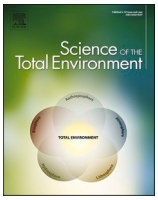
Science of the Total Environment 913 (2024) 169757

Contents lists available at ScienceDirect

Science of the Total Environment

journal homepage: www.elsevier.com/locate/scitotenv



Prenatal and childhood lead exposure is prospectively associated with biological markers of aging in adolescence

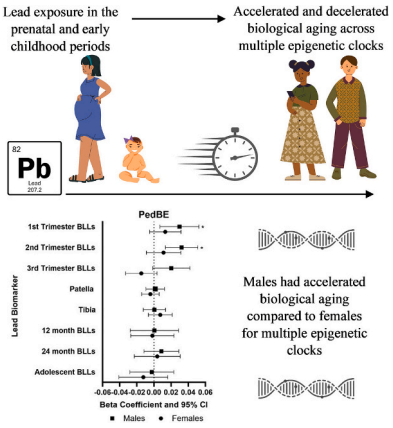
O.M. Halabicky [a,*], M.M. Téllez-Rojo [b], J.M. Goodrich [c], D.C. Dolinoy [a,c], A. Mercado-García [b], H. Hu [d], K.E. Peterson [a]

[a] Department of Nutritional Sciences, School of Public Health, University of Michigan, Ann Arbor, MI, USA
[b] Center for Nutrition and Health Research, National Institute of Public Health, Cuernavaca, Morelos, Mexico
[c] Department of Environmental Health Sciences, School of Public Health, University of Michigan, Ann Arbor, MI, USA
[d] Department of Population and Public Health Sciences, Keck School of Medicine, University of Southern California, Los Angeles, CA, USA

HIGHLIGHTS

- Altered biological aging may harm long term health.
- Early life lead exposure may influence biological aging and later health outcomes.
- Using linear regression and generalized estimating equations, early life lead levels altered adolescent biological age.
- Prenatal lead exposure appeared to be most influential for biological age.
- Lead exposed males appeared to have increased biological age compared to females.

GRAPHICAL ABSTRACT



ARTICLE INFO

Editor: Scott Sheridan

Keywords:
Lead exposure
Child development
Biological age
Epigenetic clock

ABSTRACT

Few studies have related early life lead exposure to adolescent biological aging, a period characterized by marked increases in maturational tempo. We examined associations between prenatal and childhood lead exposure and adolescent biological age (mean 14.5 years) utilizing multiple epigenetic clocks including: intrinsic (IEAA), extrinsic (EEAA), Horvath, Hannum, PhenoAge, GrimAge, Skin-Blood, Wu, PedBE, as well as DNA methylation derived telomere length (DNAmTL). Epigenetic clocks and DNAmTL were calculated via adolescent blood DNA methylation measured by Infinium MethylationEPIC BeadChips. We constructed general linear models (GLMs) with individual lead measures predicting biological age. We additionally examined sex-stratified models and lead by sex interactions, adjusting for adolescent age and lead levels, maternal smoking and education, and proportion of cell types. We also estimated effects of lead exposure on biological age using generalized estimating equations (GEE). First trimester blood lead was positively associated with a 0.14 increase in EEAA age in the GLMs though not the GEE models (95%CI 0.03, 0.25). First and 2nd trimester blood lead levels were associated

* Corresponding author.
E-mail address: oliviamh@umich.edu (O.M. Halabicky).

https://doi.org/10.1016/j.scitotenv.2023.169757
Received 3 October 2023; Received in revised form 22 December 2023; Accepted 27 December 2023
Available online 2 January 2024

with a 0.02 year increase in PedBE age in GLM and GEE models (1st trimester, 95%CI 0.004, 0.03; 2nd trimester, 95%CI 0.01, 0.03). Third trimester and 24 month blood lead levels were associated with a −0.06 and −0.05 decrease in Skin-Blood age, respectively, in GLM models. Additionally, 3rd trimester blood lead levels were associated with a 0.08 year decrease in Hannum age in GLM and GEE models (95%CI -0.15, −0.01). There were multiple significant results in sex-stratified models and significant lead by sex interactions, where males experienced accelerated biological age, compared to females who saw a decelerated biological age, with respect to IEAA, EEAA, Horvath, Hannum, and PedBE clocks. Further research is needed to understand sex-specific relationships between lead exposure and measures of biological aging in adolescence and the trajectory of biological aging into young adulthood.

1. Introduction

Despite large-scale reductions in lead exposure sources, such as leaded paint and gasoline, childhood lead exposure remains a prevalent public health concern. In fact, 1 in 3 children, or around 800 million worldwide, are estimated to have elevated blood lead levels (UNICEF, 2020). In Mexico, the national prevalence of elevated blood lead levels in children ages 1–4 years has been reported to be 17.4 % (Tellez-Rojo et al., 2022). Prevailing exposure is likely due to existing exposure sources, including residual lead paint in homes (Muller et al., 2018), industrial manufacturing sources including air and soil pollution, outdated infrastructure resulting in water contamination (Cradock et al., 2022), and everyday consumer products such as jewelry and children's toys (Centers for Disease Control and Prevention, 2022; Guney and Zagury, 2014). In Mexico specifically, the main source of lead exposure results from lead glazed ceramics which are used to cook, store, and serve foods (Romieu et al., 1994).

Environmental toxicant exposure in early life may alter biological processes that increase risk for adult disease, as outlined by the biological embedding model (Shonkoff et al., 2009). Exposure in utero and in early childhood periods may be particularly detrimental as these are critical developmental periods, where neurological and physiological developmental processes are particularly susceptible to environmental exposures such as lead (Nelson and Gabard-Durnam, 2020). Children are especially susceptible to lead exposure given their increased intestinal absorption rates of lead compared to adults (Ziegler et al., 1978), and overall hand-to-mouth activity for exploration (Hauptman et al., 2017). There is evidence for adverse associations between lead exposure in the prenatal and postnatal period and adverse adolescent and adult neurocognitive (Stiles and Bellinger, 1993; Cecil et al., 2008) and health outcomes (Navas-Acien et al., 2007; Naicker et al., 2011). Still, although the associations between lead exposure and cognition are well-known, adolescence remains an understudied period of outcome assessment (Arnold and Liu, 2020) Despite widespread investigation of deleterious associations, further research is needed to understand biological mechanisms along the causal pathway and how lead exposure may become biologically embedded and impact health across the life course.

There is increasing evidence that the toxicant and social environment may be biologically embedded through accelerated biological aging of cells and tissues (Kochmanski et al., 2017; Simons et al., 2021). Biological aging can be measured via 'epigenetic clocks' (Horvath, 2013) that quantify biological age via DNA methylation at specific CpG sites across the genome. Biological aging is thought to be predictable and correlates with chronological age. Biological age acceleration or deceleration, however, occurs when biological age is greater or less than chronological age, respectively. Environmental factors such as tobacco smoke (Carter et al., 2022) and air pollution (Wang et al., 2020; White et al., 2019), as well as social factors such as adversity (Marini et al., 2020) and neighborhood deprivation (Lawrence et al., 2020), have been associated with biological age acceleration, while maternal prenatal selective serotonin reuptake inhibitor (SSRI) usage has been linked to biological age deceleration in infants (McKenna et al., 2021). In rodent models, lead exposure has been shown to alter age-related methylation (Faulk et al., 2014).

Epigenetic clocks can provide a window into the biological aging of an individual in relation to their chronological age and are a potential predictor of long-term health (Marioni et al., 2016). Biological aging alteration has been associated with chronic disease development and mortality (Joyce et al., 2021; Hillary et al., 2020). Even in adolescence, biological age acceleration has been associated with physiological changes, such as accelerated pubertal development (Suarez et al., 2018), and alterations to brain volumes, cortical thickness, and cortisol surface area (Hoare et al., 2020), which may represent earlier aging and accelerated health risks.

Few studies have examined associations between lead exposure and biological aging through epigenetic clocks (Herrera-Moreno et al., 2021; Javed et al., 2016). These studies reported no significant associations between prenatal lead exposure and infant biological aging. In adult samples, however, increasing lead exposure levels from the 25th to 75th percentile have been associated with a ~ 0.25- year increase in biological age measured through all-cause mortality clocks (Lodge et al., 2022). There remains a gap in the literature examining adolescent populations, a critical period of physiological development.

To our knowledge, there are no studies examining associations between early life lead exposure, during critical periods of development, and adolescent biological age while considering sex-based differences. The aims of this study were twofold: 1) To examine associations between prenatal and early childhood lead exposure and multiple epigenetic clocks and estimated telomere length in adolescence and 2) examine potential effect modification of sex on the relationship between lead and biological age.

2. Materials and methods

2.1. Population

The current study included a subsample of participants in two of three cohorts comprising the Early Life Exposure in Mexico to Environmental Toxicants (ELEMENT) project. Between 1997 and 2005, women were recruited from prenatal clinics of the Mexican Social Security Institute in Mexico City Hospital Gea González (Perng et al., 2019). Offspring were followed at 12 and 24 months and again in adolescence. The analytical subsample for this study participated in an adolescent follow-up visit ($N = 521$ with available biological age variables) and had previous blood lead level data spanning from prenatal to the early childhood period. Due to missing covariates and lead exposure variables, the analytical subsample ranged from $N = 264$ to $N = 390$ depending on available data for each general linear model (GLM). The institutional review boards at the University of Michigan and the Mexico National Institute of Public Health approved the research protocols. Informed consent was obtained from parents of the participants, and assent was also received from the child participants starting at age 7 years.

2.2. Lead biomarkers

Lead was measured in the mother during the prenatal and early postpartum period as well as in childhood. Maternal blood lead was

measured at each trimester of pregnancy and child blood lead was quantified at months 12 and 24 of age. Blood samples were collected and stored in trace-metal-free tubes by trained research assistants using a standardized protocol to avoid lead contamination. These samples were measured using graphite-furnace atomic-absorption spectroscopy (model 3000; Perkin-Elmer, Chelmsford, MA, USA) at a research facility of the American British Cowdray Hospital in Mexico City. Blood samples were also collected in the adolescent follow-up visit and measured via inductively coupled plasma mass spectrometry with dynamic reaction cell mass spectrometry (ICP-DRC-MS: Agilent 8900) using digested blood at the Michigan Department of Community Health Trace Metals Laboratory, a nationally accredited facility for lead analysis. All blood lead levels (BLLs) were above the reporting limit of detection (1 μg/dl) for both instruments.

Maternal patella and tibia bone lead are considered valid measurements of cumulative prenatal lead exposure due to mobilization of lead from the bone during pregnancy and transfer through the placenta (Hu et al., 1991; Centers for Disease Control and Prevention Work Group on Lead and Pregnancy, 2010). Patella and tibia bone lead were assessed on each leg, between 1 and 12 months postpartum, using a K X-ray Fluorescence (K-XRF) instrument. The two estimates for each bone were obtained and these values were computed, averaged, and weighted by the inverse of the proportion of the measurement error corresponding to each measure as described (Téllez-Rojo et al., 2002). In this study, we considered only the first bone lead measure available for each mother; >90 % of observations were from the first month postpartum.

2.3. DNA methylation assessment

DNA isolated from blood leukocytes in adolescence were used to assay DNAm at ~850 K sites across the genome using the Infinium MethylationEPIC array (Moran et al., 2016). Within batch, samples were randomized across chips and chip positions and then hybridized and scanned at the University of Michigan Advanced Genomics Core via an Illumina iScan instrument, according to manufacturer's protocols for EPIC arrays. Data resulting from the BeadChip were processed according to a previously utilized pipeline (Goodrich et al., 2016a). Raw data including average betas, the proportion of methylated cytosines at a given site, were fed into R Project for Statistical Computing using *minfi (*Aryee et al., *2014)*. To remove unwanted technical variation estimated from control probes included on each chip, background correction, dye bias correction, and functional normalization were performed (Fortin et al., 2017). Probes that were poorly detected (in at least 5 % of samples), known to be cross-reactive, and with polymorphisms in the CpG site or the single base extension site were excluded (Jansen et al., 2021).

2.4. Biological age biomarkers

Biological age was estimated using biomarkers that were calculated from the Infinium methylation data using publicly available software by Horvath et al. (Horvath, 2013) The data was uploaded to the New Methylation Age Calculator (https://dnamage.genetics.ucla.edu/new) to calculate multiple epigenetic clocks as well as estimated telomere length. We uploaded probe data after background and dye bias correction before removing failed probes and quantile normalization (Jansen et al., 2021). Imputation and normalization through the default and recommended methods by the software were completed after uploading data to the calculator. The clocks generated for this study were the Horvath clock which was designed to be agnostic to the source of tissue type (Horvath, 2013); the Skin-Blood clock, a more robust estimator for skin, blood, or saliva samples (Horvath et al., 2018); the Hannum clock which was designed for blood samples (Hannum et al., 2013); and PhenoAge and GrimAge which are composite measures considered to be better predictors of all-cause mortality, cancers, and other adverse health outcomes compared to the original clocks (Levine et al., 2018; Lu et al., 2019a). We additionally calculated clocks which were trained on Infinium methylation datasets for children. We used the package 'methylclock in R (Pelegí-Sisó et al., 2021) to calculate the Pediatric-Buccal-Epigenetic (PedBE) clock and Wu's clock. PedBE was originally trained on buccal cell data from children ages 0 to 20 years. It performs best in buccal and saliva samples, with some utility in blood derived DNA from children (McEwen et al., 2020). Wu's clock was trained on samples from 0 to 18 year old children using blood DNA (Wu et al., 2019). To calculate PedBE and Wu, we used fully processed betas (with quantile normalization) and used the default imputation procedure (KNN) for missing values. Finally, we calculated estimated telomere length (DNAmTL) which is a validated estimator of telomere length based on DNA methylation of 140 CpG sites (Lu et al., 2019b). DNAmTL has been shown to highly correlated with measured leucocyte telomere length. Additionally, DNAmTL has been shown to correlate more strongly with chronological age and serve as a better predictor of mortality and health outcomes compared to measured leucocyte telomere length (Lu et al., 2019b).

We also calculated intrinsic epigenetic age acceleration (IEAA) and extrinsic epigenetic age acceleration (EEAA). IEAA is the residual after regressing the Horvath clock on chronological age and cell type estimates and is considered an indicator of cellular aging independent of cellular composition (Smith et al., 2019). By design, EEAA captures cellular methylation changes and extracellular changes in blood cell composition, and the method to obtain EEAA was previously described (Chen et al., 2016).

Each epigenetic clock was regressed on chronological age where the residual represented the variable to be used in the analysis, here referred to as epigenetic age. The values are then independent of chronological age and positive or negative values indicate an accelerated or decelerated biological age compared to chronological age, respectively. In total, there were 521 observations for the epigenetic age variables including IEAA, EEAA, Horvath, Hannum, PhenoAge, Skin-Blood, GrimAge, DNAmTL, PedBE, and Wu.

2.5. Covariates

Covariates were selected based on a priori knowledge and a bivariate analysis relating covariates to biological age biomarkers. All models were adjusted for sex and chronological age at the adolescent follow-up visit. Maternal years of education and smoking during pregnancy were reported at the prenatal baseline visit via a research assistant administered questionnaire. Maternal smoking during pregnancy has been associated with alterations in DNA methylation within the infant, child, and adolescent epigenome (Joubert et al., 2016; Rauschert et al., 2019). Maternal education was included as a proxy for socioeconomic status, which has been associated with longitudinal alterations to DNA methylation (Needham et al., 2012; Needham et al., 2021) and biological age acceleration (Fiorito et al., 2017). Adolescent BLLs were included as a covariate to determine if any associations between prenatal or early childhood lead exposure and biological age were actually due to concurrent adolescent BLLs. Finally, because blood is a heterogeneous tissue, we adjusted for proportions of various cell types in each sample in our models. The proportion of cell types were measured using methods based off of Infinium methylation data. Specifically, we estimated proportions of CD4+ and CD8+ T cells, natural killer (NK) cells, B cells, monocytes and granulocytes along with relative abundance of plasmablasts and 3 sub-populations of T-cells that change with aging (CD8 + CD28-CD45RA-, naive CD4+ and CD8+ T cells) using the software developed by Horvath et al. (Horvath, 2013; Lu et al., 2019a)

2.6. Statistical analysis

We first assessed Pearson's correlations between the epigenetic clocks (variables not regressed on age) and chronological age and calculated mean absolute error (MAE), a measure of the difference between chronological age and the clock's estimated age. Next, we

compared descriptive statistics for biological age and sociodemographic variables for the full sample and by sex using *t*-tests and chi-square tests. We further tested for differences in demographic and predictor variables between the included and excluded sample using t-tests and chi-square tests. To test associations between lead values (1st-3rd trimester blood, 12 & 24 month blood, and tibia and patella bone) and biological age in adolescence, we constructed generalized linear regression models (GLMs). We constructed a single model for each lead variable predicting biological age outcomes and adjusted for covariates. GLMs for IEAA and EEAA did not include cell type proportions as covariates, as these values by design take into account cell composition. All other biological age outcomes included cell covariates. As a sensitivity analysis, we log-transformed all lead variables and reran models. To consider effect modification by sex, we conducted sex-stratified models for each biological age outcomes. We next ran the GLMs while including a lead x sex interaction. To investigate significant interactions, we mean centered the lead values and plotted simple slopes for values of lead at the mean and at ±1 μg/dl increments above and below the mean for both males and females. Finally, to examine the joint effects of lead exposure across multiple collection periods on biological aging, we constructed modified generalized estimating equations (GEEs). In these adjusted models, lead exposure variables were structured as repeated measures at each time point to predict each biological age outcome. This method, unlike GLM, allows for correlation between the repeat exposure measures and assists in determining windows of susceptibility for exposures (Sánchez Brisa et al., 2011). We additionally tested for significant differences between exposure associations in each GEE model. As a secondary analysis, we also included models without cell type adjustment and examined differences between the original models (Supplemental Tables 1–3).

3. Results

In our subsample, there were 268 females (Table 1; 51.94 %). Mean age was 14.5 years and did not significantly vary by sex. Mean years of maternal education was 10.92 years and few mothers reported smoking during pregnancy (n = 17, 3.29 %). Epigenetic clocks values varied significantly between males and females. For those that did significantly vary, males appeared to have greater epigenetic age and shortened DNAmTL compared to females, except PhenoAge, where females were greater than males. Maternal BLL means at each trimester ranged from 4.95 to 5.62 μg/dl. Maternal postpartum mean bone lead measures of the tibia and patella were 9.11 and 10.69 μg/g, respectively. Child mean BLLs increased from 12 (4.36 μg/dl) to 24 months (4.64 μg/dl). When comparing the analytic and excluded samples, there were no significant differences in demographic characteristics or predictor variables except for two measures. Excluded participants had significantly greater 12 months BLLs (Analytic sample mean: 4.95 μg/dl; excluded sample mean: 5.59 μg/dl; p = 0.047) and were more likely to have mothers who reported smoking during pregnancy (Reported smoking in analytic sample: n = 17, 3.28 %; p = 0.031; Reported smoking in excluded sample: n = 54, 5.83 %).

The epigenetic clock variables and DNAmTL were significantly correlated with chronological age (Supplemental Fig. 1). All associations were positive, except DNAmTL, which showed a negative association as chronological age increased, as expected (r = −0.4053, p < 0.000). MAEs ranged from 0.43 to 3.88.

The adjusted GLMs were run to examine associations between early life lead exposure and adolescent biological age using the calculated epigenetic age measures and DNAmTL. The final covariate cell-types included CD4+ T cells, NK cells, monocytes, granulocytes, plasmablasts, CD8 + CD28-CD45RA-, and naïve CD8+ T cells as suggested by Horvath et al. (Horvath, 2013). For EEAA, a 1 μg/dl increase in 1st trimester maternal blood lead was significantly associated with a 0.14 year increase in EEAA (Table 2: 95%CI 0.03, 0.25). There were no other significant associations for IEAA and EEAA in the GLM models, however, GEE models suggest 3rd trimester maternal blood lead was associated with a 0.07 year decrease in EEAA (95%CI -0.15, −0.003) while maternal tibia bone lead was associated with a 0.08 year increase in IEAA (95%CI 0.001, 0.15). Among pediatric epigenetic clocks, a 1 μg/dl increase in 1st trimester blood lead (Table 3: 95%CI 0.004, 0.03) and 2nd trimester blood lead (95%CI 0.01, 0.03) were associated with a 0.02 year increase in PedBE age in GLMs. The GEE analysis also suggested 2nd trimester maternal blood lead as associated with a 0.02 year increase in PedBE age (95%CI 0.003, 0.03). In a secondary analysis without adjustment for cell types, 1st trimester blood (β = 0.02; 95%CI 0.001, 0.03) and tibia (β = 0.01; 95%CI 0.001, 0.02) bone lead levels were additionally associated with accelerated Wu age (Supplemental Table 1). For adult epigenetic clocks, a 1 μg/dl increase in 3rd trimester blood lead was associated with a 0.08 year decrease in Hannum age (Table 4: 95%CI -0.15, −0.01) which was replicated in the GEE findings. This association was not significant in models without cell type adjustment (Supplemental Table 1). The GEE analysis also showed maternal tibia bone lead was associated with a 0.08 year increase in Horvath age (95%CI 0.001, 0.15). A 1 μg/dl increase in 3rd trimester blood lead was associated with a decrease in 0.06 years (Table 4: 95%CI -0.09, −0.02)

Table 1
Participant characteristics.

	N	Mean ± SD (or number and %)			p-Value
		Total	Males	Females	
Child characteristics					
Age (years)	523	14.5 ± 2.11	14.50 ± 2.05	14.52 ± 2.17	0.911
IEAA	521	0.01 ± 3.28	0.32 ± 3.24	−0.26 ± 3.30	0.043*
EEAA	521	0.29 ± 4.11	0.80 ± 04.16	−0.16 ± 4.03	0.008*
Horvath	523	17.54 ± 4.20	18.21 ± 4.10	16.94 ± 4.20	0.001*
Hannum	523	11.17 ± 3.83	11.48 ± 3.82	10.90 ± 3.83	0.083
PhenoAge	523	2.79 ± 6.00	2.08 ± 5.48	3.42 ± 6.38	0.011*
Skin-Blood	523	10.51 ± 2.41	10.45 ± 2.34	10.56 ± 2.47	0.617
GrimAge	521	21.72 ± 3.48	21.88 ± 3.18	21.58 ± 3.73	0.31
PedBe	523	7.33 ± 0.63	7.36 ± 0.59	7.30 ± 0.66	0.224
Wu	523	8.36 ± 0.77	8.41 ± 0.77	8.32 ± 0.76	0.200
DNAmTL	523	7.69 ± 0.17	7.65 ± 0.16	7.73 ± 0.16	0.000*
12 month blood lead	409	4.36 ± 3.13	4.51 ± 3.04	4.24 ± 3.20	0.811
24 month blood lead	452	4.64 ± 3.44	4.71 ± 3.82	4.59 ± 3.03	0.646
Maternal characteristics					
1st Trimester blood lead (mcg/dl)	372	5.62 ± 4.25	5.39 ± 3.68	5.84 ± 4.73	0.156
2nd Trimester blood lead (mcg/dl)	379	4.95 ± 4.04	4.83 ± 3.83	5.06 ± 4.24	0.286
3rd Trimester blood lead (mcg/dl)	360	5.38 ± 4.13	5.22 ± 3.53	5.53 ± 4.63	0.232
Tibia bone lead (mcg/g)	295	9.11 ± 7.04	9.56 ± 7.70	9.01 ± 7.33	0.736
Patella bone lead (mcg/g)	336	10.69 ± 8.68	10.48 ± 8.38	11.86 ± 9.69	0.083
Years of school	520	10.92 ± 2.91	11.11 ± 2.85	10.75 ± 2.96	0.159
Smoking during pregnancy	516				0.933
Yes		17 (3.29 %)	8 (3.23 %)	9 (3.36 %)	
No		499 (96.71 %)	240 (96.71 %)	259 (96.64 %)	

* Student t-test or χ2 test was used for significant difference test.

Table 2
Generalized linear regression (GLM) and generalized estimating equation (GEE) results for cell based epigenetic clocks.

Lead values	IEAA					EEAA				
	GLM			GEE[3]		GLM			GEE	
	N	β	95 % CI	β	95%CI	N	β	95 % CI	β	95%CI
Maternal bone and blood lead[1]										
1st Trimester (mcg/dl)	361	−0.01	−0.09, 0.08	−0.02	−0.11, 0.06	**361**	**0.14**	**0.03, 0.25***	0.000	−0.07, 0.07
2nd Trimester (mcg/dl)	369	−0.02	−0.11, 0.07	−0.03	−0.11, 0.06	369	0.08	−0.03, 0.19	−0.02	−0.09, 0.06
3rd Trimester (mcg/dl)	350	−0.08	−0.17, 0.01†	−0.07	−0.16, 0.06	350	−0.03	−0.13, 0.08	**−0.07**	**−0.15, −0.003***
Tibia (g/mg)	264	−0.03	−0.09, 0.03	**0.08**	**0.001, 0.15***	264	−0.004	−0.08, 0.07	0.02	−0.04, 0.08
Patella (g/mg)	275	−0.02	−0.06, 0.03	−0.02	−0.08, 0.04	275	−0.03	−0.08, 0.03	−0.02	−0.07, 0.03
Child blood lead[1]										
12 month (mcg/dl)	288	−0.03	−0.15, 0.09	−0.05	−0.15, 0.05	288	−0.04	−0.20, 0.12	−0.07	−0.16, 0.01
24 month (mcg/dl)	331	−0.02	−0.14, 0.08	−0.01	−0.10, 0.07	331	−0.05	−0.19, 0.08	−0.06	−0.14, 0.01
Adolescent blood lead[2]	390	−0.04	−0.17, 0.08	−0.07	−0.17, 0.04	390	0.04	−0.12, 0.19	0.02	−0.07, 0.03
p-value; joint effects of lead exposure in GEE					0.2753					0.3905

[1] Models adjusted for age, sex, adolescent blood lead, maternal smoking and schooling, and proportions of cell types CD4 + T, NK, Mono, Gran, PlasmaBlast, +CD28-CD45RA-, naïve CD8+ T.
[2] Models adjusted for age, sex, maternal smoking and schooling, and proportions of cell types CD4 + T cells, natural killer cells, monocytes, granulocytes, plasmablasts, +CD28-CD45RA-, naïve CD8 + T cells.
[3] GEE $N = 515$, with 2715 observations. Model adjustment[1].

Table 3
Generalized linear regression (GLM) and generalized estimating equation (GEE) results for pediatric epigenetic clocks.

Lead values	PedBE					Wu				
	GLM			GEE[3]		GLM			GEE	
	N	β	95 % CI	β	95 % CI	N	β	95 % CI	β	95 % CI
Maternal bone and blood lead[1]										
1st Trimester (mcg/dl)	**361**	**0.02**	**0.004, 0.03***	0.01	−0.00, 0.02	361	0.01	−0.004, 0.03	0.01	−0.002, 0.03
2nd Trimester (mcg/dl)	**369**	**0.02**	**0.01, 0.03**	**0.02**	**0.003, 0.03***	369	0.003	−0.01, 0.02	0.01	−0.01, 0.02
3rd Trimester (mcg/dl)	350	−0.001	−0.01, 0.01	−0.004	−0.02, 0.01	350	0.001	−0.01, 0.02	0.002	−0.01, 0.02
Tibia (g/mg)	264	0.003	−0.01, 0.01	0.003	−0.01, 0.01	264	0.01	−0.003, 0.01	0.01	−0.0004, 0.03†
Patella (g/mg)	275	−0.001	−0.01, 0.01	0.002	−0.01, 0.01	275	0.003	−0.01, 0.01	0.01	−0.01, 0.02
Child blood lead[1]										
12 month (mcg/dl)	288	0.001	−0.02, 0.02	0.01	−0.01, 0.02	288	−0.02	−0.04, 0.01	−0.01	−0.02, 0.01
24 month (mcg/dl)	331	0.01	−0.01, 0.02	0.01	−0.01, 0.02	331	−0.004	−0.02, 0.02	0.001	−0.01, 0.02
Adolescent blood lead[2]	390	−0.1	−0.03, 0.01	−0.005	−0.02, 0.01	390	0.003	−0.02, 0.02	0.02	−0.002, 0.03†
p-value; joint effects of lead exposure in GEE					0.4095					0.5990

* $p < 0.050$.
[1] Models adjusted for age, sex, adolescent blood lead, maternal smoking and schooling, and proportions of cell types CD4 + T, NK, Mono, Gran, PlasmaBlast, +CD28-CD45RA-, naïve CD8+ T.
[2] Models adjusted for age, sex, maternal smoking and schooling, and proportions of cell types CD4 + T cells, natural killer cells, monocytes, granulocytes, plasmablasts, +CD28-CD45RA-, naïve CD8 + T cells.
[3] GEE $N = 515$, with 2715 observations. Model adjustment[1].

and 24 month blood lead associated with a decrease in 0.05 years (95% CI -0.09, −0.005) for Skin-Blood age. The GEE analysis replicated findings for 3rd trimester maternal blood lead and Skin-Blood age only, and additionally suggested a significant association between maternal tibia bone lead and a 0.04 increase in Skin-Blood age (95%CI 0.01, 0.07). In models without cell type adjustment, 1st trimester blood lead was associated with a 0.18 year increase in PhenoAge (Supplemental Table 1: 95%CI 0.05, 0.30). Considering all GEE results, differences in exposure associations at different time periods was only significant for the Skin-Blood clock ($p = 0.0003$). In our sensitivity analysis with log-transformed lead variables, there were no significant differences in model fit or in observed associations.

Throughout these analyses, sex was a significant predictor of biological age. In sex-stratified models, males and females differed in the direction of significant associations. For females, 3rd trimester lead exposure was associated with a decreased Hannum, Skin and Blood, and IEAA age (Figs. 1 and 2). For males, however, 1st trimester lead exposure was associated with increased PedBE and EEAA ages, while 2nd trimester lead exposure was associated with increased PedBE only (Figs. 2 and 3).

There were multiple significant interaction effects, all suggesting that males experienced biological age acceleration related to increasing lead exposure in both the prenatal and childhood periods. We present significant results for lead by sex interactions in GLMs in Supplemental Table 3 and an example plotted interaction in Fig. 4. Both prenatal and childhood lead exposure were associated with biological age acceleration for males and deceleration for females. For example, the interaction effects for 1st trimester, 3rd trimester, 12 month, and 24 month blood lead and males were positively significant for both IEAA and Horvath age (Supplemental Table 3). For both the EEAA and Hannum clocks, 12 month blood lead appeared to have a significant interaction where males showed accelerated biological aging. Finally, 3rd trimester BLLs significantly interacted where males saw an accelerated PedBE clock. The models reported in Supplemental Table 3 include each lead measure centered at their mean value. Investigating the margins showed that while the effect of lead on accelerated biological aging for males did reach significance in some models as lead increased, females also saw significant decelerated biological aging as lead levels increased (Fig. 4). There were no significant interactions between bone lead levels and sex.

Table 4
Generalized linear regression (GLM) and generalized estimating equation (GEE) results for adult epigenetic clocks and DNAmTL.

Lead values	Horvath					Hannum				
	GLM			GEE[3]		GLM			GEE	
	N	β	95 % CI	β	95 % CI	N	β	95 % CI	β	95 % CI
Maternal bone and blood lead[1]										
1st Trimester (mcg/dl)	361	0.004	−0.09, 0.10	−0.02	−0.11, 0.06	361	0.01	−0.06, 0.08	0.00	−0.06, 0.06
2nd Trimester (mcg/dl)	369	−0.01	−0.10, 0.07	−0.03	−0.11, 0.06	369	−0.01	−0.08, 0.06	−0.02	−0.08, 0.05
3rd Trimester (mcg/dl)	350	−0.07	−0.15, 0.02	−0.07	−0.16, 0.01	**350**	**−0.08**	**−0.15, −0.01**	**−0.07**	**−0.14, −0.002***
Tibia (g/mg)	264	−0.03	−0.09, 0.03	**0.08**	**0.001, 0.15***	264	−0.01	−0.05, 0.04	0.02	−0.04, 0.08
Patella (g/mg)	275	−0.02	−0.06, 0.03	−0.02	−0.08, 0.04	275	−0.03	−0.07, 0.002†	−0.02	−0.07, 0.03
Child blood lead										
12 month (mcg/dl)	288	−0.02	−0.15, 0.10	−0.05	−0.15, 0.05	288	−0.05	−0.15, 0.04	−0.07	−0.15, 0.01†
24 month (mcg/dl)	331	−0.02	−0.13, 0.09	−0.01	−0.10, 0.07	331	−0.08	−0.18, 0.01†	−0.06	−0.13, 0.01†
Adolescent blood lead[2]	390	−0.05	−0.17, 0.08	−0.07	−0.17, 0.04	390	0.02	−0.08, 0.11	0.01	−0.06, 0.09
p-value; joint effects of lead exposure in GEE					0.2779					0.3909

Lead values	Skin-Blood				PhenoAge					
	GLM			GEE		GLM			GEE	
	N	β	95 % CI	β	95 % CI	N	β	95 % CI	β	95 % CI
Maternal bone and blood lead[1]										
1st Trimester (mcg/dl)	361	0.000	−0.04, 0.04	−0.01	−0.05, 0.02	361	0.06	−0.04, 0.17	0.04	−0.06, 0.14
2nd Trimester (mcg/dl)	369	−0.02	−0.05, 0.02	−0.02	−0.06, 0.01	369	0.02	−0.08, 0.13	0.02	−0.08, 0.12
3rd Trimester (mcg/dl)	**350**	**−0.06**	**−0.09, −0.02***	**−0.06**	**−0.10, −0.03***	350	−0.02	−0.13, 0.08	−0.03	−0.13, 0.07
Tibia (g/mg)	264	−0.003	−0.03, 0.02	**0.04**	**0.01, 0.07***	264	0.02	−0.05, 0.09	0.08	−0.01, 0.17†
Patella (g/mg)	275	−0.01	−0.03, 0.01	0.02	−0.01, 0.04	275	0.02	−0.03, 0.08	0.03	−0.04, 0.11
Child blood lead										
12 month (mcg/dl)	288	−0.04	−0.09, 0.01	−0.04	−0.08, 0.004†	288	−0.05	−0.20, 0.10	−0.05	−0.17, 0.06
24 month (mcg/dl)	**331**	**−0.05**	**−0.09, −0.005**	−0.04	−0.07, 0.001†	331	−0.10	−0.22, 0.03	−0.08	−0.19, 0.02
Adolescent blood lead[2]	390	−0.04	−0.09, 0.01†	−0.03	−0.07, 0.02	390	−0.03	−0.18, 0.12	−0.005	−0.12, 0.11
p-value; joint effects of lead exposure in GEE[3]					**0.0003***					0.3066

Lead values	GrimAge					DNAmTL				
	GLM			GEE		GLM			GEE	
	N	β	95 % CI	β	95 % CI	N	β	95 % CI	β	95 % CI
Maternal bone and blood lead[1]										
1st Trimester (mcg/dl)	361	−0.02	−0.09, 0.05	−0.02	−0.09, 0.04	361	0.000	−0.003, 0.003	−0.000	−0.003, 0.003
2nd Trimester (mcg/dl)	369	−0.03	−0.10, 0.04	−0.04	−0.11, 0.02	369	0.002	−0.002, 0.005	0.001	−0.002, 0.004
3rd Trimester (mcg/dl)	350	0.03	−0.04, 0.10	0.004	−0.06, 007	350	0.002	−0.001, 0.005	0.001	−0.002, 0.004
Tibia (g/mg)	264	0.04	−0.01, 0.09†	0.04	−0.01, 0.10	264	−0.001	−0.003, 0.001	−0.001	−0.003, 0.002
Patella (g/mg)	275	0.02	−0.02, 0.05	0.005	−0.04, 0.05	275	0.001	−0.001, 0.002	−0.000	−0.002, 0.002
Child blood lead										
12 month (mcg/dl)	288	0.03	−0.07, 0.12	0.02	−0.06, 0.09	288	0.001	−0.003, 0.01	−0.000	−0.004, 0.003
24 month (mcg/dl)	331	0.02	−0.07, 0.10	0.004	−0.06, 007	331	0.001	−0.002, 0.005	−0.001	−0.004, 0.002
Adolescent blood lead[2]	**390**	**−0.11**	**−0.20, −0.01***	−0.04	−0.12, 0.04	390	−0.003	−0.01, 0.002	−0.003	−0.01, 0.001
p-value; joint effects of lead exposure in GEE					0.5872					0.8621

* $p < 0.050$.
[1] Models adjusted for age, sex, adolescent blood lead, maternal smoking and schooling, and proportions of cell types CD4 + T, NK, Mono, Gran, PlasmaBlast, CD8 + CD28-CD45RA-, naïve CD8 + T.
[2] Models adjusted for age, sex, maternal smoking and schooling, and proportions of cell types CD4 + T cells, natural killer cells, monocytes, granulocytes, plasmablasts, CD8 + CD28-CD45RA-. naïve CD8 + T cells.
[3] GEE N = 515, with 2715 observations. Model adjustment[1].

4. Discussion

Our findings suggest that prenatal lead exposure may alter trajectories of biological aging, as approximated via the adolescent epigenome. Results from GLMs suggest 1st trimester blood lead levels were associated with biological age acceleration in adolescence via the EEAA and PedBE clocks. However, 3rd trimester and 24 month blood lead levels were associated with a decelerated Skin-Blood age as well as Hannum age (3rd trimester only). Results from the GEE analysis largely replicated the GLM results, while also suggesting significant associations between maternal tibia bone lead and accelerated biological aging. There was a significant sex-stratified associations and lead x sex interaction throughout our analyses where males appeared to have greater associations between lead and biological age acceleration compared to females, who saw biological age deceleration. Overall, our results suggest that early life lead exposure has a long-lasting influence on biological aging, stretching into adolescence.

To our knowledge, this is the only study available examining associations between prenatal and childhood lead exposure and adolescent biological age. Two previous studies of prenatal lead exposure and infant biological age reported non-significant associations (Herrera-Moreno et al., 2021; Javed et al., 2016). We report, however, few significant associations between prenatal blood lead and altered adolescent biological age, both accelerated and decelerated aging, suggesting that associations between prenatal lead exposure and biological age become observable after early childhood. While we hypothesized that early life

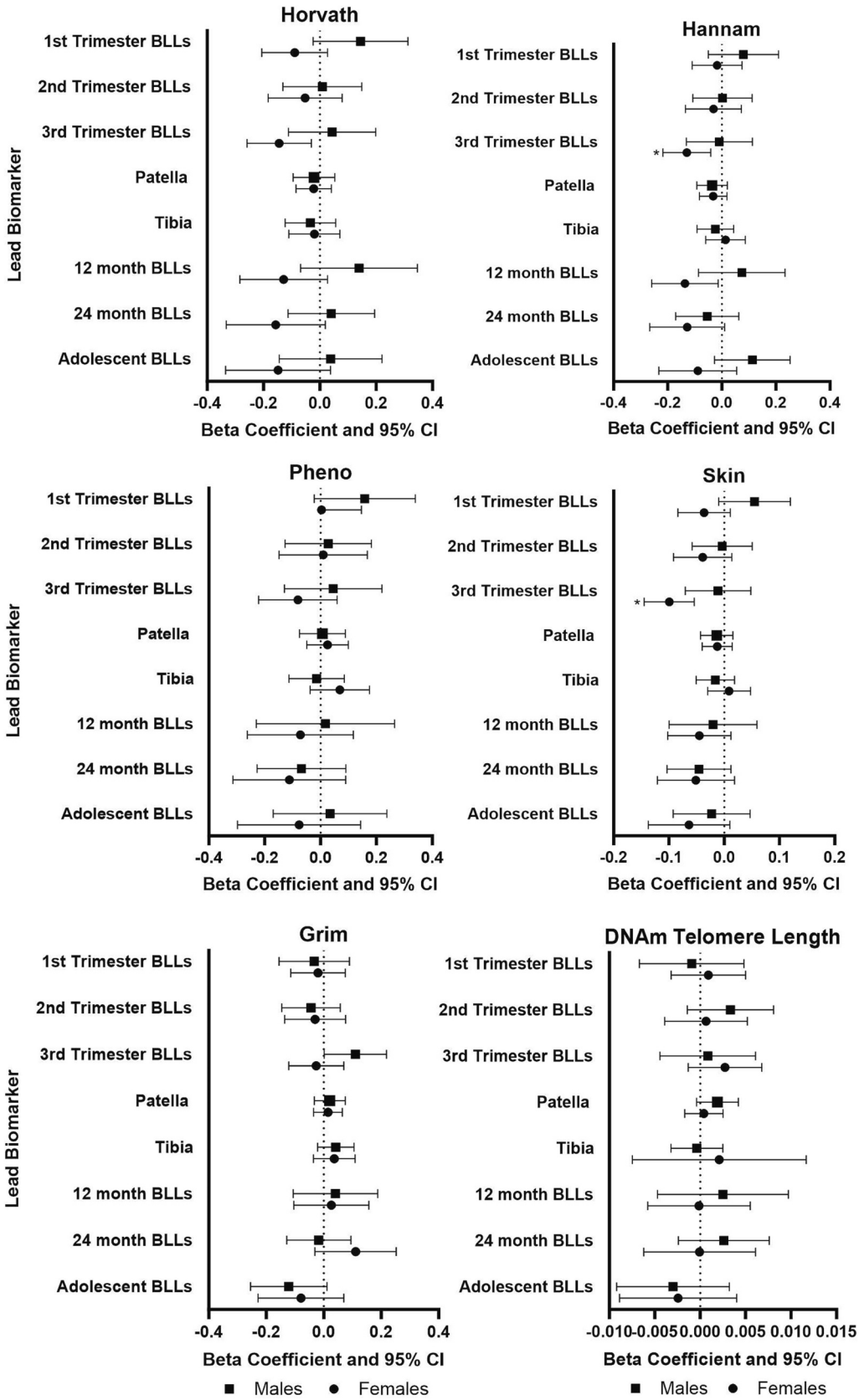


Fig. 1. Forest plot of GLM results for sex-stratified analysis of adult epigenetic clocks and DNAmTL.

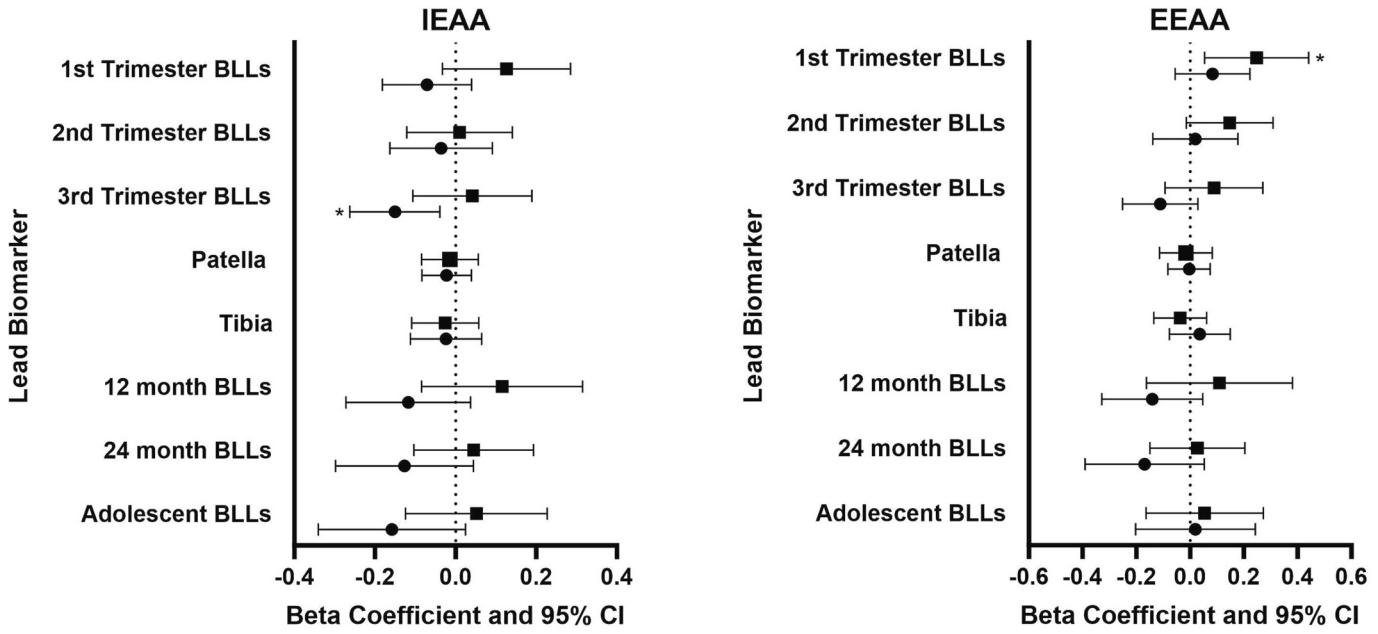


Fig. 2. Forest plot of GLM results for sex-stratified analysis of IEAA and EEAA.

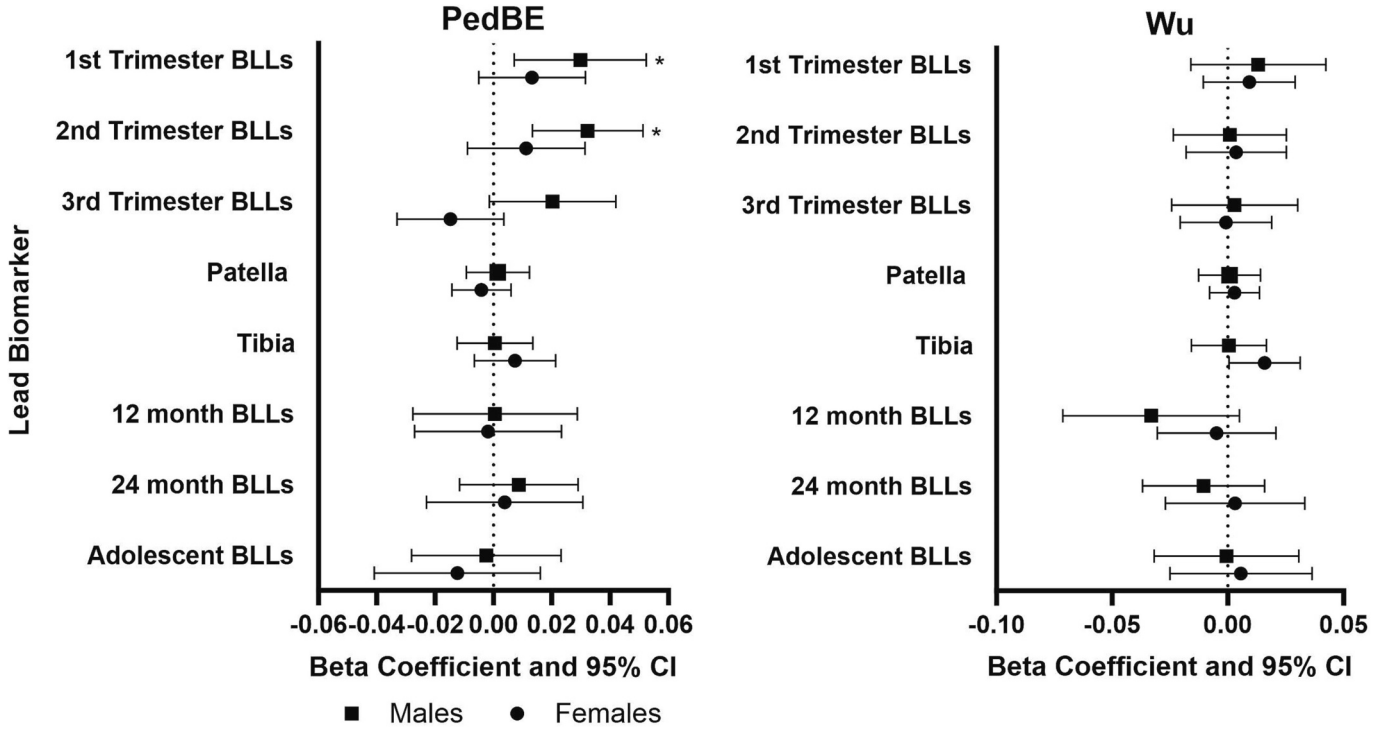


Fig. 3. Forest plot of GLM results for sex-stratified analysis of pediatric epigenetic clocks.

lead exposure would be associated with accelerated biological aging, in some GLMs we report the opposite finding, of lead being associated with biological age deceleration. This finding was replicated in our GEE analyses, where lead exposure effects showed both negative and position associations with biological aging measures. Other environmental exposures in childhood, such as pesticides, have been cross-sectionally associated with decelerated biological age of the Skin-Blood clock in children (de Prado-Bert et al., 2021). Further study is needed with prenatal lead exposure and multiple epigenetic clocks in adolescence to determine associations and build the evidence base. It would also be beneficial to observe how clock estimates change throughout adolescence into young adulthood to determine the influence of lead on biological aging during this critical developmental period, assess change in maturational tempo, and relation to health outcomes.

Our study found significant sex-stratification results and interaction effects between lead x sex where males saw a positive association between lead and biological age acceleration, compared to females who saw biological age deceleration. In general, males in our sample had greater biological age compared to females, consistent with other reports suggesting that male sex is significantly associated with accelerated age in multiple epigenetic clocks (Oblak et al., 2021; Horvath et al., 2016; Crimmins et al., 2021). Epigenome-wide studies have also found

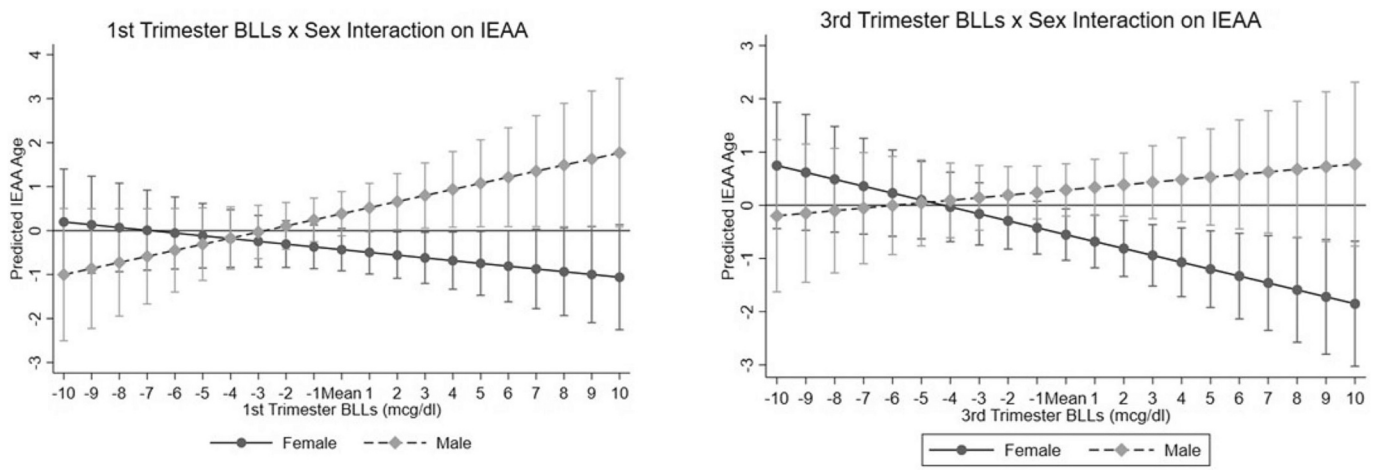


Fig. 4. Interactions between prenatal blood lead levels (BLLs) and sex for IEAA.

sex-dependent associations between lead exposure and cord blood leukocyte DNA methylation (Sen et al., 2015) and LINE-1 methylation in peri-adolescents (Goodrich et al., 2016b). Differences in associations could be due to the maturational timing in this adolescent sample (Suarez et al., 2018). For example, in a previous study of the ELEMENT cohort, 2nd trimester blood lead was associated with delayed age at menarche, which may suggest slower physiological and biological aging for females compared to males (Jansen et al., 2018). Across human and animal studies alike, there have been significant sex-based difference in associations between prenatal and postnatal exposure and later cognition (Singh et al., 2018), neurobehavior (Halabicky et al., 2022), and growth and development (Zhou et al., 2020), where males appear to be more susceptible. Further investigation considering how biological age, and whether accelerated or decelerated biological age, changes over time would help to better understand sex-differentiated associations between lead and biological aging in adolescence and into adulthood.

Currently, validated epigenetic clocks have been trained on adult and pediatric samples; there are no clocks trained specifically on adolescent populations. The rapidness of physical and biological development in the adolescent period warrants an epigenetic clock specifically trained to this age period. While our results suggest that early life lead exposure is associated with altered adolescent biological age, our findings could be influenced by the inaccuracy of the clocks in this developmental period. Horvath, the Skin and Blood, and Hannum clocks were trained for ages from birth to centurion and are theoretically able to capture the ages of our sample. However, there are questions whether such pan-tissue clocks are able to identify tissue-specific aging, such as the cardiovascular or metabolic systems, which lead is known to target (Bell et al., 2019). GrimAge was designed to capture smoking related methylation changes to predict mortality (Lu et al., 2019a), which may not be a significant factor in our sample. Further, as PhenoAge was trained on a sample 21 years and up, and GrimAge on a sample with a mean age of 66 years, these clocks may be imprecise for the age of this sample. Wu and PedBE were trained on 0–18 and 0–20 years, respectively. PedBE, however, was trained on buccal samples, which is a more homogeneous sample and has a high degree of DNA stability, and may, therefore, be less reliable in blood. Recently, a novel epigenetic clock specifically trained for adolescence and young adulthood has been developed, which may be fruitful for future research (Aanes et al., 2023).

Accelerated or decelerated biological age in adolescence has been associated with adverse health outcomes. Adolescent biological age (i.e., EEAA and IEAA) has been associated with inflammation, increased BMI 5 years later, and probability of middle aged cardiovascular disease (Huang et al., 2019). At birth, biological age declaration via the Horvath clock has been associated with increased height and fat mass at age 17 years (Simpkin et al., 2017). Horvath biological age acceleration in adolescents has been associated with accelerated puberty via Tanner staging (Binder et al., 2018), higher salivary cortisol upon awakening, and greater odds for internalizing and thought problems on the Child Behavior Checklist (Suarez et al., 2018). Additionally, greater EEAA in adolescence has been linked to alterations to brain structures including volumes, cortisol thickness, and cortical surface area (Hoare et al., 2020). Overall, there is too little evidence to suggest what altered biological age, either accelerate or decelerated, in adolescence means for later health and developmental outcomes. Longitudinal studies are, therefore, needed to determine what effects alterations in biological aging during the critical adolescent period may mean for health later in life.

Prenatal lead exposure appeared to have more significant associations with adolescent biological age in both GLM and GEE models, which mirrors findings from other studies examining the influence of early life lead exposure on neurocognitive and cardiometabolic outcomes. For example, umbilical cord blood lead, a prenatal measure, has been inversely associated with 18 month fine motor scores (Lu et al., 2023). However, others have reported significant associations of childhood, and not prenatal, lead exposure on IQ outcomes in 12 year olds (Tatsuta et al., 2020). Considering cardiometabolic health, others have reported significant associations between both prenatal and postnatal lead exposure on repeated measures of metabolic syndrome at 6 and 8 years old, while we report only effects of prenatal exposure (Muciño-Sandoval et al., 2021). While our results add evidence to the literature base, further research is needed to elucidate relationships and determine if the associations between early life lead and adolescent biological age last into later adolescence and young adulthood, or if associations become more observable in later life. For example, one study has shown that originally non-significant cross-sectional associations between childhood adversity and biological age became later significant in early adulthood (i.e., epigenetic clocks at 23 years) (Copeland et al., 2022). Further longitudinal study with repeated measures of both lead exposure and biological aging will help to determine if there are critical periods for exposure which alter the epigenome and how these relationships change across the life course.

Our results suggest no significant associations between prenatal or childhood lead exposure and adolescent DNAmTL estimated from DNA methylation. Other examinations in newborns report late-pregnancy urinary lead (considered in a mixture) was negatively association with cord blood leukocyte telomere length (Cowell et al., 2020), while others report no association with maternal blood lead or postpartum bone lead (Herrera-Moreno et al., 2021). Concurrent blood lead levels in 8-year-olds have been negatively associated with relative telomere length (Pawlas et al., 2015). It is possible the significant association between

early life lead and telomere length attenuates as adolescents age and further study is needed to clarify these associations.

4.1. Strengths and limitations

To our knowledge, this is the first study to examine associations between prenatal and childhood lead exposure and adolescent biological aging. Among strengths of the study, we examined a number of epigenetic clocks, including adult and child specific clocks. We tested multiple timepoints of lead exposure and included multiple biomarkers (i.e., blood and bone measures) in the prenatal and early childhood periods while also controlling for concurrent lead exposure in our linear regression and GEE models. Still, there are limitations to be considered. First, it is unclear whether these epigenetic clocks are accurate for predicting biological aging in this adolescent population as most of the clocks were trained on either adult or younger child populations as discussed above. Further, considering our use of blood to estimate clocks, some clocks, such as PedBE which was originally trained on buccal cells and may not be as precise in blood samples. Second, while we included multiple measures of prenatal and childhood lead exposure measured in the blood, these measures were all biomarkers of acute lead exposure as compared to chronic or cumulative exposure. While we did include maternal bone lead measures in the postpartum period, a measure of maternal cumulative exposure, we were not able to include measures of cumulative exposure in the adolescents. Future research should consider other biomarkers of chronic lead exposure, such as bone or hair measurements, to determine if exposure at specific critical periods or overall cumulative exposure is associated with biological age. Third, single timepoints of biological aging are limited and future assessment of change in biological age over time would better elucidate these relationships. Finally, while we selected covariates that are routinely adjusted for in analysis of biological aging, such as maternal smoking, there may be other prenatal or childhood experiences, such as social stressors, that are impactful on biological aging that are not captured in our analysis.

5. Conclusion

We report that prenatal lead exposure, and few childhood lead exposure measures, was a significant predictor of few biological age measures captured via epigenetic clocks in this sample of adolescents. There were significant sex-stratification associations and lead x sex interactions, where males saw a positive association between prenatal and childhood lead exposure and accelerated biological aging for multiple epigenetic clocks compared to females, who saw decelerated biological aging. Our results suggest that exposure to lead during critical periods of development as well as cumulative lead exposure may alter the rate of biological aging. While evidence is still limited, future research could examine biological aging as a pathway through which lead exposure becomes biologically embedded and increases risk for adverse health outcomes into adulthood. Additional research with repeat epigenetic clock measurements would help capture change in biological aging and maturational tempo throughout the adolescent period in relation to early life lead exposure. Such study could also help tease apart sex-differentiated associations. In addition, future research can consider biological age as a mediator between lead exposure and known associations with neurocognitive and cardiometabolic health outcomes.

Funding

This work was supported by the U.S. Environmental Protection Agency (US EPA) RD83543601 (KEP), and from the National Institute for Environmental Health Sciences (NIEHS) P01 ES02284401 (KEP), R01ES007821 (KEP), R01 ES01493 (MMTR), R01 ES013744 (MMTR), 3R24ES028502 (KEP), NIH R35 RIVER award ES031686 (DCD), the Michigan Lifestage Environmental Exposures and Disease (M-LEEaD) NIEHS P30 Core Center ES017885 (DCD), and T32ES007062 (OMH).

CRediT authorship contribution statement

O.M. Halabicky: Conceptualization, Formal analysis, Methodology, Writing – original draft, Writing – review & editing. **M.M. Téllez-Rojo:** Formal analysis, Methodology, Writing – review & editing. **J.M. Goodrich:** Conceptualization, Writing – review & editing. **D.C. Dolinoy:** Conceptualization, Writing – review & editing. **A. Mercado-García:** Investigation, Writing – review & editing. **H. Hu:** Writing – review & editing. **K.E. Peterson:** Conceptualization, Formal analysis, Funding acquisition, Project administration, Supervision, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.scitotenv.2023.169757.

References

Aanes, H., Bleka, Ø., Dahlberg, P.S., Carm, K.T., Lehtimäki, T., Raitakari, O., Kähönen, M., Hurme, M., Rolseth, V., 2023. A new blood based epigenetic age predictor for adolescents and young adults. Sci. Rep. 13 (1), 2303. https://doi.org/10.1038/s41598-023-29381-7.

Arnold, O.M., Liu, J., 2020. Blood lead levels ≤10 micrograms/deciliter and executive functioning across childhood development: a systematic review. Neurotoxicol. Teratol. 80, 106888 https://doi.org/10.1016/j.ntt.2020.106888 (PubMed PMID: 32387536).

Aryee, M.J., Jaffe, A.E., Corrada-Bravo, H., Ladd-Acosta, C., Feinberg, A.P., Hansen, K.D., Irizarry, R.A., 2014. Minfi: a flexible and comprehensive Bioconductor package for the analysis of Infinium DNA methylation microarrays. Bioinformatics 30 (10), 1363–1369. Epub 20140128. https://doi.org/10.1093/bioinformatics/btu049. PubMed PMID: 24478339; PMCID: PMC4016708.

Bell, C.G., Lowe, R., Adams, P.D., Baccarelli, A.A., Beck, S., Bell, J.T., Christensen, B.C., Gladyshev, V.N., Heijmans, B.T., Horvath, S., Ideker, T., Issa, J.-P.J., Kelsey, K.T., Marioni, R.E., Reik, W., Relton, C.L., Schalkwyk, L.C., Teschendorff, A.E., Wagner, W., Zhang, K., Rakyan, V.K., 2019. DNA methylation aging clocks: challenges and recommendations. Genome Biol. 20(1):249 https://doi.org/10.1186/s13059-019-1824-y.

Binder, A.M., Corvalan, C., Mericq, V., Pereira, A., Santos, J.L., Horvath, S., Shepherd, J., Michels, K.B., 2018. Faster ticking rate of the epigenetic clock is associated with faster pubertal development in girls. Epigenetics 13 (1), 85–94. Epub 20180215. https://doi.org/10.1080/15592294.2017.1414127. PubMed PMID: 29235933; PMCID: PMC5836971.

Carter, A., Bares, C., Lin, L., Reed, B.G., Bowden, M., Zucker, R.A., Zhao, W., Smith, J.A., Becker, J.B., 2022. Sex-specific and generational effects of alcohol and tobacco use on epigenetic age acceleration in the Michigan longitudinal study. Drug and Alcohol Dependence Reports. 4, 100077 https://doi.org/10.1016/j.dadr.2022.100077.

Cecil KM, Brubaker CJ, Adler CM, Dietrich KN, Altaye M, Egelhoff JC, Wessel S, Elangovan I, Hornung R, Jarvis K, Lanphear BP. Decreased brain volume in adults with childhood lead exposure. PLoS Med.. 2008;5(5):e112. Epub 2008/05/30. doi: https://doi.org/10.1371/journal.pmed.0050112. PubMed PMID: 18507499; PMCID: PMC2689675.

Centers for Disease Control and Prevention, 2022. Sources of Lead Exposure. Available from: https://www.cdc.gov/nceh/lead/prevention/sources.htm.

Centers for Disease Control and Prevention Work Group on Lead and Pregnancy, 2010. Guidelines for the Identification and Management of Lead Exposure in Pregnant and Lactating Women. Centers for Disease Control, Atlanta.

Chen, B.H., Marioni, R.E., Colicino, E., Peters, M.J., Ward-Caviness, C.K., Tsai, P.C., Roetker, N.S., Just, A.C., Demerath, E.W., Guan, W., Bressler, J., Fornage, M., Studenski, S., Vandiver, A.R., Moore, A.Z., Tanaka, T., Kiel, D.P., Liang, L., Vokonas, P., Schwartz, J., Lunetta, K.L., Murabito, J.M., Bandinelli, S., Hernandez, D.G., Melzer, D., Nalls, M., Pilling, L.C., Price, T.R., Singleton, A.B., Gieger, C., Holle, R., Kretschmer, A., Kronenberg, F., Kunze, S., Linseisen, J., Meisinger, C., Rathmann, W., Waldenberger, M., Visscher, P.M., Shah, S., Wray, N. R., AF, McRae, Franco, O.H., Hofman, A., Uitterlinden, A.G., Absher, D., Assimes, T.,

Levine, M.E., Lu, A.T., Tsao, P.S., Hou, L., Manson, J.E., Carty, C.L., AZ, LaCroix, Reiner, A.P., Spector, T.D., Feinberg, A.P., Levy, D., Baccarelli, A., van Meurs, J., Bell, J.T., Peters, A., Deary, I.J., Pankow, J.S., Ferrucci, L., Horvath, S., 2016. DNA methylation-based measures of biological age: meta-analysis predicting time to death. Aging (Albany NY) 8 (9), 1844–1865. Epub 2016/10/01. 10.18632/aging.101020. PubMed PMID: 27690265; PMCID: PMC5076441.

Copeland, W.E., Shanahan, L., McGinnis, E.W., Aberg, K.A., van den Oord, E.J.C.G., 2022. Early adversities accelerate epigenetic aging into adulthood: a 10-year, within-subject analysis. J. Child Psychol. Psychiatry 63 (11), 1308–1315. https://doi.org/10.1111/jcpp.13575.

Cowell, W., Colicino, E., Tanner, E., Amarasiriwardena, C., Andra, S.S., Bollati, V., Kannan, S., Ganguri, H., Gennings, C., Wright, R.O., Wright, R.J., 2020. Prenatal toxic metal mixture exposure and newborn telomere length: modification by maternal antioxidant intake. Environ. Res. 190, 110009 https://doi.org/10.1016/j.envres.2020.110009.

Cradock, A.L., Barrett, J.L., Poole, M.K., Flax, C.N., Vollmer, L., Hecht, C., 2022. Lead concentrations in US School drinking water: testing programs, prevalence, and policy opportunities, 2016–2018. Am. J. Public Health 112 (S7), S679–s89. https://doi.org/10.2105/ajph.2022.306961. PubMed PMID: 36179297; PMCID: PMC9528654.

Crimmins, E.M., Thyagarajan, B., Levine, M.E., Weir, D.R., Faul, J., 2021. Associations of age, sex, race/ethnicity, and education with 13 epigenetic clocks in a nationally representative U.S. sample: the health and retirement study. J. Gerontol. 76 (6), 1117–1123. https://doi.org/10.1093/gerona/glab016.

Faulk, C., Liu, K., Barks, A., Goodrich, J.M., Dolinoy, D.C., 2014. Longitudinal epigenetic drift in mice perinatally exposed to lead. Epigenetics 9 (7), 934–941. https://doi.org/10.4161/epi.29024.

Fiorito, G., Polidoro, S., Dugué, P.-A., Kivimaki, M., Ponzi, E., Matullo, G., Guarrera, S., Assumma, M.B., Georgiadis, P., Kyrtopoulos, S.A., Krogh, V., Palli, D., Panico, S., Sacerdote, C., Tumino, R., Chadeau-Hyam, M., Stringhini, S., Severi, G., Hodge, A. M., Giles, G.G., Marioni, R., Karlsson Linnér, R., O'Halloran, A.M., Kenny, R.A., Layte, R., Baglietto, L., Robinson, O., McCrory, C., Milne, R.L., Vineis, P., 2017. Social adversity and epigenetic aging: a multi-cohort study on socioeconomic differences in peripheral blood DNA methylation. Sci. Rep. 7 (1), 16266. https://doi.org/10.1038/s41598-017-16391-5.

Fortin, J.-P., Triche Jr., T.J., Hansen, K.D., 2017. Preprocessing, normalization and integration of the Illumina HumanMethylationEPIC array with minfi. Bioinformatics 33 (4), 558–560. https://doi.org/10.1093/bioinformatics/btw691.

Goodrich, J.M., Reddy, P., Naidoo, R.N., Asharam, K., Batterman, S., Dolinoy, D.C., 2016a. Prenatal exposures and DNA methylation in newborns: a pilot study in Durban, South Africa. Environ Sci Process Impacts 18 (7), 908–917. https://doi.org/10.1039/c6em00074f. PubMed PMID: 27359112; PMCID: PMC4945397.

Goodrich, J.M., Dolinoy, D.C., Sánchez, B.N., Zhang, Z., Meeker, J.D., Mercado-Garcia, A., Solano-González, M., Hu, H., Téllez-Rojo, M.M., Peterson, K.E., 2016b. Adolescent epigenetic profiles and environmental exposures from early life through peri-adolescence. Environmental. Epigenetics 2(3):dvw018. https://doi.org/10.1093/eep/dvw018.

Guney, M., Zagury, G.J., 2014. Bioaccessibility of As, Cd, Cu, Ni, Pb, and Sb in toys and low-cost jewelry. Environ. Sci. Technol. 48 (2), 1238–1246. https://doi.org/10.1021/es4036122.

Halabicky, O.M., Ji, X., Gur, R.E., Gur, R.C., Yan, C., Chen, A., Liu, J., 2022. Childhood lead exposure and sex-based neurobehavioral functioning in adolescence. Neurotoxicology 93, 1–8. Epub 20220819. https://doi.org/10.1016/j.neuro.2022.08.008. PubMed PMID: 35988749.

Hannum, G., Guinney, J., Zhao, L., Zhang, L., Hughes, G., Sadda, S., Klotzle, B., Bibikova, M., Fan, J.B., Gao, Y., Deconde, R., Chen, M., Rajapakse, I., Friend, S., Ideker, T., Zhang, K., 2013. Genome-wide methylation profiles reveal quantitative views of human aging rates. Mol. Cell 49 (2), 359–367. Epub 20121121. https://doi.org/10.1016/j.molcel.2012.10.016. PubMed PMID: 23177740; PMCID: PMC3780611.

Hauptman, M., Bruccoleri, R., Woolf, A.D., 2017. An update on childhood Lead poisoning. Clin. Pediatr. Emerg. Med. 18 (3), 181–192. https://doi.org/10.1016/j.cpem.2017.07.010. PubMed PMID: 29056870; PMCID: PMC5645046.

Herrera-Moreno, J.F., Estrada-Gutierrez, G., Wu, H., Bloomquist, T.R., Rosa, M.J., Just, A.C., Lamadrid-Figueroa, H., Téllez-Rojo, M.M., Wrightb, R.O., Baccarelli, A.A., 2021. Prenatal lead exposure, telomere length in cord blood, and DNA methylation age in the PROGRESS prenatal cohort. Environ. Res. Epub 2021/12/19, 112577 https://doi.org/10.1016/j.envres.2021.112577 (PubMed PMID: 34921825).

Hillary, R.F., Stevenson, A.J., McCartney, D.L., Campbell, A., Walker, R.M., Howard, D. M., Ritchie, C.W., Horvath, S., Hayward, C., McIntosh, A.M., Porteous, D.J., Deary, I. J., Evans, K.L., Marioni, R.E., 2020. Epigenetic measures of ageing predict the prevalence and incidence of leading causes of death and disease burden. Clin. Epigenetics 12 (1), 115. https://doi.org/10.1186/s13148-020-00905-6.

Hoare, J., Stein, D.J., Heany, S.J., Fouche, J.-P., Phillips, N., Er, S., Myer, L., Zar, H.J., Horvath, S., Levine, A.J., 2020. Accelerated epigenetic aging in adolescents from low-income households is associated with altered development of brain structures. Metab. Brain Dis. 35 (8), 1287–1298. https://doi.org/10.1007/s11011-020-00589-0.

Horvath, S., 2013. DNA methylation age of human tissues and cell types. Genome Biol. 14 (10) doi:gb-2013-14-10-r115.

Horvath, S., Gurven, M., Levine, M.E., Trumble, B.C., Kaplan, H., Allayee, H., Ritz, B.R., Chen, B., Lu, A.T., Rickabaugh, T.M., Jamieson, B.D., Sun, D., Li, S., Chen, W., Quintana-Murci, L., Fagny, M., Kobor, M.S., Tsao, P.S., Reiner, A.P., Edlefsen, K.L., Absher, D., Assimes, T.L., 2016. An epigenetic clock analysis of race/ethnicity, sex, and coronary heart disease. Genome Biol. 17 (1), 171. https://doi.org/10.1186/s13059-016-1030-0.

Horvath, S., Oshima, J., Martin, G.M., Lu, A.T., Quach, A., Cohen, H., Felton, S., Matsuyama, M., Lowe, D., Kabacik, S., Wilson, J.G., Reiner, A.P., Maierhofer, A., Flunkert, J., Aviv, A., Hou, L., Baccarelli, A.A., Li, Y., Stewart, J.D., Whitsel, E.A., Ferrucci, L., Matsuyama, S., Raj, K., 2018. Epigenetic clock for skin and blood cells applied to Hutchinson Gilford Progeria Syndrome and ex vivo studies. Aging (Albany NY) 10 (7), 1758–1775. https://doi.org/10.18632/aging.101508. PubMed PMID: 30048243; PMCID: PMC6075434.

Hu, H., Pepper, L., Goldman, R., 1991. Effect of repeated occupational exposure to lead, cessation of exposure, and chelation on levels of lead in bone. Am. J. Ind. Med. 20 (6), 723–735. https://doi.org/10.1002/ajim.4700200603.

Huang, R.-C., Lillycrop, K.A., Beilin, L.J., Godfrey, K.M., Anderson, D., Mori, T.A., Rauschert, S., Craig, J.M., Oddy, W.H., Ayonrinde, O.T., Pennell, C.E., Holbrook, J. D., Melton, P.E., 2019. Epigenetic age acceleration in adolescence associates with BMI, inflammation, and risk score for middle age cardiovascular disease. J. Clin. Endocrinol. Metab. 104 (7), 3012–3024. https://doi.org/10.1210/jc.2018-02076.

Jansen, E.C., Zhou, L., Song, P.X.K., Sánchez, B.N., Mercado, A., Hu, H., Solano, M., Peterson, K.E., Tellez-Rojo, M.M., 2018. Prenatal lead exposure in relation to age at menarche: results from a longitudinal study in Mexico City. J. Dev. Orig. Health Dis. 9 (4), 467–472. Epub 2018/04/30. https://doi.org/10.1017/S2040174418000223.

Jansen, E.C., Dolinoy, D., Peterson, K.E., O'Brien, L.M., Chervin, R.D., Cantoral, A., Tellez-Rojo, M.M., Solano-Gonzalez, M., Goodrich, J., 2021. Adolescent sleep timing and dietary patterns in relation to DNA methylation of core circadian genes: a pilot study of Mexican youth. Epigenetics 16 (8), 894–907. Epub 20201004. https://doi.org/10.1080/15592294.2020.1827719. PubMed PMID: 33016191; PMCID: PMC8331002.

Javed, R., Chen, W., Lin, F., Liang, H., 2016. Infant's DNA methylation age at birth and epigenetic aging accelerators. Biomed. Res. Int. 2016, 4515928 https://doi.org/10.1155/2016/4515928.

Joubert, B.R., Felix, J.F., Yousefi, P., et al., 2016. DNA Methylation in newborns and maternal smoking in pregnancy: genome-wide consortium meta-analysis. Am. J. Hum. Genet. 98 (4), 680–696. https://doi.org/10.1016/j.ajhg.2016.02.019.

Joyce, B.T., Gao, T., Zheng, Y., Ma, J., Hwang, S.-J., Liu, L., Nannini, D., Horvath, S., Lu, A.T., Bai Allen, N., Jacobs, D.R., Gross, M., Krefman, A., Ning, H., Liu, K., Lewis, C.E., Schreiner, P.J., Sidney, S., Shikany, J.M., Levy, D., Greenland, P., Hou, L., Lloyd-Jones, D., 2021. Epigenetic age acceleration reflects long-term cardiovascular health. Circ. Res. 129 (8), 770–781. https://doi.org/10.1161/CIRCRESAHA.121.318965.

Kochmanski, J., Montrose, L., Goodrich, J.M., Dolinoy, D.C., 2017. Environmental deflection: the impact of toxicant exposures on the aging epigenome. Toxicol. Sci. 156 (2), 325–335. https://doi.org/10.1093/toxsci/kfx005.

Lawrence, K.G., Kresovich, J.K., O'Brien, K.M., Hoang, T.T., Xu, Z., Taylor, J.A., Sandler, D.P., 2020. Association of neighborhood deprivation with epigenetic aging using 4 clock metrics. JAMA Netw. Open 3(11):e2024329-e. https://doi.org/10.1001/jamanetworkopen.2020.24329.

Levine, M.E., Lu, A.T., Quach, A., Chen, B.H., Assimes, T.L., Bandinelli, S., Hou, L., Baccarelli, A.A., Stewart, J.D., Li, Y., Whitsel, E.A., Wilson, J.G., Reiner, A.P., Aviv, A., Lohman, K., Liu, Y., Ferrucci, L., Horvath, S., 2018. An epigenetic biomarker of aging for lifespan and healthspan. Aging (Albany NY) 10 (4), 573–591. https://doi.org/10.18632/aging.101414. PubMed PMID: 29676998; PMCID: PMC5940111.

McEwen, L.M., O'Donnell, K.J., McGill, M.G., Edgar, R.D., Jones, M.J., MacIsaac, J.L., Lin, D.T.S, Ramadori, K., Morin, A., Gladish, N., Garg, E., Unternaehrer, E., Pokhvisneva, I., Karnani, N., Kee, M.Z.L., Klengel, T., Adler, N.E., Barr, R.G., Letourneau, N., Giesbrecht, G.F., Reynolds, J.N., Czamara, D., Armstrong, J.M., Essex, M.J., de Weerth, C., Beijers, R., Tollenaar, M.S., Bradley, B., Jovanovic, T., Ressler, K.J., Steiner, M., Entringer, S., Wadhwa, P.D., Buss, C., Bush, N.R., Binder, E. B., Boyce, W.T., Meaney, M.J., Horvath, S., Kobor, M.S., 2020. The PedBE clock accurately estimates DNA methylation age in pediatric buccal cells. Proc. Natl. Acad. Sci. U. S. A. 117 (38), 23329–23335. https://doi.org/10.1073/pnas.1820843116. Epub 20191014. PubMed PMID: 31611402; PMCID: PMC7519312.

Lodge, E.K., Dhingra, R., Martin, C.L., Fry, R.C., White, A.J., Ward-Caviness, C.K., Wani, A.H., Uddin, M., Wildman, D.E., Galea, S., Aiello, A.E., 2022. Serum lead, mercury, manganese, and copper and DNA methylation age among adults in Detroit, Michigan. Environ. Epigenet. 8(1):dvac018. Epub 20220921 https://doi.org/10.1093/eep/dvac018 (PubMed PMID: 36330039; PMCID: PMC9620967).

Lu, A.T., Quach, A., Wilson, J.G., Reiner, A.P., Aviv, A., Raj, K., Hou, L., Baccarelli, A.A., Li, Y., Stewart, J.D., Whitsel, E.A., Assimes, T.L., Ferrucci, L., Horvath, S., 2019a. DNA methylation GrimAge strongly predicts lifespan and healthspan. Aging (Albany NY) 11 (2), 303–327. https://doi.org/10.18632/aging.101684. PubMed PMID: 30669119; PMCID: PMC6366976.

Lu, A.T., Seeboth, A., Tsai, P.-C., Sun, D., Quach, A., Reiner, A.P., Kooperberg, C., Ferrucci, L., Hou, L., Baccarelli, A.A., Li, Y., Harris, S.E., Corley, J., Taylor, A., Deary, I.J., Stewart, J.D., Whitsel, E.A., Assimes, T.L., Chen, W., Li, S., Mangino, M., Bell, J.T., Wilson, J.G., Aviv, A., Marioni, R.E., Raj, K., Horvath, S., 2019b. DNA methylation-based estimator of telomere length. Aging 11 (16), 5895–5923. https://doi.org/10.18632/aging.102173.

Lu, A.-x., Wang, S.-s., Xu, X., Wu, M.-q., Liu, J.-x., Xu, M., Cao, L.-L., Wang, J., Wu, W., Li, H., Yan, C.-h., 2023. Sex-specific associations between cord blood lead and neurodevelopment in early life: the mother-child cohort (Shanghai, China). Ecotoxicol. Environ. Saf. 249, 114337 https://doi.org/10.1016/j.ecoenv.2022.114337.

Marini, S., Davis, K.A., Soare, T.W., Zhu, Y., Suderman, M.J., Simpkin, A.J., Smith, A.D. A.C., Wolf, E.J., Relton, C.L., Dunn, E.C., 2020. Adversity exposure during sensitive periods predicts accelerated epigenetic aging in children. Psychoneuroendocrinology 113, 104484. https://doi.org/10.1016/j.psyneuen.2019.104484.

Marioni, R.E., Harris, S.E., Shah, S., McRae, A.F., von Zglinicki, T., Martin-Ruiz, C., Wray, N.R., Visscher, P.M., Deary, I.J., 2016. The epigenetic clock and telomere length are independently associated with chronological age and mortality. Int. J. Epidemiol. 45 (2), 424–432. https://doi.org/10.1093/ije/dyw041.

McKenna, B.G., Hendrix, C.L., Brennan, P.A., Smith, A.K., Stowe, Z.N., Newport, D.J., Knight, A.K., 2021. Maternal prenatal depression and epigenetic age deceleration: testing potentially confounding effects of prenatal stress and SSRI use. Epigenetics 16 (3), 327–337. Epub 20200724. https://doi.org/10.1080/15592294.2020.1795604. PubMed PMID: 32660321; PMCID: PMC7901550.

Moran, S., Arribas, C., Esteller, M., 2016. Validation of a DNA methylation microarray for 850,000 CpG sites of the human genome enriched in enhancer sequences. Epigenomics 8 (3), 389–399. Epub 2015/12/18. https://doi.org/10.2217/epi.15.114. PubMed PMID: 26673039; PMCID: PMC4864062.

Muciño-Sandoval, K., Ariza, A.C., Ortiz-Panozo, E., Pizano-Zárate, M.L., Mercado-García, A., Wright, R., Maria Téllez-Rojo, M., Sanders, A.P., Tamayo-Ortiz, M., 2021. Prenatal and early childhood exposure to Lead and repeated measures of metabolic syndrome risk indicators from childhood to preadolescence. Front. Pediatr. 9 (1130) https://doi.org/10.3389/fped.2021.750316.

Muller, C., Sampson, R.J., Winter, A.S., 2018. Environmental inequality: the social causes and consequences of lead exposure. Annu. Rev. Sociol. 44 (1), 263–282. https://doi.org/10.1146/annurev-soc-073117-041222.

Naicker, N., Norris, S., Mathee, A., Richter, L., 2011. Lead exposure and the onset of puberty: birth to twenty cohort. Epidemiology (Cambridge, Mass) 22 (1).

Navas-Acien A, Guallar E, Silbergeld EK, Rothenberg SJ. Lead exposure and cardiovascular disease – a systematic review. Environ. Health Perspect. 2007;115(3): 472–82. (PubMed PMID: 105983769. Language: English. Entry Date: 20080215. Revision Date: 20150711. Publication Type: Journal Article).

Needham BL, Fernandez JR, Lin J, Epel ES, Blackburn EH. Socioeconomic status and cell aging in children. Social science & medicine (1982). 2012;74(12):1948–51. Epub 2012/04/05. doi:https://doi.org/10.1016/j.socscimed.2012.02.019. PubMed PMID: 22472277.

Needham, B.L., Straight, B., Hilton, C.E., Olungah, C.O., Lin, J., 2021. Family socioeconomic status and child telomere length among the Samburu of Kenya. Soc. Sci. Med. 283, 114182 https://doi.org/10.1016/j.socscimed.2021.114182.

Nelson, C.A., Gabard-Durnam, L.J., 2020. Early adversity and critical periods: neurodevelopmental consequences of violating the expectable environment. Trends Neurosci. 43 (3), 133–143. https://doi.org/10.1016/j.tins.2020.01.002.

Oblak, L., van der Zaag, J., Higgins-Chen, A.T., Levine, M.E., Boks, M.P., 2021. A systematic review of biological, social and environmental factors associated with epigenetic clock acceleration. Ageing Res. Rev. 69, 101348 https://doi.org/10.1016/j.arr.2021.101348.

Pawlas, N., Płachetka, A., Kozłowska, A., Broberg, K., Kasperczyk, S., 2015. Telomere length in children environmentally exposed to low-to-moderate levels of lead. Toxicol. Appl. Pharmacol. 287 (2), 111–118. https://doi.org/10.1016/j.taap.2015.05.005.

Pelegí-Sisó, D., de Prado, P., Ronkainen, J., Bustamante, M., González, J.R., 2021. methylclock: a Bioconductor package to estimate DNA methylation age. Bioinformatics 37 (12), 1759–1760. https://doi.org/10.1093/bioinformatics/btaa825.

Perng, W., Tamayo-Ortiz, M., Tang, L., Sánchez, B.N., Cantoral, A., Meeker, J.D., Dolinoy, D.C., Roberts, E.F., Martinez-Mier, E.A., Lamadrid-Figueroa, H., PXK, Song, Ettinger, A.S., Wright, R., Arora, M., Schnaas, L., Watkins, D.J., Goodrich, J.M., Garcia, R.C., Solano-Gonzalez, M., Bautista-Arredondo, L.F., Mercado-Garcia, A., Hu, H., Hernandez-Avila, M., Tellez-Rojo, M.M., Peterson, K.E., 2019. Early Life Exposure in Mexico to ENvironmental Toxicants (ELEMENT) Project. BMJ Open 9 (8), e030427. Epub 2019/08/29. https://doi.org/10.1136/bmjopen-2019-030427. PubMed PMID: 31455712; PMCID: PMC6720157.

de Prado-Bert, P., Ruiz-Arenas, C., Vives-Usano, M., Andrusaityte, S., Cadiou, S., Carracedo, Á., Casas, M., Chatzi, L., Dadvand, P., González, J.R., Grazuleviciene, R., Gutzkow, K.B., Haug, L.S., Hernandez-Ferrer, C., Keun, H.C., Lepeule, J., Maitre, L., McEachan, R., Nieuwenhuijsen, M.J., Pelegrí, D., Robinson, O., Slama, R., Vafeiadi, M., Sunyer, J., Vrijheid, M., Bustamante, M., 2021. The early-life exposome and epigenetic age acceleration in children. Environ. Int. 155, 106683 https://doi.org/10.1016/j.envint.2021.106683.

Rauschert, S., Melton, P.E., Burdge, G., Craig, J.M., Godfrey, K.M., Holbrook, J.D., Lillycrop, K., Mori, T.A., Beilin, L.J., Oddy, W.H., Pennell, C., Huang, R.-C., 2019. Maternal smoking during pregnancy induces persistent epigenetic changes into adolescence, independent of postnatal smoke exposure and is associated with cardiometabolic risk. Front. Genet. 10.

Romieu, I., Palazuelos, E., Hernandez Avila, M., Rios, C., Muñoz, I., Jimenez, C., Cahero, G., 1994. Sources of lead exposure in Mexico City. Environ. Health Perspect. 102 (4), 384–389. https://doi.org/10.1289/ehp.94102384. PubMed PMID: 7523102; PMCID: PMC1566949.

Sánchez Brisa, N., Hu, H., Litman Heather, J., Téllez-Rojo, Martha M., 2011. Statistical methods to study timing of vulnerability with sparsely sampled data on environmental toxicants. Environ. Health Perspect. 119 (3), 409–415. https://doi.org/10.1289/ehp.1002453.

Sen, A., Heredia, N., Senut, M.C., Hess, M., Land, S., Qu, W., Hollacher, K., Dereski, M.O., Ruden, D.M., 2015. Early life lead exposure causes gender-specific changes in the DNA methylation profile of DNA extracted from dried blood spots. Epigenomics 7 (3), 379–393. https://doi.org/10.2217/epi.15.2. PubMed PMID: 26077427; PMCID: PMC4501025.

Shonkoff, J.P., Boyce, W.T., McEwen, B.S., 2009. Neuroscience, molecular biology, and the childhood roots of health disparities: building a new framework for health promotion and disease prevention. JAMA 301 (21), 2252–2259. https://doi.org/10.1001/jama.2009.754.

Simons, R.L., Lei, M.-K., Klopach, E., Berg, M., Zhang, Y., Beach, S.S.R., 2021. (Re)setting epigenetic clocks: an important avenue whereby social conditions become biologically embedded across the life course. J. Health Soc. Behav. 62 (3), 436–453. https://doi.org/10.1177/00221465211009309.

Simpkin, A.J., Howe, L.D., Tilling, K., Gaunt, T.R., Lyttleton, O., McArdle, W.L., Ring, S.M., Horvath, S., Smith, G.D., Relton, C.L., 2017. The epigenetic clock and physical development during childhood and adolescence: longitudinal analysis from a UK birth cohort. Int. J. Epidemiol. 46 (2), 549–558. https://doi.org/10.1093/ije/dyw307.

Singh, G., Singh, V., Sobolewski, M., Cory-Slechta, D.A., Schneider, J.S., 2018. Sex-dependent effects of developmental lead exposure on the brain. Front. Genet. 9 (MAR). https://doi.org/10.3389/fgene.2018.00089.

Smith, J.A., Raisky, J., Ratliff, S.M., Liu, J., Kardia, S.L.R., Turner, S.T., Mosley, T.H., Zhao, W., 2019. Intrinsic and extrinsic epigenetic age acceleration are associated with hypertensive target organ damage in older African Americans. BMC Med. Genet. 12 (1), 141. https://doi.org/10.1186/s12920-019-0585-5.

Stiles, K.M., Bellinger, D.C., 1993. Neuropsychological correlates of low-level lead exposure in school-age children: a prospective study. Neurotoxicol. Teratol. 15 (1), 27–35. https://doi.org/10.1016/0892-0362(93)90042-M.

Suarez, A., Lahti, J., Czamara, D., Lahti-Pulkkinen, M., Girchenko, P., Andersson, S., Strandberg, T.E., Reynolds, R.M., Kajantie, E., Binder, E.B., Raikkonen, K., 2018. The epigenetic clock and pubertal, neuroendocrine, psychiatric, and cognitive outcomes in adolescents. Clin. Epigenetics 10 (1), 96. https://doi.org/10.1186/s13148-018-0528-6.

Tatsuta, N., Nakai, K., Kasanuma, Y., Iwai-Shimada, M., Sakamoto, M., Murata, K., Satoh, H., 2020. Prenatal and postnatal lead exposures and intellectual development among 12-year-old Japanese children. Environ. Res. 189, 109844 https://doi.org/10.1016/j.envres.2020.109844.

Téllez-Rojo, M.M., Hernández-Avila, M., González-Cossío, T., Romieu, I., Aro, A., Palazuelos, E., Schwartz, J., Hu, H., 2002. Impact of breastfeeding on the mobilization of lead from bone. Am. J. Epidemiol. 155 (5), 420–428. https://doi.org/10.1093/aje/155.5.420.

Tellez-Rojo, M.M., Bautista-Arredondo, L.F., Tamayo-Ortiz, M., Cantoral, A., Estrada-Sanchez, D., Kraiem, R., Pantic, I., Romero, M., Fuller, R., Trejo-Valdivia, B., 2022. First National Report of Blood Lead Levels in Mexican Children: An Analysis to Inform Public Health Actions and Policy. International Society for Environmental Epidemiology, Athens, Greece.

UNICEF, 2020. Pure Earth. The Toxic Truth, Children's Exposure to Lead Pollution Undermines a Generation of Future Potential.

Wang, C., Koutrakis, P., Gao, X., Baccarelli, A., Schwartz, J., 2020. Associations of annual ambient PM2.5 components with DNAm PhenoAge acceleration in elderly men: the Normative Aging Study. Environ. Pollut. 258, 113690 https://doi.org/10.1016/j.envpol.2019.113690.

White, A.J., Kresovich, J.K., Keller, J.P., Xu, Z., Kaufman, J.D., Weinberg, C.R., Taylor, J.A., Sandler, D.P., 2019. Air pollution, particulate matter composition and methylation-based biologic age. Environ. Int. 132, 105071 https://doi.org/10.1016/j.envint.2019.105071.

Wu, X., Chen, W., Lin, F., Huang, Q., Zhong, J., Gao, H., Song, Y., Liang, H., 2019. DNA methylation profile is a quantitative measure of biological aging in children. Aging (Albany NY) 11 (22), 10031–10051. Epub 20191122. 10.18632/aging.102399. PubMed PMID: 31756171; PMCID: PMC6914436.

Zhou, C.-C., He, Y.-Q., Gao, Z.-Y., Wu, M.-Q., Yan, C.-H., 2020. Sex differences in the effects of lead exposure on growth and development in young children. Chemosphere 250, 126294. https://doi.org/10.1016/j.chemosphere.2020.126294.

Ziegler, E.E., Edwards, B.B., Jensen, R.L., Mahaffey, K.R., Fomon, S.J., 1978. Absorption and retention of lead by infants. Pediatr. Res. 12 (1), 29–34. https://doi.org/10.1203/00006450-197801000-00008 (PubMed PMID: 643372).