BMJ Open

Associations of educational level with ECG-derived cardiovascular ageing in a population-based cohort: a mediation analysis from the Tromsø Study

Arya Panthalanickal Vijayakumar ,[1] Tom Wilsgaard ,[1] Henrik Schirmer ,[2,3] Haakon Lindekleiv,[1,4] Zachi I Attia,[5] Francisco Lopez-Jimenez,[5] David Leon ,[6] Olena Iakunchykova[7]

To cite: Panthalanickal Vijayakumar A, Wilsgaard T, Schirmer H, *et al*. Associations of educational level with ECG-derived cardiovascular ageing in a population-based cohort: a mediation analysis from the Tromsø Study. *BMJ Open* 2025;**15**:e088671. doi:10.1136/bmjopen-2024-088671

► Prepublication history and additional supplemental material for this paper are available online. To view these files, please visit the journal online (https://doi.org/10.1136/bmjopen-2024-088671).

Received 12 May 2024
Accepted 02 June 2025

For numbered affiliations see end of article.

Correspondence to
Dr Arya Panthalanickal Vijayakumar;
arya.p.vijayakumar@uit.no

ABSTRACT

Objective To assess the association between educational level and cardiovascular age acceleration metric derived from ECG, and to determine whether this association is mediated by established cardiovascular disease (CVD) risk factors.

Design Prospective population-based cohort study (the Tromsø Study).

Setting General population of the Tromsø municipality, Norway.

Participants The study sample consisted of 4367 participants of the Tromsø Study, who took part in both Tromsø6 (2007–2008) and Tromsø7 (2015–2016), had a 12-lead ECG obtained at Tromsø7 and did not report a history of heart attack, stroke or atrial fibrillation.

Primary outcome measures δ-age, a biomarker of cardiovascular ageing, is defined as the difference (in years) between an individual's ECG-predicted heart age and their chronological age. ECG-predicted heart age was estimated using a previously validated deep neural network.

Results Our findings indicate an inverse association between education and δ-age, with a regression coefficient per increment increase in education of −0.24 (95% CI −0.41 to −0.07) in the overall sample, −0.38 (95% CI −0.59 to −0.16) for women and −0.04 (95% CI −0.31 to 0.23) for men. Participants with the highest level of education (university/college for 4 or more years) had the lowest estimated δ-age with a regression coefficient of −0.69 years (95% CI −1.23 to −0.16) compared with the group with primary education for the overall sample, −1.05 years (95% CI −1.73 to −0.37) for women and −0.15 years (95% CI −1.03 to 0.73) for men. CVD risk factors mediated up to 75% of the association between overall education and δ-age, and 80% of the association among those with the highest education level (university/college for 4 or more years). Among women, 50% of the effect of overall education on δ-age was mediated by CVD risk factors, rising to 53% in the category with the highest level of education. However, in the subsample of men, there was no significant association between education and δ-age, and the mediation analysis produced natural direct and indirect effects pointing in opposite directions.

Conclusions Cardiovascular ageing is inversely associated with educational level, an effect that appears to be largely mediated through established risk factors.

STRENGTHS AND LIMITATIONS OF THIS STUDY

⇒ ECG-based biomarker of cardiovascular ageing was estimated for the Tromsø Study cohort by applying a novel deep learning algorithm.
⇒ The Tromsø Study boasts a population-based design with high participation proportions and minimal missing questionnaire data.
⇒ Self-reported education levels that indicate socioeconomic position were validated against national registry data.
⇒ The prospective design establishes temporality between education, risk factors and δ-age and supports a causal mediation analysis.
⇒ Assumptions regarding unmeasured confounders for the execution of the causal mediation analysis cannot be definitively proven.

INTRODUCTION

Ageing is a primary risk factor for diseases such as dementia, cardiovascular disease (CVD) and cancer.[1] Ageing can be described as a wear and tear of physiological functioning,[2 3] ultimately leading to age-related illnesses or death. Since ageing does not progress uniformly for everyone, biological ageing biomarkers reflecting the physiological deterioration of internal organs and body systems can be used to assess ageing.

Various biomarkers of biological ageing have been developed, including telomere length, the epigenetic clock, mitochondrial DNA copy number, inflammatory markers and biomarkers specific to certain organs and systems.[4–8] It has been postulated that each represents a different aspect of ageing or ageing specific to different body systems. A novel ECG-based biomarker, δ-age, has been

developed using a deep neural network applied to ECG and defined as a discrepancy between the ECG-based age and the chronological age.[7] As it is obtained directly from raw, digitalised ECG waveforms, it is easy and affordable to collect. It has also been shown that δ-age is an independent predictor of all-cause and cardiovascular mortality.[7 9] The essential feature of δ-age in comparison to other ageing biomarkers is that it appears to be measuring actual changes of the heart reflecting structural damage or dysfunction, which in turn may represent overall ageing specific to the cardiovascular system.

The process of biological ageing is intricate and is impacted by both hereditary and environmental factors.[10] Previous studies have found that telomere and epigenetic ageing occurs at a faster pace in people with lower education and socioeconomic position (SEP), resembling the pattern of association between SEP and morbidity.[11 12] This association is explained by higher exposure to risk factors among people with lower levels of education due to lifestyle and poor access to healthcare.[13 14]

Although the relationship between SEP and CVD has been studied extensively, there have not been any studies performed on SEP and biomarkers of cardiovascular ageing[15] mainly due to the lack of an adequate and specific ageing biomarker for the cardiovascular system. The advantage of exploring such an association is that it would give insights into whether SEP is related to damage to the cardiovascular system which precedes symptomatic or routinely detectable disease.

Our study aims to estimate the association of SEP indexed by education level, a widely acknowledged key SEP indicator due to its influence on health behaviours and resource access,[16] and accelerated ageing specific to the cardiovascular system indexed by ECG-based δ-age. We undertake a causal mediation analysis in order to understand if this association can be explained by exposure to CVD risk factors, namely high blood pressure (BP) and low-density lipoprotein (LDL) cholesterol, obesity, diabetes, physical inactivity, BP medication usage and smoking.

METHODS

Study design and sample

The Tromsø Study is a population-based study conducted in 1974 in the municipality of Tromsø, Norway.[17] It consists of seven waves, known as Tromsø1 through Tromsø7, and a total of 45 473 people attended one or more surveys. Questionnaires, biological samples and health examinations were used to collect data. The present study includes data collected in Tromsø6 (2007–2008) and Tromsø7 (2015–2016).[18] Selected samples and whole birth cohorts of individuals in the age range 30–87 were invited to Tromsø6, and 12 984 participated (66%). All residents of Tromsø municipality aged 40 years or older were invited to Tromsø7, and 21 083 (65%) participated. The Tromsø7 study consisted of two visits: the first visit was attended by all participants and included questionnaires and a collection of basic measurements such as height, weight, BP, blood lipids, etc. The second visit was attended by a subsample of 8346 participants, and 12-lead ECGs were collected for 7779 participants who consented to ECG. Our study included participants who took part in both Tromsø6 and Tromsø7 and had ECG measurements in Tromsø7 (N=5017), after excluding those with self-reported history of heart attack, stroke or atrial

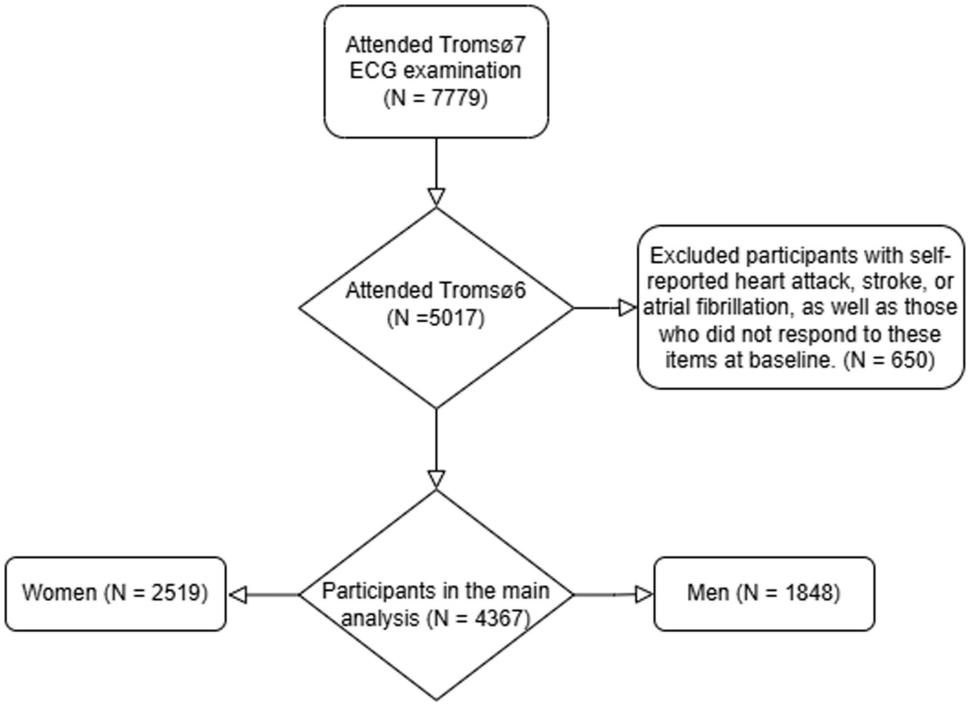

Figure 1 Flow chart displaying participants' inclusion and exclusion in the final study sample.

fibrillation, leading to a final sample of 4367 participants, as shown in the flow chart in figure 1.

Measurements

In the second visit of Tromsø7, 12-lead resting ECGs were recorded with a computer-based electrocardiograph (Cardiovit AT-104 PC; Schiller AG, Baar, Switzerland). To estimate ECG-based age, these digital ECGs were directly input into a pretrained convolutional neural network (CNN) algorithm developed and trained by our team using Mayo Clinic data.[7 9] The outcome variable, δ-age, was calculated as the difference between the CNN-derived ECG age and the chronological age.

The exposure variable was a self-reported education collected from the questionnaire in Tromsø6 with four levels: (1) primary/secondary school, modern secondary school, (2) upper secondary education or high school diploma, (3) college/university less than 4 years and (4) college/university 4 years or more. Diabetes status (yes/no), physical activity (PA), daily smoking status (yes/no) and BP medication usage (yes/no) were collected with questionnaires in Tromsø6 and included as mediator variables in the causal mediation analysis. PA level was assessed using the Saltin and Grimby leisure-time PA questionnaire[19] and categorised as sedentary, light activity, and moderate-to-vigorous activity. A history of heart attack, stroke and atrial fibrillation was self-reported. The mediators such as height, weight, diastolic BP (DBP), systolic BP (SBP), LDL cholesterol and high-density lipoprotein (HDL) cholesterol were collected as part of health examination during Tromsø6. SBP and DBP were measured on the right arm, three times with 1 min intervals after 2 min seated rest by a Dinamap ProCare 300 monitor (GE Healthcare, Norway), and the mean of the two last readings was used in the analyses. Non-fasting blood samples were obtained by venepuncture and analysed at the Department of Laboratory Medicine at the University Hospital of North Norway (ISO certification NS-EN ISO 15189:2012) within 48 hours. HDL-cholesterol was measured after separating apoB-containing lipoproteins

Table 1 Descriptive characteristics by the level of education*

Education (exposure)†				
	Primary education	Upper secondary / high school	University/college <4 years	University/college ≥4 years
n=4367	1174 (26.9%)	1490 (34.1%)	799 (18.3%)	863 (19.8%)
Outcome				
δ-age	0.05 (5.7)	0.1 (6.0)	−0.24 (6.3)	−0.41 (6.0)
Covariates†				
Age (years)	70.45 (7.50)	67.18 (8.49)	65.57 (8.87)	64.57 (8.66)
Sex (women)	772 (66%)	832 (56%)	377 (47%)	515 (60%)
Mediators				
BMI (kg/m^2)	27.29 (4.23)	27.02 (4.01)	26.85 (3.96)	25.87 (3.89)
Diastolic BP (mm Hg)	78.13 (10.35)	78.55 (10.45)	78.16 (10.25)	77.49 (10.79)
Systolic BP (mm Hg)	141.04 (21.51)	136.79 (21.23)	133.48 (20.4)	131.23 (21.01)
LDL cholesterol (mmol/L)	3.86 (0.96)	3.72 (0.90)	3.64 (0.89)	3.53 (0.92)
HDL cholesterol (mmol/L)	1.58 (0.42)	1.54 (0.43)	1.52 (0.43)	1.63 (0.45)
Diabetes	38 (3%)	55 (4%)	43 (5%)	17 (2%)
Physical activity‡				
Sedentary	209 (20%)	217 (16%)	115 (15%)	108 (13%)
Light	710 (68%)	912 (65%)	473 (61%)	527 (63%)
Moderate	119 (11%)	252 (18%)	172 (22%)	185 (22%)
Heavy	4 (0%)	15 (1%)	17 (2%)	20 (2%)
Daily smoker	259 (22%)	270 (18%)	106 (13%)	74 (9%)
BP medication (yes)	337 (29%)	314 (21%)	137 (17%)	104 (12%)

The Tromsø Study 2007–2016.
*Age is measured at Tromsø7(2015–2016), other variables were measured at Tromsø6 (2007–2008).
†Values are mean (SD) or number (per cent); row percentage is presented for educational categories, column percentages are presented for other categorical variables.
‡Except for physical activity (6% missing values) all the other variables have less than 1% missing values.
BMI, body mass index; BP, blood pressure; HDL, high-density lipoprotein; LDL, low-density lipoprotein; δ-age, predicted ECG age–chronological age.

by using heparin and manganese chloride, total cholesterol and triglycerides were measured by CHOD-PAP enzymatic colorimetric methods with commercial kits by Roche Diagnostics (Mannheim, Germany), LDL-cholesterol (mmol/L) was derived with Friedewald Equation.[20] Body mass index (BMI) (kg/m^2) was calculated by dividing weight in kilograms by height in metres squared. The mediators were selected based on findings from numerous research that examined the factors underlying the relation between SEP and CVD.[21]

Statistical analysis

Pearson correlation coefficient and the mean absolute error (MAE) between the ECG-based age and chronological age were calculated. Baseline characteristics were presented as means and SD for continuous variables, after visual inspection of histograms confirmed their approximately normal distribution. Numbers and percentages were presented for categorical variables. A directed acyclic graph for the causal association between education and CVD was created based on the review of the literature (online supplemental figure S1). Regression analyses were used to estimate the association between education and the mediators, and between mediators and δ-age. Linear regression models were used for continuous outcome variables, logistic regression for binary outcomes, and ordinal logistic regression for PA as an outcome. All regression models included age as a covariate, and the overall model also included sex. In a separate model, we used a likelihood ratio test to assess a possible interaction between sex and education. The main analyses consisted of two parts: (1) a linear regression to determine the association between education and δ-age 2) a causal mediation analysis to determine how much of the former association can be explained by the CVD risk factors, both employed in a model adjusted for age, square of age and sex. We chose to present sex-specific models a priori based on existing evidence that there are sex differences in the manifestation and pathophysiology of CVD among men and women.[22] Participants with a history of heart attack, stroke and atrial fibrillation at baseline were excluded from the sample, as previous disease was likely to lead to lifestyle changes and medication use. A sensitivity analysis was performed to verify if results are robust after exclusion of participants who reported a history of heart attack, stroke or atrial fibrillation during follow-up (self-reported disease reported at Tromsø7).

The total effects (TEs) were estimated by employing linear regression between education and δ-age after adjusting for confounders. Natural effect models estimated the natural direct and indirect effects with a 95% CI based on the robust SE using the medflex package in R.[23] In total, 396 cases had one or more missing values for BMI, SBP, DBP, LDL-cholesterol, HDL-cholesterol, diabetes status, PA, daily smoking status and BP medication. For all the regression and causal mediation analyses involving the risk factors or mediators and exposure, we used multiple imputation by chained equations with 20 imputed datasets, incorporating auxiliary variables under the missing at random assumption; the resulting estimates were then pooled according to Rubin's rule. Complete case data on 3971 participants were used in the

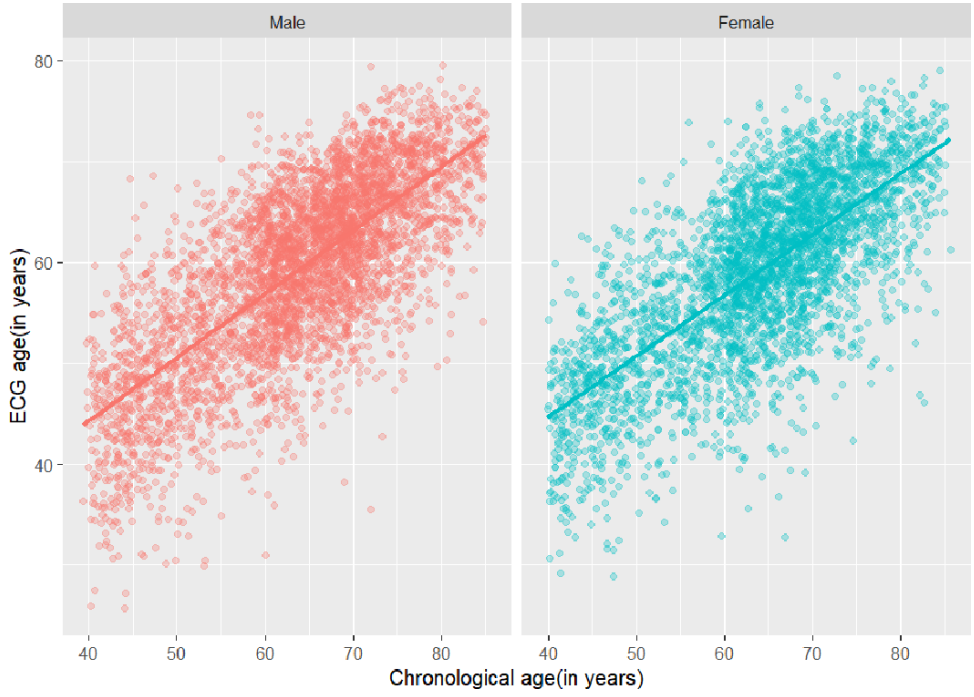

Figure 2 Scatterplot for ECG-based age and chronological age for the Tromsø Study (N=7779), with men represented in red and women in blue.

Table 2 Linear regression coefficients and ORs for the association between CVD risk factors and education*

Outcome variables	Primary education	Upper secondary /high school	University/college <4 years	University/college ≥4 years	P value for trend
Overall (N=4367)					
Linear regression coefficients (95% CI)					P value
BMI (1 SD kg/m^2)	0 (Reference)	−0.08 (−0.15 to 0.00)	−0.13 (−0.22 to −0.04)	−0.35 (−0.44 to −0.26)	<0.001
Diastolic BP (1 SD mm Hg)	0 (Reference)	0.00 (−0.07 to 0.08)	−0.08 (−0.18 to 0.01)	−0.06 (−0.15 to 0.03)	0.157
Systolic BP (1 SD mm Hg)	0 (Reference)	−0.07 (−0.14 to 0.00)	−0.17 (−0.25 to −0.08)	−0.20 (−0.28 to −0.12)	<0.001
LDL cholesterol (1 SD mmol/L)	0 (Reference)	−0.11 (−0.19 to −0.04)	−0.19 (−0.28 to −0.10)	−0.28 (−0.37 to −0.19)	<0.001
HDL cholesterol (1 SD mmol/L)	0 (Reference)	0.03 (−0.05 to 0.10)	0.08 (−0.01 to 0.17)	0.26 (0.17 to 0.35)	<0.001
ORs (95% CI)†					P value
Physical activity, per level	1 (Reference)	1.50 (1.28 to 1.77)	1.80 (1.48 to 2.18)	2.03 (1.68 to 2.45)	<0.001
Diabetes, yes/no	1 (Reference)	1.21 (0.79 to 1.85)	1.82 (1.15 to 2.88)	0.71 (0.39 to 1.27)	0.005
Daily smoking, yes/no	1 (Reference)	0.71 (0.59 to 0.87)	0.47 (0.36 to 0.61)	0.27 (0.21 to 0.36)	<0.001
BP medication, yes/no	1 (Reference)	0.81 (0.67 to 0.97)	0.69 (0.54 to 0.87)	0.48 (0.38 to 0.62)	<0.001
Women (N=2519)					
Linear regression coefficients (95% CI)					P value
BMI (1 SD kg/m^2)	0 (Reference)	−0.13 (−0.23 to −0.03)	−0.20 (−0.33 to −0.07)	−0.36 (−0.48 to −0.25)	<0.001
Diastolic BP (1 SD mm Hg)	0 (Reference)	0.02 (−0.08 to 0.12)	−0.10 (−0.23 to 0.03)	−0.07 (−0.19 to 0.04)	0.158
Systolic BP (1 SD mm Hg)	0 (Reference)	−0.03 (−0.12 to 0.06)	−0.13 (−0.25 to −0.02)	−0.19 (−0.30 to −0.09)	0.001
LDL cholesterol (1 SD mmol/L)	0 (Reference)	−0.07 (−0.17 to 0.03)	−0.20 (−0.32 to −0.07)	−0.27 (−0.39 to −0.16)	<0.001
HDL cholesterol (1 SD mmol/L)	0 (Reference)	0.14 (0.04 to 0.24)	0.21 (0.08 to 0.34)	0.41 (0.29 to 0.53)	<0.001
ORs (95% CI)†					P value
Physical activity, 4 levels	1 (Reference)	1.67 (1.32 to 2.10)	1.83 (1.37 to 2.45)	2.21 (1.69 to 2.88)	<0.001
Diabetes, yes/no	1 (Reference)	1.13 (0.61 to 2.11)	2.61 (1.35 to 5.04)	0.90 (0.40 to 2.03)	0.019
Daily smoking, yes/no	1 (Reference)	0.76 (0.60 to 0.98)	0.40 (0.28 to 0.57)	0.25 (0.17 to 0.36)	<0.001
BP Medication, yes/no	1 (Reference)	0.79 (0.62 to 1.00)	0.55 (0.39 to 0.78)	0.41 (0.29 to 0.57)	<0.001
Men (N=1848)					
Linear regression coefficients (95% CI)					P value
BMI (1 SD kg/m^2)	0 (Reference)	0.03 (−0.09 to 0.16)	−0.01 (−0.15 to 0.12)	−0.28 (−0.43 to −0.14)	<0.001
Diastolic BP (1 SD mm Hg)	0 (Reference)	0.00 (−0.13 to 0.12)	−0.05 (−0.19 to 0.09)	−0.01 (−0.16 to 0.13)	0.882
Systolic BP (1 SD mm Hg)	0 (Reference)	−0.08 (−0.20 to 0.04)	−0.17 (−0.30 to −0.03)	−0.16 (−0.30 to −0.02)	0.048
LDL cholesterol (1 SD mmol/L)	0 (Reference)	−0.12 (−0.24 to 0.00)	−0.12 (−0.26 to 0.02)	−0.21 (−0.36 to −0.06)	0.040
HDL cholesterol (1 SD mmol/L)	0 (Reference)	−0.18 (−0.31 to −0.06)	−0.14 (−0.27 to 0.00)	−0.01 (−0.15 to 0.13)	0.006
ORs (95% CI)					P value
Physical activity, 4 levels	1 (Reference)	1.36 (1.07 to 1.73)	1.72 (1.32 to 2.25)	1.90 (1.43 to 2.52)	<0.001
Diabetes, yes/no	1 (Reference)	1.28 (0.70 to 2.33)	1.45 (0.76 to 2.75)	0.59 (0.25 to 1.39)	0.103
Daily smoking, yes/no	1 (Reference)	0.62 (0.45 to 0.85)	0.53 (0.37 to 0.76)	0.30 (0.19 to 0.47)	<0.001
BP medication, yes/no	1 (Reference)	0.88 (0.65 to 1.19)	0.88 (0.63 to 1.23)	0.63 (0.43 to 0.93)	0.119

The Tromsø Study 2007–2016.
*Adjusted for age for and also for sex in the overall sample.
†ORs from binary or ordinal logistic regression models.
BMI, body mass index; BP, blood pressure; CVD, cardiovascular disease; HDL, high-density lipoprotein; LDL, low-density lipoprotein; OR, Odds ratio.

sensitivity analysis. All analyses were conducted using RStudio (Posit Team, 2025), V.2024.12.1+563 'Kousa Dogwood'.

Patient and public involvement

Patients and/or the public were not involved in the design, or conduct, or reporting, or dissemination plans of this research.

RESULTS

Out of 7779 people who had the ECG records available in Tromsø7, 5017 participants had attended and had measurements of required risk factor variables in Tromsø6. Further analysis was based on 4367 participants who have reported no history of heart attack, stroke and atrial fibrillation. Among these 4367 participants,

1174 (27.1%) had primary/secondary school level, 1490 (34.4%) had an upper secondary or high school, 799 (18.5%) had university college education with less than 4 years duration, 863 (20%) had a university college education with a duration greater than or equal to 4 years and the remaining had missing data(table 1). All continuous variables were approximately normally distributed and were described with means and SD in table 1. We observed that means and prevalence of the CVD risk factors such as BMI, DBP, SBP, LDL-cholesterol, diabetes, BP medication usage and smoking were lowest for participants with the highest education level.

The MAE and Pearson correlation coefficient between the chronological age and the ECG-based age are 6.9 years and 0.72, respectively, compared with 6.9 years and 0.83 for the original CNN model. Figure 2 further demonstrates the sex-specific correlation between chronological age and ECG-based age for the 7779 participants who underwent ECG examination in the Tromsø Study. Tables 2 and 3 present the age and sex adjusted associations between education (measured at Tromsø6) and the mediators, and between δ-age (calculated from Tromsø7) and the mediators, respectively, for the final study sample (N=4367). The differences in CVD risk factors between education groups were statistically significant: BMI, SBP, LDL-cholesterol and prevalence of diabetes, usage of BP medication and smoking were higher in the lower educational level, whereas HDL-cholesterol and PA were higher among higher educational levels (table 2). Table 3 shows that BMI, DBP, PA, diabetes, BP medication usage and smoking status were significantly associated with δ-age.

Linear regression was used to estimate the association between education and δ-age, yielding a TE of −0.24 (95% CI −0.41 to −0.07) (table 4). There was a statistically significant (p<0.01) trend of decreasing δ-age across the levels of education. Participants with university/college <4 years had an estimated −0.38 years difference as compared with those with primary education (95% CI −0.92 to 0.17), while those with university/college education ≥4 years had the strongest estimate with −0.69 years (95% CI −1.23 to −0.16). In the sex-specific models, the same pattern of associations was observed among women with a TE of −0.38 years (95% CI −0.59 to −0.16) per increment in education level(with the strongest estimate among participants with university/college education ≥4 years, −1.05 years (95% CI −1.73 to −0.37)). However, the association did not reach statistical significance for any of the education levels among men. These results were confirmed in the analysis with complete case data (TEs in online supplemental table S1).

The natural indirect effect (NIE), for the overall sample, was −0.18 years (95% CI −0.24 to −0.13) per increment in education level. Compared with the primary-education group, the NIE was −0.55 years (95% CI −0.72 to −0.39) for the highest education group, and −0.37 years (95% CI:−0.50 to −0.24) for the university/college <4 years education group. Similar patterns were observed in women and men with the estimate per increment level of education of −0.19 years (95% CI:−0.26 to −0.12) and −0.17 years (95% CI −0.25 to −0.09), respectively. The NIE through CVD risk factors included as mediators (diabetes, SBP, DBP, BMI, smoking, BP medication, LDL-cholesterol, HDL-cholesterol and PA) mediated 75% (NIE/TE) of the TE between education (per level) and δ-age and 80% of the TE between the highest level of education relative to primary education and δ-age. In the sex-specific analysis (table 4), both the direct and indirect effects were somewhat stronger in women than in men. For women, 50% of the TE between education (per level) and δ-age is mediated by the risk factors. Even though the sex-specific analysis resulted in notably different

Table 3 Linear regression coefficients for the association between δ-age and CVD risk factors*

	Overall (N=4367)		Women (N=2519)		Men (N=1848)	
Independent variables†	Coefficient (95% CI)	P value	Coefficient (95% CI)	P value	Coefficient (95% CI)	P value
BMI (1 SD kg/m^2)	0.18 (−0.02 to 0.37)	0.080	−0.04 (−0.30 to 0.21)	0.752	0.47 (0.17 to 0.78)	0.002
Diastolic BP (1 SD mm Hg)	0.57 (0.34 to 0.81)	<0.001	0.41 (0.11 to 0.72)	0.008	0.80 (0.42 to 1.18)	<0.001
Systolic BP (1 SD mm Hg)	0.18 (−0.08 to 0.44)	0.178	0.19 (−0.15 to 0.53)	0.280	0.14 (−0.26 to 0.54)	0.500
LDL cholesterol (1 SD mmol/L)	0.11 (−0.07 to 0.29)	0.241	0.18 (−0.06 to 0.42)	0.133	0.01 (−0.27 to 0.29)	0.922
HDL cholesterol (1 SD mmol/L)	−0.11 (−0.31 to 0.08)	0.247	−0.21 (−0.46 to 0.04)	0.101	0.00 (−0.30 to 0.30)	0.986
Physical activity (per level)	−0.51 (−0.80 to −0.21)	0.001	−0.75 (−1.19 to −0.31)	0.001	−0.28 (−0.68 to 0.13)	0.181
Diabetes (yes/no)	0.70 (−0.28 to 1.67)	0.161	0.00 (−1.42 to 1.43)	0.997	1.31 (−0.04 to 2.66)	0.056
Daily smoking (yes/no)	1.17 (0.69 to 1.65)	<0.001	0.89 (0.28 to 1.50)	0.004	1.54 (0.77 to 2.32)	<0.001
BP medication (yes/no)	0.72 (0.26 to 1.19)	0.002	0.90 (0.30 to 1.51)	0.004	0.47 (−0.26 to 1.20)	0.204

The Tromsø Study 2007–2016.
*Regression coefficients are presented per 1 SD unit for the continuous independent variables. All coefficients are adjusted for age and also for sex in the overall sample.
†Independent variables in this table were measured in Tromsø6.
BMI, body mass index; BP, blood pressure; CVD, cardiovascular disease; HDL, high-density lipoprotein; LDL, low-density lipoprotein; δ-age, predicted ECG age–chronological age.

Table 4 Mediation of association between education and δ-age by CVD risk factors*

	NDE (coefficient, 95% CI)†	NIE (coefficient, 95% CI)†	TE (coefficient, 95% CI)†	Proportion mediated (%)
Overall (N=4367)				
Primary education (reference)	0 (Reference)	0 (Reference)	0 (Reference)	NA‡
Upper secondary/high school	0.14 (−0.30 to 0.58)	−0.19 (−0.29 to −0.09)	−0.05 (−0.51 to 0.41)	NA
University/college <4 years	−0.01 (−0.56 to 0.54)	−0.37 (−0.50 to −0.24)	−0.38 (−0.92 to 0.17)	97
University/college ≥4 years	−0.14 (−0.67 to 0.39)	−0.55 (−0.72 to −0.39)	−0.69 (−1.23 to −0.16)	80
Total education (per level)	−0.06 (−0.23 to 0.11)	−0.18 (−0.24 to −0.13)	−0.24 (−0.41 to −0.07)	75
Women (N=2519)				
Primary education (reference)	0 (Reference)	0 (Reference)	0 (Reference)	NA
Upper secondary/high school	0.25 (−0.31 to 0.81)	−0.20 (−0.32 to −0.09)	0.04 (−0.54 to 0.63)	NA
University/college <4 years	−0.20 (−0.97 to 0.56)	−0.42 (−0.60 to −0.24)	−0.62 (−1.37 to 0.12)	68
University/college ≥4 years	−0.49 (−1.17 to 0.19)	−0.56 (−0.79 to −0.34)	−1.05 (−1.73 to −0.37)	53
Total education (per level)	−0.19 (−0.41 to 0.03)	−0.19 (−0.26 to −0.12)	−0.38 (−0.59 to −0.16)	50
Men (N=1848)				
Primary education (reference)	0 (Reference)	0 (Reference)	0 (Reference)	NA
Upper secondary/high school	0.02 (−0.71 to 0.75)	−0.15 (−0.34 to 0.05)	−0.13 (−0.88 to 0.63)	NA
University/college <4 years	0.17 (−0.65 to 1.00)	−0.28 (−0.49 to −0.07)	−0.11 (−0.94 to 0.72)	NA
University/college ≥4 years	0.38 (−0.48 to 1.24)	−0.53 (−0.79 to −0.26)	−0.15 (−1.03 to 0.73)	NA
Total education (per level)	0.13 (−0.14 to 0.40)	−0.17 (−0.25 to −0.09)	−0.04 (−0.31 to 0.23)	NA

The coefficients of NDE, NIE and TE and their corresponding 95% CI. The Tromsø Study 2007–2016.
*All models are adjusted for age and squared-age, and the overall sample adjusted for sex as well.
†TE is the overall causal effect of an exposure on an outcome, including both the NDE and the NIE. The coefficients of NDE quantify the direct impact of the exposure on the outcome, NIE implies the portion of the TE of the exposure on the outcome that is mediated by the mediators.
‡NA: the proportion mediated cannot be calculated because the NDE and NIE have opposite directions, a known limitation of this metric.
CVD, cardiovascular disease; NA, not applicable; NDE, natural direct effect; NIE, natural indirect effect; TE, total effect; δ-age, predicted ECG age–chronological age.

effects for men and women, the test of interaction did not show a significant moderation effect by sex (p=0.15). These results were robust to the exclusion of participants who reported heart attack, stroke or atrial fibrillation at Tromsø7 in addition to that in Tromsø6 (online supplemental table S2).

DISCUSSION

In this study, we demonstrate that SEP indexed by education is associated with a machine learning derived marker of cardiovascular ageing based on ECG (δ-age). A higher education level is associated with lower values of this ageing biomarker specific to the cardiovascular system with a statistically significant trend across the educational levels. Furthermore, we conducted a causal mediation analysis which demonstrated that up to 75% of the TE of education level on δ-age is mediated by well-known CVD risk factors, like diabetes, LDL-cholesterol, HDL-cholesterol, PA, BMI, BP medication and smoking measured 7 years before ageing biomarker. Consequently, these results imply that interventions directed to decrease the prevalence of CVD risk factors would narrow the gap in ageing of the cardiovascular system among people with different levels of education. The sex-specific analyses showed a stronger association between education and δ-age among women; however, these results need confirmation as the statistical test for the moderation effect of sex was not significant and should be interpreted with caution. It is also important to note that tests for interaction are often underpowered, that is, real sex differences may exist but might not be detectable. The plausible reasoning behind the sex-specific result can be found from Bloomberg and Steptoe,[24] where women and men show distinct physiological ageing trajectories influenced by education. Women with lower education had accelerated ageing compared with men, while higher education preserved a younger physiological profile. These findings imply that educational attainment can moderate sex disparities in cardiovascular health. To the best of our knowledge, this is the first study examining the differences in ageing of the cardiovascular system indexed by ECG-based biomarker among groups with different levels of education.

Although there is no gold standard measure for biological ageing, many putative biomarkers have been assessed.[4–8 25] SEP is found to have a profound influence on individuals as well as overall population health

indicated by various biological indicators.[26] Numerous studies have repeatedly shown that higher education has a protective effect on biological ageing, which is consistent with our findings for δ-age.[11 12] For instance, studies of telomeres showed that lower educational attainment was significantly associated with shorter telomere length, while neither household income nor employment status was related to telomere length.[27 28] A DNA methylation clock has also been consistently associated with different measures of SEP ranging from educational attainment to social disadvantage indices.[12 29–31] In this study, we described a similar inverse relationship between a CNN-derived biomarker of vascular ageing based on ECG and SEP indexed by education and further explored the mechanism of this relationship by considering major CVD risk factors as mediators.

Studies in different countries and populations showed that higher education is associated with a lower lifetime risk of CVD and has the inverse relation with CVD risk factor prevalence.[15 32 33] Understanding the mechanisms through which education affects the risk of developing CVD and the potential mediating function of modifiable and lifestyle-related CVD risk factors has recently gained a lot of attention. However, the relationship is complex and multifaceted with various risk factors mediating[21 34] and confounding this association.[35] In the Tromsø population which our study was based on, lifestyle variables and biological CVD risk factors were found to mediate the effect of the area level SEP measured using income, education and house ownership.[36]

In comparison to blood-based or lifestyle risk factors which are used to predict CVD events in the distant future, the δ-age that we use may be a better risk indicator than traditional CVD risk factors measured at one point of time. δ-age is derived from an individual's ECG recordings processed by a CNN that may reflect subtle variations in cardiac conduction properties and early alterations in cardiovascular physiology. By calculating δ-age as a continuous variable, researchers can assess CVD risk on a spectrum of values reflecting the cumulative exposure to the range of risk factors through the life-course including measured and unmeasured (unknown) risk factors, before the ultimate occurrence of CVD events. It has been shown before that δ-age calculated with the similar CNN as we used in this study provides additional information to predict CVD mortality over traditional risk factors.[37]

We have shown that δ-age is positively associated with BMI, BP, smoking and diabetes and inversely with PA and HDL-cholesterol, which resembles the risk factor profile of CVD.[38] While the mediation proportion was high (75%), this may be an underestimate due to measurement error in reported behavioural and lifestyle risk factors, including the difficulty in measuring lifelong exposures, as the study included mediators measured at one time point, as well as unmeasured mediators not included in the natural effects model. Diet, sleeping habits, income, early life experiences, psychological stress and air pollution are some instances of unmeasured or unaccounted variables.[15 39 40] These factors combined could have contributed to the natural direct effect of education on δ-age. It is thus remarkable that such a large proportion of association between education and δ-age was mediated by risk factors measured in our study, while studies that assessed mediated proportion for association between education/SEP and CVD reported a lower proportion of effect mediated by measured risk factors.[21 41] Understanding the mediating pathways is crucial for developing effective interventions and policies that target specific risk factors, ultimately slowing down vascular ageing.

Strengths and limitations

An intrinsic strength of δ-age as a biomarker of cardiovascular system ageing is that it reflects specific electrical conduction patterns in the heart, potentially serving as early indicators of clinically detectable disease. This study has several other strengths: it is population-based with a high participation proportion, low number of missing values in the questionnaires and rigorous data collection procedures for medical tests. Even though the educational level is self-reported, Vo *et al* validated the completeness and correctness of self-reported education in Tromsø7 against national registry data.[42] This study has a prospective study design which is ideally suited to establish the temporal sequence of the associations between education attained prior to Tromsø6, mediators measured during Tromsø6, and δ-age obtained in Tromsø7.

One limitation of our study is that the Mayo model was trained on hospital patients and can reflect accelerated ageing, potentially biasing its application to a community cohort characterised with healthy ageing. However, the similar correlation and MAE between ECG-derived and chronological age support its utility in our population. Our study also relies on some self-reported data on variables such as PA, smoking status and diabetes, which may introduce some non-differential measurement error that could bias the association towards the null. The validity of assumptions regarding unmeasured confounders that is required for the mediation analysis cannot be definitely demonstrated, so caution should be exercised when extrapolating the results to other settings. While the Tromsø Study provides valuable insights into the health of the Tromsø population, the findings may not be directly applicable to other populations with different demographic, socioeconomic characteristics and ethnical composition.

CONCLUSIONS

SEP, indexed by level of educational attainment, was associated with ageing of the cardiovascular system. Participants with a higher level of education had lower values for CNN-derived biomarker of heart ageing based on ECG (the trend to decreasing values of δ-age across educational categories). This relationship remains

unchanged even after the exclusion of participants with CVD events during the study period, supporting the conclusion that education is associated with actual damage to the cardiovascular system preceding symptomatic or routinely detectable disease. This association was mediated by established cardiovascular risk factors. This is consistent with the pattern of associations shown for established biomarkers of heart disease and CVD incidence. The findings of mediation analysis suggest that reducing inequalities due to differences in risk factor profiles between education groups may be an effective strategy to delay ageing of the cardiovascular system and prevent CVD. Our findings highlight the importance of addressing inequalities in SEP as a key public health intervention to enhance cardiovascular health and slow the ageing process.

Author affiliations
[1]Department of Community Medicine, UiT The Arctic University of Norway Faculty of Health Sciences, Tromsø, Norway
[2]Department of Clinical Medicine, University of Oslo Faculty of Medicine, Lørenskog, Norway
[3]Department of Cardiology, Akershus University Hospital, Lorenskog, Norway
[4]University Hospital of North Norway, Tromso, Norway
[5]Mayo Clinic College of Medicine and Science, Rochester, Minnesota, USA
[6]Department of Non-communicable Disease Epidemiology, London School of Hygiene & Tropical Medicine Faculty of Epidemiology and Population Health, London, UK
[7]Department of Psychology, University of Oslo Faculty of Social Sciences, Oslo, Norway

Contributors APV is responsible for the overall content as guarantor. OI, DL and APV conceived and designed the study. TW, HS and HL collected data for the study, ZIA processed the data, APV and TW performed statistical analysis, FL-J, DL, HS and OI interpreted the results. APV drafted the manuscript. OI, FJL, TW, HS, HL, ZIA and DL critically revised and approved the manuscript.

Funding This work was supported by Northern Norway Regional Health Authority, grant number HNF1636-22.

Competing interests None declared.

Patient and public involvement Patients and/or the public were not involved in the design, or conduct, or reporting, or dissemination plans of this research.

Patient consent for publication Not applicable.

Ethics approval The Tromsø Study complies with the Declaration of Helsinki and has been approved by the Regional Committee for Medical and Health Research Ethics (REK), the Data Inspectorate and the Norwegian Directorate of Health. All participants provided written informed consent. The current study was approved by REK Nord (reference 68185) and data management plan was evaluated by the Norwegian Centre for Research Data.

Provenance and peer review Not commissioned; externally peer reviewed.

Data availability statement Data may be obtained from a third party and are not publicly available. The data supporting the findings in this study are available through an application directed to The Tromsø Study by following the steps presented on their webpage. https://uit.no/research/tromsostudy.

ORCID iDs
Arya Panthalanickal Vijayakumar http://orcid.org/0009-0005-8475-3644
Tom Wilsgaard http://orcid.org/0000-0002-2709-9472
Henrik Schirmer http://orcid.org/0000-0002-9348-3149
David Leon http://orcid.org/0000-0001-9747-1762

REFERENCES

1 Niccoli T, Partridge L. Ageing as a risk factor for disease. *Curr Biol* 2012;22:R741–52.
2 Kirkwood TBL. Understanding the odd science of aging. *Cell* 2005;120:437–47.
3 Wang JC, Bennett M. Aging and atherosclerosis: mechanisms, functional consequences, and potential therapeutics for cellular senescence. *Circ Res* 2012;111:245–59.
4 Simm A, Nass N, Bartling B, *et al*. Potential biomarkers of ageing. *Biol Chem* 2008;389:257–65.
5 Horvath S, Raj K. DNA methylation-based biomarkers and the epigenetic clock theory of ageing. *Nat Rev Genet* 2018;19:371–84.
6 Rivero-Segura NA, Bello-Chavolla OY, Barrera-Vázquez OS, *et al*. Promising biomarkers of human aging: In search of a multi-omics panel to understand the aging process from a multidimensional perspective. *Ageing Res Rev* 2020;64:101164.
7 Attia ZI, Friedman PA, Noseworthy PA, *et al*. Age and Sex Estimation Using Artificial Intelligence From Standard 12-Lead ECGs. *Circ Arrhythm Electrophysiol* 2019;12:e007284.
8 Smith SM, Vidaurre D, Alfaro-Almagro F, *et al*. Estimation of brain age delta from brain imaging. *Neuroimage* 2019;200:528–39.
9 Ladejobi AO, Medina-Inojosa JR, Shelly Cohen M, *et al*. The 12-lead electrocardiogram as a biomarker of biological age. *Eur Heart J Digit Health* 2021;2:379–89.
10 Li S, Nguyen TL, Wong EM, *et al*. Genetic and environmental causes of variation in epigenetic aging across the lifespan. *Clin Epigenetics* 2020;12:158.
11 Robertson T, Batty GD, Der G, *et al*. Is socioeconomic status associated with biological aging as measured by telomere length? *Epidemiol Rev* 2013;35:98–111.
12 Steptoe A, Zaninotto P. Lower socioeconomic status and the acceleration of aging: An outcome-wide analysis. *Proc Natl Acad Sci U S A* 2020;117:14911–7.
13 Fiorito G, Polidoro S, Dugué P-A, *et al*. Social adversity and epigenetic aging: a multi-cohort study on socioeconomic differences in peripheral blood DNA methylation. *Sci Rep* 2017;7:16266.
14 McMaughan DJ, Oloruntoba O, Smith ML. Socioeconomic Status and Access to Healthcare: Interrelated Drivers for Healthy Aging. *Front Public Health* 2020;8:231.
15 de Mestral C, Stringhini S. Socioeconomic Status and Cardiovascular Disease: an Update. *Curr Cardiol Rep* 2017;19:115.
16 Winkleby MA, Jatulis DE, Frank E, *et al*. Socioeconomic status and health: how education, income, and occupation contribute to risk factors for cardiovascular disease. *Am J Public Health* 1992;82:816–20.
17 Jacobsen BK, Eggen AE, Mathiesen EB, *et al*. Cohort profile: the Tromso Study. *Int J Epidemiol* 2012;41:961–7.
18 Hopstock LA, Grimsgaard S, Johansen H, *et al*. The seventh survey of the Tromsø Study (Tromsø7) 2015-2016: study design, data collection, attendance, and prevalence of risk factors and disease in a multipurpose population-based health survey. *Scand J Public Health* 2022;50:919–29.
19 Grimby G, Börjesson M, Jonsdottir IH, *et al*. The “Saltin-Grimby Physical Activity Level Scale” and its application to health research. *Scand J Med Sci Sports* 2015;25 Suppl 4:119–25.
20 Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem* 1972;18:499–502.
21 Dégano IR, Marrugat J, Grau M, *et al*. The association between education and cardiovascular disease incidence is mediated by hypertension, diabetes, and body mass index. *Sci Rep* 2017;7:12370.

22 Woodward M. Rationale and tutorial for analysing and reporting sex differences in cardiovascular associations. *Heart* 2019;105:1701–8.
23 Steen J, Loeys T, Moerkerke B, *et al*. An R Package for Flexible Mediation Analysis using Natural Effect Models. *J Stat Softw* 2017;76:46.
24 Bloomberg M, Steptoe A. Sex and education differences in trajectories of physiological ageing: longitudinal analysis of a prospective English cohort study. *Age Ageing* 2025;54.
25 Gonzales MM, Garbarino VR, Pollet E, *et al*. Biological aging processes underlying cognitive decline and neurodegenerative disease. *J Clin Invest* 2022;132:e158453.
26 Phelan JC, Link BG, Tehranifar P. Social conditions as fundamental causes of health inequalities: theory, evidence, and policy implications. *J Health Soc Behav* 2010;51 Suppl:S28–40.
27 Adler N, Pantell MS, O'Donovan A, *et al*. Educational attainment and late life telomere length in the Health, Aging and Body Composition Study. *Brain Behav Immun* 2013;27:15–21.
28 Steptoe A, Hamer M, Butcher L, *et al*. Educational attainment but not measures of current socioeconomic circumstances are associated with leukocyte telomere length in healthy older men and women. *Brain Behav Immun* 2011;25:1292–8.
29 Fiorito G, McCrory C, Robinson O, *et al*. Socioeconomic position, lifestyle habits and biomarkers of epigenetic aging: a multi-cohort analysis. *Aging (Milano)* 2019;11:2045–70.
30 McCrory C, Fiorito G, Ni Cheallaigh C, *et al*. How does socio-economic position (SEP) get biologically embedded? A comparison of allostatic load and the epigenetic clock(s). *Psychoneuroendocrinology* 2019;104:64–73.
31 Raffington L, Belsky DW. Integrating DNA Methylation Measures of Biological Aging into Social Determinants of Health Research. *Curr Environ Health Rep* 2022;9:196–210.
32 Petrelli A, Sebastiani G, Di Napoli A, *et al*. Education inequalities in cardiovascular and coronary heart disease in Italy and the role of behavioral and biological risk factors. *Nutr Metab Cardiovasc Dis* 2022;32:918–28.
33 Ernstsen L, Strand BH, Nilsen SM, *et al*. Trends in absolute and relative educational inequalities in four modifiable ischaemic heart disease risk factors: repeated cross-sectional surveys from the Nord-Trøndelag Health Study (HUNT) 1984-2008. *BMC Public Health* 2012;12:266.
34 Kershaw KN, Droomers M, Robinson WR, *et al*. Quantifying the contributions of behavioral and biological risk factors to socioeconomic disparities in coronary heart disease incidence: the MORGEN study. *Eur J Epidemiol* 2013;28:807–14.
35 Kelli HM, Mehta A, Tahhan AS, *et al*. Low Educational Attainment is a Predictor of Adverse Outcomes in Patients With Coronary Artery Disease. *J Am Heart Assoc* 2019;8:e013165.
36 Tiwari S, Cerin E, Wilsgaard T, *et al*. Lifestyle factors as mediators of area-level socio-economic differentials in cardiovascular disease risk factors. The Tromsø Study. *SSM Popul Health* 2022;19:101241.
37 Chang CH, Lin CS, Luo YS, *et al*. Electrocardiogram-Based Heart Age Estimation by a Deep Learning Model Provides More Information on the Incidence of Cardiovascular Disorders. *Front Cardiovasc Med* 2022;9:754909.
38 Adhikary D, Barman S, Ranjan R, *et al*. A Systematic Review of Major Cardiovascular Risk Factors: A Growing Global Health Concern. *Cureus* 2022;14:e30119.
39 Medina-Inojosa BJ, Medina-Inojosa JR, Chen Z, *et al*. Abstract 16913: The Association Between Adverse Childhood Experiences and Physiologic Age as Determined by Artificial Intelligence-ECG. *Circulation* 2023;148:A16913.
40 Rajai N, Medina-Inojosa JR, Sheffeh MA, *et al*. Abstract 13378: Association Between Social Connection and Biological Age as Determined by Artificial Intelligence-Enabled Electrocardiography. Circulation 2022;146:A13378.
41 Powell KL, Stephens SR, Stephens AS. Cardiovascular risk factor mediation of the effects of education and Genetic Risk Score on cardiovascular disease: a prospective observational cohort study of the Framingham Heart Study. *BMJ Open* 2021;11:e045210.
42 Vo CQ, Samuelsen P-J, Sommerseth HL, *et al*. Validity of self-reported educational level in the Tromsø Study. *Scand J Public Health* 2023;51:1061–8.