SUPPLEMENTARY DATA

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

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SUPPLEMENTARY DATA

Supplementary Table 1. Comparison of Included Vs. Excluded participants

Variable	Exclude	Included	p-value
n	42,430	2,597	
Chronological Age (SD)	48.4 (10.6)	60.0 (11.1)	<0.001
Systolic Blood Pressure (SD)	120.0 (16.4)	123.4 (15.1)	<0.001
BMI (SD)	26.1 (4.8)	26.8 (4.1)	<0.001
Male, N(%)	27,280 (64%)	1,884 (73%)	<0.001
Smoking, N(%)	3,280 (8%)	130 (5%)	<0.001
Outcomes			
Mortality, N(%)	1,730 (4%)	144 (6%)	0.024
Participants with >1 hospitalization, N(%)	567 (1%)	276 (10%)	<0.001
Diagnosis			
Acute Myocardial Infarction, N(%)	430 (1%)	83 (3%)	<0.001
Chronic Kidney Disease, N(%)	35 (0%)	18 (1%)	<0.001
Diabetes Mellitus, N(%)	1,769 (4%)	431 (16%)	<0.001
Peripheral Artery Disease, N(%)	121 (0%)	51 (2%)	<0.001
Transient Ischemic Attack, N(%)	97 (0%)	36 (1%)	<0.001
Presence of Atherosclerosis, N(%)	1,569 (4%)	359 (14%)	<0.001
Hypertension Diagnosis, N(%)	5,219 (12%)	966 (37%)	<0.001
Ischemic Heart Disease, N(%)	1,308 (3%)	316 (12%)	<0.001
Cerebrovascular Accident (Stroke), N(%)	231 (1%)	51 (2%)	<0.001
Percutaneous Coronary Intervention, N(%)	228 (1%)	181 (7%)	<0.001
Coronary Artery Bypass Graft, N(%)	175 (0%)	58 (2%)	<0.001
Medication usage			
Aspirin Use, N(%)	959 (2%)	392 (15%)	<0.001
Plavix Use, N(%)	183 (0%)	78 (3%)	<0.001
Statin Use, N(%)	490 (1%)	566 (22%)	<0.001
GLP-1 Receptor Agonist Use, N(%)	136 (0%)	11 (0%)	0.474
SGLT2 Inhibitor Use, N(%)	14 (0%)	5 (0%)	<0.001

SUPPLEMENTARY DATA

Supplementary Table 2. Laboratory Blood Test Parameters and Corresponding Units of Measurement used for Blood age calculations.

Abbreviation	Full Name	Unit
ALP	Alkaline Phosphatase	IU/l
ALB	Albumin	g/dl
ALT	Alanine Aminotransferase	IU/l
AST	Aspartate Aminotransferase	IU/l
BASO%	Basophils (percentage)	%
BILIT	Total Bilirubin	mg/dl
BUN	Blood Urea Nitrogen	mg/dl
CA	Calcium	mg/dl
CHOLT	Total Cholesterol	mg/dl
CL	Chloride	meq/l
CREA	Creatinine	mg/dl
EOS%	Eosinophils (percentage)	%
GGT	Gamma-Glutamyl Transferase	IU/l
GLC	Glucose	mg/dl
GLOBT	Globulin	g/dl
HCT	Hematocrit	%
HGB	Hemoglobin	g/dl
K+	Potassium	meq/l
LDH	Lactate Dehydrogenase	IU/l
LDL	Low-Density Lipoprotein	mg/dl
LYMPH%	Lymphocytes (percentage)	%
MCHC	Mean Corpuscular Hemoglobin Concentration	g/dl
MCH	Mean Corpuscular Hemoglobin	pg
MCV	Mean Corpuscular Volume	fL
MONO%	Monocytes (percentage)	%
MPV	Mean Platelet Volume	fL
NA+	Sodium	meq/l
NEUTR%	Neutrophils (percentage)	%
PLT	Platelets	K/microL
PROT	Total Protein	g/dl
P	Phosphorus	mg/dl
RBC	Red Blood Cell Count	M/microL
RDW	Red Cell Distribution Width	%
TRIG	Triglycerides	mg/dl
UA	Uric Acid	mg/dl
WBC	White Blood Cell Count	K/microL

Supplementary Table 2. † Adjusted for sex, baseline chronological age, smoking, BMI, and baseline hypertension; offset = log(follow-up time in years).‡ Negative binomial overdispersion (shape≈0.25; θ≈4); random-intercept SD ≈ 1.32 (95% CrI 1.13–1.51) in the group model; continuous model similar. CrI = credible interval.

SUPPLEMENTARY DATA

Supplementary Table 3. STROBE Statement—Checklist of items that should be included in reports of cohort studies

	Item No	Recommendation
Title and abstract	1	p.1 – Title and structured abstract summarizing objectives, methods, results, and conclusions.
Introduction		
Background/rationale	2	pp.2-3 – Introduction, paragraphs 1–2: explains rationale and limitations of chronological age as a risk marker.
Objectives	3	p.3 – Last paragraph of Introduction: study hypotheses and aims clearly defined.
Methods		
Study design	4	p.3 – Methods, paragraph 1: retrospective longitudinal cohort study design described.
Setting	5	pp.3–4 – The institute for Medical Screening, Sheba Medical Center; recruitment 2006–2019.
Participants	6	pp.4-5 – Eligibility criteria, inclusion/exclusion, and participant numbers.
Variables	7	pp.4–5 – Exposure (Agediff), outcomes, and covariates clearly defined.
Data sources/ measurement	8*	pp.4–6 – Data collection, laboratory analyses, and linkage to national mortality registry.
Bias	9	pp.5–6 – Addressed via data validation, exclusion of implausible values, and covariate control.
Study size	10	p.5 – Based on all eligible participants meeting inclusion criteria (n=2,597).
Quantitative variables	11	pp.5–6 – Continuous variable handling, residual diagnostics (martingale residuals, dfbeta).
Statistical methods	12	pp.5–6 – Cox and Poisson models, cluster-robust SEs, Bayesian validation, proportional hazards checks.
Results		
Participants	13*	p.7 – Eligibility criteria, inclusion/exclusion, and participant numbers.
Descriptive data	14*	p.7 – Baseline characteristics by sex and overall cohort (Table 1).
Outcome data	15*	pp.9-10 – Mortality and hospitalization data (Tables 1–2, Figures 1–4).
Main results	16	pp.7–8 – Continuous and threshold-based Agediff analyses, HRs and IRRs with CIs.
Other analyses	17	pp.8 – Sensitivity, Bayesian mixed-effects, and cut-point optimization analyses.
Discussion		
Key results	18	pp.9-10- Main results
Limitations	19	pp.10-11– Discussion, paragraph 6: limitations including bias, confounding, and generalizability.

Interpretation	20	pp.9-10 – Discussion, integration with prior work, and clinical meaning of Agediff threshold.
Generalisability	21	p.11 – Discussion, paragraph 1: applicability of findings to other populations.

* Information separately for exposed and unexposed groups.

Supplementary Table 4. Time-dependent Cox model for mortality with extended adjustment (sensitivity analysis)

Variable	HR	95% CI	p-value
Agediff (per 1 year)	1.11	1.04 – 1.18	0.002
Chronological age	1.11	1.08 – 1.13	<0.001
Sex	0.94	0.56 – 1.57	0.81
Smoking	0.48	0.17 – 1.37	0.17
BMI	0.96	0.91 – 1.02	0.20
Hypertension	1.33	0.77 – 2.28	0.31
Ischemic heart disease (IHD)	0.68	0.27 – 1.71	0.41
Diabetes mellitus	1.04	0.63 – 1.73	0.87
Atherosclerosis	2.23	0.96 – 5.17	0.061
Acute myocardial infarction (AMI)	1.68	0.85 – 3.34	0.14
Chronic kidney disease (CKD)	2.23	1.02 – 4.85	0.043

Supplementary Table 5. Association of biological age acceleration with hospitalization burden

Model family	Exposure (ΔAge)	Adjustment	Effect (IRR/ Posterior IRR)	95% interval/ CrI	p-value	Notes
Quasi-Poisson	Continuous, per +1 year	Unadjusted	.084	1.05–1.12	2.11×10^{-7}	Offset=log (follow-up years)
Quasi-Poisson	Continuous, per +1 year	Adjusted†	1.06	1.03–1.09	3.68×10^{-5}	Robust to overdispersion (quasi-likelihood)
Bayesian Negative Binomial	Continuous, per +1 year	Random intercept (ID)	1.08	1.06–1.12	—	Convergence: $\hat{R}\approx1.00–1.01$; ESS adequate; shape≈0.25‡

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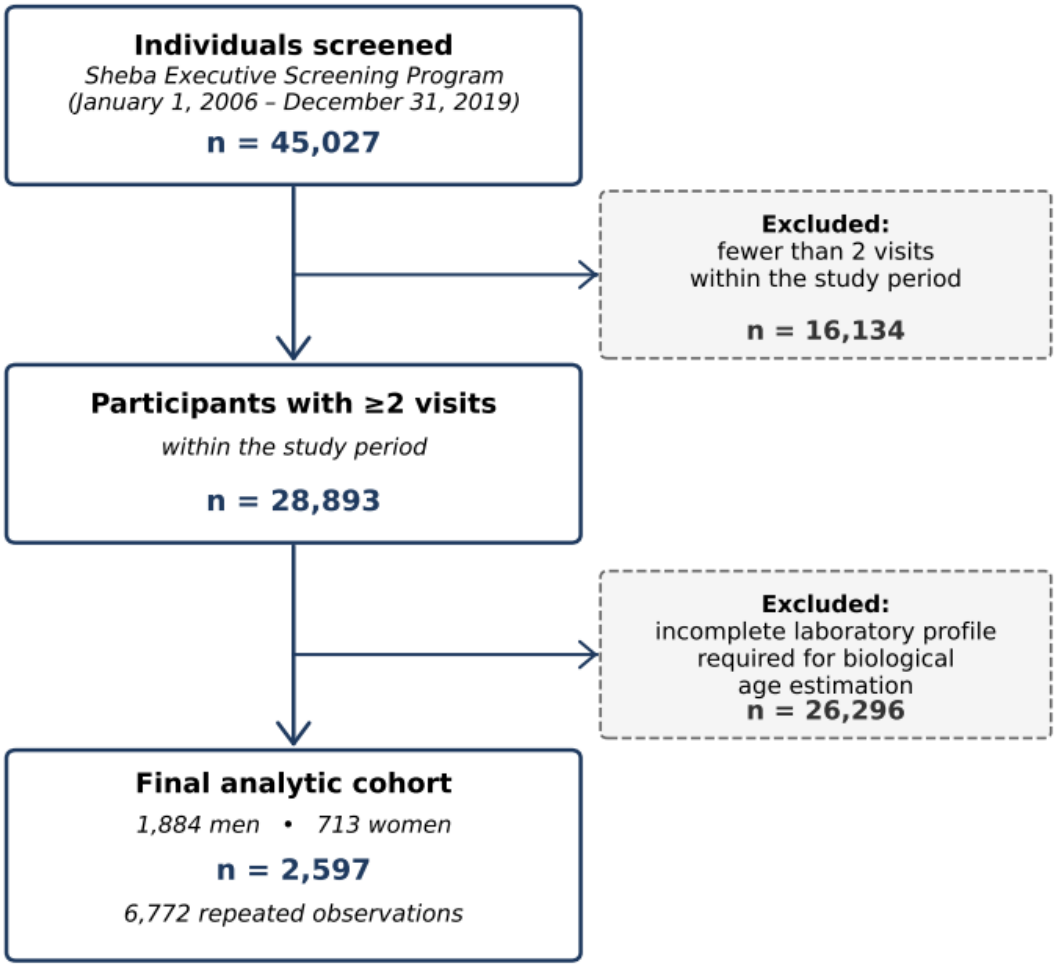

Supplementary Figure 1. Flow diagram of participant selection. Eligible participants were aged ≥18 years at their first visit and had at least two assessments between January 1, 2006, and December 31, 2019. Of 45,027 individuals screened at the Sheba Executive Screening Program during this period, 28,893 had ≥2 visits within the study time frame. After restricting the cohort to participants with complete laboratory profiles required for biological age estimation, the final analytic cohort comprised 2,597 individuals (1,884 men and 713 women), contributing 6,772 repeated observations

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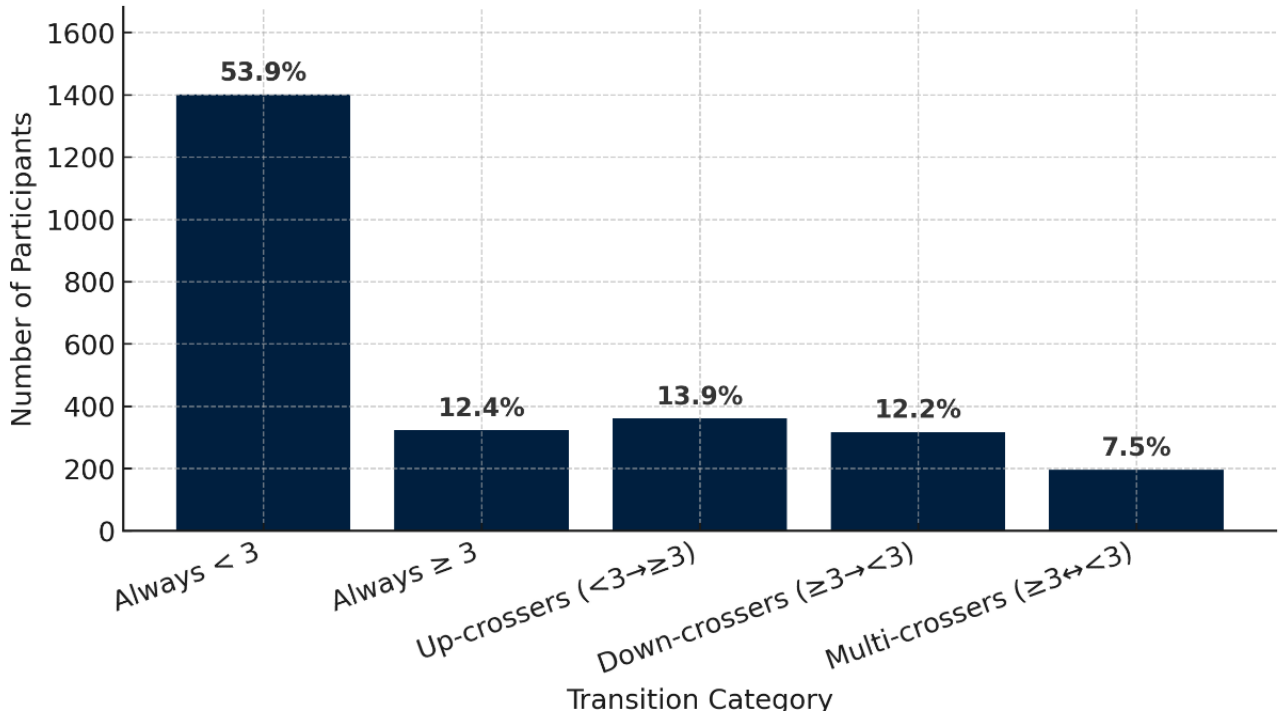

Supplementary Figure 2. Bars represent the proportion of participants classified according to longitudinal changes in Age $_{diff}$ Age category. "Always <3" and "Always ≥3" indicate stable trajectories, whereas "Up-crossers" and "Down-crossers" represent individuals who shifted between groups across follow-up visits. A small subset ("Multi-crossers") exhibited fluctuating transitions.