



**HAL**  
open science

## Seasonal variation in telomere dynamics in African striped mice

Francois Criscuolo, Neville Pillay, Sandrine Zahn, Carsten Schradin

► **To cite this version:**

Francois Criscuolo, Neville Pillay, Sandrine Zahn, Carsten Schradin. Seasonal variation in telomere dynamics in African striped mice. *Oecologia*, 2020. hal-02985514

**HAL Id: hal-02985514**

**<https://hal.science/hal-02985514>**

Submitted on 2 Nov 2020

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.

## Seasonal variation in telomere dynamics in African striped mice

Francois Criscuolo<sup>1</sup>, Neville Pillay<sup>2</sup>, Sandrine Zahn<sup>1</sup> & Carsten Schradin<sup>1,2</sup>

<sup>1</sup>Université de Strasbourg, CNRS, IPHC UMR 7178, F-67000 Strasbourg, France

<sup>2</sup>School of Animal, Plant and Environmental Sciences, University of the Witwatersrand, South Africa.

Key words: telomere length, telomere lengthening, seasonal variation, age, mice.

Corresponding authors: F. Criscuolo, [francois.criscuolo@iphc.cnrs.fr](mailto:francois.criscuolo@iphc.cnrs.fr)

## **ABSTRACT**

Telomere shortening has been used as an indicator of aging and is believed to accelerate under harsh environmental conditions. This can be attributed to the fact that telomere shortening has often been regarded as non-reversible and negatively impacting fitness. However, studies of laboratory mice indicate that they may be able to repair telomere loss to recover from environmental harshness, as indicated by recent studies in hibernating rodents. We studied seasonal variation in telomere dynamics in African striped mice (*Rhabdomys pumilio*) living in a highly seasonal environment. In our annual species, individuals born in the moist spring (high food availability) need to survive the harsh dry summer (low food availability) to be able to reproduce in the following spring. We studied the effect of the harsh dry vs. the benign moist season on telomere dynamics. We also tested whether telomere length and changes in telomere length were associated with the probability that individuals disappeared from the population during the dry season. Male but not female striped mice showed age-related telomere erosion. Telomeres were longer at the beginning of the dry season compared to the rest of the year. Telomeres increased significantly in length during the moist season. Neither telomere length at the onset of the dry season nor telomere loss over the dry season predicted whether or not individuals disappeared. In conclusion, our data suggest that seasonal attrition and restoring of telomeres also occurs in non-hibernating wild rodents living in hot food restricted environments.

1 **INTRODUCTION**

2 Telomeres are DNA sequences that cap the end of linear chromosomes and protect genome integrity  
3 (Chan and Blackburn 2004). Telomeres erode progressively with cell replication and accumulation of  
4 stress-derived damages (Ilmonen et al. 2008; Kotrschal et al. 2007; Olonikov 1973), which is considered  
5 as one main indicator of the ageing rate of cells and of organisms. Hence, a straightforward initial  
6 telomere-ageing hypothesis was that telomere length is shortened progressively with age, and that  
7 faster shortening rate actually reflects senescence, and reduced individual fitness (Dantzer and Fletcher  
8 2015; Salomons et al. 2009). Accordingly, both telomere length and telomere shortening have been  
9 suggested to predict survival and/or longevity in different taxa (Fairlie et al. 2016; Heidinger et al. 2012;  
10 Whittemore et al. 2019).

11  
12 A potential causal role of telomeres on life history trajectories was supported by studies showing that  
13 telomere length / shortening may reflect life stress effects like the costs of reproduction (Ilmonen et al.  
14 2008; Kotrschal et al. 2007), and are related to immune status and response (Crisuolo et al. 2018b).  
15 However, contradictory results have been published challenging the idea of continuous telomere  
16 shortening with age or its link with higher mortality (Cerchiara et al. 2017; Martin-Ruiz et al. 2005; Seeker  
17 et al. 2018). This highlighted a key element for our understanding of telomere role in animal ageing,  
18 which is that the telomere – fitness relationships are indeed more complex than initially thought,  
19 possibly because telomere dynamics evolved differently under distinct environmental pressures (Tian et  
20 al. 2018). Whether telomeres are causal in ageing or rather just a biomarker of somatic maintenance is  
21 still an open question (Simons 2015; Young 2018). This seems to be especially true for small mammals in  
22 which telomere shortening is often not part of the cell replicative ageing process (Gorbunova et al. 2008;  
23 Tian et al. 2018). For example, in laboratory mice, telomeres do not always shorten with cell divisions,  
24 (Gomes et al. 2010; Gorbunova et al. 2008). However, other recent studies in laboratory mouse strains

25 (e.g. C57BL/6 mice housed under *ad libitum* in standard laboratory conditions), showed some shortening  
26 in telomere length as mice aged, at rates predictive of individual longevity (Vera et al. 2012). In addition,  
27 sex differences exist in telomere length (Barrett and Richardson 2011), resulting in additional complexity  
28 related to sex-specific telomere length inheritance or aging. Field studies of telomere dynamics on small  
29 mammals to test these contrasting results obtained from controlled laboratory strains are mostly absent.

30 Even in other taxa than rodents, some studies found that telomeres may either be stable (cows:  
31 (Seeker et al. 2018)) or even increase over time (great tits: (Atema et al. 2018)). In other mammals, such  
32 as wild Soay sheep, telomere lengthening over an individual's lifetime was described (Fairlie et al. 2016)  
33 (for a bird example, see also (Spurgin et al. 2018)). Remarkably, a recent study on hibernating rodents  
34 have highlighted compensatory telomere lengthening when environmental conditions are becoming  
35 favorable after winter (Hoelzl et al. 2016a). Three explanations have been proposed for an increase of  
36 telomere length over time. Although (1) measurement errors or statistical adjustment in telomere length  
37 assessment need to be checked (Nettle and Bateson 2017; Simons et al. 2014; Verhulst et al. 2013),  
38 biological explanations have to be considered (Bateson and Nettle 2016), such as (2) variation in cell  
39 population on which telomere lengths are measured, or (3) in activated telomere maintenance processes  
40 (e.g. telomerase activity or alternative lengthening pathway) leading to actual elongation of telomere  
41 ends (Blackburn 1991; Cesare and Reddel 2010; Criscuolo et al. 2018a), a process which may be  
42 energetically costly (Hoelzl et al. 2016a; Nowack et al. 2019). If telomere size elongation in juvenile or  
43 adult somatic cells is more common than initially believed (Fairlie et al. 2016; Hausmann et al. 2004;  
44 Olsson et al. 2018), this would have important consequences for our understanding of how telomeres  
45 interact with fitness-related traits. For instance, it may highlight why long telomeres correlate better  
46 with individual fitness than short telomeres (Bauch et al. 2014). Importantly, we should test whether  
47 there is further evidence for an increase in telomere length in individuals living under different harsh  
48 environmental conditions than studied so far and whether this is related to fitness.

49 In wild-derived house mice strains (*Mus musculus*) kept under laboratory conditions, individuals  
50 showed accelerated telomere loss with immune, reproductive or social stress but non-significant  
51 relationships with mortality (Ilmonen et al. 2008; Kotrschal et al. 2007). Thus, one important emerging  
52 question is whether the idea that the shortening of telomeres under environmentally harsh conditions is  
53 long-lasting (De Punder et al. 2018; Reichert et al. 2014), or whether shortened telomeres can remain  
54 stable in size or even increase when environmental conditions become favorable. Whether telomere  
55 lengthening is a selected maintenance process improving individual fitness *via* increased lifespan, for  
56 example in hibernating mammals (Hoelzl et al. 2016b; Turbill et al. 2012), remains an unanswered  
57 question. To our knowledge, only four published studies described seasonal variation in telomere length  
58 (one on small wild mammals) (Fairlie et al. 2016; Hoelzl et al. 2016a; Quirici et al. 2016b; Stier et al.  
59 2014). Two studies approached this question indirectly through cross-sectional sampling, assessing  
60 differences in telomere length between individuals experiencing different environments during growth  
61 or reproduction (Quirici et al. 2016a; Stier et al. 2014). Studies in Soay sheep and edible dormice  
62 provided some evidence that changes in telomere length are more related to seasonal variation in  
63 environmental conditions (like high population densities and feeding conditions before and/or during the  
64 winter) than to age (Fairlie et al. 2016; Hoelzl et al. 2016a).

65 To better understand how telomere dynamics are affected by environmental harshness,  
66 longitudinal studies are needed, where individuals are sampled repeatedly under different  
67 environmental conditions. Here, we investigated telomere dynamics in African striped mice (*Rhabdomys*  
68 *pumilio*). Striped mice inhabit a highly seasonal environment with high food abundance in the moist  
69 spring being followed by the food restricted harsh dry season in summer. We sampled individuals  
70 repeatedly, at the onset of the dry season (January), after they experienced 3 months of harshness in the  
71 middle of the dry season (March), 7 months after they experienced harshness at the end of the onset of  
72 the moist season (July), and at the end of the favourable moist season with high food abundance

73 (October). We predicted telomere shortening to be faster during the harsh food-restricted dry season  
74 than during the favourable moist season, and that telomere length at the onset of the dry season is  
75 related to disappearance risk of individuals, which we used as proxy for dry season survival and thus  
76 fitness.  
77

78 **MATERIAL AND METHODS**

79 **Study species**

80 The striped mouse is an omnivorous annual species: individuals are born in the moist spring (August to  
81 December) with peak food availability, then have to survive the following hot summer dry season  
82 (January to May) with very low food availability, before reproducing in the following spring; less than 1%  
83 of striped mice survive to reproduce in a second spring (Schradin et al. 2012). Depending on the  
84 harshness of the dry season, 1% to 50% of the striped mice survive the dry season to reproduce, such  
85 that dry season survival is a key determinant of individual fitness. In the breeding season, striped mice  
86 can live solitarily but more often live in communally breeding groups with 2-4 breeding females and 1  
87 breeding male, and adult offspring remain philopatric, such that all striped mice live in social groups  
88 during the dry season (Schradin et al. 2010; Schradin et al. 2019).

89

90 **Data collection**

91 The study was conducted in Goegap Nature Reserve in South Africa (S 29 41.56, E 18 1.60). Goegap lies  
92 within the semi-desert Succulent Karoo biome, which is characterized by cold and moist winters followed  
93 by spring with high food abundance and hot dry summers with low food abundance. During the current  
94 study, food availability measured as the number of food plant species per 4 m<sup>2</sup> monitoring plot changed  
95 from 2.8 in January to 2.0 in March, 6.4 in August (after the July sampling), and 3.6 in October. Our field  
96 site was 7 ha and contained 12 social groups. Because the population was monitored throughout the  
97 year, we knew all individuals present on the field site, their life history, and their group affiliation  
98 (Schradin et al. 2019). Trapping continued even after our study such that we knew which individuals  
99 disappeared during the study and did not re-appear afterwards. During our normal monitoring, every  
100 group was trapped for 3 days 2 to 4 times every month (i.e. 6 to 12 days per month).



101 Tail tissue can be used to evaluate somatic telomere dynamics and are representative of telomere  
102 dynamics of overall body soma (Daniali et al. 2013). Collecting tail tissue has the advantage of being non-  
103 lethal and allowing repeated sampling, which is essential for longitudinal studies. We repeatedly  
104 collected tissue samples (1-2mm tail snips) from all individuals present on the field site (n=80, age 11 –  
105 737 days) during extensive trapping sessions (always for 7 days) in 2018 at: (i) the beginning of January  
106 (onset dry season); (ii) March (middle dry season); (iii) End of July, beginning of August (onset of the  
107 moist and breeding season); and (iv) October (end of the moist / breeding season). Individuals were born  
108 in the moist season (August and December) in 2016 (n=9) or 2017 (n=48), with 14 individuals being born  
109 between January and July 2018. Season of birth was unknown for 9 immigrants born in 2016 (n=2), 2017  
110 (n=6) and undefined (n=1). Tail clips were stored in 95% ethanol and transported to Strasbourg, France,  
111 for analysis.

112 For the onset of the dry season (January), we had samples from 54 individuals (29 females, 25  
113 males, age 15-501 days). For the onset of the moist season (end of July), we had samples from 26  
114 individuals (16 females, 10 males, age 9-652 days). In addition, intra-individual changes in telomere  
115 length was evaluated through repeated tissue sampling of the same individual in (i) January and March  
116 for 36 individuals (21 females, 15 males, individual variation in telomere dynamics over the dry season),  
117 (ii) July and October for 26 individuals (16 females, 10 males, individual variation in telomere dynamics  
118 over the moist season), and (iii) 6 individuals sampled monthly during all four months (Figure 1).

119 Fieldwork and tissue sampling were carried out under the necessary licenses. All data were  
120 collected following protocols approved by the Animal Ethics Screening Committee of the University of  
121 the Witwatersrand (AESC 2012/37/2A, AESC 2014/40/B).

122

123 **Telomere analysis**

124 Striped mice DNA was extracted from tail tissue using Nucleospin Tissue kit (Macherey-Nagel, Düren,  
125 Germany). DNA quality and concentration were assessed with a Nano-Drop spectrophotometer  
126 (NanoDrop Technologies, Thermo Fisher Scientific) and diluted to the concentration of 2.5 ng/mL.  
127 Relative telomere length was measured in striped mice on extracted DNA from tail tissue, using the  
128 methodology of real-time qPCR that originated from the work of Cawthon and collaborators on human  
129 samples (Cawthon 2002). For our study, the qPCR amplification protocol has been adapted as follows:  
130 (i) qPCR gives us as a final results a ratio (T/S) of amplification cycles (Cq) of the telomere sequence (T)  
131 and of a genomic gene sequence (S) that must be specific or preferably a non-variable copy number  
132 (non-VCN) gene (Smith et al. 2011). Final T/S values were calculated by taking into account each plate  
133 efficiencies, following (Pfaffl 2001). For the present study, we designed amplification primers for a region  
134 using aligned sequences of PGK1 genes from other closely related organisms extracted from the NCBI  
135 data base (Huchon et al. 2017). We used a 150 bp portion of the phosphoglycerate gene (PGK1), which  
136 was found non-variable in copy number within our population of striped mice (Cq values ranged  
137 between 24-25.6 for 90% of the samples). Primer sequences for the non-VCN gene were: *Forward*  
138 *primer*: 5'-CGGAGACACCGCCACTTG-3' and *Reverse primer*: 5'-AAGGCAGGAAAATACTAAACAT-3'. The  
139 forward and reverse primers for the telomere sequence were: 5'-  
140 CGGTTTGGTTGGGTTTGGGTTTGGGTTTGGGTTTGGGTT-3' and 5'-  
141 GGCTTGCCTTACCCTTACCCTTACCCTTACCCTTACCCT-3', respectively.  
142 (ii) Amplifications of T and S were done on distinct 96-wells plates due to differences in amplification  
143 temperatures using an automated thermocycler (Master-cycler Realplex; Eppendorf, Hamburg,  
144 Germany). The reactions conditions were set as follows: for the telomere amplification, 95°C for 2 min –  
145 followed by 30 cycles of 95°C for 15 sec, 56°C for 15 sec, 72°C for 30 sec; for the PGK1 amplification, 95°C  
146 for 2 min – followed by 30 cycles of 95°C for 15 sec, 60°C for 15 sec, 72°C for 30 sec. All amplification  
147 runs were ended by a melting curve program (steady increase in amplification temperature from 56°C (T)

148 or 60°C (S)) to verify the absence of non-specific amplification events (*i.e.* lack of specificity of the  
149 primers or primer-dimer artefacts, see below). A negative control (without DNA) was also present on  
150 each plate to check for potential contamination.

151 (iii) Amplifications were performed with 5 ng of DNA (2 uL) per well, in a final volume of 10 uL containing  
152 5 uL of GoTaq qPCR mix (Promega, Madison, WI, USA), 0.5 uL of each primers at an initial concentration  
153 of 5 nM and 2 uL of ultra-pure water.

154 (iv) Samples were amplified in duplicates, on 4 different runs (each comprising 1 telomere plate and 1  
155 PGK1 plate). Each 96-wells plate included a dilution curve of the same randomly-chosen reference  
156 sample, serially diluted from 10 to 0.625 ng/mL of DNA, to check for consistency in amplification  
157 efficiency among plates ( $r^2$  of the dilution curves were comprised between 0.931 – 0.998 (telomere) and  
158 0.995 – 0.998 (PGK1)). In addition, 5 samples were repeated over all plates. Mean amplification  
159 efficiencies were of  $96.0 \pm 0.44$  % for PGK1 and  $97.8 \pm 0.97$  % for telomere. We did not include any plate  
160 effect in our models since there was no indications for an inter-plate variation in the T/S ratio values  
161 (Linear model, estimates  $-0.002 \pm 0.017$ ,  $t = -0.13$ ,  $P = 0.896$ ).

162

### 163 **qPCR repeatabilities and effect of storage time**

164 As suggested by (Nettle et al. 2019), we evaluated the repeatability of our measures using intra-class  
165 correlation coefficients (ICC, *R* package ICC, (Wolak et al. 2012)) both for intra-plate (duplicates) and  
166 inter-plates variability in T/S ratio (see also (Eisenberg 2016)). Intra-plate ICC was of 0.864 and inter-plate  
167 ICC was of 0.836 (Cicchetti 1994). We also checked whether ICC varied with time of storage, *i.e.* time of  
168 sampling (from January to October): all samples were analysed together in May 2018, such that samples  
169 from January had the longest storage time and those samples from October had shortest storage time.  
170 Measurement repeatability (intra-individual) did not change with time from January (ICC = 0.851), March  
171 (0.835), July (0.769) and October 2018 (0.901). In addition, we found that T/S ratios were correlated

172 within individuals measured both in January and March ( $n=35$ ,  $t_{1,34} = 2.413$ ,  $P = 0.021$ ), and both in July  
173 and October ( $n=30$ ,  $t_{1,29} = 2.198$ ,  $P = 0.036$ ; see Figure1 in ESM 1). Additional qPCR methodological  
174 details are presented in ESM1.

175

## 176 **Statistical analysis**

177 Data were analysed using R version 3.5.1 (R Foundation for statistical Computing Platform, 2018), using  
178 the packages *lme4* (function *lmer*) (Bates et al. 2014) and *Factominer* (Lê et al. 2008). We tested for  
179 normality with both a Shapiro-Wilk's test and QQ plots distribution (*fitdistrplus* package, (Delignette-  
180 Muller and Dutang 2015)). We used a Linear Mixed Model (LMM) to analyse the relationship between  
181 telomere length ( $n=154$  measures) and the following explanatory variables: *Age*, *sex*, and *season* and all  
182 interactions. Season was coded in 4 categories : beginning of the dry season (January), middle of the dry  
183 season (March), beginning of the moist season (July) and middle of the moist season (October). So, our  
184 statistical analysis considers the 4 sampling points and tested for differences among 4 season categories.  
185 Posthoc multiple comparisons among the 4 season categories were conducted using the function *glht*  
186 from the *multcomp* package (Hothorn et al. 2008). Individual ID was included as random factor (to  
187 control for repeated sampling). Because age, body mass and body size are interdependent, we also used  
188 a principal component analysis (PCA) to merge those variables into one PC axis (ESM2). PCA1 turned out  
189 to be non-significant in all models, so we kept only *Age* as a fixed factor in the final analysis. The date of  
190 birth was initially used as a fixed factor when testing variance in individual relative telomere length, but  
191 it was not significant (LMM, estimates  $0.002 \pm 0.001$ ,  $t = 1.556$ ,  $P = 0.124$ ) and not considered in our final  
192 models. Individual changes in telomere length over the dry and moist seasons were tested using paired *t*-  
193 tests. To test the predictive value of relative telomere length on disappearance risk of individual striped  
194 mice from the population (Vuarin et al. 2019) , we used a linear mixed model with binomial distribution  
195 of the survival variable (0 and 1, the latter indicating the presence of the individual). Individuals were

196 regarded as not having disappeared when they were still trapped in July or afterwards (monthly trapping  
197 of groups continued for 1.5 years). Of the 54 individuals sampled in January, 19 survived until July and 35  
198 disappeared. July was chosen as the censoring date, since this is when breeding dispersal peaks, such  
199 that most cases of disappearance that occur before this time can be assumed to be due to mortality  
200 (Vuarin et al. 2019). Explanatory variables included in the models were telomere lengths measured in  
201 January, March and July (n=52 measures, with Individual ID as random factor). In addition, telomere  
202 length rate of change (*i.e.* the within individual differences in telomere lengths measured within the dry  
203 season or TROC, n=92 measures) was tested separately. Using the correction for the regression to the  
204 mean (RtM) effect ((Verhulst et al. 2013), ESM4) or using, as we present in the main text, uncorrected  
205 TROC with the initial telomere length as a covariate (Bateson et al. 2019; Hoelzl et al. 2016b) did not  
206 change the output of the models . We choose to present models obtained after model selection using  
207 the Akaike Information Criterion corrected for small sample sizes (*Mumin* R package, (Barton 2020),  
208 ESM3).  
209

210 **RESULTS**

211 There was an overall significant variation in relative telomere length over the seasons (Table 1, Figure 2).  
212 Posthoc Tukey contrasts indicated that, overall, relative telomere lengths were longest at the start of the  
213 dry season in January and shortest in March and July. Relative telomere lengths were significantly longer  
214 in October than in July ( $P = 0.035$ ; while there was no significant difference between March and October,  
215  $P = 0.994$ ). Individual telomere dynamics (Figure 3) demonstrated that, during the dry season, telomere  
216 length of most individuals declined (paired  $t$ -test,  $t_{35} = 3.486$ ,  $P = 0.001$ , mean difference in relative  
217 telomere length =  $-0.238$  T/S ratio) whereas during the moist season, telomere lengths of most  
218 individuals increased (paired  $t$ -test,  $t_{31} = 4.373$ ,  $P < 0.001$ , mean difference in relative telomere length =  
219  $0.170$  T/S ratio).

220 The difference in relative telomere length between sexes depended on age (interaction term Age  
221 x Sex:  $P = 0.031$ , Table 1). Telomeres of males shortened significantly during ageing while female  
222 telomeres did not (Figure 4, linear regression, males:  $df = 62$ ,  $t = -2.91$ ,  $P = 0.005$ ; females:  $df = 88$ ,  $t = -$   
223  $0.09$ ,  $P = 0.930$ ).

224 Individual disappearance risk during the dry season (*i.e.* from January to July) was not  
225 significantly predicted by the relative telomere length measured at the beginning of the dry season ( $n =$   
226  $52$ , Table 2A) nor by TROC within the dry season (January to March, Table 2B). TROC corrected for the  
227 regression to the mean stayed non-significant for predicting disappearance risk ( $0.956 \pm 0.866$ ,  $z = 0.110$ ,  
228  $P = 0.912$ ). None of the other explanatory variables (sex, age, and interaction terms, all  $P > 0.308$ )  
229 predicted individual disappearance.

230

231 **DISCUSSION**

232 Here we showed that (i) the degree of telomere length degradation differed between the harsh dry and  
233 the favorable moist season in African striped mice, and (ii) that this effect appeared independent of an  
234 age effect which was only evident in males. In fact, on average, telomere length even increased in  
235 individuals from July to October (more so in females), demonstrating that the telomere loss during the  
236 preceding dry season cannot be simply explained by age (Figure 3). This indicates that environmental  
237 harshness influences telomere dynamics: telomeres degrade under harsh conditions but can increase  
238 afterwards, at least in females. Thus, our study indicates that a small mammal living under harsh  
239 conditions characterized by heat and low food abundance shows similar telomere dynamics as small  
240 hibernating mammals after winter but can be subsequently restored, at least partially, during periods of  
241 higher food availability (Hoelzl et al. 2016a).

242 Telomere shortening might indicate energetic costs (Crisuolo et al. 2018b; Ilmonen et al. 2008;  
243 Nettle et al. 2015; Reichert et al. 2014). In our study, food was two times more abundant during the peak  
244 of the moist season than during the dry season, such that the dry season was much harsher than the  
245 moist season. Accordingly, telomeres shortened significantly during the dry season. Physiological factors  
246 that are suspected to negatively affect telomeres in other species include high metabolic rate via  
247 increased oxidative stress ((Reichert and Stier 2017) but see (Boonekamp *et al.* 2017) and (Turbill et al.  
248 2012)) and corticosterone levels ((Hausmann et al. 2012; Tissier et al. 2014) but see (Gil *et al.* 2019)).  
249 Also, in striped mice, the food restricted dry season triggers several physiological mechanisms (Rimbach  
250 et al. 2016; Schradin 2008a; Schradin 2008b; Schradin et al. 2014), enabling them to reduce both resting  
251 metabolic rate and daily energy expenditure (Rimbach et al 2018a). Metabolic rate and corticosterone  
252 levels in striped mice are increased during the moist season (Rimbach et al. 2018a; Rimbach et al. 2018b;  
253 Schradin 2008c) which would predict a decrease in telomere length, while in opposite we observed an  
254 increase. Instead, the low energy availability in the dry season might lead to a decreased investment in

255 cell maintenance and telomere degradation. In the harsh summer, striped mice might rather invest in  
256 mechanisms that improve immediate dry season survival while increased food abundance in the  
257 subsequent spring favours energy investment into telomere maintenance.

258 We found age-related telomere erosion in males but not in females. Males also tended to have  
259 slightly longer telomeres than females (Table 1,  $P = 0.076$ ). If males start their yearly cycle with longer  
260 telomeres (Figure 3), this may give a mechanistic explanation for faster shortening thereafter, since long  
261 telomeres are expected to be more susceptible to damage and less to maintenance than short telomeres  
262 (Bauch et al. 2014). A trade-off based effect due to sex-specific investment in reproduction vs. telomere  
263 maintenance should be explored in the future, based, for instance, on an earlier reproductive investment  
264 in males during the dry season (Schradin and Pillay 2005). Further, females may better protect their  
265 telomeres or invest in soma maintenance in order to transfer longer telomeres to the next generation, as  
266 suggested for another ageing-related mechanism, oxidative stress, in two distinct mammal species  
267 (*Mungos mungo* and *Urocitellus columbianus*, (Viblanco et al. 2018; Vitikainen et al. 2016). Future studies  
268 should investigate sex specific telomere dynamics depending on sex specific investment into  
269 reproduction and whether the shield hypothesis protecting offspring from maternal oxidative stress  
270 (Blount et al. 2016) extends to telomere inheritance.

271 Only 37% of the individuals sampled at the onset of the dry season were still present on the field  
272 site after the dry season, the rest having disappeared. In our current study, individuals with shorter  
273 telomeres were not more likely to disappear during the food restricted dry season. This leads to the  
274 hypothesis that telomere length in striped mice is uncoupled from short-term survival because of high  
275 extrinsic mortality risks. An alternative may be that, like other small rodents (Seluanov et al. 2007),  
276 striped mice maintain a high telomerase activity throughout their lives and consequently telomere  
277 length is not a good cell ageing marker. However, in wild mice strains (*Mus musculus* (Criscuolo et al.  
278 2018b; Kotrschal et al. 2007)) and in a laboratory study conducted on Sudanian grass rat (*Arvicanthis*



279 *ansorgei*, (Grosbellet et al. 2015)), the rate of telomere shortening increased with stress caused by  
280 circadian desynchronization. Telomere shortening in mice should then be more closely related to survival  
281 (Ilmonen et al. 2008), since a faster telomere loss may reflect a concomitant deterioration of the  
282 organism's functioning (*i.e.* as an intrinsic ageing process, (Harris et al. 2016; Whittemore et al. 2019)).  
283 We did not find such a relationship since individuals with increased telomere shortening during the dry  
284 season were not more likely to disappear. In future, we need more data to test whether telomere length  
285 is correlated with reproductive success in striped mice.

286         It is currently widely debated whether reported increases in telomere length represents  
287 measurement errors or are of biological relevance (Bateson and Nettle 2016; Steenstrup et al. 2013). To  
288 date, negative and positive variation in telomere length over life have been found in free-living mammals  
289 and birds sampled longitudinally (Fairlie et al. 2016; Spurgin et al. 2018), illustrating the potential  
290 biological relevance of telomere lengthening. In striped mice, telomeres even increased in many  
291 individuals during the moist season (Figure 3). Although a long-term study with several dry and moist  
292 seasons differing in harshness is needed to definitively show an effect of harshness on telomere  
293 dynamics, the observed lengthening of telomeres raises questions about both its ultimate function (*i.e.*  
294 link with reproductive success) and proximate mechanism (*i.e.* seasonal changes in telomerase activity or  
295 changes in the rate of cell renewal). It is important to note that even though we were not able to  
296 statistically test for true lengthening (which needs at least 3 repeated samples, (Simons et al. 2014),  
297 because we used repeated tail sampling for telomere measurements, attributing telomere lengthening  
298 to a change in cell population is less likely than in studies using repeated blood sampling. One ultimate  
299 hypothesis may be related to breeding. Since the moist season is also the breeding season for striped  
300 mice, the lengthening of somatic telomeres may improve reproductive success either because it is  
301 associated with enhanced adult physiological processes or because it ensures better offspring quality.  
302 Therefore, elongating telomeres in breeding adults may be of key importance for fitness. Such an

303 increase in telomere length was previously found in the edible dormouse (*Glis glis*), also during the active  
304 breeding season (Turbill *et al.* 2013). Our study sheds lights on the fact that telomere elongation is not  
305 specific to hibernating rodents. Itsuggests that telomere dynamics change seasonally in a sex specific  
306 way in free-living striped mice, and that, as previously found in hibernating small mammals (Hoelzl *et al.*  
307 2016a; Nowack *et al.* 2019), telomeres can even lengthen after a harsh season when environmental  
308 conditions become favourable. This indicates that telomere dynamics are plastic and future studies need  
309 to focus on the ultimate function and proximate mechanisms of increasing telomere length in striped  
310 mice specifically and free-living animals generally.

311

#### 312 **ACKNOWLEDGEMENTS**

313 This study was supported by the Succulent Karoo Research Station, the University of the Witwatersrand,  
314 and the CNRS. We are grateful to Goegap Nature Reserve and for the help of several field managers and  
315 assistants. Data are available online at figshare doi: xxxxxx.

316

317 **FIGURES legends**

318 **Figure1**

319 Schematic representation of the repeated sampling design of the study. In total, we collected 154  
320 samples from 80 individuals (starting with 54 mice in January and adding 26 mice in July), but not all  
321 individuals were sampled repeatedly. 36 individuals were sampled both at the start (January) and the  
322 middle (March) of the dry season. 32 individuals were sampled both at the start (July) and the middle  
323 (October) of the moist season. Altogether 6 individuals were sampled during all four months.

324 **Figure 2**

325 Year-round variation in relative telomere length in adult striped mice (156 samples from 80 individuals).  
326 Relative telomere length was repeatedly measured in 36 individuals over part of the dry season (two  
327 sampling sessions in January and March) and in 32 individuals over the moist season (July and October).  
328 \* indicates significant levels of posthoc Tukey contrasts: \*\*\* < 0.001, \*\* < 0.01, \* < 0.05. Boxplots  
329 indicate minimum and maximum, median (black line), first and third quartile and mean (diamonds)  
330 values. See text for details.

331

332 **Figure 3**

333 Individual variation in relative telomere length in male (left panel) and female (right panel) adult striped  
334 mice over the beginning and middle of the dry season (January and February, black circles, 36  
335 individuals) and beginning and middle of the moist season (July and October, grey circles, 26  
336 individuals). Increases in telomere length were found in 24 of 32 individuals in the moist season.  
337 Numbers for the dry season was 11 of 36 individuals. Dashed lines indicate the 6 individuals that were  
338 sampled 4 times over the year.

339

340 **Figure 4**

341 Relative telomere length of striped mice in relation to sex over the entire study period. The Sex x Age  
342 interaction was significant ( $P = 0.031$ , Table 1), indicating that males underwent an age-related telomere  
343 erosion (linear regression,  $P = 0.005$ ) while females did not (linear regression,  $P = 0.930$ ). See text for  
344 details. Confidence intervals 95% are indicated in grey.  
345

346

347 **Table 1:** Linear mixed model explaining the effects of season (coded using the 4 monthly sampling  
348 points, January / March for the dry season and July / October for the moist season), age, and sex on  
349 individual relative telomere length in wild striped mice. The model encompasses repeated telomere data  
350 from individuals sequentially sampled within seasons (individual ID as random factor) but includes all  
351 individual measurements (in total 154 telomere measurements among 80 individuals). Seasons are  
352 defined as dry (January and March) and moist (July and October, see figure 1). Significant fixed effects (in  
353 bold) are considered at  $P < 0.05$  and tendencies (in italics) are considered at  $P < 0.1$ , P values being  
354 obtained using the *anova* R function. The model presented was selected based on AICc values. Posthoc  
355 comparisons were done using the *multcomp* R package.

356

Response variable: Relative telomere length		Estimates $\pm$ SE	df	Z value	P
<i>Random factors</i>					
	Individual ID	0.019 $\pm$ 0.014			
	Residual variance	0.063 $\pm$ 0.251			
<i>Fixed factors</i>					
	Intercept	0.935 $\pm$ 0.057	128.2	16.275	<b>&lt; 0.001</b>
	Sex (M)	0.150 $\pm$ 0.084	100.6	1.794	<i>0.076</i>
	Age	0.001 $\pm$ 0.001	114.4	0.436	0.664
	Sex x Age	-0.001 $\pm$ 0.001	100.8	-2.192	<b>0.031</b>
	Season	-0.199 $\pm$ 0.062	104.9	10.723	<b>&lt; 0.001</b>
<i>Tukey Contrast posthoc</i>				<b>Z value</b>	<b>P</b>
	January - March	0.181 $\pm$ 0.056		3.263	<b>0.006</b>
	January - July	0.334 $\pm$ 0.063		5.280	<b>&lt; 0.001</b>
	January - October	0.164 $\pm$ 0.063		2.593	<b>0.047</b>
	March - July	0.152 $\pm$ 0.068		2.234	0.113
	March - October	-0.018 $\pm$ 0.068		-0.259	0.994
	July - October	-0.170 $\pm$ 0.063		-2.692	<b>0.035</b>

357  
358  
359

360 **Table 2:** Predictors of survival of striped mice until the beginning of the 2018 breeding season (*i.e.* July).  
 361 A generalized linear model was used, following a binary distribution of individual survival (0 =  
 362 disappearance) using (A) the individual telomere lengths measured at the start of the dry season (*i.e.*  
 363 January) and (B) the rate of change in telomere length (TROC) over the dry season (*i.e.* January – March).  
 364 Results were also not significant when using TROC corrected for the regression to the mean effect (see  
 365 Results). Estimates  $\pm$  SE are given for males vs. females. Significant fixed effects (in bold) are considered  
 366 for  $P < 0.05$ . The models presented here were selected from AICc values.

<b>A. Response variable: Survival</b>	<b>Estimates <math>\pm</math> SE</b>	<b>Z value</b>	<b>P</b>
<i>Random factors</i> Individual ID	5276 $\pm$ 72.63		
<i>Fixed factors</i>			
Intercept	-18.899 $\pm$ 6.633	-2.849	<b>0.004</b>
Relative telomere length (T/S ratio)	0.664 $\pm$ 6.218	0.107	0.915
Sex (M)	-9.389 $\pm$ 23.168	-0.405	0.685
Age	-0.063 $\pm$ 2.902	-0.180	0.798
Sex x Age	0.039 $\pm$ 0.052	0.749	0.454
Relative telomere length x Sex	6.063 $\pm$ 14.284	0.424	0.671
<b>B. Response variable: Survival</b>	<b>Estimates <math>\pm</math> SE</b>	<b>Z value</b>	<b>P</b>
<i>Random factors</i> Individual ID	NA		
<i>Fixed factors</i>			
Intercept	-1.070 $\pm$ 1.703	-0.629	0.530
Telomere rate of change (T/S ratio)	0.390 $\pm$ 1.926	0.203	0.839
Age	0.002 $\pm$ 0.003	0.781	0.435
Sex (M vs. F)	-2.791 $\pm$ 1.395	-2.001	<b>0.045</b>
Initial relative telomere length	1.144 $\pm$ 1.900	0.602	0.547
Age x Sex	0.017 $\pm$ 0.012	1.363	0.173

367  
 368

369

370 **Literature cited**

- 371 Atema E, Mulder E, Van Noordwijk AJ, Verhulst S (2018) Ultra-long telomeres shorten with age  
372 in nestling great tits but are static in adults and mask attrition of short telomeres.  
373 *Molecular Ecology Resources* 19:648-658
- 374 Barrett ELB, Richardson DS (2011) Sex differences in telomeres and lifespan. *Aging Cell* 10:913-  
375 921. doi: 10.1111/j.1474-9726.2011.00741.x
- 376 Barton K (2020) MuMIn: Multi-Model Inference. . R package version 1.43.17. doi:  
377 <https://CRAN.R-project.org/package=MuMIn>
- 378 Bates D, Mächler M, Bolker B, Walker S (2014) Fitting linear mixed-effects models using lme4.  
379 arXiv preprint arXiv 1808.00864:1406.5823
- 380 Bateson M, Eisenberg DTA, Nettle D (2019) Controlling for baseline telomere length biases  
381 estimates of the rate of telomere attrition. *Royal Society Open Science* 6:190937. doi:  
382 10.1098/rsos.190937 10.6084/m9.figshare.c
- 383 Bateson M, Nettle D (2016) The telomere lengthening conundrum - it could be biology. *Aging*  
384 *Cell* 16:312-319
- 385 Bauch C, Becker PH, Verhulst S (2014) Within the genome, long telomeres are more informative  
386 than short telomeres with respect to fitness components in a long-lived seabird.  
387 *Molecular Ecology* 23:300-310. doi: 10.1111/mec.12602
- 388 Blackburn EH (1991) Structure and function of telomeres. *Nature* 6319:569
- 389 Blount JD, Vitikainen EIK, Stott I, Cant MA (2016) Oxidative shielding and the cost of  
390 reproduction. *Biological Reviews* 91:483-497. doi: 10.1111/brv.12179
- 391 Boonekamp JJ, Bauch C, Mulder E, Verhulst S (2017) Does oxidative stress shorten telomeres?  
392 *Biology Letters* 13:20170164
- 393 Cawthon RM (2002) Telomere measurement by quantitative PCR. *Nucleic Acids Research* 30:e47
- 394 Cerchiara JA, Risques RA, Prunkard D, Smith JR, Kane OJ, BOERSMA PD (2017) Telomeres  
395 shorten and then lengthen before fledging in Magellanic penguins (*Spheniscus*  
396 *magellanicus*). *Aging* 9:487-493
- 397 Cesare AJ, Reddel RR (2010) Alternative lengthening of telomeres: models, mechanisms and  
398 implications. *Nature Reviews Genetics* 11:319-330. doi: 10.1038/nrg2763
- 399 Chan SRWL, Blackburn EH (2004) Telomeres and telomerase. *Philosophical Transactions of the*  
400 *Royal Society B: Biological Sciences* 359:109-122. doi: 10.1098/rstb.2003.1370
- 401 Cicchetti DV (1994) Guidelines, criteria, and rules of thumb for evaluating normed and  
402 standardized assessment instruments in psychology. *Psychological Assessment* 6:284-  
403 290. doi: 10.1037/1040-3590.6.4.284
- 404 Criscuolo F, Smith S, Zahn S, Heidinger BJ, Haussmann MF (2018a) Experimental manipulation of  
405 telomere length: does it reveal a corner-stone role for telomerase in the natural  
406 variability of individual fitness? *Philosophical Transactions of the Royal Society B:*  
407 *Biological Sciences* 373:20160440. doi: 10.1098/rstb.2016.0440 10.6084/m9
- 408 Criscuolo F, Sorci G, Behaim-Delarbre M, Zahn S, Faivre B, Bertile F (2018b) Age-related  
409 response to an acute innate immune challenge in mice: proteomics reveals a telomere

410 maintenance-related cost. *Proceedings of the Royal Society B: Biological Sciences*  
411 285:20181877. doi: 10.1098/rspb.2018.1877

412 Daniali L et al. (2013) Telomeres shorten at equivalent rates in somatic tissues of adults. *Nature*  
413 *Communications* 4:1597. doi: 10.1038/ncomms2602

414 Dantzer B, Fletcher QE (2015) Telomeres shorten more slowly in slow-aging wild animals than in  
415 fast-aging ones. *Experimental Gerontology* 71:38-47. doi: 10.1016/j.exger.2015.08.012

416 De Punder K, Heim C, Przesdzing I, Wadhwa PD, Entringer S (2018) Characterization in humans  
417 of *in vitro* leucocyte maximal telomerase activity capacity and association with stress.  
418 *Philosophical Transactions of the Royal Society B: Biological Sciences* 373. doi:  
419 10.1098/rstb.2016.0441 10.6084/m9

420 Delignette-Muller M-L, Dutang C (2015) Fitdistrplus: An R package for fitting distributions.  
421 *Journal of Statistical Software* 64:1-34

422 Eisenberg DTA (2016) Telomere length measurement validity: the coefficient of variation is  
423 invalid and cannot be used to compare quantitative polymerase chain reaction and  
424 Southern blot telomere length measurement techniques. *International Journal of*  
425 *Epidemiology*:dyw191. doi: 10.1093/ije/dyw191

426 Fairlie J, Holland R, Pikington JG, Pemberton JM, Harrington L, Nussey DH (2016) Lifelong  
427 leukocyte telomere dynamics and survival in a free-living mammal. *Aging Cell* 15:140-148

428 Gil D, Alfonso-Iñiguez S, Pérez-Rodríguez L, Muriel J, Monclús R (2019) Harsh conditions during  
429 early development influence telomere length in an altricial passerine: Links with  
430 oxidative stress and corticosteroids. *Journal of Evolutionary Biology* 32:111-125. doi:  
431 10.1111/jeb.13396

432 Gomes NMV, Shay JW, Wright WE (2010) Telomere biology in Metazoa. *FEBS Letters* 584:3741-  
433 3751. doi: 10.1016/j.febslet.2010.07.031

434 Gorbunova V, Bozzella MJ, Seluanov A (2008) Rodents for comparative aging studies: from mice  
435 to beavers. *Age* 30:111-119. doi: 10.1007/s11357-008-9053-4

436 Grosbellet E et al. (2015) Circadian desynchronization triggers premature cellular aging in a  
437 diurnal rodent. *The FASEB Journal* 29:4794-4803. doi: 10.1096/fj.14-266817

438 Harris SE et al. (2016) Longitudinal telomere length shortening and cognitive and physical  
439 decline in later life: The Lothian Birth Cohorts 1936 and 1921. *Mechanisms of Ageing and*  
440 *Development* 154:43-48. doi: 10.1016/j.mad.2016.02.004

441 Hausmann MF, Longenecker AS, Marchetto NM, Juliano SA, Bowden RM (2012) Embryonic  
442 exposure to corticosterone modifies the juvenile stress response, oxidative stress and  
443 telomere length. *Proceedings of the Royal Society B: Biological Sciences* 279:1447-1456.  
444 doi: 10.1098/rspb.2011.1913

445 Hausmann MF, Winkler DW, Huntington CE, Nisbet ICT, Vleck CM (2004) Telomerase  
446 Expression Is Differentially Regulated in Birds of Differing Life Span. *Annals of the New*  
447 *York Academy of Sciences* 1019:186-190. doi: 10.1196/annals.1297.029

448 Heidinger BJ, Blount JD, Boner W, Griffiths K, Metcalfe NB, Monaghan P (2012) Telomere length  
449 in early life predicts lifespan. *Proceedings of the National Academy of Sciences*  
450 109:1742-1748. doi: 10.1073/pnas.1113306109

451 Hoelzl F, Cornils JS, Smith S, Moodley Y, Ruf T (2016a) Telomere dynamics in free-living edible  
452 dormice (*Glis glis*): the impact of hibernation and food supply. *The Journal of*  
453 *Experimental Biology* 219:2469-2474. doi: 10.1242/jeb.140871



454 Hoelzl F, Smith S, Cornils JS, Aydinonat D, Bieber C, Ruf T (2016b) Telomeres are elongated in  
455 older individuals in a hibernating rodent, the edible dormouse (*Glis glis*). *Scientific*  
456 *Reports* 6:36856. doi: 10.1038/srep36856

457 Hothorn T, Bretz F, Westfall P, Heiberger RM (2008) Multcomp: simultaneous inference for  
458 general linear hypotheses. R package version, 1-0.

459 Huchon D, Stepan SJ, Schenk JJ (2017) Muroid rodent phylogenetics: 900-species tree reveals  
460 increasing diversification rates. *Plos One* 12:e0183070. doi:  
461 10.1371/journal.pone.0183070

462 Ilmonen P, Kotrschal A, Penn DJ (2008) Telomere attrition due to infection. *Plos One* 3:e2143.  
463 doi: 10.1371/journal.pone.0002143.t001

464 Kotrschal A, Ilmonen P, Penn DJ (2007) Stress impacts telomere dynamics. *Biology Letters*  
465 3:128-130. doi: 10.1098/rsbl.2006.0594

466 Lê S, Josse J, Husson F (2008) FactoMineR: An R Package for Multivariate Analysis. *Journal of*  
467 *Statistical Software* 25:2-18

468 Martin-Ruiz CM, Gussekloo J, Van Heemst D, Von Zglinicki T, Westendorp RGJ (2005) Telomere  
469 length in white blood cells is not associated with morbidity or mortality in the oldest old:  
470 a population-based study. *Aging Cell* 4:287-290

471 Nettle D, Bateson M (2017) Detecting telomere elongation in longitudinal datasets: analysis of a  
472 proposal by Simons, Stulp and Nakagawa. *PeerJ* 5:e3265. doi: 10.7717/peerj.3265

473 Nettle D, Monaghan P, Gillespie R, Brilot B, Bedford T, Bateson M (2015) An experimental  
474 demonstration that early-life competitive disadvantage accelerates telomere loss.  
475 *Proceedings of the Royal Society B: Biological Sciences* 282:20141610-20141610. doi:  
476 10.1098/rspb.2014.1610

477 Nettle D, Seeker LA, Nussey D, Froy H, Bateson M (2019) Consequences of measurement error  
478 in qPCR telomere data: A simulation study. *PloS one* 14:e0216118. doi: 10.1101/491944

479 Nowack J, Tarmann I, Hoelzl F, Smith S, Giroud S, Ruf T (2019) Always a price to pay: hibernation  
480 at low temperatures comes with a trade-off between energy savings and telomere  
481 damage. *Biology Letters* 15:20190466. doi: 10.1098/rsbl.2019.0466

482 Olonikov AM (1973) A theory of marginotomy. *J Theor Biol.* 41:181-190

483 Olsson M, Wapstra E, Friesen C (2018) Ectothermic telomeres: it's time they came in from the  
484 cold. *Philosophical Transactions of the Royal Society B: Biological Sciences*  
485 373:20160449. doi: 10.1098/rstb.2016.0449

486 Pfaffl MW (2001) A new mathematical model for relative quantification in real-time RT-PCR.  
487 *Nucleic Acids Research* 29:2003-2007

488 Quirici V, Guerrero CJ, Krause JS, Wingfield JC, Vásquez RA (2016a) The relationship of telomere  
489 length to baseline corticosterone levels in nestlings of an altricial passerine bird in  
490 natural populations. *Frontiers in Zoology* 13. doi: 10.1186/s12983-016-0133-5

491 Quirici V, Guerrero CJ, Krause JS, Wingfield JC, Vásquez RA (2016b) The relationship of telomere  
492 length to baseline corticosterone levels in nestlings of an altricial passerine bird in  
493 natural populations. *Frontiers in Zoology* 13:1. doi: 10.1186/s12983-016-0133-5

494 Reichert S, Stier A (2017) Does oxidative stress shorten telomeres in vivo? A review. *Biology*  
495 *Letters* 13:20170463. doi: 10.1098/rsbl.2017.0463

496 Reichert S et al. (2014) Increased brood size leads to persistent eroded telomeres. *Frontiers in*  
497 *Ecology and Evolution* 2. doi: 10.3389/fevo.2014.00009

498 Rimbach R, Blanc S, Zahariev A, Gatta M, Pillay N, Schradin C (2018a) Seasonal variation in  
499 energy expenditure in a rodent inhabiting a winter-rainfall desert. *Journal of*  
500 *Comparative Physiology B*. doi: 10.1007/s00360-018-1168-z

501 Rimbach R, Jäger J, Pillay N, Schradin C (2018b) Food Availability Is the Main Driver of Seasonal  
502 Changes in Resting Metabolic Rate in African Striped Mice (*Rhabdomys pumilio*).  
503 *Physiological and Biochemical Zoology* 91:826-833. doi: 10.1086/696828

504 Rimbach R, Pillay N, Schradin C (2016) Both thyroid hormone levels and resting metabolic rate  
505 decrease in African striped mice when food availability decreases. *The Journal of*  
506 *Experimental Biology*. doi: 10.1242/jeb.151449

507 Salomons HM, Mulder GA, van de Zande L, Haussmann MF, Linskens MHK, Verhulst S (2009)  
508 Telomere shortening and survival in free-living corvids. *Proceedings of the Royal Society*  
509 *B: Biological Sciences* 276:3157-3165. doi: 10.1098/rspb.2009.0517

510 Schradin C (2008a) Differences in prolactin levels between three alternative male reproductive  
511 tactics in striped mice (*Rhabdomys pumilio*). *Proceedings of the Royal Society of London*  
512 *B* 275:1047-1052

513 Schradin C (2008b) Seasonal changes in testosterone and corticosterone levels in four social  
514 categories of a desert dwelling sociable rodent. *Horm & Behav* 53:573-579

515 Schradin C (2008c) Seasonal changes in testosterone and corticosterone levels in four social  
516 classes of a desert dwelling sociable rodent. *Hormones and Behavior* 53:573-579. doi:  
517 10.1016/j.yhbeh.2008.01.003

518 Schradin C, König B, Pillay N (2010) Reproductive competition favours solitary living while  
519 ecological constraints impose group-living in African striped mice. *Journal of Animal*  
520 *Ecology* 79:515-521

521 Schradin C et al. (2012) Social flexibility and social evolution in mammals: a case study of the  
522 African striped mouse (*Rhabdomys pumilio*). *Molecular Ecology* 21:541-553. doi:  
523 10.1111/j.1365-294X.2011.05256.x

524 Schradin C, Pillay N (2005) Demography of the striped mouse (*Rhabdomys pumilio*) in the  
525 succulent karoo: a unique population in an extreme environment. *Mammalian Biology*  
526 70:84-92

527 Schradin C, Pillay N, Bertelsmeier C (2019) Social flexibility and environmental unpredictability in  
528 African striped mice. *Behavioral Ecology and Sociobiology* 73:94

529 Schradin C, Raynaud J, Arrivé M, Blanc S (2014) Leptin levels in free ranging striped mice  
530 (*Rhabdomys pumilio*) increase when food decreases: the ecological leptin hypothesis.  
531 *General and Comparative Endocrinology* 206:139-145. doi: 10.1016/j.ygcen.2014.06.024

532 Seeker LA et al. (2018) Longitudinal changes in telomere length and associated genetic  
533 parameters in dairy cattle analysed using random regression models. *Plos One*  
534 13:e0192864. doi: 10.1371/journal.pone.0192864

535 Seluanov A et al. (2007) Telomerase activity coevolves with body mass, not lifespan. *Aging Cell*  
536 6:45-52

537 Simons MJP (2015) Questioning causal involvement of telomeres in aging. *Ageing Research*  
538 *Reviews*. doi: 10.1016/j.arr.2015.08.002

539 Simons MJP, Stulp G, Nakagawa S (2014) A statistical approach to distinguish telomere  
540 elongation from error in longitudinal datasets. *Biogerontology* 15:99-103. doi:  
541 10.1007/s10522-013-9471-2

542 Smith S, Turbill C, Penn DJ (2011) Chasing telomeres, not red herrings, in evolutionary ecology.  
543 Heredity 107:372-373. doi: 10.1038/hdy.2011.14

544 Spurgin LG et al. (2018) Spatio-temporal variation in lifelong telomere dynamics in a long-term  
545 ecological study. *Journal of Animal Ecology* 87:187-198. doi: 10.1111/1365-2656.12741

546 Steenstrup T, Hjelmborg JvB, Kark JD, Christensen K, Aviv A (2013) The telomere lengthening  
547 conundrum--artifact or biology? *Nucleic Acids Research* 41:e131-e131. doi:  
548 10.1093/nar/gkt370

549 Stier A et al. (2014) Starting with a handicap: phenotypic differences between early- and late-  
550 born king penguin chicks and their survival correlates. *Functional Ecology* 28:601-611.  
551 doi: 10.1111/1365-2435.12204

552 Tian X et al. (2018) Evolution of telomere maintenance and tumour suppressor mechanisms  
553 across mammals. *Philosophical Transactions of the Royal Society B: Biological Sciences*  
554 373:20160443. doi: 10.1098/rstb.2016.0443 10.6084/m9

555 Tissier ML, Williams TD, Criscuolo F (2014) Maternal Effects Underlie Ageing Costs of Growth in  
556 the Zebra Finch (*Taeniopygia guttata*). *Plos One* 9:e97705. doi:  
557 10.1371/journal.pone.0097705

558 Turbill C, Ruf T, Smith S, Bieber C (2013) Seasonal variation in telomere length of a hibernating  
559 rodent. *Biology Letters* 9:20121095-20121095. doi: 10.1098/rsbl.2012.1095

560 Turbill C, Smith S, Deimel C, Ruf T (2012) Daily torpor is associated with telomere length change  
561 over winter in Djungarian hamsters. *Biology Letters* 8:304-307. doi:  
562 10.1098/rsbl.2011.0758

563 Vera E, Bernardes de Jesus B, Foronda M, Flores Juana M, Blasco Maria A (2012) The Rate of  
564 Increase of Short Telomeres Predicts Longevity in Mammals. *Cell Reports* 2:732-737. doi:  
565 10.1016/j.celrep.2012.08.023

566 Verhulst S, Aviv A, Benetos A, Berenson GS, Kark JD (2013) Do leukocyte telomere length  
567 dynamics depend on baseline telomere length? An analysis that corrects for 'regression  
568 to the mean'. *European Journal of Epidemiology* 28:859-866. doi: 10.1007/s10654-013-  
569 9845-4

570 Viblanc VA et al. (2018) Maternal oxidative stress and reproduction: Testing the constraint, cost  
571 and shielding hypotheses in a wild mammal. *Functional Ecology* 32:722-735. doi:  
572 10.1111/1365-2435.13032

573 Vitikainen EIK et al. (2016) Evidence of Oxidative Shielding of Offspring in a Wild Mammal.  
574 *Frontiers in Ecology and Evolution* 4. doi: 10.3389/fevo.2016.00058

575 Vuarin P, Pillay N, Schradin C (2019) Elevated basal corticosterone levels increase disappearance  
576 risk of light but not heavy individuals in a long-term monitored rodent population.  
577 *Hormones and Behavior* 113:95-102

578 Whitemore K, Vera E, Martínez-Nevado E, Sanpera C, Blasco MA (2019) Telomere shortening  
579 rate predicts species life span. *Proceedings of the National Academy of Sciences*  
580 116:15122-15127. doi: 10.1073/pnas.1902452116

581 Wolak ME, Fairbairn DJ, Paulsen YR (2012) Guidelines for estimating repeatability. *Methods in*  
582 *Ecology and Evolution* 3:129-137. doi: 10.1111/j.2041-210X.2011.00125.x

583 Young AJ (2018) The role of telomeres in the mechanisms and evolution of life-history trade-offs  
584 and ageing. *Philosophical Transactions of the Royal Society B: Biological Sciences*  
585 373:20160452. doi: 10.1098/rstb.2016.0452

586

587

588