

BMJ Open Protocol for Pertussis Immunisation and Food Allergy (PIFA): a case-control study of the association between pertussis vaccination in infancy and the risk of IgE-mediated food allergy among Australian children

Marie J Estcourt,^{1,2} Julie A Marsh,^{1,3} Dianne E Campbell,^{4,5} Michael S Gold,⁶ Katrina J Allen,^{7,8} Peter Richmond,^{1,9} Claire S Waddington,^{1,9} Thomas L Snelling^{1,10}

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For numbered affiliations see end of article.

Correspondence to

Dr Thomas L Snelling;
Tom.Snelling@telethonkids.org.au

ABSTRACT

Introduction Atopic diseases, including food allergy, have become a predominant cause of chronic illness among children in developed countries. In Australia, a rise in hospitalisations among infants coded as anaphylaxis to foods coincided with the replacement of whole-cell pertussis (wP) vaccine with subunit acellular pertussis (aP) vaccine on the national immunisation schedule in the late 1990s. Atopy is characterised by a tendency to mount T helper type 2 (Th2) responses to otherwise innocuous environmental antigens. Compared with infants who receive aP as their first pertussis vaccine, those who receive wP appear less likely to mount Th2 immune responses to either vaccine or extraneous antigens. We therefore speculate that removal of wP from the vaccine schedule contributed to the observed rise in IgE-mediated food allergy among Australian infants.

Methods and analysis This is a retrospective individually matched case-control study among a cohort of Australian children born from 1997 to 1999, the period of transition from wP to aP vaccines; we include in the cohort children listed on Australia's comprehensive population-based immunisation register as having received a first dose of either pertussis vaccine by 16 weeks old. 500 cohort children diagnosed as having IgE-mediated food allergy at specialist allergy clinics will be included as cases. Controls matched to each case by date and jurisdiction of birth and regional socioeconomic index will be sampled from the immunisation register. Conditional logistic regression will be used to estimate OR ($\pm 95\%$ CI) of receipt of wP (vs aP) as the first vaccine dose among cases compared with controls.

Ethics and dissemination The study is approved by all relevant human research ethics committees: Western Australia Child and Adolescent Health Services (2015052EP), Women's and Children's Hospital (HREC/15/WCHN/162), Royal Children's Hospital (35230A) and Sydney Children's Hospital Network (HREC/15/SCHN/405). Outcomes will be disseminated through publication and scientific presentation.

Trial registration number NCT02490007.

Strengths and limitations of this study

- The transition from whole-cell pertussis (wP) to acellular pertussis (aP) in Australia represents a natural experiment of the effect of infant pertussis vaccination on subsequent food allergy on a scale not achievable by a randomised controlled trial.
- The study is nested within Australia's comprehensive, prospective, population-based immunisation register, meaning that ascertainment of vaccination status is likely to be accurate, and any misclassification is likely to be non-differential for cases and controls.
- We included only cases diagnosed as IgE-mediated food allergy in specialist tertiary allergy clinics.
- Not all children diagnosed with IgE-mediated food allergy had a formal food challenge, the gold standard for diagnosis.
- We assume that the probability of receipt of wP as the first pertussis vaccine dose (rather than aP) was dependent on date and jurisdiction of birth, but independent of any other risk factors for food allergy. It is not possible to verify if this assumption is valid.

INTRODUCTION

Epidemiology of atopic disease in Australia and internationally

As a group, atopic diseases (eczema, asthma, rhinoconjunctivitis and food allergy) are now the most prevalent chronic diseases of children in resource-rich industrialised countries; at least one in four Australian children is affected by these conditions.¹ Whereas the incidence of aeroallergic atopic diseases (asthma and rhinoconjunctivitis) began rising several decades ago and peaked in Australia in the 1980s,² since the late 1990s a 'second wave' of non-aero allergic atopy has emerged characterised by severe IgE-mediated food

allergies in children.³ For example, one Australian allergy clinic observed a 12-fold rise in consultations for food allergy in children between 1995 and 2006.⁴ Fatal food anaphylaxis rose almost 10% per year from 1997 to 2013.⁵ In the most comprehensive study of its kind, the prevalence of food sensitisation among 12-month-old Victorian infants was 18% (95% CI 17% to 19%), with 10% having challenge-proven food allergy.⁶ The most common allergies were to peanut 3.0%, raw egg 8.9% and sesame 0.8%. The prevalence of allergic eczema among infants was found to be even higher, 26.7% (95% CI 25.0% to 28.4%).⁶ A similar phenomenon has also been described in the UK⁷ and the USA.^{8,9} A characteristic feature of this epidemic has been the onset of symptoms in early infancy and persistence of symptoms into adolescence.³

The rise in atopic disease and sudden rise in food allergy, in particular, suggest one or more causal environmental triggers. It has been widely suggested that lifestyle change, in particular declining exposure to infection—the hygiene hypothesis¹⁰—is responsible. However, this does not by itself explain the more abrupt onset of food-related allergy in Australia.¹¹

A temporal association exists between the onset of the epidemic of food allergies in Australia and the transition from the use of vaccines containing the whole-cell pertussis (wP) antigen to those containing the acellular pertussis (aP) antigen. Use of aP began to replace that of wP in the mid-1990s and by early 1999; most scheduled childhood pertussis vaccines were aP.¹² In the USA and in the UK, a temporal association between the phasing out of wP vaccines and the increase in hospitalisations for food allergy is not clear (unpublished data, personal communication) although coding of hospitalisation records for allergies may be both insensitive and non-specific so any associations could be easily obscured. In light of the contrasting immunological effects of wP compared with aP (reviewed below), it is plausible that a causal relationship underlies the ecological association observed in Australia.

Immunological basis of atopic disease

A reciprocal relationship exists between T helper cell type 1 and type 2 (Th1 and Th2) immunity due to cross-regulation of their respective effector cell populations.¹³ The balance between Th1 and Th2 is established during early infancy. Atopy is caused by a dysregulation of this balance in the developing immune system, characterised by an immune phenotype that is heavily biased towards Th2 immunity or ‘Th2 polarised’, the consequent overproduction of IgE to one or more allergens, and IgE-mediated inflammation.

The developing immune system appears most susceptible to Th2 polarisation in the critical early months after birth. This period is pivotal in the transition from the Th1-suppressed/Th2-dominant phenotype needed to avoid rejection in utero, to a more Th1/Th2-balanced phenotype. Newborns are exposed to an array of new antigenic proteins from infection and other natural

environmental exposures, including gut flora and food components. The development of tolerance to these natural exposures represents an early challenge to the developing immune system. It appears that the development of tolerance can be influenced by a range of factors, which in turn modify the risk of food allergy. The best studied of these are optimal bacterial colonisation, breast milk, prebiotics, vitamins and polyunsaturated fatty acids¹⁴; however, so far no broadly effective strategies have been identified to promote the natural development of Th1/Th2 balance or to prevent the development of food-related and other allergies in infants.

Immune profile of pertussis vaccines

Th1 responses are needed for clearing pertussis infection.¹⁵ wP stimulates these adaptive responses via the presence of bacterial cell wall components which stimulate the Th1 immune pathway.¹⁶ Stimulation of the Th1 pathway also results in local and systemic adverse vaccine reactions, which was the driving reason for phasing out use of wP. In contrast, aP typically induces strong Th2 responses^{15–17} with the production of antigen-specific antibodies. While this aP response provides immediate antibody-derived protection from disease, the absence of Th1 stimulation may skew the developing immune response towards one characterised by Th2 responses. Children who receive at least one dose of wP in infancy appear better protected against pertussis than children who receive aP only.¹⁸ The most recent meta-analysis of vaccine efficacy comparing wP and aP estimates efficacy at 94% (95% CI 88% to 97%) and 84% (95% CI 81% to 87%), respectively.¹⁹ Moreover, a number of studies detail the Th2 immune bias induced in some infants who received aP-only schedules, resulting in excessive IgE production against vaccine antigens.^{20,21} A recent study has shown that the Th polarisation induced by infant immunisation can persist into adolescence and adulthood and is maintained after booster vaccination.²² This could be due to the combined effect of: (1) carry over of the Th2-biased in utero phenotype, (2) the presence of alum and pertussis toxin, which have Th2-adjuvanting properties and (3) the absence of the balancing Th1-stimulating ligands present in wP. These effects manifest especially among children with evidence of an underlying Th2-skewed phenotype.^{20,21}

Broader immunomodulating properties of pertussis

The Th2 polarising effect of the initial dose of aP appears to extend beyond vaccine-specific responses; there may be a significant ‘bystander’ effect with upregulation of circulating IgE to a broad range of antigens following subsequent doses of aP. Importantly, these Th2-stimulatory effects appear to extend to food allergens,²³ especially in early infancy when immune memory against allergens is most susceptible to programming.^{24,25} The administration of additional pertussis vaccine doses in later childhood in children primed with aP only is associated with frequent injection-site reactions. In Australia, this contributed to the removal of the 18-month-old

pertussis booster given in 2003, which in turn led to reduced protection against pertussis among preschool aged children.²⁶ These adverse reactions have been linked to the presence of high levels of vaccine-specific, Th2-polarised, Th-memory cells.²⁷ Children primed with aP only also exhibit high titres of total IgE, including tetanus (T)-specific^{28 29} and pertussis-specific IgE.²⁸ These responses are rarely observed among children who have received wP-containing vaccines, including those who have received mixed vaccine schedules of wP and aP.³⁰ A recent study of a birth cohort examined the effect of delaying aP vaccination on atopic outcomes. This study found that delay of aP by 1 month resulted in a reduction in eczema in infants and use of eczema medications.³¹ To date, no other studies have found a relationship between aP vaccines and allergy. A study from the 1990s found no significant difference in overall rates of skin prick test (SPT) reactivity or allergic disease among young Swedish children who received wP compared with those who received aP.³² In that study population, the frequency of food allergy was low (~2%) compared with that observed among contemporary Australian infants. A more recent prospective birth cohort of children from the Isle of Wight also failed to find any difference in the frequency of atopic outcomes among children receiving wP versus aP.³³

Based on these observations, we speculate that removal of wP from the infant vaccine schedule has contributed to the observed rise in IgE-mediated food allergy among Australian infants.

AIM

To assess the possible food allergy-preventive benefit of using wP compared with aP for pertussis vaccination in childhood.

OBJECTIVES AND OUTCOME MEASURES

Primary objective

To determine if Australian children born between 1997 and 1999 (inclusive) who received wP as their first pertussis vaccine dose in infancy were less likely to subsequently develop IgE-mediated food allergy compared with contemporaneous children who received aP as their first pertussis vaccine dose.

Secondary objectives

1. To determine if Australian children born in the years 1997 to 1999 (inclusive) who received at least one dose of a wP vaccine at any age were less likely to subsequently develop IgE-mediated food allergy compared with contemporaneous children who received only aP vaccines.
2. To determine if Australian children born in the years 1997 to 1999 (inclusive) who received wP vaccines exclusively were less likely to subsequently develop IgE-mediated food allergy compared with

contemporaneous children who received only aP vaccines.

Prespecified hypotheses

Primary hypothesis

Among Australian children with documented evidence of receiving a pertussis vaccine before age 16 weeks, a record of receipt of a wP vaccine as dose one is less common than aP vaccine among those subsequently diagnosed with food allergy compared with date of birth, socioeconomic index and jurisdiction-matched cohort controls.

Secondary hypothesis I

Among Australian children with documented evidence of receiving a pertussis vaccine before age 16 weeks, a record of receipt of at least one dose of wP vaccine at any age is less common than aP vaccine at all ages among those subsequently diagnosed with food allergy compared with date of birth, socioeconomic index and jurisdiction-matched cohort controls.

Secondary hypothesis II

Among Australian children with documented evidence of receiving a pertussis vaccine before age 16 weeks, a record of receipt of one or more wP vaccine doses exclusively (ie, no aP vaccine) is less common than one or more aP vaccine doses exclusively (ie, no wP vaccines) among those subsequently diagnosed with food allergy compared with date of birth, socioeconomic index and jurisdiction-matched cohort controls.

METHODS AND ANALYSIS

Study design

This is a retrospective individually matched case-control study of Australian children born during the period of transition from use of wP-containing pertussis vaccines to aP-containing pertussis vaccines (year of birth 1997–1999 inclusive) and who are registered on the Australian Immunisation Register (AIR; prior to 2016 known as the Australian Childhood Immunisation Register) and who have received their first dose of pertussis vaccine before age 16 weeks. Cases are drawn from private allergy clinics and allergy clinics associated with tertiary teaching hospitals around Australia. Five hundred children identified as having IgE-mediated food allergy, from reviewing case histories against standardised inclusion criteria, will be enrolled. Cohort controls will be drawn from a deidentified database of the AIR held by the National Centre for Immunisation Research and Surveillance of Vaccine Preventable Diseases. Each case will be individually matched with up to 10 cohort controls based on year of birth, socioeconomic index and jurisdiction.

Project coordination and epidemiological analysis is conducted from the Wesfarmers Centre of Vaccines and Infectious Diseases at the Telethon Kids Institute Western Australia.

Primary outcome measure

The primary outcome is IgE-mediated food allergy diagnosed by a registered specialist paediatric allergist.

Definition of exposure of interest (vaccination)

For the primary analysis, exposure is defined as AIR-documented receipt of either a wP-containing vaccine or receipt of an aP-containing vaccine as the first vaccine against pertussis and given before age 16 weeks (strictly <112 days) irrespective of any subsequent pertussis vaccinations.

For the secondary analyses, exposure is defined as either:

1. AIR-documented receipt of *one or more* doses of wP at any age or only aP-containing vaccines for all pertussis vaccinations. All other sequences of vaccines will be coded as non-applicable and excluded from the analysis.
2. AIR-documented receipt of *only* wP-containing vaccine for all pertussis vaccinations or *only* aP-containing vaccines for all pertussis immunisations. All other sequences of vaccines will be coded as non-applicable and excluded from the analysis.

Vaccination status for each case and its corresponding matched controls is referenced from the age of allergy diagnosis in the case.

Study setting

Cases are drawn from private allergy clinics and from allergy clinics associated with tertiary teaching hospitals in the Australian states of New South Wales (NSW), Victoria, South Australia and Western Australia; matched cohort controls are selected from among children registered as resident in the same state and recorded as having received at least one pertussis vaccine dose on the population-based vaccine register (AIR).

Participant identification

Eligibility criteria

All cases and cohort controls must be registered on AIR as having had a first dose of any pertussis-containing vaccine before age 16 weeks and during the period in which the transition from wP to aP vaccine occurred: 1 January 1997 to 31 December 1999.

Overall description of trial participants

To maximise the study efficiency in the case, identification has been concentrated among children born when aP accounted for between 25% and 75% of infant vaccine doses administered. The changeover from wP to aP occurred with slightly different timing in the different jurisdictions of Australia so the exact dates vary across jurisdictions. In the first instance, cases have been identified from among clinic attendees in NSW, Victoria, Queensland and Western Australia who are AIR-registered and born from 1 October 1998 to 30 June 1999. For South Australia, cases have been drawn from AIR-registered children born from 1 July 1997 to

30 October 1998 owing to the earlier uptake of aP in that state.

Five hundred cases in total will be enrolled. If fewer than 500 cases are identified from among children born within the initial birthdate range, CI will expand this birthdate range for NSW, Victoria, Queensland and Western Australia to 1 June 1998–30 October 1999; for South Australia, the birthdate range will be expanded to 1 April 1997–30 April 1999. If there are still insufficient eligible cases identified, the birthdate range will be progressively increased by 3-month increments in each direction for all jurisdictions until 500 cases are identified or until the limits of 1 January 1997 and 31 December 1999 are reached (whichever occurs first).

Case definition

Cases are considered to have IgE-mediated food allergy on the basis of (1) a documented history of consistent clinical symptoms following ingestion of an implicated food and (2) evidence of sensitisation to that food via either positive SPT or elevated-specific IgE to the implicated food, with onset after the first pertussis-containing vaccine but before age 15 years.

To meet the case definition of IgE-mediated food allergy, the case must satisfy BOTH:

The clinical notes or a clinical letter arising from the allergy consult explicitly documents the presence of one or more of the following features:

1. urticaria
2. angioedema
3. emesis
4. vocal hoarseness
5. persistent cough
6. wheeze
7. stridor
8. collapse
9. hypotension

with onset of at least one feature within 1 hour of ingestion of the suspected food where this can reasonably be inferred from statements such as 'immediate' or 'within x minutes' where x is <60

AND

Documented evidence of allergic sensitisation to the implicated food through EITHER:

1. specific IgE positive to suspected food (serum-specific IgE >0.35 kU/L)
2. positive SPT to suspected food (weal diameter >3 mm) where evidence of sensitisation must be at the time of consultation or within 6 months after the clinical encounter.

Other acceptable terms:

When reviewing case histories, the below terms are considered synonymous:

1. urticaria—hives, rash, welts
2. angioedema—oedema, swelling of lips or eyes
3. emesis—vomit or vomiting
4. vocal hoarseness—horse voice, raspy voice
5. persistent cough

6. wheeze
7. stridor
8. collapse—faint, loss of consciousness
9. hypotension—low blood pressure.

The case definition has been agreed on by specialist allergists associated with the study as consistent with international expert consensus for the definition of IgE-mediated food allergy.

Determination of exposure (vaccination)

The primary exposure of interest for cases and cohort controls is the first received pertussis vaccine as recorded on the AIR. The Australian vaccination schedule recommends three sequential priming vaccines against pertussis at approximately 2, 4 and 6 months of age. Cases and controls will have either received aP or wP together with T and diphtheria, either with or without hepatitis B as part of commercially combined vaccine preparations. All children will have been eligible for booster doses of aP at age 18 months old and at age 4 years old.

Ethics and dissemination

A waiver of consent was sought and approved on the basis that the study poses negligible risk to participants and that seeking individual consent for access to data was unfeasible and may lead to ascertainment bias.

STATISTICS

Sample size and power considerations

A study involving 500 sets of cases and controls, with 10 matched controls for each case, has 80% power to detect a 23% lower risk (OR 0.77) of food allergy among children who received wP as their first dose of pertussis vaccine compared with those who received aP as their first dose. This assumes that: (1) 50% of cohort controls receive a first pertussis vaccine dose of wP on average, (2) the correlation coefficient for exposure (first dose of wP) between cases and matched controls is 0.5 and (3) a two-sided significance level of 5%. We believe that a smaller effect size will not influence vaccine policy.

Bias

Management of confounding

Since routine vaccines in Australia are delivered almost exclusively via the National Immunisation Programme, date of birth (and therefore date of vaccination) and jurisdiction are considered to be the only relevant factors associated with vaccine type received (wP vs aP) and, therefore, the only relevant potential confounders. We will minimise confounding by these factors by direct matching. In so far as the *type* of vaccine received is expected to be independent of ethnicity, sex, family size, birth order, pet ownership and all other factors putatively or known to be related to food allergy, there are unlikely to be any other relevant confounders of the association between vaccine type and allergy; inability to match or

adjust for other factors poses negligible threat to study validity.

Minimising information bias

The case definition is intended to be pragmatic and yet specific for IgE-mediated food allergy, excluding many non-allergic food reactions potentially misclassified as allergy by non-specialists and in-hospital coding. For completeness, we will conduct an a priori sensitivity analyses which will include as cases only those children meeting the case definition who also have (1) challenge-proven food allergy and (2) evidence of sensitisation at or higher than the following levels: SPTs: 8 mm for cow milk, 7 mm for egg and 8 mm for peanut or IgE serum responses: 15 kU/L for cow milk, 7 kU/L for egg and 14 kU/L for peanut. These are the documented levels for 95% positive predictive value (PPV) as defined by Sporik *et al*³⁴ and Sampson.³⁵ Ascertainment of vaccination status will be from the AIR for both cases and controls and will not rely on either parental recall or recording in medical records. The AIR record of cases will only be ascertained after verification from the site Principle Investigator (PI) that the case definition is fully met. Children with food allergy will be excluded as cases if they are not registered on AIR. There is no reason to expect that the accuracy or completeness of AIR should be different for cases and controls, so any inaccuracy is likely to be non-differential.

Minimisation of selection bias

Cases are sampled only from among children presenting to specialist-led private allergy clinics and allergy clinics at tertiary paediatric centres. To ensure correct classification of cases, we will not sample cases from non-tertiary Australian hospitals, from cases diagnosed and managed by non-specialists or those without confirmation of sensitisation. We nonetheless expect cases will be generally representative of all Australian children with true IgE-mediated food allergy in the birth cohort. Cohort controls will be sampled from date of birth, jurisdiction and socioeconomic index-matched children from the AIR, a comprehensive population-based register of all Medicare-registered children in Australia. Since cohort controls are sampled at random, they will provide an unbiased estimate of the vaccine status of the baseline source population for each case by date of birth, jurisdiction and Index of Relative Socio-economic Advantage and Disadvantage (IRSAD) score (Australian Bureau of Statistics (ABS) -assigned socioeconomic index by postcode).

Description of statistical methods

The study population characteristics will be summarised by case or control status using frequency and proportion for categorical or binary variables, means and SDs for symmetric continuous distributions and medians and IQRs for asymmetric distributions. Conditional logistic regression will be used to perform hypothesis tests of the association between pertussis vaccine type (wP or aP) and IgE-mediated food allergy. Results will be summarised

using ORs and presented with associated 95% CIs. Since controls will be sampled from the AIR irrespective of past or future case status, the OR will be considered to be an unbiased estimator of the relative risk of food allergy among children receiving wP compared with aP vaccine.

Analysis of outcome measures

Primary analysis population

All case and control individuals recorded on the AIR as receiving at least one dose of pertussis vaccine before age 16 weeks will be included in the primary analysis, irrespective of whether they received any further doses of pertussis vaccine.

Primary analysis

Conditional logistic regression will be used to evaluate the association between receipt of wP versus aP as the first pertussis vaccine and diagnosis of IgE-mediated food allergy. Direct matching will be on date of birth (± 7 days), jurisdiction (Australian state or territory) and IRSAD decile of the most recent Medicare-listed postcode (1st to 10th); no factors or interactions will be adjusted for in the a priori analysis.

For the primary analysis, a child's exposure will be coded as *either*:

aP1: first pertussis vaccine dose of aP, with subsequent doses either wP or aP or none

or

wP1: first pertussis vaccine dose of wP, with subsequent doses either wP or aP or none.

The comparison of primary interest will be between aP1 and wP1 vaccinated children.

Secondary analysis population

As for the primary analysis population, however, the comparison of wP-only and aP-only vaccinated children (secondary analysis II) will exclude any children who received a mixture of aP and wP vaccines.

Secondary analysis

As for the primary analysis to evaluate the association between pertussis vaccines received (wP only or mixed wP/aP in any combination, compared with aP only) and IgE-mediated food allergy.

For secondary analyses: (1), a child's exposure will be coded as either:

aP_only: all pertussis vaccine doses as aP, none as wP

or

wP_mix: at least one dose of wP at any age.

The comparison of interest is between aP only and at least one dose of wP vaccine.

For secondary analyses: (2), a child's exposure will also be coded as either:

aP_only: all pertussis vaccine doses as aP, none as wP

or

wP_only: all pertussis vaccine doses as wP, none as aP.

The comparison of interest is between aP-only and wP-only vaccinated children. All mixed aP/wP vaccinated children will be excluded from the analysis. [Table 1](#) provides a summary of the coding of exposure for the primary and secondary analyses.

Any analyses other than those outlined above or in the sensitivity analyses and case subgroup analysis below will be declared as unplanned and post hoc.

Matching procedures

For each identified case, up to 10 children will be randomly sampled without replacement from the AIR database from among all children born on the same day as the case ± 7 days and from the same jurisdiction and from a postcode with the same IRSAD decile (1st to 10th). Children will only be included once as a case. Each cohort control will be associated with one unique case. Consistent with the case-cohort method,³⁶ sampling of controls will be from the register cohort without regard to case status (ie, children will be eligible to be a control irrespective of whether they are at any stage a case). In

Table 1 Summary of the coding of exposure for the primary and secondary analyses

First dose, study eligibility requires first dose before age 16 weeks	Second dose	Third dose	Primary coding, first wP versus first aP	Secondary (1) coding, any wP versus only aP	Secondary (2) coding, only wP versus only aP
wP	Missing	Missing	wP1	wP_mix	wP_only
wP	wP	Missing	wP1	wP_mix	wP_only
wP	wP	wP	wP1	wP_mix	wP_only
wP	aP	aP or missing	wP1	wP_mix	Not included
wP	aP or missing	aP	wP1	wP_mix	Not included
aP	wP or missing	wP	aP1	wP_mix	Not included
aP	wP	wP or missing	aP1	wP_mix	Not included
aP	aP	aP	aP1	aP_only	aP_only
aP	aP	Missing	aP1	aP_only	aP_only
aP	Missing	Missing	aP1	aP_only	aP_only

aP, acellular pertussis; wP, whole-cell pertussis.

total, up to 5000 controls will be sampled. IRSAD deciles will be ascertained from ABS data from the 2011 census since it is likely that only the most recent Medicare post-code is available for the majority of the participants.

Sensitivity analysis

In the first instance, a sensitivity analysis will be performed on the case definition for the primary analysis based on confirmation of food allergy via a food challenge. Confirmation by food challenge is considered the gold standard for food allergy, but in clinical practice, this has more usually been reserved for cases in which there is diagnostic uncertainty or to confirm resolution of the food allergy. If there is a significant change in the interpretation of the results based on the sensitivity analysis compared with the primary analysis, then further sensitivity analyses will also be performed for the secondary analyses.

As an additional sensitivity analysis, the case definition will require documentation of an SPT weal of 8 mm for cow milk, 7 mm for egg or 8 mm for peanut or food-specific serum IgE of 15 kU/L for cow milk, 7 kU/L for egg or 14 kU/L for peanut. These more conservative cutoffs correspond to almost 100% and 95% PPV of IgE-mediated allergy for the SPT and ss-IgE, respectively.

Case subgroup analysis

To investigate the potential heterogeneity of responses following exposure to specific vaccine formulations, we will conduct an a priori subgroup analysis by acellular vaccine brand (excluding children who have received mixed acellular vaccine types). All other subgroup analyses will be declared as unplanned and post hoc.

DISCUSSION

Allergy represents a significant disease burden in developed countries. The temporal association between an apparent increase in severe food allergy and the replacement of whole cell with aP vaccine in Australia warrants further investigation. Food allergy is not only important in its own right, it is also associated with eczema and with asthma (fourfold increased risk) in later childhood.^{37,38}

This study has been designed to comply with both the Strengthening the Reporting of Observational Studies in Epidemiology recommendations and also the more specific recommendations put forward by Sharpe *et al*³⁹ for case-cohort studies. The strengths of this study include objective allergy definition and access to an established prospective population-based vaccine register for determination of vaccination status of both cases and cohort controls. Access to patient lists of allergist-diagnosed food allergy from large paediatric referral centres along with detailed prospectively collected immunisation records from the AIR provides a unique opportunity to investigate a potential relationship between these events. The transition from wP to aP in Australia represents a natural experiment of the effect of infant wP on subsequent

food allergy, on a scale unachievable by a randomised controlled trial.

The disadvantages of this study are that it is reliant on existent databases for which there is little way to assess the validity or accuracy of data entry. The AIR database has been lauded as a highly effective means of tracking vaccination in the Australian population; however, there is evidence of under-reporting of administered vaccines,⁴⁰ and there has been no validation of the quality of data entry into the database. Finally, we assume that the type of pertussis vaccine administered was dependent on jurisdiction and date of birth, but not otherwise dependent on factors which are also risk factors for allergy. While this assumption appears entirely reasonable, we are not able to assess its validity.

Author affiliations

¹Wesfarmers Centre of Vaccines & Infectious Diseases, Telethon Kids Institute, West Perth, Western Australia, Australia

²School of Population and Global Health, University of Western Australia, Crawley, Western Australia, Australia

³Centre for Applied Statistics, University of Western Australia, Crawley, Western Australia, Australia

⁴Department of Allergy and Immunology, The Children's Hospital at Westmead, Westmead, New South Wales, Australia

⁵Sydney Medical School, University of Sydney, Sydney, New South Wales, Australia

⁶School of Medicine, University of Adelaide, Women's and Children's Health Network, North Adelaide, South Australia, Australia

⁷Centre for Food and Allergy Research, Murdoch Children's Research Institute, Melbourne, Australia

⁸Department of Paediatrics, University of Melbourne, Royal Children's Hospital Melbourne, Victoria, Australia

⁹Child Health Research, Princess Margaret Hospital for Children, Subiaco, Western Australia, Australia

¹⁰School of Public Health, Curtin University, Bentley, Western Australia, Australia

Contributors TLS, MSG, DEC, KJA and CSW were responsible for study concept, design and funding acquisition. MJE, TLS, CSW and JAM wrote the protocol. MJE drafted the manuscript and coordinated manuscript preparation and revision. DEC, MSG, KJA and PR specified the clinical definition of food allergy for the study. JAM and TLS developed the statistical analysis plan. All authors provided critical evaluation and revision of the manuscript and have given final approval of the manuscript accepting responsibility for all aspects.

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Competing interests None declared.

Patient consent Not required.

Ethics approval This study was approved by the Human Research Ethics Committees (HREC) in each jurisdiction. The PIFA study is approved by the Child and Adolescent Health Service Human Research Ethics Committee, Perth, Western Australia (ref 2015052EP), Adelaide Women's and Children's Hospital HREC, South Australia (ref HREC/15/WCHN/162), Royal Children's Hospital HREC, Victoria, Australia (ref 35230A) and Sydney Children's Hospital Network HREC, New South Wales, Australia (ref HREC/15/SCHN/405).

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REFERENCES

- Asher MI, Montefort S, Björkstén B, *et al.* Worldwide time trends in the prevalence of symptoms of asthma, allergic rhinoconjunctivitis, and eczema in childhood: ISAAC Phases One and Three repeat multicountry cross-sectional surveys. *Lancet* 2006;368:733–43.
- Robertson CF, Roberts MF, Kappers JH. Asthma prevalence in Melbourne schoolchildren: have we reached the peak? *Med J Aust* 2004;180:273–6.
- Prescott S, Allen KJ. Food allergy: riding the second wave of the allergy epidemic. *Pediatr Allergy Immunol* 2011;22:155–60.
- Mullins RJ. Paediatric food allergy trends in a community-based specialist allergy practice, 1995–2006. *Med J Aust* 2007;186:618–21.
- Mullins RJ, Wainstein BK, Barnes EH, *et al.* Increases in anaphylaxis fatalities in Australia from 1997 to 2013. *Clin Exp Allergy* 2016;46:1099–110.
- Osborne NJ, Koplin JJ, Martin PE, *et al.* Prevalence of challenge-proven IgE-mediated food allergy using population-based sampling and predetermined challenge criteria in infants. *J Allergy Clin Immunol* 2011;127:668–76.
- Venter C, Pereira B, Grundy J, *et al.* Prevalence of sensitization reported and objectively assessed food hypersensitivity amongst six-year-old children: a population-based study. *Pediatr Allergy Immunol* 2006;17:356–63.
- Lin RY, Anderson AS, Shah SN, *et al.* Increasing anaphylaxis hospitalizations in the first two decades of life: New York State, 1990–2006. *Ann Allergy Asthma Immunol* 2008;101:387–93.
- Ross MP, Ferguson M, Street D, *et al.* Analysis of food-allergic and anaphylactic events in the National Electronic Injury Surveillance System. *J Allergy Clin Immunol* 2008;121:166–71.
- Strachan DP. Family size, infection and atopy: the first decade of the "hygiene hypothesis". *Thorax* 2000;55 Suppl 1(Suppl 1):2S–10.
- Allen KJ, Martin PE. Clinical aspects of pediatric food allergy and failed oral immune tolerance. *J Clin Gastroenterol* 2010;44:391–401.
- Torvaldsen S, Hull BP, McIntyre PB. Using the Australian Childhood Immunisation Register to track the transition from whole-cell to acellular pertussis vaccines. *Commun Dis Intell Q Rep* 2002;26:581–3.
- Coffman RL, Seymour BW, Lebman DA, *et al.* The role of helper T cell products in mouse B cell differentiation and isotype regulation. *Immunol Rev* 1988;102:5–28.
- West CE, Videky DJ, Prescott SL. Role of diet in the development of immune tolerance in the context of allergic disease. *Curr Opin Pediatr* 2010;22:635–41.
- Mills KH, Ryan M, Mcguirk P, *et al.* The immunology of bordetella pertussis infection. *Biologicals* 1999;27:77.
- Barlow WE, Davis RL, Glasser JW, *et al.* The risk of seizures after receipt of whole-cell pertussis or measles, mumps, and rubella vaccine. *N Engl J Med* 2001;345:656–61.
- Feunou PF, Bertout J, Loch C. T- and B-cell-mediated protection induced by novel, live attenuated pertussis vaccine in mice. Cross protection against parapertussis. *PLoS One* 2010;5:e10178.
- Mills KH, Ryan M, Ryan E, *et al.* A murine model in which protection correlates with pertussis vaccine efficacy in children reveals complementary roles for humoral and cell-mediated immunity in protection against Bordetella pertussis. *Infect Immun* 1998;66:594–602.
- Sheridan SL, Ware RS, Grimwood K, *et al.* Number and order of whole cell pertussis vaccines in infancy and disease protection. *JAMA* 2012;308:454–6.
- Fulton TR, Phadke VK, Orenstein WA, *et al.* Protective Effect of Contemporary Pertussis Vaccines: A Systematic Review and Meta-analysis. *Clin Infect Dis* 2016;62:1100–10.
- Dannemann A, van Ree R, Kulig M, *et al.* Specific IgE and IgG4 immune responses to tetanus and diphtheria toxoid in atopic and nonatopic children during the first two years of life. *Int Arch Allergy Immunol* 1996;111:262–7.
- Rowe J, Macaubas C, Monger T, *et al.* Heterogeneity in diphtheria-tetanus-acellular pertussis vaccine-specific cellular immunity during infancy: relationship to variations in the kinetics of postnatal maturation of systemic th1 function. *J Infect Dis* 2001;184:80–8.
- Bancroft T, Dillon MB, da Silva Antunes R, *et al.* Th1 versus Th2 T cell polarization by whole-cell and acellular childhood pertussis vaccines persists upon re-immunization in adolescence and adulthood. *Cell Immunol* 2016;304:35–43.
- Mascart F, Hainaut M, Peltier A, *et al.* Modulation of the infant immune responses by the first pertussis vaccine administrations. *Vaccine* 2007;25:391–8.
- Holt PG, Rowe J, Kusel M, *et al.* Toward improved prediction of risk for atopy and asthma among preschoolers: a prospective cohort study. *J Allergy Clin Immunol* 2010;125:653–9.
- Sly PD, Boner AL, Björkstén B, *et al.* Early identification of atopy in the prediction of persistent asthma in children. *Lancet* 2008;372:1100–6.
- Quinn P, Gold M, Royle J, *et al.* Recurrence of extensive injection site reactions following DTaP or dTpa vaccine in children 4–6 years old. *Vaccine* 2011;29:4230–7.
- Rowe J, Yerkovich ST, Richmond P, *et al.* Th2-associated local reactions to the acellular diphtheria-tetanus-pertussis vaccine in 4- to 6-year-old children. *Infect Immun* 2005;73:8130–5.
- Ryan EJ, Nilsson L, Kjellman N, *et al.* Booster immunization of children with an acellular pertussis vaccine enhances Th2 cytokine production and serum IgE responses against pertussis toxin but not against common allergens. *Clin Exp Immunol* 2000;121:193–200.
- Zhang L, Prietsch SO, Axelsson I, *et al.* Acellular vaccines for preventing whooping cough in children. *Cochrane Database Syst Rev* 2014:CD001478.
- Holt PG, Snelling T, White OJ, *et al.* Transiently increased IgE responses in infants and pre-schoolers receiving only acellular Diphtheria-Pertussis-Tetanus (DTaP) vaccines compared to those initially receiving at least one dose of cellular vaccine (DTwP) - Immunological curiosity or canary in the mine? *Vaccine* 2016;34:4257–62.
- Kiraly N, Koplin JJ, Crawford NW, *et al.* Timing of routine infant vaccinations and risk of food allergy and eczema at one year of age. *Allergy* 2016;71:541–9.
- Nilsson L, Kjellman NI, Björkstén B. A randomized controlled trial of the effect of pertussis vaccines on atopic disease. *Arch Pediatr Adolesc Med* 1998;152:734–8.
- Venter C, Stowe J, Andrews NJ, *et al.* No association between atopic outcomes and type of pertussis vaccine given in children born on the Isle of Wight 2001–2002. *J Allergy Clin Immunol Pract* 2016;4:1248–50.
- Sporik R, Hill DJ, Hosking CS. Specificity of allergen skin testing in predicting positive open food challenges to milk, egg and peanut in children. *Clin Exp Allergy* 2000;30:1541–6.
- Sampson HA. Update on food allergy. *J Allergy Clin Immunol* 2004;113:805–19. quiz 20.
- Barlow WE, Ichikawa L, Rosner D, *et al.* Analysis of case-cohort designs. *J Clin Epidemiol* 1999;52:1165–72.
- Lowe AJ, Hosking CS, Bennett CM, *et al.* Skin prick test can identify eczematous infants at risk of asthma and allergic rhinitis. *Clin Exp Allergy* 2007;37:1624–31.
- Kjaer HF, Eller E, Andersen KE, *et al.* The association between early sensitization patterns and subsequent allergic disease. The DARC birth cohort study. *Pediatr Allergy Immunol* 2009;20:726–34.
- Sharp SJ, Poulaliou M, Thompson SG, *et al.* A review of published analyses of case-cohort studies and recommendations for future reporting. *PLoS One* 2014;9:e101176.
- Hull BP, McIntyre PB. A re-evaluation of immunisation coverage estimates from the Australian Childhood Immunisation Register. *Commun Dis Intell* 2000;24:161–4.