Accelerated epigenetic clock aging in peripheral blood is associated with preterm birth (PTB)

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Background

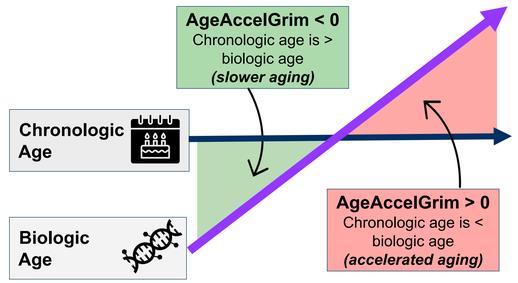
- Preterm birth (PTB) remains the largest cause of neonatal morbidity and mortality among non-anomalous babies delivered in the United States
- Despite the magnitude of this clinical problem, prediction of PTB remains imperfect, and the underlying pathophysiology is poorly understood
  - Some established PTB risk factors (e.g., racism, discrimination, history of childhood abuse) are associated with chronic stress and increased allostatic load, which may result in biologic changes including accelerated biologic and cellular aging
- Epigenetic clocks use CpG DNA methylation (DNAm) to estimate biologic age in relation to chronologic age
  - Biologic age acceleration is associated with cancer, heart disease, and a shortened lifespan
- AgeAccelGrim is a novel epigenetic clock that combines 7 DNAm components (Horvath, S. *Genome Biol* 2013)
  - AgeAccelGrim is a publicly available, online web tool.
  - Inputs include individual's CpG methylation data as measured on the EPIC 450K methylation platform and chronologic age. (https://horvath.genetics.ucla.edu/html/dnamage/)
  - AgeAccelGrim produces a single numeric outcome indicative of an individual's biologic age in relation to chronologic age (see top figure, center panel); negative numbers indicate slower aging while higher numbers indicate accelerated aging

Objective

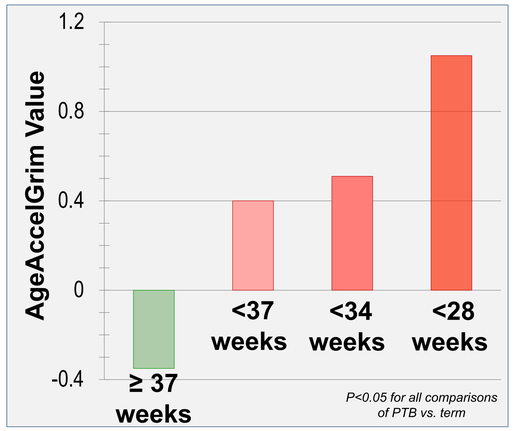
To determine if maternal biologic aging (measured by AgeAccelGrim) is associated with early PTB

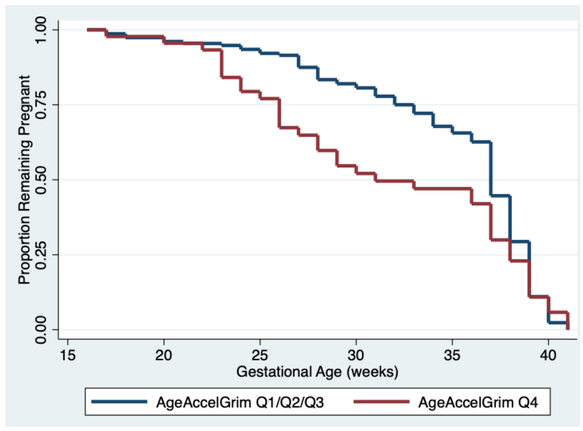
Methods

- Prospective cohort of pregnant individuals with singleton, non-anomalous gestations, at high risk for spontaneous PTB due to obstetric history ± diagnosis of a short cervix <25mm (current gestation)
- Peripheral blood samples were obtained <28 weeks' gestation
- Medical and obstetric history, antenatal course, and pregnancy outcomes abstracted from electronic medical record by trained research assistants
- Genome-wide CpG methylation was measured using the Illumina® EPIC 450K Array BeadChip.
- AgeAccelGrim and its 7 DNAm components were estimated by the Horvath DNAm age online tool. Participants were grouped into quartiles based on AgeAccelGrim score (lowest quartile = lowest scores / slowest aging; highest quartile = highest scores / fastest aging)
- **Primary outcome**: PTB < 34 weeks
- **Secondary outcomes**: PTB < 37 and < 28 weeks; individual components of the AgeAccelGrim
- Data were analyzed by chi, t-test, Kaplan-Meier survival curves, and logistic regression. Regression models controlled *a priori* for factors known to be associated with PTB and/or DNAm, including chronologic age, low socioeconomic status, gestational age at blood sample, and peripheral cell counts (as calculated by online Horvath tool)



Among individuals at high risk for preterm birth, those who delivered preterm were more likely to have an accelerated mid-pregnancy peripheral blood biologic age





Kaplan-Meier Survival curve, adjusted for maternal age, cell counts, low SES, and gestational age at sample. Patients with an AgeAccelGrim value in Q4, are compared to those with lower AgeAccelGrim values (Q1/Q2/Q3).

Results

- 163 participants were included. AgeAccelGrim correlated with chronologic age (Rho=0.84, p< 0.001). Baseline characteristics are shown below in Table 1:

Characteristic	PTB <34 weeks N=63	Delivery ≥ 34 weeks N=100	p-value
Maternal age, mean years ± SD	30.9 ± 6.3	31.9 ± 6.5	0.372
Black race	24 (38.1)	38 (38.0)	0.990
Hispanic ethnicity	7 (11.1)	23 (23.0)	0.056
Tobacco use disorder	11 (17.5)	10 (10.0)	
Low socioeconomic status*	16 (25.4)	38 (38.0)	0.096
Earliest prior delivery, mean weeks ± SD**	26.4 ± 7.5	26.3 ± 7.4	0.941
Gestational age at time of blood sample, mean weeks ± SD	20.2 ± 5.9	17.3 ± 6.0	0.003
Male fetus	35 (56.5)	53 (53.0)	0.668

*defined as public or no medical insurance ± less than high school education ± annual household income <$25,000.
**among 114 multiparous patients

- **Median acceleration was -0.35 for those delivering at term, and was inversely proportional to each PTB cutoff**, including <37 (+0.40; p=0.049), <34 (+0.51; p=0.036), and <28 (+1.05, p=0.001) weeks, figure left bottom in center panel
- DNAm of all 7 clock components = higher for those with PTB <34 wks, but most p=NS (Table):

Epigenetic clock component & brief description	PTB <34 weeks N=63	Delivery ≥ 34 weeks N=100	p-value
DNAmADM Estimated DNAm of adrenomedullin – a vasodilator that increases with HTN, heart failure	1.40 (-7.51, 10.6)	0.60 (-9.24, 7.03)	0.148
DNAmB2M Estimated DNAm of Beta-2 microglobulin, a biomarker associated with CVD, inflammation	12976 (-37903, 60496)	3074 (-46129, 46289)	0.377
DNAmCystatinC Estimated DNAm of Cystatin C, a biomarker of kidney function, CVD	968 (-8691, 13317)	-1128 (-11529, 9430)	0.170
DNAmGDF15 Estimated DNAm of growth differentiation factor 15, in TGF-beta sub-family; implicated in aging, mitochondrial dysfunction	15.9 (-41, 53)	-7.2 (-51, 48)	0.315
DNAmLeptin Estimated DNAm of leptin, a hormone with roles in appetite regulation	90 (-640, 648)	-110 (-1018, 813)	0.535
DNAmPAI1 Estimated DNAm of plasminogen activation inhibitor 1, a protein released in response to inflammation	**538 (-950, 1432)**	**-110 (-1486, 1069)**	**0.043**
DNAmTIMP1 Estimated DNAm of tissue inhibitor of metalloproteinases; TIMP1 naturally inhibits MMPs, and TIMP1 gene transcription is inducible in response to cytokines	97 (-325, 512)	-28 (-339, 279)	0.223

- **In regression models** adjusting for maternal age, cell counts, low SES, and gestational age at sample, AgeAccelGrim was not significantly associated with PTB <37 weeks. **However, patients with the highest quartile AgeAccelGrim values were at higher risk of PTB:**
  - < 34 weeks: aOR 2.4, 95% CI 1.1-5.1)
  - < 28 weeks: (aOR 3.9, 95% CI 1.6-9.4)
- In Kaplan-Meier survival analyses, those in the highest AgeAccelGrim quartile delivered earliest (log-rank p< 0.001), figure right bottom in center panel

Conclusions

Accelerated biologic aging is associated with PTB among high-risk patients. Elucidating factors that slow biologic aging may improve birth outcomes.