

Box 1: Summary of the studies conducted on telomerase and on telomerase-related fitness links.

Telomerase may act on individual performance either via a telomere length or a non-telomere length maintenance pathway (see main text). However, when looking at the potential role of telomerase as a determinant of inter-individual differences in performances and fitness, it is important to keep in mind that telomere length regulation is complex involving extensive protein interactions. Therefore, while this review focuses exclusively on telomerase, searching for links between fitness and other telomere length regulatory components are also of particular interest, including shelterin proteins [1], DNA repairing factors (e.g. Werners helicase), non-telomerase dependent lengthening of telomeres (Alternative Lengthening of Telomeres, ALT) (reviewed in [2]), proteins that bind with telomerase [3, 4], and microRNAs [5]. It is likely that an integrated approach investigating the interaction of all the different regulatory factors of telomere length in relation to a variety of life-history traits will prove powerful for understanding the role of telomeres in determining individual fitness.

Species	Telomerase activity (TA) or gene expression (TE)	Other components of telomere length (TL) regulation [#]	Sign and nature of the relationship with physiological / fitness traits	
Mammals[§]				
Humans <i>Homo sapiens</i>	TA in embryos TA in germ and stem cells, in lymphocytes TE differential expression in centenarians		Protection against cancer Cell proliferation Lifespan? Healthy ageing?	[6, 7] [8]
Chiroptera Leaf-nosed bat <i>Hipposideros armiger</i> Leschenault's rousette <i>Rousettus leschenaultia</i>	TA in several adult tissues		Relationship with heterothermy	[9]
Ringtailed lemur <i>Lemur catta</i> Spider monkey <i>Ateles geoffroyi</i> Squirrel monkey <i>Saimiri sciureus</i> Rhesus monkey <i>Macaca mulatta</i> Orangutan <i>Pongo pygmaeus</i> Pigmy chimpanzee <i>Pan paniscus</i>	No activity detected TA detected in fibroblast cell culture after transfection with human hTERT	Possibly ALT		[10]

Dog <i>Canis lupus familiaris</i>	TA in germ cells Polymorphisms in <i>TERT</i> gene		No link with lifespan	See [11] [12]
Cat <i>Felis catus</i> Pig <i>Sus scrofa</i> Sheep <i>Ovis aries</i>	TA in germ cells			See [11]
Laboratory mice <i>mus musculus</i>	TA in testes, ovary, breast, colon and liver			[13]
<i>Mus spretus</i>	TA only in cell cultures TE (mTERT) even in telomerase negative tissues			[13]
Long-Evans rats	TA in white cells		Increased in response to stress	[14]
Laboratory mice <i>mus musculus</i>		ALT	Telomere length maintenance in early embryo development	[15]
15 rodents species	TA in somatic tissues		Inversely related to body mass No relation with lifespan Protection against cancer	[16, 17]
Leporidae European white rabbit <i>O. cuniculus</i> Black-tailed Jack rabbit <i>O. californicus</i> Swamp rabbit <i>S. aquaticus</i>	No activity detected		Protection against cancer	[18]
Ochotonidae North american pika <i>O. princeps</i>	TA in ear snips cells			
Horse <i>Equus equus</i> Donkeys <i>Equus asinus</i>	No activity detected (even in tumor) TA in testis			See [11]
Indian muntjak <i>Muntiacus muntjack</i>	No activity detected			[11, 18]
Birds				
Chicken <i>Gallus gallus</i>	TA in embryos and in reproductive and immune tissues TE in blood cells		Proliferative potential of the tissues	[19, 20]
Leach's storm petrel <i>Oceanodroma leucorhoa</i> Common tern <i>Sterna hirundo</i> Zebra finch <i>Taeniopigya guttata</i> Tree swallow <i>Tachycineta bicolor</i>	TA in several tissues at adult stage TA in several tissues at young stage		Proliferative potential of the tissues Lifespan	[21, 22]
Reptiles				
Frillneck lizard <i>Chlamydosaurus kingii</i>	TE in blood cells		Positive relationship with TL	[23]

Sic-lined racerunner <i>Cnemidophorus sexlineatus</i>	TA in several tissues		Regenerative capacity	[24]
Common wall lizard <i>Podarcis muralis</i> Carolina anole <i>Anolis carolinensis</i>	TE in tail blastoma		Regenerative capacity	[25]
Mud turtles <i>Kinosternon flavescens</i> Snapping turtle <i>Chelydra serpentina</i> Painted turtle <i>Chrysemys picta</i> Omate boc turtle <i>Terrapene ornata</i>	TA in the gonads only TA in the gonads and other tissues		Suggested relationship with regeneration and absence of apparent senescence	[26] See [11]
Amphibians				
African clawed frog <i>Xenopus leavis</i>	TA in testes, spleen, liver and embryos		A role not only related to DNA replication during embryogenesis	[27]
Crustacea				
Lobster <i>Homarus americanus</i>	TA in several tissues		A parallel in made with indeterminate growth	[28]
Green sea crab <i>Carcinus maenas</i>	TA in several tissues		Tissue regeneration Absence of age-related cancer	[29]
Fish				
Rainbow trout <i>Oncorhynchus mykiss</i> Zebrafish <i>Danio Rerio</i> Dogfish shark <i>Squalus acanthias</i> Little skate <i>Raja erinacae</i> Mackerel <i>Scomber scombrus</i> American eel <i>Anguilla rostrate</i> Killifish <i>Fundulus heteroclitus</i> Japanese medaka <i>Oryzias latipes</i> Flounder <i>Pleuronectes americanus</i>	TA in several tissues TA in several tissues	Non-canonical function of telomerase	A parallel in made with indeterminate growth Myelopoiesis. Young survival, adult fertility and lifespan determinant Tissue regeneration. No association with lifespan	[30] [31-33] [29, 34, 35]
Insects				
Isoptera Blattaria Lepidoptera Hymenoptera Trichoptera Coleoptera Sternorrhyncha	TA in head gonads or whole body protein extracts, in both adults and embryos Higher TA activity in queens of honeybees		Potential role in cell proliferation and organism development Queen determination developmental process / longevity	[36-38] [39]
Diptera Orthoptera Zygentoma Phasmida	No activity detected			[36, 40]
Echinodermata				

Red sea urchin <i>S. franciscanus</i> Green sea urchin <i>L. variagatus</i>	TA in mature eggs and adult various tissues		No link with lifespan	See [11]
Golden star tunicate <i>Botryllus schlosseri</i>	TA in germ and embryonic tissues and in adult bud rudiments		Necessary for progenitor and stem cells	[41]
Lower metazoan				
Porifera	TA in somatic and germ cells		Not obligatory for proliferative capacity	See [11]
Cnidaria	TA in gonads and ctenophore		NA	[42]
Annelids <i>Aeolosoma viride</i>	TE in blastemal		Regeneration	[43]
Planarian flatworm <i>Schmidtea mediterranea</i>	TE expression in adult stem cells		Somatic telomere maintenance during sexual reproduction	
Plants				
<i>Arabidopsis thaliana</i>	TA in callus		Growth and development	[44]
<i>Melandrium album</i>	TA in germinating seeds and roots		Growth and development	[45]

⁵ TERT gene has been cloned in platypus *Ornithorhynchus anatinus* [46]

References

- [1] de Lange, T. 2005 Shelterin: the protein complex that shapes and safeguards human telomeres. *Genes & Development* **19**, 2100-2110. (doi:10.1101/gad.1346005).
- [2] Cesare, A.J. & Reddel, R.R. 2010 Alternative lengthening of telomeres: models, mechanisms and implications. *Nature Reviews Genetics* **11**, 319-330. (doi:10.1038/nrg2763).
- [3] Forsythe, H.L., Jarvis, J.L., Turner, J.W., Elmore, L.W. & Holt, S.E. 2001 Stable Association of hsp90 and p23, but Not hsp70, with Active Human Telomerase. *Journal of Biological Chemistry* **276**, 15571-15574. (doi:10.1074/jbc.C100055200).
- [4] Kappei, D., Butter, F., Benda, C., Scheibe, M., Draškovič, I., Stevense, M., Novo, C.L., Basquin, C., Araki, M., Araki, K., et al. 2013 HOT1 is a mammalian direct telomere repeat-binding protein contributing to telomerase recruitment. *The EMBO Journal* **32**, 1681-1701. (doi:10.1038/emboj.2013.105).
- [5] Okada, M., Kim, H.W., Matsu-ura, K., Wang, Y.-G., Xu, M. & Ashraf, M. 2016 Abrogation of Age-Induced MicroRNA-195 Rejuvenates the Senescent Mesenchymal Stem Cells by Reactivating Telomerase. *Stem Cells* **34**, 148-159. (doi:10.1002/stem.2211).

- [6] Wright, W.E., Piatyszek, M.A., Rainey, W.E., Byrd, W. & Shay, J.W. 1996 Telomerase activity in human germline and embryonic tissues and cells. *Developmental Genetics* **18**, 173-179.
- [7] Cong, Y.S., Wright, W.E. & Shay, J.W. 2002 Human Telomerase and Its Regulation. *Microbiology and Molecular Biology Reviews* **66**, 407-425. (doi:10.1128/mmbr.66.3.407-425.2002).
- [8] Atzmon, G., Cho, M., Cawthon, R.M., Budagov, T., Katz, M., Yang, X., Siegel, G., Bergman, A., Huffman, D.M., Schechter, C.B., et al. 2009 Genetic variation in human telomerase is associated with telomere length in Ashkenazi centenarians. *Proceedings of the National Academy of Sciences* **107**, 1710-1717. (doi:10.1073/pnas.0906191106).
- [9] Wang, L., McAllan, B.M. & He, G. 2011 Telomerase activity in the bats *Hipposideros armiger* and *Rousettus leschenaultia*. *Biochemistry (Moscow)* **76**, 1017-1021. (doi:10.1134/s0006297911090057).
- [10] Steinert, S., White, D.M., Zou, Y., Shay, J.W. & Wright, W.E. 2002 Telomere Biology and Cellular Aging in Nonhuman Primate Cells. *Experimental Cell Research* **272**, 146-152. (doi:10.1006/excr.2001.5409).
- [11] Gomes, N.M.V., Shay, J.W. & Wright, W.E. 2010 Telomere biology in Metazoa. *FEBS Letters* **584**, 3741-3751. (doi:10.1016/j.febslet.2010.07.031).
- [12] McAloney, C.A., Silverstein, K.A., Modiano, J.F. & Bagchi, A. 2014 Polymorphisms within the Telomerase Reverse Transcriptase gene (TERT) in four breeds of dogs selected for difference in lifespan and cancer susceptibility. *BMC Veterinary Research* **10**, 20.
- [13] Prowse, K.R. & Greider, C.W. 1995 Developmental and tissue-specific regulation of mouse telomerase and telomere length. *Proceedings of the National Academy of Sciences USA* **92**, 4818-4822.
- [14] Beery, A.K., Lin, J., Biddle, J.S., Francis, D.D., Blackburn, E.H. & Epel, E.S. 2012 Chronic stress elevates telomerase activity in rats. *Biology Letters* **8**, 1063-1066. (doi:10.1098/rsbl.2012.0747).
- [15] Liu, L., Bailey, S.M., Okuka, M., Muñoz, P., Li, C., Zhou, L., Wu, C., Czerwiec, E., Sandler, L., Seyfang, A., et al. 2007 Telomere lengthening early in development. *Nature Cell Biology* **9**, 1436-1441. (doi:10.1038/ncb1664).

- [16] Gorbunova, V., Bozzella, M.J. & Seluanov, A. 2008 Rodents for comparative aging studies: from mice to beavers. *Age* **30**, 111-119. (doi:10.1007/s11357-008-9053-4).
- [17] Gorbunova, V. & Seluanov, A. 2009 Coevolution of telomerase activity and body mass in mammals: From mice to beavers. *Mechanisms of Ageing and Development* **130**, 3-9. (doi:10.1016/j.mad.2008.02.008).
- [18] Forsyth, N.R., Elder, F.F.B., Shay, J.W. & Wright, W.E. 2005 Lagomorphs (rabbits, pikas and hares) do not use telomere-directed replicative aging in vitro. *Mechanisms of Ageing and Development* **126**, 685-691. (doi:10.1016/j.mad.2005.01.003).
- [19] Delany, M.E., Krupkin, A.B. & Miller, M.M. 2000 Organization of telomere sequences in birds: evidence for arrays of extreme length and for in vivo shortening. *Cytogenetics and Cell Genetics* **90**, 139-145.
- [20] Venkatesan, R.N. & Price, C. 1998 Telomerase expression in chickens: Constitutive activity in somatic tissues and down-regulation in culture. *Proceedings of the National Academy of Sciences, USA* **95**, 14763-14768.
- [21] Haussmann, M., Winkler, D., Huntington, C., Nisbet, I. & Vleck, C. 2007 Telomerase activity is maintained throughout the lifespan of long-lived birds. *Experimental Gerontology* **42**, 610-618. (doi:10.1016/j.exger.2007.03.004).
- [22] Haussmann, M.F., Winkler, D.W., Huntington, C.E., Nisbet, I.C.T. & Vleck, C.M. 2004 Telomerase Expression Is Differentially Regulated in Birds of Differing Life Span. *Annals of the New York Academy of Sciences* **1019**, 186-190. (doi:10.1196/annals.1297.029).
- [23] Ujvari, B., Biro, P.A., Charters, J.E., Brown, G., Heasman, K., Beckmann, C. & Madsen, T. 2016 Curvilinear telomere length dynamics in a squamate reptile. *Functional Ecology*. (doi:10.1111/1365-2435.12764).
- [24] Christiansen, J.L., Henderson, E.R., Budke, B., Lynch, M., Lu, Q. & Johnson, J. 2001 A final report of studies of the Hayflick limit in Reptiles, a test of potential immortality. . *Proceedings of the Iowa Space Grant Consortium* **10**.

- [25] Alibardi, L. 2016 Immunocalization of telomerase in cells of lizard tail after amputation suggests cell activation for tail regeneration. *Tissue and Cell* **48**, 63-71. (doi:10.1016/j.tice.2015.10.004).
- [26] Christiansen, J., Johnson, J., Henderson, E.R., Budke, B. & Lynch, M. 2001 The relationship between telomeres, telomerase, reptilian lifespan, and reptilian tissue regeneration. *Proceedings of the Iowa Space Grant Consortium* **1-10**.
- [27] Mantell, L.L. & Greider, C.W. 1994 Telomerase activity in germline and embryonic cells of *Xenopus*. *The EMBO Journal* **13**, 3211-3217.
- [28] Klapper, W., Kühne, K., Singh, K.K., Heidorn, K., Parwaresch, R. & Krupp, G. 1998 Longevity of lobsters is linked to ubiquitous telomerase expression. *FEBS Letters* **439**, 143-146.
- [29] Elmore, L.W., Norris, M.W., Sircar, S., Bright, A.T., McChesney, P.A., Winn, R.N. & Holt, S.E. 2008 Upregulation of Telomerase Function During Tissue Regeneration. *Exp Biol Med* **233**, 958. (doi:10.3181/0712-RM-345).
- [30] Klapper, W., Heidorn, K., Kühne, K., Parwaresch, R. & Krupp, G. 1998 Telomerase activity in immortal fish. *FEBS Letters* **434**, 409-412.
- [31] Alcaraz-Pérez, F., Garcia-Castillo, J., Garcia-Moreno, D., Lopez-Munoz, A., Anchelin, M., Angosto, D., Zon, L.I., Muleno, V. & Cayuela, M.L. 2014 A non-canonical function of telomerase RNA in the regulation of developmental myelopoiesis in zebrafish. *Nature Communications* **5**, 3228. (doi:10.1038/ncomms4228 | www.nature.com/naturecommunications).
- [32] Anchelin, M., Alcaraz-Perez, F., Martinez, C.M., Bernabe-Garcia, M., Mulero, V. & Cayuela, M.L. 2013 Premature aging in telomerase-deficient zebrafish. *Disease Models & Mechanisms* **6**, 1101-1112. (doi:10.1242/dmm.011635).
- [33] Artandi, S.E., Henriques, C.M., Carneiro, M.C., Tenente, I.M., Jacinto, A. & Ferreira, M.G. 2013 Telomerase Is Required for Zebrafish Lifespan. *PLoS Genetics* **9**, e1003214. (doi:10.1371/journal.pgen.1003214).
- [34] Hatakeyama, H., Nakamura, K.-I., Izumiya-Shimomura, N., Ishii, A., Tsuchida, S., Takubo, K. & Ishikawa, N. 2008 The teleost *Oryzias latipes* shows telomere shortening with age despite

considerable telomerase activity throughout life. *Mechanisms of Ageing and Development* **129**, 550-557. (doi:10.1016/j.mad.2008.05.006).

[35] Bednarek, D., González-Rosa, Juan M., Guzmán-Martínez, G., Gutiérrez-Gutiérrez, Ó., Aguado, T., Sánchez-Ferrer, C., Marques, Inês J., Galardi-Castilla, M., de Diego, I., Gómez, Manuel J., et al. 2015 Telomerase Is Essential for Zebrafish Heart Regeneration. *Cell Reports* **12**, 1691-1703. (doi:10.1016/j.celrep.2015.07.064).

[36] Korandová, M., Krůček, T., Vrbová, K. & Frydrychová, R.Č. 2014 Distribution of TTAGG-specific telomerase activity in insects. *Chromosome Research* **22**, 495-503. (doi:10.1007/s10577-014-9436-6).

[37] Sasaki, T. & Kishi, S. 2013 Molecular and chemical genetic approaches to developmental origins of aging and disease in zebrafish. *Biochimica et Biophysica Acta - Molecular Basis of Disease* **1832**, 1362-1370.

[38] Gong, H., Zhu, W., Zhang, J., Li, X., Meng, Q., Zhou, G., Wang, M., Wang, H., Miao, L., Qin, Q., et al. 2015 TTAGG-repeat telomeres and characterization of telomerase in the beet armyworm, *Spodoptera exigua* (Lepidoptera: Noctuidae). *Insect Molecular Biology* **24**, 358-367. (doi:10.1111/imb.12163).

[39] Korandová, M. & Frydrychová, R.Č. 2015 Activity of telomerase and telomeric length in *Apis mellifera*. *Chromosoma* **125**, 405-411. (doi:10.1007/s00412-015-0547-4).

[40] Sasaki, T. & Fujiwara, H. 2000 Detection and distribution patterns of telomerase activity in insects. *European Journal of Biochemistry* **267**, 3025-3031.

[41] Laird, D.J. & Weissman, I.L. 2004 Telomerase maintained in self-renewing tissues during serial regeneration of the urochordate *Botryllus schlosseri*. *Developmental Biology* **273**, 185-194. (doi:10.1016/j.ydbio.2004.05.029).

[42] Chen, C.F. 2016 The Roles of Telomerase in Regeneration during Aging and the Telomeric DNA Sequence Identification in *Aeolosoma viride*. *The FASEB Journal* **30**, 1051.1052.

[43] Tan, T.C.J., Rahman, R., Jaber-Hijazi, F., Felix, D.A., Chen, C., Louis, E.J. & Aboobaker, A. 2012 Telomere maintenance and telomerase activity are differentially regulated in asexual and sexual

worms. *Proceedings of the National Academy of Sciences, USA* **109**, 4209-4214.

(doi:10.1073/pnas.1118885109).

[44] Fitzgerald, M.S., Riha, K., Gao, F., Ren, S., McKnight, T.D. & Shippen, D.E. 1999 Disruption of the telomerase catalytic subunit gene from *Arabidopsis* inactivates telomerase and leads to a slow loss of telomeric DNA. *P Natl Acad Sci USA* **96**, 14813–14818.

[45] Riha, K., Fajkus, J., Siroky, J. & Vyskot, B. 1998 Developmental Control of Telomere Lengths and Telomerase Activity in Plants. *The Plant Cell* **10**, 1691-1698.

[46] Hrdličkov, R., Nehyba, J., Lim, S.L., Grützner, F. & Bose, H.R. 2012 Insights into the evolution of mammalian telomerase: Platypus TERT shares similarities with genes of birds and other reptiles and localizes on sex chromosomes. *BMC Genomics* **13**, 216. (doi:10.1186/1471-2164-13-216).