Biochemical Society Transactions (2019)
https://doi.org/10.1042/BST20190105

PORTLAND PRESS

Review Article

Long-lived animals with negligible senescence: clues for ageing research

Peter Stenvinkel[1] **and Paul G. Shiels**[2]

[1]Division of Renal Medicine, Department of Clinical Science, Intervention and Technology, Karolinska Institutet, Campus Flemingsberg, Stockholm, Sweden; [2]Wolfson Wohl Translational Research Centre, University of Glasgow, Garscube Estate, Switchback Road, Bearsden, Glasgow G61 1QH, U.K.

Correspondence: Peter Stenvinkel (peter.stenvinkel@ki.se)

Among several theories to explain the complicated process of human ageing, the mitochondrial oxidative stress hypothesis has received recent attention. Considering that lifespan and ageing rates vary considerably across taxa, a better understanding of factors that lead to negligible or extremely rapid senescence in mammals may generate novel approaches to target human ageing. Several species, such as naked mole rats, ocean quahog, rockfish and Greenland shark, have been identified that exhibit negligible senescence and superior resistance to age-related diseases. Considering that the available literature suggests that their outstanding stress resistance is linked to maintenance of protein homeostasis and robust mitochondrial functions, treatments that target protein modification and upregulation of matrix antioxidants may have implications for extending human health span.

Introduction

The principles of ageing and an inescapable advancement towards death have always captivated the interest of mankind. Today the hunt for the 'fountain of youth' has been replaced by the search for geroprotectors; i.e. pharmacological or nutraceutical compounds that prevent ageing [1]. Though life and health spans have been prolonged in many experimental animals, it is arduous to conduct studies to test if these regimens also prolong life and health span in humans [2]. As the main cause of accelerated mammalian ageing is energy excess, caloric restriction without malnutrition is currently the most effective non-genetic intervention to delay ageing phenotypes in a wide range of mammals from flies [3] to primates [4]. A problem with this strategy is that caloric restriction may have adverse effects [4] and reduce the quality of life in humans. An alternative approach to arrest ageing would be to use drugs that have caloric restriction mimetic effects [5]. Sirtuins are believed to mediate the prolongevity effect of calorie restriction; thus, sirtuin activators have attracted interest within gerontology. As the human ageing process is slow and gradual, clinical trials of anti-senescence drugs [6], nuclear factor erythroid 2-related factor 2 (NRF2)-agonists [7], NAD-dependent deacetylase sirtuin-1 (SIRT1) agonists [8] and/or dietary interventions that prolong lifespan in animals, such as caloric [4] and methionine [9] restriction, would need to be extremely long.

Considering that the global number of individuals >80 years old will triple by 2050, ageing is a considerable challenge for biomedical research that will have a dramatic influence on human morbidity and mortality [10]. Diseases of ageing are influenced by mechanisms that show strong evolutionary conservation and ageing is the main risk factor for a number of 'burden of life style' diseases, such as cancer, cardiovascular disease (CVD), obesity, type-2 diabetes, osteoporosis, Alzheimer and chronic kidney disease (CKD), that have become exceedingly prevalent in the modern society [7]. Detailed studies of relatively large groups of patients displaying features of accelerated ageing, such as CKD, may be an opportunity to learn more about slow ageing processes [11]. The majority of chronic diseases that accumulate with age share common underpinning mechanisms, such as low-grade inflammation, oxidative stress, mitochondrial dysfunction, metabolic imbalances and senescence [12]. The

Received: 3 June 2019
Revised: 9 July 2019
Accepted: 10 July 2019

Version of Record published:
31 July 2019

© 2019 The Author(s). Published by Portland Press Limited on behalf of the Biochemical Society

Biochemical Society Transactions (2019)
https://doi.org/10.1042/BST20190105

role of cellular senescence in diseases that accumulate with age is a growing topic in gerontology; cellular senescence being integral to the physiology of ageing, longevity and diseases that accumulate with age, either by protein–protein interactions, allocation of common genes or prevailing signalling pathways [13]. A major problem in modern health care is the growing gap between health span and lifespan; a gap that has major economic and societal implications. Thus, rather than focus on increasing lifespan, gerontology research should aim to increase health span. Strategies that target central ageing structures in the whole cluster of 'burden of life style' diseases would likely reduce the gap between health span and lifespan [7].

Studies of longevity in the animal kingdom—could it provide clues for human ageing?

Studies of long-lived animals that stay healthy and capable of reproduction until death (i.e. long health span) could provide clues on how to target central human ageing processes [7]. Although the lifespan trajectories in different model organisms depict remarkable similarities in the ageing process with humans [14], many wild animals may not have the time to develop features of senescence, as they die of disease, are predated or starve before they reach sufficiently advanced years [15,16]. In order to activate extra resources for survival during stressful periods of the animals life, such as hibernation, it has been speculated that ageing programmes are dampened during such periods [17].

One of the biggest challenges in gerontology is to identify changes in cells that are related to senescence (cell cycle arrest with loss of a cells ability of division and growth), instead of just chronology [18]. Senescence has been considered trivial in wild animals. However, current evidence implies that it is widespread in natural populations of mammals and birds, affecting survival and reproduction of older individuals. A problem with existing data on longevity in animals is that they often consist of maximum values for lifespan (usually single observations) rather than sound and statistically robust estimates of their mean longevity [19]. Delineating the physiological and biochemical causes of ageing process in animals is a developing area of research for several reasons. These include the possible assets for human well-being, the relation of ageing to growth and reproduction and the responses of organisms to natural conditions and stress.

From our archaic past, when our common nematode-like ancestors aged exceptionally rapidly, mutations have expanded human lifespan more than 2000-fold [20]. Given that protracted longevity in animals is associated with augmented stress resistance, longevity requires uninterrupted and large-scale alterations of organs to suspend senescence. Studies of animals with particularly long lifespans would be an approach to better understand mechanisms linked to superior functionality and learn about ageing processes and find central targets for extending health span [21]. A recent study that included a variety of birds and mammals with different lifespans and body sizes showed that telomere shortening rate predicts lifespan in animals [22]. In the following, we discuss what we can learn from animal species that during evolution have adapted systems that render them extraordinary long-lived for their size (Figure 1).

Naked mole rats

Among long-lived animals, none has received as much recent attention as the naked mole rat (*Heterocephalus glaber*), a native of Eastern Africa that has emerged as a model organism for studies of ageing-related diseases and negligible senescence [23]. In contrast to other mammals, naked mole rats do not conform to the Gompertzian law of age-related mortality risk [24]; i.e. their risk of death does not increase with age. During evolution, these rodents have adapted to severe constant hypoxia in underground burrows with limited gas exchange. They can survive up to 18 min of anoxia without apparent injury, due to fructose-driven glycolysis [25]. It has been reported that hypoxia-inducible factors (HIF)-1α activation is another important mechanism for their remarkable hypoxia adaption [26]. Naked mole rats live >5 times longer than similarly sized rodents and >9 times longer than laboratory rats and show sustained good health during their lifetime. Notably, naked mole rats are subjected to high levels of oxidative stress and show increased levels of oxidative damage and less robust repair mechanisms than their short-lived rodent counter-parts, but still maintain a superior health span; this is not consistent with the free radical theory of ageing [27]. Beside superior anoxia tolerance, they are resistant to age-related diseases, such as neurodegeneration, CVD and cancer [21]. Out of 1000's of necropsies only two cases of cancer have been reported in naked mole rats [28]. In addition, naked mole rats experience no changes in body composition, or decline in genomic and proteomic integrity due to elevated proteasome quality control mechanisms [29]. Their diet (well-off in tubers and roots) provide not only a rich source of

© 2019 The Author(s). Published by Portland Press Limited on behalf of the Biochemical Society

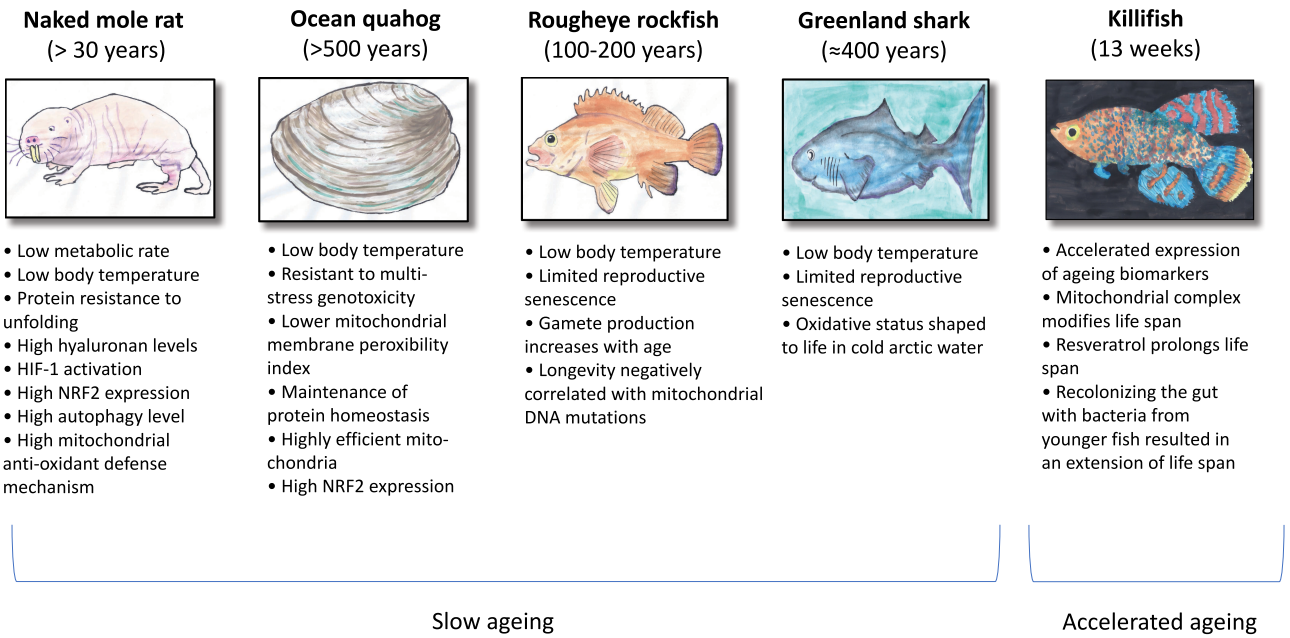


Figure 1. Animals of interest in ageing research.

Reported characteristics of four long-lived animals (naked mole rat, ocean quahog, rougheye rockfish, Greenland shark) with negligible senescence and one animal showing accelerated ageing (turquoise killifish).

anti-inflammatory and anti-oxidative polyphenols, but is also restricted in calories and methionine. It is likely that their diet in combination with low body temperature in a hypoxic and hypercapnic milieu has resulted in a series of metabolic adaptions that contribute to their exceptional resilience against age-associated diseases. Efficient protein stability and integrity with less urea-induced protein unfolding, less protein ubiquitination and higher proteasome activity [30], high levels of autophagy [31], high hyaluronan skin expression [32], a superior capacity for mitochondria to consume H_2O_2 [33] and overexpression of NRF2 [23] are reported factors liable for maintaining superior damage control mechanisms (Figure 1).

Secrets of longevity found in oceans

Fish (the most diverse class of vertebrates with >20 000 existing species) provide an enormous spectrum of longevity that could be used for gerontology research. Although some marine animals, such as sea turtles, sharks and sturgeons are reported to have a long lifespan, they have not been extensively used for studies of ageing. In contrast to the common contention that turtles (*Chrysemys picta*) do not age, data spanning >20 years from a wild population of turtles has shown that survival declines among adults [34]. Fish exhibit three types of senescence; rapid (such as eels, killi fish and pacific salmon), gradual (such as guppy and platyfish) and negligible (such as sturgeons and rockfish). Given recent data indicating that senolytic drugs, such as quercetin and navitoclax, may be an option to arrest human ageing processes [35] comparative studies of fish with rapid or negligible senescence would be of interest [36]. Whereas lower environmental temperature and dietary restriction retards the ageing process in species showing gradual senescence, the reason(s) why only some species of fish exhibit negligible senescence are undisclosed [36]. The existence of fish with negligible senescence allow extensive studies of cellular, genetic and biochemical characteristics of slow ageing. It has been reported that muscle extracts from fish with negligible senescence, such as sturgeon, have protective effects on senescence induced by oxidative stress [37]. It would also be interested to find out if long-lived fish exhibit an enhanced capacity to repair DNA damage, protect protein function or exhibit a continued turnover of tissue. The rockfish is a long-lived fish with a documented age range of 100–200 years, that has attracted interest as a model system due to intermediate growth (i.e. it continues to increase in body size throughout life) and limited reproductive senescence. In contrast to other mammals, gamete production in female rockfish increases with age and size fecundity is positively correlated with age [38]. Among long-lived fish, sharks have received recent attention since

© 2019 The Author(s). Published by Portland Press Limited on behalf of the Biochemical Society

Biochemical Society Transactions (2019)
https://doi.org/10.1042/BST20190105

they live much longer than realized previously [39]. Radiocarbon studies of the eye lens in the Greenland shark (*Somniosus microcephalus*) show that they may reach an age of almost 400 years; i.e. they are the longest-lived vertebrate documented [40] and surpass the maximum age of >200 years reported in bowhead whales (*Balaena mysticetus*) [41].

The most long-lived marine species are bivalve molluscs, such as the ocean quahog, which provide the widest range of lifespans available for interspecies comparisons. The maximal lifespan in different populations of bivalve molluscs (*Spisula solidissima vs. Arctica Icelandica*) ranges between 36 to >500 years (age is counted by growth rings on the shell) despite the similar size and living conditions [42]. Thus, *Arctica Icelandica* is the longest-lived non-colonial metazoan known on the planet. As bivalves accommodate a protracted sphere of environmental conditions and express markedly contrasting metabolic phenotypes they constitute a well-founded model to study ageing processes [42]. Their exceptional longevity appears independent of telomere length regulation [43]. Moreover, whereas their longevity-related differences in peroxidation index seem resilient to environmental conditions, such as temperature, season, nutrition and age [44] they are resistant to multi-stress genotoxicity [45]. To elucidate the underpinning features of successful cardiac ageing, age-related changes in proteosome activity, inflammatory markers, heat shock proteins and mitochondrial complexes were analysed in the heart of *A. Icelandica* [46]. The results showed that maintenance of protein homeostasis preserves cardiac function and that levels of oxidatively damaged proteins did not change over the first 120 years of their life [46]. Treaster et al. [47] have also emphasized the relationship between proteostasis and longevity through estimation of function, unfolding and aggregation. As inhibition of Hsp90 protects from atherosclerosis via cytoprotective mechanisms that are NRF2-dependent [48] and SKN-1/NRF2 is essential for the health span gains provided by metformin treatment in *C. elegans* [49], a role for NRF2 as a pro-longevity signalling pathway across phyla is supported. Indeed, a transcriptomics-based screening recently identified that inhibition of Hsp90 deferred ageing by augmented protein homeostasis in *C. elegans* [1].

Free radical and mitochondrial theories of ageing

According to the free radical theory of ageing (reactive oxygen species [ROS] arise as bi-products of metabolism), long-lived species should be more resistant to oxidative damage and senescence than short-lived species. However, as research using 18 different genetic manipulations of the antioxidant defense system showed no effect on lifespan in mice (except deletion of the *Sod1* gene), the evidence argues against the hypothesis that oxidative damage plays a dominant role in ageing [50]. Data from animal species other than mice have shown equivocal results. It was reported that augmented resistance to oxidative stress and maintenance of protein homeostasis preserves cardiac function in *A. Icelandica* [46] and may be a prerequisite for their extreme longevity [51]. When differences in markers of oxidative stress were compared in mollusc species with a six-fold difference in lifespan, Gruber et al. [51] reported correlations between DNA oxidation and biological, but not chronological age, implying that nucleic acid oxidation reflected intrinsic ageing mechanisms. On the other hand, since surrogate markers of oxidative damage in red blood cells and muscle did not correlate to lifespan in Greenland sharks, the free radical theory of ageing do not apply to all long-lived species [52]. In the case of the Greenland shark, the oxidative status may rather be shaped to the life in cold and deep artic water [52].

It has been suggested that the cumulative effect of biochemical damage in mitochondria causing mitochondrial mutations plays a key role in many chronic diseases associated with premature ageing, such as CKD, type-2 diabetes and neurodegenerative diseases. The transcriptional coactivator peroxisome proliferator-activated receptor gamma co-activator 1 (PGC1α), the master regulator of mitochondrial biogenesis, allow animals to conform their energy metabolism to their current state of nutrition and their environment. It is likely that the evolution of differences in the distribution and density of mitochondria play a major role for longevity, aerobic capacity and protection in extreme conditions. In the animal kingdom, the levels of mitochondrial enzymes vary more than 100-fold, from fish white muscle to the flight muscles of birds [53]. The Atlantic bluefin tuna (*Thunnus thynnus*) is a migratory fish with a remarkable swimming endurance and high metabolic demands, which stands out from other fish because they possess red muscle (due to an exceptionally high content of mitochondria. In a recent study, Dumesic et al. [54] demonstrated that the upstream open reading frame (uORF), which slows down the production of PGC1α, was completely absent in the Atlantic blue fish tuna. Their finding indicates that disruption of uORF may be an important evolutionary adaption in the animal kingdom that allows high metabolic demand.

If the rate of mitochondrial mutation is negatively correlated with longevity, the accumulation of mitochondrial mutations should associate with the degree of senescence [13]. Indeed, Blier et al. [42] reported lower

© 2019 The Author(s). Published by Portland Press Limited on behalf of the Biochemical Society

ROS production and a lower mitochondrial membrane peroxibility index in the long-lived *A. Icelandica,* supporting the concept that mitochondrial dysfunction-mediated dysregulation of molecular and metabolic pathways boosts senescence. Considering that *A. Icelandica* produces markedly lower rates of H_2O_2 compared to short-lived species and carries mitochondria that are remarkably efficient in energy transduction, extreme longevity seems to be associated with robust and efficient mitochondria that produce low levels of H_2O_2 and ROS [42]. Given the reported links between mitochondrial dysfunction and cellular senescence [55], and recent data showing that NRF2 activation plays a role in mitochondrial quality control [56], it is not surprising that the NRF2 signalling pathway plays an important role in the fine-tuning of gene expression in bivalve molluscs [57]. In accordance, a superior antioxidant mitochondrial defence system [33] together with overexpression of NRF2 [23] characterizes the exceptionally long-lived naked mole rat. The evolvement of kelch-like ECH-associated protein 1 (KEAP1) provided mammals with a more precise way to regulate NRF2 activity [58]. Considering that the lifespan of rockfish (*Sebastes*) is negatively correlated with the rate of mitochondrial DNA mutations [38], this further support the concept that mitochondria play a main role for longevity in the animal kingdom. Data in birds and mammals corroborate these findings and show an inverse relationship between longevity and evolutionary rate of mitochondrial proteins [59]. A comparative study in 21 bird species showed that red blood cells from long-lived birds produced less mitochondrial ROS than red blood cells of short-lived birds [60]. However, although the majority of studies report that a reduced lifespan in mammals is related to an increase in somatic mitochondrial mutations [61,62], Vermulst et al. [63] reported that mitochondrial point mutations do not limit the natural lifespan of mice. In accordance, a study in *C. elegans* reported that mitochondrial DNA damage does not control lifespan [64].

What can we learn from species with accelerated aging?

A better understanding of senescence and life history strategies of remarkable short-lived species could also provide valuable clues about factors which favoured the selection of species with a reduced lifespan. Among longevity minimalists, mayflies (*Ephemeroptera*) are exceptional, since these rapidly senescencing organisms have a lifespan of only 1–2 days [65]. For the purpose of screening the potential of pharmaceutical agents with senescence-retarding effects, the extraordinary short-lived killifish (*Notobranchius furzeri*), living in shallow transitory pools on the Savannah in south-eastern Africa, is an interesting vertebrate in ageing research (Figure 1). This seasonal turquoise fish is the shortest-lived vertebrate that can be cultured in captivity and has a maximal lifespan of about 13 weeks. Their early sexual maturation is tied with accelerated development and expression of ageing biomarkers at a cellular level [66]. Since killifish significantly resemble mammals (including humans) with respect to the molecular and cellular traits of ageing, they are of definite interest to gerontology research [67]. It is notable that killifish do not display a significant age-related decline in reproductive functions during their extremely short lifespan [68]. Furthermore, considering their documented high degree of Klotho gene conservation killifish may serve as a model system for studies of molecular mechanisms and pathways that determine the anti-ageing effects of klotho [69]. Baumgart et al. [70] performed a longitudinal study in killifish correlating variations in gene expression and found that mitochondrial respiratory chain complex 1 served as a hub for a group of genes whose expression correlated negatively with lifespan. This observation further strengthens the hypothesis that the mitochondrion is central to ageing processes and is in keeping with the original mitochondrial, telomere ribosome biogenesis (MTR) postulate of Shiels & Davies [71]. This espouses that interplay between these three intracellular features governs intracellular energy availability and, thus, determines the capacity of a cell for proteosynthesis in response to macromolecular damage and/or redox stress. Taken together, killifish seems to be a valid vertebrate model to identify pharmacological modulators and protein targets associated with aging. Indeed, supplementation with a sirtuin agonist (resveratrol) prolonged lifespan and retarded the expression of age-dependent attributes in killifish [66]. The observation that recolonizing the gut of middle-age killifish with bacteria from younger killifish resulted in an extension of lifespan [72] support links between the gut microbiome and longevity [73].

Perspectives

- Ageing is a considerable challenge for biomedical research that will have a dramatic influence on human morbidity and mortality.

© 2019 The Author(s). Published by Portland Press Limited on behalf of the Biochemical Society

Biochemical Society Transactions (2019)
https://doi.org/10.1042/BST20190105

- An outstanding stress resistance is a common trait of long-lived mammals and recent literature position maintenance of protein homeostasis and robust mitochondrial function in the centre of preservation of long life in the animal kingdom (Figure 1).
- A better understanding of the biology of exceptionally long-lived animals with negligible senescence may contribute to better understanding of ageing processes and lead to novel interventions that extend human health span.

Abbreviations

CKD, chronic kidney disease; CVD, cardiovascular disease; KEAP1, kelch-like ECH-associated protein 1; NRF2, nuclear factor erythroid 2-related factor 2; ROS, reactive oxygen species; SIRT1, sirtuin-1; uORF, upstream open reading frame.

Acknowledgements

The Heart and Lung Foundation supported Peter Stenvinkel's research.

Competing Interests

The Authors declare that there are no competing interests associated with the manuscript.

References

1 Janssens, G.E., Lin, X.X., Millan-Ariño, L., Kavšek, A., Sen, I., Seinstra, R.I. et al. (2019) Transcriptomics-based screening identifies pharmacological inhibition of Hsp90 as a means to defer aging. *Cell Rep.* **27**, 467–480 https://doi.org/10.1016/j.celrep.2019.03.044
2 Kirkland, J.L. (2016) Translating the science of aging into therapeutic interventions. *Cold Spring Harb. Perspect. Med.* **6**, a025908 https://doi.org/10.1101/cshperspect.a025908
3 Lian, T., Wu, Q., Hodge, B.A., Wilson, K.A., Yu, G. and Yang, M. (2018) *Drosophila* Gut-A nexus between dietary restriction and lifespan. *Int. J. Mol. Sci.* **19**, E3810 https://doi.org/10.3390/ijms19123810
4 Pifferi, F., Terrien, J., Perret, M., Epelbaum, J., Blanc, S., Picq, J.L. et al. (2019) Promoting healthspan and lifespan with caloric restriction in primates. *Commun. Biol.* **2**, 107 https://doi.org/10.1038/s42003-019-0348-z
5 Handschin, C. (2016) Caloric restriction and exercise 'mimetics': ready for prime time? *Pharmacol. Res.* **103**, 158–166 https://doi.org/10.1016/j.phrs.2015.11.009
6 Zhu, Y., Tchkonia, T., Pirtskhalava, T., Gower, A.C., Ding, H., Giorgadze, N. et al. (2015) The Achilles' heel of senescent cells: from transcriptome to senolytic drugs. *Aging Cell* **14**, 644–658 https://doi.org/10.1111/acel.12344
7 Stenvinkel, P., Meyer, C.J., Block, G.A., Chertow, G.M. and Shiels, P.G. (2019) Understanding the role of the cytoprotective transcription factor NRF2 - Lessons from evolution, the animal kingdom and rare progeroid syndromes. *Nephrol. Dial. Transpl.*, pii: gfz120 https://doi.org/10.1093/ndt/gfz120 [Epub ahead of print]
8 Lee, S.H., Lee, J.H., Lee, H.Y. and Min, K.J. (2019) Sirtuin signaling in cellular senescence and aging. *BMB Rep.* **52**, 24–34 https://doi.org/10.5483/BMBRep.2019.52.1.290
9 Lee, B.C., Kaya, A. and Gladyshev, V.N. (2016) Methionine restriction and life-span control. *Ann. N. Y. Acad. Sci.* **1363**, 116–124 https://doi.org/10.1111/nyas.12973
10 Niccoli, T. and Partridge, L. (2012) Ageing as a risk factor for disease. *Curr. Biol.* **22**, R741–RR52 https://doi.org/10.1016/j.cub.2012.07.024
11 Kooman, J.P., Kotanko, P., Schols, A.M., Shiels, P.G. and Stenvinkel, P. (2014) Chronic kidney disease and premature ageing. *Nat. Rev. Nephrol.* **10**, 732–742 https://doi.org/10.1038/nrneph.2014.185
12 Fougère, B., Boulanger, E., Nourhashémi, F., Guyonnet, S. and Cesari, M. (2017) Chronic inflammation: accelerator of biological aging. *J. Gerontol. A Biol. Sci. Med. Sci.* **72**, 1218–1225 https://doi.org/10.1093/gerona/glw240
13 Tacutu, R., Budovsky, A., Yanai, H. and Fraifeld, V.E. (2011) Molecular links between cellular senescence, longevity and age-related diseases - a systems biology perspective. *Aging (Albany NY)* **3**, 1178–1191 https://doi.org/10.18632/aging.100413
14 Mitchell, S.J., Scheibye-Knudsen, M., Longo, D.L. and de Cabo, R. (2015) Animal models of aging research: implications for human aging and age-related diseases. *Annu. Rev. Anim. Biosci.* **3**, 283–303 https://doi.org/10.1146/annurev-animal-022114-110829
15 Finch, C.E. (2009) Update on slow aging and negligible senescence - a mini review. *Gerontology* **55**, 307–311 https://doi.org/10.1159/000215589
16 Buffenstein, R. (2008) Negligible senescence in the longest living rodent, the naked mole-rat: insights from a successfully aging species. *J. Comp. Physiol. B* **4**, 439–445 https://doi.org/10.1007/s00360-007-0237-5
17 Skulachev, M.V., Severin, F.F. and Skulachev, V.P. (2015) Aging as an evolvability-increasing program which can be switched off by organism to mobilize additional resources for survival. *Curr. Aging Sci.* **8**, 95–109 https://doi.org/10.2174/1874609808666150422122401
18 Jeyapalan, J.C. and Sedivy, J.M. (2008) Cellular senescence and organismal aging. *Mech. Ageing Dev.* **129**, 467–474 https://doi.org/10.1016/j.mad.2008.04.001

© 2019 The Author(s). Published by Portland Press Limited on behalf of the Biochemical Society

19 Che-Castaldo, J.P., Byrne, A., Perišin, K. and Faust, L.J. (2019) Sex-specific median life expectancies from ex situ populations for 330 animal species. *Sci. Data* **6**, 190019 https://doi.org/10.1038/sdata.2019.19
20 Kenyon, C.J. (2010) The genetics of aging. *Nature* **464**, 504–512 https://doi.org/10.1038/nature08980
21 Stenvinkel, P., Painer, J., Kuro-O, M., Lanaspa, M., Arnold, W., Ruf, T. et al. (2018) Novel treatment strategies for chronic kidney disease: insights from the animal kingdom. *Nat. Rev. Nephrol.* **14**, 265–284 https://doi.org/10.1038/nrneph.2017.169
22 Whittemore, K., Vera, E., Martinez-Nevado, E., Sanpera, C. and Blasco, M.A. (2019) Telomere shortening rate predicts species life span. *Proc. Natl Acad. Sci. U.S.A.*, pii: 201902452 https://doi.org/10.1073/pnas.1902452116 [Epub ahead of print]
23 Lewis, K.N., Wason, E., Edrey, Y.H., Kristan, D.M., Nevo, E. and Buffenstein, R. (2015) Regulation of Nrf2 signaling and longevity in naturally long-lived rodents. *Proc. Natl Acad. Sci. U.S.A.* **112**, 3722–3727 https://doi.org/10.1073/pnas.1417566112
24 Ruby, J.G., Smith, M. and Buffenstein, R. (2018) Naked mole-rat mortality rates defy gompertzian laws by not increasing with age. *eLife* **7**, e31157 https://doi.org/10.7554/eLife.31157
25 Park, T.J., Reznick, J., Peterson, B.L., Blass, G., Omerbasic, D., Bennett, N.C. et al. (2017) Fructose-driven glycolysis supports anoxia resistance in the naked mole-rat. *Science* **356**, 307–311 https://doi.org/10.1126/science.aab3896
26 Xiao, B., Wang, S., Yang, G., Sun, X., Zhao, S., Lin, L. et al. (2017) HIF-1α contributes to hypoxia adaptation of the naked mole rat. *Oncotarget* **8**, 109941–109951 https://doi.org/10.18632/oncotarget.22767
27 Rodriguez, K.A., Wywial, E., Perez, V.I., Lambert, A.J., Edrey, Y.H., Lewis, K.N. et al. (2011) Walking the oxidative stress tightrope: a perspective from the naked mole-rat, the longest-living rodent. *Curr. Pharm. Des.* **17**, 2290–2307 https://doi.org/10.2174/138161211797052457
28 Delaney, M.A., Ward, J.M., Walsh, T.F., Chinnadurai, S.K., Kerns, K., Kinsel, M.J. et al. (2016) Initial case reports of cancer in naked mole-rats (Heterocephalus glaber). *Vet. Pathol.* **53**, 691–696 https://doi.org/10.1177/0300985816630796
29 Lewis, K.N., Andziak, B., Yang, T. and Buffenstein, R. (2013) The naked mole-rat response to oxidative stress: just deal with it. *Antioxid. Redox Signal.* **19**, 1388–1399 https://doi.org/10.1089/ars.2012.4911
30 Pérez, V.I., Buffenstein, R., Masamsetti, V., Leonard, S., Salmon, A.B., Mele, J. et al. (2010) Protein stability and resistance to oxidative stress are determinants of longevity in the longest-living rodent, the naked mole-rat. *Proc. Natl Acad. Sci. U.S.A.* **106**, 3059–3064 https://doi.org/10.1073/pnas.0809620106
31 Zhao, S., Lin, L., Kan, G., Xu, C., Tang, Q., Yu, C. et al. (2014) High autophagy in the naked mole rat may play a significant role in maintaining good health. *Cell Physiol. Biochem.* **33**, 321–332 https://doi.org/10.1159/000356672
32 Tian, X., Azpurua, J., Hine, C., Vaidya, A., Myakishev-Rempel, M., Ablaeva, J. et al. (2013) High-molecular-mass hyaluronan mediates the cancer resistance of the naked mole rat. *Nature* **499**, 346–349 https://doi.org/10.1038/nature12234
33 Munro, D., Baldy, C., Pamenter, M.E. and Treberg, J.R. (2019) The exceptional longevity of the naked mole-rat may be explained by mitochondrial antioxidant defenses. *Aging Cell* **18**, e12916 https://doi.org/10.1111/acel.12916
34 Warner, D.A., Miller, D.A., Bronikowski, A.M. and Janzen, F.J. (2016) Decades of field data reveal that turtles senesce in the wild. *Proc. Natl Acad. Sci. U.S.A.* **113**, 6502–6507 https://doi.org/10.1073/pnas.1600035113
35 Tchkonia, T. and Kirkland, J.L. (2018) Aging, cell senescence, and chronic disease: emerging therapeutic strategies. *JAMA* **320**, 1319–1320 https://doi.org/10.1001/jama.2018.12440
36 Patnaik, B.K., Mahapatro, N. and Jena, B.S. (1994) Ageing in fishes. *Gerontology* **40**, 113–132 https://doi.org/10.1159/000213582
37 Mikhailova, M.V., Belyaeva, N.F., Kozlova, N.I., Zolotarev, K.V., Mikhailov, A.N., Berman, A.E. et al. (2017) Protective action of fish muscle extracts against cellular senescence induced by oxidative stress. *Biomed. Khim.* **63**, 351–355 https://doi.org/10.18097/pbmc20176304351
38 Hua, X., Cowman, P., Warren, D. and Bromham, L. (2015) Longevity is linked to mitochondrial mutation rates in rockfish: a test using poisson regression. *Mol. Biol. Evol.* **32**, 2633–2645 https://doi.org/10.1093/molbev/msv137
39 Cressey, D. (2017) Sharks can live a lot longer than researchers realized. *Nature* **549**, 316–317 https://doi.org/10.1038/nature.2017.22626
40 Nielsen, J., Hedeholm, R., Heinemeier, J., Bushnell, P.G., Christiansen, J.S., Olsen, J. et al. (2016) Eye lens radiocarbon reveals centuries of longevity in the Greenland shark (Somniosus microcephalus). *Science* **353**, 702–704 https://doi.org/10.1126/science.aaf1703
41 George, J.C., Bada, J., Zeh, J., Scott, L., Brown, S.E., O'Hara, T. et al. (1999) Age and growth estimates of bowhead whales (*Balaena mysticetus*) via aspartic acid racemization. *Can. J. Zool.* **77**, 571–580 https://doi.org/10.1139/z99-015
42 Blier, P.U., Abele, D., Munro, D., Degletagne, C., Rodriguez, E. and Hagen, T. (2017) What modulates animal longevity? Fast and slow aging in bivalves as a model for the study of lifespan. *Semin. Cell Dev. Biol.* **70**, 130–140 https://doi.org/10.1016/j.semcdb.2017.07.046
43 Gruber, H., Schaible, R., Ridgway, I.D., Chow, T.T., Held, C. and Philipp, E.E. (2014) Telomere-independent ageing in the longest-lived non-colonial animal, *Arctica islandica*. *Exp. Gerontol.* **51**, 38–45 https://doi.org/10.1016/j.exger.2013.12.014
44 Munro, D. and Blier, P.U. (2015) Age, diet, and season do not affect longevity-related differences in peroxidation index between *Spisula solidissima* and *Arctica islandica*. *J. Gerontol. A Biol. Sci. Med. Sci.* **70**, 434–443 https://doi.org/10.1093/gerona/glu054
45 Ungvari, Z., Sosnowska, D., Mason, J.B., Gruber, H., Lee, S.W., Schwartz, T.S. et al. (2013) Resistance to genotoxic stresses in *Arctica islandica*, the longest living noncolonial animal: is extreme longevity associated with a multistress resistance phenotype? *J. Gerontol. A Biol. Sci. Med. Sci.* **68**, 521–529 https://doi.org/10.1093/gerona/gls193
46 Sosnowska, D., Richardson, C., Sonntag, W.E., Csiszar, A., Ungvari, Z. and Ridgway, I. (2014) A heart that beats for 500 years: age-related changes in cardiac proteasome activity, oxidative protein damage and expression of heat shock proteins, inflammatory factors, and mitochondrial complexes in *Arctica islandica*, the longest-living noncolonial animal. *J. Gerontol. A Biol. Sci. Med. Sci.* **69**, 1448–1461 https://doi.org/10.1093/gerona/glt201
47 Treaster, S.B., Ridgway, I.D., Richardson, C.A., Gaspar, M.B., Chaudhuri, A.R. and Austad, S.N. (2014) Superior proteome stability in the longest lived animal. *Age (Dordr)* **36**, 9597 https://doi.org/10.1007/s11357-013-9597-9
48 Lazaro, I., Oguiza, A., Recio, C., Lopez-Sanz, L., Bernal, S., Egido, J. et al. (2017) Interplay between HSP90 and Nrf2 pathways in diabetes-associated atherosclerosis. *Clin. Investig. Arterioscler.* **29**, 51–59 https://doi.org/10.1016/j.arteri.2016.10.003
49 Onken, B. and Driscoll, M. (2010) Metformin induces a dietary restriction-like state and the oxidative stress response to extend *C. elegans* Healthspan via AMPK, LKB1, and SKN-1. *PLoS ONE* **5**, e8758 https://doi.org/10.1371/journal.pone.0008758
50 Pérez, V.I., Bokov, A., Van Remmen, H., Mele, J., Ran, Q., Ikeno, Y. et al. (2009) Is the oxidative stress theory of aging dead? *Biochim. Biophys. Acta* **1790**, 1005–1014 https://doi.org/10.1016/j.bbagen.2009.06.003

© 2019 The Author(s). Published by Portland Press Limited on behalf of the Biochemical Society

51 Gruber, H., Wessels, W., Bonyton, P., Xu, J., Wohlgemuth, S., Leeuwenburgh, C. et al. (2015) Age-related cellular changes in the long-lived bivalve *A. islandica*. *Age (Dordr)* **37**, 90 https://doi.org/10.1007/s11357-015-9831-8
52 Costantini, D., Smith, S., Killen, S.S., Nielsen, J. and Steffensen, J.F. (2017) The Greenland shark: a new challenge for the oxidative stress theory of ageing? *Comp. Biochem. Physiol. A Mol. Integr. Physiol.* **203**, 227–232 https://doi.org/10.1016/j.cbpa.2016.09.026
53 Moyers, C.D. (2003) Controlling muscle mitochondrial content. *J. Exp. Biol.* **206**, 4385–4391 https://doi.org/10.1242/jeb.00699
54 Dumesic, P.A., Egan, D.F., Gut, P., Tran, M.T., Parisi, A., Chatterjee, N. et al. (2019) An evolutionarily conserved uORF regulates PGC1α and oxidative metabolism in mice, flies, and bluefin tuna. *Cell Metab.* **30**, 190–200 https://doi.org/10.1016/j.cmet.2019.04.013
55 Habiballa, L., Salmonowicz, H. and Passos, J.F. (2019) Mitochondria and cellular senescence: implications for musculoskeletal ageing. *Free Radic. Biol. Med.* **132**, 3–10 https://doi.org/10.1016/j.freeradbiomed.2018.10.417
56 Kasai, S., Yamazaki, H., Tanji, K., Engler, M.J., Matsumiya, T. and Itoh, K. (2019) Role of the ISR-ATF4 pathway and its cross talk with Nrf2 in mitochondrial quality control. *J. Clin. Biochem. Nutr.* **64**, 1–12 https://doi.org/10.3164/jcbn.18-37
57 Wang, H., Pan, L., Si, L. and Miao, J. (2018) The role of Nrf2-Keap1 signaling pathway in the antioxidant defense response induced by PAHs in the calm *Ruditapes philippinarum*. *Fish Shellfish Immunol.* **80**, 325–334 https://doi.org/10.1016/j.fsi.2018.06.030
58 Maher, J. and Yamamoto, M. (2010) The rise of antioxidant signaling - the evolution and hormetic actions of Nrf2. *Toxicol. Appl. Pharmacol.* **244**, 4–15 https://doi.org/10.1016/j.taap.2010.01.011
59 Galtier, N., Blier, P.U. and Nabholz, B. (2009) Inverse relationship between longevity and evolutionary rate of mitochondrial proteins in mammals and birds. *Mitochondrion* **9**, 51–57 https://doi.org/10.1016/j.mito.2008.11.006
60 Delhaye, J., Salamin, N., Roulin, A., Criscuolo, F., Bize, P. and Christe, P. (2016) Interspecific correlation between red blood cell mitochondrial ROS production, cardiolipin content and longevity in birds. *Age (Dordr)* **38**, 433–443 https://doi.org/10.1007/s11357-016-9940-z
61 Trifunovic, A., Wredenberg, A., Falkenberg, M., Spelbrink, J.N., Rovio, A.T., Bruder, C.E. et al. (2004) Premature ageing in mice expressing defective mitochondrial DNA polymerase. *Nature* **429**, 417–423 https://doi.org/10.1038/nature02517
62 Yang, J.N., Seluanov, A. and Gorbunova, V. (2013) Mitochondrial inverted repeats strongly correlate with lifespan: mtDNA inversions and aging. *PLoS ONE* **8**, e73318 https://doi.org/10.1371/journal.pone.0073318
63 Vermulst, M., Bielas, J.H., Kujoth, G.C., Ladiges, W.C., Rabinovitch, P.S., Prolla, T.A. et al. (2006) Mitochondrial point mutations do not limit the natural lifespan of mice. *Nat. Genet.* **39**, 540–543 https://doi.org/10.1038/ng1988
64 Ng, L., Ng, L., van Breugel, M., Halliwell, B. and Gruber, J. (2019) Mitochondrial DNA damage does not determine *C. elegans* lifespan. *Front. Genet.* **10**, 311 https://doi.org/10.3389/fgene.2019.00311
65 Carey, J.R. (2002) Longevity minimalists: life table studies of two species of northern Michigan adult mayflies. *Exp. Geront.* **37**, 567–570 https://doi.org/10.1016/S0531-5565(01)00180-2
66 Valenzano, D.R., Terzibasi, E., Genade, T., Cattaneo, A., Domenici, L. and Cellerino, A. (2006) Resveratrol prolongs lifespan and retards the onset of age-related markers in a short-lived vertebrate. *Curr. Biol.* **16**, 296–300 https://doi.org/10.1016/j.cub.2005.12.038
67 Platzer, M. and Englert, C. (2016) Nothobranchius furzeri: a model for aging research and more. *Trends Genet.* **32**, 543–552 https://doi.org/10.1016/j.tig.2016.06.006
68 Vrtílek, M., Žák, J., Blažek, R., Polačik, M., Cellerino, A. and Reichard, M. (2018) Limited scope for reproductive senescence in wild populations of a short-lived fish. *Naturwissenschaften* **105**, 68 https://doi.org/10.1007/s00114-018-1594-5
69 Zupkovitz, G., Kabiljo, J., Martin, D., Laffer, S., Schöfer, C. and Pusch, O. (2018) Phylogenetic analysis and expression profiling of the Klotho gene family in the short-lived African killifish *Nothobranchius furzeri*. *Dev. Genes Evol.* **228**, 255–265 https://doi.org/10.1007/s00427-018-0619-6
70 Baumgart, M., Priebe, S., Groth, M., Hartmann, N., Menzel, U., Pandolfini, L. et al. (2016) Longitudinal RNA-seq analysis of vertebrate aging identifies mitochondrial complex i as a small-molecule-sensitive modifier of lifespan. *Cell Syst.* **2**, 122–132 https://doi.org/10.1016/j.cels.2016.01.014
71 Shiels, P. and Davies, R.W. (2004) Ageing and death in neurones. In *Molecular Biology of the Neuron* (Davies, R.W. and Morris, B.J., eds), pp. 435–464, Oxford University Press, UK
72 Smith, P., Willemsen, D., Popkes, M., Metge, F., Gandiwa, E., Reichard, M. et al. (2017) Regulation of life span by the gut microbiota in the short-lived African turquoise killifish. *eLife* **6**, e27014 https://doi.org/10.7554/eLife.27014
73 Maynard, C. and Weinkove, D. (2018) The gut microbiota and ageing. *Subcell Biochem.* **90**, 351–371 https://doi.org/10.1007/978-981-13-2835-0_12

© 2019 The Author(s). Published by Portland Press Limited on behalf of the Biochemical Society