Can aging be reversed?

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Abstract
One of the hallmarks of aging and aging-related diseases is a disruption in the homeostasis of mitochondrial function. Why this disruption occurs and how it leads to the reduced function of mitochondria is still a topic of much research and scientific attention. In this midst, Gomes et al's recent article in Cell brings forth an essential and exciting finding on reversibility of mitochondrial dysfunction with age (Gomes, Price et al. 2013). This reversibility is enabled through restoration of intracellular communication between mitochondria and nucleus, which falls short during the aging process. To explain this, there are a number of key players that are going to be discussed first.

Key Words: Aging, Mitochondria, Sirtuins, Metabolism, Skeletal muscle

In the 1950's, Denham Harman proposed that aging is the result of accumulated cellular damage imposed by free radical species (molecules with unpaired electrons in their outer shells) (Harman 1956; Harman 1983). Shortly after this, the mitochondrial theory of aging gained momentum. This theory holds that the mitochondrion can act as one of the key players in aiding the production of superoxide species and so contribute to cellular damage (Jacobs 2003; Cui, Kong et al. 2012).

Mitochondrion is a self-contained energy powerhouse organelle with its own small genome producing ATP through the electron transport chain (ETC) (Cadenas and Davies 2000). Although ETC is very efficient, approximately 2% of oxygen is incompletely oxidized during the ETC process and results in superoxide radicals that can be converted to more damaging free radical species (Cadenas 2004). It is the production of these free radicals that can cause cellular perturbations through cumulated damage to the DNA, proteins, and lipids. Interestingly, mild chronic oxidative damage can increase cell growth and division while high amounts can cause oxidative damage, senescence and cell death, all phenomena associated with aging (Zu, Liu et al. 2010).

SIRT1 is part of a family of genes called Sir2 (sirtuins), a conserved NAD^+-dependent deacetylases predominantly localized in the nucleus (Jin, Yan et al. 2007), and is associated with caspase-mediated apoptosis when localized in the cytosol. SIRT1 has been identified as an aging gene activated by resveratrol (a natural phenol) (Borra, Smith et al. 2005) and is best known for controlling physiological responses to stress, metabolism, and aging (Ghosh,

Liu et al. 2013; Satoh, Brace et al. 2013). The expression of SIRT1 is elevated in a number of tissues following calorie restriction (CR) (Chen, Bruno et al. 2008), an intervention that extends lifespan and protects organisms from secondary aging related diseases such as cancer (Lin and Fang 2013). There exists some controversial data on the role of SIRT1 on HIF-1α activity. There are articles showing that without SIRT1, levels of HIF-1α escalate and HIF-1α dependent gene expression is upregulated. This HIF-1α stabilization is through deacetylation by SIRT1 (Chen, Dioum et al. 2011). However, SIRT1 has also been shown to repress HIF-1α activity (Lim, Lee et al. 2010). Hypoxia-inducible factor 1 alpha (HIF-1α) adapts cells to stresses like hypoxia and promotes steps in tumor progression and aggressiveness (Gruber, Greiner et al. 2004). Some of the genes it regulates include heme oxygenase-1 (HO-1), vascular endothelial growth factor (VEGF), erythropoietin (EPO), and inducible nitric oxide synthase (iNOS), all hallmarks of cancer and the aging process (Gregg 2003; Kang, Kim et al. 2005; Lim, Lee et al. 2010). Gomes et al's finding contributes to understanding HIF-1α stabilization through SIRT1 induction of VHL (an E3 ubiquitin ligase, short for Von Hippel-Lindau) at the protein level. Previously, VHL's role in stabilizing HIF-1α had also been established (Nicholas 2008).

One reason why CR is involved in extending lifespan is because intracellular NAD^+ and NADH levels are modulated by nutrient deprivation, energy consumption, or hypoxia. CR increases the NAD^+: NADH ratio and extends the lifespan of yeast by activating Sir2. In mammals, this is partly done by SIRT1 promoting the functionality of mitochondrial genes through deacetylating PGC1-α, a powerful regulator of ROS removal and mitochondrial metabolism (Austin and St-Pierre 2012).

Now can aging be reversed?

Gomes et al has found that deleting SIRT1 in 2-4 month mice accelerated aging. They also observed that this process is marked by a decrease in all mitochondrial genes responsible for oxidative phosphorylation and no change in nuclear genome.

To find whether increase in NAD^+ could rescue SIRT1 activity, Gomes et al injected 22-month-old mice twice daily for a week with nicotinamide mono nucleotide (NMN) – a molecule known to increase levels of NAD^+.

They found that deleting SIRT1 accelerates aging, whereas raising NAD^+ levels in old mice not only played a role in nuclear genome regulation but also in regulating and restoring the mitochondrial oxidative phosphorylation function in a SIRT1-dependent manner. If the compound was given early enough—prior to excessive mutation accumulation—within days, some aspects of the aging process could be reversed. It is also noteworthy that raising the NAD^+ levels (in the form of NMNAT1) in the nucleus improved mitochondrial function whereas raising NAD^+ levels in the Golgi/Cytoplasm/Mitochondria (in the form of NMNAT2 and NMNAT3) did not improve mitochondrial function, further

pointing to the essential role of nucleus in the process.

To examine how SIRT1 exerts its function on mitochondrial function, Gomes et al inevitably investigated its role on PGC1-α regulation. However, surprisingly, enhancement of mitochondrial function was independent of PGC1-α. Further analysis showed that SIRT1 stabilizes HIF-1α which then modulates c-myc's ability to activate TFAM (responsible for mitochondrial biogenesis) (Hock and Kralli 2009). Therefore, with aging and a concurrent decrease in NAD^+ levels, *SIRT1* loses its ability to regulate HIF-1α, c-myc, and consequently TFAM; thus creating a pseudohypoxic state that disrupts PGC-1α/β-independent nuclear-mitochondrial communication and contributing to the decline in mitochondrial function over time. This loss of communication reduces the cell's ability to make energy, and thus signs of aging and disease become apparent, a process that is reversible thanks to Gomes et al's study. In the end, although this study is an important contribution to the field of cancer research as well as mitochondrial and metabolic diseases, the findings are of the effect of treatments and not causal. Therefore, firstly the causal correlation need to be investigated more in depth and the effect of treatments on skeletal muscle physiology should also be addressed (i.e. wheel run, and exercise exhaustion and endurance tests).

Figure 1

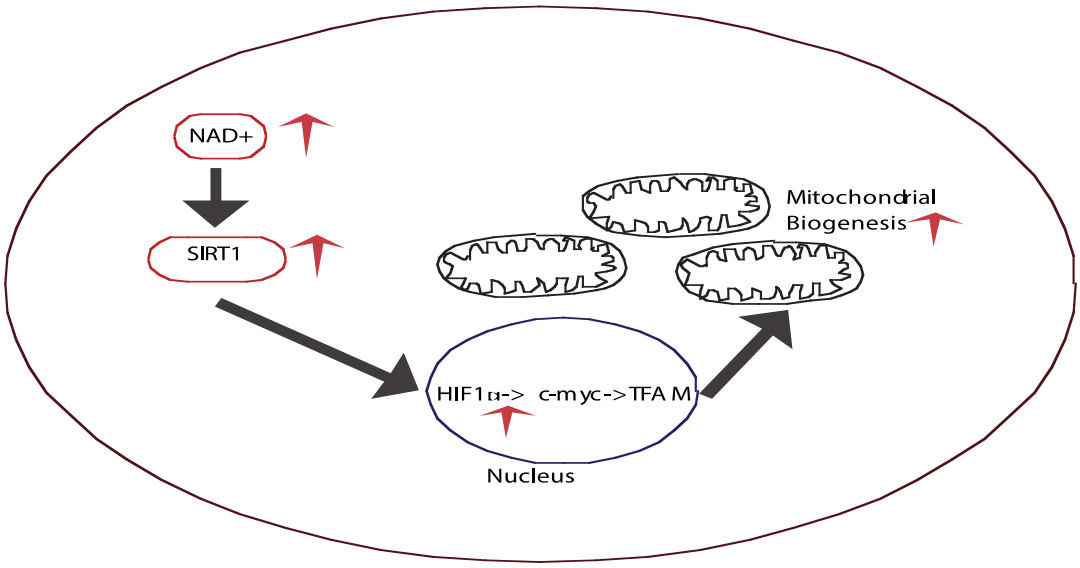

Figure 1. Simplified reconstruction of Gomes et al.'s mitochondrial biogenesis mechanism through increase in NAD^+ levels which in turn increases SIRT1 levels that can stabilize nuclear HIF1-α. This stabilization leads to increase of mitochondrial function through TFAM upregulation.

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Reference:

Austin, S. and J. St-Pierre (2012). "PGC1α and mitochondrial metabolism – emerging concepts and relevance in ageing and neurodegenerative disorders." Journal of Cell Science **125**(21): 4963-4971.

Borra, M. T., B. C. Smith, et al. (2005). "Mechanism of Human SIRT1 Activation by Resveratrol." Journal of Biological Chemistry **280**(17): 17187-17195.

Cadenas, E. (2004). "Mitochondrial free radical production and cell signaling." Molecular Aspects of Medicine **25**(1–2): 17-26.

Cadenas, E. and K. J. A. Davies (2000). "Mitochondrial free radical generation, oxidative stress, and aging." Free Radical Biology and Medicine **29**(3–4): 222-230.

Chen, D., J. Bruno, et al. (2008). "Tissue-specific regulation of SIRT1 by calorie restriction." Genes & Development **22**(13): 1753-1757.

Chen, R., E. M. Dioum, et al. (2011). "Hypoxia increases Sirtuin 1 expression in a hypoxia inducible factor-dependent manner." Journal of Biological Chemistry.

Cui, H., Y. Kong, et al. (2012). "Oxidative Stress, Mitochondrial Dysfunction, and Aging." Journal of Signal Transduction **2012**: 13.

Ghosh, S., B. Liu, et al. (2013). "Resveratrol activates SIRT1 in a Lamin A-dependent manner." Cell Cycle **12**(6): 872-876.

Gomes, A. P., N. L. Price, et al. (2013). "Declining NAD+ Induces a Pseudohypoxic State Disrupting Nuclear-Mitochondrial Communication during Aging." Cell **155**(7): 1624-1638.

Gregg, L. S. (2003). "Targeting HIF-1 for cancer therapy." Nature Reviews Cancer **3**(10): 721-732.

Gruber, G., R. Greiner, et al. (2004). "Hypoxia-inducible factor 1 alpha in high-risk breast cancer: an independent prognostic parameter?" Breast Cancer Res **6**(3): R191 - R198.

Harman, D. (1956). "Aging: A Theory Based on Free Radical and Radiation Chemistry." Journal of Gerontology **11**(3): 298-300.

Harman, D. (1983). "Free radical theory of aging: Consequences of mitochondrial aging." AGE **6**(3): 86-94.

Hock, M. B. and A. Kralli (2009). "Transcriptional Control of Mitochondrial Biogenesis and Function." Annual Review of Physiology **71**(1): 177-203.

Jacobs, H. T. (2003). "The mitochondrial theory of aging: dead or alive?" Aging Cell **2**(1): 11-17.

Jin, Q., T. Yan, et al. (2007). "Cytoplasm-localized SIRT1 enhances apoptosis." Journal of Cellular Physiology **213**(1): 88-97.

Kang, M., H. Kim, et al. (2005). "The effect of age and calorie restriction on HIF-1-responsive genes in aged liver." Biogerontology **6**(1): 27-37.

Lim, J.-H., Y.-M. Lee, et al. (2010). "Sirtuin 1 Modulates Cellular Responses to Hypoxia by Deacetylating Hypoxia-Inducible Factor 1±." Molecular Cell **38**(6): 864-878.

Lim, J.-H., Y.-M. Lee, et al. (2010). "Sirtuin 1 Modulates Cellular Responses to Hypoxia by Deacetylating Hypoxia-Inducible Factor 1α." Molecular Cell **38**(6): 864-878.

Lin, Z. and D. Fang (2013). "The Roles of SIRT1 in Cancer." Genes & Cancer.

Nicholas, C. D. (2008). "Hypoxia, HIF1 and glucose metabolism in the solid tumour." Nature Reviews Cancer **8**(9): 705-713.

Satoh, A., C. S. Brace, et al. (2013). "Sirt1 Extends Life Span and Delays Aging in Mice through the Regulation of Nk2 Homeobox 1 in the DMH and LH." Cell metabolism **18**(3): 416-430.

Zu, Y., L. Liu, et al. (2010). "SIRT1 Promotes Proliferation and Prevents Senescence Through Targeting LKB1 in Primary Porcine Aortic Endothelial Cells." Circulation Research **106**(8): 1384-1393.