Asthma is associated with carotid arterial injury in children: The Childhood Origins of Asthma (COAST) Cohort

Matthew C. Tattersall, Michael D. Evans, Claudia E. Korcarz, Carol Mitchell, Elizabeth Anderson, Douglas F. DaSilva, Lisa P. Salazar, James E. Gern, Daniel J. Jackson, Robert F. Lemanske, Jr., James H. Stein

1 Department of Medicine, Division of Cardiovascular Medicine, University of Wisconsin School of Medicine and Public Health, Madison, Wisconsin, United States of America, 2 Department of Biostatistics and Medical Informatics, University of Wisconsin-Madison, Madison, Wisconsin, United States of America, 3 Department of Pediatrics, University of Wisconsin School of Medicine and Public Health, Madison, Wisconsin, United States of America

* mtattersall@medicine.wisc.edu

Abstract

Background
Asthma is associated with an increased cardiovascular disease (CVD) risk in adults, but the impact of asthma and atopic conditions on CVD risk in children is less well established. We hypothesized that children in the Childhood Origins of Asthma (COAST) Cohort with asthma and atopic conditions would have early carotid arterial injury.

Methods
The COAST study is a longitudinal birth cohort of children at increased risk of developing asthma. Children underwent ultrasonography measuring far wall right carotid bifurcation (RCB) and common carotid artery (RCCA) intima-media thickness (IMT; a measure of arterial injury). Multivariable linear regression models adjusted for age, gender, race, blood pressure, and body-mass index were used to assess associations of asthma and markers of arterial injury.

Results
The 89 participants were a mean (standard deviation) 15.3 (0.6) years old and 42% were female; 28 asthmatics had atopic disease, 34 asthmatics were without other atopic disease, and 15 non-asthmatics had atopic disease. This study population was compared to 12 controls (participants free of asthma or atopic disease). Compared to controls (589 μm), those with atopic disease (653 μm, p = 0.07), asthma (649 μm, p = 0.05), or both (677 μm, p = 0.005) had progressively higher RCB IMT values (P_trend = 0.011). In adjusted models, asthmatic and/or atopic participants had significantly higher RCB IMT than those without asthma or atopic disease (all p<0.03). Similar relationships were found for RCCA IMT.
Conclusion
Adolescents with asthma and other atopic diseases have an increased risk of subclinical arterial injury compared to children without asthma or other atopic disease.

Introduction
Asthma and cardiovascular disease (CVD) pose significant public health burdens. Asthma is a highly prevalent condition afflicting over 25 million individuals in the United States.[1–2] CVD is the leading cause of death for adults in the United States.[3–4] Inflammation and immune activation are central in the pathophysiology of both asthma and CVD.[5–8] Prior studies have demonstrated increased CVD risk among individuals with other chronic inflammatory diseases.[5,8–10].

We and others have demonstrated that in adults, asthma is associated with increased CVD risk, likely because of a common inflammatory pathophysiology.[5,11–15] Prior studies, however, were not performed in asthma-specific cohorts with well-characterized asthma subtypes; moreover, the adults that were studied had confounding CVD risk factors. To our knowledge, no prior study has investigated the associations among asthma, atopy, and subclinical CVD in a well-characterized asthma cohort of children.

The purpose of the present study was to investigate the associations among asthma, atopy and subclinical CVD in the Childhood Origins of Asthma (COAST) study, a birth cohort of children at increased risk for the development of asthma and atopic conditions. We hypothesized that the presence of asthma and atopic disease in children is associated with greater arterial injury manifested as a thicker carotid intima-media thickness.

Methods
Participants
The COAST study is an NHLBI funded observational birth cohort designed to investigate genetic and environmental factors contributing to the development of asthma in childhood.[16] To qualify for COAST enrollment, at least one parent was required to have respiratory allergies (defined as one or more positive aeroallergen skin tests) and/or a history of physician-diagnosed asthma.[16] A total of 289 newborns were enrolled from November of 1998 through May 2000; additional children with asthma meeting these same inclusion criteria were enrolled in the COAST study between 9 and 11 years of age. We recruited 90 participants that presented for an onsite annual COAST examination between August 2014 and August 2015; one had type 1 diabetes mellitus and was excluded from the analysis. Details on the design of COAST have been published previously.[16] Participant consent/assent was obtained at the time of their annual COAST examination. This study was approved by the University of Wisconsin Institutional Review Board. All COAST participants presenting for their annual onsite examination were eligible for the current study.

Carotid ultrasonography
B-mode ultrasound images of the far walls of the right common carotid artery (RCCA) and right carotid artery bifurcation (RCB) were obtained by registered sonographers (Siemens Acuson S2000, Malvern, PA, USA) with a 9L4 transducer. Because of our participants' young age, we needed to be sensitive to the duration of the ultrasound exam, so we imaged right
carotid artery only, as in several prior studies [17–19]. The RCCA was defined as the distal 10 mm of the carotid artery proximal to the RCB. The proximal 10 mm of the RCB were measured after being exported in DICOM format to a syngo Ultrasound Workplace reading station (Siemens Medical, Malvern, PA). Maximum segmental IMT measurements were obtained from end-diastolic still frames using a semi-automated border detected program and averaging tracings from 3 distinct cardiac cycles (Arterial Health Package, Siemens Medical, Malvern, PA) for IMT measurement [20–21]. The mean of the maximum measurements of the RCCA and the RCB were calculated [22–23]. Carotid IMT measurements are very reproducible in the UW Atherosclerosis Imaging Research Program (AIRP) lab, the intra-class correlation coefficients (ICC) for intra-reader reproducibility for mean CCA and bulb/ICA IMT are 0.98–0.99 in the UW AIRP. For scan-rescan variability, for 44 repeated studies, the Pearson correlation coefficient is 0.98 for maximum CCA IMT. Mean differences were 0.006 (0.034)–0.0007 (0.0.036) mm. There were no outliers noted on limit of agreement analyses for matched segments [24–25].

Asthma definition
In COAST, asthma assessments occurred at the end of the 6th, 8th, 11th, and 13th years of life. The presence of asthma at any of those exams qualified the participant as asthmatic for the study. A diagnosis of asthma in COAST has been previously reported and was based on the documented presence of one or more of the following characteristics in the previous year [26,27] (1) physician diagnosis of asthma, (2) use of physician-prescribed albuterol for coughing or wheezing episodes, (3) use of a daily controller medication, (4) step-up plan including use of albuterol or short-term use of inhaled corticosteroids during illness, or (5) use of prednisone for asthma exacerbation.

Atopic disease definition
Atopic disease was defined as the presence of physician-diagnosed atopic dermatitis, eczematous skin conditions or allergic rhinitis. Atopic dermatitis was defined as documentation by a health care provider in the participant’s medical record. There were two participants with physician documented “possible” atopic dermatitis. For proper adjudication, a review of 3 additional forms administered at ages 5–8 was performed and included the following: a study physician questionnaire, a parent-report questionnaire, and the scheduled physical exam, all of these three forms had to be consistent with atopic dermatitis for these participants to be classified as “atopic”. Allergic rhinitis was defined as documentation by a health care provider in the participant’s medical record.

Laboratory data
Peripheral blood samples were collected at the 13 year visit. The blood was collected in sterile heparinized tubes, kept at room temperature, and then processed the day of collection. Total IgE was measured by fluoroenzyme immunoassays using an automated instrument (Unicap 100; Pharmacia and Upjohn Diagnostics, Kalamazoo, Mich). The sensitivity for detection of total IgE was 2 kU/L [28–29]. Peripheral blood eosinophil numbers were measured by standard methods [29].

Pulmonary function testing
Spirometry was performed using the Jaeger MasterScope system (Jaeger-Toennies GmbH, Hoechberg, Germany) according to protocols described by the Childhood Asthma Research
and Education (CARE) Network [27,30]. The family was instructed to give the child their prescribed asthma medications but to hold albuterol and caffeinated food products for 6 hours prior to the annual visit. If the child was ill or taking albuterol for symptoms, the visit was rescheduled. Fractional exhaled nitric oxide (FeNO) was measured as reported previously[31] using the NIOX system (Aerocrine, Stockholm, Sweden) according to American Thoracic Society online measurement standards adapted for children [32]. The expiratory flow rate was 0.05 L/s. Exhalation times were at least 6 seconds with a 2-second analysis period. Children were required to have 3 measurements within 10% or 2 measurements within 5% for acceptability. Measurements were made before the performance of spirometry [31].

**Statistical methods**

Descriptive statistics are reported as means (standard deviations [SD]) for continuous and percentages for categorical variables. Analysis of variance (ANOVA) was used to compare baseline descriptive continuous variables among the groups; Fisher’s Exact tests were used for categorical variables. Before adjustment for confounders, the study was powered to detect a $50 \pm 60 \mu m$ difference in carotid IMT values with a $\beta = 0.9$ and $\alpha = 0.05$ with a total of 64 participants. A multivariate linear regression model was used to assess the association of carotid IMT and asthma/atopic status, adjusting for biologic confounders. A series of *a priori* models were created by adding potential known biological confounders into each model. Model 1: unadjusted, Model 2: adjusted for age, gender and race, Model 3: fully adjusted, Model 2 + systolic blood pressure and body-mass index. Sensitivity analyses were performed evaluating effect modification by Sex, IgE level and lung function markers. Statistical significance was set at a two-sided $p<0.05$. We report nominal $p$ values in the context of the study we performed for the group comparisons, all hypotheses tested are reported in the paper Analyses were performed in SAS (Version 9.2, Cary, NC: SAS Institute Inc.) and R (Version 3.3.0, Vienna, Austria: R Core Team (2016). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/).

**Results**

**Descriptive characteristics**

The 89 participants were mean (SD) 15.3 (0.6) years old, 58% were male, and 89.7% Caucasian. The 28 asthmatics with other atopic disease (As$^-$At$^+$), 34 asthmatics without other atopic disease (As$^+$At$^-$) and 15 non-asthmatics with atopic disease (As$^-$At$^+$) were compared to 12 participants free of asthma and atopic disease (As$^-$At$^-$). These groups were similar in age, gender, and blood pressure distributions (all $p>0.25$), but As$^-$At$^+$ participants tended to have higher body-mass index ($p = 0.06$), Table 1. By design, our study was enriched with asthmatic participants ($p = 0.0006$) Table 2.

**Carotid bulb IMT**

In unadjusted models, the As$^-$At$^+$ participants had the lowest RCB IMT values (589 $\pm$ 73 $\mu m$) compared to both asthmatic groups As$^+$At$^-$ (649 $\pm$ 78 $\mu m$, $p = 0.05$) and A$^+$A$^+$ (677 $\pm$ 107 $\mu m$, $p = 0.005$) (Table 3). In fully adjusted models, the As$^+$At$^-$ ($\beta = 77.4 \mu m$, 95% confidence intervals [CI], 10.3–144.5 $\mu m$, $p = 0.02$) and both asthmatic groups, As$^+$At$^-$ ($\beta = 64.2 \mu m$, 95% CI 6.0–122.3 $\mu m$, $p = 0.03$) and As$^+$A$^+$ ($\beta = 85.8 \mu m$, 95% CI, 24.2–147.4 $\mu m$, $p = 0.007$) had significantly thicker RCB compared to the As$^-$At$^+$ participants.
In unadjusted models the asthmatic groups (As\textsuperscript{+} At\textsuperscript{-} and As\textsuperscript{+} At\textsuperscript{+} participants) tended to have higher CCA IMT than As\textsuperscript{-} At\textsuperscript{-} participants ($\beta = 38.9 \mu m$, 95% CI -6.1–84.0 $\mu m$, $p = 0.09$ compared to As\textsuperscript{+} At\textsuperscript{-} and $\beta = 44.2 \mu m$, 95% CI -2.1–90.5 $\mu m$, $p = 0.06$ As\textsuperscript{+} At\textsuperscript{+}, respectively. In adjusted models, this trend persisted Table 3.

Sensitivity analyses

Because total IgE and lung function measures could plausibly confound or mediate an association between asthma and arterial injury\cite{33, 34}, we performed sensitivity analyses to assess the effects of both IgE and lung function in our adjusted models. We examined models that included % predicted FEV\textsubscript{1}, FEV\textsubscript{1}/FVC, total IgE and their interactions with asthma/atopic status for both RCCA and RCB. In these models, the size and significance of the group differences were very similar to those in the models without these terms and there was no evidence

### Table 1. Descriptive statistics of included COAST participants.

| Characteristic | Neither (A-A) (N = 12) | Atopic\textsuperscript{-} only (A-A\textsuperscript{-}) (N = 15) | Asthma only (A+A) (N = 34) | Atopy\textsuperscript{+} & Asthma (A+A\textsuperscript{+}) (N = 28) | p value |
|----------------|-------------------------|-------------------------|---------------------------|--------------------------------|--------|
| Age, years (SD)| 15.1 (0.3)              | 15.1 (0.4)              | 15.3 (0.7)                | 15.4 (0.6)                     | 0.26   |
| Body-mass index, kg/m\textsuperscript{2} (SD) | 20.3 (3.2)              | 22.2 (4.2)              | 23.7 (5.9)                | 24.6 (4.4)                     | 0.06   |
| Male gender, % (N) | 50% (6)                | 60% (9)                 | 53% (18)                  | 68% (19)                       | 0.61   |
| Race, % (N) | Caucasian | 92% (11)                | 100% (15)                | 91% (31)                       | 86% (24) | 0.26   |
| Asian | 8% (1) | 0% | 0% | 0% | 0% | 0.38   |
| Systolic Blood Pressure, mmHg (SD) | 114 (9) | 111 (9) | 113 (12) | 114 (8) | 0.70   |
| Diastolic Blood Pressure, mmHg (SD) | 64 (5) | 63 (4) | 65 (5) | 64 (4) | 0.68   |
| Asthma status at Exam 13 | None | 100% (12) | 100% (15) | 26% (9) | 14% (4) | N/A   |
| Intermittent | 0% | 0% | 18% (6) | 39% (11) |        |
| Mild-persistent | 0% | 0% | 47% (16) | 29% (8) |        |
| Moderate-persistent | 0% | 0% | 6% (2) | 18% (5) |        |
| Severe | 0% | 0% | 3% (1) | 0% |        |
| Medication Use at year 15 | Oral corticosteroid | 0% | 0% | 6% (2) | 11% (3) | 0.57   |
| Inhaled corticosteroid | 0% | 0% | 50% (17) | 61% (17) | <0.0001 |
| Leukotriene antagonist | 0% | 7% (1) | 24% (8) | 14% (4) | 0.21   |
| Albuterol | 17% (2) | 0% | 74% (25) | 86% (24) | <0.0001 |
| Long acting beta agonist | 0% | 0% | 32% (11) | 21% (6) | 0.01   |
| Total IgE, median [25\textsuperscript{th}, 75\textsuperscript{th}] | 45 [17,88] | 59 [20,132] | 83 [53,474] | 136 [51,320] | 0.08   |
| Blood eosinophil, median [25\textsuperscript{th}, 75\textsuperscript{th}] | 82 [61,128] | 198 [112,309] | 216 [92,450] | 176 [101,280] | 0.27   |
| Fractional exhaled nitric oxide, median [25\textsuperscript{th}, 75\textsuperscript{th}] | 13 [12,19] | 13 [9,18] | 20 [10,38] | 18 [11,34] | 0.33   |
| Right Carotid Artery Bifurcation $\mu$m (SD) | 589 (73) | 653 (86) | 649 (78) | 677 (107) | 0.04   |
| Right Common Carotid IMT $\mu$m (SD) | 558 (42) | 590 (76) | 597 (65) | 602 (74) | 0.26   |

Continuous measures are summarized as either mean (standard deviation, SD) or median [25th, 75th], and compared across groups using an analysis of variance. Total IgE, blood eosinophil counts, and exhaled nitric oxide were log-transformed for analysis. Categorical measures are summarized as rates (%) and compared across groups using Fisher’s exact test. Participants may select more than one race category so totals may sum to >100%.

\*Physician Diagnosed Atopic Disease (atopic dermatitis and/or allergic rhinitis)

https://doi.org/10.1371/journal.pone.0204708.t001

Common carotid artery IMT

In unadjusted models the asthmatic groups (As\textsuperscript{+} At\textsuperscript{-} and As\textsuperscript{+} At\textsuperscript{+} participants) tended to have higher CCA IMT than As\textsuperscript{-} At\textsuperscript{-} participants ($\beta = 38.9 \mu m$, 95% CI -6.1–84.0 $\mu m$, $p = 0.09$ compared to As\textsuperscript{+} At\textsuperscript{-} and $\beta = 44.2 \mu m$, 95% CI -2.1–90.5 $\mu m$, $p = 0.06$ As\textsuperscript{+} At\textsuperscript{+}, respectively. In adjusted models, this trend persisted Table 3.

Sensitivity analyses

Because total IgE and lung function measures could plausibly confound or mediate an association between asthma and arterial injury\cite{33, 34}, we performed sensitivity analyses to assess the effects of both IgE and lung function in our adjusted models. We examined models that included % predicted FEV\textsubscript{1}, FEV\textsubscript{1}/FVC, total IgE and their interactions with asthma/atopic status for both RCCA and RCB. In these models, the size and significance of the group differences were very similar to those in the models without these terms and there was no evidence
Table 2. Descriptive statistics of included COAST participants in this study compared to those not in this study.

| Characteristic                        | Included Participants N = 89 | Not Included Participants N = 147 | p value |
|--------------------------------------|------------------------------|-----------------------------------|---------|
| Age, years (SD)                      | 15.3 (0.6)                   | 15 (0)                            | N/A     |
| Body-mass index, kg/m² (SD)          | 23.3 (5.0)                   | 22.3 (4.9)                        | 0.17    |
| Male gender, % (N)                   | 58% (52)                     | 54% (79)                          | 0.50    |
| Race % (N)                           |                              |                                   |         |
| Caucasian                            | 96% (85)                     | 90% (132)                         | 0.21    |
| African-American                     | 7% (6)                       | 10% (15)                          | 0.48    |
| Asian                                | 2% (2)                       | 1% (2)                            | 0.64    |
| Asthma status at Exam                |                              |                                   |         |
| None                                 | 44% (40)                     | 67% (99)                          | 0.0006  |
| Intermittent                         | 20% (17)                     | 12% (17)                          | 0.13    |
| Mild-persistent                      | 28% (24)                     | 14% (20)                          | 0.02    |
| Moderate-persistent                  | 8% (7)                       | 6% (9)                            | 0.58    |
| Severe                               | 1% (1)                       | 1% (2)                            | 1       |
| Medication use                       |                              |                                   |         |
| Oral corticosteroid                  | 6% (5)                       | 3% (4)                            | 0.30    |
| Inhaled corticosteroid               | 38% (34)                     | 18% (26)                          | 0.0006  |
| Leukotriene antagonist               | 15% (13)                     | 4% (6)                            | 0.006   |
| Albuterol                            | 57% (51)                     | 33% (48)                          | 0.0002  |
| Long acting beta agonist             | 19% (17)                     | 6% (9)                            | 0.004   |
| Total IgE, median [25th, 75th]        | 88 [39, 320]                 | 92 [28, 325]                      | 0.60    |
| Blood eosinophil, median [25th, 75th]| 192 [93, 327]                | 141 [64, 300]                     | 0.37    |
| Fractional exhaled nitric oxide, median [25th, 75th]| 17 [11, 32]              | 14 [9, 22]                        | 0.08    |

Continuous measures are summarized as either mean (SD) or median [25th, 75th], and compared across groups using student’s t-test. Total IgE, blood eosinophil counts, and exhaled nitric oxide were log-transformed for analysis. Categorical measures are summarized as rates (%) and compared across groups using Fisher’s exact test. Participants may select more than one race category so totals may sum to >100%

https://doi.org/10.1371/journal.pone.0204708.t002

Table 3. Association of asthma & atopy with subclinical carotid injury.

| Asthma–& Other Atopic Disease'' + | Asthma + & Other Atopic Disease' - | Asthma + & Other Atopic Disease'' + |
|-----------------------------------|-----------------------------------|-----------------------------------|
| N = 15                            | N = 34                            | N = 28                            |
| β value (95% CI)                  | p value                           | β value (95% CI)                  | p value                           |
| Right Carotid Artery Bifurcation Intima-Media Thickness |
| Model 1                           | 64.5 (-5.1–134.1)                 | 0.07                              | 60.2 (0.6–119.8)                  | 0.05                                  |
| Model 2                           | 61.7 (-8.0–131.4)                 | 0.08                              | 56.2 (-3.8–116.1)                 | 0.07                                  |
| Model 3                           | 77.4 (10.3–144.5)                 | 0.02                              | 64.2 (6.0–122.3)                  | 0.03                                  |

Right Common Carotid Artery Intima-Media Thickness

| Model 1                           | 31.9 (-20.1–83.9)                 | 0.23                              | 38.9 (-6.1–84.0)                  | 0.09                                  |
| Model 2                           | 30.0 (-22.9–82.8)                 | 0.26                              | 37.9 (-8.0–83.7)                  | 0.10                                  |
| Model 3                           | 30.1 (-23.6–83.8)                 | 0.27                              | 34.5 (-12.7–81.7)                 | 0.15                                  |

No Asthma, Atopy (A A ) as reference group. Model 1: Unadjusted. Model 2: Adjusted for age, gender, race. Model 3: Fully adjusted, Model 2 + Systolic blood pressure and body-mass index. CI = confidence interval

''Physician-Diagnosed Atopic Disease

https://doi.org/10.1371/journal.pone.0204708.t003
of interactive effects with the 3 asthmatic/atopic groups compared to the non-asthmatic, non-atopic controls for RCCA or RCB IMT. Based on result from the Swiss Study on Air Pollution and Lung and Heart Disease in Adults (SAPALDIA)[35], we examined effect modification by sex, but did not find a sex by group interaction (p = 0.72).

Discussion

In a contemporary birth cohort, asthmatic children with or without other atopic disease had thicker RCB and RCCA walls compared to non-asthmatic/non-atopic children, indicating evidence of arterial injury, that is antecedent to the development of CVD. These relationships were identified among adolescents and persisted both after adjustment for several potential biological confounders and in the application of sensitivity analyses. To our knowledge, this is the first report demonstrating the presence of carotid arterial injury in a well-characterized cohort of asthmatic and atopic children. Arterial injury and subsequent atherosclerosis occur more rapidly in the carotid bulb than in the common carotid artery due to the anatomy of the bifurcation and flow disturbances resulting in regions of low and oscillatory shear stress [36–37]. Therefore, the early development of significantly thicker carotid bifurcation IMT in these asthmatic and atopic children identifies an early marker of subclinical arterial changes [36–37]. The differences noted in this study are clinically relevant, since CCA IMT differences of 30–50 μm are associated with approximately 4–6% increases in myocardial infarction and strokes over a decade in adults [38].

Asthma has been associated with increased CVD risk in adults, although the strength of this association has varied widely. Prior reports were from case-control studies and retrospective insurance claims analyses, had differing asthma definitions, or contained homogenous groups of individuals taking older asthma treatments with very few studies investigating subclinical CVD and asthma [13–15,33–34,39–42]. There are limited population level data that describe the association of arterial injury and asthma. Two previous studies have investigated this association in adults and young adults in CVD specific cohorts. In the Atherosclerosis Risk in Communities (ARIC) study, women with adult onset but not childhood onset asthma had increased carotid IMT[34]. Men with either childhood or adult onset asthma did not have increased carotid IMT. In the Atherosclerosis Risk Factors in Male Youngsters and Bruneck studies, allergic conditions such as allergic rhinitis and asthma were associated with thicker carotid or femoral IMT, or more carotid plaque progression in young male and middle-aged adults, respectively[33]. Two reports have examined the association of asthma and carotid IMT in asthmatic children. One demonstrated that children with mild asthma had thicker carotid IMT in a simple, unadjusted analysis; more recently, a report from the SAPALDIA study demonstrated increased carotid IMT in adolescent asthmatic boys but not asthmatic girls [35, 43]. There are less data on atopic conditions and CVD risk. Recently, cross-sectional data from the National Health and Nutrition Examination Survey demonstrated that adults and children with atopic dermatitis have increased risk factors for CVD [44–45]. In the Atherosclerosis Risk Factors in Male Youngsters and Bruneck studies, participants with allergic rhinitis were combined with asthmatics and this group had increased carotid IMT compared to controls [33].

Our study adds significant insight into the association of asthma and CVD. First, we describe the associations of asthma, atopy, and arterial injury in adolescents. Atopic conditions have been associated with CVD risk factors, but prior studies have not reported the effect of atopic conditions with and without asthma on carotid injury. Second, while our sample size is modest, our report is one of the largest studies to date to investigate arterial injury with asthma and atopy. The Atherosclerosis Risk Factors in Male Youngsters study excluded asthmatics
and female participants; the Bruneck study had 20 participants with asthma (2.4%); in SAPAL-DIA, there were 28 asthmatics (19 male, 9 female) [33,35]. Adolescents with and without asthma are ideally suited for studying early arterial injury as their physiology is less confounded by the comorbidities of aging which cause arterial injury and increase CVD risk. Indeed, prior studies included participants with a high prevalence of traditional CVD risk factors. In the COAST study, participants were healthy children free from diabetes mellitus and hypertension and the systolic blood pressures between the non-atopic, non-asthmatic group did not differ from the asthmatic and atopic groups (p = 0.26). Third, this is the first study to investigate the association of asthma, atopy, and carotid IMT in an asthma-specific cohort, with rigorous definitions of the exposure variables. Similar to two previous but limited reports, we found increased arterial injury among early onset asthmatics. Prior studies in adults demonstrated increased arterial injury and CVD events with late-onset asthma; however, few studies have examined the CVD association with early onset asthma and atopic conditions [12,34,41,42]. Our study supports previous reports of arterial injury in early onset asthmatics and suggests that both the early and late onset asthma phenotypes may increase CVD risk.

Early arterial injury occurs in children with other chronic inflammatory diseases such as systemic lupus erythematosus, juvenile idiopathic arthritis and human immunodeficiency virus and predicts higher CVD risks throughout life [46–48]. Asthma is a heterogeneous inflammatory syndrome with different phenotypes each with unique pathophysiology, and the previously reported inconsistent effect sizes and directions for the association of asthma and CVD in prior investigations may be a reflection of this [49]. Atopic dermatitis and allergic rhinitis are also chronic inflammatory disorders that commonly co-exist with childhood asthma [50].

The mechanism(s) by which inflammation in asthma and atopic disease can result in arterial injury and increased CVD risk have not been clarified. Childhood asthma is often associated with a disturbed balance in T-helper cell polarization. T-helper cell polarization occurs primarily in a Type 1 (T\(_1\)) or Type 2 (T\(_2\)) inflammatory profile and childhood asthma and atopy demonstrate a skewed T\(_2\) pattern of inflammation. T\(_2\) high asthma is accompanied by elevated levels of IL-4, a pro-atherogenic cytokine that activates the vascular endothelium leading to recruitment and activation of mononuclear cells, increased cellular adhesion molecule expression, and stimulation of 15-lipo-oxygenase production, which can oxidize low-density lipoproteins [51–52]. In addition, mast cells play a central role in asthma and atopic conditions; these mast cells produce the cysteinyl leukotrienes (C\(_4\), D\(_4\) and E\(_4\)) and induce acute and chronic inflammation [53]. Animal studies have demonstrated that atherosclerotic plaques contain elevated levels of leukotrienes and that blockade of leukotriene B\(_4\) reduces monocyte recruitment and atheroma progression [54–55]. Thus, there may be several mechanisms by which the inflammation of early onset asthma and atopy result in arterial injury and increased CVD risk.

**Limitations**

Participants from the COAST cohort without asthma or atopic disease served as controls; however, these individuals may be prone to a misclassification bias. To be enrolled in the COAST cohort, children were determined to be at high risk for developing asthma or atopic diseases and therefore some of these children may go on to develop asthma/atopy in the future as they age and/or harbor a subclinical physiology similar to the asthmatic/atopic participants. Nevertheless, this misclassification bias would have influenced our results toward the null. At the time of this study, there were two COAST participants in the control groups that were not diagnosed with asthma at the age 13 year old asthma assessment, but had used albuterol in the
past year. These individuals may have been misclassified as not asthmatic, but again, this would bias the results toward the null. This study is observational and the associations we identified do not imply causation. This study was performed in children who were free of many of the CVD confounders that accrue with age. We did not have lipid parameters available for adjustment, however in our full models we adjusted for body mass index which has found to be a sensitive predictor of dyslipidemia in children [56]. Although we adjusted for known biologic confounders, there may have been unmeasured confounders that could have resulted in residual confounding. Although our sensitivity analyses may have been somewhat underpowered, we were able to identify a strong, consistent effect using a modest sample size that was robust to covariate adjustment and that corroborate results found in prior investigations therefore reinforcing the validity of our results.

**Conclusions**

Even at a young age, children with asthma and other atopic disease have increased risk of subclinical arterial injury compared to children without asthma or atopy. Given the high prevalence of childhood asthma and atopic conditions, further investigations to confirm this association and elucidate underlying mechanisms are needed.

**Disclosures**

Dr. Stein: Inventor, Ultrasonic Apparatus and Method for Providing Quantitative Indication of Risk of Coronary Heart Disease, Patent # US 6,730,023 B2. Assignee: Wisconsin Alumni Research Foundation, Madison, WI. Filed November 7, 2002. Awarded May 4, 2004. It is used to estimate carotid arterial age based on carotid wall thickness measures. Not used in this study.

**Supporting information**

S1 File.
(XLSX)

**Author Contributions**

**Conceptualization:** Matthew C. Tattersall, Claudia E. Korcarz, Douglas F. DaSilva, James H. Stein.

**Data curation:** Michael D. Evans, Claudia E. Korcarz, Carol Mitchell, Elizabeth Anderson, Lisa P. Salazar, James E. Gern, Daniel J. Jackson, Robert F. Lemanske, Jr., James H. Stein.

**Formal analysis:** Michael D. Evans.

**Writing – original draft:** Matthew C. Tattersall, James E. Gern, Daniel J. Jackson, Robert F. Lemanske, Jr., James H. Stein.

**Writing – review & editing:** Elizabeth Anderson, James E. Gern, Daniel J. Jackson, Robert F. Lemanske, Jr.

**References**

1. Akinbami LJ, Moorman JE, Bailey C, Zahran HS, King M, Johnson CA, et al. Trends in asthma prevalence, health care use, and mortality in the United States, 2001–2010. NCHS Data Brief. 2012;(94):1–8. Epub 2012/05/24. PMID: 22617340.

2. Wisconsin Department of Health Services DoPH, Bureau of Environmental and Occupational Health. The Burden of Asthma in Wisconsin 2013. 2013.
3. Yuan H, White RC, Pettito F. The Burden of Heart Disease and Stroke in Wisconsin 2010. Wisconsin Heart Disease and Stroke Prevention Program, Wisconsin Department of Health Services.2010.

4. Go AS, Mozaffarian D, Roger VL, Benjamin EJ, Berry JD, Blaha MJ, et al. Heart disease and stroke statistics—2014 update: a report from the American Heart Association. Circulation. 2014; 129(3):e28–e292. Epub 2013/12/20. https://doi.org/10.1161/01.cir.0000441139.02102.80 PMID: 24352519.

5. Hansson GK. Inflammation, atherosclerosis, and coronary artery disease. N Engl J Med. 2005; 352(16):1685–95. Epub 2005/04/22. https://doi.org/10.1056/NEJMoa043430 PMID: 15843671.

6. Ridker PM, Hennekens CH, Buring JE, Rifai N. C-reactive protein and other markers of inflammation in the prediction of cardiovascular disease in women. N Engl J Med. 2000; 342(12):836–43. Epub 2000/03/25. https://doi.org/10.1056/NEJM200003233421202 PMID: 10733371.

7. Ridker PM, Cushman M, Stampfer MJ, Tracy RP, Hennekens CH. Inflammation, aspirin, and the risk of cardiovascular disease in apparently healthy men. N Engl J Med. 1997; 336(14):973–9. Epub 1997/04/03. https://doi.org/10.1056/NEJM199704033361401 PMID: 9077376.

8. Ridker PM, Danielson E, Fonseca FA, Genest J, Gotto AM Jr., Kastelein JJ, et al. Rosuvastatin to prevent vascular events in men and women with elevated C-reactive protein. N Engl J Med. 2008; 359(21):2195–207. Epub 2008/11/11. https://doi.org/10.1056/NEJMoa0807646 PMID: 18997196.

9. Solomon DH, Karlson EW, Rimm EB, Cannuscio CC, Mandl LA, Manson JE. Cardiovascular morbidity and mortality in women diagnosed with rheumatoid arthritis. Circulation. 2003; 107(9):1303–7. Epub 2003/03/12. PMID: 12628952.

10. Stein JH, Hsue PY. Inflammation and Arterial Injury in Individuals With Human Immunodeficiency Virus Infection. JAMA Cardiol. 2016; 1(4):481–2. Epub 2016/07/22. https://doi.org/10.1001/jamacardio.2016.1169 PMID: 27438326.

11. Tattersall MC, Guo M, Korcarz CE, Gepner AD, Kaufman JD, Liu KJ, et al. Asthma predicts cardiovascular disease events: the multi-ethnic study of atherosclerosis. Arterioscler Thromb Vasc Biol. 2015; 35(6):1520–5. Epub 2015/04/25. https://doi.org/10.1161/ATVBAHA.115.305452 PMID: 25908767.

12. Tattersall MC, Barnett JH, Korcarz CE, Hagen EW, Peppard PE, Stein JH. Late-Onset Asthma Predicts Cardiovascular Disease Events: The Wisconsin Sleep Cohort. J Am Heart Assoc. 2016; 5(9). Epub 2016/08/26. https://doi.org/10.1161/jaha.116.003448 PMID: 27559071.

13. Iribarren C, Tolstykh IV, Miller MK, Sobel E, Eisner MD. Adult asthma and risk of coronary heart disease, cerebrovascular disease, and heart failure: a prospective study of 2 matched cohorts. Am J Epidemiol. 2012; 176(11):1014–24. Epub 2012/11/10. https://doi.org/10.1093/aje/kws181 PMID: 23139248.

14. Iribarren C, Tolstykh IV, Eisner MD. Are patients with asthma at increased risk of coronary heart disease? Int J Epidemiol. 2004; 33(4):743–8. Epub 2004/05/08. https://doi.org/10.1093/ije/dyh081 PMID: 15131088.

15. Schanen JG, Iribarren C, Shahar E, Punjabi NM, Rich SS, Soricie PD, et al. Asthma and incident cardiovascular disease: the Atherosclerosis Risk in Communities Study. Thorax. 2005; 60(8):633–8. Epub 2005/08/03. https://doi.org/10.1136/thx.2004.026484 PMID: 16061703.

16. Lemanske RF Jr. The childhood origins of asthma (COAST) study. Pediatr Allergy Immunol. 2002; 13 Suppl 15:38–43. Epub 2002/06/21. https://doi.org/10.1034/j.1411-9768.2002.00206.x PMID: 12068583.

17. Currier JS, Kendall MA, Zackerin R, Henry WK, Alston-Smith B, Torriani FJ, et al. Carotid artery intima-media thickness and HIV infection: traditional risk factors overshadow impact of protease inhibitor exposure. AIDS. 2005; 19(9):927–33. PMID: 15905673.

18. Schanen JG, Iribarren C, Shahar E, Punjabi NM, Rich SS, Soricie PD, et al. Asthma and incident cardiovascular disease: the Atherosclerosis Risk in Communities Study. Thorax. 2005; 60(8):633–8. Epub 2005/08/03. https://doi.org/10.1136/thx.2004.026484 PMID: 16061703.

19. Lemanske RF Jr. The childhood origins of asthma (COAST) study. Pediatr Allergy Immunol. 2002; 13 Suppl 15:38–43. Epub 2002/06/21. https://doi.org/10.1034/j.1411-9768.2002.00206.x PMID: 12068583.

20. Currier JS, Kendall MA, Zackerin R, Henry WK, Alston-Smith B, Torriani FJ, et al. Carotid artery intima-media thickness and HIV infection: traditional risk factors overshadow impact of protease inhibitor exposure. AIDS. 2005; 19(9):927–33. PMID: 15905673.

21. Stein JH, Korcarz CE, Hurst RT, Lonn E, Kendall CB, Mohler ER, et al. Use of carotid ultrasound to identify subclinical vascular disease and evaluate cardiovascular disease risk: a consensus statement from the American Society of Echocardiography Carotid Intima-Media Thickness Task Force. Endorsed by the Society for Vascular Medicine. J Am Soc Echocardiogr. 2008; 21(2):93–111; quiz 89–90. Epub 2008/02/12. https://doi.org/10.1016/j.echo.2007.11.011 PMID: 18261694.
22. Urbina EM, Williams RV, Alpert BS, Collins RT, Daniels SR, Hayman L, et al. Noninvasive assessment of subclinical atherosclerosis in children and adolescents: recommendations for standard assessment for clinical research: a scientific statement from the American Heart Association. Hypertension. 2009; 54(5):919–50. Epub 2009/09/05. https://doi.org/10.1161/HYPERTENSIONAHA.109.192639 PMID: 19729599.

23. Bhuiyan AR, Srinivasan SR, Chen W, Paul TK, Berenson GS. Correlates of vascular structure and function measures in asymptomatic young adults: the Bogalusa Heart Study. Atherosclerosis. 2006; 189(1):1–7. Epub 2006/03/30. https://doi.org/10.1016/j.atherosclerosis.2006.02.011 PMID: 16569409.

24. Tattersall MC, Gassett A, Korcarz CE, Gepner AD, Kaufman JD, Liu KJ, et al. Predictors of carotid thickness and plaque progression during a decade: the Multi-Ethnic Study of Atherosclerosis. Stroke. 2014; 45(11):3257–62. Epub 2014/09/13. https://doi.org/10.1161/STROKEAHA.114.005669 PMID: 25213342.

25. Johnson HM, Piper ME, Jorenby DE, Fiore MC, Baker TB, Stein JH. Risk factors for subclinical carotid atherosclerosis among current smokers. Prev Cardiol. 2010; 13(4):166–71. Epub 2010/09/24. https://doi.org/10.1111/j.1751-7141.2010.00068.x PMID: 20860659.

26. Jackson DJ, Gangnon RE, Evans MD, Roberg KA, Anderson EL, Pappas TE, et al. Wheezing rhinovirus illnesses in early life predict asthma development in high-risk children. Am J Respir Crit Care Med. 2008; 178(7):667–72. Epub 2008/06/21. https://doi.org/10.1164/rccm.200802-309OC PMID: 18565953.

27. Guilbert TW, Singh AM, Danov Z, Evans MD, Jackson DJ, Burton R, et al. Decreased lung function after preschool wheezing rhinovirus illnesses in children at risk to develop asthma. J Allergy Clin Immunol. 2011; 128(3):532–8.e1–10. Epub 2011/09/01. https://doi.org/10.1016/j.jaci.2011.06.037 PMID: 21878241.

28. Neaville WA, Tisler C, Bhattacharya A, Ankiam K, Gilbertson-White S, Hamilton R, et al. Developmental cytokine response profiles and the clinical and immunologic expression of atopy during the first year of life. J Allergy Clin Immunol. 2003; 112(4):740–6. Epub 2003/10/18. https://doi.org/10.1016/S0091-6749(03)00712-X PMID: 14564354.

29. Anderson HM, Lemanske RF Jr., Arron JR, Holweg CTJ, Rajamanickam V, Gangnon RE, et al. Relationships among aeroallergen sensitization, peripheral blood eosinophils, and peristin in pediatric asthma development. J Allergy Clin Immunol. 2017; 139(3):790–6. Epub 2016/08/04. https://doi.org/10.1016/j.jaci.2016.05.033 PMID: 27484037.

30. Strunk RC, Szeffer SJ, Phillips BR, Zeiger RS, Chinchilli VM, Larsen G, et al. Relationship of exhaled nitric oxide to clinical and inflammatory markers of persistent asthma in children. J Allergy Clin Immunol. 2003; 112(5):883–92. Epub 2003/11/12. https://doi.org/10.1067/0091 jaci.2003.08.014 PMID: 14610474.

31. Jackson DJ, Virmig CM, Gangnon RE, Evans MD, Roberg KA, Anderson EL, et al. Fractional exhaled nitric oxide measurements are most closely associated with allergic sensitization in school-age children. J Allergy Clin Immunol. 2009; 124(5):949–53. Epub 2009/09/15. https://doi.org/10.1016/j.jaci.2009.07.024 PMID: 19748661.

32. Baraldi E, de Jongste JC. Measurement of exhaled nitric oxide in children, 2001. Eur Respir J. 2002; 20(1):223–37. Epub 2002/08/09. PMID: 12166573.

33. Knoiflach M, Kiechl S, Mayr A, Willeit J, Poewe W, Wick G. Allergic rhinitis, asthma, and atherosclerosis in the Bruneck and ARMY studies. Arch Intern Med. 2005; 165(21):2521–6. Epub 2005/11/30. https://doi.org/10.1001/archinte.165.21.2521 PMID: 16314550.

34. Onufrač S, Abramson J, Vaccarino V. Adult-onset asthma is associated with increased carotid atherosclerosis among women in the Atherosclerosis Risk in Communities (ARIC) study. Atherosclerosis. 2007; 195(1):129–37. Epub 2006/10/19. https://doi.org/10.1016/j.atherosclerosis.2006.09.004 PMID: 17045272.

35. Dratva J, Caviezel S, Schaffner E, Stolz D, Rotte T, Kuenzli N, et al. Is there a gender-specific association between asthma and carotid intima-media thickness in Swiss adolescents? Eur J Pediatr. 2018; 177(5):699–707. Epub 2018/02/08. https://doi.org/10.1007/s00431-018-3107-0 PMID: 29411143.

36. Ku DN, Giddens DP, Zarins CK, Glagov S. Pulsatile flow and atherosclerosis in the human carotid bifurcation. Positive correlation between plaque location and low oscillating shear stress. Arteriosclerosis. 1985; 5(3):293–302. Epub 1985/05/01. PMID: 3994585.

37. Tzou WS, Douglas PS, Srinivasan SR, Bond MG, Tang R, Li S, et al. Distribution and predictors of carotid intima-media thickness in young adults. Prev Cardiol. 2007; 10(4):181–9. Epub 2007/10/06. PMID: 17917514.

38. Den Ruijter HM, Peters SA, Anderson TJ, Britton AR, Dekker JM, Eijkmans MJ, et al. Common carotid intima-media thickness measurements in cardiovascular risk prediction: a meta-analysis. Jama. 2012; 308(8):796–803. Epub 2012/08/23. https://doi.org/10.1001/jama.2012.9630 PMID: 22910757.
39. Toren K, Lindholm NB. Do patients with severe asthma run an increased risk from ischaemic heart disease? Int J Epidemiol. 1996; 25(3):617–20. Epub 1996/06/01. PMID: 8671564.

40. Enright PL, Ward BJ, Tracy RP, Lasser EC. Asthma and its association with cardiovascular disease in the elderly. The Cardiovascular Health Study Research Group. J Asthma. 1996; 33(1):45–53. Epub 1996/01/01. PMID: 8621370.

41. Onufrait SJ, Abramson JL, Austin HD, Holguin F, McClellan WM, Vaccarino LV. Relation of adult-onset asthma to coronary heart disease and stroke. Am J Cardiol. 2008; 101(9):1247–52. Epub 2008/04/26. https://doi.org/10.1016/j.amjcard.2007.12.024 PMID: 1843952.

42. Lee HM, Truong ST, Wong ND. Association of adult-onset asthma with specific cardiovascular conditions. Respir Med. 2012; 106(7):948–53. Epub 2012/03/27. https://doi.org/10.1016/j.rmed.2012.02.017 PMID: 22445771.

43. Cakmak A, Zeyrek D, Cece H, Erel O. The relationship between carotid intima media thickness and oxidative stress in asthmatic children. Asian Pac J Allergy Immunol. 2010; 28(4):256–61. Epub 2011/02/23. PMID: 21337909.

44. Silverberg JI. Association between adult atopic dermatitis, cardiovascular disease, and increased heart attacks in three population-based studies. Allergy. 2015; 70(10):1300–8. Epub 2015/07/07. https://doi.org/10.1111/all.12685 PMID: 26148129.

45. Silverberg JI. Atopic disease and cardiovascular risk factors in US children. J Allergy Clin Immunol. 2016; 137(3):398–40.e1. Epub 2015/12/19. https://doi.org/10.1016/j.jaci.2015.09.012 PMID: 26679356.

46. Liisson J, Zagura M, Zilmer K, Salum E, Heilman K, Piir A, et al. Increased carotid artery intima-media thickness and myeloperoxidase level in children with newly diagnosed juvenile idiopathic arthritis. Arthritis Res Ther. PMID: 172015.

47. Schanberg LE, Sandborg CX, Barnhart HX, Ardoin SP, Yow E, Evans GW, et al. Premature atherosclerosis in pediatric systemic lupus erythematosus: risk factors for increased carotid intima-media thickness in the atherosclerosis prevention in pediatric lupus erythematosus cohort. Arthritis Rheum. 2009; 60(5):1496–507. Epub 2009/05/01. https://doi.org/10.1002/art.24469 PMID: 19404953.

48. Charakida M, Donald AE, Green H, Storry C, Clapson M, Caslake M, et al. Early structural and functional changes of the vasculature in HIV-infected children: impact of disease and antiretroviral therapy. Circulation. 2005; 112(1):103–9. Epub 2005/06/29. https://doi.org/10.1161/CIRCUITSPHASIC.A.104.517144 PMID: 15983247.

49. Wenzel SE. Asthma phenotypes: the evolution from clinical to molecular approaches. Nat Med. 2012; 18(5):716–25. Epub 2012/05/09. https://doi.org/10.1038/nm.2678 PMID: 22561835.

50. Kay AB. Allergy and allergic diseases. First of two parts. N Engl J Med. 2001; 344(1):30–7. Epub 2001/01/04. https://doi.org/10.1056/NEJM200101043440106 PMID: 11136958.

51. Lee YW, Kuhn H, Kaiser S, Hennig B, Daugher A, Toborek M. Interleukin 4 induces transcription of the 15-lipoxygenase I gene in human endothelial cells. J Lipid Res. 2001; 42(5):783–91. Epub 2001/05/16. PMID: 11352986.

52. Walch L, Massade L, Dufilho M, Brunet A, Rendu F. Pro-atherogenic effect of interleukin-4 in endothelial cells: modulation of oxidative stress, nitric oxide and monocyte chemoattractant protein-1 expression. Atherosclerosis. 2006; 187(2):285–91. Epub 2005/10/27. https://doi.org/10.1016/j.atherosclerosis.2005.09.016 PMID: 16249002.

53. Di Gennaro A, Haeggstrom JZ. The leukotrienes: immune-modulating lipid mediators of disease. Adv Immunol. 2012; 116:51–92. Epub 2012/10/16. https://doi.org/10.1016/B978-0-12-394300-2.00002-8 PMID: 23063073.

54. Aiello RJ, Bourassa PA, Lindsey S, Weng W, Freeman A, Showell HJ. Leukotriene B4 receptor antagonism reduces monocytic foam cells in mice. Arterioscler Thromb Vasc Biol. 2002; 22(3):443–9. Epub 2002/03/09. PMID: 11884288.

55. Spanbroek R, Grabner R, Lotzer K, Hildner M, Urbach A, Ruhling K, et al. Expanding expression of the 5-lipoxygenase pathway within the arterial wall during human atherogenesis. Proc Natl Acad Sci U S A. 2003; 100(3):1238–43. Epub 2003/01/29. https://doi.org/10.1073/pnas.242716099 PMID: 12552108.

56. Zhu Y, Shao Z, Jing J, Ma J, Chen Y, Li X, et al. Body Mass Index Is Better than Other Anthropometric Indices for Identifying Dyslipidemia in Chinese Children with Obesity. PLoS One. 2016; 11(3): e0149392. Epub 2016/03/11. https://doi.org/10.1371/journal.pone.0149392 PMID: 26963377.