

Birth Order and Cord Immunoglobulin E: Results Using a High-Sensitivity Immunoglobulin E Protocol

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Key Words

Allergic disease · Atopic disease · Birth order · Immunoglobulin E · Epidemiology

Abstract

Background: Studies have shown an inverse association between birth order and allergic disease risk; some but not all have shown an inverse association between cord blood immunoglobulin E (IgE) and birth order. We further examined the relationship between birth order and cord blood IgE in a racially diverse birth cohort. **Methods:** Women were interviewed about their pregnancy history, and their babies' cord blood was collected to measure total IgE using a high-sensitivity protocol (lower detection limit 0.01 IU/ml). We analyzed cord IgE as both a continuous and categorical measure. **Results:** Of the 733 children, 171 (23%) were first born, 92 (13%) were first born with the mother having prior pregnancies but no live births, and 470 (64%) were born second or later. By birth order, the geometric means $\pm$ standard deviations were: first born 0.26 ± 4.2 IU/ml, first born after prior pregnancies 0.35 ± 3.9 IU/ml, second born 0.30 ± 4.8 IU/ml, third born 0.28 ± 5.1 IU/ml, and fourth born or greater 0.28 ± 4.5 IU/ml (trend $p = 0.51$). Other factors considered

(maternal allergic disease history, age, race, exposure to smoking and cats/dogs during pregnancy, fetal gender, season of delivery) neither modified nor confounded these relationships. **Conclusions:** Unlike some previous reports, there was no association between total cord IgE level and birth order. Mechanisms other than cord IgE should be studied in the quest to understand the role of birth order in allergic disease risk. Categorization of a continuous measure of IgE may incorrectly create statistically significant results.

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Introduction

Although a biological mechanism has not yet been identified, a large body of evidence indicates that later birth order is inversely associated with the risk of childhood allergic diseases [1]. In the context of the hygiene hypothesis, Strachan [2] proposed that birth order could be associated with frequency of infection, since having older siblings may increase the likelihood of exposure to infectious disease leading to a protective effect against childhood allergic disease. However, there is no scientific consensus on the underlying biological mecha-

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nism(s). Thus, alternative explanations have been pursued. One such approach is to consider the role of programming in utero. It has been proposed that maternal immunoglobulin E (IgE) levels decrease with each pregnancy (tolerance induction hypothesis) and that a tendency towards lower IgE is transmitted to subsequent offspring (tolerance transmission hypothesis) [3].

To explore the latter hypothesis, we analyzed the relationship between the pregnancy history of mothers enrolled in our ongoing cohort study and the total IgE in the cord blood of their children. We used a high-sensitivity protocol for IgE (lowest detection limit of 0.01 IU/ml) and analyzed the data in several ways in order to compare our results to prior research. Further, these analyses will demonstrate that variation in results can be introduced by turning continuous variables into categorical variables.

Material and Methods

The Wayne County Health, Environment, Allergy and Asthma Longitudinal Study (WHEALS) is an ongoing cohort study in southeastern Michigan designed to examine the relationships between early life exposures such as pets, infections and endotoxin and allergic diseases in early childhood. We recruit 21- to 49-year-old women from a predefined geographic area who are in their second trimester of pregnancy, are seeing a Henry Ford Health System medical group clinician at 1 of 5 clinics, and plan to stay in the Detroit area for at least 2 years after delivery. Prior to meeting the women at the clinics to discuss participation, a letter introducing the study is mailed to them.

We ask women to complete an interview at the time of recruitment about their pregnancy and asthma and allergy histories, as well as information about the asthma and allergy histories of their babies' fathers. Collected cord blood samples are sent frozen by our molecular epidemiology laboratory to the Medical College of Georgia for analyses of total IgE. Measurements are performed using the Pharmacia UniCAP system (Pharmacia-Upjohn Diagnostic Division, Kalamazoo, Mich., USA), according to the manufacturer's protocol for high sensitivity. The high-sensitivity protocol provides an assay range of 0.01–100 IU/ml IgE. One percent of all assays are repeated in a different assay run on a different day to provide a precise estimate of intra-assay reliability over time. Thirty-four (5%) of our samples had undetectable measures of IgE which were assigned a value of 0.01 IU/ml. All IgE units are given in international units per milliliter.

Recruitment began in September 2003 and is ongoing. Women are required to speak English well enough to provide written informed consent. This research was approved by the Henry Ford Health System IRB.

At the time of this writing, 3,086 letters had been mailed to potentially eligible women. We were unable to meet with 905 (29.3%) women prior to delivery to confirm eligibility for several reasons including appointment cancellations/changes and the inability of the study to have recruiters present at all clinic appoint-

ments due to resource constraints; 267 (8.7%) women were determined to be ineligible after meeting with them to confirm eligibility criteria; 129 women (4.2%) did not speak English well enough to provide acceptable written informed consent; 792 contacted eligible women (44.4% of the known eligible women) declined to participate in the study, and 993 (55.7%) provided written informed consent for study participation.

Thus far, we have conducted IgE analyses on 733 cord blood samples. Women's pregnancy histories include information about the number, timing, duration and outcomes of all prior pregnancies including those not carried to term. Using this information, children were assigned a pregnancy/birth order group based on information from the maternal predelivery interview: no prior pregnancies, prior pregnancies but no live births, 1 prior live birth, 2 prior live births, or 3 or more prior live births. There were not enough participants to analyze children whose mothers' prior pregnancies were recognized as spontaneous abortions versus elective abortions.

Using information from the interview, we classified mothers as having a history of allergic disease if they reported ever having a doctor diagnosis of asthma, eczema, hay fever or allergic rhinitis, or took prescription medication for asthma or allergies. The maternal allergic history, gender of the baby, season of delivery, self-reported maternal race, maternal tobacco smoke exposure during pregnancy, maternal age and the presence of dogs/cats in the home during pregnancy were investigated as effect modifiers and then as confounders of the relationship between prior live births and total IgE in the children's cord blood.

We used robust descriptive statistics (geometric means and standard deviations) to describe the relationship between the non-normally distributed cord IgE and the number of prior live births. We assessed associations between the number of prior live births and continuous cord blood total IgE for all children and within race and gender stratum using nonparametric Spearman correlations. To evaluate differences in IgE between the prior live birth categories, the 2-sided Jonckheere-Terpstra test, a nonparametric trend test, was used in order to take advantage of the intrinsic order of the increasing number of prior live births [4]. We used stratum-specific 2-sided Jonckheere-Terpstra tests to evaluate effect modification and linear regression models of log-transformed cord IgE measures to assess potential confounders of the relationship between the number of prior live births and the outcome of total cord blood IgE. We assessed confounding by evaluating the coefficients for birth order in adjusted and unadjusted models using a change in effect criteria of at least 15% to warrant adjustment.

To further address outliers and examine those children without elevated IgE levels, we repeated these analyses for only those children with IgE <10 IU/ml and then again for only those with IgE <2 IU/ml.

After completing the analyses of a continuous measure of cord blood IgE, we analyzed the data with a categorical approach using a variety of cord blood IgE cutpoints in order to facilitate comparisons with prior published research. We further stratified the categorical results for the cutpoint of 0.5 IU/ml for variables often used in analyses involving cord blood levels (baby's sex, maternal age, maternal race). For the categorical analyses, we conducted Cochran-Armitage trend tests (excluding children born to mothers who have had prior pregnancies, but no live births).

Table 1. Descriptive information about the WHEALS population by pregnancy history prior to this live birth

	All women (n = 733)	No prior pregnancies (n = 171)	At least 1 prior pregnancy but no live births (n = 92)	Number of prior live births		
				1 (n = 257)	2 (n = 129)	≥3 (n = 84)
Age at recruitment						
21–25 years	225 (30.7)	80 (46.8)	39 (42.4)	77 (29.9)	23 (17.8)	6 (7.2)
26–30 years	236 (32.2)	46 (26.9)	28 (30.4)	92 (35.8)	43 (33.3)	27 (32.1)
31–35 years	185 (25.2)	35 (20.5)	17 (18.5)	66 (25.7)	40 (31.0)	27 (32.1)
36–40 years	82 (11.2)	10 (5.8)	7 (7.6)	21 (8.2)	22 (17.1)	22 (26.2)
40+ years	5 (0.7)	0 (0)	1 (1.1)	1 (0.4)	1 (0.8)	2 (2.4)
Ethnicity						
African-American/black	441 (60.2)	86 (50.3)	69 (75.0)	153 (59.5)	79 (61.2)	54 (64.3)
Caucasian/white	209 (28.5)	62 (36.3)	16 (17.4)	75 (29.2)	35 (27.1)	21 (25.0)
American Indian/Alaskan native	6 (0.8)	0 (0)	2 (2.2)	3 (1.2)	1 (0.8)	0 (0)
Asian	20 (2.7)	7 (4.1)	0 (0)	8 (3.1)	2 (1.6)	3 (3.6)
Pacific islander	1 (0.1)	0 (0)	0 (0)	1 (0.4)	0 (0)	0 (0)
Refused	2 (0.3)	1 (0.6)	0 (0)	0 (0)	1 (0.8)	0 (0)
Other	54 (7.4)	15 (8.8)	5 (5.4)	17 (6.6)	11 (8.5)	6 (7.1)
Mother smoked during pregnancy						
Yes	93 (12.7)	27 (15.8)	11 (12.0)	31 (12.1)	15 (11.6)	9 (10.7)
No	640 (87.3)	144 (84.2)	81 (88.0)	226 (87.9)	114 (88.4)	75 (89.3)
Mother lived with cat/dog during pregnancy ¹						
Yes	285 (38.9)	75 (43.9)	40 (43.5)	95 (37.0)	47 (36.4)	28 (33.7)
No	447 (61.1)	96 (56.1)	52 (56.5)	162 (63.0)	82 (63.6)	55 (66.3)
Baby gender ²						
Male	359 (49.7)	82 (48.5)	41 (45.6)	137 (54.1)	63 (49.2)	36 (43.9)
Female	363 (50.3)	87 (51.5)	49 (54.4)	116 (45.9)	65 (50.8)	46 (56.1)
Season of delivery						
December–February	161 (22.0)	43 (25.2)	21 (22.8)	52 (20.2)	31 (24.0)	14 (16.6)
March–May	209 (28.5)	44 (25.7)	25 (27.2)	70 (27.3)	44 (34.1)	26 (31.0)
June–August	177 (24.1)	40 (23.4)	20 (21.7)	63 (24.5)	28 (21.7)	26 (31.0)
September–November	186 (25.4)	44 (25.7)	26 (28.3)	72 (28.0)	26 (20.2)	18 (21.4)
Household smoker during pregnancy ³						
Yes	201 (27.4)	49 (28.7)	28 (30.4)	69 (26.8)	33 (25.6)	22 (26.2)
No	532 (72.6)	122 (71.3)	64 (69.6)	188 (73.2)	96 (74.4)	62 (73.8)
Maternal allergic disease history ¹						
Yes	295 (40.3)	69 (40.4)	45 (48.9)	90 (35.0)	53 (41.4)	38 (45.2)
No	437 (59.7)	102 (59.6)	47 (51.1)	167 (65.0)	75 (58.6)	46 (54.8)

Figures in parentheses are percentages.

¹ Unknown for 1 mother. ² Unknown for 11 babies. ³ Includes mother.

Results

Table 1 presents descriptive information about the study population by birth order groupings. As expected, mothers with more prior live births tended to be older than the other groups. The majority of the mothers identified themselves as African-American/black (60.2%).

The overall geometric mean $\pm$ SD of cord blood total IgE for all 733 babies was 0.29 ± 4.6 IU/ml; 64% of the blood samples had IgE values <0.5 IU/ml. The geometric

means for those with and without prior live births were 0.29 ± 4.8 (n = 470) and 0.29 ± 4.1 IU/ml (n = 263), respectively. The geometric means for those with less than 3 prior live births and those with 3 or more prior live births were 0.29 ± 4.6 (n = 649) and 0.28 ± 4.5 IU/ml (n = 84), respectively. The Spearman correlation between the number of prior live births and cord blood total IgE was $r = 0.05$ and $p = 0.19$ (black mothers, $r = -0.001$, $p = 0.97$; white mothers, $r = 0.07$, $p = 0.29$; male babies, $r = 0.03$, $p = 0.63$; female babies, $r = 0.07$, $p = 0.20$).

Table 2. WHEALS babies' cord blood total IgE geometric means $\pm$ SD (IU/ml) by maternal pregnancy history

	No prior pregnancies	At least 1 prior pregnancy/ no live births	Prior live births		
			1	2	≥ 3
All babies					
Geometric	0.26 $\pm$ 4.2	0.35 $\pm$ 3.9	0.30 $\pm$ 4.8	0.28 $\pm$ 5.1	0.28 $\pm$ 4.5
n	171	92	257	129	84
p value	0.51				
Babies of black mothers					
Geometric	0.38 $\pm$ 3.4	0.32 $\pm$ 3.8	0.39 $\pm$ 4.9	0.40 $\pm$ 5.3	0.30 $\pm$ 4.6
n	86	69	153	79	54
p value	0.84				
Babies of white mothers					
Geometric	0.14 $\pm$ 3.9	0.41 $\pm$ 5.2	0.14 $\pm$ 3.7	0.16 $\pm$ 3.6	0.25 $\pm$ 4.5
n	62	16	75	35	21
p value	0.32				
Female babies					
Geometric	0.25 $\pm$ 3.6	0.38 $\pm$ 3.9	0.26 $\pm$ 4.4	0.28 $\pm$ 5.2	0.22 $\pm$ 4.1
n	87	49	116	65	46
p value	0.90				
Male babies					
Geometric	0.27 $\pm$ 5.1	0.31 $\pm$ 4.1	0.33 $\pm$ 5.2	0.27 $\pm$ 5.0	0.42 $\pm$ 4.6
n	82	41	137	63	36
p value	0.23				
Mother <30 years old at birth					
Geometric	0.26 $\pm$ 3.9	0.31 $\pm$ 3.7	0.30 $\pm$ 4.5	0.25 $\pm$ 4.3	0.19 $\pm$ 3.9
n	117	62	145	58	25
p value	0.82				
Mother ≥ 30 years old at birth					
Geometric	0.24 $\pm$ 5.1	0.45 $\pm$ 4.3	0.29 $\pm$ 5.2	0.31 $\pm$ 5.9	0.34 $\pm$ 4.6
n	54	30	112	71	59
p value	0.26				

p values for the Jonckheere-Terpstra test that excludes children born to mothers who have had prior pregnancies, but no live births.

Table 2 presents the geometric means by birth order for all children and then separately for children born to mothers who identified themselves as African-American/black or white, for male and female children and for children born to mothers at least 30 years of age at birth or less than 30 years of age at birth. There was no statistically significant trend between the number of prior live births and cord IgE geometric means ($p = 0.51$) for all children. There were no statistically significant trends by birth order when we examined the geometric means stratified by race (black or white), maternal age, maternal allergic disease history, maternal smoking during pregnancy, mother living with a smoker during pregnancy, mother living with cats/dogs during pregnancy, baby's sex and season of birth (data shown only for race, baby's

sex and maternal age). None of these factors were confounders of the association between birth order and cord blood IgE level.

After repeating these analyses for only those children with IgE <10 IU/ml and again for only those with IgE <2 IU/ml, neither set of results differed significantly from the unrestricted analysis.

Table 3 presents the percentages of children who have IgE above various cutpoints within the birth order groups as well as p values for the trend tests. Among these unadjusted results, no level has an associated p value <0.05 for the Cochran-Armitage trend test. Several cutpoints were near statistical significance (IgE >0.4 IU/ml, $p = 0.079$; IgE >0.6 IU/ml, $p = 0.074$; IgE >0.9 IU/ml, $p = 0.10$). For these groups, the trends indicate that those with no prior

Table 3. Percentages of WHEALS babies with cord blood total IgE greater than various cutpoints by maternal pregnancy history

All babies	No prior pregnancies (n = 171)	At least 1 prior pregnancy/ no live births (n = 92)	Prior live births		
			1 (n = 257)	2 (n = 129)	≥3 (n = 84)
Cord IgE >0.20 IU/ml	57.9	69.6	59.5	54.3	57.1
p value	0.64				
Cord IgE >0.30 IU/ml	42.7	55.4	46.3	45.7	50.0
p value	0.31				
Cord IgE >0.35 IU/ml	36.3	45.7	42.0	41.9	42.9
p value	0.29				
Cord IgE >0.40 IU/ml	32.2	41.3	37.7	39.5	42.9
p value	0.079				
Cord IgE >0.5 IU/ml	26.3	34.8	33.5	33.3	35.7
p value	0.12				
Cord IgE >0.6 IU/ml	21.6	29.4	29.6	31.0	31.0
p value	0.074				
Cord IgE >0.7 IU/ml	18.7	26.1	24.9	28.7	23.8
p value	0.16				
Cord IgE >0.8 IU/ml	17.0	26.1	21.4	27.9	21.4
p value	0.12				
Cord IgE >0.9 IU/ml	15.2	22.8	18.7	24.8	20.2
p value	0.10				

p values for the Cochran-Armitage test that excludes children born to mothers who have had prior pregnancies, but no live births.

pregnancies were less likely than any other group to have a high level of cord IgE.

Table 4 presents the percentages of children who have IgE >0.5 IU/ml within the birth order groups stratified by factors often considered important in cord blood IgE analyses (baby's gender, maternal race, maternal age). There were no statistically significant trends ($p < 0.05$). However, although not statistically significant, similar trends of first-born children being less likely to have cord IgE >0.5 IU/ml were seen for all babies ($p = 0.12$), babies born to white women ($p = 0.062$) and women at least 30 years of age ($p = 0.068$), as well as for male babies ($p = 0.12$).

Discussion

We found no association between birth order and cord blood IgE levels, even after considering the potential effects of numerous additional factors. If we had only conducted a categorical analysis, we would have interpreted the results (lower birth order is associated with lower

cord IgE) as showing a tendency towards the opposite of what others have previously published (lower birth order associated with higher cord IgE). This would have been erroneous.

Prior evidence to support an association between birth order, immune response and neonatal IgE has been mixed. In their cohort of 223 children in Scotland, Devereux et al. [5] reported that cord blood mononuclear cell-proliferative responses after stimulation with *Mycobacterium tuberculosis* purified protein derivative, timothy grass and house dust mite tended to be higher in samples from first-born children compared with later-born children (mean and median values not presented; none of the reported p values were <0.1). The reported associations were stronger when they used categories based on the number of the mother's total pregnancies (dust mite, $p = 0.021$; purified protein derivative, $p = 0.07$; timothy grass, data not provided). The analyses were unadjusted.

Karmaus et al. [6] reported that children born third or later in their family were less likely (adjusted odds ratio 0.59; 95% confidence interval 0.41–0.83) than first-born

Table 4. Percentages of WHEALS babies with cord blood total IgE >0.5 IU/ml by maternal pregnancy history

	No prior pregnancies	At least 1 prior pregnancy/ no live births	Prior live births		
			1	2	≥3
All babies					
Cord IgE >0.5 IU/ml	26.3	34.8	33.5	33.3	35.7
n	171	92	257	129	84
p value	0.12				
Babies of black mothers					
Cord IgE >0.5 IU/ml	33.7	34.8	41.2	43.0	38.9
n	86	69	153	79	54
p value	0.38				
Babies of white mothers					
Cord IgE >0.5 IU/ml	12.9	31.3	16.0	17.1	33.3
n	62	16	75	35	21
p value	0.062				
Female babies					
Cord IgE >0.5 IU/ml	25.3	30.6	31.9	35.4	28.3
n	87	49	116	65	46
p	0.39				
Male babies					
Cord IgE >0.5 IU/ml	28.1	39.0	35.0	30.2	47.2
n	82	41	137	63	36
p value	0.12				
Mothers <30 years old at birth					
Cord IgE >0.5 IU/ml	26.5	37.1	33.8	25.9	24.0
n	117	62	145	58	25
p value	0.86				
Mothers ≥30 years old at birth					
Cord IgE >0.5 IU/ml	25.9	30.0	33.0	39.4	40.7
n	54	30	112	71	59
p value	0.068				

p values for the Cochran-Armitage test that excludes children born to mothers who have had prior pregnancies, but no live births.

children to be in a higher total cord IgE group (ordinal logistic model; total IgE groups: <0.2, 0.2 to <0.5, ≥0.5 kU/l; IgE assay lower detection limit 0.2 kU/l) in a cross-sectional analysis of more than 800 children from the Isle of Wight birth cohort. This research team later reported a statistically significant Spearman correlation ($p = 0.008$) between cord serum IgE and birth order of live offspring for this same cohort [3]. However, it could be argued that their actual correlation of -0.092 is weak and not indicative of an association.

In a study of cord blood from more than 6,000 German children, decreased IgE values were associated with increasing parity (IgE assay lower detection limit 0.35 kU/l) [7]. However, this relationship was not statistically significant, in spite of the large sample size. Further details of the association were not provided.

No association was found between first born and cord IgE in a cohort of 291 Dominican and African-American children born to women living in Northern Manhattan (IgE assay lower detection limit 0.5 IU/ml) [8]. Similarly, in a birth cohort of 1,027 Dutch children, there was no association found between IgE in blood collected in the first week of life (heel stick) and the presence of older siblings (IgE assay lower detection limit 0.5 IU/ml) [9].

Prior research using cord blood IgE has been conducted with a categorical data analytical approach [6]. This can be misleading. Prior studies of cord blood IgE and birth order have used >0.5 IU/ml as a measure of high cord blood IgE [6]. With this approach, all values >0.5 IU/ml are treated the same. If one group has more children with cord blood IgE above this level, it will appear as though that group is associated with higher IgE. This

obscures the actual order and magnitude of the measurements.

If in a study population, for example, subgroup A has children with cord IgE measurements of 0.40, 0.51, 0.51, 0.51 and 0.51 IU/ml and children in subgroup B have measurements of 0.49, 0.49, 0.49, 0.61 and 0.61 IU/ml, 80% of children in group A would have high IgE compared with 40% of children in group B if a cutpoint of >0.5 IU/ml were used. This is a relative risk of 2.0 (risk in A versus risk in B). However, the mean of group A is 0.488 IU/ml, while the mean of group B is 0.53 IU/ml – an approximately 8% increase from group A to B. This is neither the same magnitude nor direction of the relative risk. An analysis of a continuous measure must address rank as well as magnitude of measurements from different subgroups.

In our analyses, dichotomizing the continuous outcome could have led us to erroneously conclude that, based on marginal statistical significance, early birth order (fewer older siblings) was associated with lower risk of high IgE. In comparison, the results were not at all statistically significant when analyzed as a log-transformed continuous variable. This is an example in which dichotomization of a continuous variable has created evidence of a misleading association. This type of statistical event has been discussed in the literature [10–12].

Because accrual of cord blood IgE is ongoing in our study, we projected the *p* value for the Jonckheere-Terpstra trend test for a larger sample size based on associations in our observed data ($n = 733$). With a sample size of 1,000, we estimate that the Jonckheere-Terpstra trend test would have a *p* value of 0.41. This means that if we saw the same patterns in a larger data set, we would still not see a statistically significant trend.

With, as of yet, no biological mechanism to explain the generally well-accepted birth order effect on allergic disease, the proposed hypotheses of Karmaus et al. [3] (the maternal induction and the maternal transference hypotheses) have been interesting pathways to pursue. However, the focus on cord IgE has not been the simple answer that clarifies the biological mechanism of how birth order is actually related to subsequent allergic disease risk. The effects of birth order on allergic disease risk probably come from complex interactions between several factors: (1) the in utero environment influenced by normal changes to the immune system associated with pregnancy, maternal environmental exposures, infections and inflammation; (2) a child's environment in early life influenced by older siblings and exposure to other children, and (3) a complex profile of genetic factors that

influence immune responses to environmental exposures. Some have suggested that later birth order may have the effect of delayed sensitization [13]. We look forward to studying this in our cohort as the children age in the coming years.

There are numerous strengths to our study. Our study is comprised of a diverse population which allowed us to consider whether race affected the relationships. We also considered numerous factors that neither modified nor confounded the associations studied.

Ideally, we would have had a higher participation rate. Additionally, although we collected details on all prior pregnancy outcomes, we did not have a sufficient sample size to examine women with prior spontaneous versus elective abortions among women with prior pregnancies but no prior live births. We think this may be important to the study due to the recognized crucial role that the maternal immune system plays in pregnancy maintenance [14]. Also, we do not have information on change in partner from previous pregnancies. However, we are not aware of any scientific consensus stating that the birth order effect is only seen when the second-born child shares the father with the first born. Change in father may represent greater interval between pregnancies and exposure to new male antigens.

We did not conduct any tests to rule out maternal contamination of the cord blood. Scirica et al. [15] succinctly summarized the reasons why this is not necessary in their recent analysis of predictors of cord blood IgE in which they did not screen their samples for maternal cord blood contamination. Briefly, most of the IgE detected in the cord blood is probably from the fetus itself since (1) maternal IgE does not cross the placenta [16], and (2) by the 11th week of gestation, the fetus is capable of producing IgE [17]. They further explain that although some studies have measured both IgE and IgA in cord blood, the rates of contamination were low [7, 18–20].

Focusing on cord blood IgE alone has not provided insight into understanding the biological mechanism underlying the relationship between birth order and allergic disease risk. An additional focus on mechanisms and measures such as regulatory T cells has been suggested [21]. The focus on the in utero environment and the maternal-fetal interface during pregnancy is emerging as an important area of research demanding further investigation and understanding. Better understanding of within-woman change in the immune system over the course of pregnancy and the persistence of any changes after initial and subsequent pregnancies would be useful in the elucidation of factors affecting allergic risk in children.

Our continued analyses of these data will focus on the relationship between time since prior pregnancy and cord IgE, as well as the relationship between pregnancy history and maternal IgE. Future work will also examine the relationship between the cord blood IgE level and the

subsequent risk of allergic disease. These are all important relationships that deserve careful analysis as we work to tease out the hypothesized complex link between pregnancy history and allergic disease risk.

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