Accelerated epigenetic aging in newborns with Down syndrome

Keren Xu[1] Shaobo Li[1] Ivo S. Muskens[1] Natalina Elliott[2] Swe Swe Myint[1] Priyatama Pandey[1] Xiaomei Ma[3] Catherine Metayer[4] Beth A. Mueller[5] Kyle M. Walsh[6] Irene Roberts[2] Steve Horvath[7] Joseph L. Wiemels[1] Adam J. de Smith[1]

[1] *Center for Genetic Epidemiology, Department of Population and Public Health Sciences, Keck School of Medicine of the University of Southern California, Los Angeles, CA, USA*
[2] *Department of Paediatrics and MRC Molecular Haematology Unit, Weatherall Institute of Molecular Medicine, Oxford University and BRC Blood Theme, NIHR Oxford Biomedical Centre, Oxford, UK*
[3] *Department of Chronic Disease Epidemiology, Yale School of Public Health, New Haven, CT, USA*
[4] *School of Public Health, University of California, Berkeley, Berkeley, CA, USA*
[5] *Public Health Sciences Division, Fred Hutchinson Cancer Research Center, Seattle, WA, USA*
[6] *Department of Neurosurgery, Duke University, Durham, NC, USA*
[7] *Department of Biostatistics, Fielding School of Public Health, University of California, Los Angeles, Los Angeles, CA, USA*

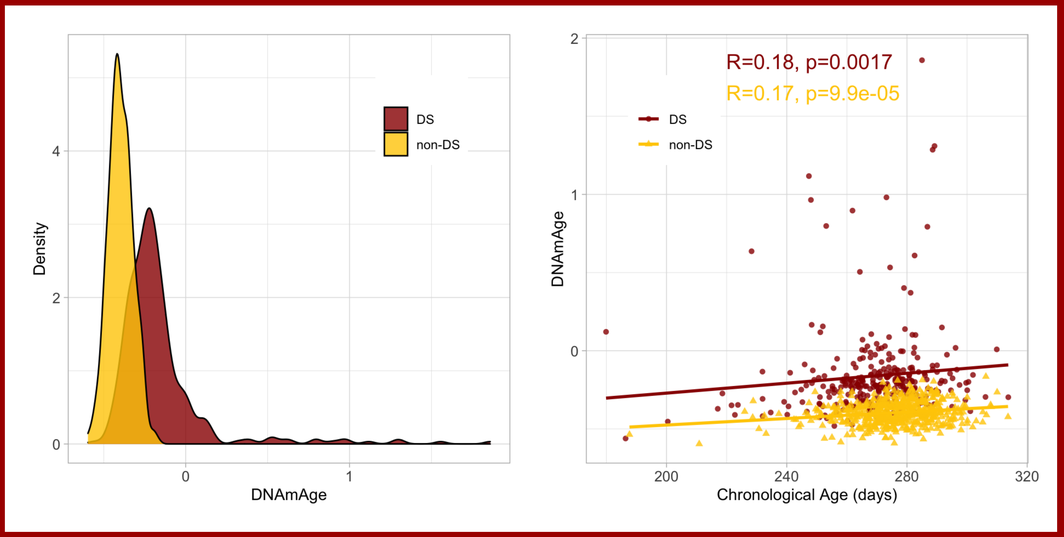
Introduction

Accelerated aging is a hallmark of Down syndrome (DS), with adults experiencing early-onset Alzheimer's disease and premature aging of skin, hair, and immune and endocrine systems. Accelerated epigenetic aging was found in blood and brain tissue of adults with DS,[1] but when this premature aging begins is unknown. We investigated whether accelerated aging in DS is already detectable in blood at birth.

Methods

- Dried bloodspots were obtained from 347 newborns with DS and 567 newborns without DS from California or Washington. DNA was isolated, bisulfite-converted, and assayed on Illumina MethylationEPIC DNA methylation (DNAm) arrays.[2]
- We calculated epigenetic age (DNAmAge) using a published epigenetic clock (391 CpGs)[3] and performed reference-based deconvolution of blood cell proportions using the "Identifying Optimal Libraries" algorithm.[4]
- *GATA1* was sequenced in a subset of 184 newborns with DS to identify somatic mutations associated with transient abnormal myelopoiesis.
- We compared DNAmAge between DS and non-DS newborns using linear regression adjusting for chronological age from conception (gestational age plus age at blood sampling), sex, batch, blood cell proportions, and genetic ancestry using EPISTRUCTURE.[5] Age acceleration was calculated as the deviation from expected DNAmAge based on its linear association with chronological age in non-DS newborns. We repeated analyses excluding 61 newborns (60 DS) exceeding mean+1SD for nucleated red blood cell (nRBC) proportions and 30 *GATA1*-positive DS newborns to address potential confounding. We tested for association between *GATA1* mutation variant allele frequency (VAF) and DNAmAge in DS newborns.

Down syndrome is associated with **increased DNAmAge** in **newborns**, with an age acceleration of 237 days.



Keck School of Medicine of USC
Department of Population and Public Health Sciences

Results

- Mean chronological age from conception was 269 days in DS and 276 in non-DS newborns. Chronological age was significantly positively correlated with DNAmAge in both DS and non-DS newborns (**Figure**).
- Blood cell proportions were significantly associated with DS status and DNAmAge, including strong correlation between nRBCs and DNAmAge (r=0.29, $p=5.8 * 10^{-19}$).
- Adjusting for cell proportions, DS was significantly associated with increased DNAmAge (beta=0.2419, $p=6.42 * 10^{-22}$), with an age acceleration of 237 days (**Table**). This association remained after excluding high nRBC newborns and *GATA1*-positive DS newborns (beta=0.1285, $p=3.18 * 10^{-11}$, age acceleration=127 days).
- Among newborns with DS, *GATA1* mutations were associated with increased DNAmAge ($p=6.65 * 10^{-12}$), with age acceleration of 115 days per 10% increase in VAF.

Table. Linear regression comparing DNAmAge between DS and non-DS newborns.

term	estimate (95% CI)	p	obs
Model 1			
DS	0.3502 (0.3126,0.3878)	8.3e-63	835
Chronological age	0.0003 (-0.0006,0.0011)	0.527	835
Age accel. for DS	355.23 days		
Model 2			
DS	0.2419 (0.1940,0.2898)	6.42e-22	835
Chronological age	0.0008 (0.0001,0.0015)	0.0224	835
Age accel. for DS	**236.89 days**		
Model 3			
DS	0.1285 (0.0911,0.1658)	3.18e-11	768
Chronological age	0.0005 (0.0001,0.0009)	0.0238	768
Age accel. for DS	**126.50 days**		

Note:

Model 1 adjusted for sex, 9 EPISTRUCTURE PCs, and batch; Model 2 additionally adjusted for blood cell proportions; Model 3 is model 2 excluding high nRBC subjects and subjects with *GATA1* mutation.

Conclusion

Our results support that accelerated aging in blood in DS begins prenatally, with implications for the pathophysiology of immunosenescence and other aging-related traits in DS.

References

1. Horvath S, Garagnani P, Bacalini MG, et al. Accelerated epigenetic aging in Down syndrome. *Aging Cell*. 2015;14(3):491–495.
2. Muskens IS, Li S, Jackson T, et al. The genome-wide impact of trisomy 21 on DNA methylation and its implications for hematopoiesis. *Nature Communications*. 2021;12(1, 1):821.
3. Horvath S, Oshima J, Martin GM, et al. Epigenetic clock for skin and blood cells applied to Hutchinson Gilford Progeria Syndrome and ex vivo studies. *Aging (Albany NY)*. 2018;10(7):1758–1775.
4. Koestler DC, Jones MJ, Usset J, et al. Improving cell mixture deconvolution by identifying optimal DNA methylation libraries (IDOL). *BMC Bioinformatics*. 2016;17(1):120.
5. Rahmani E, Shenhav L, Schweiger R, et al. Genome-wide methylation data mirror ancestry information. *Epigenetics & Chromatin*. 2017;10(1):1.