EPIGENETIC CHANGES ASSOCIATED WITH EARLY-LIFE AMBIENT AIR POLLUTION EXPOSURE IN THE CANDLE STUDY

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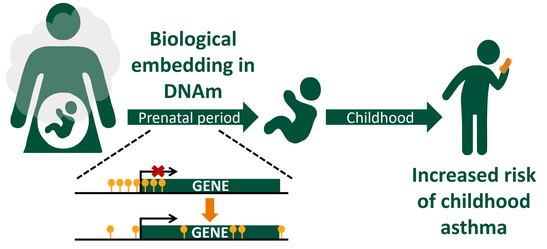
Background

Previous research shows prenatal air pollution exposure alters cord blood DNA methylation (DNAm), **however…**



previous findings are mainly **based on European cohorts** and have **limited applicability to other populations**. Additionally, **few studies link DNAm changes to health outcomes**.

Objective

Investigate DNAm changes associated with prenatal air pollution exposure in the racially **diverse CANDLE study**.

Study population

- 1503 families recruited from Shelby County, Tennessee between 2006-2011 as part of CANDLE study
- This study uses a subset of 515 CANDLE participants with air pollution and DNAm data, approved by UofM Bannatyne REB (HS23413)

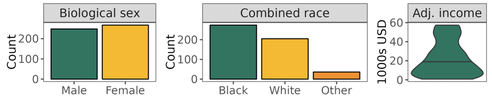


Figure 1. Study population (N=515) characteristics. The study population exhibits a similar distribution in child combined race to the CANDLE cohort and the Shelby country population. Participant combined race is determined from parental reported race.

Methods

Air pollution estimates

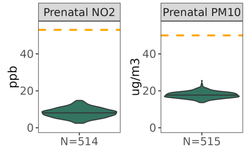


Figure 2. Annual individual address-level pregnancy period air pollution exposure. Estimated using advanced spatiotemporal model. Orange dashed lines represent annual air pollution exposure limits set by the U.S. EPA.

Cord blood DNAm

- Illumina EPIC array, preprocessing/normalization using *minfi* and *watermelon*
- Batch correction using *combat*, *minfi* used to estimate cell type proportions

Genetic ancestry

- Illumina Infinium Global Screening Array, QC and filtering performed in GenomeStudio, PLINK, and R
- Filtered SNPs used to define ancestry based on similarity coordinate values derived from multidimensional scaling

Health outcomes

- Asthma, wheeze, and atopy assessed at age 4 based on parental report and/or asthma medication use and/or physician diagnosis

A priori selected covariates

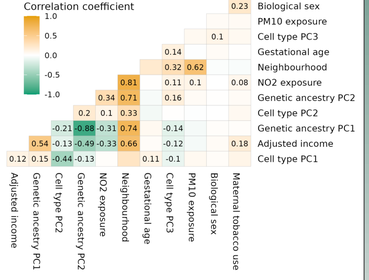


Figure 3. Study variables correlations. Only correlation coefficients of significant (p<0.05) associations are displayed. Neighborhood variable is specific to prenatal NO_2 models.

Statistical analysis

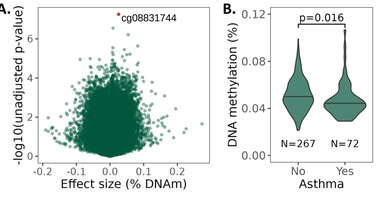
- Robust linear regressions to assess air pollution-DNAm relationship
- Differentially methylated regions identified using *comb-p*
- Gene set enrichment analysis performed using *missMethyl*
- Causal mediation analysis performed on DNAm sites/DMRs significantly associated with health outcome

Results

Prenatal NO_2 Exposure

Prenatal NO_2 is associated with higher DNAm at cg08831744, located in *IGFB3* promoter

Figure 4. (A) Prenatal NO_2 EWAS results (N=514). Red point = significant (FDR p<0.05) DNAm site. **(B) Association of cg08831744 DNAm with asthma.** Lower DNAm is significantly (two-sided t-test) associated with asthma (N=339), but not wheeze (N=420) or atopy (N=411; not shown). DNAm at cg08831744 (N=339) does not mediate the effects of prenatal NO_2 on asthma (p=0.15; not shown).



Differentially methylated regions (DMRs) associated with prenatal NO_2 occur in genes enriched for cell adhesion

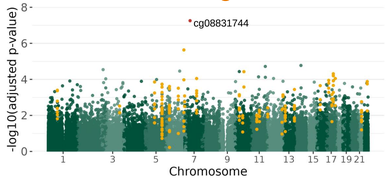


Figure 5. Significance and genomic location of DNAm sites from NO_2 EWAS. Red point = cg08831744 (not in a DMR). Yellow points = DNAm sites in DMRs (N=28).

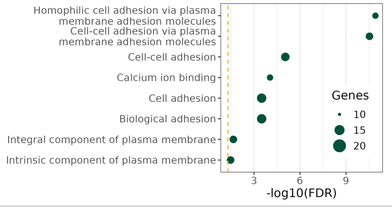
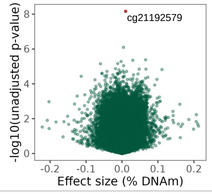


Figure 6. Significant (FDR p<0.05) gene ontology (GO) terms. Vertical yellow dashed line = 5% FDR. DMRs mapping to protocadherin gamma (*PCDHG*) subfamilies A and B were enriched in all GO terms.

Prenatal PM_{10} Exposure

Prenatal PM_{10} is associated with higher DNAm at cg21192579, located in *MYO7B* promoter

Figure 7. Prenatal PM_{10} EWAS results (N=515). Red point = significant (FDR p<0.05) DNAm site. Two-sided t-tests suggest cg08831744 DNAm is not (p>0.05) associated with asthma (N=339), wheeze (N=420), or atopy (N=411; not shown).



A differentially methylated region (DMR) in *XYLT1* mediates the effect of prenatal PM_{10} exposure on asthma, wheeze and atopy at age 4

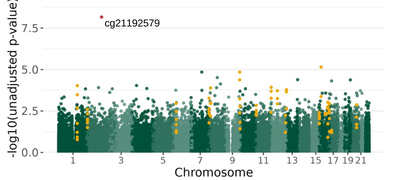


Figure 8. Significance and genomic location of DNAm sites from PM_{10} EWAS. Red point = cg21192579 (not in a DMR). Yellow points = DNAm sites in DMRs (N=19). Red dashed box = two DNAm sites in *XYLT1* DMR; both exhibit lower DNAm with PM_{10}.

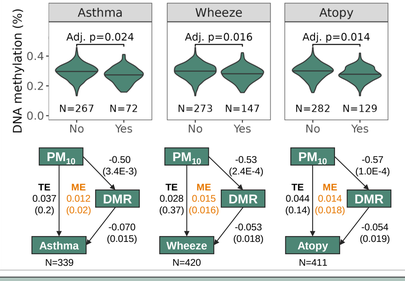


Figure 9. Associations between mean DNAm across *XYLT1* DMR and child health outcomes. Mean DNAm is significantly (two-sided t-tests, p<0.05) associated with asthma (N=339), wheeze (N=420), and atopy (N=411).

Figure 10. Mean DNAm at *XYLT1* DMR significantly (p<0.05) mediates the effects of prenatal PM_{10} on health outcomes. TE = total effect; ME=mediation effect. Effect estimates presented, p-value in brackets.

Conclusions

- Prenatal NO_2 and PM_{10} exposure alters cord blood DNAm in CANDLE participants
- These novel observations provide insight into robust DNAm changes that are shared across racially diverse populations
- These findings can be used to help develop methods to treat and prevent childhood asthma and atopic disease across populations exposed to high levels of air pollution

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Thanks

CIHR IRSC Canadian Institutes of Health Research / Instituts de recherche en santé du Canada
The VADA Program Visual and Automated Disease Analytics Graduate Training Program
Canadian Society of Allergy and Clinical Immunology / Société canadienne d'allergie et d'immunologie clinique
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