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INVITED REVIEW

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How telomere dynamics are influenced by the balance between mitochondrial efficiency, reactive oxygen species production and DNA damage

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Abstract

It is well known that oxidative stress is a major cause of DNA damage and telomere attrition. Most endogenous reactive oxygen species (ROS) are produced in the mitochondria, producing a link between mitochondrial function, DNA integrity and telomere dynamics. In this review we will describe how ROS production, rates of damage to telomeric DNA and DNA repair are dynamic processes. The rate of ROS production depends on mitochondrial features such as the level of inner membrane uncoupling and the proportion of time that ATP is actively being produced. However, the efficiency of ATP production (the ATP/O ratio) is positively related to the rate of ROS production, so leading to a trade-off between the body's energy requirements and its need to prevent oxidative stress. Telomeric DNA is especially vulnerable to oxidative damage due to features such as its high guanine content; while repair to damaged telomere regions is possible through a range of mechanisms, these can result in more rapid telomere shortening. There is increasing evidence that mitochondrial efficiency varies over time and with environmental context, as do rates of DNA repair. We argue that telomere dynamics can only be understood by appreciating that the optimal solution to the trade-off between energetic efficiency and telomere protection will differ between individuals and will change over time, depending on resource availability, energetic demands and life history strategy.

KEYWORDS
DNA damage, energetics, oxidative stress, telomerase

1 | INTRODUCTION

The ATP that is required to drive cellular processes is primarily created in the mitochondria in a process that consumes oxygen and carbohydrate substrates; the details are complex (and are summarised in the next section) but involve the transfer of electrons through a series of complexes and the movement of protons across the inner mitochondrial membrane (IMM). However, some electrons leak to react with molecular oxygen and form the incompletely reduced radical superoxide ($O_2^{\bullet-}$) and other downstream reactive oxygen species (ROS). These include hydrogen peroxide (H_2O_2) and hydroxyl radicals ($^{\bullet}OH$); in this review we refer collectively to reactive molecules as ROS, although other reactive molecules are also formed with for example, nitrogen. ROS have many beneficial roles in cellular

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processes (Murphy et al., 2011). They are involved as signalling molecules activating pathways to initiate biological processes, such as stem cell renewal and capacitation of human sperm, processes referred to as redox biology (Schieber & Chandel, 2014). ROS also act as a first immune defence against pathogens and as activators of T- and b-cells, thus being vital parts of the immune system (Murphy et al., 2011; Schieber & Chandel, 2014).

However, when levels of ROS exceed those necessary for cellular signalling and overwhelm the detoxification capacity of a biological system, they can cause oxidative damage to a wide range of biomolecules, including DNA. We refer to this state of ROS imbalance as one of oxidative stress; the fact that oxygen is both essential for aerobic metabolism and yet also a potential cause of damage is referred to as the "oxygen paradox". As highlighted in the review title of Markkanen (2017)—"Not breathing is not an option"—the oxidative damage arising from mitochondrial respiration is to some extent inevitable, but may have major consequences for the viability of cells, tissues and the whole organism. This leads to complex interactions between energy expenditure and rates of senescence, leading to diverse life history trade-offs (Hou & Amunugama, 2015). One cellular component that is especially vulnerable to oxidative damage is the telomeric region of DNA (for reasons given below); this is particularly relevant to life history evolution since damage to the telomeres can result in faster attrition, leading to more rapid cellular and organismal ageing. Furthermore, since ROS accelerate mitochondrial decay, with consequences for ROS control, the process is one in which ageing mitochondria can enter a vicious cycle of increasing ROS production (Barja, 2004; Finkel & Holbrook, 2000; Harman, 1956; Pańczyszyn et al., 2020; Shigenaga et al., 1994).

A vast body of evidence from the biomedical literature reveals the complexities of telomere dynamics, elevated attrition from oxidative stress (von Zglinicki, 2002), and epigenetic master regulators of telomere dynamics such as histone and DNA methyltransferases (Blasco, 2007). One review summarizes these effects with the statement "no other chromosome structure has been linked to major human health issues as tightly as telomeres, and in particular their length, to the point that telomere length has become an obliged biomarker for anyone analysing the impact of any factor (either environmental or genetic) into human fitness, a forteriori in aged populations" (Kappei & Londoño-Vallejo, 2008). Therefore, understanding the mechanisms underlying mitochondrial ROS generation and their consequences for oxidative balance and telomere dynamics is crucial for evaluating alternative hypotheses for the evolution of senescence. This is perhaps no better demonstrated than in the work by Tanaka et al. (1998, 2000) who reported that two thirds of Japanese people that lived to be a 100 years old shared the same single nucleotide change in the genetic code for a subunit of complex I in the mitochondria, compared to 45% for the rest of the population. Thus, this genotype alone makes you 50% more likely to live to a 100. Furthermore, the study by Tanaka et al. (1998) showed that people with this genotype were only half as likely to end up in hospital—for any reason at all—in the second half of their lives and less likely to suffer from age-related disease. The underlying reason for these results is likely to be that people carrying these mutations have a reduced rate of free-radical leakage from the mitochondria (Tanaka et al., 2000). At any given moment, this advantage may be imperceptible. Over a lifetime, however, the cumulative benefits of a reduced attack rate of aggressive molecules leaking from the thousands of mitochondria in each cell will reduce the risk of cancer and other disease, and so be evolutionarily significant. However, as we reveal later, this mutation is likely to result in the mitochondria being less efficient at producing ATP.

Some ROS molecules are neutralised by innate antioxidants, such as superoxide dismutase (SOD), whose evolution itself is a testimony to the detrimental effects of reactive molecules and their actions. Other ROS may be scavenged by exogenous, dietary antioxidants, whose relative importance has much been debated (Catoni et al., 2008; Costantini et al., 2010; McGraw et al., 2010; Pamplona & Costantini, 2011; Seifried et al., 2007) not least because dietary antioxidant supplementation has become increasingly questioned in humans due to their potential procancerous consequences; cancer cells have a much higher ROS production than normal cells so benefit more from antioxidant defences than normal cells (Ahmed & Lingner, 2018). However, given the extensive previous consideration of the role of antioxidants, including in the ecological literature (Buttemer et al., 2010; Catoni et al., 2008; McGraw et al., 2010), we consider the subject of antioxidant defences to be outside the scope of this review. Instead, we primarily focus on the costs of oxidative stress from the perspective of telomere biology. We first consider how the rate of endogenous ROS production is not fixed but varies with mitochondrial state. This generates a trade-off between the benefits of minimising ROS versus maximising the energetic efficiency of the mitochondria. We then explore how ROS can cause DNA damage, with consequences for the rate of telomere attrition, and review the mechanisms that have evolved to remove or repair damaged DNA bases (focussing on telomeric regions). We evaluate how interactions between mitochondrial energy generation, ROS management, and repair of oxidatively damaged DNA have influenced patterns of telomere loss and hence the rate of senescence. To date very few empirical studies have attempted to integrate these processes (exceptions being Casagrande et al., 2020; Olsson, Friesen, et al., 2018), but recent conceptual and technical advances have opened up this field, and we conclude by highlighting the opportunities that have now arisen to make real advances in this area.

2 | MITOCHONDRIAL FUNCTION

2.1 | The link between ATP production and oxidative stress

The production of ATP by eukaryote mitochondria is dependent on the existence of a proton gradient across the IMM. The gradient is produced by the pumping of protons across the IMM, a multistep process termed the electron transport system (ETS) or electron transport chain that consumes oxygen (and is the major reason for

oxygen consumption by the body) (Echtay, 2007; Mailloux et al., 2013). The existence of this proton-motive force causes protons to flow back across the inner membrane (Figure 1a). Most of these protons flow through the ATP synthase enzyme that straddles the IMM; the enzyme contains a complex ring shape that creates a pore in the membrane through which the protons pass. Their movement through the ATP synthase molecule drives the phosphorylation of ADP to form ATP. The "coupling" between the oxygen-consuming and ATP-producing components of this process is termed oxidative phosphorylation (OXPHOS) (Mailloux & Harper, 2011; Mailloux et al., 2013).

However, the proton gradient can also be dissipated by protons "leaking" across the IMM without passing through ATP synthase, so without producing any ATP (Figure 1a). This leak of protons across the membrane means that ATP synthesis becomes less efficient, since energy and oxygen must then be consumed in pumping additional protons across the membrane in order to restore the proton gradient (so-called LEAK respiration; see Table 1 for a full list of terms and definitions) (Mailloux & Harper, 2011; Salin et al., 2015). During this LEAK respiration, oxygen consumption is uncoupled from ATP production, since oxygen is consumed by the ETS but without generating any ATP. The rate at which oxygen is being consumed to offset the proton leak will in part depend on the proportion of time that the mitochondria spend "idle" (i.e., not producing ATP): periods without ATP production allow the proton gradient to build up, so increasing the rate of leakage (Mailloux & Harper, 2011). LEAK respiration thus varies over time, but can account for up to 30% of basal metabolic rate in endotherms (Mookerjee et al., 2010).

One benefit of a leakier (and hence less efficient) mitochondrial membrane is that it reduces the rate of production of ROS. ROS are highly reactive molecules that have a number of functions (e.g., signalling, phagocytosis) (Murphy et al., 2011), but when in excess can cause oxidative damage to cellular lipids, proteins and DNA, leading to a state of oxidative stress (Finkel & Holbrook, 2000). As a result ROS are thought to be an important contributor to cellular ageing (Brand, 2000), and so to be a selective force in shaping the evolution of ageing and life history strategies (Beckman & Ames, 1998; Dowling & Simmons, 2009; Hou & Amunugama, 2015; Metcalfe & Alonso-Alvarez, 2010; Monaghan et al., 2009). The mitochondria are the major endogenous source of ROS in living cells, since ROS are continuously produced (in the form of the highly reactive superoxide molecule) as a byproduct from the ETS (Figure 1a; Brand, 2000; Murphy, 2009). In vitro assays have shown that the rate of ROS production positively correlates with the strength of the proton gradient across the IMM (and hence the degree of coupling) (Mailloux & Harper, 2011). As a consequence, a leakier IMM produces fewer ROS, as does the synthesis of ATP since this reduces the proton gradient (Brand, 2000; Mookerjee et al., 2010; Murphy, 2009; Roussel et al., 2019). It is important to note that the positive relationship between the proton gradient and the rate of ROS production is an accelerating rather than linear one, and so the ratio of ROS production

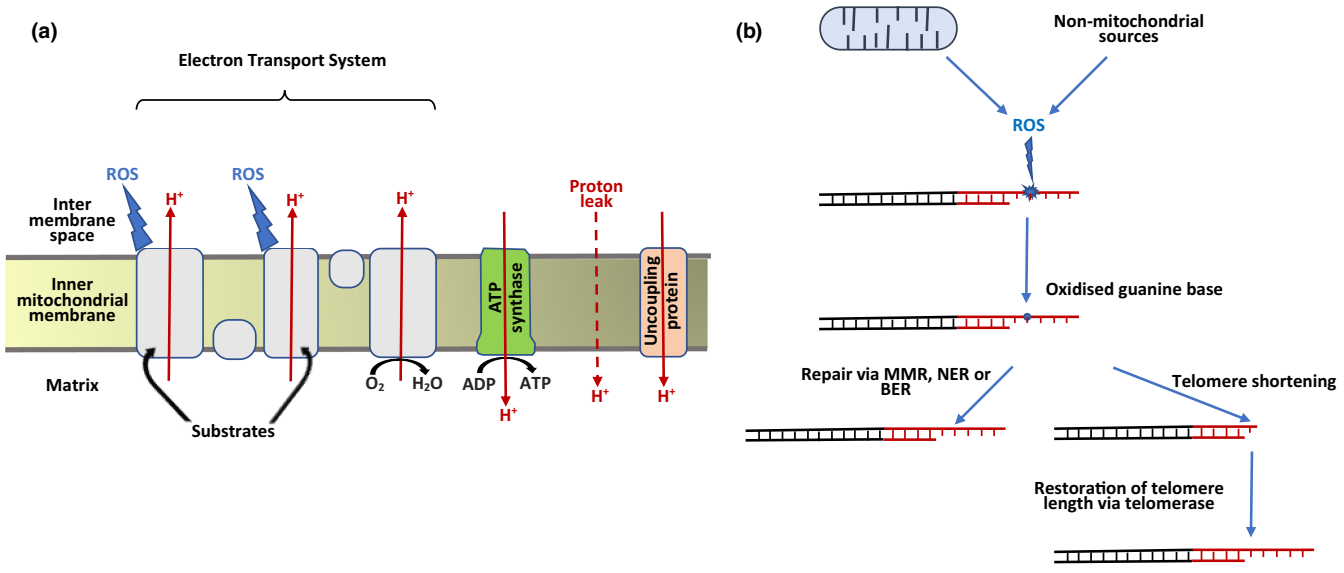

FIGURE 1 (a) Diagram illustrating the basic processes in the mitochondria that influence the production of reactive oxygen species (ROS) and hence telomere attrition. The inner mitochondrial membrane (IMM) contains the various complexes that make up the ETS, which uses substrates and oxygen to pump protons across the IMM, so creating a proton gradient. The back-flow of these protons through ATP synthase drives the production of ATP from ADP. ROS are produced as a byproduct of the proton gradient. Both the rate of ROS production and the efficiency of ATP production are reduced if protons leak back across the IMM or if they pass through uncoupling proteins. (b) ROS produced either by the mitochondria or other processes can cause oxidative damage to telomeric regions of the DNA (shown in red), with the commonest form of damage being oxidized bases, especially guanine. This damage can be repaired by a number of processes (e.g., mismatch repair [MMR], nucleotide excision repair [NER] and base excision repair [BER]). Repair is less likely in the single stranded terminal section of the telomere. If this oxidative damage is not repaired then telomeres will shorten faster, although in some situations the telomeres can be restored by the enzyme telomerase

TABLE 1 Definitions of the main terms and indices used to describe mitochondrial function and efficiency, together with an indication of whether a high value of an index indicates a high or low efficiency of converting oxygen into ATP

Term or index	Definition
OXPHOS	Oxidative phosphorylation—the conversion by the mitochondria of ATP to ADP, consuming oxygen in the process
ROUTINE	Rate of oxygen consumption when mitochondria are producing ATP
LEAK	Rate of oxygen consumption required to maintain the proton gradient across the inner mitochondrial membrane (IMM) at times when no ATP is being produced
Mitochondrial (un)coupling	(Breaking of) the link between maintaining the IMM proton gradient and generating ATP
Uncoupling proteins (UCP)	Endogenous proteins that increase leakage of proteins across IMM, so increasing LEAK and reducing efficiency of ATP production
Mitochondrial efficiency	Efficiency of producing ATP, measured using one of the indices below
Indices	
Respiratory control ratio (RCR)	ROUTINE/LEAK (high values = more efficient)
Net phosphorylation efficiency	1 – (LEAK/ROUTINE) (high values = more efficient)
Leak: routine flux control ratio, $FCR_{L/R}$	LEAK/ROUTINE (high values = less efficient)
ATP/O (or P/O)	ATP produced/(2*ROUTINE) (high values = more efficient)

to ATP production increases sharply as the need for ATP generation decreases (Roussel et al., 2019).

Since a leakier IMM requires a greater oxygen consumption to maintain the proton gradient, at the intraspecific level this can result in a negative correlation between whole-animal metabolic rate and the rate of ROS production (Salin, Auer, Rudolf, et al., 2015). This is the opposite of the commonly-assumed pattern of a high metabolic rate causing greater oxidative stress (Sohal & Weindruch, 1996), but supports Brand's "uncoupled to survive" hypothesis which posits that animals with more uncoupled mitochondria could have a reduced rate of ageing (Brand, 2000; Speakman et al., 2004). With respect to variation in telomere dynamics within species, individuals with more uncoupled mitochondria should therefore show slower rates of telomere attrition, due to the reduced production of ROS and hence lower level of oxidative damage to DNA strands.

2.2 | Mitochondrial function is dynamic

It is becoming increasingly apparent that both the rate and efficiency with which mitochondria produce ATP varies both between individual organisms and over time, and this has consequences for the rate of ROS production and hence the risk of oxidative damage to telomeres (Koch et al., 2021). Before exploring this further, however, it is important to consider what we mean by "mitochondrial efficiency", since authors have used a range of different metrics to describe mitochondrial function. The rate at which mitochondria are consuming oxygen while producing ATP is usually called the ROUTINE or State 3 respiration rate; it comprises the oxygen routinely required to offset the leak as well as that used in driving the ATP synthase enzyme (i.e., OXPHOS+LEAK). The respiratory control ratio (RCR, Table 1) measures the degree of coupling, with higher values indicating tighter coupling (i.e., there is relatively little oxygen being wasted in combating the leakage of protons across the IMM (Chance & Williams, 1955)). There are several variants on this ratio (Table 1), such as the net phosphorylation efficiency (Shama et al., 2016), which is a proxy for the proportion of oxygen consumed that is contributing to OXPHOS, and the $FCR_{L/R}$, which gives the equivalent proportion that is contributing to LEAK (Stier et al., 2017). A high net phosphorylation efficiency, or low $FCR_{L/R}$, indicates a high level of coupling so that little oxygen is being used to offset the leakage of protons across the IMM. All of these ratios have been taken to be measures of mitochondrial efficiency (Koch et al., 2021)—but it is important to note that they are all entirely based on measures of oxygen consumption and do not involve any quantification of ATP production. This is an important caveat, since they do not measure the animal's efficiency at performing work.

The level of mitochondrial coupling can change in response to environmental conditions. For example, penguin chicks facing food shortage have been shown to increase mitochondrial coupling, so increasing the efficiency of fuel use, with responses becoming stronger if the period of food shortage becomes prolonged; similar responses have been found in short-term fasts in ducklings (Monternier et al., 2017). However, this comes at a cost of increased ROS levels, since it requires a higher proton gradient across the IMM (Salin, Villasevil, Anderson, Auer, et al., 2018). Moreover, a true assessment of an animal's mitochondrial efficiency and thus its ability to work at a high rate requires quantification of ATP production as well as oxygen consumption. Until recently this has rarely been done since it is technologically challenging, and so was generally restricted to assays in a standard laboratory setting using mammalian subjects. This situation is now changing, as more flexible methods have been developed to allow simultaneous measurement of both oxygen consumption and ATP production in small tissue samples from non-model organisms (Bryant et al., 2018; Salin et al., 2012, 2016). This has revealed substantial variation amongst individual animals in the

amount of ATP produced per unit of oxygen consumed by the mitochondria (termed the ATP/O or P/O ratio; high values = efficient mitochondria; Table 1). For example, up to twofold variation in the ATP/O ratio exists amongst individual brown trout *Salmo trutta* of the same age, sex, body size and nutritional state (Salin, Villasevil, Anderson, Selman, et al., 2018). Some individuals are thus twice as efficient as others of the same species at producing ATP within their mitochondria—a finding which is completely hidden in conventional measures of metabolic rate, since measurements of whole-animal oxygen consumption do not tell us anything about ATP production (Salin, Auer, Rey, et al., 2015).

Direct measurements of ATP production have also revealed that the RCR, net phosphorylation efficiency and $FCR_{L/R}$ are not necessarily reliable indicators of the efficiency with which the mitochondria produce ATP. Indeed, in some situations the RCR can be negatively related to ATP/O, even though it is routinely presumed to be positively related to ATP production efficiency (Salin, Villasevil, Anderson, Selman, et al., 2018). It is thus important to realise the assumptions and limitations of the various indices of mitochondrial efficiency (Koch et al., 2021); the ATP/O ratio perhaps has the greatest ecological relevance since it relates whole-animal metabolic rate to energy production by the cells.

The nonlinear relationship between the proton gradient across the IMM and the rate of ROS production makes it feasible for ROS to be managed by slightly increasing the leak of protons through the membrane, without overly compromising energy production. This is termed "mild uncoupling", and is thought to be achieved by activation of both the uncoupling proteins (UCPs) and adenine nucleotide translocase (ANT), molecules that are embedded in the membrane itself (Figure 1a; Echtay, 2007; Mailloux & Harper, 2011; Mailloux et al., 2013). The family of UCPs is evolutionarily well conserved although the exact role of each isoform is still being determined (Woyda-Ploszczyca & Jarmuszkiewicz, 2017). Current evidence suggests that UCP2 and UCP3 are the major players in the regulation of ROS levels within mammalian mitochondria (Brand, 2000; Echtay, 2007; Mailloux et al., 2013; Toime & Brand, 2010), while a similar role may be played by avian UCP (avUCP) in birds (Rey et al., 2010) and UCP1 in fish (Bryant et al., 2018). When activated, these UCPs allow protons to pass through the IMM, thus reducing the strength of the proton gradient. Their precise mode of functioning is still being determined, but it is clear that their activity is responsive to ROS levels. UCP2 and UCP3 are deactivated by the attachment of a glutathione molecule, and there is evidence that this occurs when superoxide levels are low (Mailloux et al., 2013). If superoxide levels then rise within the mitochondrial matrix, this triggers the deglutathionylation of UCP2 and UCP3, which has two consequences: the glutathione is a potent scavenger of the superoxide radicals, while the activation of the UCPs triggers the leakage of protons across the IMM, so reducing the rate of ROS production (Mailloux & Harper, 2011; Mailloux et al., 2013).

This reversible process of glutathionylation of UCP2 and UCP3 thus provides the means to regulate the production of ROS by the mitochondria, and thus the risk of oxidative stress. Preventing their production in this way is thought to be energetically more efficient than employing antioxidants to scavenge them after they have been generated (Mailloux & Harper, 2011). The regulation can be rapid, since levels of both the membrane potential and of ROS within the mitochondria have been found to cycle over timescales of less than a minute (Mailloux & Harper, 2011). However, the cost to the cell (and therefore the animal) of reducing the rate of ROS production is that the efficiency of energy generation is reduced. There is thus a dynamic trade-off between the need to be energetically efficient and the need to reduce oxidative damage to biomolecules including telomeres.

There is increasing evidence—primarily from the otherwise unrelated fields of gerontology and sports science - that the efficiency with which the mitochondria produce ATP can influence aerobic performance. For instance, recent research has shown that the ATP/O ratio is significantly and positively correlated with maximum aerobic capacity, muscle strength and locomotor speed in humans (Coen et al., 2013; Conley, 2016; Conley et al., 2013; Distefano et al., 2018; Zane et al., 2017). Meanwhile in the field of agriculture there has been interest in using information on mitochondrial traits when selecting for fast-growing domesticated animals (Bottje, 2015; Bottje & Carstens, 2009), and the efficiency of ATP production has been shown to be positively related to the efficiency of growth in domesticated pigs and chickens (Fu et al., 2017; Hudson et al., 2017). The same has been shown in wild-derived brown trout, where individual variation in the ATP/O ratio in liver mitochondria is a significant predictor of their growth rate and growth efficiency, at each of two different levels of food availability (Salin et al., 2019). Intraspecific variation in the efficiency of the mitochondria thus becomes key to how the whole animal can perform.

3 | TELOMERE DAMAGE AND REPAIR

3.1 | Evidence that ROS cause oxidative damage to telomeres

We now turn to consider how the ROS produced by the mitochondria have consequences for telomere dynamics. Oxidative stress can have a detrimental effect on telomeres through different routes acting both during, and independent from, cell replication. It can inflict both single-strand breaks (SSBs) and double-strand breaks (DSBs) in DNA, so affecting the telomere sequence, and can have strikingly inhibitory effects on telomerase activity, a major telomere repair pathway (Ahmed & Lingner, 2018). ROS are capable of generating around 100 different types of oxidatively damaged bases (Cadet & Wagner, 2013). It has been estimated that as many as 20,000 DNA base lesions are generated every day in every cell under unstressed conditions (Markkanen, 2017). Other work has estimated the number of oxidized bases as 1200 per cell cycle, with 3000 SSBs and 50 DSBs (Chen et al., 2020). The most studied base to receive oxidative damage is guanine. Its low oxidation potential makes it particularly susceptible to reaction with singlet oxygen,

leading to the formation of 8-oxo-7,8-dihydroguanine (8-oxoGua), a reaction that is estimated to occur on average 100–500 times per day in the genome of each human cell (Poetsch, 2020), rising to 100,000 times in cancer cells (Chen et al., 2020). These numbers are quite extraordinary, given that a single unrepaired DSB can cause cell cycle arrest (Chen et al., 2020).

There is evidence that telomeric DNA is particularly reactive with ROS and so prone to oxidative damage. The base sequence of telomeres (T_2AG_3), with its high guanine content, makes telomeres more prone to 8-oxoGua formation in vitro than the rest of the genome (Kawanishi & Oikawa, 2004). This is also partly due to the preferential binding of Fe^{2+} to telomere repeats, which allows for hydroxyl radical production via Fenton reactions (Fouquerel et al., 2016; Markkanen, 2017). As a result, double-stranded telomeric DNA is more susceptible to cleavage by ROS than nontelomeric DNA (Oikawa & Kawanishi, 1999). Furthermore, higher frequencies of H_2O_2 or alkylating-agent induced SSBs and DNA gaps are found in telomeres than in microsatellites and bulk genome DNA (reviewed in Fouquerel et al., 2016). However, the shielding effect against oxidation of the proteins that protect telomeres (e.g., the shelterin complex in mammals) is strongly suggested by there being 50% fewer lesions on naked telomeres than on mini-satellites (Fouquerel et al., 2016). Interestingly, recent work reveals a rapid rate of evolution in the proteins associated with telomeric regions, demonstrating that a highly unconserved protein machinery protects the highly-conserved telomere and its functions (Saint-Leandre & Levine, 2020). Other reasons suggesting that telomeres are particularly vulnerable to oxidative damage are that the DNA damage response (DDR) is repressed within intact telomeres and that parts of the telomere sequence is single-stranded, that is, there is no template from which to recognise and replace nucleotides. It should be noted, however, that much of the above evidence comes from in vitro studies and it is less clear if oxidative lesions arise more frequently in vivo at telomeres compared to the rest of the genome (Fouquerel et al., 2016).

Two of the leaders in this field, Ahmed and Lingner (2020) conclude that "the consequences of persistent oxidative lesions on telomere integrity are not well understood" and report position effects of 8-oxo-dG (the deoxyriboside form of 8-oxoGua) when present within a DNA substrate in vitro, either inhibiting or stimulating telomerase production, depending on its location in the telomeric DNA (Fouquerel et al., 2016). To what extent such effects are detectable, and make interpretation of telomere dynamics in vivo problematic, is currently not well understood (Ahmed & Lingner, 2020). The impact of DSBs depends on their genomic position, with DSBs near telomeric regions being much more likely to result in gross chromosomal rearrangements (GCR) and chromosomal instability than DSBs occurring within interstitial regions (Silva et al., 2017). Ultimately, telomeres may fuse to form dicentric chromosomes, a sequence of events that can lead to chromothripsis, a form of genetic catastrophe with tens of thousands of genomic rearrangements that occur primarily widely across different tumour types (Cleal & Baird, 2020).

3.2 | Does oxidative stress cause faster telomere shortening in vivo?

The main process during which telomeres suffer loss of base pairs and so shorten is at replication, the process where two identical replicas are formed from one original DNA molecule. This leads to well-described telomere loss in all cells that lack telomerase, such as in most somatic endotherm cells. This process of replication-related attrition occurs for a number of reasons: (i) replication cannot proceed to the very end of the chromosome, resulting in telomere loss at every round of replication (the end-replication problem). (ii) Stochastic loss of telomere sequence may occur when single-stranded highly stable G-quadruplexes, including the t-loop structures, are enzymatically unwound, and when (iii) TERRA RNA forms hybrid DNA/RNA structures that interfere with replication. (iv) Telomere replication is unidirectional and driven from subtelomeric regions, and so stalled replication forks cannot be rescued by converging forks coming from the chromosome end (Ahmed & Lingner, 2018). The most common telomeric mutation, 8-oxo-Gua, is not considered a blocking lesion per se. Thus, the cell bypasses the 8-oxo-Gua lesion during replication. The overall in vivo mutation frequency of 8-oxo-Gua lesions in templating DNA without post-replication repair is estimated to be around 19%, which is much lower than expected considering the replication bypass problem (Markkanen, 2017). Error-free bypass of 8-oxo-Gua lesions to counteract their mutational effect is achieved by invoking replicative DNA polymerases (Pols; see Markkanen, 2017 for excellent review).

The question then arises as to whether the oxidative damage to telomeric regions that arises due to ROS produced by the mitochondria causes a faster rate of telomere shortening, and whether this occurs in vivo. The answer appears to be yes (Figure 1b): in their recent review, Reichert and Stier (2017) concluded that 10 of 18 correlative and seven of eight experimental studies supported the hypothesis that oxidative stress caused shortening of telomeres. Part of the incentive for that review was the demonstration of a lack of relationship between simultaneous sampling of telomere attrition and oxidative stress markers (Boonekamp et al., 2017). Barnes et al. (2019) argue that mechanistic information linking oxidative stress to in vivo telomere attrition and viability can more reliably be extracted from tissues in vivo (rather than from cell culture in vitro), especially associated with inflammation. Considerable evidence links oxidative stress to short telomeres, greater chromosomal instability and inflammation that must necessarily be associated with compromised vigour (Barnes et al., 2019). For example, Graham and Meeker (2017) argue that at a normal rate of attrition, a prostate cancer cell would need more than 100 years to achieve the critical telomere length observed in prostate tumours with their high ROS levels. Thus, telomere-associated oxidative stress pathology affects viability in vivo.

The effect of innate antioxidants, such as peroxiredoxins (*PRDX1*), on telomere stability becomes obvious in *PRDX1* knock out cells: treating such cells with pro-oxidants leads to massive telomeric instability from elevated ROS levels (Ahmed & Lingner, 2020).

Its applied value has been predicted in cancer treatment (increasing ROS to kill tumors), but to what extent corresponding variation in *PRDX1* shows covariation with viability in vivo in the wild is not known. Importantly, most of this information has been researched in model organisms such as yeast (*Saccharomyces cerevisae*), roundworms (*Caenorhabditis elegans*), fruit fly (*Drosophila melanogaster*), zebra fish (*Danio rerio*), medaka (*Oryzias laticeps*), and mouse (*Mus musculus*) (Wojtczyk-Miaskowska & Schlichtholz, 2018). However, there is very limited information on ROS-telomere relations on non-model taxa in the wild.

3.3 | Repair of oxidative damage to telomeric DNA

It is clear that organisms have been under intense selection to evolve the much conserved DNA repair pathways we see today, involving around 100 enzymes, each with their specific role of repair, and more enzymes predicted to be discovered (Cadet & Davies, 2017). DSBs are repaired via homology directed repair (HDR), nonhomologous end-joining (NHEJ), and alternative-NHEJ. HDR and NHEJ are repressed in intact telomeres whereas alt-NHEJ also acts within telomeric repeats. The DNA mismatch repair (MMR) system corrects single nucleotide damage, nucleotide excision repair (NER) removes modifications that distort the helical structure, and direct reversal (DR) occurs in some taxa through which UV-induced damage is inversed without a template (Figure 1b; Marcomini & Gasser, 2015; Silva et al., 2017; Wojtczyk-Miaskowska & Schlichtholz, 2018). The base excision repair system (BER) excises bases that are chemically altered, such as through endogenous ROS exposure, but cannot repair sequences within G-quadruplexes (for reviews, see Ahmed & Lingner, 2018; Fouquerel et al., 2016; Jia et al., 2015; Poetsch, 2020). BER excises 8-oxoGua using the repair enzyme 8-oxoguanine DNA glycosylase (OGG1), leaving an apurine site (AP site). The process is rapid: 8-oxoGua sites have a half-life of around 11 min on average (Markkanen, 2017). The interplay of newly emerging DNA lesions versus repair fidelity results in a steady state of a few thousand 8-oxoGua sites and c. 15,000–30,000 AP sites in total per cell (Poetsch, 2020). Interestingly, it has been shown that ROS-induced SSBs are not directly produced by ROS itself, but are a by-product of base-excision repair processing of presumably oxidized bases (Ahmed & Lingner, 2020).

It is of relevance to ecologists that there is a circadian rhythm in OGG1 and 8-oxo-Gua repair, possibly associated with different risks of oxidative stress at different times of day (Markkanen, 2017). Another strong temporal trend in the rate of DNA repair is associated with organismal age. Many studies have confirmed that the capacity for DNA repair declines with age, resulting in the accumulation of unresolved or misrepaired DNA damage with a considerably increased risk of genomic instability (Chen et al., 2020). This coincides with a decline in telomere length with age in most endotherms (and at least some ectotherms), and with a corresponding increase in overall pathology (Chakravarti et al.,2021; Kruk et al., 1995; Mensà et al., 2019).

The enzyme telomerase can restore telomere length (Figure 1b), but is usually downregulated in the majority of adult endotherm somatic cells (although still present in the cells of ectotherms (Olsson, Friesen, et al., 2018)). In those cells in which it is expressed, the telomere attrition that occurs during cell replication may be countered by telomerase adding DNA repeats to the 3' end using an internal moiety as a template. However, even in such cells, less than one telomere in 10 is elongated by telomerase per cell cycle on average (Marcomini & Gasser, 2015). The likelihood of telomere extension is inversely related to telomere length (yeast telomeres of 100 bp being about six times more likely to be lengthened than those of 300 bp), indicating that telomerase is more associated with repair than it is with the routine process of cell replication (Webb et al., 2013). It is worth stressing that, despite the existence of a range of repair mechanisms, some oxidative damage persists within telomeric regions, possibly due to suppression of DNA repair proteins by the shelterin protein TRF2 (Silva et al., 2017).

4 | THE ECOLOGICAL IMPORTANCE OF MITOCHONDRIAL ROS PRODUCTION FOR TELOMERE DYNAMICS

Our aim with this section is to discuss whether the interactions between the rates of ROS production, of telomere damage and of repair seem to affect telomere dynamics in vivo, and whether these interactions are likely to have ecological or life history consequences. The evidence of fitness consequences is currently thin due to a paucity of studies, but this picture is likely to change as more sophisticated analytical approaches are developed. As identified by Fouquerel et al. (2016), methods to explicitly quantify telomere lesions are still considered relatively blunt, which is challenging for research in this area given the relatively low abundance of telomeric DNA in the genome (c. 0.025% of the genome). We focus here on easy-to-use assays that can be adopted by ecologists and evolutionary biologists to test hypotheses relating to telomere dynamics, including in natural populations. An obvious approach to assess the effects of oxidative stress on telomere attrition is to quantify relationships between parameters such as free radicals, oxidative stress markers, DNA damage/repair (e.g., assays of 8-oxo-dG or 8-oxo-Gua, chosen because they are relatively stable molecules), and telomere length and attrition. Olsson et al. (Olsson, Friesen, et al., 2018) adopted this approach in a study of wild-caught painted dragon lizards (*Ctenophorus pictus*) that were kept in the laboratory throughout the mating season. The prediction was tested that increased excision-repair will elongate telomeres. DNA repair was analysed using Flow FISH, based upon hybridization of synthetic DNA/RNA binding to telomeres in a sequence-specific manner. Less successful hybridization is expected at compromised (unrepaired) telomeric sequence. Once the telomeric sequence is repaired, it is expected to regain the ability to hybridize, resulting in more identified sequence repeats. It was found that higher superoxide levels at the beginning of the mating season predicted shorter telomeres at the end of the

mating season, while 8-oxo-dG repair rates positively influenced subsequent telomere length (Olsson, Friesen, et al., 2018). Thus, this lends support to oxidative stress telomere attrition being repaired by the BER pathway, resulting in longer telomeres. However, the study also showed that telomere lengths late in life were more related to superoxide levels measured earlier in life than superoxide measured at the same time as the telomeres. This temporal separation of cause and effect highlights the complexity in hypothesis testing of the effects of oxidative stress on telomere biology. It also suggests that there are long-term mechanistic processes involved in life history trade-offs that are determined in early life and set organisms on an attrition and, perhaps, ageing trajectory. This supports similar lifelong effects of an individual animal's early life telomere dynamics on its lifespan or late life fitness attributes, found in a range of species (Boonekamp et al., 2014; Entringer et al., 2018; Heidinger et al., 2012; Martens et al., 2021).

Olsson, Friesen, et al. (2018) also found a significant relationship between mitochondrial "content" (a combined measure of mitochondrial mass and volume) in blood and superoxide levels in blood, suggesting that more mitochondria leads to elevation of superoxide levels. However, this need not always be the case: Salin, Auer, Rudolf, et al. (2015) found no relation in trout between intraspecific variation in liver density of mitochondria (measured as cytochrome c oxidase [COX] activity) and levels of the ROS molecule H_2O_2 (produced from superoxide formed in the mitochondria). Instead, they found that an individual's liver H_2O_2 production was negatively related to its metabolic rate - consistent with the uncoupled to survive hypothesis (Brand, 2000). It should also be borne in mind that the rate of production of ROS per mitochondria depends on its level of coupling, so can change over time. Indeed, it has been shown that brown trout responded to food shortage by reducing the size of their liver (and hence liver mitochondrial content), but this was matched by an increase in the whole-liver level of H_2O_2 due to increased coupling in the mitochondria that remain (Salin, Villasevil, Anderson, Auer, et al., 2018).

Recent research on metabolically regulated telomere attrition shows great potential for explaining links between energy production, reduction in resources available for costly telomere maintenance, stress and levels of glucocorticoids (GC) (the "metabolic telomere attrition hypothesis"; Casagrande & Hau, 2019; Casagrande et al., 2020). Experimental elevation of GC in nestling great tits *Parus major* resulted in shorter telomeres associated with relevant shifts in mitochondrial parameters (e.g., LEAK respiration, mitochondrial efficiency) but no effects on oxidative stress, which lends support to short-term metabolic attrition effects (Casagrande et al., 2020). This concurs with the growing evidence that exposure to stressors in general is associated with faster telomere attrition (Chatelain et al., 2020). However, again, if the effects of oxidative stress are not manifested until some time after ROS exposure, then such effects would be overlooked in short-term experiments.

Indirect evidence for the importance of ROS for telomere attrition can also be derived from patterns of antioxidation and DNA damage/repair. Olsson et al. (2012) and Olsson et al. (2012) took such approaches when examining sex-specific effects of innate production of SOD and associated DNA damage/repair. In painted dragon lizards, males had four times higher levels of SOD than females, whereas at the end of the mating season females had higher levels of DNA damage/repair (8-oxo-dG; (Olsson, Healey, et al., 2012)). Experimental enhancement of SOD in males by using a SOD/catalase mimetic, Eukarion 134 (EUK) made them better able to maintain their breeding coloration through the mating season, and males receiving EUK had significantly less DNA damage/repair levels (Olsson, Tobler, et al., 2012).

The above evidence of individual and temporal variation in patterns of oxidative stress, telomere damage and repair suggests that there is much scope for variation in mitochondrial function to influence telomere dynamics (Figure 1). There are an increasing number of experimental studies that show mitochondrial coupling increases with declining nutritional state (Bourguignon et al., 2017; Monternier et al., 2014, 2017; Roussel et al.,2018; Salin, Villasevil, Anderson, Auer, et al., 2018), such that animals facing food shortage maximise the efficiency of ATP production but at a cost of greater ROS production and risk of oxidative stress (Salin, Villasevil, Anderson, Auer, et al., 2018). The same shift towards greater mitochondrial efficiency has been found in other contexts where there is an elevated energetic demand, such as in the liver of lactating mice *Mus musculus* (Mowry et al., 2017) and the flipper muscles of King penguin chicks *Aptenodytes patagonicus* when they first enter the sea and so need to have high sustained muscle efficiency (Roussel et al., 2020). Similar trends are found in human skeletal muscle mitochondrial efficiency when undergoing intense exercise training (Fiorenza et al., 2019). In all cases there may be a cost to this increased efficiency of energy production in terms of a greater rate of telomere attrition (or investment in repair to prevent this), but these might nonetheless be adaptive responses given that the benefits are immediately apparent whereas the cost may not be paid until later (as in the painted dragon lizard example above (Olsson, Friesen, et al., 2018)).

5 | MITOCHONDRIA AND TELOMERES: KNOWLEDGE GAPS AND OPPORTUNITIES

There has previously been speculation that intraspecific variation in mitochondrial function or efficiency could underlie physiological trade-offs involving oxidative stress, and so explain observed variation in a diverse array of organismal fitness-related traits such as immune function, sexual ornamentation or the degree or quality of animal mating displays (Hill, 2014; Koch et al., 2017, 2021; Koch & Hill, 2018). We are now proposing that it should also influence the rate of telomere attrition. It seems clear that analysis of mitochondrial function can explain individual variation in whole-organism traits (Koch et al., 2021)—for instance, two animals can achieve the same growth rate despite having differing food intakes due to differences in their ATP/O (Salin et al., 2019). By the same logic, we can argue that this should lead to different rates of telomere loss, with the animal that has a higher ATP/O being able to grow more

efficiently, but potentially paying a cost in terms of greater rates of ROS production and so having a faster rate of telomere attrition. However, the extent to which a trade-off operating at the cellular level can scale up to create an effect seen at the whole organism is unclear, since there may be feedback or buffering mechanisms operating at these higher levels of biological organisation. This therefore needs to be tested—and in a setting where the relevant physiological and life-history trade-offs will be revealed. The research to date on mitochondrial function and efficiency has almost entirely been done in a laboratory or domesticated animal context, in which animals are typically kept in benign conditions with unlimited access to resources. This can result in little evidence of the trade-offs that would apply in the "real" world (Metcalfe & Monaghan, 2013; Reznick et al., 2000).

There is thus a real need to extend this approach into a more ecological context, testing for the first time how intraspecific variation in mitochondrial efficiency is traded off against telomere protection, and how this trade-off differs between environmental contexts. The methodological challenges of measuring mitochondrial function in wild animals are not trivial, given that samples must be kept unfrozen and analysed within hours, but there are now studies that have done this (Nord et al., 2021; Stier et al., 2019). Outcomes are likely to be taxon-specific—for instance, there is evidence in the toad *Bufo bufo* that temperature affects both ROS levels and mitochondrial efficiency, with the rate of ROS production per unit of ATP generated shown to be greater at higher temperatures (Roussel & Voituron, 2020). Conversely, cold- and warm-adapted populations of the spotted snow skink *Niveoscincus ocellatus* differed in how mitochondrial ROS production responded to environmental temperature, although their telomere dynamics were similar (Fitzpatrick et al., 2019). There are likely to be significant differences in such traits between endotherms and ectotherms, since it has recently become evident that ectotherms express telomerase, and maintain or even increase telomere length, at later life stages than do endotherms (Olsson et al., 2018). Evidence is also emerging of among-species variation in mitochondrial function, hinting at differences in the outcome of the trade-off between energetic efficiency and protection against oxidative damage. For instance, when comparing across the limited number of mammalian species studied to date, ATP/O is highest in small-bodied species (Boël et al., 2020), but shows a greater response to temporal variation in metabolic rate in larger species (Boël et al., 2019). There is also evidence of within-species variation in mitochondrial flexibility that is related to anticipated environmental variation: the plasticity in UCP expression (i.e., degree of mitochondrial uncoupling) in relation to temperature has been shown to vary among subspecies of Atlantic killifish *Fundulus heteroclitus*, with the subspecies more exposed to cold temperatures showing greater thermal plasticity in mitochondrial efficiency (Bryant et al., 2018).

There is scope for much more work of this kind, exploring how environmental conditions select for particular outcomes to our proposed trade-off between mitochondrial efficiency and telomere protection. For instance, we can predict that mitochondrial efficiency should take into account the long-term costs of greater oxidative damage, so should vary over time within individuals. We can predict that mitochondrial efficiency should be prioritised over protection against oxidative damage to telomeres in short-lived species, in semelparous species as they approach their only breeding season, and in situations where either resources are very limited or energetic demands highest. Conversely, animals living in benign conditions where energy intake is not limiting should be more likely to have lower mitochondrial efficiency, so as to protect against oxidative damage and so delay rates of senescence.

However, information on the temporal dynamics of mitochondrial function is currently lacking due to the need to cull animals in order to take the tissue samples required for mitochondrial measurements. While it is possible to make cross-sectional comparisons of animals experimentally allocated to treatment groups, these generally do not reveal the speed with which mitochondrial function can change, nor do they allow for assessment of the longer term consequences. This problem has been partially solved by measuring mitochondrial function in nucleated blood samples, allowing longitudinal measures (Stier et al., 2017). This approach showed that mitochondrial function in red blood cells of pied flycatchers *Ficedula hypoleuca* was repeatable within individuals over the course of a breeding season, but showed a shift to greater mitochondrial efficiency when parents were feeding chicks (i.e., working hardest) (Stier et al., 2019). However, whether measurements in blood cells can be taken to be representative of mitochondrial function in other tissues is still not fully clear (Koch et al., 2021), since mitochondrial function can differ among tissues (e.g., Salin et al., 2019). If these issues can be resolved then longitudinal, repeat sampling could shed real light on the interactions between stressors, glucocorticoid hormones, ROS levels, DNA damage/repair and rates of telomere attrition, since all can be measured from blood samples. Such approaches could also involve telomerase sampling, which is known to be repressed by both oxidative stress (Ahmed & Lingner, 2018) and other physiological stress (Kurz et al., 2004), causing compromised repair of the telomere sequence (Kurz et al., 2004). Given that mitochondria are maternally inherited, there is also a need for quantitative genetics experiments—ideally in the wild—using mixed paternity clutches where paternal and maternal effects on mitochondrial function and ROS production can be partitioned. If mitochondrial function shows similarities between close relatives, this opens the possibility of cross-sectional but sequential sampling of family members as an alternative approach when longitudinal sampling proves impossible. There is much fertile ground that needs to be explored.

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CONFLICT OF INTEREST

The authors declare no conflicts of interest.

AUTHOR CONTRIBUTIONS

The authors cowrote the review.

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REFERENCES

Ahmed, W., & Lingner, J. (2018). Impact of oxidative stress on telomere biology. *Differentiation*, *99*, 21–27. https://doi.org/10.1016/j.diff.2017.12.002

Ahmed, W., & Lingner, J. (2020). PRDX1 counteracts catastrophic telomeric cleavage events that are triggered by DNA repair activities post oxidative damage. *Cell Reports*, *33*(5), 108347. https://doi.org/10.1016/j.celrep.2020.108347

Barja, G. (2004). Aging in vertebrates, and the effect of caloric restriction: A mitochondrial free radical production-DNA damage mechanism? *Biological Reviews of the Cambridge Philosophical Society*, *79*(2), 235–251. https://doi.org/10.1017/S1464793103006213

Barnes, R. P., Fouquerel, E., & Opresko, P. L. (2019). The impact of oxidative DNA damage and stress on telomere homeostasis. *Mechanisms of Ageing and Development*, *177*, 37–45. https://doi.org/10.1016/j.mad.2018.03.013

Beckman, K. B., & Ames, B. N. (1998). The free radical theory of aging matures. *Physiological Reviews*, *78*(2), 547–581. https://doi.org/10.1152/physrev.1998.78.2.547

Blasco, M. A. (2007). The epigenetic regulation of mammalian telomeres. *Nature Reviews Genetics*, *8*, 299–309. https://doi.org/10.1038/nrg2047

Boël, M., Romestaing, C., Duchamp, C., Veyrunes, F., Renaud, S., Roussel, D., & Voituron, Y. (2020). Improved mitochondrial coupling as a response to high mass-specific metabolic rate in extremely small mammals. *Journal of Experimental Biology*, *223*(5), jeb215558. https://doi.org/10.1242/jeb.215558

Boël, M., Romestaing, C., Voituron, Y., & Roussel, D. (2019). Allometry of mitochondrial efficiency is set by metabolic intensity. *Proceedings of the Royal Society B-Biological Sciences*, *286*(1911), 20191693. https://doi.org/10.1098/rspb.2019.1693

Boonekamp, J. J., Bauch, C., Mulder, E., & Verhulst, S. (2017). Does oxidative stress shorten telomeres? *Biology Letters*, *13*(5), 20170164. https://doi.org/10.1098/rsbl.2017.0164

Boonekamp, J. J., Mulder, G. A., Salomons, H. M., Dijkstra, C., & Verhulst, S. (2014). Nestling telomere shortening, but not telomere length, reflects developmental stress and predicts survival in wild birds. *Proceedings of the Royal Society B-Biological Sciences*, *281*, 20133287. https://doi.org/10.1098/rspb.2013.3287

Bottje, W. (2015). Mitochondrial physiology. In C. G. Scanes (Ed.), *Sturkie's avian physiology* (6th ed., pp. 39–51). Elsevier Science Publishing.

Bottje, W. G., & Carstens, G. E. (2009). Association of mitochondrial function and feed efficiency in poultry and livestock species. *Journal of Animal Science*, *87*, E48–E63.

Bourguignon, A., Rameau, A., Toullec, G., Romestaing, C., & Roussel, D. (2017). Increased mitochondrial energy efficiency in skeletal muscle after long-term fasting: Its relevance to animal performance. *Journal of Experimental Biology*, *220*(13), 2445–2451. https://doi.org/10.1242/jeb.159087

Brand, M. D. (2000). Uncoupling to survive? The role of mitochondrial inefficiency in ageing. *Experimental Gerontology*, *35*, 811–820. https://doi.org/10.1016/S0531-5565(00)00135-2

Bryant, H. J., Chung, D. J., & Schulte, P. M. (2018). Subspecies differences in thermal acclimation of mitochondria function and the role of uncoupling proteins in killifish. *Journal of Experimental Biology*, *221*(24), UNSP jeb186320. https://doi.org/10.1242/jeb.186320

Buttemer, W. A., Abele, D., & Costantini, D. (2010). From bivalves to birds: Oxidative stress and longevity. *Functional Ecology*, *24*, 971–983. https://doi.org/10.1111/j.1365-2435.2010.01740.x

Cadet, J., & Davies, K. J. A. (2017). Oxidative DNA damage & repair: An introduction. *Free Radical Biology and Medicine*, *107*, 2–12. https://doi.org/10.1016/j.freeradbiomed.2017.03.030

Cadet, J., & Wagner, J. R. (2013). DNA base damage by reactive oxygen species, oxidizing agents, and UV radiation. *Cold Spring Harbor Perspectives in Biology*, *5*(2), a012559. https://doi.org/10.1101/cshperspect.a012559

Casagrande, S., & Hau, M. (2019). Telomere attrition: Metabolic regulation and signalling function? *Biology Letters*, *15*(3), 20180885. https://doi.org/10.1098/rsbl.2018.0885

Casagrande, S., Stier, A., Monaghan, P., Loveland, J. L., Boner, W., Lupi, S., Trevisi, R., & Hau, M. (2020). Increased glucocorticoid concentrations in early life cause mitochondrial inefficiency and short telomeres. *Journal of Experimental Biology*, *223*(15), jeb222513. https://doi.org/10.1242/jeb.222513

Catoni, C., Peters, A., & Schaefer, H. M. (2008). Life history trade-offs are influenced by the diversity, availability and interactions of dietary antioxidants. *Animal Behaviour*, *76*, 1107–1119. https://doi.org/10.1016/j.anbehav.2008.05.027

Chakravarti, D., LaBella, K. A., & DePinho, R. A. (2021). Telomeres: History, health, and hallmarks of aging. *Cell*, *184*(2), 306–322. https://doi.org/10.1016/j.cell.2020.12.028

Chance, B., & Williams, G. R. (1955). Simple and rapid assay of oxidative phosphorylation. *Nature*, *175*(4469), 1120–1121. https://doi.org/10.1038/1751120a0

Chatelain, M., Drobniak, S. M., & Szulkin, M. (2020). The association between stressors and telomeres in non-human vertebrates: A meta-analysis. *Ecology Letters*, *23*(2), 381–398. https://doi.org/10.1111/ele.13426

Chen, Y., Geng, A. K., Zhang, W. N., Qian, Z., Wan, X. P., Jiang, Y., & Mao, Z. Y. (2020). Fight to the bitter end: DNA repair and aging. *Ageing Research Reviews*, *64*, 101154. https://doi.org/10.1016/j.arr.2020.101154

Cleal, K., & Baird, D. M. (2020). Catastrophic endgames: Emerging mechanisms of telomere-driven genomic instability. *Trends in Genetics*, *36*(5), 347–359. https://doi.org/10.1016/j.tig.2020.02.001

Coen, P. M., Jubrias, S. A., Distefano, G., Amati, F., Mackey, D. C., Glynn, N. W., Manini, T. M., Wohlgemuth, S. E., Leeuwenburgh, C., Cummings, S. R., Newman, A. B., Ferrucci, L., Toledo, F. G. S., Shankland, E., Conley, K. E., & Goodpaster, B. H. (2013). Skeletal muscle mitochondrial energetics are associated with maximal aerobic capacity and walking speed in older adults. *Journals of Gerontology Series A-Biological Sciences and Medical Sciences*, *68*, 447–455. https://doi.org/10.1093/gerona/gls196

Conley, K. E. (2016). Mitochondria to motion: Optimizing oxidative phosphorylation to improve exercise performance. *Journal of Experimental Biology*, *219*(2), 243–249. https://doi.org/10.1242/jeb.126623

Conley, K. E., Jubrias, S. A., Cress, M. E., & Esselman, P. (2013). Exercise efficiency is reduced by mitochondrial uncoupling in the elderly. *Experimental Physiology*, *98*, 768–777. https://doi.org/10.1113/expphysiol.2012.067314

Costantini, D., Rowe, M., Butler, M. W., & McGraw, K. J. (2010). From molecules to living systems: Historical and contemporary issues in oxidative stress and antioxidant ecology. *Functional Ecology*, *24*, 950–959. https://doi.org/10.1111/j.1365-2435.2010.01746.x

Distefano, G., Standley, R. A., Zhang, X. L., Carnero, E. A., Yi, F., Cornnell, H. H., & Coen, P. M. (2018). Physical activity unveils the relationship

between mitochondrial energetics, muscle quality, and physical function in older adults. *Journal of Cachexia Sarcopenia and Muscle*, *9*(2), 279–294. https://doi.org/10.1002/jcsm.12272

Dowling, D. K., & Simmons, L. W. (2009). Reactive oxygen species as universal constraints in life-history evolution. *Proceedings of the Royal Society B*, *276*, 1737–1745. https://doi.org/10.1098/rspb.2008.1791

Echtay, K. S. (2007). Mitochondrial uncoupling proteins - What is their physiological role? *Free Radical Biology and Medicine*, *43*, 1351–1371. https://doi.org/10.1016/j.freeradbiomed.2007.08.011

Entringer, S., de Punder, K., Buss, C., & Wadhwa, P. D. (2018). The fetal programming of telomere biology hypothesis: An update. *Philosophical Transactions of the Royal Society B Biological Sciences*, *373*(1741), 20170151. https://doi.org/10.1098/rstb.2017.0151

Finkel, T., & Holbrook, N. J. (2000). Oxidants, oxidative stress and the biology of ageing. *Nature*, *408*(6809), 239–247.

Fiorenza, M., Lemminger, A. K., Marker, M., Eibye, K., Iaia, F. M., Bangsbo, J., & Hostrup, M. (2019). High-intensity exercise training enhances mitochondrial oxidative phosphorylation efficiency in a temperature-dependent manner in human skeletal muscle: Implications for exercise performance. *FASEB Journal*, *33*(8), 8976–8989. https://doi.org/10.1096/fj.201900106RRR

Fitzpatrick, L. J., Olsson, M., Parsley, L. M., Pauliny, A., Pinfold, T. L., Pirtle, T., While, G. M., & Wapstra, E. (2019). Temperature and telomeres: Thermal treatment influences telomere dynamics through a complex interplay of cellular processes in a cold-climate skink. *Oecologia*, *191*(4), 767–776. https://doi.org/10.1007/s00442-019-04530-w

Fouquerel, E., Parikh, D., & Opresko, P. (2016). DNA damage processing at telomeres: The ends justify the means. *DNA Repair*, *44*, 159–168. https://doi.org/10.1016/j.dnarep.2016.05.022

Fu, L., Xu, Y., Hou, Y. E., Qi, X., Zhou, L., Liu, H., Luan, Y. U., Jing, L. U., Miao, Y., Zhao, S., Liu, H., & Li, X. (2017). Proteomic analysis indicates that mitochondrial energy metabolism in skeletal muscle tissue is negatively correlated with feed efficiency in pigs. *Scientific Reports*, *7*, 45291. https://doi.org/10.1038/srep45291

Graham, M. K., & Meeker, A. (2017). Telomeres and telomerase in prostate cancer development and therapy. *Nature Reviews Urology*, *14*(10), 607–619. https://doi.org/10.1038/nrurol.2017.104

Harman, D. (1956). Aging: A theory based on free radical and radiation chemistry. *The Journal of Gerontology*, *11*, 298–300. https://doi.org/10.1093/geronj/11.3.298

Heidinger, B. J., Blount, J. D., Boner, W., Griffiths, K., Metcalfe, N. B., & Monaghan, P. (2012). Telomere length in early life predicts lifespan. *Proceedings of the National Academy of Sciences of the United States of America*, *109*, 1743–1748. https://doi.org/10.1073/pnas.1113306109

Hill, G. E. (2014). Cellular respiration: The nexus of stress, condition, and ornamentation. *Integrative and Comparative Biology*, *54*(4), 645–657. https://doi.org/10.1093/icb/icu029

Hou, C., & Amunugama, K. (2015). On the complex relationship between energy expenditure and longevity: Reconciling the contradictory empirical results with a simple theoretical model. *Mechanisms of Ageing and Development*, *149*, 50–64. https://doi.org/10.1016/j.mad.2015.06.003

Hudson, N. J., Bottje, W. G., Hawken, R. J., Kong, B., Okimoto, R., & Reverter, A. (2017). Mitochondrial metabolism: A driver of energy utilisation and product quality? *Animal Production Science*, *57*(11), 2204–2215. https://doi.org/10.1071/an17322

Jia, P. P., Her, C. T., & Chai, W. H. (2015). DNA excision repair at telomeres. *DNA Repair*, *36*, 137–145. https://doi.org/10.1016/j.dnarep.2015.09.017

Kappei, D., & Londoño-Vallejo, J. A. (2008). Telomere length inheritance and aging. *Mechanisms of Ageing and Development*, *129*, 17–26. https://doi.org/10.1016/j.mad.2007.10.009

Kawanishi, S., & Oikawa, S. (2004). Mechanism of telomere shortening by oxidative stress. *Annals of the New York Academy of Sciences*, *1019*, 278–284. https://doi.org/10.1196/annals.1297.047

Koch, R. E., Buchanan, K. L., Casagrande, S., Crino, O., Dowling, D. K., Hill, G. E., Hood, W. R., McKenzie, M., Mariette, M. M., Noble, D. W. A., Pavlova, A., Seebacher, F., Sunnucks, P., Udino, E., White, C. R., Salin, K., & Stier, A. (2021). Integrating mitochondrial aerobic metabolism into ecology and evolution. *Trends in Ecology & Evolution*, *4*, 321–332. https://doi.org/10.1016/j.tree.2020.12.006

Koch, R. E., & Hill, G. E. (2018). Behavioural mating displays depend on mitochondrial function: A potential mechanism for linking behaviour to individual condition. *Biological Reviews*, *93*, 1387–1398. https://doi.org/10.1111/brv.12400

Koch, R. E., Josefson, C. C., & Hill, G. E. (2017). Mitochondrial function, ornamentation, and immunocompetence. *Biological Reviews*, *92*(3), 1459–1474. https://doi.org/10.1111/brv.12291

Kruk, P. A., Rampino, N. J., & Bohr, V. A. (1995). DNA-damage and repair in telomeres - relation to aging. *Proceedings of the National Academy of Sciences of the United States of America*, *92*(1), 258–262. https://doi.org/10.1073/pnas.92.1.258

Kurz, D. J., Decary, S., Hong, Y., Trivier, E., Akhmedov, A., & Erusalimsky, J. D. (2004). Chronic oxidative stress compromises telomere integrity and accelerates the onset of senescence in human endothelial cells. *Journal of Cell Science*, *117*(11), 2417–2426. https://doi.org/10.1242/jcs.01097

Mailloux, R. J., & Harper, M. E. (2011). Uncoupling proteins and the control of mitochondrial reactive oxygen species production. *Free Radical Biology and Medicine*, *51*, 1106–1115. https://doi.org/10.1016/j.freeradbiomed.2011.06.022

Mailloux, R. J., McBride, S. L., & Harper, M. E. (2013). Unearthing the secrets of mitochondrial ROS and glutathione in bioenergetics. *Trends in Biochemical Sciences*, *38*, 592–602. https://doi.org/10.1016/j.tibs.2013.09.001

Marcomini, I., & Gasser, S. M. (2015). Nuclear organization in DNA end processing: Telomeres vs double-strand breaks. *DNA Repair*, *32*, 134–140. https://doi.org/10.1016/j.dnarep.2015.04.024

Markkanen, E. (2017). Not breathing is not an option: How to deal with oxidative DNA damage. *DNA Repair*, *59*, 82–105. https://doi.org/10.1016/j.dnarep.2017.09.007

Martens, D. S., Van Der Stukken, C., Derom, C., Thiery, E., Bijnens, E. M., & Nawrot, T. S. (2021). Newborn telomere length predicts later life telomere length: Tracking telomere length from birth to child- and adulthood. *EBioMedicine*, *63*, 103164. https://doi.org/10.1016/j.ebiom.2020.103164

McGraw, K. J., Cohen, A. A., Costantini, D., & Hõrak, P. (2010). The ecological significance of antioxidants and oxidative stress: A marriage between mechanistic and functional perspectives. *Functional Ecology*, *24*, 947–949. https://doi.org/10.1111/j.1365-2435.2010.01772.x

Mensà, E., Latini, S., Ramini, D., Storci, G., Bonafe, M., & Olivieri, F. (2019). The telomere world and aging: Analytical challenges and future perspectives. *Ageing Research Reviews*, *50*, 27–42. https://doi.org/10.1016/j.arr.2019.01.004

Metcalfe, N. B., & Alonso-Alvarez, C. (2010). Oxidative stress as a life-history constraint: The role of reactive oxygen species (ROS) in shaping phenotypes from conception to death. *Functional Ecology*, *24*, 984–996.

Metcalfe, N. B., & Monaghan, P. (2013). Does reproduction cause oxidative stress? An open question. *Trends in Ecology & Evolution*, *28*, 347–351. https://doi.org/10.1016/j.tree.2013.01.015

Monaghan, P., Metcalfe, N. B., & Torres, R. (2009). Oxidative stress as a mediator of life history trade-offs: Mechanisms, measurements and interpretation. *Ecology Letters*, *12*, 75–92. https://doi.org/10.1111/j.1461-0248.2008.01258.x

Monternier, P. A., Marmillot, V., Rouanet, J. L., & Roussel, D. (2014). Mitochondrial phenotypic flexibility enhances energy savings

during winter fast in king penguin chicks. *Journal of Experimental Biology*, *217*(15), 2691–2697. https://doi.org/10.1242/jeb.104505

Monternier, P.-A., Teulier, L., Drai, J., Bourguignon, A., Collin-Chavagnac, D., Hervant, F., Rouanet, J.-L., & Roussel, D. (2017). Mitochondrial oxidative phosphorylation efficiency is upregulated during fasting in two major oxidative tissues of ducklings. *Comparative Biochemistry and Physiology a-Molecular & Integrative Physiology*, *212*, 1–8. https://doi.org/10.1016/j.cbpa.2017.06.018

Mookerjee, S. A., Divakaruni, A. S., Jastroch, M., & Brand, M. D. (2010). Mitochondrial uncoupling and lifespan. *Mechanisms of Ageing and Development*, *131*, 463–472. https://doi.org/10.1016/j.mad.2010.03.010

Mowry, A. V., Donoviel, Z. S., Kavazis, A. N., & Hood, W. R. (2017). Mitochondrial function and bioenergetic trade-offs during lactation in the house mouse (*Mus musculus*). *Ecology and Evolution*, *7*(9), 2994–3005. https://doi.org/10.1002/ece3.2817

Murphy, M. P. (2009). How mitochondria produce reactive oxygen species. *Biochemical Journal*, *417*, 1–13. https://doi.org/10.1042/BJ20081386

Murphy, M. P., Holmgren, A., Larsson, N.-G., Halliwell, B., Chang, C. J., Kalyanaraman, B., Rhee, S. G., Thornalley, P. J., Partridge, L., Gems, D., Nyström, T., Belousov, V., Schumacker, P. T., & Winterbourn, C. C. (2011). Unraveling the biological roles of reactive oxygen species. *Cell Metabolism*, *13*, 361–366. https://doi.org/10.1016/j.cmet.2011.03.010

Nord, A., Metcalfe, N. B., Page, J. L., Huxtable, A., McCafferty, D. J., & Dawson, N. J. (2021). Avian red blood cell mitochondria produce more heat in winter than in autumn. *FASEB Journal*, *35*, e21490. https://doi.org/10.1096/fj.202100107R

Oikawa, S., & Kawanishi, S. (1999). Site-specific DNA damage at GGG sequence by oxidative stress may accelerate telomere shortening. *FEBS Letters*, *453*(3), 365–368. https://doi.org/10.1016/s0014-5793(99)00748-6

Olsson, M., Friesen, C. R., Rollings, N., Sudyka, J., Lindsay, W., Whittington, C. M., & Wilson, M. (2018). Long-term effects of superoxide and DNA repair on lizard telomeres. *Molecular Ecology*, *27*(24), 5154–5164. https://doi.org/10.1111/mec.14913

Olsson, M., Healey, M., Perrin, C., Wilson, M., & Tobler, M. (2012). Sex-specific SOD levels and DNA damage in painted dragon lizards (*Ctenophorus pictus*). *Oecologia*, *170*, 917–924. https://doi.org/10.1007/s00442-012-2383-z

Olsson, M., Tobler, M., Healey, M., Perrin, C., & Wilson, M. (2012). A significant component of ageing (DNA damage) is reflected in fading breeding colors: An experimental test using innate antioxidant mimetics in painted dragon lizards. *Evolution*, *66*(8), 2475–2483. https://doi.org/10.1111/j.1558-5646.2012.01617.x

Olsson, M., Wapstra, E., & Friesen, C. (2018). Ectothermic telomeres: It's time they came in from the cold. *Philosophical Transactions of the Royal Society B-Biological Sciences*, *373*(1741), 20160449. https://doi.org/10.1098/rstb.2016.0449

Pamplona, R., & Costantini, D. (2011). Molecular and structural antioxidant defenses against oxidative stress in animals. *American Journal of Physiology-Regulatory Integrative and Comparative Physiology*, *301*(4), R843–R863. https://doi.org/10.1152/ajpregu.00034.2011

Pańczyszyn, A., Boniewska-Bernacka, E., & Goc, A. (2020). The role of telomeres and telomerase in the senescence of postmitotic cells. *DNA Repair*, *95*, 102956. https://doi.org/10.1016/j.dnarep.2020.102956

Poetsch, A. R. (2020). The genomics of oxidative DNA damage, repair, and resulting mutagenesis. *Computational and Structural Biotechnology Journal*, *18*, 207–219. https://doi.org/10.1016/j.csbj.2019.12.013

Reichert, S., & Stier, A. (2017). Does oxidative stress shorten telomeres in vivo? A Review. *Biology Letters*, *13*(12), 20170463. https://doi.org/10.1098/rsbl.2017.0463

Rey, B., Roussel, D., Romestaing, C., Belouze, M., Rouanet, J.-L., Desplanches, D., Sibille, B., Servais, S., & Duchamp, C. (2010). Up-regulation of avian uncoupling protein in cold-acclimated and hyperthyroid ducklings prevents reactive oxygen species production by skeletal muscle mitochondria. *BMC Physiology*, *10*, 5. https://doi.org/10.1186/1472-6793-10-5

Reznick, D., Nunney, L., & Tessier, A. (2000). Big houses, big cars, superfleas, and the costs of reproduction. *Trends in Ecology & Evolution*, *15*, 421–425. https://doi.org/10.1016/S0169-5347(00)01941-8

Roussel, D., Boel, M., Mortz, M., Romestaing, C., Duchamp, C., & Voituron, Y. (2019). Threshold effect in the H_2O_2 production of skeletal muscle mitochondria during fasting and refeeding. *Journal of Experimental Biology*, *222*(4), UNSP jeb196188. https://doi.org/10.1242/jeb.196188

Roussel, D., Boel, M., & Romestaing, C. (2018). Fasting enhances mitochondrial efficiency in duckling skeletal muscle by acting on the substrate oxidation system. *Journal of Experimental Biology*, *221*(4), UNSP jeb172213. https://doi.org/10.1242/jeb.172213

Roussel, D., Marmillot, V., Monternier, P. A., Bourguignon, A., Toullec, G., Romestaing, C., & Duchamp, C. (2020). Skeletal muscle metabolism in sea-acclimatized king penguins. II. Improved efficiency of mitochondrial bioenergetics. *Journal of Experimental Biology*, *223*, jeb.233684

Roussel, D., & Voituron, Y. (2020). Mitochondrial costs of being hot: Effects of acute thermal change on liver bioenergetics in toads (*Bufo bufo*). *Frontiers in Physiology*, *11*, 153. https://doi.org/10.3389/fphys.2020.00153

Saint-Leandre, B., & Levine, M. T. (2020). The Telomere paradox: Stable genome preservation with rapidly evolving proteins. *Trends in Genetics*, *36*(4), 232–242. https://doi.org/10.1016/j.tig.2020.01.007

Salin, K., Auer, S. K., Rey, B., Selman, C., & Metcalfe, N. B. (2015). Variation in the link between oxygen consumption and ATP production, and its relevance for animal performance. *Proceedings of the Royal Society B-Biological Sciences*, *282*(1812), 20151028. https://doi.org/10.1098/rspb.2015.1028

Salin, K., Auer, S. K., Rudolf, A. M., Anderson, G. J., Cairns, A. G., Mullen, W., & Metcalfe, N. B. (2015). Individuals with higher metabolic rates have lower levels of reactive oxygen species in vivo. *Biology Letters*, *11*(9), 20150538. https://doi.org/10.1098/rsbl.2015.0538

Salin, K., Luquet, E., Rey, B., Roussel, D., & Voituron, Y. (2012). Alteration of mitochondrial efficiency affects oxidative balance, development and growth in frog (*Rana temporaria*) tadpoles. *Journal of Experimental Biology*, *215*, 863–869.

Salin, K., Villasevil, E. M., Anderson, G. J., Auer, S. K., Selman, C., Hartley, R. C., Mullen, W., Chinopoulos, C., & Metcalfe, N. B. (2018). Decreased mitochondrial metabolic requirements in fasting animals carry an oxidative cost. *Functional Ecology*, *32*, 2149–2157. https://doi.org/10.1111/1365-2435.13125

Salin, K., Villasevil, E. M., Anderson, G. J., Lamarre, S. G., Melanson, C. A., McCarthy, I., Selman, C., & Metcalfe, N. B. (2019). Differences in mitochondrial efficiency explain individual variation in growth performance. *Proceedings of the Royal Society B-Biological Sciences*, *286*(1909), 20191466. https://doi.org/10.1098/rspb.2019.1466

Salin, K., Villasevil, E. M., Anderson, G. J., Selman, C., Chinopoulos, C., & Metcalfe, N. B. (2018). The RCR and ATP/O indices can give contradictory messages about mitochondrial efficiency. *Integrative and Comparative Biology*, *58*, 486–494. https://doi.org/10.1093/icb/icy085

Salin, K., Villasevil, E. M., Auer, S. K., Anderson, G. J., Selman, C., Metcalfe, N. B., & Chinopoulos, C. (2016). Simultaneous measurement of mitochondrial respiration and ATP production in tissue homogenates and calculation of effective P/O ratios. *Physiological Reports*, *4*(20), e13007. https://doi.org/10.14814/phy2.13007

Schieber, M., & Chandel, N. S. (2014). ROS function in redox signaling and oxidative stress. *Current Biology*, *24*(10), R453–R462. https://doi.org/10.1016/j.cub.2014.03.034

Seifried, H. E., Anderson, D. E., Fisher, E. I., & Milner, J. A. (2007). A review of the interaction among dietary antioxidants and reactive

oxygen species. *Journal of Nutritional Biochemistry*, *18*(9), 567–579. https://doi.org/10.1016/j.jnutbio.2006.10.007

Shama, L. N. S., Mark, F. C., Strobel, A., Lokmer, A., John, U., & Wegner, K. M. (2016). Transgenerational effects persist down the maternal line in marine sticklebacks: Gene expression matches physiology in a warming ocean. *Evolutionary Applications*, *9*(9), 1096–1111. https://doi.org/10.1111/eva.12370

Shigenaga, M. K., Hagen, T. M., & Ames, B. N. (1994). Oxidative damage and mitochondrial decay in aging. *Proceedings of the National Academy of Sciences of the United States of America*, *91*(23), 10771–10778. https://doi.org/10.1073/pnas.91.23.10771

Silva, B. A., Jones, T. J., & Murnane, J. P. (2017). Differences in the recruitment of DNA repair proteins at subtelomeric and interstitial I-SceI endonuclease-induced DNA double-strand breaks. *DNA Repair*, *49*, 1–8. https://doi.org/10.1016/j.dnarep.2016.10.008

Sohal, R. S., & Weindruch, R. (1996). Oxidative stress, caloric restriction, and aging. *Science*, *273*(5271), 59–63.

Speakman, J. R., Talbot, D. A., Selman, C., Snart, S., McLaren, J. S., Redman, P., Krol, E., Jackson, D. M., Johnson, M. S., & Brand, M. D. (2004). Uncoupled and surviving: Individual mice with high metabolism have greater mitochondrial uncoupling and live longer. *Aging Cell*, *3*(3), 87–95. https://doi.org/10.1111/j.1474-9728.2004.00097.x

Stier, A., Bize, P., Hsu, B. Y., & Ruuskanen, S. (2019). Plastic but repeatable: Rapid adjustments of mitochondrial function and density during reproduction in a wild bird species. *Biology Letters*, *15*(11), 20190536. https://doi.org/10.1098/rsbl.2019.0536

Stier, A., Romestaing, C., Schull, Q., Lefol, E., Robin, J. P., Roussel, D., & Bize, P. (2017). How to measure mitochondrial function in birds using red blood cells: A case study in the king penguin and perspectives in ecology and evolution. *Methods in Ecology and Evolution*, *8*(10), 1172–1182. https://doi.org/10.1111/2041-210x.12724

Tanaka, M., Gong, J. S., Zhang, J., Yamada, Y., Borgeld, H. J., & Yagi, K. (2000). Mitochondrial genotype associated with longevity and its inhibitory effect on mutagenesis. *Mechanisms of Ageing and Development*, *116*(2–3), 65–76. https://doi.org/10.1016/s0047-6374(00)00149-4

Tanaka, M., Gong, J. S., Zhang, J., Yoneda, M., & Yagi, K. (1998). Mitochondrial genotype associated with longevity. *Lancet*, *351*(9097), 185–186. https://doi.org/10.1016/s0140-6736(05)78211-8

Toime, L. J., & Brand, M. D. (2010). Uncoupling protein-3 lowers reactive oxygen species production in isolated mitochondria. *Free Radical Biology and Medicine*, *49*(4), 606–611. https://doi.org/10.1016/j.freeradbiomed.2010.05.010

von Zglinicki, T. (2002). Oxidative stress shortens telomeres. *Trends in Biochemical Sciences*, *27*(7), 339–344. https://doi.org/10.1016/S0968-0004(02)02110-2

Webb, C. J., Wu, Y., & Zakian, V. A. (2013). DNA repair at telomeres: Keeping the ends intact. *Cold Spring Harbor Perspectives in Biology*, *5*(6), a012666. https://doi.org/10.1101/cshperspect.a012666

Wojtczyk-Miaskowska, A., & Schlichtholz, B. (2018). DNA damage and oxidative stress in long-lived aquatic organisms. *DNA Repair*, *69*, 14–23. https://doi.org/10.1016/j.dnarep.2018.07.003

Woyda-Ploszczyca, A. M., & Jarmuszkiewicz, W. (2017). The conserved regulation of mitochondrial uncoupling proteins: From unicellular eukaryotes to mammals. *Biochimica Et Biophysica Acta-Bioenergetics*, *1858*(1), 21–33. https://doi.org/10.1016/j.bbabio.2016.10.003

Zane, A. C., Reiter, D. A., Shardell, M., Cameron, D., Simonsick, E. M., Fishbein, K. W., Studenski, S. A., Spencer, R. G., & Ferrucci, L. (2017). Muscle strength mediates the relationship between mitochondrial energetics and walking performance. *Aging Cell*, *16*(3), 461–468. https://doi.org/10.1111/acel.12568

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