the most significant difference between mitochondria isolated from the livers of WT and *Gbe1*$^{+/-}$ mice was found in the presence of succinate + rotenone or succinate + CCCP (Fig. 1C and D). The common feature of these two conditions is that CI is not involved in electron transfer, since electrons are introduced to the chain at CII and reverse electron transfer through ubiquinone to CI is prevented either via inhibition of CI by rotenone or via depolarization of mitochondria by CCCP. Thus, ROS measured under these conditions originate mostly from CIII. Additionally, leveling ROS generation of WT and *Gbe1*$^{+/-}$ mitochondria fueled with glutamate + malate upon addition of rotenone speaks for CIII being responsible for the higher ROS release from *Gbe1*$^{+/-}$ liver mitochondria also in the presence of CI substrates.

There are several factors that determine CIII-dependent superoxide release. The main is the CoQ redox state, which depends on the CoQ content, relative activities of respiratory complexes as well as on organization of the respiratory chain into supercomplexes [18,19]. Interestingly, the alterations of CoQ content as well as of respiratory supercomplexes organization have been postulated to play a role in various metabolic diseases due to their impact on

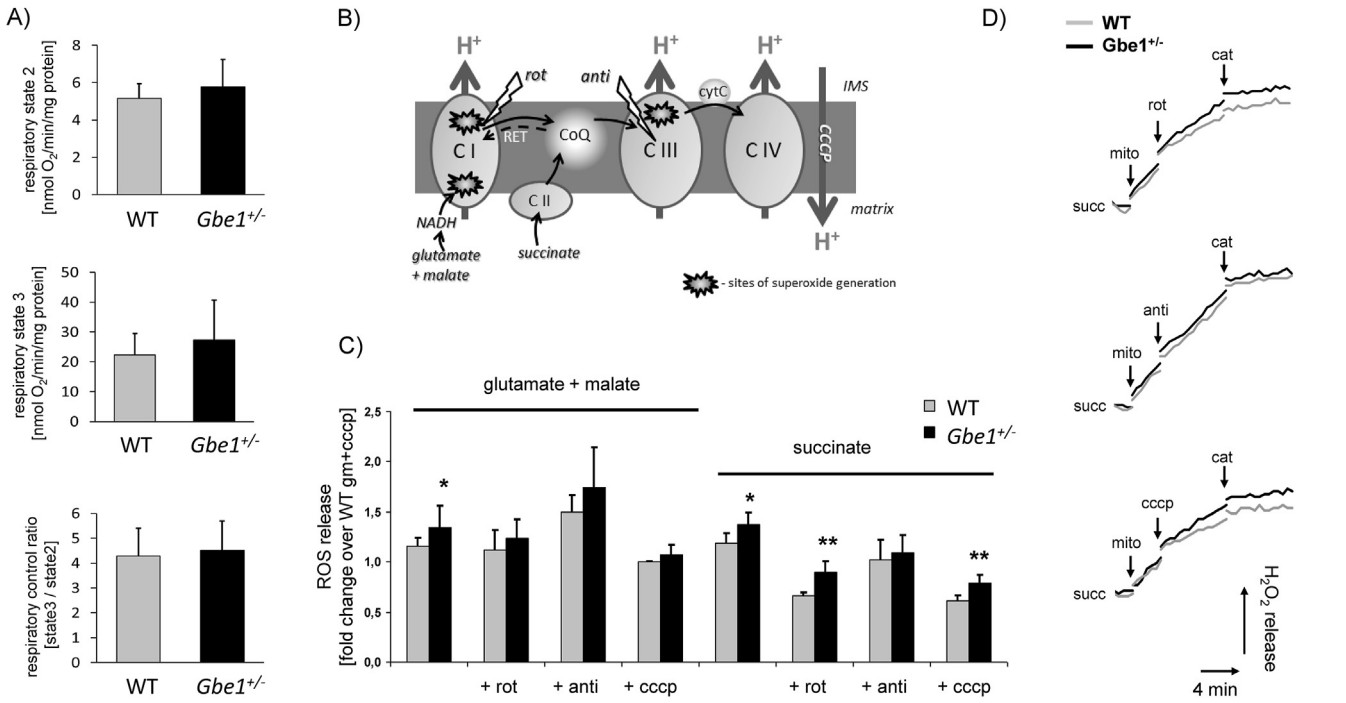

Fig. 1. ROS production and respiration rates in isolated liver mitochondria from wild-type (WT) and *Gbe1*$^{+/-}$ mice. **(A)** Mitochondrial respiration rates in presence of glutamate and malate only (state 2) and after ADP addition (state 3). Graphs present mean values ± SD from n = 4 independent experiments. The measurements were performed with 0.15 mg of mitochondrial protein in 2.1 ml final volume of the measurement medium. **(B)** Mitochondrial respiratory chain - sites of action of the applied inhibitors. **(C)** ROS production by isolated mouse liver mitochondria. In each experiment the preparations from one WT mouse and one *Gbe1*$^{+/-}$ mouse livers were prepared and analyzed in parallel. The results were normalized to the values measured in particular experiment for WT mitochondria in the presence of glutamate + malate + CCCP. Graph presents mean values ± SD from n = 4 independent experiments. *p < 0.05, **p < 0.01 compared to WT, according to Student's t-test. **(D)** Representative traces of Amplex red oxidation by WT (grey lines) and *Gbe1*$^{+/-}$ (black lines) mice liver mitochondria in selected experimental options. For each measurement, 30 μg of mitochondrial protein was used in 0.5 ml of the measurement medium. anti: antimycin A, cat: catalase, CoQ – coenzyme Q, cyt *c* – cytochrome *c*, IMS – mitochondrial intermembrane space, rot: rotenone, succ: succinate.

mitochondrial ROS production [20,21]. Apart from CoQ redox state, ROS release from CIII can be affected by cytochrome *c* content in the intermembrane space as well as by total CIII levels and activities [18]. In our study, similar ROS production rates in the presence of antimycin A indicate comparable maximal capacity of CIII to produce superoxide in both groups. Moreover, the content of representative respiratory chain proteins did not differ between WT and Gbe1$^{+/-}$ mice mitochondria, showing similar content and stoichiometry of particular respiratory complexes (Fig. 2B). We detected the increased CI activity in the liver mitochondria from *Gbe1*$^{+/-}$ mice (with CII and CIV activities not differing between the groups) (Fig. 2A), however, this does not clarify the elevated ROS production under conditions when electron transfer bypassed CI. Thus, the exact reason of higher CIII-dependent ROS release in mitochondria from Gbe1-deficient livers remains to be determined.

In addition to studying isolated mitochondria, we also investigated the content and activity of mitochondrial respiratory complexes in total liver homogenates. In contrast to what was seen in isolated mitochondria, the activities of CI were similar in homogenates from WT and *Gbe1*$^{+/-}$ mice (Fig. 2C). Determination of mitochondrial markers (CS activity and the levels of proteins characteristic for different mitochondrial compartments: matrix protein HSP60, TIM23 from the inner mitochondrial membrane and TOM20 from the outer mitochondrial membrane) revealed, that mitochondrial content did not differ significantly between the groups (Fig. 2C and D). In the liver homogenates from *Gbe1*$^{+/-}$ mice the levels of respiratory chain proteins seemed slightly increased, but without reaching statistical significance (Fig. 2D). The observed discrepancy between the results obtained for isolated mitochondria and whole tissue homogenates concerning CI activity may reflect the fact, that mitochondrial preparations contain a subpopulation of all mitochondria present in the tissue [22,23], which should always be taken into consideration in experiments based on subcellular fractionation. The obtained results indicate, that in *Gbe1*$^{+/-}$ mouse livers a subpopulation of mitochondria with lower CI activity was not present in the final mitochondrial pellet, possibly due to its higher fragility and disruption during the isolation procedure. Increased CI activity in isolated *Gbe1*$^{+/-}$ liver mitochondria preparations can reflect an adaptive response to compensate for lower CI activity in another mitochondrial subpopulation. Interestingly, in our previous studies performed with aged *Gbe1*$^{+/-}$ mice, we detected lower complex I activity in liver homogenates from *Gbe1*$^{+/-}$ animals than from WT littermates, indicative of respiratory chain damage progressing with age [8].

To date, the issue of mitochondrial dysfunctions has not been explored in GSD IV, however there are reports showing mitochondrial alterations in other types of GSDs, for instance in type I, II and V [24–26]. It is also known that many glycogen storage disorders are accompanied by impaired autophagy, which can translate to less efficient clearance of damaged mitochondria and, subsequently, dysfunction of these organelles [27]. Whether such impairment also occurs in GSD IV remains to be determined, as the data concerning this disease are strongly limited due to its rare prevalence. However, autophagy appears worth exploring in the context of mitochondrial dysfunction observed in Gbe1-deficient tissues.

Our results point to new directions in investigating the pathogenesis of GSD IV that go beyond focusing on excessive glycogen accumulation alone. We observed an increase in mitochondrial ROS production in the GSD IV model with unaltered glycogen levels, which additionally shows that not only glycogen levels but also its structure is important in the regulation of cellular bioenergetics.

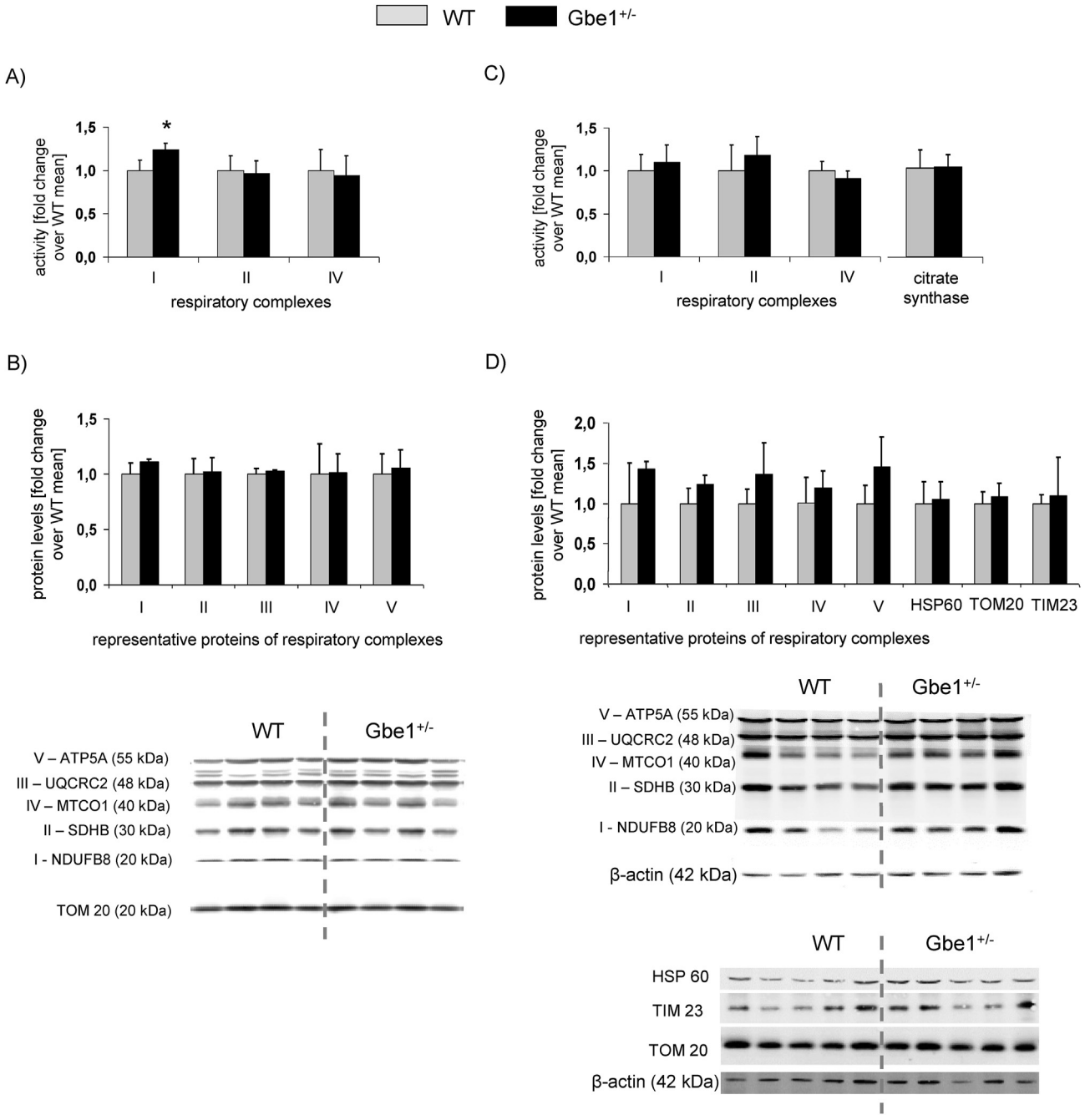

Fig. 2. Activities and levels of respiratory complexes in isolated liver mitochondria (A, B) and in liver homogenates (C, D) from wild-type (WT) and *Gbe1*$^{+/-}$ mice. **(A)** Total activities of respiratory complexes in the isolated liver mitochondria (n = 3). **(B)** Respiratory proteins content in the isolated liver mitochondria (n = 4). **(C)** Total activities of respiratory complexes and of citrate synthase in the liver homogenates (n = 5). **(D)** Respiratory proteins content in the liver homogenates (n = 5). Bar graphs present means ± standard deviations. Data were normalized against mean values measured for WT samples. For each of the respiratory complexes the levels of one representative subunit was determined, as indicated next to the Western blot image. As loading controls, TOM20 was used for isolated mitochondria samples and β-actin was used for liver homogenates. *$p < 0.05$ compared to WT, according to Student's t-test. Roman numerals I–V refer to particular respiratory complexes. Below the graphs, the representative Western blot result is presented.

Author contributions

J.D., J.G. and D.M. designed the study, G.T. and J. Duran prepared the animals, D.M., M.B. and G.T. performed the experiments, D.M. and M.B. analyzed the data, D.M. wrote the manuscript, J.G., J.D., G.T. and J. Duran revised and edited the manuscript, J.G., J.D. and D.M. provided the funding.

All the authors read and approved the final manuscript.

Declaration of competing interest

The authors declare no conflict of interests.

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References

[1] R. Meléndez, E. Meléndez-Hevia, M. Cascante, How did glycogen structure evolve to satisfy the requirement for rapid mobilization of glucose? A problem of physical constraints in structure building, J. Mol. Evol. 45 (1997) 446–455.
[2] D. Stapleton, C. Nelson, K. Parsawar, D. McClain, R. Gilbert-Wilson, E. Barker, B. Rudd, K. Brown, W. Hendrix, P. O'Donnell, G. Parker, Analysis of hepatic glycogen-associated proteins, Proteomics 10 (2010) 2320–2329.
[3] S.W. Moses, R. Parvari, The variable presentations of glycogen storage disease type IV: a review of clinical, enzymatic and molecular studies, Curr. Mol. Med. 2 (2002) 177–188.
[4] C. Hedberg-Oldfors, A. Oldfors, Polyglucosan storage myopathies, Mol. Aspect. Med. 46 (2015) 85–100.
[5] Y.C. Lee, C.J. Chang, D. Bali, Y.T. Chen, Y.T. Yan, Glycogen-branching enzyme deficiency leads to abnormal cardiac development: novel insights into glycogen storage disease IV, Hum. Mol. Genet. 20 (2011) 455–465.
[6] H.O. Akman, T. Sheiko, S.K. Tay, M.J. Finegold, S. Dimauro, W.J. Craigen, Generation of a novel mouse model that recapitulates early and adult onset glycogenosis type IV, Hum. Mol. Genet. 20 (2011) 4430–4439.
[7] H.O. Akman, V. Emmanuele, Y.G. Kurt, B. Kurt, T. Sheiko, S. DiMauro, W.J. Craigen, A novel mouse model that recapitulates adult-onset glycogenosis type 4, Hum. Mol. Genet. 24 (2015) 6801–6810.
[8] D. Malinska, G. Testoni, J. Duran, A. Brudnicka, J.J. Guinovart, J. Duszynski, Hallmarks of oxidative stress in the livers of aged mice with mild glycogen branching enzyme deficiency, Arch. Biochem. Biophys. 695 (2020) 108626.
[9] C.L. Quinlan, I.V. Perevoshchikova, M. Hey-Mogensen, A.L. Orr, M.D. Brand, Sites of reactive oxygen species generation by mitochondria oxidizing different substrates, Redox Biol. 1 (2013) 304–312.
[10] G.S. Shadel, T.L. Horvath, Mitochondrial ROS signaling in organismal homeostasis, Cell 163 (2015) 560–569.
[11] S.R. Pieczenik, J. Neustadt, Mitochondrial dysfunction and molecular pathways of disease, Exp. Mol. Pathol. 83 (2007) 84–92.
[12] D. Johnson, H. Lardy, Isolation of liver or kidney mitochondria, Methods Enzymol. 10 (1967) 94–96.
[13] D. Malinska, A.P. Kudin, G. Debska-Vielhaber, S. Vielhaber, W.S. Kunz, Chapter 23 Quantification of superoxide production by mouse brain and skeletal muscle mitochondria, Methods Enzymol. 456 (2009) 419–437.
[14] A.V. Kuznetsov, D. Strobl, E. Ruttmann, A. Königsrainer, R. Margreiter, E. Gnaiger, Evaluation of mitochondrial respiratory function in small biopsies of liver, Anal. Biochem. 305 (2002) 186–194.
[15] A.P. Kudin, G. Debska-Vielhaber, W.S. Kunz, Characterization of superoxide production sites in isolated rat brain and skeletal muscle mitochondria, Biomed. Pharmacother. 59 (2005) 163–168.
[16] L. Bleier, I. Wittig, H. Heide, M. Steger, U. Brandt, S. Dröse, Generator-specific targets of mitochondrial reactive oxygen species, Free Radic. Biol. Med. 78 (2015) 1–10.
[17] A.M. Gusdon, G.A. Fernandez-Bueno, S. Wohlgemuth, J. Fernandez, J. Chen, C.E. Mathews, Respiration and substrate transport rates as well as reactive oxygen species production distinguish mitochondria from brain and liver, BMC Biochem. 16 (2015) 22.
[18] L. Bleier, S. Dröse, Superoxide generation by complex III: from mechanistic rationales to functional consequences, Biochim. Biophys. Acta 1827 (2013) 1320–1331.
[19] R. Moreno-Loshuertos, J.A. Enríquez, Respiratory supercomplexes and the functional segmentation of the CoQ pool, Free Radic. Biol. Med. 100 (2016) 5–13.
[20] D.J. Fazakerley, R. Chaudhuri, P. Yang, G.J. Maghzal, K.C. Thomas, J.R. Krycer, S.J. Humphrey, B.L. Parker, K.H. Fisher-Wellman, C.C. Meoli, N.J. Hoffman, C. Diskin, J.G. Burchfield, M.J. Cowley, W. Kaplan, Z. Modrusan, G. Kolumam, J.Y. Yang, D.L. Chen, D. Samocha-Bonet, J.R. Greenfield, K.L. Hoehn, R. Stocker, D.E. James, Mitochondrial CoQ deficiency is a common driver of mitochondrial oxidants and insulin resistance, Elife 7 (2018) e32111.
[21] I. Ramírez-Camacho, W.R. García-Niño, M. Flores-García, J. Pedraza-Chaverri, C. Zazueta, Alteration of mitochondrial supercomplexes assembly in metabolic diseases, Biochim. Biophys. Acta (BBA) - Mol. Basis Dis. 1866 (2020) 165935.
[22] P.A. Figueiredo, S.K. Powers, R.M. Ferreira, F. Amado, H.J. Appell, J.A. Duarte, Impact of lifelong sedentary behavior on mitochondrial function of mice skeletal muscle, J. Gerontol. A Biol. Sci. Med. Sci. 64 (2009) 927–939.
[23] M. Picard, D. Ritchie, K.J. Wright, C. Romestaing, M.M. Thomas, S.L. Rowan, T. Taivassalo, R.T. Hepple, Mitochondrial functional impairment with aging is exaggerated in isolated mitochondria compared to permeabilized myofibers, Aging Cell 9 (2010) 1032–1046.
[24] J.A. Lim, L. Li, O. Kakhlon, R. Myerowitz, N. Raben, Defects in calcium homeostasis and mitochondria can be reversed in Pompe disease, Autophagy 11 (2015) 385–402.
[25] J.J. Kaczor, H.A. Robertshaw, M.A. Tarnopolsky, Higher oxidative stress in skeletal muscle of McArdle disease patients, Mol. Genet. Metab. Rep. 12 (2017) 69–75.
[26] J.H. Cho, G.Y. Kim, B.C. Mansfield, J.Y. Chou, Sirtuin signaling controls mitochondrial function in glycogen storage disease type Ia, J. Inherit. Metab. Dis. (2018), https://doi.org/10.1007/s10545-018-0192-1.
[27] B.L. Farah, P.M. Yen, D.D. Koeberl, Links between autophagy and disorders of glycogen metabolism - perspectives on pathogenesis and possible treatments, Mol. Genet. Metabol. 129 (2020) 3–12.