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Preterm birth, vaccination and neurodevelopmental disorders: a cross-sectional study of 6- to 12-year-old vaccinated and unvaccinated children

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Abstract

From about 8% to 27% of extremely preterm infants develop symptoms of autism spectrum disorder, but the causes are not well understood. Preterm infants receive the same doses of the recommended vaccines and on the same schedule as term infants. The possible role of vaccination in neurodevelopmental disorders (NDD) among premature infants is unknown, in part because pre-licensure clinical trials of pediatric vaccines have excluded ex-preterm infants. This paper explores the association between preterm birth, vaccination and NDD, based on a secondary analysis of data from an anonymous survey of mothers, comparing the birth history and health outcomes of vaccinated and unvaccinated homeschool children 6 to 12 years of age. A convenience sample of 666 children was obtained, of which 261 (39%) were unvaccinated, 7.5% had an NDD (defined as a learning disability, Attention Deficit Hyperactivity Disorder and/or Autism Spectrum Disorder), and 7.7% were born preterm. No association was found between preterm birth and NDD in the absence of vaccination, but vaccination was significantly associated with NDD in children born at term (OR 2.7, 95% CI: 1.2, 6.0). However, vaccination coupled with preterm birth was associated with increasing odds of NDD, ranging from 5.4 (95% CI: 2.5, 11.9) compared to vaccinated but non-preterm children, to 14.5 (95% CI: 5.4, 38.7) compared to children who were neither preterm nor vaccinated. The results of this pilot study suggest clues to the epidemiology and causation of NDD but question the safety of current vaccination practices for preterm infants. Further research is needed to validate and investigate these associations in order to optimize the impact of vaccines on children's health.

Introduction

Preterm birth (defined as birth occurring before 37 completed weeks of gestation) is known as a major risk factor for neurodevelopmental deficits, including cerebral palsy, intellectual disability, cognitive and speech delays, motor deficits, and visual impairment associated with retinopathy of prematurity. In particular, preterm birth is the leading cause of neurodevelopmental disorders (NDD) and disability, including the development of autism spectrum disorder (ASD) [1-3], but the underlying mechanisms are not well understood. Preterm infants receive the same doses of the recommended vaccines on the same schedule as term infants in order to protect them from several infections [4-7]. However, the possible role of vaccination in the development of NDD in premature infants has not been assessed, partly because pre-licensure clinical trials of pediatric vaccines have routinely excluded ex-preterm infants, and because of the assumed overall safety of vaccinations [8-15].

This paper presents additional results of a survey designed to compare the health outcomes of vaccinated and unvaccinated children educated at home, based on mothers' anonymous reports on the birth histories and physician-diagnosed illnesses in their children. The analysis explores the possible role of vaccination in NDD among children born preterm.

Preterm birth and vaccination

In 2012, 450,000 babies (11%) were born preterm in the United States, resulting in 35% of all infant deaths in that year, more than any other single cause. Worldwide, an estimated 15 million infants were born preterm in 2010, of which about 13 million survived beyond the first month. In addition to the acute complications of prematurity, which include respiratory distress, intracranial hemorrhage, necrotizing enterocolitis and retinopathy, 345,000 (2.7%) preterm infants were estimated to have moderate to severe neurodevelopmental impairment; a further 567,000 (4.4%) had mild neurodevelopmental impairment, and many more had specific learning or behavioral disorders [4]. Advances in medical care have led to increased rates of preterm birth and decreased preterm mortality rates. However, neurodevelopmental disabilities have increased, especially in infants born at ≤25 weeks' gestation, with nearly half of surviving extremely preterm infants having significant disabilities [16]. Total annual costs of preterm birth in the United States exceed $26 billion per year, with an overall average cost of approximately $51,600 per preterm infant [17].

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In a follow-up study of 11-year-old children born extremely preterm, the rate of ASD was 8% compared to zero percent among their classmates born at term [18] – a rate at least 5 times higher than recent population estimates for ASD (14.7 per 1,000, or 1 in 68 children aged 8 years) [19]. In another study involving 84 children born at <27 weeks of gestation who were screened for ASD at 6.5 years, 23 (27.4%) tested positive for ASD. The ASD-positive group had a significantly higher frequency of neonatal complications than the ASD-negative group [20].

Since special efforts are made to vaccinate preterm infants, the effects of prematurity are difficult to separate from those of vaccination. Given the benefits of vaccination, it has not been thought necessary to do so. On the other hand, vaccine safety assessment in preterm infants is particularly important due to the frequency of adverse events associated with prematurity itself [21]. Adverse cardiorespiratory events including apnea, bradycardia and desaturations (oxygen saturation <90%) are well documented following vaccination in many preterm infants, yet vaccination is strongly recommended regardless of such events, since the prevention of infection is considered paramount. Pertussis-containing pediatric vaccines in particular can increase apnea and bradycardia in ex-preterm infants, typically after the 2-month dose [22,23]. In a study evaluating the safety of hexavalent vaccines (DTaP-IPV-Hib) involving 78 preterm infants, vaccination triggered transient cardiorespiratory events in 47% (15% apnea, 21% bradycardia, 42% desaturations), and those with pre-existing cardiorespiratory symptoms had a five- to eight-fold increased risk of cardiorespiratory events post-vaccination [24].

Age at vaccination of preterm infants is inversely associated with adverse effects. In one study of 27 preterm infants vaccinated at ≤70 days of age, 9 (33.3%) developed apnea, bradycardia or desaturations compared to none among those vaccinated at >70 days of age [25]. A study of nearly 14,000 extremely low-birth-weight (ELBW) infants reported a 3.7-fold increase in Rule-Out Sepsis evaluations in the immediate post-vaccination period as well as high rates of apnea, bradycardia and intubations [26]. Pre-licensure clinical trials of pediatric combination vaccines have, however, often specifically excluded ex-preterm infants, even though these vaccines are routinely administered to all infants regardless of gestational age at 2, 4 and 6 months of age [27].

At present, over 95% of all U.S. children receive the CDC-recommended vaccines [28,29] in accordance with the policy that universal vaccination is essential for maintaining herd immunity [30]. Vaccination is estimated to have prevented millions of illnesses and hospitalizations and 732,000 premature deaths among U.S. children born during 1995-2013, at an overall cost savings of $1.38 trillion [31]. Much less is known about the extent and cost of vaccine-associated injury, which can include severe morbidity and death [32]. Since 1988 over 16,038 claims have been filed with the Vaccine Injury Compensation Program, which was created in 1986 to compensate individuals and their families for injuries caused by vaccines. Total compensation paid since the program began is approximately $3.2 billion [33]. Since only about one percent of serious vaccine injuries are officially reported [34], the true extent and cost of vaccine-associated damage on a population basis is likely to be much higher.

A complicating factor in evaluating the vaccination program is that individual vaccines against infectious diseases have nonspecific effects on morbidity and mortality that extend beyond prevention of the targeted disease. In some instances these effects are beneficial; in others they increase morbidity and mortality [35]. For instance, the measles and Bacille Calmette- Guérin (BCG) vaccines reportedly reduce overall morbidity and mortality [36], whereas the diphtheria-tetanus-pertussis [37] and the hepatitis B vaccines [38] are associated with increased morbidity and mortality risks. These nonspecific effects are consistently reported in studies carried out in low-income countries but require replication in higher-income populations.

Exploring the association between preterm birth, vaccination and neurodevelopmental disorders

While the safety of vaccines is officially assured, observational studies have involved only a limited number of vaccines and vaccine ingredients, and none has reported on the long-term outcomes of the present vaccination schedule [39], which has been expanded and accelerated in recent decades [40]. The current childhood vaccination program now includes 48 doses of vaccines for 14 diseases from birth to age 6 years compared to 3 vaccinations for 7 diseases in the 1970s [41]. Given the many unknowns related to the long-term effects of vaccines and their ingredients, the U.S. Institute of Medicine has recommended that studies be carried out to compare health outcomes in vaccinated and unvaccinated groups of children and to determine the cumulative effects of vaccines [12].

A difficulty in comparing vaccinated and unvaccinated children has been the apparent absence of a readily available pool of unvaccinated children. The growing population of homeschool children (*i.e.,* children educated at home) is a suitable source for such studies, as vaccination rates are lower in homeschool children [42]. Homeschool families and children are approximately representative of US families and children in general, with an approximately equal median income to that of married-couple families nationwide, somewhat more years of formal education than parents in the general population, and a higher average family size of just over three children compared to the national average of just over two children [43-45]. Geographically, homeschooling families are slightly overrepresented in the south; children from all racial/ethnic backgrounds are involved in homeschooling; about 23% are nonwhite; and the age distribution of homeschool children in grades K-12 (ages 6 to 18) is similar to that of children nationwide [46]. About 3.4% (1.8 million) of the school-age population was homeschooled in the 2011-2012 school year [47].

An opportunity to address the question of the possible role of vaccination in NDD associated with preterm birth arose from the availability of data from a cross-sectional pilot study of the birth histories and health outcomes of vaccinated and unvaccinated homeschool children ages 6 to 12, carried out by the authors [48]. The first aim of the study was to compare vaccinated and unvaccinated children on a broad range of health outcomes, based on mothers' reports in an anonymous online survey. The collected data included physician-diagnosed acute and chronic illnesses, medications and the use of health services, as well as pregnancy experiences and birth histories. The second aim of the study was to determine whether an association found between vaccination and NDD, if any, remained significant after controlling for other measured factors.

This report presents additional findings on the independent association and/or interaction between preterm birth, vaccination and NDD.

Methods

Study planning

A partnership was formed with the National Home Education Research Institute (NHERI) to carry out the proposed study. NHERI has been involved in educational research on homeschooling for

many years and has strong and extensive contacts with homeschool organizations and families throughout the country (www.nheri.org). The study was approved by the Institutional Review Board of Jackson State University.

Study design

The study was cross-sectional in design, aimed at determining the association between vaccination and health outcomes, and was based on a survey of homeschooling mothers on the overall health of their vaccinated and unvaccinated biological children. Contact information on homeschool families was unavailable. Hence, there was no defined population or sampling frame from which a randomized study could be carried out and response rates could be determined. However, the goal of the pilot study was rather to obtain a convenience sample of unvaccinated children of sufficient size to test for significant differences in outcomes between the groups.

We proceeded by selecting 4 states (Florida, Louisiana, Mississippi, and Oregon) for the survey (Stage 1). NHERI compiled a list of statewide and local homeschool organizations, totaling 84 in Florida, 18 in Louisiana, 12 in Mississippi and 17 in Oregon. Initial contacts were made in June 2012. NHERI sent emails to the leaders of each statewide organization requesting their support for the study; this was followed by a second email, explaining the study purpose and background, which the leaders were asked to forward to their members (Stage 2). Prospective respondents were provided with a link to the questionnaire. With funding limited to a one-year study period, we sought to obtain as many responses as possible, contacting families only indirectly through homeschool organizations. This is considered a pilot study for a future national study of homeschool children.

Biological mothers of homeschool children ages 6-12 years were asked to serve as respondents in order to standardize data collection. They were asked to confirm their consent to participate, to indicate their home state and zip code of residence, and to confirm that they had biological children ages 6 to 12. This age-range was selected because children have completed most of their vaccinations by then, and would have been diagnosed with the common diseases of childhood if they were to develop them. The communications company Qualtrics (http://qualtrics.com) hosted the survey website. Mothers were asked to use their children's vaccination records to complete the online survey and to check off items from a list that pertained to them and their child or children, including pregnancy-related conditions and medications used, birth histories (including preterm birth—yes or no), vaccinations, physician-diagnosed illnesses, medications used by the child, and the use of health services. Vaccination was defined as receipt of one or more of the recommended vaccines. NDD, a derived outcome measure, was defined as a diagnosis of one or more of the following: Autism Spectrum Disorder (ASD), Attention Deficit Hyperactivity Disorder (ADHD), and a learning disability. Only closed-ended questions were used. The data were analyzed using SAS (Version 9.3) to determine the strength of associations between vaccination and health outcomes in terms of odds ratios (OR) and 95% Confidence Intervals (CI). Odds ratios describe the strength of the association between two categorical variables measured simultaneously and are appropriate measures of that relationship in a cross-sectional study [49].

Recruitment and informed consent

To increase trust and community engagement, homeschool organizational leaders were asked to sign Memoranda of Agreement and to provide the total number of member families. This was an attempt to interest and recruit as many homeschool families as possible. Non-responding organizations were sent a second email notice, but few provided the requested information. Follow-up calls to the leaders suggested that they had contacted their members about the study. The online survey remained open for three months in the summer of 2012. Financial incentives to complete the survey were neither available nor offered. The questionnaire required only factual, binary responses, allowing for rapid data entry, with the aim of improving both response and completion rates. The introductory letter and survey questions were stated in a neutral way so that, if biases existed, they would be evenly distributed across comparison groups. Our letter to parents began:

"Dear Parent, This study concerns a major current health question: namely, whether vaccination is linked in any way to children's long-term health. Vaccination is one of the greatest discoveries in medicine, yet little is known about its long-term impact. The objective of this study is to evaluate the effects of vaccination by comparing vaccinated and unvaccinated children in terms of a number of major health outcomes …"

Results

The survey yielded a sample of 666 children, of whom 261 (39%) were unvaccinated and 51 (7.7%) were born preterm. Fifty children (7.5%) had a neurodevelopmental disorder (NDD). The overall diagnostic category of NDD was created for analytic purposes in view of the relatively small number of children with the individual diagnoses, and because the diagnoses of LD, ADHD and ASD are closely related and overlapping [50]; that is, about 50% of children with ASD meet the diagnostic criteria for ADHD [51]. In this study the most common diagnoses were LD only (n = 15), ADHD only (n = 9), and ASD only (n = 9), followed by various combinations of these diagnoses (Figure 1).

Table 1 shows the characteristics of the survey respondents. Mothers averaged about 40 years of age, were typically white, college graduates, Christian and married, with household incomes between $50,000 and $100,000. The children as a group were similarly mostly white (88%), with a slight preponderance of females (52%). With regard to vaccination status, 261 (39%) were unvaccinated, 208 (31%) were partially vaccinated, and 197 (30%) had received all of the recommended vaccinations. All data analyses are based on these numbers.

Summary of the initial findings

To summarize the initial results of the study [48], vaccinated children were significantly less likely than the unvaccinated to have

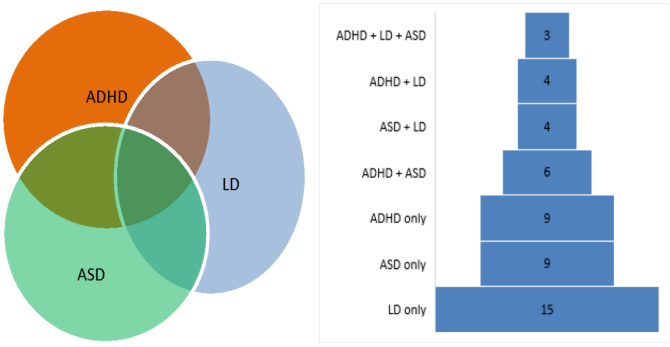


Figure 1. The overlap and distribution of physician-diagnosed neurodevelopmental disorders, based on mothers' reports

Table 1. Characteristics of the respondents [a]

	Mean (SD)[a]
Age (n=407)	40.59 (6.7)
	Number (%)[a]
Race	
White	382 (92.5)
Non-White	21 (7.6)
Total	413
Education	
High School Graduate or Less	35 (8.5)
Some College	114 (27.5)
College Graduate	187 (45.2)
Post-Graduates	78 (18.5)
Total	414
Total Gross Household Income	
< $49,999	123 (30.8)
$50,000-100,000	182 (45.5)
> $100,000	95 (23.8)
Total	400
Religious Affiliation	
Christianity	375 (91.2)
Non-Christianity	36 (8.8)
Total	411
Marital Status	
Married	386 (93.7)
Not Married	26 (6.3)
Total	412

[a] Missing observations are excluded.

been diagnosed with chickenpox and pertussis, but significantly more likely to have been diagnosed with pneumonia, otitis media, allergies and NDD (that is, ASD, ADHD, and/or a learning disability). The vaccinated were also more likely to use allergy medication, to have had myringotomies with tube placement, visited a doctor for a health problem in the previous year, and been hospitalized at some time in the past.

The second aim of the study was to determine whether the association found between vaccination and NDD remained significant after controlling for other measured factors. The factors associated with NDD in unadjusted regression analyses were vaccination (OR 3.7, 95% CI: 1.7, 7.9), male gender (OR 2.1, 95% CI: 1.1, 3.8), adverse environment (defined as living within 1-2 miles of a furniture manufacturing factory, lumber factory, waste site or landfill) (OR 2.9, 95% CI: 1.2, 7.4), maternal use of antibiotics in pregnancy (OR 2.3, 95% CI: 1.1, 4.8), and preterm birth (OR 4.9, 95% CI: 2.4, 10.3). After adjustment, the factors that remained significantly associated with NDD were vaccination (OR 3.1, 95% CI: 1.4, 6.8), male gender (OR 2.3, 95% CI: 1.2, 4.3), and preterm birth (OR 5.0, 95% CI: 2.3, 11.6). However, in a final adjusted model with interaction, vaccination, but not preterm birth, remained associated with NDD, while the interaction of preterm birth and vaccination was associated with a 6.6-fold increased odds of NDD (95% CI: 2.8, 15.5), suggesting a synergistic effect.

As noted, initial bivariate analyses suggested that NDD was significantly associated both with preterm birth (OR 4.9, 95% CI: 2.4, 10.3) and vaccination (OR 3.7, 95% CI: 1.7, 7.9), and more strongly so with preterm birth than vaccination (see Table 2, below). Vaccination was also significantly associated with the specific diagnoses of learning disability (OR 5.2, 95% CI: 1.5, 17.5), ADHD (OR 4.3, 95% CI: 1.2, 14.5), Autism Spectrum Disorder (OR 4.3, 95% CI: 1.2, 14.5) as well as the combined diagnosis of NDD (OR 3.7, 95% CI: 1.7, 7.9) (Table 3, below).

These findings suggested that preterm birth and vaccination could be independent risk factors for NDD. However, since most preterm infants receive the same vaccinations on the same schedule as those recommended for term infants, interpretation of the findings remains unclear.

Interactions between preterm birth, vaccination and NDD

Given the apparent synergism in the combination of vaccination and preterm birth on NDD found in regression analyses [48], and the relatively small numbers available for analysis, this report presents additional findings on the association and interaction between preterm birth (yes, no), vaccination status (yes, no) and NDD (yes, no). Stratified analyses with Odds Ratios and 95% Confidence Intervals were used to quantify the strength of the associations. The analytic process can be envisioned as a 2 X 2 table with the cells in Row 1 labeled A and B, and those in Row 2 labeled C and D. The odds ratio is calculated by the formula AD/BC. Here we examine different combinations of preterm birth and vaccination and their association with NDD. Calculations based on the data in Table 4 (below) suggest a complex picture of the association between prematurity, vaccination and NDD.

Most children in the sample were vaccinated and not preterm (n=367; 55%). The second largest group comprised those who were neither vaccinated nor preterm (n=249; 37%); the third largest comprised those who were preterm and vaccinated (n=37; 6%), and the smallest group those who were preterm and unvaccinated (n=12; 2%).

While acknowledging that our study group is relatively small and not a representative sample of U.S. children, the following observations are noted. First, of the 37 children who were both preterm and vaccinated (P/V), 12 (32%) had an NDD, consistent with studies suggesting that a high proportion of preterm infants are later diagnosed with ASD [18-20]. Second, of the 12 children who were preterm but unvaccinated (P/V-), none had an NDD. Third, among the 367 children

Table 2. Unadjusted associations between preterm birth, vaccination, and neurodevelopmental disorders (NDD)

	Neurodevelopmental Disorders		**Chi-Square**	**OR (95% CI)**	**P-value**
	Yes n (%)[a]	No n (%)[b]			
Preterm birth					
Yes	12 (24.0)	37 (6.0)	22.9	4.9 (2.4-10.3)	<0.001
No	38 (76.0)	578 (93.8)			
Vaccination Status					
Yes	42 (84.0)	363 (58.9)	12.2	3.7 (1.7-7.9)	< 0.001
No	8 (16.0)	253 (41.1)			

[a]Values in parentheses indicate % of total (n=50).
[b]Values in parentheses indicate % of totals for respective factors.

Table 3. Vaccination status and types of NDD

Condition	**Vaccination Status**	**n (%)**	**OR (95% CI)**	**P-value***
ADHD	Vaccinated (N=405)	19 (4.69)	4.3 (1.3, 14.5)	0.013
	Unvaccinated (n=261)	3 (1.15)		
ASD	Vaccinated (N=405)	19 (4.69)	4.3 (1.2, 14.5)	0.013
	Unvaccinated (n=261)	3 (1.15)		
Learning Disability	Vaccinated (N=405)	23 (5.68)		
	Unvaccinated (n=261)	3 (1.15)	5.2 (1.5, 17.5)	0.003
Any NDD	Vaccinated (N=405)	42 (10.37)	3.7 (1.7, 7.9)	0.005
	Unvaccinated (n=261)	8 (3.03)		

* From Fisher's exact test.

Table 4. Preterm birth and vaccination status among children with NDD and controls

Neurodevelopmental Disorders					
	Yes		No		
	Preterm	Not Preterm	Preterm	Not Preterm	Totals
Vaccinated	12	30	25	337	404
Not vaccinated	0	8	12	241	261
Totals	12	38	37	578	665

in the sample who were not preterm but vaccinated (P-/V), 30 (8%) had an NDD. Fourth, of the 249 children who were neither preterm nor vaccinated (P-/V-), only 8 (3%) had an NDD. These observations suggest that birth history strongly affects the likelihood of NDD in vaccinated children, given that the combination of preterm birth and vaccination accounts for a substantial proportion of NDD diagnoses in the overall sample. In contrast, cases of NDD are under-represented in preterm but unvaccinated children. These suggestions are further supported and amplified by analyses of the odds of NDD given various combinations of preterm birth and vaccination (see Table 5).

Table 5 shows the following:

1) Preterm birth without vaccination (P/V-) was not associated with NDD.

2) Term birth with vaccination (P-/V) was associated with a significant 2.7-fold increase in the odds of NDD.

3) Preterm birth with vaccination (P/V) was associated with a significant 5.4-fold increase in the odds of NDD compared to the odds of NDD given term birth and vaccination (P-/V).

4) Preterm birth with vaccination (P/V) was associated with a nonsignificant 12.3-fold increased odds of NDD compared to preterm birth without vaccination (P/V-) (not technically significant because no child in the sample with an NDD was both preterm and unvaccinated).

5) Preterm birth with vaccination (P/V) was associated with a significant 14.5-fold increased odds of NDD compared to being neither preterm nor vaccinated (P-/V-).

Figure 2 depicts the findings reported in Table 5. In summary, the results suggest that: preterm birth without vaccination is not associated with NDD; vaccination (in a term infant) is associated with a 2.7-fold increased odds of NDD, compared to an unvaccinated child born at term (P-/V-); and the combination of preterm birth and vaccination (P/V) is associated with progressively increased odds of NDD (depending on birth history and vaccination status), with a 5.4-fold increase compared to vaccination alone, a 12.3-fold (nonsignificant) increase compared to preterm birth alone (*i.e.,* unvaccinated), and a 14.5-fold increase compared to term birth without vaccination. These results (depicted in Figure 2, below) suggest that preterm birth without vaccination is not associated with NDD whereas vaccination is (regardless of birth history), and that vaccination coupled with preterm birth greatly increases the odds of NDD compared to vaccination alone.

Discussion

Prematurity is a known risk factor for neurodevelopmental disorders, with 8% to 27% of extremely preterm infants showing symptoms of autism at 6 years of age [20]. Late preterm infants are also reportedly more prone to long-term neurologic sequelae than previously recognized [16,17]. Premature infants typically receive the same doses of recommended vaccines and on the same schedule as term infants, yet the possible role of vaccination in preterm-associated NDD has not been investigated. A unique feature of this study is that it included birth history and pregnancy experiences, based on mothers' reports. The present analysis focused on the association and possible interaction of preterm birth (yes, no) and vaccination (yes, no) in relation to NDD (yes, no).

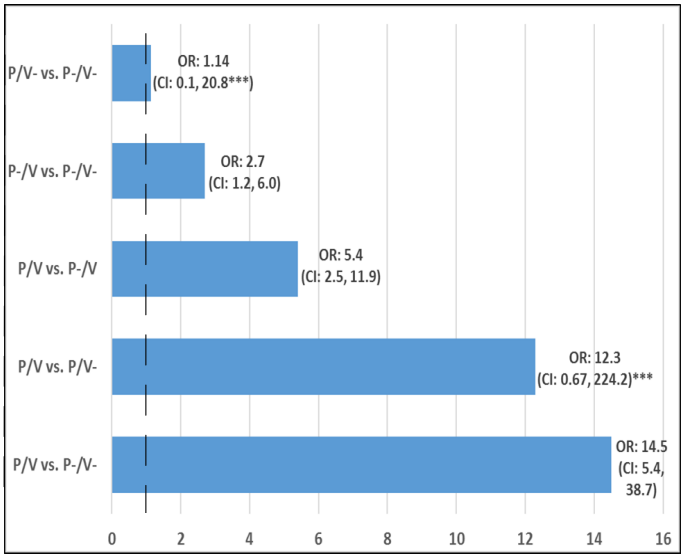
In our initial report [48], logistic regression analyses revealed that both preterm birth and vaccination (receipt of one of more of the recommended vaccines) were significantly associated with NDD after controlling for other factors, suggesting independent effects. However, in a final regression model with interaction, preterm birth combined with vaccination was associated with a 6.6-fold increased odds of NDD, suggesting a synergistic effect.

This second report focused on the association between preterm birth, vaccination and NDD. Because of the relatively small numbers, stratified analyses with Odds Ratios and 95% Confidence Intervals were used to examine and quantify different combinations of preterm birth and vaccination in relation to NDD. Vaccination was found to be significantly and independently associated with NDD, whereas preterm birth without vaccination was not. However, vaccination

Table 5. Association between combinations of preterm birth and vaccination status, and neurodevelopmental disorders (NDD)

Combinations of Exposures	NDD		OR (95% CI)	P-value*
	Yes	No		
1. Preterm and Unvaccinated (P/V-)	0	12	1.14 (0.1, 20.8)**	1
Not Preterm and Unvaccinated (P-/V-)	8	241		
2. Not Preterm and Vaccinated (P-/V)	30	337	2.7 (1.2, 6.0)	0.012
Not Preterm and Unvaccinated (P-/V-)	8	241		
3. Preterm and Vaccinated (P/V)	12	25	5.4 (2.5, 11.9)	< 0.001
Not Preterm and Vaccinated (P-/V)	30	337		
4. Preterm and Vaccinated (P/V)	12	25	12.3 (0.67, 224.2)**	0.024
Preterm and Unvaccinated (P/V-)	0	12		
5. Preterm and Vaccinated (P/V)	12	25	14.5 (5.4, 38.7)	< 0.001
Not Preterm and Unvaccinated (P-/V-)	8	241		

* From Fisher's exact test.
** Calculated by adding 0.5 to each cell because of zero count.



*P = preterm; P- = not preterm; V = vaccinated; V- = not vaccinated.
**CI = Confidence Interval.
***Calculated by adding 0.5 to each cell because of zero count.

Figure 2. Combinations of Preterm birth and Vaccination Status* and the Odds Ratio of Neurodevelopmental Disorders with 95% CI**

coupled with preterm birth greatly increased the odds of NDD over that of vaccination alone, especially compared to being born at term and unvaccinated, suggesting that vaccination may adversely affect neurodevelopmental outcomes in preterm infants. Indeed, the apparent synergism between preterm birth and vaccination in regard to NDD suggests the possibility that rising rates of NDD could be driven in part by the vaccination of growing numbers of late preterm newborns, the latter of which account for 74% of all preterm birth and about 8% of total births [53].

While additional studies are needed to verify and explain the present findings, a tentative hypothesis of the mechanisms linking preterm birth and vaccination with NDD is outlined as follows. Receipt of one or more vaccines could precipitate NDD in some preterm infants by exacerbating a preexisting inflammatory state associated with prematurity, leading to hepatic encephalopathy and hypoxic-ischemic brain damage. Impaired liver function is a predisposing factor for preterm birth [54,55] and the latter is associated with increased risks of hypoxic-ischemic brain injury [56]. A possible biochemical basis for vaccination-associated NDD in preterm infants could involve the spillage of a membranolytic biliary metabolite from the maternal liver into the circulation and its transfer to the fetus, contributing thereby to the pathogenesis of preterm birth itself [55] and possibly being further increased to neurotoxic concentrations by the impact of vaccination on the infant's liver. Consistent with this hypothesis, liver dysfunction is reported as an adverse effect of vaccination [57] and as a feature of children with autism [58,59]. Furthermore, hyperbilirubinemia is associated with hypoxic-ischemic brain damage [60] and is a feature of the preterm infant as well as children with later-onset cognitive disorders and ASD [61,62].

Strengths and limitations

Strengths of the study include: the relatively large sample of unvaccinated children (N=261); the demographically homogeneous sample of mainly white, higher-income and college-educated homeschooling families, in which the children studied were 6 to 12 years of age; and the recruitment of biological mothers as respondents, which made it possible to test hypotheses about the role of pregnancy-related factors, birth history and vaccination in specific conditions. Contact information on the families and the membership of homeschool organizations was unavailable, which meant that neither the number of homeschool families contacted nor the survey response rate could be determined. However, the study was not intended to be a representative survey of homeschool children but to achieve a sample of sufficient size to test for significant differences in outcomes. Homeschoolers were targeted for the study because their vaccination completion rates are lower than those of children in general. Although respondents were self-selected, efforts were made to reduce or eliminate biases of several kinds. To minimize recall bias, respondents were asked to use their child's vaccination records; to enhance reliability, closed-ended questions were used; to enhance validity, parents were asked to report only physician-diagnosed illnesses.

Study limitations include the relatively small number of preterm children and children with NDD. To preserve anonymity, the reported health outcomes could not be validated by clinical records. However, self-reports are accepted as a valid proxy for official records when the latter are unavailable [63]. A further potential limitation is that morbidity may have been under-estimated in unvaccinated children, as they were less likely than the vaccinated to see a physician for a routine checkup in the past year (57.6% vs. 37.2%, $p < 0.001$; OR 2.3, 95% CI: 1.7, 3.2). This could be due to the fact that such visits usually involve vaccinations, which non-vaccinating families would be expected to refuse. However, unvaccinated children were more likely than the vaccinated to be diagnosed with chickenpox and pertussis, which would have involved one or more visits to the pediatrician. This suggests that observed differences in health outcomes were not due to under-ascertainment of disease.

Conclusions

This study compared the birth histories and health outcomes of vaccinated and unvaccinated children and sought to determine the association, if any, between vaccination, preterm birth and neurodevelopmental disorders (NDD). Vaccination (*i.e.,* receipt of one of more of the recommended vaccines) was significantly associated with NDD, while preterm birth without vaccination was not. Preterm birth coupled with vaccination, however, was associated with a synergistic increase in the odds of NDD, suggesting the possibility that vaccination could precipitate adverse neurodevelopmental outcomes in preterm infants. These results provide clues to the epidemiology and causation of NDD but question the safety of current vaccination programs for preterm infants. Further research is needed to validate and investigate these findings in order to optimize the impact of vaccines on children's health.

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Conflict of interest

The first author has advocated for further research on the health outcomes of routine vaccination. None of the authors has a financial conflict of interest to declare.

Author contributions

ARM designed the study, contributed to data analysis and interpretation, and wrote successive drafts of the paper. AB contributed to data analyses and edited the paper. BJ contributed to data analyses and editing. BR designed the study, contributed to data collection, and edited the paper. All authors read and approved the final version of the paper.

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Disclaimer

This study, approved by the Institutional Review Board of Jackson State University, was completed prior to Dr. Mawson's tenure-track appointment at Jackson State University.

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