

Original Article

Difference between Biological Age and Chronological Age Predicts Mortality and Hospitalization in a Longitudinal Adult Cohort

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ABSTRACT: Biological age (BA) reflects overall health status and may deviate from chronological age (CA). Whether the magnitude of this difference (Age_{diff}) independently predicts hospitalization and mortality remains unknown. Retrospective longitudinal cohort of 2,597 adults (27.5% women), aged 21–92 years, participating in an executive screening program at Sheba Medical Center, Israel, contributing 6,772 clinical encounters between 2006 and 2020, mean follow-up 9.2 (2.2) years. BA was estimated to use a previously established deep-learning model trained on routine clinical laboratory tests. Bayesian time-dependent Cox models evaluated associations between Age_{diff} and all-cause mortality, and negative-binomial models were applied to hospitalizations, adjusting for age, sex, BMI, smoking, and baseline comorbidities. The Age_{diff} associated with a hazard ratio (HR) of 1.5 for mortality was determined for each chronological age, and $Age_{diff} \times age$ interaction was tested. Each 1-year increase in Age_{diff} was associated with a 15% higher mortality hazard (HR 1.15, 95% CI 1.10–1.21) and a 6% higher hospitalization rate (incidence rate ratio 1.06, 95% CI 1.03–1.09). The $Age_{diff} \times age$ interaction was not significant ($p = 0.56$). The Age_{diff} required to reach HR = 1.5 decreased with chronological age, from approximately 5 years at age 40 to 3 years at age 80. $Age_{diff} \geq 3$ years was associated with a clinically significant increase in mortality. In this cohort, a BA estimated from routine laboratory results exceeding CA by 3 or more years was associated with increased hospitalization and mortality risk, suggesting potential utility for risk stratification in clinical settings.

Keywords: Biological age, aging clock, mortality, hospitalization, blood biomarkers, longitudinal

INTRODUCTION

Individuals of the same chronological age (CA) often follow markedly different biological trajectories, some septuagenarian's experience frailty, multimorbidity, and recurrent hospitalizations, while others remain cognitively and physically resilient well into advanced ages. Clinical outcomes cannot be predicted by CA

because of this heterogeneity [1, 2]. Biological age (BA) is a better proxy for functional reserve and disease vulnerability than CA. BA is expensive to estimate because it usually is estimated from a constellation of molecular, biochemical, or clinical parameters [3].

The difference between biological and chronological age, often termed the age gap or Age_{diff} , can be a predictor of morbidity and adverse outcomes [4, 5]. Most existing

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BA prediction models rely on medical records, demographic, or clinical characteristics [3]. These models require information that is sometimes expensive to assemble in a healthcare facility and result in predictions that do not adequately assess physiological reserve variability. On the other hand, costs, associated with BA estimation can be reduced if they are derived from routine clinical laboratory information. Numerous laboratory test-based BA estimators have been proposed, including DNA-methylation, inflammatory, and clinical indices, and multi-omics approaches such as proteomics, glycomics, and microbiome profiling [1, 4–9]. Despite their theoretical promise, these BA estimators are less useful in clinical practice because they require specialized laboratory testing that are not always available or affordable.

Recent advances in machine learning have facilitated the development of BA estimators derived solely from routine blood chemistry results. We evaluated the clinical utility of a routine blood test-based biological aging model. Using a large real-world cohort with over a decade of follow-up, we examined whether the biological–chronological age gap ($\text{Age}_{\text{diff}} = \text{BA} - \text{CA}$) and its progression over time were associated with adverse health outcomes [4, 5]. Using classical and Bayesian survival models, we explored whether Age_{diff} was associated with long-term health outcomes such as hospitalization and mortality. We identified a model-derived exploratory reference point for Age_{diff} that may help identify individuals at elevated risk of future hospitalization or mortality.

METHODS

A retrospective cohort was sampled from the Executive Screening Program at Sheba Medical Center, a tertiary academic hospital conducting about 9,000 standardized evaluations annually. The program serves predominantly employed adults who undergo comprehensive preventive health assessments on a voluntary basis. Each visit followed a uniform protocol administered by trained physicians and certified staff, including detailed medical and lifestyle history, anthropometry, vital signs, and physical examination. Fasting blood and urine samples were analyzed in a single central laboratory operating under internal and external quality-assurance procedures. Eligible participants were aged ≥ 18 years at their first visit and had at least two assessments between January 1, 2006, and December 31, 2019. Of 45,027 individuals screened, 28,893 had ≥ 2 visits within this time frame. After restricting the cohort to participants with complete laboratory profiles required for biological age estimation, 2,597 individuals (713 women, 1,884 men, Supplementary Fig. 1) were included in the final analysis,

contributing 6,772 repeated observations (median 2 visits, IQR 2–3). To assess potential selection effects, characteristics of included and excluded individuals are presented in Supplementary Table 1. Follow-up extended through May 25, 2025, with a median duration of 9.2 years (SD 2.2), during which mortality and hospitalization outcomes were ascertained.

The institutional review board of Sheba Medical Center approved the study; individual informed consent was waived due to the use of de-identified secondary data

Exposure variable

Biological age based on routine blood tests was estimated using the BloodAge clock, available via the online SenoClock platform (www.deeplongevity.com/senoclock) developed by Deep Longevity, Hong Kong (subsidiary of Regent Pacific 00575.HK). [10] The SenoClock- BloodAge algorithm is a modular ensemble of 21 deep neural networks (DNNs) trained on more than 60,000 samples from routine blood biochemistry and cell count tests. A total of 36 standard blood-based biomarkers: covering metabolic, hepatic, renal, hematologic, and inflammatory domains were included in the biological age estimation (Supplementary Table 2). De-identified laboratory data were securely transmitted for age prediction, and BA estimates were returned for research use only. We defined $\text{Age}_{\text{diff}} = \text{BA} - \text{CA}$, representing the difference between biological and chronological age; higher values suggest accelerated aging.

In the present study, BloodAge was applied as an externally derived measure without local retraining or recalibration. To our knowledge, the model has not been formally validated in an Israeli population and is therefore used here as a real-world application rather than a formal transportability or validation framework.

Outcomes

The primary clinical endpoints were all-cause mortality and hospitalizations. Mortality was ascertained through deterministic linkage with the Israeli Ministry of Health National Mortality Registry using national identification numbers, with follow-up censored on May 25, 2025. Hospitalization data: including admission and discharge dates, durations, and ICD-coded discharge diagnoses, were extracted from Sheba's electronic health-record system for the same observation period.

Covariates

Baseline covariates were collected at the first visit through physician-administered assessments. These included sex,

age, body-mass index (BMI), systolic blood pressure, and smoking status. Comorbidities were recorded such as diabetes mellitus, hypertension, ischemic heart disease, myocardial infarction, chronic kidney disease, cerebrovascular disease, and peripheral artery disease. Medication use (e.g., statins, antiplatelets, GLP-1 receptor agonists, SGLT2 inhibitors) and history of coronary revascularization (PCI or CABG) were verified through medical records.

Statistical analysis

Continuous variables are presented as mean (SD) or median (IQR) for skewed data, and categorical variables as n (%). Between-group comparisons used t tests or Wilcoxon rank-sum tests for continuous data (as appropriate) and χ^2 tests for categorical data. Biological age (BA), chronological age (CA), and Age_{diff} were considered as years; Age_{diff} was defined as $\text{BA} - \text{CA}$. Implausible physiologic entries (e.g., $\text{BMI}=0$, systolic blood pressure coded as 0) were treated as missing.

Descriptive cohort characteristics: Baseline characteristics were shown for the entire cohort also stratified by sex. Person-time was accrued from the index visit until death or administrative censoring. Repeat visits were included in longitudinal models. Covariate adjustment for baseline conditions used data obtained at the index visit.

Cox models [11] were estimated to determine the Age_{diff} threshold associated with a need for risk-mitigation intervention. Clinically significant high risk was defined as having a 50% higher risk of mortality ($\text{HR}=1.5$) at all CAs. Independent variables in the Cox model included Age_{diff} (updated at each visit), normalized age and the $\text{Age}_{\text{diff}} \times \text{age}$ interaction. Cluster-robust standard errors by participant accounted for repeated observations. The models were adjusted for sex, baseline CA, BMI, smoking, and baseline hypertension. Models were adjusted only for variables independent of the biological age algorithm, to prevent over-fitting [12]. Proportional-hazards assumptions were examined using global and covariate-specific Schoenfeld residuals [13]. Approximate linearity of the Age_{diff} hazard association was examined using martingale residuals, and individual influence was assessed with dfbeta statistics, ensuring model stability and robustness. These diagnostic procedures were performed according to standard survival modeling guidelines [14].

Hospitalizations: health-care utilization was modelled as panel counts using quasi-Poisson regression with a log link and a person-time offset [15], with Age_{diff} (per +1 year) as the exposure and the same covariate set specified above. We assessed dispersion and residual

diagnostics and report incidence rate ratios (IRRs) with 95% CI's.

Kaplan–Meier curves [16] stratified by baseline $\text{Age}_{\text{diff}} \geq 3$ vs < 3 years with numbers at risk and a log-rank test were created. For visualization purpose, a smoothed overlay was generated by applying a LOESS smoother ($\text{span} = 0.25$) to the estimated cumulative event probabilities ($1 - S(t)$) derived from the Kaplan–Meier curves, this smoothing was used for visualization only, and all statistical inference was based on the original stepwise Kaplan–Meier estimates. Baseline characteristics were compared between $\text{Age}_{\text{diff}} < 3$ and ≥ 3 years using t-tests for continuous variables and chi-square tests for categorical variables.

Sensitivity and post-hoc analyses were performed. To assess the potential impact of residual confounding, additional Cox models including baseline comorbidities (e.g., diabetes, cardiovascular disease, and chronic kidney disease) were fitted. Robustness of hospitalization models to over-dispersion and within-person correlation was evaluated using Bayesian negative-binomial mixed-effects models with participant-level random intercepts, reporting posterior incidence rate ratios (IRRs) with 95% credible intervals. Age-quartile empirical cut-point optimization (based on the log-rank statistic) was performed and compared with model-based $\text{HR}=1.5$ thresholds.

Missing data, and censoring. Less than 4% of data were missing. There was no missing data for BA, CA, Age_{diff} , deaths, and hospitalizations. We used complete-case analysis for adjusted models. Participants were administratively censored at last known contact; numbers at risk for each time interval are shown for the Kaplan–Meier display.

Reporting guideline

This cohort study was reported in accordance with the STROBE statement for cohort studies. A completed STROBE checklist is provided in the Supplementary Table 3.

Software

Analyses were conducted in R version 4.5.0 using the packages: survival, survminer, brms, cmdstanr, dplyr, tidyr, ggplot2, cowplot, readxl, Loess and MASS.

RESULTS

A total of 2,597 participants (713 women [27.5%], 1,884 men) had 6,772 assessments (median 2 visits, IQR 2–3); 62.8% had two visits, 22.8% three, and 12.5% four or

more. Mean follow-up was 9.2 (2.2) years, yielding approximately 23,892 person-years of observation. At the index visit, the mean CA was 60.0 ± 11.1 years, and mean BA was 61.6 (11.7) years, with an average Age_{diff} (BA – CA) of 1.5 (3.0) years. Women had a smaller Age_{diff} than men (1.1 vs 1.7 years; $p < 0.001$). Men had higher systolic blood pressure (120.6 vs 114.1 mmHg; $p < 0.001$) and BMI (26.2 vs 24.3 kg/m²; $p < 0.001$); smoking prevalence

was low and similar between sexes ($\approx 5\%$). Cardiometabolic morbidity was more frequent among men; ischemic heart disease 16% vs 2%, myocardial infarction 4% vs 1%, atherosclerosis 18% vs 4% (all $p < 0.001$), consistent with greater use of medications used to treat these conditions. Baseline characteristics are summarized in Table 1. During follow-up, 144 deaths (5.6%) and 1,269 hospitalizations were recorded.

Table 1. Baseline characteristics of first visit for all participants (2006-2019).

Variable	Total	Women	Men	P- value	Missing N%
n	2597	713	1884		
Average Follow-up years (SD)	9.2 (2.2)	9.3 (2.2)	9.14 (2.1)	0.023	0
Average number of visits (SD)	2.6 (1.0)	2.6 (1.0)	2.6 (1.0)	0.85	0
Chronological Age (SD)	60.0 (11.1)	59.9 (11.0)	60.1 (11.2)	0.731	0
Biological Age (SD)	61.6 (11.7)	61.1 (11.8)	61.8 (11.7)	0.173	0
Chronological age- Biological Age_{diff} (SD)	1.5 (3.0)	1.1 (2.9)	1.7 (3.1)	<.001	0
Systolic blood pressure (SD)	118.9 (27.6)	114.06 (27.9)	120.62 (27.3)	<.001	96 (4%)
BMI (SD)	25.7 (6.7)	24.27 (6.6)	26.18 (6.6)	<.001	107 (4%)
Smoking, N (%)	130 (5%)	40 (6%)	90 (5%)	0.442	0
Outcomes					
Mortality	144 (6%)	36 (5%)	108 (6%)	0.560	0
Participants with > 1 hospitalization	276 (11%)	70 (10%)	206 (11%)	0.036	0
Diagnosis					
Acute Myocardial Infarction (heart attack), N (%)	83 (3%)	5 (1%)	78 (4 %)	<.001	0
Chronic Kidney Disease, N (%)	18 (1%)	1 (0%)	17 (1%)	0.067	0
Diabetes Mellitus, N (%)	431 (16%)	71 (10%)	360 (19%)	<.001	0
Peripheral Artery Disease, N (%)	51 (2%)	7 (1%)	44 (2%)	0.039	0
Transient Ischemic Attack, N (%)	36 (1%)	5 (1%)	31 (2%)	0.099	0
Presence of atherosclerosis, N (%)	359 (14%)	26 (4%)	333 (18%)	<.001	0
Hypertension diagnosis, N (%)	966 (37%)	186 (26%)	780 (41%)	<.001	0
Ischemic Heart Disease, N (%)	316 (12%)	15 (2%)	301 (16%)	<.001	0
Cerebrovascular Accident (stroke), N (%)	51 (2%)	11 (2%)	40 (2%)	0.428	0
Percutaneous Coronary Intervention, N (%)	181 (7 %)	6 (1%)	175 (9 %)	<.001	0
Coronary Artery Bypass Graft, N (%)	58 (2%)	0 (0%)	58 (3%)	<.001	0
Medication usage					
Aspirin use, N (%)	392 (15%)	61 (9%)	331 (18%)	<.001	0
Plavix use, N (%)	78 (3.0%)	6 (0.8%)	72 (4%)	<.001	0
Statin use, N (%)	566 (22%)	130 (18%)	436 (23%)	0.008	0
GLP-1 receptor agonist use, N (%)	11 (0%)	1 (0%)	10 (1%)	0.303	0
SGLT2 inhibitor use, N (%)	5 (0%)	1 (0%)	4 (0%)	1.000	0

In the study cohort, biological age was highly correlated with chronological age ($r = 0.966$), with a mean absolute error (MAE) of 2.46 years. These values are within the expected range reported in the original model development study ($r \approx 0.91$; MAE ≈ 5.5 years) [10], and likely reflect cohort-specific characteristics rather than differences in underlying model performance.

Using all the repeated assessments in time-to-event Cox modeling, the $\text{Age}_{\text{diff}} \times \text{age}$ interaction was not significant ($p = 0.56$), suggesting no effect modification of Age_{diff} by age itself. Although the $\text{Age}_{\text{diff}} \times \text{age}$ interaction was not statistically significant, model-derived estimates suggest that as CA increased, the Age_{diff} associated with a mortality hazard of 1.5 decreased; from

approximately ~ 5 years at 40 years of age to ~ 3 years at age 80. Based on this model-derived relationship, an Age_{diff} of 3 years was used as exploratory reference point corresponding to a clinically meaningful increase in mortality risk (Fig. 1). This threshold was exceeded in 1,966 (29%) measurements occurring in 725 (28%) participants.

To assess the effect of the 3-year Age_{diff} model-derived reference point, we evaluated the strength of association between Age_{diff} and mortality on the continuous scale. Using a time-dependent Cox framework with counting-process intervals (Age_{diff} updated at each visit; covariates fixed at baseline), with values defined at the beginning of each interval and used to predict

subsequent risk, ensuring correct temporal alignment between exposure and outcome, and cluster-robust standard errors by participant ID, each 1-year increment Age_{diff} was associated with a 15% higher hazard of death (HR 1.15, 95% CI 1.10–1.21; $p = 2.6 \times 10^{-8}$) after adjustment for sex, baseline chronological age, BMI, smoking, and baseline hypertension. This approach minimizes the risk of immortal time bias by assigning exposure status within discrete observation intervals and preserves correct temporal alignment between exposure and outcome. Model diagnostics supported proportional hazards (global Schoenfeld $p = 0.42$) and an approximately linear log-hazard for Age_{diff} (martingale

residuals); influence analyses (dfbeta) did not identify outliers materially altering the coefficient. Figure 2 illustrates these continuous associations, with adjusted HRs and 95% CIs, confirming the robustness and dose-response nature of the Age_{diff} mortality relationship. Sensitivity analyses with additional adjustment for baseline comorbidities showed consistent results, with Age_{diff} remaining significantly associated with mortality (HR 1.11, 95% CI, 1.04–1.18; $p = 0.002$; Supplementary Table 4). Yielding consistent results and reducing the likelihood that the observed association is driven by short-term disease processes or reverse causation.

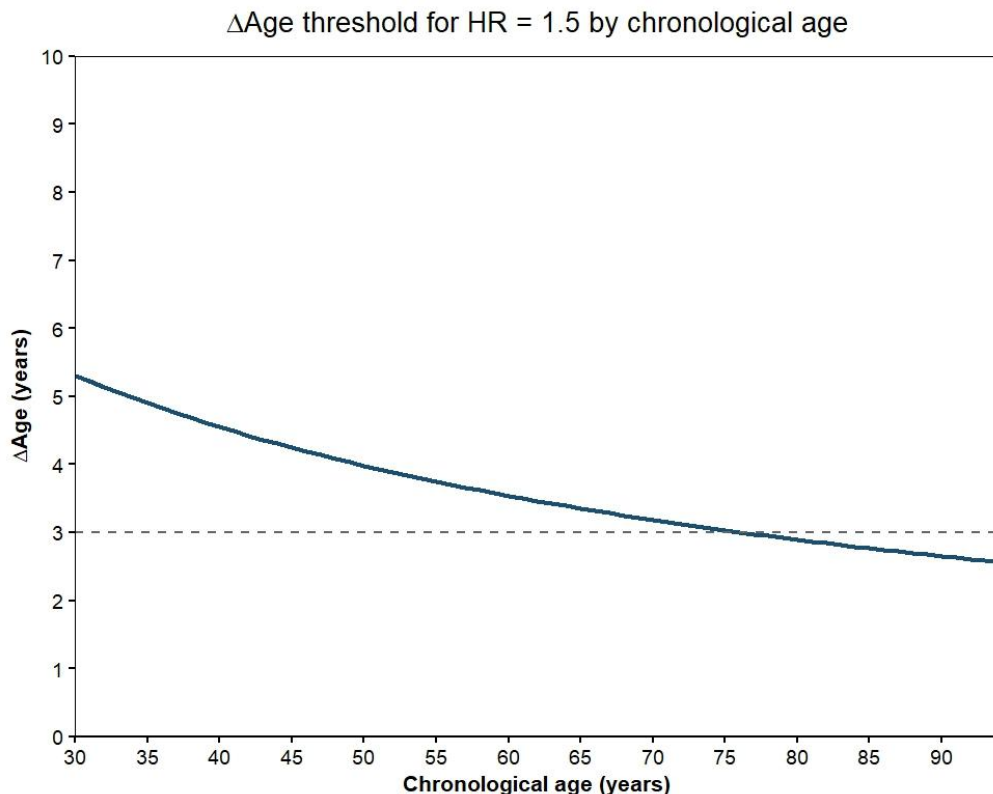


Figure 1. Model-derived Age_{diff} required to reach HR=1.5 across chronological age (30–94 years). Estimates are from a time-dependent Cox model using all repeated assessments; the $\text{Age}_{diff} \times \text{age}$ interaction was not significant ($p = 0.56$). Model-derived estimates suggest that the required Age_{diff} declines with age (≈ 5 years at age 40 to ≈ 3 years at age 80). The dashed line marks a pragmatic, model-derived 3-year reference threshold.

During the follow-up period, there were 1,269 hospitalizations (mean of 0.49 (1.1) per participant), with approximately 80% of participants experiencing no hospitalizations, indicating a highly skewed count distribution. Event-level summaries showed similar patterns: total hospitalization counts did not differ significantly between men and women (956 vs. 313 events; $p = 0.305$). Similarly, while participants with $\text{Age}_{diff} \geq 3$ years had a higher cumulative number of hospitalizations compared with those with $\text{Age}_{diff} < 3$

years (464 vs. 805 events), this difference did not reach statistical significance ($p = 0.065$).

To appropriately model this outcome, we used quasi-Poisson regression with a log person-time offset, allowing for overdispersion relative to the Poisson assumption while estimating incidence rate ratios (IRRs), with the number of hospitalizations as the dependent variable, and sex, baseline CA and hypertension status, BMI, and smoking history as covariates. Each 1-year increase in Age_{diff} was associated with a greater admission rate (IRR 1.06, 95% CI, 1.03–1.09; $p < 0.0001$; Fig. 3).

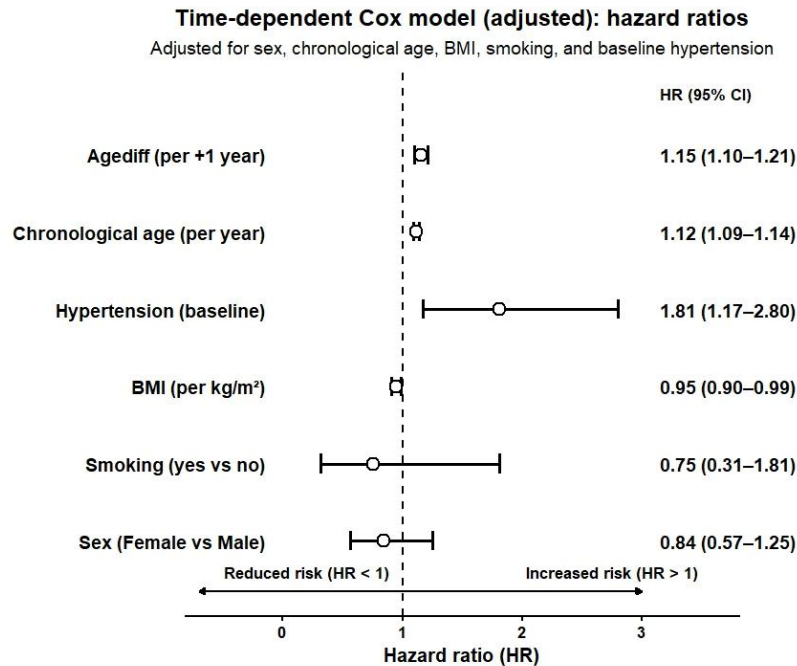


Figure 2. Forest-style display of hazard ratios (HR) from a Cox model that treats Age diff (biological–chronological age gap) as a continuous exposure (per +1 year). Models were fit in a counting-process (start–stop) framework over repeated visits with cluster-robust standard errors (cluster[id]) and were adjusted for sex, baseline chronological age, smoking, BMI, and baseline hypertension. Points denote HR estimates; horizontal bars show 95% CIs; the dashed vertical line marks HR=1. The x-axis is linear (0–3). Directional arrows below the axis indicate reduced risk (left of 1) and increased risk (right of 1). Variable names are displayed at the left margin; the rightmost column reports HR (95% CI). Abbreviations: HR=hazard ratio; CI=confidence interval; BMI=body-mass index; HTN=hypertension; Age_{diff}=BA–CA (biological age minus chronological age).

To further assess the robustness of these findings to distributional assumptions and within-person correlation, we implemented Bayesian negative binomial mixed-effects models with participant-level random intercepts. These models explicitly account for excess dispersion and heterogeneity across individuals, yielding a consistent effect estimate (posterior IRR 1.08, 95% CrI 1.06–1.12, Supplementary Table 5) with moderate dispersion (shape) ≈ 0.25 . In addition, visit-level count models with clustered standard errors produced similar results (adjusted IRR 1.08, 95% CI, 1.04–1.11; $p < 0.0001$), supporting the robustness of the association across modeling strategies.

Collectively, these complementary approaches, addressing overdispersion, excess zeros, and within-subject correlation, demonstrate that biological-age acceleration is consistently associated with a greater hospitalization burden, independent of measured confounders. For clinical interpretation, a 3-year increase in Age_{diff} corresponds to an approximately 19% higher hospitalization rate (Supplementary Table 4).

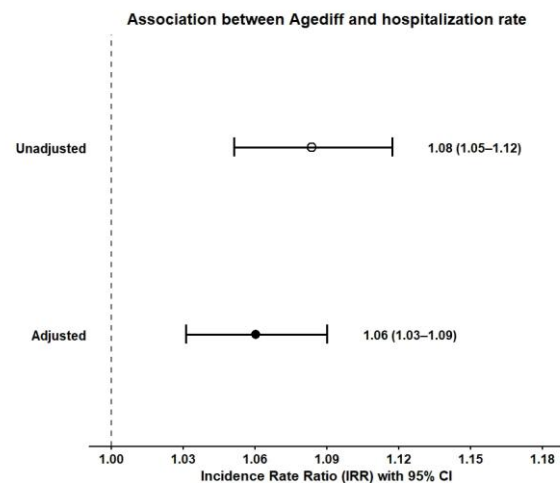


Figure 3. Incidence rate ratios (IRR) with 95% CIs from quasi-Poisson models (log link) including an offset for follow-up time. Continuous exposure Age_{diff} (BA–CA) per +1 year. Open circles = unadjusted; filled circles = adjusted for sex, baseline chronological age, smoking, BMI, and baseline hypertension. Vertical dashed line marks IRR=1.

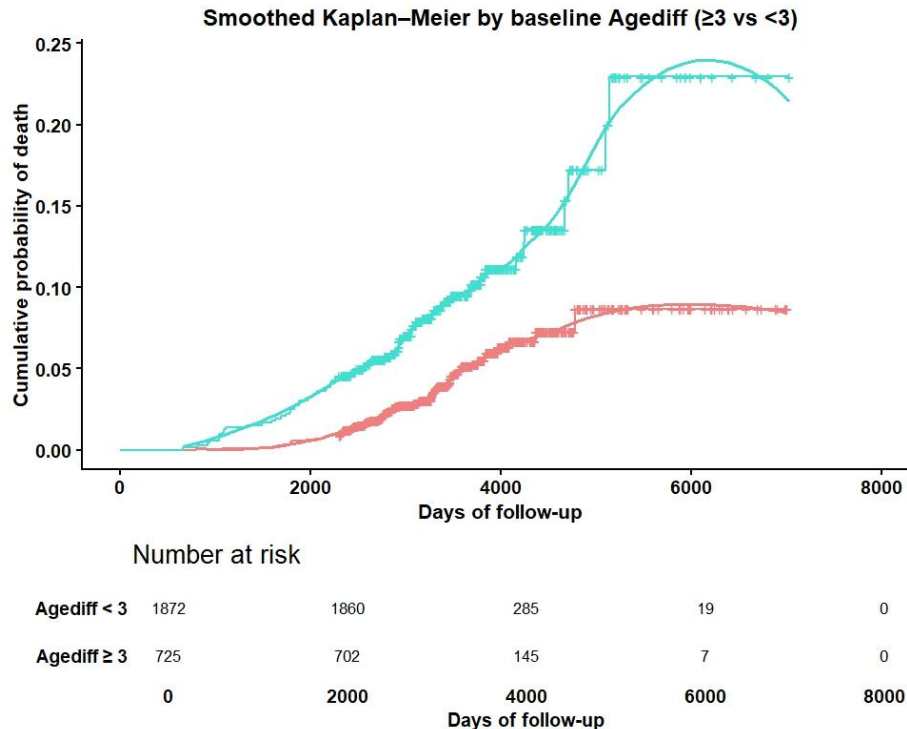


Figure 4. Kaplan–Meier curves depicting the cumulative probability of death overtime according to baseline age acceleration status, shown with a LOESS-smoothed overlay for visualization. Participants with biological age ≥ 3 years older than chronological age ($\text{Age}_{\text{diff}} \geq 3$; turquoise line) exhibited substantially higher mortality risk compared with those with smaller differences ($\text{Age}_{\text{diff}} < 3$; coral line). Tick marks denote censoring events. Numbers at risk are displayed below the plot at selected time points.

Kaplan–Meier curves showing the mortality-time relationship are shown in Figure 4 stratified for $\text{Age}_{\text{diff}} \geq 3$ vs < 3 , with a smoothed overlay for visualization. There is a clear separation of the curves over 8,000 days of follow-up ($n=725$ and $n=1,872$, respectively). At 8,000 days, cumulative mortality was 23.0% (95% CI, 12.7–32.0) in the ≥ 3 -year group compared with 8.7% (95% CI, 5.2–12.0) in the < 3 -year group; the difference was significant by the log-rank test ($\chi^2=26.16$; $p = 3.14 \times 10^{-7}$). The characteristics of participants who had $\text{Age}_{\text{diff}} \geq 3$ vs < 3 are shown in Table 2. The mean Age_{diff} was 5.5 years with the largest differences in baseline characteristics between groups being in diabetes and hypertension. Transition patterns of Age_{diff} across repeated visits demonstrated substantial longitudinal variability (Supplementary Fig. 2). While 53.9% of participants remained consistently below the ≥ 3 -year threshold, a considerable proportion exhibited transitions over time, including up-crossers (13.9%), down-crossers (12.2%), and multi-directional changes (7.5%).

DISCUSSION

In this longitudinal cohort of 2,597 adults contributing 6,772 repeated assessments ($\approx 23,892$ person-years),

Age_{diff} calculated from routine blood tests was independently associated with hospitalization and mortality after adjustment for measured covariates. Following adjustment for sex, baseline CA, BMI, smoking, and hypertension, each additional year of Age_{diff} was associated with a 15% greater hazard of death (HR 1.15, 95% CI, 1.10–1.21). Similarly, Age_{diff} was predictive of an increased risk for hospitalization (IRR 1.06, 95% CI, 1.03–1.09). However, the magnitude of this association was modest and should be interpreted with caution, particularly given the relatively low hospitalization rate in this cohort. These results are consistent with prior work showing that BA indices outperform chronological age for risk discrimination [6, 17–22]. Compared with more established biological age measures such as epigenetic or proteomic clocks, this laboratory-based Age_{diff} approach relies on widely available tests and is more readily applicable in routine clinical settings, although it may provide less biological specificity.

Because the BA model used in the current study relies on standard chemistry and hematology panels, Age_{diff} can be recomputed during any clinical encounter. Age_{diff} and risk factor profiles change in parallel (Supplementary Fig. 1), and the observed transition patterns indicate that

Age_{diff} status is not fixed over time, with individuals moving between risk categories across follow-up. These findings suggest that repeated assessment of Age_{diff} may better capture the dynamic nature of biological aging

compared to a single measurement. Accordingly, Age_{diff} may serve as a useful indicator associated with increased risk, with potential relevance for longitudinal risk stratification and patient monitoring.

Table 2. First visit baseline characteristics, stratified by <3 vs ≥3 years Age_{diff}.

Variable	Age _{diff} < 3y	Age _{diff} ≥ 3y	p-value
n	1872	725	
Average follow-up (years)	9.2 (2.0)	9.3 (2.4)	0.47
Chronological age (SD)	59.6 (11.0)	61.3 (11.4)	0.00037
Biological age (SD)	59.5 (11.1)	66.8 (11.6)	<0.0001
Age _{diff} (BA–CA), years	-0.0 (1.6)	5.5 (2.0)	<0.0001
BMI (SD)	25.4 (6.6)	26.4 (6.8)	0.00028
Female, n (%)	544 (29%)	169 (23%)	0.0038
Smoking, n (%)	102 (5%)	28 (4%)	0.12
Outcomes			
Mortality, n (%)	75 (4%)	69 (10%)	<0.0001
Participants with >1 hospitalization, n (%)	181 (10%)	95 (13%)	0.013
Diagnosis			
Acute myocardial infarction, n (%)	58 (3%)	25 (3%)	0.74
Chronic kidney disease, n (%)	5 (0%)	13 (2%)	<0.0001
Diabetes mellitus, n (%)	232 (12%)	189 (26%)	<0.0001
Peripheral artery disease, n (%)	30 (2%)	21 (3%)	0.048
Transient ischemic attack, n (%)	25 (1%)	11 (2%)	0.87
Presence of atherosclerosis, n (%)	249 (13%)	110 (15%)	0.24
Hypertension diagnosis, n (%)	659 (35%)	307 (42%)	0.00086
Ischemic heart disease, n (%)	223 (12%)	93 (13%)	0.57
Cerebrovascular accident (stroke), n (%)	31 (2%)	20 (3%)	0.097
Percutaneous coronary intervention, n (%)	134 (7%)	47 (6%)	0.6
Coronary artery bypass graft, n (%)	35 (2%)	23 (3%)	0.062
Medication usage			
Aspirin use, n (%)	260 (14%)	132 (18%)	0.007
Plavix use, n (%)	53 (3%)	25 (3%)	0.48
Statin use, n (%)	390 (21%)	176 (24%)	0.064
GLP-1 receptor agonist use, n (%)	4 (0%)	7 (1%)	0.014
SGLT2 inhibitor use, n (%)	1 (0%)	4 (1%)	0.023

We considered that a 50% increase in the hazard of death is clinically significant. This was associated with an approximate Age_{diff} of 3 years (Fig. 1). We therefore used an Age_{diff} of 3 years as a model-derived exploratory reference point that may help contextualize increased mortality risk. Age_{diff} ≥ 3 years occurred in 1,966 (29%) measurements occurring in 725 (28%) participants. This difference was supported by the Kaplan–Meier curve showing clear separation between Age_{diff} groups over the entire 8,000-day observation period. The cumulative mortality for participants with Age_{diff} ≥ 3 years was 23.0% compared with 8.7% for Age_{diff} <3 years (log-rank $\chi^2=26.16$; $p = 3.14 \times 10^{-7}$; Fig. 4). However, the continuous association between Age_{diff} and mortality represents the primary and most robust finding.

Calculating BA and determining the Age_{diff} is a simple measurement that might help identify individuals at higher risk in selected clinical settings. Based on this analysis, an Age_{diff} of 3 years may serve as a practical

reference point, although further validation in independent cohorts is required before clinical use. Accordingly, the clinical applicability of these findings should be interpreted cautiously outside similar screening populations.

This study has limitations: 1) Age_{diff} is a surrogate marker for disease-mortality relationships. It cannot be considered causal. It may also reflect, in part, underlying disease burden captured by its component biomarkers. 2) The use of a proprietary, externally developed biological age algorithm. The BloodAge model was not retrained or recalibrated in the current population and has not been formally validated in an Israeli cohort. As such, potential population-specific biases and limitations in transportability cannot be excluded. Moreover, the performance metrics reported in this study reflect internal consistency rather than formal external validation. In addition, the proprietary nature of the model limits full transparency and independent reproducibility and may

affect its applicability across laboratory platforms. 3) The hospitalizations we measured were in a single health-care system. We may have underestimated hospitalization rates by not capturing patient admission to hospitals outside of our system. This potential under-ascertainment is likely to be non-differential and may have attenuated, rather than inflated, the observed association. 4) Our study population was generally healthy undergoing voluntary executive screening at a single center, with male predominance and under-representation of women, potentially limiting the generalizability of our results. In addition, restriction on participants with repeated visits and complete laboratory profiles may have introduced selection effects, potentially enriching the analytic cohort for individuals with greater healthcare engagement and higher disease burden.

Conclusions

Our findings show that Age_{diff} , modelled as a time-varying factor might provide a simple, laboratory-based measure that may capture clinically relevant risk for both mortality and hospitalization. Future studies are needed to evaluate the generalizability of these findings and to further examine the potential role of Age_{diff} , including thresholds such as >3 years, across diverse populations and clinical settings. Risk factors and their relative contribution to higher mortality should be identified as targets for risk mitigation. Prospective studies of risk factor interventions and their impact on mortality will be needed to determine the utility of Age_{diff} as a clinically useful tool to identify patients who may benefit from this approach.

Authors contribution

AG conceived the study, performed the statistical analyses, interpreted the data, and drafted the manuscript. EB contributed to the conceptualization and provided critical input on data interpretation and manuscript editing. EHL substantially contributed to the study concept and design, data interpretation, statistical analysis, and critical revision of the manuscript for important intellectual content. DK, NH, and MCS assisted with data processing and reviewed the manuscript. SS and YM contributed to data acquisition and clinical interpretation. TS contributed to the initial study concept and critically reviewed the manuscript. All authors reviewed and approved the final version of the manuscript.

Disclosures

None to declare.

Ethics Approval and Consent to Participate

This study was approved by the Institutional Review Board (Helsinki Committee) of Sheba Medical Center. All procedures were conducted in accordance with the Declaration of Helsinki and relevant institutional guidelines. As this was a retrospective

analysis of anonymized data, informed consent was waived by the institutional ethics committee.

Data and Code Availability

The data underlying this study contain sensitive clinical information and are therefore not publicly available. De-identified datasets and analysis code are available from the corresponding author upon reasonable request and after completion of a data-sharing agreement, in accordance with institutional policies. The biological age algorithm (BloodAge/SenoClock) is proprietary and not accessible for independent reproduction.

Supplementary Materials

The Supplementary data are available online at: www.aginganddisease.org/EN/10.14336/AD.2026.00260.

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