

## Supplemental Information for:

### When does early-life telomere length predict survival? A case study and meta-analysis

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## 1. Supplementary tables

**Table S1: relative telomere length (TL) was not associated with survival from fledging to independence (model 1).** Results of a generalised linear model with survival as the dependent variable (yes = 1, no = 0).

Model	Parameter	Estimate	Standard error	Statistic	P	N died	N survived
GLM	Intercept	0.890	0.196	4.543	<0.001	70	206
	Relative TL	0.006	0.140	0.045	0.964		
	Sex (male)	0.367	0.279	1.317	0.188		

**Table S2: relative telomere length (TL) was not associated with survival from independence to maturity (including late fledged males and females, model 2).** Results of a generalised linear model with survival as the dependent variable (yes = 1, no = 0).

Model	Parameter	Estimate	Standard error	Statistic	P	N died	N survived
GLM	Intercept	-0.078	0.297	-0.262	0.794	41	68
	Relative TL	0.057	0.208	0.274	0.784		
	Sex (male)	1.067	0.413	2.583	0.010		

**Table S3: relative telomere length (TL) was not associated with survival from independence to maturity (including all males hatched at all times, model 3).** Results of a generalised linear model with survival as the dependent variable (yes = 1, no = 0).

Model	Parameter	Estimate	Standard error	Statistic	P	N died	N survived
GLM	Intercept	1.105	0.223	4.945	<0.001	27	81
	Relative TL	0.158	0.228	0.694	0.488		

**Table S4: relative telomere length (TL) was not associated with survival from maturity to death (including breeding males and females, model 4).** Breeding status (zero offspring = 0, greater than zero offspring = 1) was included as a fixed factor to control for social status. An interaction between TL and breeding status was removed from the final model because the effect was not statistically significant. Dataset included 18 females and 47 males.

Model	Parameter	Estimate	Standard error	Statistic	P	N died	N censored
CoxPH	Relative TL	0.022	0.133	0.166	0.868	60	2
	Sex (male)	0.017	0.311	0.056	0.955		

**Table S5: relative telomere length (TL) was not associated with survival from maturity to death within males only (including all males that survived to maturity regardless of breeding status, model 5).** Breeding status (zero offspring = 0, greater than zero offspring = 1) was included as a fixed factor to control for social status. An interaction between TL and breeding status was removed from the final model because the effect was not statistically significant. Dataset included 40 subordinate helpers and 45 dominant breeders.

Model	Parameter	Estimate	Standard error	Statistic	P	N died	N censored
CoxPH	Relative TL	0.129	0.113	1.149	0.250	81	4
	Breeding status (breeding)	-1.253	0.242	-5.183	<0.001		

**Table S6: relative telomere length (TL) was not associated with lifetime survival (included all males over all life-stages, model 6).** Breeding position (zero offspring = 0, greater than zero offspring = 1) as a fixed factor to control for social status. An interaction between TL and breeding status was removed from the final model because the effect was not statistically significant. Dataset included 104 subordinate helpers and 45 dominant breeders.

Model	Parameter	Estimate	Standard error	Statistic	P	N died	N censored
CoxPH	Relative TL	0.140	0.091	1.545	0.122	140	9
	Breeding status (breeding)	-1.633	0.207	-7.884	<0.001		

**Table S7: the intercept only model (null model) including the variance components study ID, effect ID and species ID, shows an overall negative association between early-life telomere length and mortality.** There was high heterogeneity between studies ( $I^2 = 88.57\%$ ).

Fixed effects	Estimate	Std. error	Z value	P value	Lower CI	Upper CI
Intercept	-0.164	0.065	-2.524	0.012	-0.291	-0.037
Random effects	Estimate	$I^2$				
Between study variance (study ID)	0.018	26.606				
Within study variance (effect ID)	0.003	4.993				
Species variance (species ID)	0.039	56.967				

$Q_{(d.f. = 31)} = 133.872$ ,  $p < 0.001$ ;  $AIC_c = 27.488$ .

**Table S8: phylogeny explained only a small proportion of variation when included into the intercept only model with the variance components study ID, effect ID and species ID (phylogenetic model).** Heterogeneity was similar in the phylogenetic model ( $I^2 = 89.13\%$ ) and did not improve the model fit  $\Delta AIC_c = 2.812$  compared to null model.

Fixed effects	Estimate	Std. error	Z value	P value	Lower CI	Upper CI
Intercept	-0.173	0.076	-2.276	0.023	-0.323	-0.024
Random effects	Estimate	$I^2$				
Between study variance (study ID)	0.020	27.459				
Within study variance (effect ID)	0.004	4.893				
Species variance (species ID)	0.037	50.315				
Phylogenetic variance	0.005	6.465				

$Q_{(d.f. = 31)} = 133.872$ ,  $p < 0.001$ ;  $AIC_c = 30.300$ .

**Table S9: No relationship between telomere length and the mortality period measured, but effect sizes are impacted by method and year of publication.** Likewise, maximum lifespan (mean corrected) had no significant effect. Publication year and telomere measurement method (full model;  $I^2 = 82.4\%$ ). For the mortality period measured the reference category is 'growth to independence' and for method the reference category is 'qPCR'. Species ID was excluded because it explained zero variance after including the moderator variables. Lifespan was also substituted with  $\log_{10}(\text{lifespan})$  to test for a non-linear effect but was not significant (estimate = -0.362, se = 0.297, z = -1.220, P = 0.222).

Fixed effects	Estimate	Std. error	Z value	P value	Lower CI	Upper CI
Intercept	-0.231	0.093	-2.477	0.013	-0.414	-0.048
Mortality period measured						
Growth/independence to adulthood	-0.036	0.104	-0.351	0.726	-0.239	0.167
Post-maturity	-0.030	0.124	-0.240	0.811	-0.274	0.214
Lifetime	0.022	0.117	0.185	0.853	-0.208	0.252
Method						
TRF	0.511	0.181	2.821	0.005	0.156	0.866
Maximum lifespan (mean centred)	-0.016	0.010	-1.635	0.102	-0.036	0.003
Publication Year (mean centred)	0.070	0.020	3.429	<0.001	0.030	0.109
Random effects	Estimate	$I^2$				
Between study variance (study ID)	0.040	63.609				
Within study variance (effect ID)	0.012	18.789				
Species variance (species ID)	0	0				

$QE_{(d.f. = 25)} = 92.860$ ,  $p < 0.001$ ;  $QM_{(d.f. = 6)} = 15.612$ ,  $p = 0.016$ ;  $AIC_c = 39.48$ .

**Table S10: across all studies including information on sex, the relationship between telomere length and mortality was stronger in females than males, although the confidence intervals overlap.** In both models, effect ID explained zero variance, however, due to the overlapping of the variance explained, either study or species ID explained zero variation in males or females.

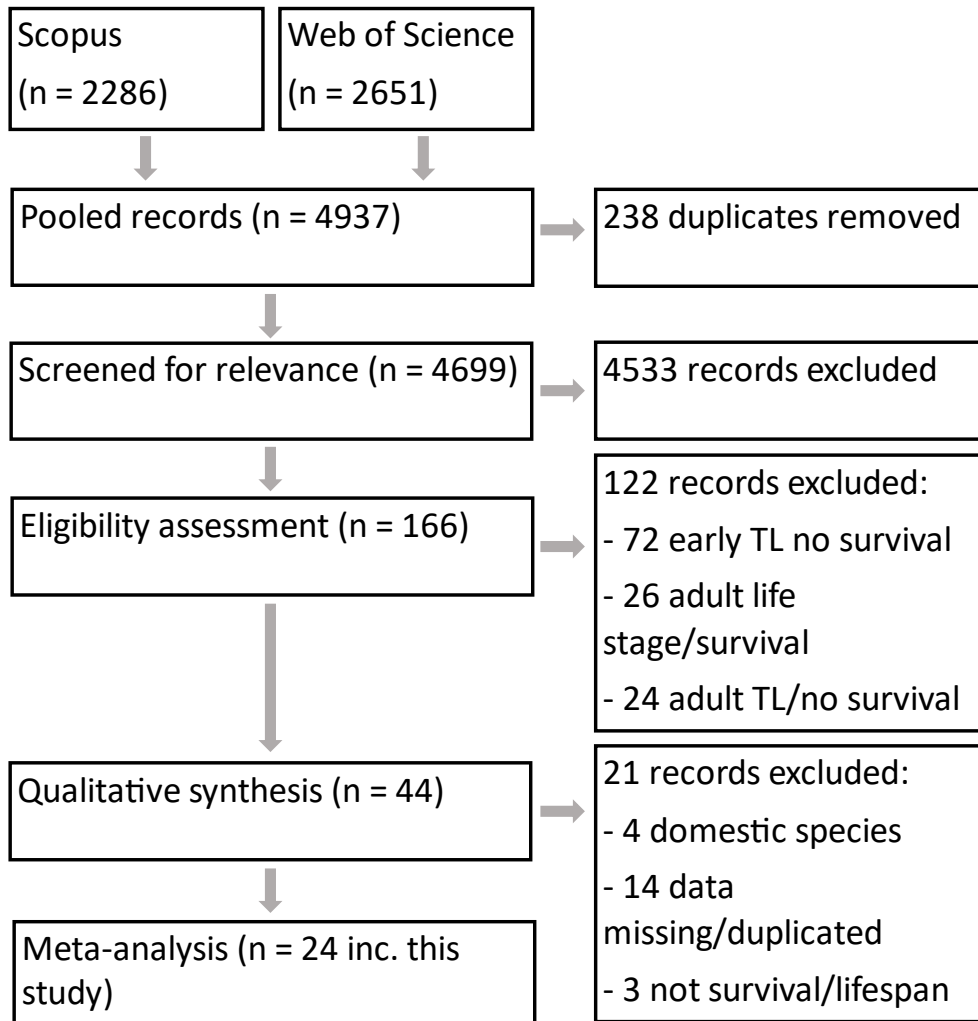
<b>Females</b>						
<b>Fixed effects</b>	<b>Estimate</b>	<b>Std. error</b>	<b>Z value</b>	<b>P value</b>	<b>Lower CI</b>	<b>Upper CI</b>
Intercept	-0.112	0.063	-1.895	0.058	-0.241	0.004
<b>Random effects</b>	<b>Estimate</b>	<b>I<sup>2</sup></b>				
Between study variance (study ID)	0.051	76.243				
Within study variance (effect ID)	0	0				
Species variance (species ID)	0	0				
<b>Males</b>						
<b>Fixed effects</b>	<b>Estimate</b>	<b>Std. error</b>	<b>Z value</b>	<b>P value</b>	<b>Lower CI</b>	<b>Upper CI</b>
Intercept	-0.068	0.043	-1.593	0.111	-0.152	0.016
<b>Random effects</b>	<b>Estimate</b>	<b>I<sup>2</sup></b>				
Between study variance (study ID)	0	0				
Within study variance (effect ID)	0	0				
Species variance (species ID)	0.013	48.501				

**Table S11: Bayesian mixed effect model assessment to test whether there is evidence of publication bias and the potential effects on the overall estimate.** We used a mixed effect modelling approach implemented in a Bayesian framework with the *MCMCglmm* package (Hadfield, 2010). Random effects included study ID (between study effects), species (species replication between studies) and phylogenetic relatedness (to account for evolutionary non-independence of species included in each study; see methods for details). First, we included an intercept only model (null model) to replicate the *metafor* analysis in the main text and to estimate the change in the overall effect when accounting for publication bias (Trim-and-fill method). Model chains were run for 5005000 iterations with a thin of 500 and a burn-in of 1000. For the residuals and random terms we used relatively un-informative prior settings (G: V = 1, nu = 1, alpha.mu = 0, alpha.V = 1000; R: V = 1, nu = 0.002). Fixed effect priors scaled to have a mean of zero and standard deviation of one (default priors). To obtain how the meta-analytic effect may change without publication bias we performed a Trim-and-fill analysis using both the LO and RO estimators. Using the same set of priors above, we then performed a full model including the fixed covariates: telomere measurement method, mortality period, maximum lifespan, and publication year. Here, we found that under a Bayesian framework, the estimates of each of the moderators were comparable to the *metafor* analysis presented in the manuscript.

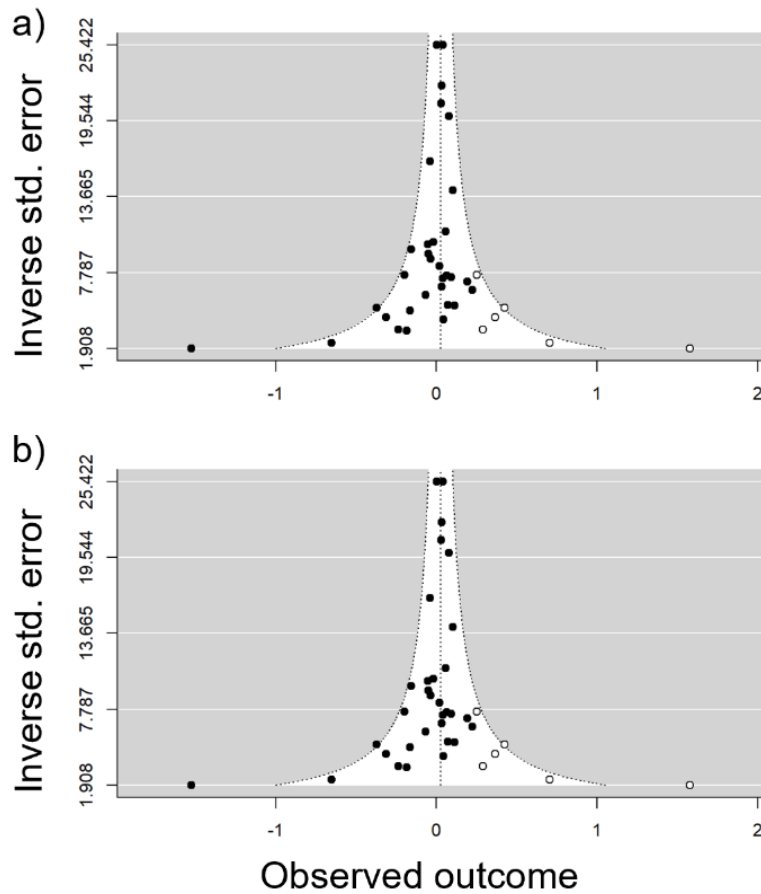
<b>Null model</b>				
<b>Fixed effects</b>	<b>Estimate</b>	<b>Lower CI</b>	<b>Upper CI</b>	<b>P</b>
Intercept	-0.203	-0.550	0.152	0.154
<b>Random effects</b>	<b>Estimate</b>	<b>Lower CI</b>	<b>Upper CI</b>	<b>I<sup>2</sup></b>
Between study variance (study ID)	0.044	1.039×10 <sup>-9</sup>	0.132	28.25 %
Species variance (species ID)	0.043	5.870×10 <sup>-11</sup>	0.131	28.22 %
Animal	0.072	4.624×10 <sup>-10</sup>	0.287	29.41 %
Total I <sup>2</sup>				93.50 %
<b>Full model</b>				
<b>Fixed effects</b>	<b>Estimate</b>	<b>Lower CI</b>	<b>Upper CI</b>	<b>P</b>
Intercept	-0.229	-0.599	0.113	0.148
Mortality period measured				
Growth/independence to adulthood	-0.084	-0.300	0.149	0.435
Post-maturity	-0.039	-0.285	0.215	0.755
Lifetime	0.004	-0.244	0.261	0.978
<b>Method</b>				
<b>TRF</b>	<b>0.518</b>	<b>0.076</b>	<b>0.990</b>	<b>0.023</b>
Maximum lifespan (mean corrected)	-0.012	-0.044	0.018	0.416
<b>Publication year</b>	<b>0.070</b>	<b>0.019</b>	<b>0.122</b>	<b>0.008</b>
<b>Random effects</b>	<b>Estimate</b>	<b>Lower CI</b>	<b>Upper CI</b>	<b>I<sup>2</sup></b>
Between study variance (study ID)	0.032	8.764×10 <sup>-11</sup>	0.102	25.01 %
Species variance (species ID)	0.033	5.151×10 <sup>-10</sup>	0.107	25.66 %
Animal	0.061	1.486×10 <sup>-11</sup>	0.245	29.49 %
Total I <sup>2</sup>				92.12 %



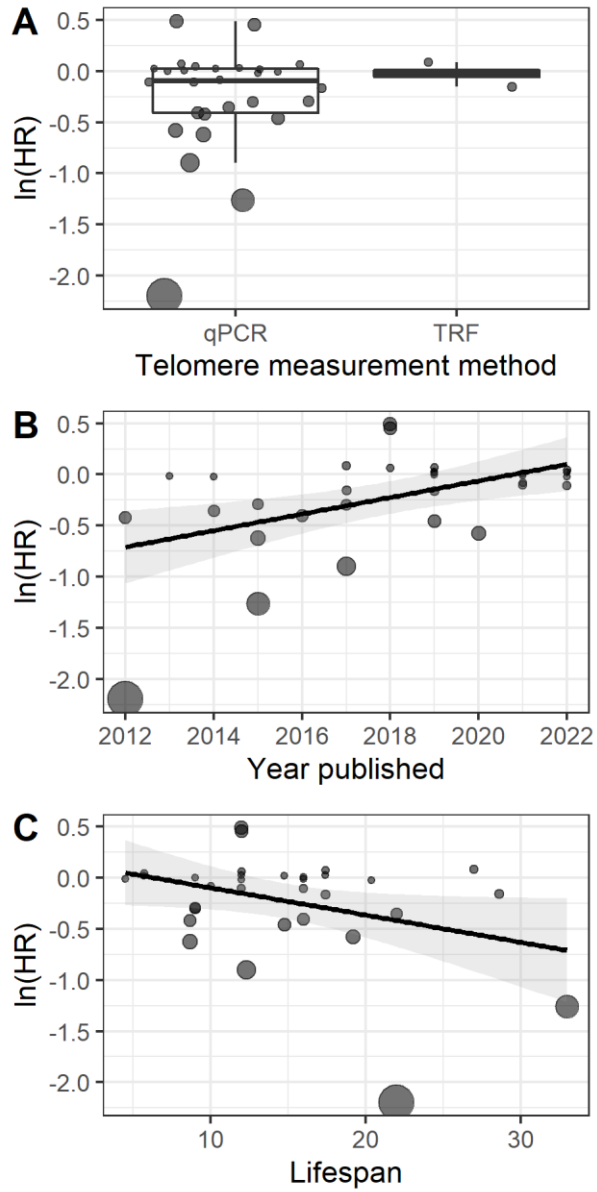
## 2. Supplementary figures



**Figure S1:** Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) flow chart including the number of records and reasons for exclusion at each step. Literature search: 01 February 2022, Monash University library.



**Figure S2: Trim-and-fill applied to the Bayesian mixed effect modelling described in Table S11.** A) the estimator L0 identified six missing studies on the right side. B) the estimator R0 identified the same six studies on the right side as L0.



**Figure S3: Mortality risk ( $\ln(\text{HR})$ ) effect sizes plotted for each meta-regression moderator.** (a) telomere measurement method (qPCR vs TRF), (b) year of publication, and (c) species maximum lifespan. Plot of non-adjusted estimates not accounting for study non-independence. Regression lines are plotted for visualisation purposes using the raw data. The size of the points indicates standard error.

### 3. References

Hadfield, J. D. (2010). MCMC Methods for Multi-Response Generalized Linear Mixed Models:

The MCMCglmm R Package. *Journal of Statistical Software*, 33, 1–22. doi:

10.18637/jss.v033.i02