Climate change, age acceleration, and the erosion of fitness in polar bears

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Abstract

Climate change is increasingly disrupting evolved life history strategies and decreasing population viability in wild species[1]. The magnitude and pace at which environments will change mean the persistence of wild populations will depend substantially on their ability to adapt genetically. However, we know very little about the capacity for evolutionary change in response to climate warming. We mapped the effects of climate change, beginning with the decline of cellular function through to the erosion of fitness and adaptive potential in an intensively studied polar bear (*Ursus maritimus*) population in western Hudson Bay, Canada. Using estimates of epigenetic age acceleration, an indicator of declining cellular function associated with exposure to stress[2], we found that polar bears aged approximately one year faster, on average, for each degree Celsius temperature increase they experienced. Declining cellular function should reduce fitness[3,4] and counter adaptive evolution in rapidly changing environments. Individuals who reproduced early had higher lifetime reproductive success; however, this was before the onset of rapid warming. Fitness benefits associated with early reproduction declined with warming, and today, bears have similar lifetime reproductive success regardless of when they first reproduce. Finally, using a large pedigree[5], we found no evidence for genetic variation associated with reproductive success in this population—the population is not evolving in response to the changing environment. The physiological costs of climate change accumulate across lifetimes to degrade cellular function and, ultimately, adaptive capacity. These findings warn that adaptive responses to warming could be the exception rather than the rule.

Main

Climate change is causing extreme environmental fluctuations and persistent warming, exposing species to conditions increasingly distant from those they evolved to tolerate[7]. Species have typically responded to environmental change through range shifts[8], changes in seasonally timed behaviours[9], and population declines[10]. However, the accumulation of our past emissions and current emission targets commits the planet to ongoing warming for the foreseeable future[11]. Continued long-term environmental change means the persistence of many species will ultimately rely on their capacity to adapt genetically to the changed environment. However, the extent to which populations can respond evolutionarily to the rapid pace of climate warming is poorly understood.

We document the effects of more than a half-century of climate change on an intensively studied polar bear (*Ursus maritimus*) population, from the progressive degradation of cellular processes to the erosion of fitness and adaptive potential. Using modern biomedical techniques, we first tested for epigenetic age acceleration, an estimate of the decline in cellular function associated with an individual's cumulative experience of stress across their lifetime. Faster aging is associated with deteriorating metabolic and physiological processes important for cell maintenance and function[3]. Deteriorating cellular function should eventually reduce survival and fecundity and may eventually counter adaptive evolution in response to the changing environment, so we next tested for links between epigenetic age acceleration in individuals and the erosion of fitness and adaptive potential at the population level (Figure 1). Our results offer a window into whether we might expect widespread adaptive change by wildlife populations to levels of warming that will soon be reached globally. The Arctic has warmed approximately four times faster than the global mean and is now 3°C warmer, on average, than it was at the onset of

63 rapid warming[12]. If current emission targets are met, the rest of the planet will warm by ~3°C by
64 the end of the century[13].

65 The western Hudson Bay polar bear population is at the southern edge of the Arctic and
66 has been subject to standardized annual sampling and individual-based monitoring since 1980.
67 Polar bears rely on sea ice for travel and mating and as a platform to hunt their primary prey,
68 ringed (*Pusa hispida*) and bearded (*Erignathus barbatus*) seals. After sea ice retreats in spring,
69 their prey becomes inaccessible, and polar bears must fast, relying on accumulated fat reserves
70 for growth, reproduction, and survival[14]. As a result of air and sea surface temperature anomalies
71 associated with warming[15], the ice-free season in the Hudson Bay region has lengthened by
72 approximately ten days per decade since the early 1980s[16,17]. During this time the western
73 Hudson Bay polar bear population has declined by nearly 50% to its current estimated size of
74 just over 600 individuals[18]. Sea ice loss is firmly tied to this decline and is the most significant
75 climate-related threat to this population[16,17,19–21]. Longer ice-free seasons increase the bears'
76 fasting period on land, and each additional day of fasting requires metabolizing approximately
77 one kilogram of body mass[22]. Bears also risk losing stored body mass as thinning winter ice and
78 rapid spring melts force longer swims between ice floes. Swimming is five times more
79 energetically expensive for bears than walking, and dramatically longer swims are required for
80 even small changes in sea ice[23].

81 **Increase in age acceleration with climate warming**

82 In 1956, Hans Selye observed that 'Every stress leaves an indelible scar, and the organism pays
83 for its survival after a stressful situation by becoming a little older'[24]. Biomedical research has
84 since established that an organism's cumulative experience of stressful environmental conditions
85 is reflected in its cellular aging rate or biological age. When an organism experiences excess

86 stress, molecular wear and tear cause its cellular function to decline, making its cells appear
87 biologically older than their chronological age would suggest. This phenomenon is biological
88 age acceleration[25,26]. If the accumulated experience of environmental stressors associated with
89 climate warming causes declines in cellular function, there should eventually be declines in
90 survival and fecundity that affect a population's capacity to adapt to the changing environment.
91 We measured biological age acceleration in western Hudson Bay polar bears using an epigenetic
92 approach recently developed in biomedicine[3,28,29]. Epigenetic age acceleration is estimated using
93 the residual difference between chronological and predicted biological age[27]. Epigenetic age
94 acceleration is currently the most accurate biomedical measure of cumulative experiences of
95 environmental stressors across lifetimes[26], increasing, for example, in response to smoking[28] and
96 early life adversity[30]. It is also the best biomarker of morbidity and all-cause mortality in
97 humans[3,4].

98 Epigenetic age is measured through DNA methylation, a process in which methyl groups
99 are attached to DNA molecules at cytosine guanine dinucleotides (CpG or CG sites)[25]. This
100 process plays a role in cell fate and gene regulation[31]. DNA methylation patterns at some CpG
101 sites change so predictably across lifespans that they can be used to build "epigenetic clocks" for
102 predicting chronological age in humans[27], mouse models[32], and many other mammals[33–38].
103 Epigenetic clocks predict chronological age two- to three-fold more accurately than earlier
104 biological aging approaches such as telomere shortening[39]. Critical for our purposes, epigenetic
105 age acceleration can be estimated from archived tissue sampled from individuals with known
106 chronological ages. These samples are available for the western Hudson Bay population. This
107 methodological advance provided a lens with which we could view changes in the accumulation
108 of stress across lifetimes and the associated deterioration of cellular function in polar bears

sampled across 40 years, starting before the onset of significant climate warming through to the recent period of rapid warming.

We built an epigenetic clock for polar bears with archived tissue samples (Methods – *Building the epigenetic clock for polar bears*). Our clock (Supplemental Data File 1) is based on blood and skin tissue DNA methylation rates from 144 male and female individuals evenly sampled across ages 0–30 between 1988–2016 (Supplemental Data File 2; Methods – *Field data collection*). We used elastic net regression and epigenome-wide association surveys to identify 125 CpG sites in the polar bear genome where DNA methylation is strongly associated with chronological age but unrelated to sex (Extended Data Figure 1). We narrowed these sites down from an initial 33,674 candidate CpG sites on the mammalian DNA methylation array[40] that align to the polar bear genome (Supplemental Data File 3). Our clock accurately tracked epigenetic age over the lifetimes of bears with repeat samples (Figure 2 A-E) and predicted the chronological ages of an independent sample of 134 test bears within a median absolute error of 2.07 years (Figure 2 F), or approximately 5% of the maximum polar bear lifespan[41]. This performance is on par with the most accurate epigenetic clocks built for humans and other wildlife[27,34,35]. We used our epigenetic clock to estimate epigenetic age acceleration through time for the 134 test bears not used for clock development.

We found a signal of epigenetic age acceleration with time that paralleled climatic warming and lengthening ice-free periods (Table 1; Figure 3). This suggests the accumulation of environmental stressors experienced by western Hudson Bay polar bears is associated with deteriorating cellular function through time. Polar bears born more recently aged faster as the climate warmed, and the absence of a significant statistical interaction between birth year and sex in our model suggests both males and females responded similarly (Table 1; Figure 3). A

causal role of sea ice loss leading to decreased cell function is supported by a spike in epigenetic age acceleration in 1990 (Figure 3). This spike coincides with one of the earliest spring melts on record in western Hudson Bay and one of the largest observed declines in polar bear survival[31]. On average, bears born in the 2010s were biologically 2.6 years older than bears born in the 1960s (Figure 3). Considering a 30-year lifespan, near the maximum for this population[42], a 2.6-year increase in epigenetic age is equivalent to an 8.7% increase in aging rate. Because of our sampling approach, however, this increase is likely a conservative estimate. We evenly selected samples from individuals across age classes (Methods – *Building the epigenetic clock for polar bears*), but polar bear survival declines significantly as they approach their early 20s[42]. Age acceleration is associated with increased morbidity and mortality[3,4], so the 20–30-year-old bears available to sample were likely among the healthiest of their cohort. This suggests we oversampled healthy bears and underestimated age acceleration.

Evolutionary change and adaptive potential

The decline of processes important for cellular function and maintenance through time should eventually erode fitness[2]. This could dampen or counter adaptive responses to the changing environments. To test for evidence of the erosion of fitness in association with age acceleration and the warming environment, we explored changes in resource allocation and the costs and timing of reproduction[43,44]. Individuals take energy and nutrients from the environment and allocate them to self-maintenance and reproduction. Resources devoted to one area are not immediately available for the other, meaning the optimal allocation of energy to self-maintenance and reproduction depends on the ecological setting[44]. Reproduction is expensive, so reproducing early in life is expected to come at the expense of self-maintenance and longevity[43] which should be detectable as accelerated aging. Indeed, we observed a weak correlation

between age acceleration and age at first reproduction for 100 western Hudson Bay polar bears with estimates of both age acceleration and age at first reproduction ($R^2_m = 0.12$; Table 1; Extended Data Figure 2).

When environments are variable and survival is uncertain, reproduction late in life is also uncertain, so theory predicts selection for investing energy in reproducing earlier in life[43]. We could estimate lifetime reproductive success, a common measure of fitness, for 628 individuals from the long-term study (Methods – *Estimating life history traits*). We found that in the 1980s, early reproducing bears had the highest lifetime reproductive success, suggesting that reproducing early was indeed adaptive before the onset of high levels of warming (Table 1). However, the fitness advantage of reproducing early in life declined through the 1990s. By 2000, bears produced the same number of offspring over their lifetimes regardless of how young they were when they first reproduced (Figure 4 A). The fitness advantage of reproducing early in life has eroded with time in association with warming and declining cellular function.

Finally, building from the links between cellular decline and increased lifetime stress, reduced fitness, and the warming climate, we explored the overall rate of recent adaptive evolution in the western Hudson Bay polar bear population. All credible climate forecasts project ongoing warming for the foreseeable future. This means that population persistence will depend on a population's overall capacity to adapt genetically to the changing environment. We used our estimates of lifetime reproductive success and a population pedigree that spanned the 40-year study[5] to test for genetic differences among individuals in their ability to reproduce (i.e., the contribution of selection to the genetically based increase in population mean fitness). We did so by estimating the additive genetic variance in individual relative fitness, which can be thought of as an estimate of the overall effect of selection on the ability of individuals to reproduce[45]. Fitting

178 an animal model[46] with the 4634-individual pedigree (923 dams, 443 sires), we found the
179 additive genetic variance underlying lifetime reproductive success in this population is
180 approximately zero (V_A(w) = 0.006; Extended Data Table 1). This suggests selection on traits
181 related to individual fitness is not contributing to adaptive change in this population. This lack of
182 evidence for adaptive change is consistent with observed low survival rates and population
183 declines [18], patterns expected for populations experiencing substantial environmental change
184 with limited capacity to respond adaptively[47]. In western Hudson Bay, adaptive evolution in
185 polar bears appears to be countered by the negative effects of the rapidly changing environment.
186 Without gene flow from differently adapted populations or changes in selective regimes, this
187 population likely has minimal adaptive capacity to cope with the ongoing long-term
188 environmental change associated with climate warming.

189 **Conclusions**

190 We mapped the process by which climate warming negatively affects wild populations from
191 deteriorating cellular function within individuals across their lifetimes to fitness losses and the
192 erosion of adaptive potential. Age acceleration reflects an individual's cumulative lifetime
193 experiences with its environment[3]. As the climate warms, there are good and bad years, and thus
194 good and bad environmental experiences. Age acceleration captures both[26]. Regardless of
195 interannual variation, the net consequence of warming for polar bears across their lifetimes is the
196 deterioration of cellular function. With increasing exposure to harsh environments, declines in
197 abundance, and little evidence for adaptive capacity, the western Hudson Bay polar bear
198 population faces a uncertain future.

199 The increased cumulative experience of stress across lifetimes and the erosion of fitness
200 and adaptive potential in association with warming are instructive for understanding whether and

how other populations might respond adaptively to environmental change in the coming decades. Warming in the Arctic has abruptly altered the ecosystem, and this abruptness outpaced the adaptive capacity of western Hudson Bay polar bears. Recent modelling suggests that similarly abrupt exposures to intolerable temperatures across large portions of species' ranges could be widespread in the coming decades[48]. At the current rate of warming, more than 30% of species could be exposed to temperatures beyond those they evolved to tolerate by 2100[48,49]. As species pass their thermal thresholds, the capacity for populations and ecosystems to adapt will diminish. With the recent signing of the United Nations Montreal-Kunming Global Biodiversity Framework, monitoring and conserving adaptive potential has become mandated in international policy. This is a major positive step toward safeguarding biodiversity given ongoing climate change and other threats. However, our results, and forecasts of future species exposure to warming[48,49], warn that adaptive responses to warming could be the exception rather than the rule. Significant conservation efforts on all fronts and concerted efforts to halt warming will be necessary to safeguard biodiversity for future generations.

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Table 1. Four decades of warming is linked to epigenetic aging and fitness declines in an intensively studied polar bear (*Ursus maritimus*) population in western Hudson Bay, Canada. Bears born more recently and those that reproduce earlier in life age faster epigenetically. The fitness benefit of earlier reproduction, estimated using lifetime reproductive success, declined for later-born bears. We report the fixed effect covariates and their coefficients from Bayesian generalized linear models testing relationships between epigenetic age acceleration, year of birth, age at first reproduction, and lifetime reproductive success for male and female polar bears. Probability of direction (P direction) describes the probability that a coefficient is either positive or negative, expressed as a percentage between 50% and 100%. Marginal R^2 (R^2_m) describes the proportion of variation in the response explained by the fixed effects relative to random effects, and conditional R^2 (R^2_c) describes the variation explained by fixed effects and random effects if included in the model. For all models, we used consevative weakly informative priors with mean = 0 and standard deviation = 1. We fit all models using the *brms* package v2.20.4 in R v4.3.1, with 4 chains and 10,000 iterations including 5,000 warmup iterations. Posterior predictive checks are in Extended Data Figure 3.

Variable	Covariates	Coefficient (95% CrI)	R^2m	R^2c	P direction
Age acceleration[§] (n = 134)	Year of birth	0.18 (-0.04, 0.40)	0.07	0.36	95
	Sex (M)	0.28 (-0.15, 0.71)			90
Age acceleration[§] (n = 100)	Age at first reproduction	-0.33 (-0.61, -0.03)	0.12	0.31	98
	Sex (M)	0.60 (0.07, 1.10)	-	-	98
Lifetime reproductive success[†] (n = 628)	Age at first reproduction	-0.16 (-0.22, -0.10)	-	0.15	100
	Year of birth	-0.34 (-0.43, -0.25)	-	-	100
	Sex (M)	-0.10 (-0.20, 0.01)		-	96
	Age at first reproduction: Year of birth	0.18 (0.08, 0.27)		-	100

[†]Models were specified using a negative binomial response distribution with a log link function.

[§]Models were specified using a Gaussian link function.

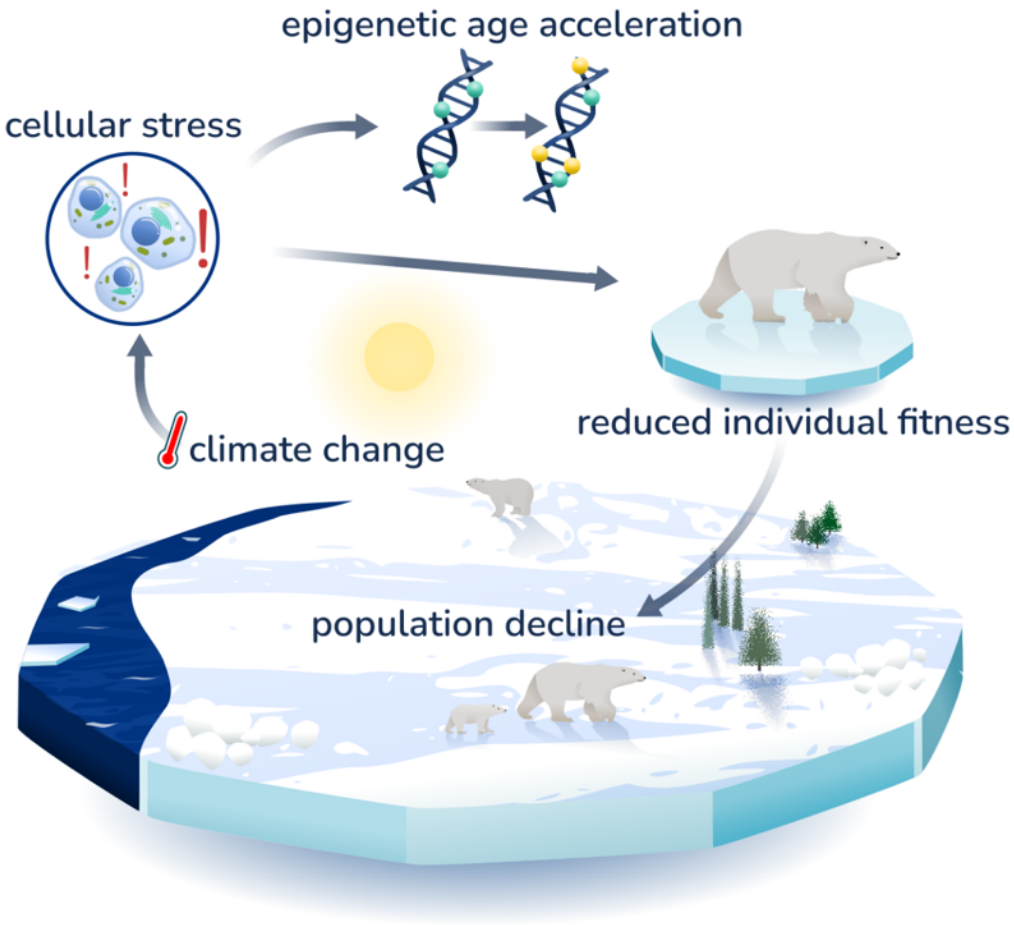

Figure 1. Epigenetic age acceleration predicts polar bear (*Ursus maritimus*) population declines caused by climate change. Global concerns over declining polar bear populations prompted formation of an international agreement on polar bear populations in the mid 1960s. Annual sampling and individual-based monitoring were standard in the western Hudson Bay subpopulation by 1980. Epigenetic aging rates accelerated over time with climate change (n = 134). Estimates of reproductive success for bears born between 1980 and 2000 (n = 628) suggest individual fitness has declined over time, coinciding with the erosion of advantages conferred by once-adaptive traits (n = 628) and a lack of adaptive capacity (n = 4,634). We linked declines in individual fitness to epigenetic age acceleration (n = 100).

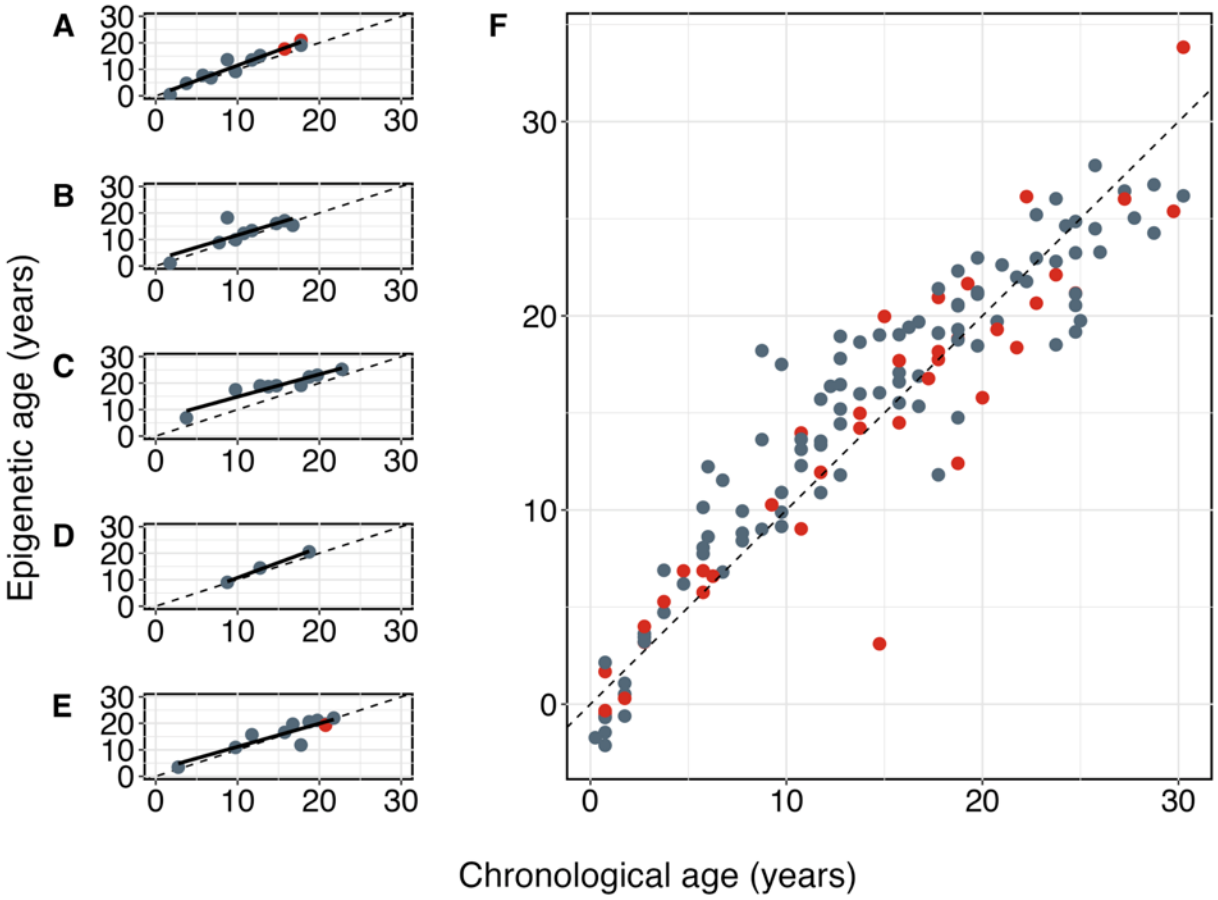

Figure 2. Our epigenetic clock for western Hudson Bay polar bears (*Ursus maritimus*) tracks the aging of bears repeatedly sampled over their lifetimes (A–E) and accurately predicts chronological age within a median absolute error of 2.07 years for n = 134 bears (F). Epigenetic clocks estimate age using patterns of methylation at cytosine guanine dinucleotides on DNA molecules. We built our clock with archived blood and skin tissue samples from 144 male and female individuals aged 0–30 sampled between 1988–2016. Red points represent blood samples and grey points represent skin. The dotted line is a guide for a 1:1 relationship between chronological and epigenetic age. Points above this line indicate age acceleration, i.e., individuals or samples that are epigenetically older than their chronological age.

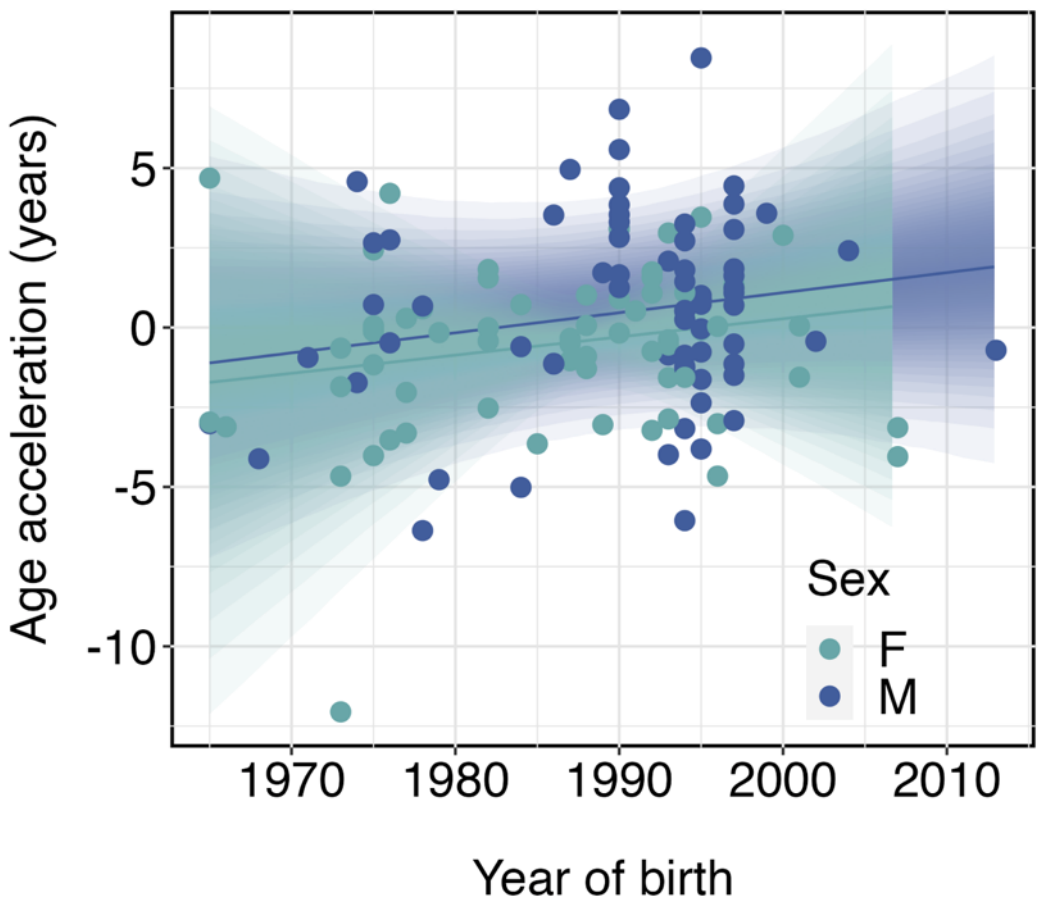

Figure 3. Epigenetic age acceleration, a cumulative measure of lifetime stressful exposures, has increased for both male (blue) and female (seafoam) polar bears (*Ursus maritimus*) in western Hudson Bay, Canada since annual monitoring began in the 1980s. Points represent measurements of epigenetic age acceleration from individuals born as early as 1965 and sampled between 1988 and 2016. The line and ribbon represent the mean and 95% credible interval around the posterior distributions, plotted separately and estimated using Bayesian generalized linear models.

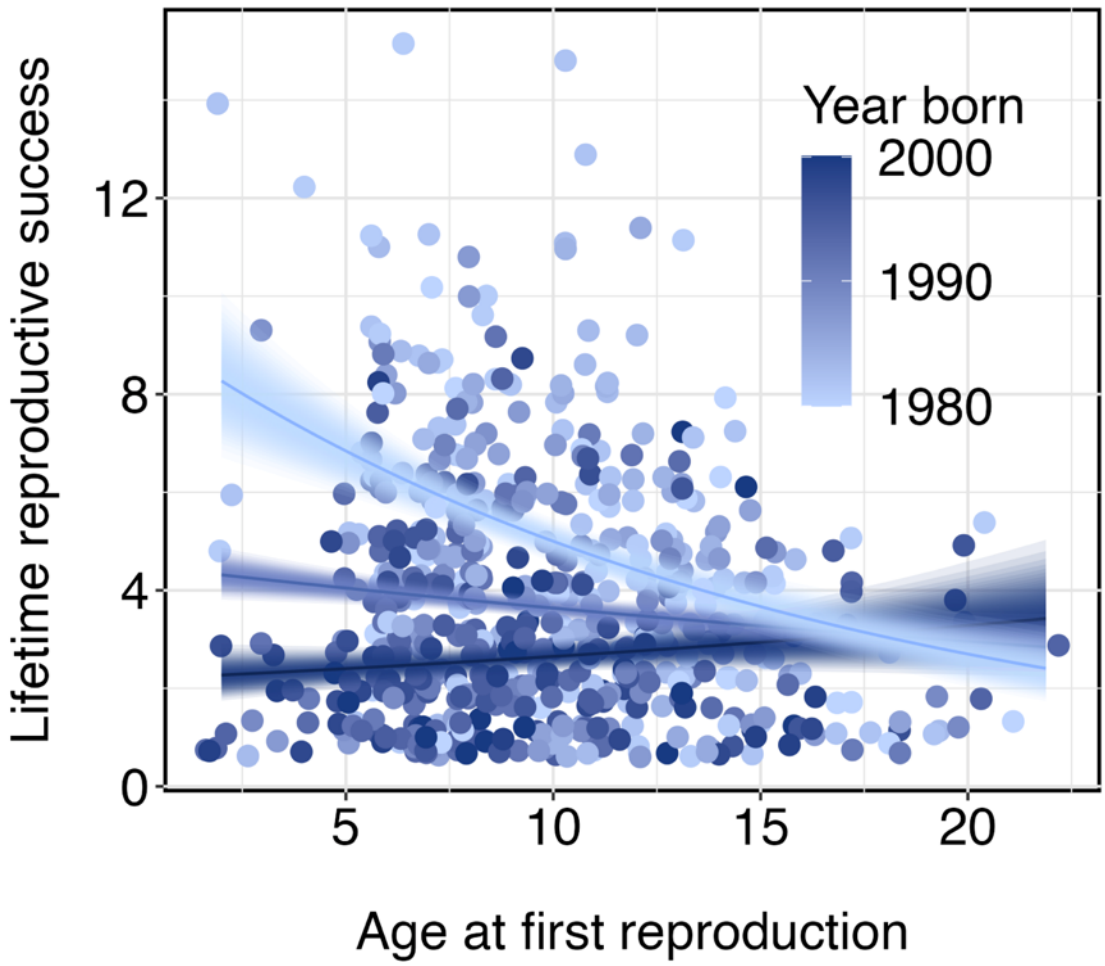

Figure 4. The fitness benefit of reproducing at younger ages eroded over two decades of climate change for n = 628 western Hudson Bay polar bears (*Ursus maritimus*). Reproducing at younger ages redirects energy from survival and self-maintenance, but theory predicts it can provide a fitness advantage in environments where reproduction is uncertain. In the 1980s when annual sampling began in this population (light blue), reproducing at younger ages increased fitness, which we estimated using lifetime reproductive success. Lifetime reproductive success declined for younger reproducers through the 1990s to 2000, at which point the fitness benefit of younger reproduction was lost (gradient from light to dark blue). Points show individual bear observations and lines are the predicted means of the posterior distributions in 1980, 1990, and 2000. Blue ribbons are 95% credible intervals.

Methods

Summary

We studied biological aging, fitness, and the capacity for adaptation in the western Hudson Bay polar bear (*Ursus maritimus*) population. Using a pedigree previously constructed for the population, we estimated age at first reproduction and lifetime reproductive success. We measured biological aging by first building an epigenetic clock for polar bears and then using it to gauge the rate at which the cells of individual bears from the population aged epigenetically relative to their chronological age. Using Bayesian generalized linear models, we tested for an increase in the rate of epigenetic aging over time, a relationship between age at first reproduction and the rate of epigenetic aging, and changes in lifetime reproductive success with age at first reproduction over time. Finally, we used an animal model, a type of mixed-effects model that estimates the additive genetic variance of traits like lifetime reproductive success[1], to estimate the capacity of western Hudson Bay polar bears to adapt to climate change.

Field data collection

Since 1966, polar bears have been captured in northeastern Manitoba near Churchill, Canada as part of a long-term study[2]. Bears are chemically immobilized, sexed, and fitted with unique ear tags and tattoos on the upper lip for later identification in case of recaptures. Skin samples are extracted either from pinna tissue remaining after ear-tagging or using a biopsy punch of superficial rump fat[2]. Blood samples are drawn from femoral blood into a sterile Vacutainer and stored at -80° C[2]. All capture and handling protocols are approved annually by Environment and Climate Change Canada's Prairie and Northern Region Animal Care Committee and wildlife research permits are issued by the Province of Manitoba and by Parks Canada. A standardized

sampling program was initiated in 1980 and continues today, with the exclusion of 1985 and 1986. As part of this program, adult females and their cubs of the year are sampled in February and March. Chronological age is either derived from known years of birth for cubs-of-the-year or based on cementum annulus deposition from an extracted vestigial premolar tooth for bears first captured as adults[3].

Estimating life history traits

We estimated polar bear ages at first reproduction and lifetime reproductive success using previously established pedigree relationships. The western Hudson Bay polar bear population pedigree contains 4,634 polar bears (443 sires, 923 dams, and 1,130 founders, i.e., individuals of unknown parentage) from over six generations sampled between 1966 and 2019. Field sampling data from females and cubs-of-the-year provided offspring-dam associations. Additional linkage information came from parentage analyses using multi-locus microsatellites to genotype individuals[2]. We removed three individuals whose sex classifications were inconsistent with parentage data; these individuals were classified either as male dams or female sires. Additional information about the pedigree construction, capture, handling, and sampling protocols for the western Hudson Bay subpopulation is described in more detail in[2]. We used the pedigree to estimate lifetime reproductive success and age at first reproduction for all individuals who were confirmed parents of at least one other bear in the pedigree. We defined lifetime reproductive success as the total number of other bears in the pedigree for which an individual was a confirmed parent. We defined age at first reproduction as the age of the individual when its first known offspring was born. In our analyses considering age at first reproduction and lifetime reproductive success, we removed individuals with potentially biased data, using only data from individuals born between 1980 and 2000 (n = 628 bears). Sampling between 1966–1979 was less

consistent than post-1980 sampling. By 2019 bears born after 2000 would not have reached 20 years, or the approximate age of senescence for western Hudson Bay polar bears[4], which might have resulted in artificially low estimates of lifetime reproductive success. While we expected some error in the absolute ages at first reproduction and lifetime reproductive success because of gaps in the pedigree, we assumed relative comparisons among individuals were unbiased between 1980 and 2000.

Building the epigenetic clock for polar bears

Epigenetic clocks predict chronological ages based on methylation of CpG dinucleotides, where a cytosine is followed by a guanine[5]. Many CpGs change with chronological age. The Illumina HorvathMammalMethylChip40 array (Illumina Inc., San Diego, CA, USA; hereafter mammalian array) was designed to analyze methylation at CpG sequences highly conserved across all mammalian species, measuring a total of 37,449 unique sequences per sample at a single nucleotide resolution. The high-throughput array can process 96 samples simultaneously, making this approach useful for aging samples from long-term ecological projects that store many samples over multiple years.

To build our epigenetic clock, we randomly selected from 6,135 blood and skin samples collected from western Hudson Bay polar bears aged 0–30 over the sampling period. We stratified sampling based on individual age, tissue type, sex, and year of sample collection. We ensured at least some samples came from bears that were sampled more than once over their lifetimes to test for consistency in individual aging rates over time (Figure 2). To test for consistency in DNA methylation rates between tissues, we also included several samples with blood and skin collected from the same individual at the same time. Our final 288 samples included 150 female and 138 male samples from 223 unique individuals, collected between

1988–2018, of which 111 were blood and 177 were skin. Five individuals were sampled either 4, 9, 10, 11, or 13 times throughout their lives and another 25 individuals were sampled twice. All sample details, including ages, sexes, and dates of sampling are available in Supplementary Data File 1.

We isolated genomic DNA from blood and skin samples using the Qiagen DNeasy Blood and Tissue Kit 250 (Qiagen, Hilden, Germany). We dissected approximately 25 mg of frozen skin samples on a pre-chilled plate placed on dry ice to prevent thawing of the entire tissue. We then cut the skin tissue into smaller pieces, placed it in 1.5 mL microcentrifuge tubes, and digested it overnight in Buffer ATL (Qiagen) and Proteinase K (Qiagen) solution at 56 °C. We also digested 50 µL volumes of blood samples in Proteinase K (Qiagen) and PBS (pH 7.4, 1X, Gibco) solution at 56 °C for 10 minutes. After tissue digestion, we extracted genomic DNA from the samples as per the manufacturer's protocol (Document #HB-0540-002, Version #04/2016) and eluted the samples in two 100 µL volumes of elution buffer (Qiagen) consecutively to increase yield. We measured the concentration of gDNA using the NanoDrop2000 spectrophotometer (Thermo Scientific, Wilmington, USA). Next, we treated 750 ng of each genomic DNA sample with sodium bisulfite using the EZ-96 DNA Methylation-Gold Kit (shallow-well format) (Zymo Research, CA, USA) as per the manufacturer's protocol (Document #D5007, Version #2.1.6). We eluted the bisulfite-converted DNA in 12 µL of elution buffer (Zymo Research), after which we amplified 4 µL from each sample to be hybridized onto the mammalian array following the Infinium HD Methylation Assay protocol (Document #15019519, Version #07).

We measured DNA methylation by imaging the hybridized chips on the same day they were stained using the iScan instrument (Illumina Inc., San Diego, CA, USA). We normalized

468 the raw intensity data (IDAT) files from the chip scans using the recommended pipeline in the
469 *minfi* package in R (Aryee et al. 2014). Normalized intensity data, hereafter values, quantify
470 the degree of methylation at each of the 37,449 sites on the mammal chip with a value between 0
471 for no methylation and 1 for 100% methylation at each site.

472 The design of the mammalian array, while appropriate for any mammal species, presents
473 some practical challenges in terms of accurately quantifying methylation in the genomes of
474 specific species. First, all 37,449 CpG sequences on the array might not bind to the genomes of
475 all species because of species-specific CpG differences. Alternatively, a single sequence might
476 bind multiple times in the genome of a given species because probes were designed with up to
477 three degenerate bases to facilitate matching in case of cross-species mutations[6]. Sequences that
478 bind multiple times can confound methylation signals coming from multiple sites at once[6].
479 Methylation can also vary by sex if CpG sequences are located on the sex chromosomes[7], a
480 particularly important concern for the mammalian array because of species-specific locations of
481 CpG sequences on chromosomes.

482 To minimize potential confounds from sex-specific site methylation and non-binding or
483 multiple-binding probes, we narrowed our probe search space before building our clock. First,
484 we aligned the CpG sequences on the mammalian array to a reference polar bear genome (NCBI
485 Genome assembly ASM1731132v1
486 https://www.ncbi.nlm.nih.gov/datasets/genome/GCF_017311325.1/) using the *QuasR* package
487 v1.40.1[8] in R v4.3.1[9]. We selected only the 33,674 sites that bound uniquely. We also limited our
488 search space with an epigenome-wide association study (EWAS), a technique which correlates
489 phenotypic traits with epigenetic modifications including DNA methylation. We used our EWAS
490 to isolate sites with methylation patterns related to age but not different between sexes. We fit

three linear models with site-specific CpG methylation proportions as the response and combinations of age and sex as predictors using the *limma* package v3.56.2 in R[10]. In the first model, we tested the effects of sex on methylation while controlling for ages and tissue types of samples. We also fit two models including only either blood or skin samples to isolate the effects of age on the proportion of methylation in either tissue. We excluded 3,740 CpG sequences significantly associated with sex ($p < 0.05$) and 29,573 that were not sufficiently associated with age ($p > 10e^{-6}$). We erroneously excluded another 23 (0.007%) CpG sites. We used a final 3,328 of the 37,449 CpG sites on the HorvathMammalMethylChip40 array in the model we used to build our clock (Supplementary Data File 2).

We fit our DNAm clock using a training set of 144 samples (Supplementary Data File 1) from our original 288 samples. We first screened unreliable samples from the training data by predicting epigenetic ages in our samples using the universal clock for mammals[11]. We screened 10 samples from the training data either because their scans failed on the iScan or the normalized values did not pass quality control tests. Because we included bears from a single population, we were also concerned that potential relatedness between individuals used to build the clock might bias its predictions. We used the *GeneAlEx 6.5* software[12] to assess relatedness between individuals and screened any individuals from the training data with a relatedness index > 0.25. We also screened any individuals with repeat samples from the training data. We used the training data to fit the ~ age clock model using the cv.glmnet function in the *glmnet* package v4.1-8 in R[13], setting = 0.5 to combine the benefits of both ridge and lasso regression. This compromise reduces the variance in age predictions at the cost of some bias. We used 10-fold cross-validation to select the optimal regularization parameter. We validated our clock by using it

to predict the chronological ages of all remaining samples (n = 134; Supplementary Data File 1). CpG sites and values are available in Supplementary Data File 3.

Statistical analysis

Generalized linear models

We used Bayesian generalized linear models to test for a change in epigenetic age acceleration over time, for a relationship between age acceleration and age at first reproduction, and for a relationship between age at first reproduction and lifetime reproductive success. We fit all models using the *brms* package v2.20.4 in R[14], with 4 chains and 10,000 iterations including 5,000 warmup iterations. For the first two models — age acceleration over time and with age at first reproduction — we specified a Gaussian link function and used weakly informative prior slopes with a mean of 0 and standard deviation of 1. We tested for a relationship between birth year and age acceleration using n = 134 bears aged 0–30 born between 1966–2013. We tested for a relationship between age at first reproduction and age acceleration for n = 100 bears with epigenetic age estimates and known offspring. We included an effect for sex in both models to control for differences in age acceleration between male and female bears. We also included random effects in both models to account for individual differences and multiple measures from individuals. We included n = 628 bears born between 1980–2000 in our model testing the effect of age at first reproduction on lifetime reproductive success. For this model we specified a negative binomial response distribution with a log link function, using weakly informative prior slopes with a mean of 0 and a standard deviation of 1. We included an interaction between age at first reproduction and birth year.

Animal model

We used an animal model[1] to estimate the additive genetic variance of lifetime reproductive success for 628 polar bears. We used the pedigree data to create a genetic relatedness matrix. We fit this matrix as the random effect '*animal*' to estimate additive genetic variance (V_A). Phenotypic variance (V_P) is partitioned into additive genetic variance (V_A) and a residual variance (V_R) component, which is interpreted as the environmental effect. We further partitioned the residual variance by including maternal variance (V_M, or the identity of individual's dam) and cohort variance (V_{YBirth}, or year of birth). We also included sex as a fixed effect in the model. We used a log link function, an inverse-Gamma distribution for the random effect variances, and a wide normal distribution for the prior distribution of fixed effects[15]. We fit the animal model in the package *MCMCglmm* v2.35[16] in R with 1,000,000 iterations and 20,000 warmup iterations. The *MCMCglmm* package allows incomplete pedigrees and uses Bayesian inference and Markov chain Monte Carlo (MCMC) methods.

Code availability: All code is available from https://github.com/ljnewediuk/PB_life-history.

Data availability: All data needed to recreate main analyses are available from https://github.com/ljnewediuk/PB_life-history. Additional data will be made available in the figshare repository upon acceptance.

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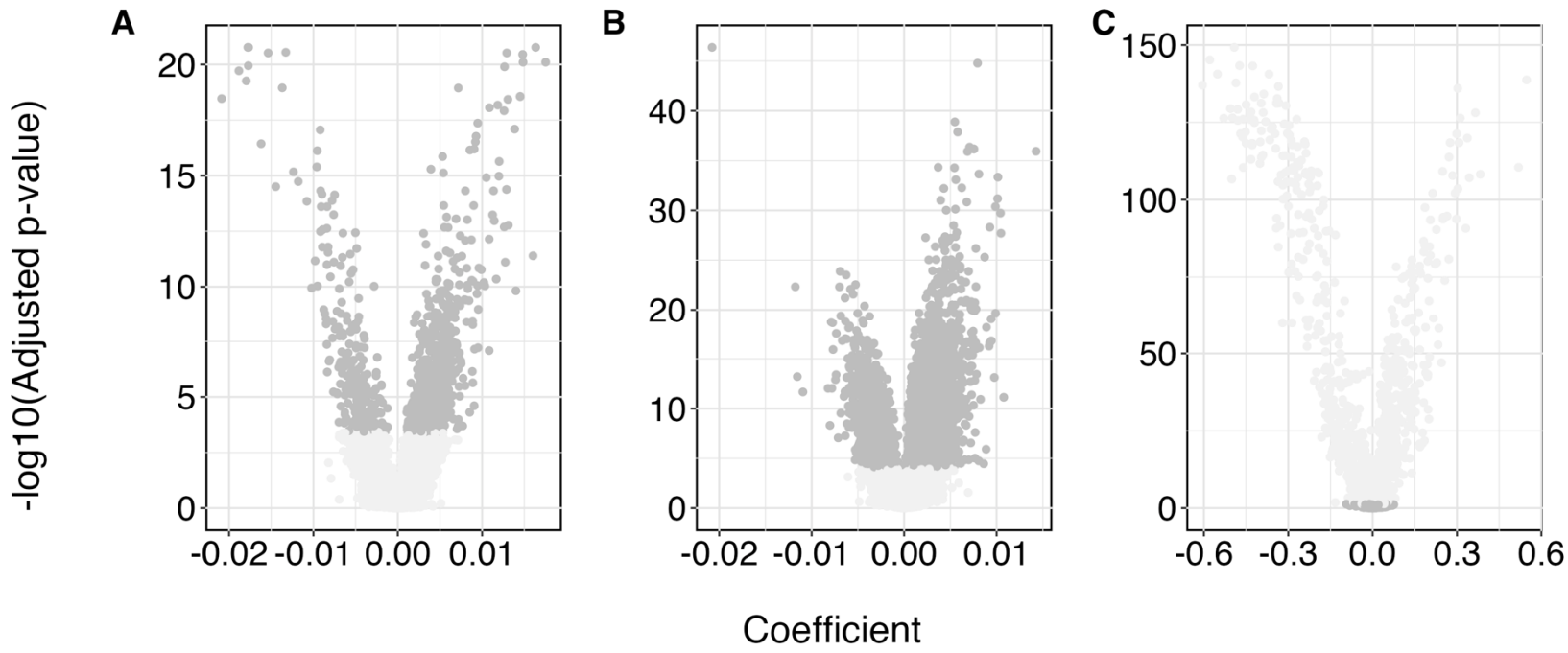
Supplementary information: Supplementary information is available in separate files supplemental_data_file1.csv, supplemental_data_file2.csv, and supplemental_data_file3.txt.

Extended Data

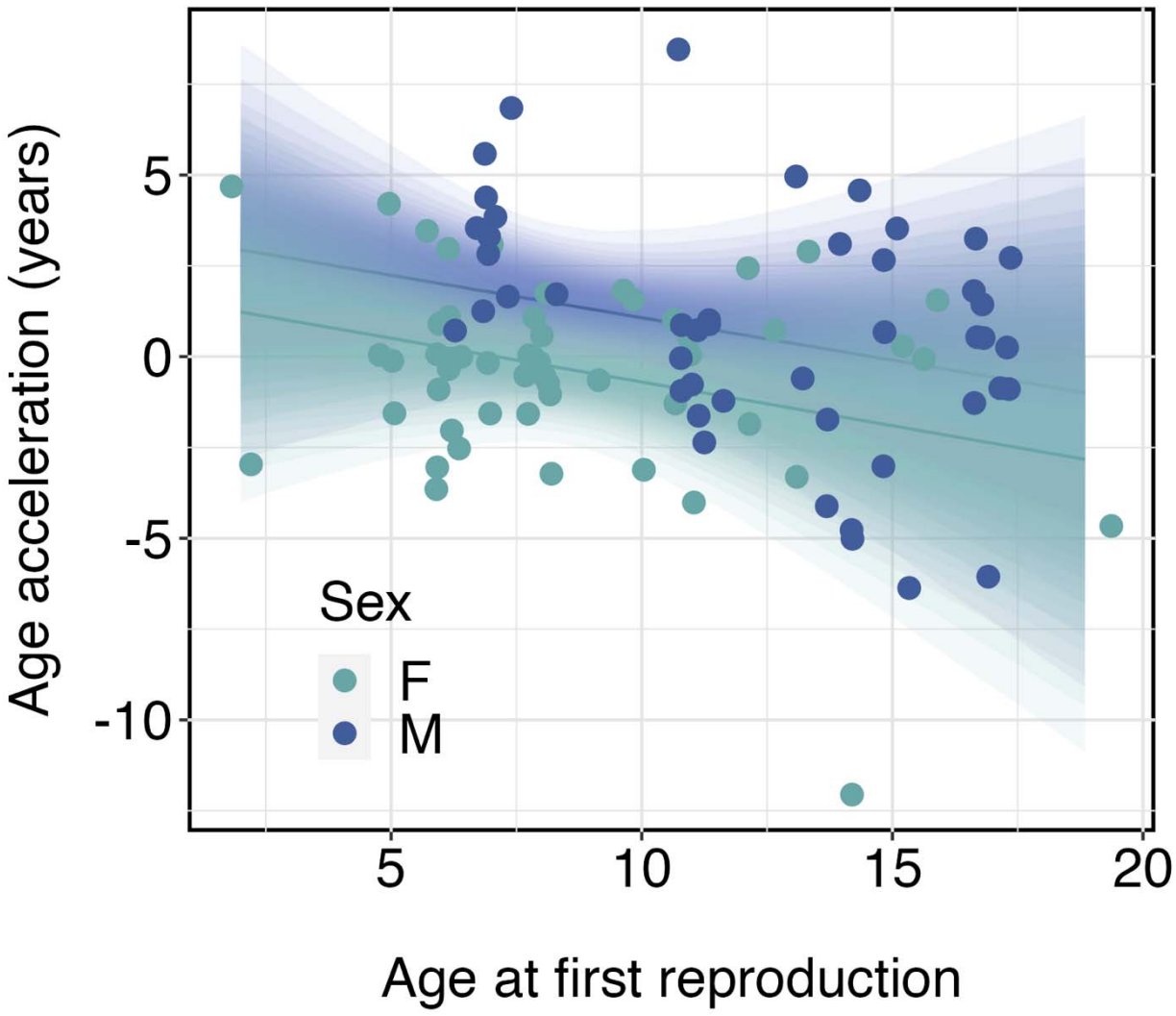
Extended Data Table 1. Phenotypic mean and estimated random-effect sizes of lifetime reproductive success for Western Hudson Bay polar bears born between 1980 and 2000 using a univariate animal model with a 4634-individual pedigree documented between 1966 and 2019 in northeastern Manitoba near Churchill, Canada. N_{ind} indicates the number of individuals with an estimate of observed lifetime reproductive success, a measure of relative fitness. V_P is the total phenotypic variance and is the sum of the variance components.Variance components; V_A=additive genetic 'animal', V_M=maternal 'dam' (i.e., identity of individual's dam), V_{YBirth}=cohort 'year of birth', and V_R=residual 'units'.Values in parentheses represent standard deviations (SD) or standard errors (SE).

Trait	**N_{Ind}**	**$N_{females}$**	**N_{males}**	**Mean (SD)**	***V_P*** **(SE)**	***V_A*** **(SE)**	***V_M*** **(SE)**	***V_{YBirth}*** **(SE)**	***V_R*** **(SE)**
Lifetime reproductive success	447	301	146	3.69 (2.37)	0.154 ($2.45x10^{-5}$)	0.007 ($2.90x10^{-5}$)	0.006 ($2.72x10^{-5}$)	0.037 ($6.59x10^{-5}$)	0.104 ($7.24x10^{-5}$)

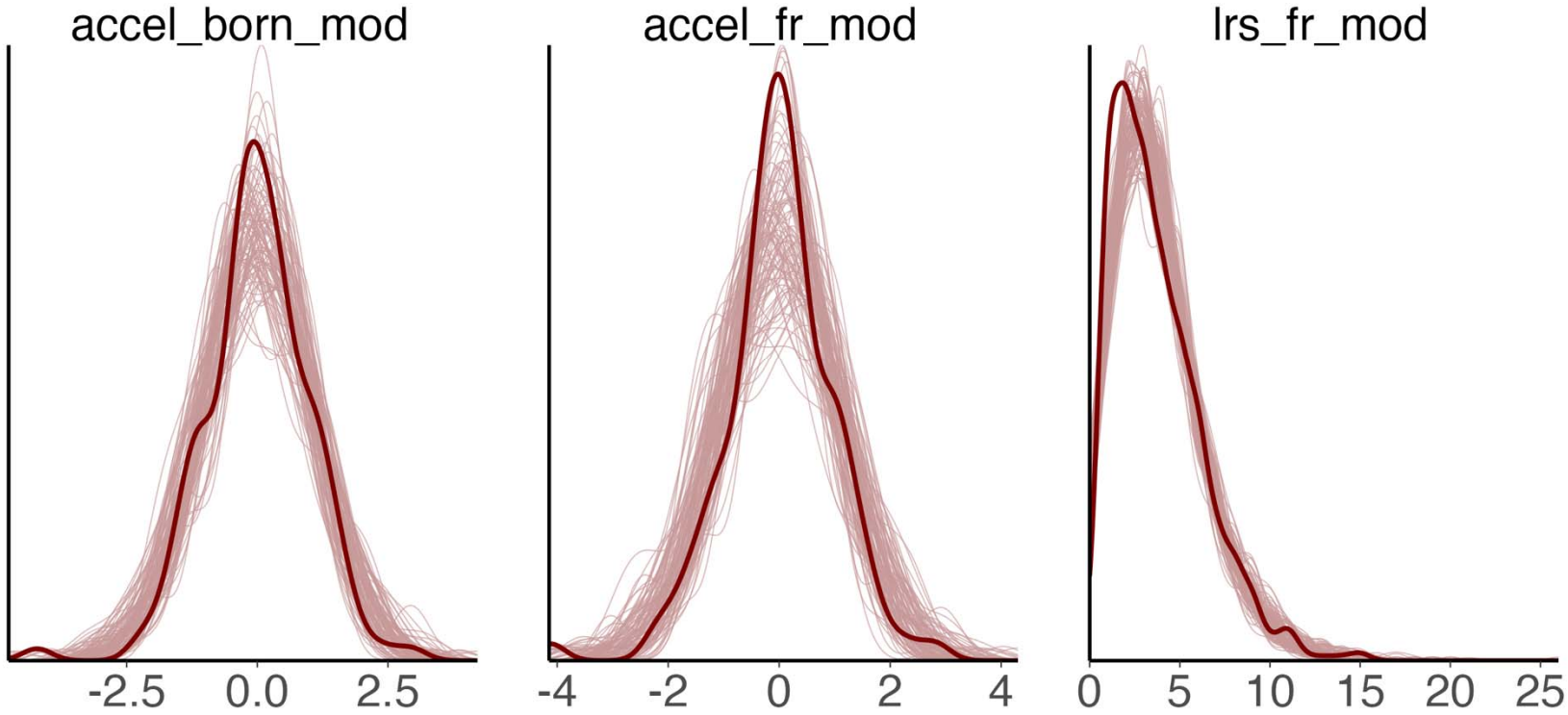


615

616 **Extended Data Figure 1.** Volcano plots showing sites included in clock (dark grey) and excluded from the clock (light grey) because
617 the coefficients from blood (a) and skin (b) samples were either not significantly associated with age, or both skin and blood samples
618 were associated with sex (c).



Extended Data Figure 2. Effect of age at first reproduction on epigenetic age acceleration for n = 100 male (blue) and female (seafoam) western Hudson Bay polar bears (Ursus maritimus). Points represent measurements of age acceleration from individuals sampled between 1988 and 2016, and the line and ribbon represent the mean and 95% percentile interval around the posterior distribution estimated using a Bayesian generalized linear model.



Extended Data Figure 3. Plots showing posterior predictive checks for Bayesian generalized linear models. Draws from the posterior predictive distribution (thin lines) are compared against the observed data (thick lines). Models include change in age acceleration with birth year (accel_born_mod), change in age acceleration with age at first reproduction (accel_fr_mod), and change in lifetime reproductive success with age at first reproduction (lrs_fr_mod).