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Research Article

Accelerated Epigenetic Aging and Prospective Morbidity and Mortality Among U.S. Veterans

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Abstract

Background: Epigenetic aging measures have promise as surrogate health outcomes in randomized control trials and observational cohort studies. The value of these measures, however, will reflect the extent to which they are associated with prospective health outcomes in real-world medical settings.

Methods: Using data from 2 216 post-9/11 veterans from the VISN 6 MIRECC's Post-Deployment Mental Health Study, we examined whether accelerated epigenetic aging, assessed by DunedinPACE, was associated with prospective chronic disease morbidity, predicted healthcare costs, and mortality over an average of 13.1 years of electronic health record follow-up.

Results: Veterans with faster DunedinPACE aging scores developed more chronic disease over the subsequent 5 years (*RR*, 1.25; 95% CI, 1.14–1.36), 10 years (*RR*, 1.31; 95% CI, 1.21–1.40), and 15 years (*RR*, 1.36; 95% CI, 1.22–1.52). Faster aging scores were also associated with increases in predicted healthcare costs over the next 5 years (β = 0.08; 95% CI, 0.03–0.13), 10 years (β = 0.23, 95% CI, 0.15–0.31), and 15 years (β = 0.21; 95% CI, 0.11–0.30). Faster DunedinPACE aging scores were associated with greater risk for incident myocardial infarction (84%), stroke (38%), diabetes (56%), cancer (25%), liver disease (44%), and renal disease (34%), as well as greater risk of mortality due to all-causes (38%) and chronic disease (74%). These results remained when adjusting for demographic, biomarker, and smoking covariates.

Conclusions: Our findings suggest DunedinPACE is a biomarker of accelerated aging that is prospectively associated with chronic disease morbidity and mortality, as assessed using health records from an integrated healthcare system.

Keywords: Biological aging, Chronic disease, DNA methylation, Mortality, Veterans

Epigenetic measures of aging derived from DNA methylation (DNAm) developed over the last decade can assess biological aging using tissue samples collected at a single point in time (1–6). These new measures have the potential to allow for the identification of individuals with accelerated aging who could be targeted by geroprotective interventions. Slowing the rate at which individuals are aging—defined in the geroscience hypothesis as a common cause of chronic disease morbidity and mortality (7,8)—would be expected to improve health across many chronic disease pathways and organ systems (9–11). If epigenetic measures can index change in biological aging and are associated with prospective health, they would be invaluable surrogate health outcomes for randomized control trials testing interventions that aim to slow aging and improve health (12), as well as observational studies investigating health trajectories (13). With additional validation,

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such measures also have the potential to serve as clinical biomarkers of future health for use in clinical settings.

There is promising evidence that second- and third-generation epigenetic measures of aging (1,2) are associated with future health (1,3,4,14,15), particularly third-generation measures developed using longitudinal biomarker data (1,15) (eg, DunedinPACE (2)). However, realizing the potential clinical and research value of epigenetic measures of aging requires evaluating whether these biomarkers are associated with prospective chronic disease morbidity and mortality in real-world medical settings (16). To do so, we integrated survey, epigenetic, and electronic health record (EHR) data from 2 216 U.S. military veterans who served after September 11, 2001 (17). The cohort was 37.4 years old at enrollment with an average follow-up of 13.1 years, which afforded an observation window spanning early adulthood into midlife and older age, periods that often include the onset of chronic disease (18).

Method

Participants and Study Design

Participants were enrolled in the VISN 6 Mid-Atlantic Mental Illness Research, Education, and Clinical Center's (MIRECC's) Post-Deployment Mental Health Study (PDMH) (17), a multisite study of veterans who served in the post-9/11 period. The Durham, Richmond, and Salisbury Veteran's Affairs (VA) Medical Centers' Institutional Review Boards approved the PDMH study protocol and all participants provided informed consent. The study included participants with DNAm and VA EHR data (Supplementary Figure 1), resulting in a sample of 2 216 veterans followed for an average of 13.1 years (standard deviation, *SD* = 2.8; Supplementary Figure 2).

Measures

Genomic DNAm data generation and processing

Whole blood was collected during baseline assessments and analyzed using the Infinium HumanMethylation450 or MethylationEPIC Beadchip (Illumina Inc., San Diego, CA) to derive DNAm data (19,20). Internal replicates were included and checked for consistency using single nucleotide polymorphisms on each array. Quality control was performed using the minfi (21) and ChAMP (22) R packages (R Foundation for Statistical Computing, Vienna, Austria). Samples were excluded if average fluorescence signal intensity was below 2 000 arbitrary units or < 50% of the mean intensity of all samples, > 10% of probes were not detectable (p value > .001), if a sex mismatch was detected, or if the sample was deemed an outlier on principal component analysis plots. In total, 134 samples were removed due to quality control. Probe quality control and data normalization were performed within each batch using the R package wateRmelon (23). Probes not detected (detection p value > .001) in > 10% of samples and those hybridizing to multiple locations in the genome were removed. Raw beta values were normalized using the dasen approach (24) and batch and chip adjustments were completed using ComBat in the R package sva (25). Methylation values reflected the resulting normalized and adjusted beta values.

DunedinPACE. Epigenetic aging was assessed by applying the DunedinPACE algorithm to PDMH DNAm data to produce normalized DunedinPACE values (21,22,26). The algorithm (26) is derived from reliable CpG probes (25) and produces aging scores that represent years of biological aging per chronological year. Statistical tests used continuous DunedinPACE scores unless otherwise noted (ie, aging quartiles were created for visualization and interpretation only).

Technical DNAm covariates. A dummy variable was created to denote if DNAm data was generated using 450k or EPIC V1 chips. Estimated white blood cell counts (27) (T lymphocytes [CD4+ and CD8+], B cells [CD19+], monocytes [CD14+], NK cells [CD56+], and neutrophils) were derived using FlowSorted.Blood.450k and FlowSorted.Blood.EPIC packages and were used as covariates in all models (excluding neutrophils to avoid multicollinearity).

DNAm smoking. Lifetime exposure to tobacco smoke (28) was calculated for participants using a DNAm measure (29). These methylation smoking scores were moderately associated with self-reported smoking ($r = 0.55$, $p < .001$).

PC-based second-generation epigenetic clocks: PhenoAge and GrimAge. We derived 2 second-generation clocks derived from principle components (PC) generated using data from 78 464 CpGs, PC-PhenoAge (30), and PC-GrimAge (3). The PC-based second-generation clocks provide improved reliability for aging estimates (13) compared to the original PhenoAge and GrimAge algorithms (described in detail previously) (3,30). Principle components-based clock estimates were generated using established algorithms (12) and residualized on chronological age to provide a measure of age acceleration.

Electronic health record data

Prospective health outcomes and clinical biomarkers were derived using the VA EHR. Supplementary Method 1 provides a detailed description of EHR data processing. Veterans enrolled in the PDMH from 2005 to 2016 (13), which resulted in follow-up periods ranging from 7.3 to 18.5 years (88–222 months). Electronic health record data coverage (Supplementary Table 1) was predominantly based on the timing of veterans' baseline assessment (and resulting length of EHR follow-up). Baseline assessment year was not associated with DunedinPACE (Supplementary Figure 3).

Charlson Comorbidity Index. Charlson Comorbidity Index (31) (CCI) scores assessed chronic disease burden and were derived using diagnostic ICD-9 and ICD-10 codes (32) ascertained from outpatient, inpatient, and purchased care data (ie, community care referrals from VA providers and/or paid by VA sources). Baseline values were calculated on the date of enrollment in the PDMH and were updated for each follow-up period. Charlson Comorbidity Index scores were used to calculate 10-year CCI-predicted mortality risk.

Nosos risk adjustment score. Nosos risk adjustment scores (33,34) represent predicted annual healthcare costs for VA patients based on the Centers for Medicare and Medicaid Hierarchical Condition Categories risk adjustment model. This algorithm was updated to include items specific to the VA, such as priority status and computed costs (34). Nosos scores are normalized to a mean of 1.0, such that greater values represent higher predicted patient costs (eg, a score of 1.25 equals 25% higher predicted costs).

Chronic disease onset. Chronic disease onset for each of the disease categories in the CCI was ascertained at baseline, as

well as at follow-up to a censor date of December 31, 2023. Combined diagnosis data were used to create a measure of time to first chronic disease onset across categories.

Clinical biomarkers

Clinical biomarkers included body mass, blood pressure (BP), and heart rate (*HR*), which were selected based on their routine collection during VA clinical encounters. Body mass was calculated using the standard formula via height and weight. Blood pressure was assessed using systolic BP—diastolic BP was not included to avoid multicollinearity due to a high correlation with systolic BP ($r = .75$, $p < .001$). Heart rate was assessed using pulse. All measures were assessed at baseline using a 2-year lookback period, producing data for over 75% of veterans who had clinical encounters with these biomarkers assessed (Supplementary Table 1).

Mortality

Dates of death and all-cause mortality status were ascertained using the VA EHR. Months from PDMH baseline to date of death defined time to mortality to a censor date of January 21, 2024. In total, 92 deaths were observed over the follow-up. National Death Index data included in the VA Mortality Data Repository (MDR) (35) provided primary cause of death, which was used to classify deaths as related to acute causes or chronic disease. A subset of recent deaths did not have a cause of death data ($n = 22$), as MDR data is currently censored to December 31, 2021 (32). Further excluding acute mortality events ($n = 30$; due to overdoses, accidents, death by suicide, infection, and homicide) left 40 deaths due to chronic diseases, largely cardiovascular diseases ($n = 17$), and cancer ($n = 15$).

Demographics

Participants reported their age, sex, race, ethnicity, years of education, and smoking status (coded: never smoker, 0; past smoker, 1; current smoker, 2). Sex, race, and ethnicity self-reports were confirmed using sex chromosomes and ancestry from genetic data.

Data Analysis

We tested the association of DunedinPACE epigenetic aging scores with CCI and Nosos scores at baseline, then with change to 5-, 10-, and 15-year follow-ups. Models of 5-, 10-, and 15-year change in CCI and Nosos scores controlled for baseline CCI and Nosos scores, respectively. Analyses of CCI scores used zero-inflated Poisson regression models to account for CCI distributions (see Supplementary Figure 4) and analyses of Nosos scores used linear regression models. We next tested the association between aging scores and incident onset of chronic diseases and mortality using Cox proportional-hazard models. Finally, we conducted 3 additional sets of analyses to complement the primary results: (a) stratifying by sex, then race and ethnicity, (b) moderating our primary models by methylation chip, and (c) providing results for 2 PC-based second-generation epigenetic measures of aging. For each set of models, we report estimates controlling for demographic (age, sex, race and ethnicity, and years of education) and DNAm technical covariates (chip type, white blood cell counts), then results controlling for clinical biomarkers (body mass, BP, *HR*), and 2 measures of smoking (self-reported smoking and DNAm-derived tobacco smoke exposure). Tables also include bivariate associations controlling for age. Poisson regression models used Monte Carlo simulation to account for missing data, linear regression models used full maximum likelihood estimation (full results excluding any participants with missing data are reported in Supplementary Table 2), whereas Cox proportional-hazard models included only participants with full data. Cox proportional-hazard models also excluded individuals who had outcome conditions at baseline (eg, models predicting diabetes excluded veterans with diabetes at baseline). Inspection of Schoenfeld residual plots and estimates of the interaction of time with aging scores suggest survival curves met the proportional hazard assumption. Models were run in MPLUS version 8.3 (36) using 2-tailed tests with an a priori significance level of 0.05 and all estimates were scaled to 1 *SD* DunedinPACE aging score.

Results

The 2 216 veterans (472 women, 1 744 men) included 1 077 non-Hispanic Black and 1 139 non-Hispanic White veterans, with a mean age of 37.4 years (*SD* = 10.1) at baseline.

Accelerated Aging and Chronic Disease Burden

Veterans with faster DunedinPACE aging scores had greater chronic disease burden at baseline (β, 0.23; 95% CI, 0.10–0.35; $p < .001$). Veterans with faster aging scores developed greater chronic diseases burden over the subsequent 5 years (β, 0.22; 95% CI, 0.13–0.31; $p < .001$), 10 years (β, 0.24; 95% CI, 0.16–0.31; $p < .001$), and 15 years (β, 0.31; 95% CI, 0.20–0.36; $p < .001$). These associations represented 25% (*RR*, 1.25; 95% CI, 14–36%), 27% (*RR*, 1.27; 95% CI, 17–41%), and 36% (*RR*, 1.36; 95% CI, 95% CI, 22–52%) greater relative risk, respectively. Results remained when controlling for clinical biomarkers and smoking measures (Table 1). The size of the associations increased as follow-up periods increased in length. At the 5-, 10-, and 15-year follow-ups, the fastest-aging veterans had 0.41, 0.92, and 1.84 higher CCI scores than the slowest-aging veterans (Figure 1; Supplementary Figure 5), corresponding to 5.3, 6.2, and 12.0 times greater relative—and 1.3%, 4.7%, and 16.5% greater absolute—increases in 10-year CCI-predicted mortality risk, respectively (Supplementary Table 3).

Accelerated Aging and Predicted Healthcare Costs

Veterans with faster DunedinPACE aging scores had higher predicted healthcare costs at baseline (β, 0.11; 95% CI, 0.06–0.15, $p < .001$). Veterans with faster aging scores had greater increases in predicted costs over the next 5 years (β, 0.08; 95% CI, 0.03–0.13; $p < .001$), 10 years (β, 0.23; 95% CI, 0.15–0.31; $p < .001$), and 15 years (β, 0.21, 95% CI, 0.11–0.30, $p < .001$). These results largely remained when controlling for clinical biomarkers and smoking measures (Table 1). Similar to the CCI, associations increased in size as the follow-up periods increased in length. At the 5-, 10-, and 15-year follow-ups, fastest aging veterans had 11%, 40%, and 38% greater increases in predicted healthcare costs compared to the slowest aging veterans (Supplementary Table 3). With estimated annual costs of $14 950 per veteran patient in 2021 (31), these represent $1 645, $5 980, and $5 681 greater increases in annual healthcare expenditures, respectively.

Accelerated Aging and Chronic Disease Incidence

Veterans with faster DunedinPACE aging scores were at increased risk for the onset of any chronic disease comprising

Table 1. Association of DunedinPACE and Prospective Health Among post-9/11 Veterans

Association With DunedinPACE	Age-adjusted Bivariate Association		Adding Demographic and Technical Covariates		Adding Clinical Biomarkers		Adding Self-reported and Methylation Smoking	
N = 2 216	*β*	95% CI	*β*	95% CI	*β*	95% CI	*β*	95% CI
Chronic disease burden (Charlson Comorbidity Index score)								
Baseline CCI score	0.25**	[0.14, 0.36]	0.23**	[0.10, 0.35]	0.20**	[0.07, 0.33]	0.19*	[0.01, 0.36]
5-Year change in CCI	0.20**	[0.13, 0.28]	0.22**	[0.13, 0.31]	0.17**	[0.07, 0.26]	0.14**	[0.04, 0.25]
10-Year change in CCI	0.19**	[0.12, 0.26]	0.24**	[0.16, 0.31]	0.19**	[0.11, 0.27]	0.18**	[0.10, 0.26]
15-Year change in CCI	0.29**	[0.19, 0.38]	0.31**	[0.20, 0.42]	0.27**	[0.16, 0.38]	0.30**	[0.18, 0.42]
Predicted annual VA healthcare costs (Nosos risk adjustment score)								
Baseline Nosos score	0.09**	[0.04, 0.12]	0.11**	[0.06, 0.15]	0.11**	[0.03, 0.10]	0.04	[−0.01, 0.09]
5-Year change in Nosos	0.09**	[0.05, 0.14]	0.08**	[0.03, 0.13]	0.07**	[0.01, 0.12]	0.03	[−0.03, 0.10]
10-Year change in Nosos	0.24**	[0.17, 0.31]	0.23**	[0.15, 0.31]	0.23**	[0.14, 0.31]	0.22**	[0.12, 0.31]
15-Year change in Nosos	0.22**	[0.13, 0.31]	0.21**	[0.11, 0.30]	0.19**	[0.09, 0.29]	0.15**	[0.04, 0.26]

Notes: Each model adds covariates to the model, first demographics (sex, race and ethnicity, and education) and technical covariates (chip type, cell proportions), then clinical biomarkers (body mass, blood pressure, and heart rate), and then self-reported smoking and smoking methylation scores. CCI outcomes were estimated with Poisson regression using Monte Carlo simulation to account for missing data; Nosos outcomes were estimated with linear regression using full information maximum likelihood. CI = confidence interval.
*$p < .05$.
**$p < .01$.

the CCI (*HR*, 1.29; 95% CI, 1.21–1.39; $p < .001$). When testing individual chronic diseases, faster aging was associated with greater risk for incident myocardial infarction (84%), stroke (38%), peripheral vascular disease (55%), diabetes (56%), chronic pulmonary disease (19%), cancer (25%), liver disease (44%), and renal disease (34%; Figure 2). Results remained when controlling for clinical biomarkers and smoking (Table 2), with the exception of peripheral vascular disease and chronic pulmonary disease. Figure 3 illustrates diabetes onset by DunedinPACE aging score categories.

Accelerated Aging and Mortality

Veterans with faster DunedinPACE aging scores were more likely to die due to all causes (*HR*, 1.38; 95% CI, 1.12–1.72, $p = .016$). Notably, when excluding mortality due to acute events, the association between aging scores and mortality was approximately twice as strong (*HR*, 1.74, 95% CI, 1.27–2.39, $p < .001$). DunedinPACE remained associated with mortality due to chronic disease when also controlling for clinical covariates and smoking (Table 2). Aging scores were not associated with mortality due to acute events (*HR*, 0.96, 95% CI, 0.63–1.33, $p = .836$).

Results Stratified by Sex, Race, and Ethnicity

We examined associations for CCI and Nosos scores when stratifying by sex, then by race and ethnicity. Men and women veterans showed largely similar associations of DunedinPACE with CCI and Nosos scores at baseline, as well as change over the next 5, 10, and 15 years (Supplementary Table 4). Non-Hispanic Black veterans and non-Hispanic White veterans also showed similar associations of DunedinPACE with CCI and Nosos scores at baseline, as well as change over the next 5, 10, and 15 years (Supplementary Table 5). There were no consistent moderations by sex, race, or ethnicity across the CCI or Nosos score outcomes.

Results Stratified by Methylation Chip

We also examined whether associations for DunedinPACE with CCI and Nosos scores varied by methylation chip. Methylation chip did not significantly moderate the association between DunedinPACE and any outcome (Supplementary Table 6).

Results for PC-based Second-generation Epigenetic Clocks

We focused on DunedinPACE epigenetic aging scores in our study, as DunedinPACE currently represents the most widely used third-generation epigenetic measure trained on longitudinal biomarker data. However, we also tested associations for 2 PC-based second-generation epigenetic clocks (12), PC-PhenoAge and PC-GrimAge. Consistent with prior studies, the 3 measures of aging were moderately correlated ($.33 \le r \le .52$, all ps $< .001$). When accounting for demographic and technical covariates, PC-PhenoAge was not consistently associated with prospective health. In contrast, PC-GrimAge was largely associated with CCI and Nosos scores, as well as a number of specific chronic diseases, all-cause mortality, and chronic disease mortality. Descriptively, the magnitude of most associations for PC-GrimAge was comparable to those for DunedinPACE, though DunedinPACE associations were larger when assessing longitudinal change in CCI scores, particularly over longer follow-up periods. Full results for DunedinPACE, PC-PhenoAge, and PC-GrimAge are presented in Supplementary Table 7.

Discussion

Veterans with faster DunedinPACE aging scores developed more chronic disease, showed larger increases in predicted healthcare costs, and were at greater risk of premature mortality over the follow-up, as observed in an average of 13.1 years of VA health records. The sizes of these prospective associations appear clinically significant. After 10 years, veterans with faster aging developed approximately 1 additional chronic disease (0.92 points on the CCI), corresponding to a 4.7% larger increase in 10-year predicted mortality risk compared to veterans with slower aging (a 5.6% vs 0.9% increase, 6.2 times greater increase in relative risk). Veterans

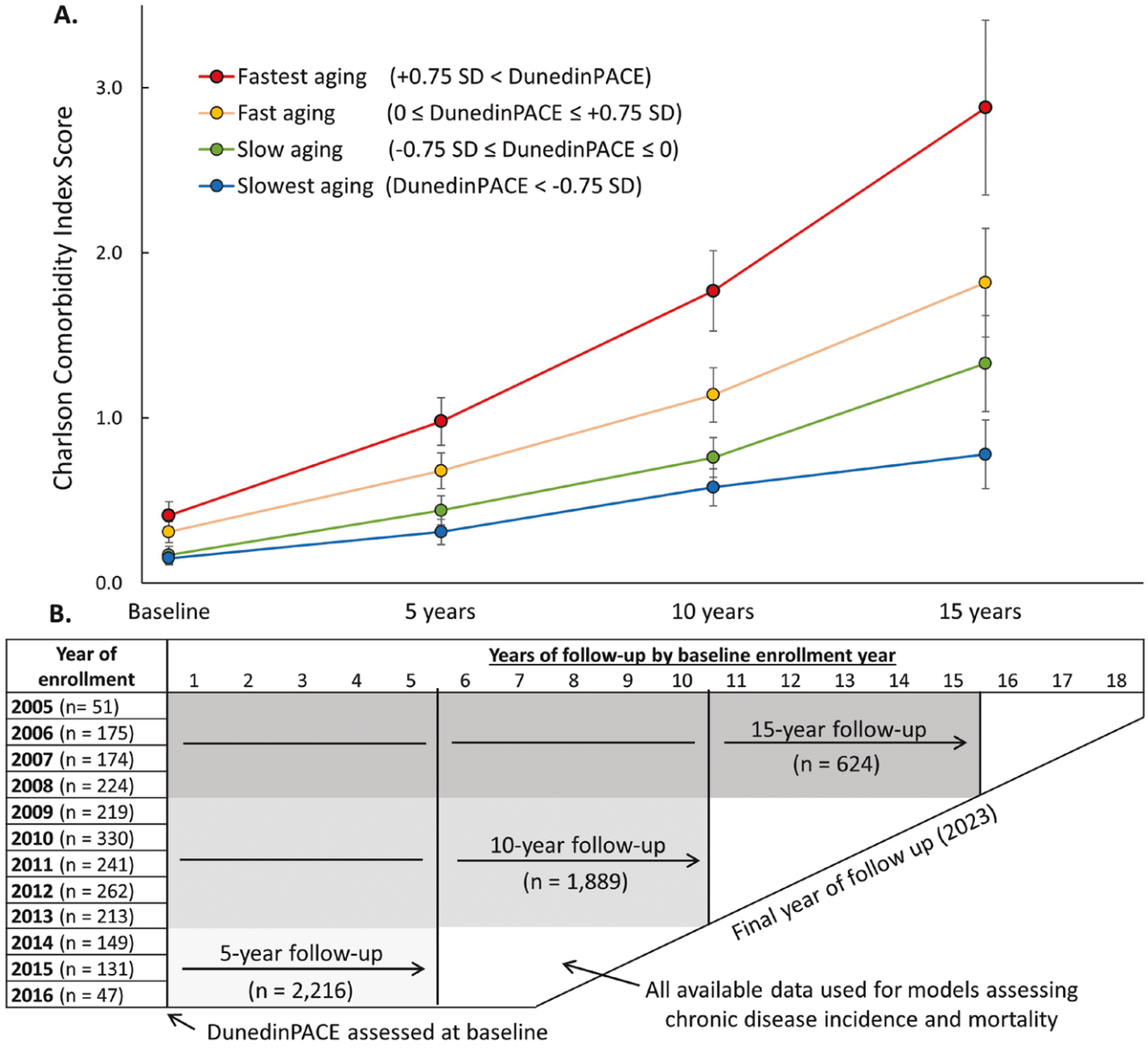


Figure 1. Panel **A** presents CCI scores over time grouped by DunedinPACE aging scores. Four groups were created by standardizing DunedinPACE scores and creating cutoffs at the mean and 0.75 *SD* above and below the mean, corresponding to DunedinPACE values of "slowest aging" ≤ 0.98 (n = 517, 23.3%), "slow aging" between 0.98 and 1.07 (n = 656, 29.6%), "fast aging" between 1.07 and 1.15 (n = 561, 25.3%), and 1.15 ≤ for "fastest aging" (n = 482, 21.8%). Groups were created using a priori *SD* cutoffs to rough quartiles for illustrative purposes—all models used full DunedinPACE aging scores. Panel **B** presents the study sample by year of enrollment in the PDMH and years of follow-up. Baseline PDMH enrollment included the blood draw used to derive DNA methylation data from whole blood.

with faster aging also had a 40% larger increase in predicted healthcare costs over the next 10 years, representing $5 980 higher annual costs per VA patient compared to veterans with slower aging. In terms of specific chronic disease morbidity and mortality, a 1 *SD* higher DunedinPACE aging score was associated with a 32% increased risk of developing any chronic disease—including increased risk for incident myocardial infarction (61%), stroke (32%), diabetes (85%), cancer (28%), liver disease (52%), and renal disease (46%)—and a 64% increased risk of death due to chronic disease. These associations accounted for numerous covariates—including chronological age, demographic and technical covariates, clinical biomarkers (body mass, BP, and *HR*), self-reported smoking, and smoking methylation scores—and excluded individuals with the relevant chronic disease at baseline. Notably, associations were largely similar for men and women veterans, as well as for non-Hispanic Black veterans and non-Hispanic White veterans.

Our results provide additional support and validation for the use of epigenetic aging measures as surrogate health outcomes in observational studies of health (13) and randomized control trials (12) aiming to slow aging (16). Although prior studies have linked epigenetic aging to a subset of prospective health outcomes using research cohorts (3,4,37,38), particularly mortality (3–6,14,15), none have used EHR data from an integrated healthcare system in a real-world medical setting. The sizes of the associations between DunedinPACE and chronic disease morbidity and mortality in our study were largely comparable to those reported in the original validation of DunedinPACE (3). Belsky and colleagues found

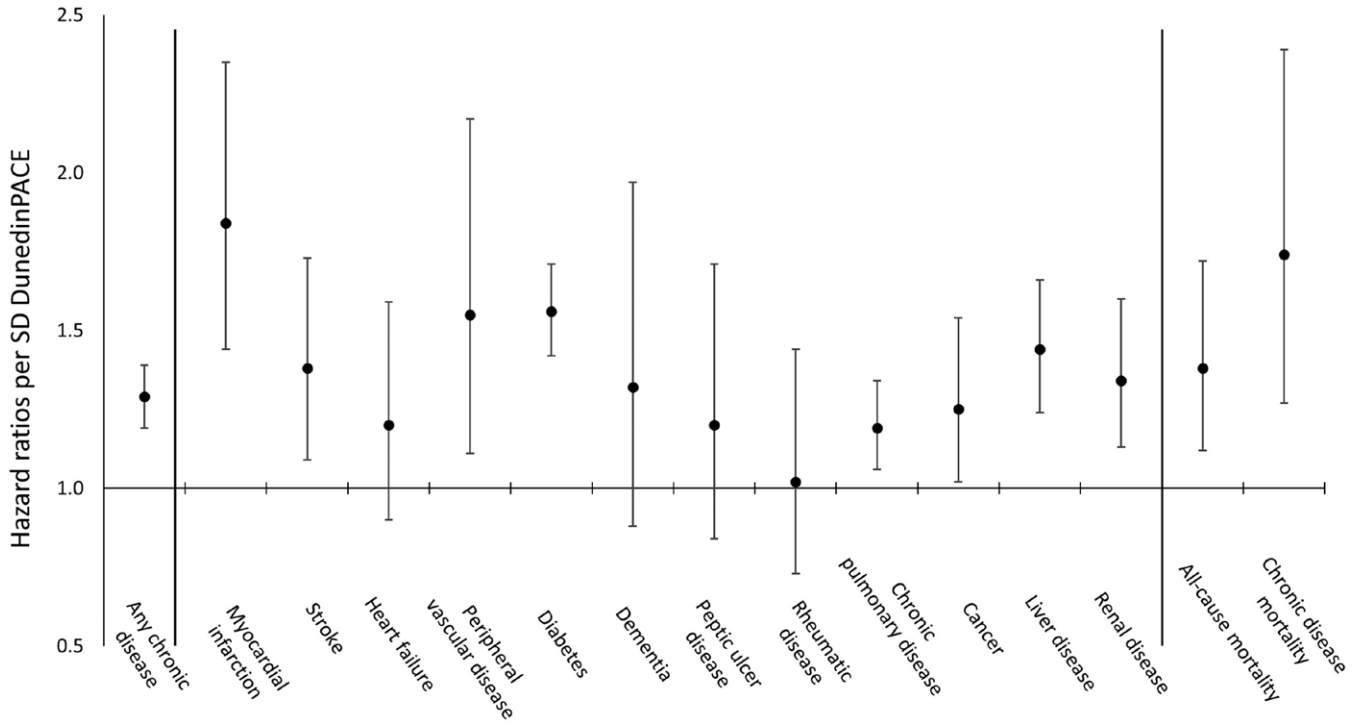


Figure 2. Visualization of the *HR*s for each of the CCI chronic disease categories over the follow-up period. Effects represent *HR*s per 1 *SD* difference in DunedinPACE. All estimates include demographic and technical DNAm covariates. Number of cases and excluded participants for each estimate are presented in Table 2. Error bars represent 95% confidence intervals.

DunedinPACE was associated with 23% increased risk of incident chronic disease, 37% increased risk of stroke, and 26%–65% increased risk of mortality across 2 research cohorts—older men veterans in the Normative Aging Study (mean age, 77 years) and individuals in the Framingham Offspring study (mean age, 66 years) (3). Similarly, we found DunedinPACE was associated with a 28% increased risk of incident chronic disease, 38% increased risk of stroke, 38% increased risk of all-cause mortality, and 74% increased risk of chronic disease mortality when controlling for demographic characteristics. Although the cohorts differed in composition—including age and sex—associations across samples were notably consistent.

Empirical evidence that epigenetic aging scores are associated with incident morbidity and mortality in an integrated healthcare system highlights the potential clinical and research applications of aging biomarkers. The associations presented in this study remained clinically relevant, even when accounting for common demographic characteristics, clinical biomarkers, and smoking. For example, the size of the association between DunedinPACE and change in chronic disease burden over the next decade was equivalent to that of 8.8 years chronological aging. The differences between all-cause and chronic disease mortality were similarly notable. The association between DunedinPACE and all-cause mortality was attenuated when accounting for the full range of covariates, particularly smoking behavior and methylation scores; however, the association with chronic disease death was not attributable to smoking or other covariates. Stronger associations with chronic disease death would be expected by a measure indexing accelerated aging, when compared to all-cause mortality, which includes deaths due to acute causes that DunedinPACE would not be expected to directly predict (eg, motor vehicle accidents). Finally, the DNAm data underlying aging scores derived using current algorithms would be available for any future refinements to those algorithms (such as the revised PC-based algorithms). As refinements occur, new algorithms can be validated with data from cohorts such as the PDMH. Ideally, epigenetic aging measures will eventually reach levels of reliability and validity that provide predictive utility for individual patients and clinical providers.

These results have particular relevance to the Veterans Health Administration (VHA). The post-9/11 cohort (currently over 5 million of the 17.9 million living U.S. veterans (39)) is a growing proportion of patients served by the VHA (40) and has different characteristics compared to previous service cohorts, such as greater numbers of women (39–41). Our findings show epigenetic aging is associated with prospective health across men and women, as well as across non-Hispanic Black and non-Hispanic White veterans, suggesting that future uses for epigenetic aging scores would benefit the patient populations that will utilize VHA services. Notably, the post-9/11 cohort of veterans is approaching midlife and older age (41), periods when chronic disease morbidity and mortality become more pronounced. This risk is also an opportunity. The VHA is the largest integrated healthcare system in the United States and implementing interventions to address risk (14,16,42) for accelerated aging—such as unhealthy behaviors (14) or PTSD (20)—could delay or prevent the development of ill health for a large number of veterans. If successful, efforts to slow aging using behavioral treatments (43) or other potentially geroprotective interventions could reduce healthcare costs and prolong veterans' independence, health, and well-being as they grow older. VA clinical trials could also provide data and guidance for

Table 2. Association of DunedinPACE, Chronic Disease Incidence, and Mortality Among Post-9/11 Veterans

Association With DunedinPACE	Chronic Disease at Baseline and Follow-up		Age-adjusted Bivariate Association		Adding Demographics and Technical Covariates		Adding Clinical Biomarkers		Adding Self-reported and Methylation Smoking	
	Baseline Dx	Dx Onset	*HR*	95% CI	*HR*	95% CI	*HR*	95% CI	*HR*	95% CI
Any chronic disease category										
Chronic disease onset	*n* = 364	*n* = 789	1.29**	[1.21, 1.39]	1.28**	[1.19, 1.39]	1.24**	[1.13, 1.37]	1.32**	[1.21, 1.45]
Chronic disease categories										
Myocardial infarction	*n* = 5	*n* = 56	1.64**	[1.32, 2.04]	1.84**	[1.44, 2.35]	1.73**	[1.26, 2.37]	1.61**	[1.17, 2.22]
Stroke	*n* = 21	*n* = 109	1.30*	[1.06, 1.61]	1.38**	[1.09, 1.73]	1.47**	[1.15, 1.88]	1.32*	[1.04, 1.68]
Heart failure	*n* = 13	*n* = 53	1.38*	[1.07, 1.78]	1.20	[0.90, 1.59]	1.05	[0.76, 1.44]	1.07	[0.77, 1.49]
Peripheral vascular disease	*n* = 17	*n* = 41	1.56**	[1.16, 2.08]	1.55**	[1.11, 2.17]	1.56*	[1.06, 2.31]	1.29	[0.87, 1.90]
Diabetes	*n* = 7	*n* = 482	1.54**	[1.41, 1.67]	1.56**	[1.42, 1.71]	1.40**	[1.25, 1.57]	1.85**	[1.66, 2.06]
Dementia	*n* = 12	*n* = 29	1.39	[0.99, 1.97]	1.32	[0.88, 1.97]	1.50	[0.94, 2.39]	1.22	[0.77, 1.94]
Peptic ulcer disease	*n* = 17	*n* = 36	1.29	[0.94, 1.77]	1.20	[0.84, 1.71]	1.09	[0.73, 1.62]	1.14	[0.76, 1.72]
Rheumatic disease	*n* = 14	*n* = 43	1.07	[0.79, 1.45]	1.02	[0.73, 1.44]	1.18	[0.80, 1.74]	0.93	[0.63, 1.38]
Chronic pulmonary disease	*n* = 234	*n* = 325	1.20**	[1.07, 1.34]	1.19**	[1.06, 1.34]	1.25**	[1.08, 1.44]	1.08	[0.94, 1.24]
Cancer	*n* = 42	*n* = 111	1.21*	[1.00, 1.45]	1.25*	[1.02, 1.54]	1.35*	[1.07, 1.71]	1.28*	[1.01, 1.63]
Liver disease	*n* = 24	*n* = 206	1.35**	[1.18, 1.55]	1.44**	[1.24, 1.66]	1.34**	[1.13, 1.58]	1.52**	[1.27, 1.81]
Renal disease	*n* = 26	*n* = 153	1.30**	[1.11, 1.51]	1.34**	[1.13, 1.60]	1.36**	[1.11, 1.67]	1.46**	[1.20, 1.78]
Mortality										
All-cause mortality	–	*n* = 92	1.28*	[1.02, 1.59]	1.38**	[1.12, 1.72]	1.25	[0.97, 1.62]	1.06	[0.82, 1.38]
Chronic disease mortality	–	*n* = 40	1.55**	[1.16, 2.06]	1.74**	[1.27, 2.39]	1.59*	[1.10, 2.32]	1.63**	[1.13, 2.36]

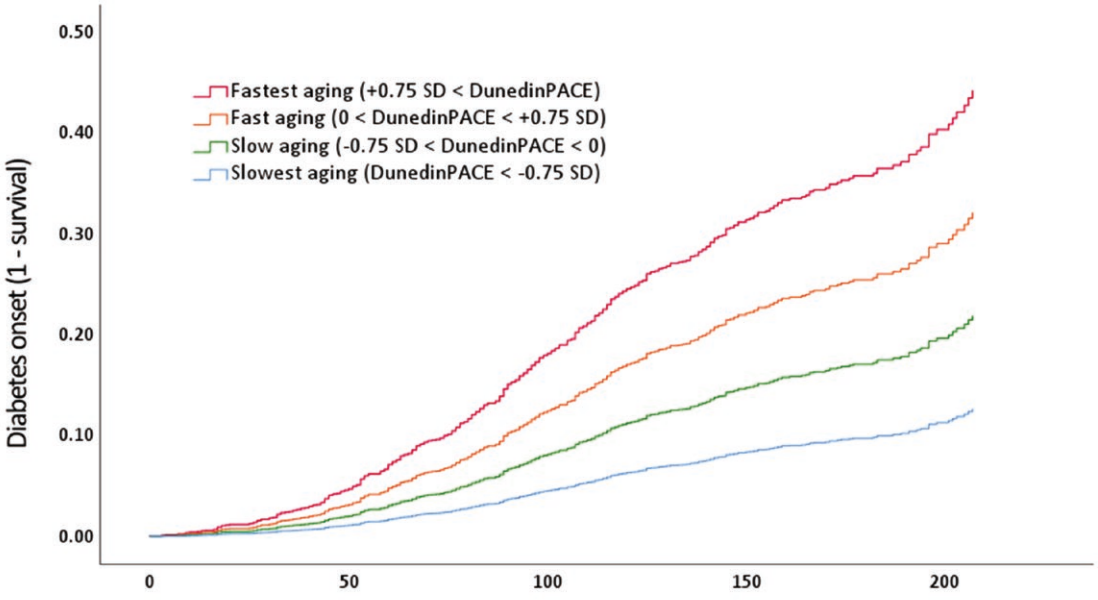
Notes: Each disease specific model excluded participants with the disease at baseline from the cohort *N* of 2 216. Models assessing the inclusion of clinical biomarkers and smoking measures were run with those sets of covariates separately to reduce missing data for the models that included smoking measures. Demographic and technical covariates models also excluded 21 participants who were missing data, clinical biomarker models excluded 561 participants with missing biomarker and education data, and models including self-reported smoking excluded 29 participants missing self-reported smoking or education data. Demographic variables included age, sex, race and ethnicity, education, and technical covariates included chip type and white blood cell proportions. CI = confidence interval; Dx = diagnosis; *HR* = hazard ratio.
*$p < .05$.
**$p < .01$.

implementing interventions in other non-VA populations and healthcare systems.

This study has limitations relevant to interpreting our findings. First, the EHR-derived health outcomes can only capture VA clinical encounters or referrals to community care through VA sources. Although it is unlikely that aging and chronic disease onset vary systematically by the amount or type of community care that veterans access through private insurance, it is possible our data are not representative of all veterans' health. Second, these results controlled for 3 health-relevant clinical biomarkers (body mass, BP, and HR) that are routinely collected during clinical care, including primary care. Other biomarkers might provide additional clinical utility in predicting future health, but could also introduce new bias due to selection effects, as tests are generally ordered due to providers' or veteran patients' health concerns, age, or other systematic reasons. Future studies would benefit from comparisons between DNAm measures of aging and other clinical biomarkers collected across full samples. Third, it is not clear to what extent results would generalize to nonveteran populations. It will be important to validate these results in other healthcare systems. Fourth, although the longitudinal associations adjusted for baseline health status, clinical biomarkers, and multiple measures of smoking, it is possible that additional unmeasured confounders explained the observed associations. Establishing causal links between epigenetic aging measures and health outcomes will benefit from experimental designs, such as randomized clinical trials, that can show interventions reliably produce changes in epigenetic aging (44), and that those changes in aging scores translate to improved health (45). Fifth, the sample averaged 38 years old at baseline and 51 years old at follow up. Many of the diseases associated with aging show increased incidence at older ages (eg, dementia) and there may be greater statistical power to detect effects in cohorts with older average ages with increased disease prevalence. Finally, the length of the EHR follow-up varied based on the year that veterans enrolled in the PDMH cohort. Although aging scores were not correlated with enrollment date and results replicated both when accounting for missing data and using listwise deletion, continuing to replicate our findings as the lengths of EHR observation increases in duration would provide additional confidence in our findings.

Conclusion

Epigenetic aging scores were associated with increased risk for chronic disease morbidity and mortality, as observed in VA medical records for 2 216 U.S. veterans (11) who served after September 11, 2001. Consistent with the geroscience hypothesis, faster aging was associated with poorer prospective health across multiple chronic disease categories and organ systems. Epigenetic measures of aging, such as DunedinPACE, might be useful surrogate outcomes for clinical trials and observational cohort studies. With additional validation, epigenetic measures of aging might serve as clinical



No. at risk					
Full sample	2188	2116	1853	999	211
Faster aging	470	434	346	165	27
Fast aging	556	533	455	234	49
Slow aging	651	640	568	321	61
Slowest aging	511	509	484	279	74

Figure 3. Visualization of diabetes onset (1—survival) across the follow-up period, as an illustrative example of chronic disease incidence. The model included demographic and technical covariates and excluded 28 veterans with a baseline diagnosis of diabetes or missing covariate data. The 4 groups were created by standardizing DunedinPACE aging scores and creating cutoffs, as with Figure 1. *No. at risk* represents veterans at risk of diabetes onset at each period on the *x*-axis. Over the follow up, 45 of 511 (8.8%) slowest aging veterans, 110 of 651 (16.9%) slow aging veterans, 145 of 556 (26.1%) fast aging veterans, and 179 of 470 (38.1%) of fastest aging veterans developed diabetes. Compared to the slowest aging veterans, all other groups were more likely to develop diabetes; slow aging *HR*, 1.90 (95% CI, 1.33–2.70), fast aging *HR*, 3.21 (95% CI, 2.27–4.56), fastest aging *HR*, 5.13 (95% CI, 3.57–7.38, all *p*s < .001).

biomarkers of future health risk that can identify individuals who might be candidates for geroprotective interventions.

Supplementary Material

Supplementary data are available at *The Journals of Gerontology, Series A: Biological Sciences and Medical Sciences* online.

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Conflict of Interest

Drs. Terrie Moffitt, Avshalom Caspi, and Karen Sugden are named as an inventor on a license issued by Duke University for the DunedinPACE. The algorithm to calculate DunedinPACE is publicly available on Github, https://github.com/danbelsky/DunedinPACE. The other authors declare no conflict.

Data Availability

Data from the Post Deployment Mental Health (PDMH) Study are part of a Veterans Affairs data repository and are available to researchers who request access through the VISN 6 MIRECC and follow the appropriate data access protocols. Medical record data from the Veteran Affairs Corporate Data Warehouse are available to researchers who request and are approved for access through the Office of Research and Development (ORD) Data Access Request Tracker (DART). Mortality Data Repository (MDR) data is available to researchers who request MDR data through an MDR Data Request.

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Disclaimer

The views expressed in this article are those of the authors and do not necessarily reflect the position or policy of the VA, the U.S. government or any other affiliated institution.

Author Access to Data

Dr. Kyle J. Bourassa had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

References

1. Rutledge J, Oh H, Wyss-Coray T. Measuring biological age using omics data. *Nat Rev Genet*. 2022;23(12):715–727. https://doi.org/10.1038/s41576-022-00511-7
2. Moqri M, Herzog C, Poganik JR, et al.; Biomarkers of Aging Consortium. Biomarkers of aging for the identification and evaluation of longevity interventions. *Cell*. 2023;186(18):3758–3775. https://doi.org/10.1016/j.cell.2023.08.003
3. Belsky DW, Caspi A, Corcoran DL, et al. DunedinPACE, a DNA methylation biomarker of the pace of aging. *Elife*. 2022;11:e73420. https://doi.org/10.7554/eLife.73420
4. Lu AT, Quach A, Wilson JG, et al. DNA methylation GrimAge strongly predicts lifespan and healthspan. *Aging (Albany NY)*. 2019;11(2):303–327. https://doi.org/10.18632/aging.101684
5. Horvath S. DNA methylation age of human tissues and cell types [published correction appears in Genome Biol. 2015;16:96. doi: 10.1186/s13059-015-0649-6]. *Genome Biol*. 2013;14(10):R115. https://doi.org/10.1186/gb-2013-14-10-r115
6. Marioni RE, Shah S, McRae AF, et al. DNA methylation age of blood predicts all-cause mortality in later life. *Genome Biol*. 2015;16(1):25. https://doi.org/10.1186/s13059-015-0584-6
7. Kaeberlein M. Longevity and aging. *F1000prime Rep*. 2013;5:5. https://doi.org/10.12703/P5-5
8. López-Otín C, Blasco MA, Partridge L, Serrano M, Kroemer G. The hallmarks of aging. *Cell*. 2013;153(6):1194–1217. https://doi.org/10.1016/j.cell.2013.05.039
9. Barzilai N, Cuervo AM, Austad S. Aging as a biological target for prevention and therapy. *JAMA*. 2018;320(13):1321–1322. https://doi.org/10.1001/jama.2018.9562
10. Hillary RF, Stevenson AJ, McCartney DL, et al. Epigenetic measures of ageing predict the prevalence and incidence of leading causes of death and disease burden. *Clin Epigenetics*. 2020;12(1):115. https://doi.org/10.1186/s13148-020-00905-6
11. Moffitt TE. Behavioral and social research to accelerate the geroscience translation agenda. *Ageing Res Rev*. 2020;63:101146. https://doi.org/10.1016/j.arr.2020.101146
12. Higgins-Chen AT, Thrush KL, Wang Y, et al. A computational solution for bolstering reliability of epigenetic clocks: implications for clinical trials and longitudinal tracking. *Nat Aging*. 2022;2(7):644–661. https://doi.org/10.1038/s43587-022-00248-2
13. Bourassa KJ, Sbarra DA. Trauma, adversity, and biological aging: behavioral mechanisms relevant to treatment and theory. *Transl Psychiatry*. 2024;14(1):285. https://doi.org/10.1038/s41398-024-03004-9
14. McCrory C, Fiorito G, Hernandez B, et al. GrimAge outperforms other epigenetic clocks in the prediction of age-related clinical phenotypes and all-cause mortality. *J Gerontol A Biol Sci Med Sci*. 2021;76(5):741–749. https://doi.org/10.1093/gerona/glaa286
15. Faul JD, Kim JK, Levine ME, Thyagarajan B, Weir DR, Crimmins EM. Epigenetic-based age acceleration in a representative sample of older Americans: associations with aging-related morbidity and mortality. *Proc Natl Acad Sci U S A*. 2023;120(9):e2215840120. https://doi.org/10.1073/pnas.2215840120
16. Moqri M, Herzog C, Poganik JR, et al. Validation of biomarkers of aging. *Nat Med*. 2024;30(2):360–372. https://doi.org/10.1038/s41591-023-02784-9
17. Brancu M, Wagner HR, Morey RA, et al.; VA Mid-Atlantic MIRECC Workgroup. The Post-Deployment Mental Health (PDMH) study and repository: a multi-site study of US Afghanistan and Iraq era veterans. *Int J Methods Psychiatr Res*. 2017;26(3):e1570. https://doi.org/10.1002/mpr.1570
18. Christensen K, Doblhammer G, Rau R, Vaupel JW. Ageing populations: the challenges ahead. *Lancet*. 2009;374(9696):1196–1208. https://doi.org/10.1016/S0140-6736(09)61460-4
19. Kimbrel NA, Garrett ME, Evans MK, et al.; VA Mid-Atlantic MIRECC Workgroup. Large epigenome-wide association study identifies multiple novel differentially methylated CpG sites associated with suicidal thoughts and behaviors in veterans. *Front Psychiatry*. 2023;14:1145375. https://doi.org/10.3389/fpsyt.2023.1145375
20. Bourassa KJ, Garrett ME, Caspi A, et al.; VA Mid Atlantic MIRECC Workgroup. Posttraumatic stress disorder, trauma, and accelerated biological aging among post-9/11 veterans. *Transl Psychiatry*. 2024;14(1):4. https://doi.org/10.1038/s41398-023-02704-y
21. Aryee MJ, Jaffe AE, Corrada-Bravo H, et al. Minfi: a flexible and comprehensive Bioconductor package for the analysis of Infinium DNA methylation microarrays. *Bioinformatics*. 2014;30(10):1363–1369. https://doi.org/10.1093/bioinformatics/btu049
22. Morris TJ, Butcher LM, Feber A, et al. ChAMP: 450k chip analysis methylation pipeline. *Bioinformatics*. 2014;30(3):428–430. https://doi.org/10.1093/bioinformatics/btt684
23. Pidsley R, Wong CCY, Volta M, Lunnon K, Mill J, Schalkwyk LC. A data-driven approach to preprocessing Illumina 450K methylation array data. *BMC Genomics*. 2013;14:293. https://doi.org/10.1186/1471-2164-14-293
24. Leek JT, Johnson WE, Parker HS, Jaffe AE, Storey JD. The sva package for removing batch effects and other unwanted variation in high-throughput experiments. *Bioinformatics*. 2012;28(6):882–883. https://doi.org/10.1093/bioinformatics/bts034
25. Sugden K, Hannon EJ, Arseneault L, et al. Patterns of reliability: assessing the reproducibility and integrity of DNA methylation measurement. *Patterns (NY)*. 2020;1(2):100014. https://doi.org/10.1016/j.patter.2020.100014
26. Belsky DW. *DunedinPACE calculator*. 2022. https://github.com/danbelsky/DunedinPACE
27. Houseman EA, Accomando WP, Koestler DC, et al. DNA methylation arrays as surrogate measures of cell mixture distribution. *BMC Bioinform*. 2012;13(1):1–6.

28. Joehanes R, Just AC, Marioni RE, et al. Epigenetic signatures of cigarette smoking. *Circ Cardiovasc Genet*. 2016;9:436–447. https://doi.org/10.1161/CIRCGENETICS.116.001506
29. Sugden K, Hannon EJ, Arseneault L, et al. Establishing a generalized polyepigenetic biomarker for tobacco smoking. *Transl Psychiatry*. 2019;9(1):92. https://doi.org/10.1038/s41398-019-0430-9
30. Levine ME, Lu AT, Quach A, et al. An epigenetic biomarker of aging for lifespan and healthspan. *Aging (Milano)*. 2018;10(4):573–591. https://doi.org/10.18632/aging.101414
31. Charlson ME, Pompei P, Ales KL, MacKenzie CR. A new method of classifying prognostic comorbidity in longitudinal studies: development and validation. *J Chronic Dis*. 1987;40(5):373–383. https://doi.org/10.1016/0021-9681(87)90171-8
32. Glasheen WP, Cordier T, Gumpina R, Haugh G, Davis J, Renda A. Charlson Comorbidity Index: *ICD-9* update and *ICD-10* translation. *Am Health Drug Benefits*. 2019;12(4):188–197.
33. Wagner T, Moran E, Shen ML, Gehlert E. *HERC's Guide to the Nosos Risk Adjustment Score*. Health Economics Resource Center, U.S. Department of Veterans Affairs; 2024.
34. Wagner TH, Upadhyay A, Cowgill E, et al. Risk adjustment tools for learning health systems: a comparison of DxCG and CMS-HCC V21. *Health Serv Res*. 2016;51(5):2002–2019. https://doi.org/10.1111/1475-6773.12454
35. Department of Veterans Affairs, Office of Mental Health and Suicide Prevention. Joint Department of Veterans Affairs and Department of Defense Mortality Data Repository. *Data compiled from the National Death Index*. www.mirecc.va.gov/suicideprevention/documents/VA_DoD-MDR_Flyer.pdf
36. Muthén LK and Muthéén BO. *Mplus User's Guide*. 7th ed. Los Angeles, CA: Muthén & Muthén; 1998–2012.
37. Fransquet PD, Wrigglesworth J, Woods RL, Ernst ME, Ryan J. The epigenetic clock as a predictor of disease and mortality risk: a systematic review and meta-analysis. *Clin Epigenetics*. 2019;11(1):62. https://doi.org/10.1186/s13148-019-0656-7
38. Kuiper LM, Polinder-Bos HA, Bizzarri D, et al. Epigenetic and metabolomic biomarkers for biological age: a comparative analysis of mortality and frailty risk [published correction appears in J Gerontol A Biol Sci Med Sci. 2024 Apr 1;79(4):]. *J Gerontol A Biol Sci Med Sci*. 2023;78(10):1753–1762. https://doi.org/10.1093/gerona/glad137
39. Department of Veteran Affairs. National Center for Veterans Analysis and Statistics. *Veteran population*. 2024. https://www.va.gov/vetdata/veteran_population.asp
40. Department of Veteran Affairs. National Center for Veterans Analysis and Statistics. *Profile of post-9/11 veterans*. 2016. https://www.va.gov/vetdata/docs/SpecialReports/Post_911_Veterans_Profile_2016.pdf
41. Barroso A. *The changing profile of the U.S. military: Smaller in size, more diverse, more women in leadership*. Pew Research Center; 2019. https://pewrsr.ch/2N93uRV
42. Bourassa KJ, Halverson TF, Garrett ME, et al; VA Mid Atlantic MIRECC Workgroup. Demographic characteristics and epigenetic biological aging among post-9/11 veterans: associations of DunedinPACE with sex, race, and age. *Psychiatry Res*. 2024;336:115908. https://doi.org/10.1016/j.psychres.2024.115908
43. Waziry R, Ryan CP, Corcoran DL, et al. Effect of long-term caloric restriction on DNA methylation measures of biological aging in healthy adults from the CALERIE trial. *Nat Aging*. 2023;3(3):248–257. https://doi.org/10.1038/s43587-022-00357-y
44. Sehgal R, Borrus D, Kasamato J, et al.; DNAm aging biomarkers community, community Longevity interventional studies. DNAm aging biomarkers are responsive: insights from 51 longevity interventional studies in humans. *bioRxiv*. 2024:10.22.619522. https://doi.org/10.1101/2024.10.22.619522
45. Campisi J, Kapahi P, Lithgow GJ, Melov S, Newman JC, Verdin E. From discoveries in ageing research to therapeutics for healthy ageing. *Nature*. 2019;571(7764):183–192. https://doi.org/10.1038/s41586-019-1365-2