

## REVIEW

## Early life circumstances and atopic disorders in childhood

R. M. D. Bernsen<sup>\*†</sup>, J. C. van der Wouden<sup>\*</sup>, N. J. D. Nagelkerke<sup>†</sup> and J. C. de Jongste<sup>‡</sup>

<sup>\*</sup>Department of General Practice, Erasmus MC University Medical Center Rotterdam, Rotterdam, The Netherlands, <sup>†</sup>Department of Community Medicine, Faculty of Medicine and Health Sciences, United Arab Emirates University, Al Ain, United Arab Emirates and <sup>‡</sup>Department of Pediatrics, Subdivision of Pediatric Respiratory Medicine, Erasmus MC Sophia Children's Hospital Rotterdam, Rotterdam, The Netherlands

### Clinical and Experimental Allergy

#### Summary

The prevalence of childhood atopic disorders has risen dramatically in the last decades of the past century. Risk factors for the development of these disorders have been studied extensively. This review focuses on the role of early life risk factors such as pre-natal development, perinatal circumstances, birth order and childhood vaccinations.

#### Correspondence:

Roos M. D. Bernsen, Department of  
Community Medicine, Faculty of  
Medicine and Health Sciences, United  
Arab Emirates University, PO Box 17666,  
Al Ain, United Arab Emirates.  
E-mail: rmdbernsen@gmail.com

#### Introduction

Childhood atopic disorders, which constitute a significant burden for affected children and their parents [1], have shown an epidemic rise in western societies during recent decades [2, 3]. The reasons for this rise are poorly understood, as is the aetiology of these disorders in general. Although there is general consensus on the importance of a genetic predisposition for atopic disorders [4], only changes in environmental factors can explain this rise. There are two – not mutually exclusive – possibilities, namely, that substances that promote atopy have been added to the environment or that factors that provided protection from allergic disease were lost from the environment. In 1989, Strachan, pursuing the second possibility, put forward the 'hygiene hypothesis', stating that hygienic living conditions with little exposure to pathogens at an early age would increase the risk of atopy [5]. This idea was prompted by the finding that subjects raised in large families had less hayfever than those raised in small families who presumably live under more hygienic conditions than children with many siblings. This so-called 'sibling effect' is closely related to, and difficult to distinguish from, the phenomenon called the 'birth order

effect', that children with no or a few older siblings have a higher risk of atopic disorders than children with many older siblings. Since the hypothesis was published, many studies have been empirically testing it and several findings appear to support it. For instance, exposure to probiotics [6], farm environment (associated with endotoxin exposure) [7, 8], orofecal pathogens [9] and helminths [10] all seem to reduce the risk of atopic disorders. However, the birth order effect cannot be explained by early infections [11–13].

Whatever factors are responsible for the development of atopic disorders, it is now recognized that priming of the immune system occurs early in infancy [14], or even before birth [15, 16], and it is therefore key to study the relationship between early life circumstances and the development of asthma and allergy during childhood. Here, we will focus on foetal development (anthropometric measures at birth and gestational age), obstetric complications, birth order and vaccinations in infancy as risk factors for atopic disorders in childhood and speculate on possible mechanisms underlying the relationships observed.

In this paper, we use 'atopic disorders' for a wide variety of diseases and symptoms such as asthma, hayfever,

allergic rhinitis (AR), atopic dermatitis (AD), skin prick test positivity and elevated specific IgE. We have chosen this comprehensive definition in order to cover the wide range of outcomes that we found.

### **Anthropometric measures at birth and gestational age**

The increasing evidence that the foetal environment can have life-long health effects [17, 18] (the 'Barker hypothesis') has stimulated research on the relationship between foetal development and atopic disorders [19–40]. Unfortunately, no clear picture emerges from these studies as the results are often contradictory, which may in part be due to the large heterogeneity in study design and outcome measures. For example, while atopy was determined in childhood in most studies, it was measured in adulthood in others. While several studies reported low gestational age (or low birth weight) to be a risk factor for the subsequent development of asthma or atopic disease [19–26], this relationship was challenged by others [27–31]. Similarly, while some studies found a link between high gestational age, or high birth weight, and either a high risk of asthma, atopic disease or a high IgE concentration [24, 26, 32–34], others did not [21, 25, 27, 29, 31, 35, 36]. One study reported that the lymphocyte-subsets profile at birth of pre-term infants differed from that of term infants [37], suggesting that a possible link between pre-maturity and atopy may be detectable already at birth. A positive association of neonatal head circumference with total IgE concentration [29, 32, 36, 38] and with asthma [26] has been reported, prompting Godfrey *et al.* [32] to hypothesize that this reflects the long-term effect of sustaining foetal brain growth at the expense of the growth of the trunk, in particular the thymus. However, this hypothesis was subsequently challenged by the finding that thymus size is not related to allergic disease [39] and that the relationship between the ratio of head circumference to birth weight, a better indicator of relative brain growth, and allergy is actually negative [34, 40], although one later study reported this relationship to be positive [25].

The positive association between pre-maturity or low birth weight and risk of asthma is usually explained by the fact that pre-maturity causes disturbed lung growth, which would result in reduced airway calibre and give rise to wheezing symptoms during viral infections, symptoms that are often misdiagnosed as asthma. There may, however, be more complex mechanisms underlying the relationships between foetal development and atopic disorders, which will be discussed below.

### **Pre-natal and obstetric complications**

Since perinatal factors, including obstetric events, have been reported to be related to the development of child-

hood atopic disorders [41], many studies have focused on the relationship between complications and interventions during delivery and atopy. Several studies found that caesarean section is a risk factor for subsequent development of asthma and wheezing in both childhood [23, 42–45] and adulthood [46] and sensitization in children [43, 47, 48]. However, other studies failed to reproduce these results [25, 49]. Other complications during labour also showed positive relationships, e.g. vacuum extraction with allergy [25]. It has been hypothesized that incubator care, being more common after complicated deliveries, would delay the contact of the newborn with microbes and thus affect the T-helper type 2 (Th2) → Th1 shift [23] and bias the immune system towards atopic reactions, but no empirical study supports this hypothesis. In fact, it is unknown whether these relationships are causal, or due to confounding by some underlying condition associated with both obstetric complications and atopic disorders. In fact, such confounding was suggested by a study [50] comparing B lymphocytes in cord blood between neonates born by vaginal delivery and those born by caesarean section: a higher percentage of positivity to cell surface markers of B lymphocyte subpopulations in infants born by elective caesarean section was found. Similarly, a recent study found that total IgE in cord blood was higher in children born by caesarean section compared with vaginally delivered children [47]. However, as the authors suggest, other factors such as gestational age and birth order might also play a role.

### **Birth order**

Strachan's [5] observation, that individuals with many siblings have a smaller risk of allergy than children without siblings, which prompted the hygiene hypothesis in 1989, has been confirmed and further explored by many studies summarized in a review by Karmaus *et al.* [51]. A study that assessed the independent effects of birth order and sibship size on allergy in the siblings of 700 families [52] showed that it is the number of older siblings, rather than the size of the sibship *per se*, that is (inversely) related to the risk of allergy. Although a recent Danish study found that the number of younger siblings was negatively associated with the risk of AR [53], this does not necessarily imply a protective effect of younger siblings on the development of AR as other interpretations are possible [54]. The mechanism underlying this sibling effect or birth order effect is a puzzle worth solving, because it could potentially help in preventing as much as 30% of all atopy cases [51].

### *Age of onset of birth order effect*

An important issue is the age at which the birth order effect develops, as it may clarify its aetiology. There is

increasing evidence that this occurs very early in life. For example, IgE measurements in cord blood have been shown to be negatively associated with birth order [55–57], murine experiments have demonstrated that the immune status of the placenta changes with parity [58] and a negative relationship between parity and maternal atopy [59] and parity and maternal IgE [60] was found. These results provide strong evidence that the birth order effect already operates before birth and that possibly the immune status of women changes after multiple births. This last hypothesis could not be confirmed in a large follow-up study [61], while the findings of a smaller follow-up study supported the hypothesis [62]. Surely, this pre-natal birth order effect does not preclude possible additional effects of exposure to pathogens from other siblings in the family. While this pre-natal effect of parity could be due to maternal exposure to pathogens from her older children during pregnancy, a change in the maternal–foetal interaction with increasing parity [55] would seem a more plausible explanation.

#### *Maternal alloantigen reactivity*

Although a positive relationship between parity and human lymphocyte antigen (HLA) antibodies was found already in 1984 [63], the association between HLA antibodies and atopy of the offspring was not investigated until 2005 when Prescott et al. [64] explored maternal–foetal interaction. They found a positive relationship between maternal reactivity to foetal alloantigens and allergic disease at 6 years of age and a positive relationship between foetal IFN- $\gamma$  alloantigen response and the degree of HLA mismatch between mother and child. This study also found that the number of previous pregnancies was associated with stronger maternal responses to foetal alloantigens, which was related to a higher risk of allergy at 6 years. This finding is surprising as it would suggest absence of the birth order effect, or even a reversed birth order effect. Indeed, in their study population there was no relationship between parity and allergy at the age of 6 years (S. L. Prescott, personal communication). This positive association between parity and maternal response to foetal alloantigens raises the question of whether mothers were sensitized to the alloantigens of the child by exposure to these alloantigens during previous pregnancies in which the fetus presented the same (paternally inherited) alloantigens. Data from a different, older, study on the HLA antigen distribution in families with at least one child with AD [65] provide evidence that this is indeed the case. In this study, 19 children with AD from 17 families, their 36 unaffected siblings and their parents were HLA typed to explore the association between HLA and atopy. The authors, however, did not consider the role of sensitization to alloantigens, which can be examined using data provided in Table 1 of their paper: for nine out

of a total of 19 atopic children, the mother had been exposed to exactly one paternal HLA type before she became pregnant with the atopic child. We call these nine mother–child pairs ‘informative’, because in these pairs we could assess how often the paternal HLA type presented by the atopic child was equal or not equal (each inherited with probability 0.5 if not related to atopy of the child) to the one the mother had been exposed to before. It turned out that eight mothers had been exposed to the same alloantigens as presented by the atopic child, a significantly (two tailed  $P$ -value:  $20/512 < 0.05$ ) higher number than expected by chance. This result is in agreement with the result of stronger maternal responses with increasing parity, found by Prescott et al. Surprisingly, also in the study by MacKie and Dick [65], no relationship between birth order and childhood atopy was observed. These two studies together suggest that maternal alloantigen reactivity increases with increasing parity. Previous exposure of the mother to alloantigens presented by the child increases the risk of childhood allergy. This is intriguing, because the effect of such a mechanism would contradict the empirically established birth order effect.

#### *Genetic susceptibility*

Another aspect of the birth order effect is its genetic susceptibility, i.e. are there genes, of the mother or the child, that are predictors of the birth order effect, so that some mother–child pairs benefit from the birth order effect while others do not? Unfortunately, little is known in this area, although the identification of loci involved in this effect would both provide an insight into the underlying mechanism of the birth order effect and outline which groups of people would benefit from preventive actions. Also, the genetic constitution of the mother could act as an effect modifier in the relationship between alloantigen presentation and maternal alloantigen reactivity, and thus influence the atopic status of the child. Undoubtedly, the availability of high throughput equipment for identifying genotypes and gene expression patterns will make it easier to address these issues in the near future.

#### *Other conditions related to birth order*

Another approach to identify mechanisms underlying the birth order effect is by comparison with other conditions related to birth order, such as pre-eclampsia [66]. This disorder results from deficient trophoblast invasion, with the highest risk in nulliparous women and in women with a new partner [67], suggesting that mothers develop tolerance to alloantigens after multiple births from the same partner. A possible explanation for these relationships is that during childbirth foetal cells enter the

maternal circulation and stimulate the mother's immune system to develop tolerance to foetal antigens. Another result of these intriguing studies is that miscarriage, pre-eclampsia and impaired foetal growth appear to represent a spectrum of disorders secondary to deficient trophoblast invasion [66]. Also, pre-eclampsia is related to the immunophenotype of neonates [68]. The exact relevance of these phenomena for the development of atopy and asthma remains to be elucidated.

### Infant and childhood vaccinations

We confine ourselves to infant vaccinations that are routinely administered in the western world, viz.: diphtheria-tetanus-pertussis (whole-cell or acellular) (DTP) (sometimes supplemented with a polio vaccine), measles-mumps-rubella (MMR) and *Haemophilus influenzae* type B (Hib). For the new meningococcal C vaccine (included as a routine childhood immunization in the United Kingdom in 2000 and in the Netherlands in 2003), no epidemiological studies on the relationship studied in this paper are as yet available.

In the context of the hygiene hypothesis, a potential relationship between vaccinations and atopy could go either way: either vaccinations prevent infections, and thus contribute to the 'hygiene effect', or vaccinated children, at a very young age, are exposed to (attenuated or inactivated) pathogens or components of pathogens, thus possibly promoting Th1 proliferation, a favourable situation according to the hygiene hypothesis. In addition, exposure to vaccine additives, such as aluminium-containing adjuvants, might also change the risk of developing atopy.

#### *Diphtheria-tetanus-pertussis*

An increased IgE serum concentration after stimulation with extracts of *Bordetella pertussis* was found in murine models [69] and in newborns after whole-cell pertussis vaccination [70]. Many epidemiological studies on the role of the DTP vaccination (or the pertussis component alone) in the development of atopic disorders have been conducted since. In a recent review, Koppen et al. [71] concluded that childhood vaccinations are not associated with a higher risk of allergy, but a few studies were not included, mainly because they appeared after its publication [72-77]. A Japanese cross-sectional study in 82 children aged 0-3 years concluded that DTP-vaccinated children had a significantly higher risk of asthma, AR or AD than DTP-unvaccinated children [72]. A cross-sectional study in France in 718 adolescents (of whom only 24 were not vaccinated) concluded that childhood immunizations in general, and especially immunizations with live attenuated vaccines, possibly protect from the development of atopic disease [73]. A study in Australian

young adults [74] showed a weak support for the hypothesis that childhood vaccinations may lead to a higher risk of asthma, but the authors state that this result may be due to recall bias. A case-control study in UK concluded that DTP vaccination does not increase the risk of hayfever [75]. Unfortunately, the conclusions of all these studies may be flawed because the reason for not receiving the DTP vaccination is rarely properly documented and may be health related [78]. If parents are more inclined to refrain from vaccinations if their children are at risk of atopy, studies could be subject to bias similar to confounding by indication. This bias could for example either conceal an existing effect of vaccinations on atopy or yield a spurious difference in favour of unvaccinated children. Bernsen et al. [76] tried to avoid this possible bias in two studies in the Netherlands. In the first one, we studied data from health records of 6-year-old children and concluded that pertussis-vaccinated children (or DTP-vaccinated children, as there was much overlap) had a significantly smaller risk of atopic disorders. Despite the advantage that the (parents of) unvaccinated children refrained from vaccinations for religious reasons (not health related), residual confounding could not be ruled out as all unvaccinated children attended the same school and little was known about the characteristics of both groups. In a second study in the Netherlands, in the same religious population but from a much larger area [77], we obtained questionnaire data of 1875 children aged 8-12 years recruited from 38 primary schools and found no difference in the prevalence of atopic disorders between DTP-vaccinated and DTP-unvaccinated children. In this study, the main reason for refraining from vaccinations was also religious and differences between the vaccinated and the unvaccinated group were thoroughly explored and adjusted for, thus providing convincing evidence that in this population the DTP vaccination is not associated with a higher risk of atopic disorders.

#### *Measles-mumps-rubella*

Many studies on the effects of MMR vaccination have been carried out [71, 75]. Generally, their findings are more consistent than those of the DTP vaccination, and support the conclusion that MMR vaccination is not associated with a higher risk of atopic disorders.

#### *Haemophilus influenzae type B*

Only a few studies have reported on the relationship between Hib vaccination and atopic disorders. A cross-sectional survey of 1943 schoolchildren aged 5-14 years in Eastern Germany [79] found a positive association between Hib vaccination and asthma (odds ratio (OR): 1.86 (CI<sub>95%</sub>: 1.05-3.32)), but the authors caution that residual confounding cannot be ruled out. A cohort study

in USA [80] on the relationship of Hib vaccination with asthma involving almost 170 000 children and using computerized data on vaccinations and medical care encounters with follow-up from birth to a maximum of 6 years of age also found a weak association (OR = 1.18, CI<sub>95%</sub>: 1.02–1.36), which the authors at least partially ascribe to health care utilization or information bias. In these observational studies, information bias and residual confounding cannot be excluded. A nested case-control study in 7–8-year-old Dutch and German children [81] with complete data for 572 children found no evidence for an association between the Hib vaccination and respiratory symptoms or allergic sensitization.

Finally, our cross-sectional study in the Netherlands comparing Hib-vaccinated and Hib-unvaccinated children within the subpopulation of DTP-vaccinated children found no evidence for a higher risk of atopic disorders among the Hib-vaccinated children compared with the Hib-unvaccinated children [82]. Biological plausibility for a causal relationship is not evident: one study showed a mixed Th1–Th2 response after Hib vaccination of 19 adults [83].

## Discussion and recommendations

### *Heterogeneous study results*

The reported relationships between perinatal variables (anthropometric measurements and obstetric complications) and atopic disorders as summarized in this paper are often inconsistent, which raises the question of whether these relationships are real, whether they are results of residual confounding or perhaps mere chance findings. Many factors could have caused studies to have conflicting results. As an example, we consider the relationship between low birth weight and atopy. If medical consumption is involved in the assessment of the outcome (as in 'doctor-diagnosed asthma'), one could imagine that low-birth-weight groups differ in medical consumption behaviour from normal-birth-weight groups. Also, studies in different populations could yield different results. For example, exposure to smoke (pre- and post-natal) could induce a positive relationship between low birth weight and asthma in populations with a high prevalence of smoking, whereas the relationship is absent in populations with a low smoking prevalence. Also, the age of the study subjects might influence the conclusions, as the atopic phenotype is highly age-dependent: an effect present in childhood might have disappeared in adulthood. Another explanation for inconsistent results of studies, with asthma as an outcome, could be that asthma is not a single entity, but a heterogeneous syndrome with different underlying pathology [84]. Therefore, it is recommendable for future studies to phenotype the patient with

asthma symptoms carefully, and search for phenotype-specific effects.

### *Pre-natal priming – an explanation for the birth order effect?*

The mechanism underlying the *in utero* birth order effect deserves special attention. Recent research has shown that the maternal–foetal interaction (and thus pre-natal development) is influenced by parity and genetic make-up [85] and even by the gender of older siblings [86]. According to Burton's theory [66], efficient trophoblast invasion is related to tolerance, implying that in general women with higher parity are more tolerant to foetal antigens. Combining this hypothesis with the association between pre-eclampsia and neonatal immunophenotype could give us clues regarding where to look for explanations of the birth order effect. Further, again hypothesized by Burton and Jauniaux [66], miscarriage, pre-eclampsia and impaired foetal growth represent different consequences of a deficient trophoblast invasion. A combination of these hypotheses with the association between pre-eclampsia and immunophenotype of the neonate could offer new starting points for exploring the mechanism underlying the relationship between foetal development and atopic disorders. For instance, we could hypothesize that low birth weight could be related to deficient trophoblast invasion, while at the same time deficient trophoblast invasion could be related to childhood atopy, thus explaining the association between low birth weight and atopic disease in some studies. A role of alloantigens in both allergy and pre-eclampsia has already been discussed by Karmaus and Johnson [87]. These hypotheses could potentially lead to the development of preventive measures, for instance by artificially inducing tolerance to paternal tissues before pregnancy.

Results conflicting with the idea of mothers developing tolerance to alloantigens after multiple births come from studies on maternal alloantigen reactivity [64, 65]. These results suggest a decreasing tolerance with increasing parity. However, in both studies no birth order effect (i.e. a lower risk of atopy in children with a higher number of older siblings) was observed. It would be interesting to repeat the study by MacKie and Dick [65] in a larger population where the (commonly found) birth order effect is present.

### *Vaccinations*

Available evidence is rather convincing that the current regular childhood vaccinations do not increase the risk of atopic disorders. The only exception may be the Meningococcal C vaccination, which, to our knowledge, has not yet been studied in this respect. In order to strengthen public confidence in vaccination programmes and thereby

enhance vaccination coverage, we recommend to conduct a cross-sectional study about 10 years after the introduction of this vaccination (and similarly any other new routine vaccinations), comparing vaccinated and unvaccinated children of about 8–12 years of age.

### Recommendations for future research

Obviously, it is difficult to explain all controversies in published studies. We feel that the highest priority should be given to unravelling the complex mechanisms underlying the relationships between maternal genes, maternal atopy and maternal alloantigen reactivity on the one hand and childhood atopy on the other. Pregnancy outcome is the result of a 9 month period where interactions, poorly understood so far, but vital for the child's health, have taken place. We would recommend a relatively large follow-up study including mothers, both atopic and non-atopic, and their successive children with measurements of maternal genotype, maternal and child HLA, maternal IgE, maternal and foetal responses during successive pregnancies and childhood atopy. Also, the duration of the relationship with the partner and information on new relationships would be desirable in order to explore any associations with alloantigen reactivity. We have seen so far that the mother's reproductive history has an impact on different aspects of a child's constitution and health, which is not limited to atopic disorders: parity was related to the risk of pre-eclampsia [67], birth order was related to the association between genetic risk of type 2 diabetes and neonatal head circumference [85], the number of older brothers was negatively associated with male birth weight [86] and male sexual inclination appeared to be related to the number of older brothers [88]. Maybe combining all these pieces of evidence will reveal the key mechanism underlying the birth order effect. Therefore, we can agree with a recent statement by Karmaus and Johnson [87] that research should focus on a comparative disease approach, trying to integrate findings from different areas of disease. In this way, research on a molecular level combined from different areas could be used to generate and test hypotheses on the mechanism underlying the birth order effect.

### Conclusions

Atopic disorders are a considerable burden for affected children and their parents. Research should focus on an integrative approach where knowledge of several disciplines involved in the early immunopathology of atopic disorders is combined. In this way, mechanisms underlying the development of these disorders could be detected and this could possibly lead to effective new preventive strategies. New vaccinations should be monitored as to their effects on childhood allergy, both in order to prevent

a possible increase of the allergy prevalence and to promote high vaccination coverage.

### References

- O'Connell EJ. The burden of atopy and asthma in children. *Allergy* 2004; 59 (Suppl. 78):7–11.
- Wieringa MH, Vermeire PA, Brunekreef B, Weyler JJ. Increased occurrence of asthma and allergy: critical appraisal of studies using allergic sensitization, bronchial hyper-responsiveness and lung function measurements. *Clin Exp Allergy* 2001; 31: 1553–63.
- Law M, Morris JK, Wald N, Luczynska C, Burney P. Changes in atopy over a quarter of a century, based on cross sectional data at three time periods. *Br Med J* 2005; 330:1187–8.
- Blumenthal MN. The role of genetics in the development of asthma and atopy. *Curr Opin Allergy Clin Immunol* 2005; 5: 141–5.
- Strachan DP. Hay fever, hygiene, and household size. *Br Med J* 1989; 299:1259–60.
- Kalliomaki M, Salminen S, Arvilommi H, Kero P, Koskinen P, Isolauri E. Probiotics in primary prevention of atopic disease: a randomised placebo-controlled trial. *Lancet* 2001; 357:1076–9.
- Braun-Fahrlander C, Gassner M, Grize L *et al*. Prevalence of hay fever and allergic sensitization in farmer's children and their peers living in the same rural community. SCARPOL team. Swiss study on childhood allergy and respiratory symptoms with respect to air pollution. *Clin Exp Allergy* 1999; 29:28–34.
- Riedler J, Eder W, Oberfeld G, Schreuer M. Austrian children living on a farm have less hay fever, asthma and allergic sensitization. *Clin Exp Allergy* 2000; 30:194–200.
- Matricardi PM, Rosmini F, Riondino S *et al*. Exposure to foodborne and orofecal microbes versus airborne viruses in relation to atopy and allergic asthma: epidemiological study. *Br Med J* 2000; 320:412–7.
- Yazdanbakhsh M, Matricardi PM. Parasites and the hygiene hypothesis: regulating the immune system? *Clin Rev Allergy Immunol* 2004; 26:15–24.
- Ponsonby AL, Couper D, Dwyer T, Carmichael A, Kemp A. Relationship between early life respiratory illness, family size over time, and the development of asthma and hay fever: a seven year follow up study. *Thorax* 1999; 54:664–9.
- Cullinan P, Harris JM, Newman Taylor AJ *et al*. Can early infection explain the sibling effect in adult atopy? *Eur Respir J* 2003; 22:956–61.
- Gibbs S, Surridge H, Adamson R, Cohen B, Bentham G, Reading R. Atopic dermatitis and the hygiene hypothesis: a case-control study. *Int J Epidemiol* 2004; 33:199–207.
- van der Velden VH, Laan MP, Baert MR, de Waal Malefyt R, Neijens HJ, Savelkoul HF. Selective development of a strong Th2 cytokine profile in high-risk children who develop atopy: risk factors and regulatory role of IFN-gamma, IL-4 and IL-10. *Clin Exp Allergy* 2001; 31:997–1006.
- Jones CA, Holloway JA, Warner JO. Does atopic disease start in foetal life? *Allergy* 2000; 55:2–10.
- Warner JO, Jones CA, Kilburn SA, Vance GH, Warner JA. Prenatal sensitization in humans. *Pediatr Allergy Immunol* 2000; 11 (Suppl. 13):6–8.

- 17 Barker DJ, (ed.). *Fetal and infant origins of adult disease*. London: BMJ Publishing, 1992.
- 18 Roseboom TJ, van der Meulen JH, Ravelli AC, Bleker OP. Link between prenatal exposure to the "Winter of Famine" and long-term medical consequences. *Ned Tijdschr Geneesk* 2000; 144:2488–91.
- 19 Arshad SH, Stevens M, Hide DW. The effect of genetic and environmental factors on the prevalence of allergic disorders at the age of two years. *Clin Exp Allergy* 1993; 23:504–11.
- 20 von Mutius E, Nicolai T, Martinez FD. Prematurity as a risk factor for asthma in preadolescent children. *J Pediatr* 1993; 123:223–9.
- 21 Schaubel D, Johansen H, Dutta M, Desmeules M, Becker A, Mao Y. Neonatal characteristics as risk factors for preschool asthma. *J Asthma* 1996; 33:255–64.
- 22 Braback L, Hedberg A. Perinatal risk factors for atopic disease in conscripts. *Clin Exp Allergy* 1998; 28:936–42.
- 23 Annesi-Maesano I, Moreau D, Strachan D. In utero and perinatal complications preceding asthma. *Allergy* 2001; 56:491–7.
- 24 Xu B, Pekkanen J, Laitinen J, Jarvelin MR. Body build from birth to adulthood and risk of asthma. *Eur J Public Health* 2002; 12:166–70.
- 25 Bernsen RM, de Jongste JC, Aardoom HA, Koes BW, van der Wouden JC. Perinatal characteristics and obstetric complications as risk factors for asthma, allergy and eczema at the age of six years. *Clin Exp Allergy* 2005; 35:1135–40.
- 26 Pekkanen J, Xu B, Jarvelin MR. Gestational age and occurrence of atopy at age 31 – a prospective birth cohort study in Finland. *Clin Exp Allergy* 2001; 31:95–102.
- 27 Fergusson DM, Crane J, Beasley R, Horwood LJ. Perinatal factors and atopic disease in childhood. *Clin Exp Allergy* 1997; 27:1394–401.
- 28 Hagstrom B, Nyberg P, Nilsson PM. Asthma in adult life – is there an association with birth weight? *Scand J Prim Health Care* 1998; 16:117–20.
- 29 Gregory A, Doull I, Pearce N *et al*. The relationship between anthropometric measurements at birth: asthma and atopy in childhood. *Clin Exp Allergy* 1999; 29:330–3.
- 30 Sin DD, Spier S, Svenson LW *et al*. The relationship between birth weight and childhood asthma: a population-based cohort study. *Arch Pediatr Adolesc Med* 2004; 158:60–4.
- 31 Laerum BN, Svanes C, Wentzel-Larsen T *et al*. The association between birth size and atopy in young North-European adults. *Clin Exp Allergy* 2005; 35:1022–7.
- 32 Godfrey KM, Barker DJ, Osmond C. Disproportionate fetal growth and raised IgE concentration in adult life. *Clin Exp Allergy* 1994; 24:641–8.
- 33 Olesen AB, Ellingsen AR, Olesen H, Juul S, Thestrup-Pedersen K. Atopic dermatitis and birth factors: historical follow up by record linkage. *Br Med J* 1997; 314:1003–8.
- 34 Bolte G, Schmidt M, Maziak W *et al*. The relation of markers of fetal growth with asthma, allergies and serum immunoglobulin E levels in children at age 5–7 years. *Clin Exp Allergy* 2004; 34:381–8.
- 35 Strachan DP, Harkins LS, Johnston ID, Anderson HR. Childhood antecedents of allergic sensitization in young British adults. *J Allergy Clin Immunol* 1997; 99:6–12.
- 36 Leadbitter P, Pearce N, Cheng S *et al*. Relationship between fetal growth and the development of asthma and atopy in childhood. *Thorax* 1999; 54:905–10.
- 37 Berrington JE, Barge D, Fenton AC, Cant AJ, Spickett GP. Lymphocyte subsets in term and significantly preterm UK infants in the first year of life analysed by single platform flow cytometry. *Clin Exp Immunol* 2005; 140:289–92.
- 38 Oryszczyn MP, Annesi-Maesano I, Campagna D, Sahuquillo J, Huel G, Kauffmann F. Head circumference at birth and maternal factors related to cord blood total IgE. *Clin Exp Allergy* 1999; 29:334–41.
- 39 Benn CS, Jeppesen DL, Hasselbalch H *et al*. Thymus size and head circumference at birth and the development of allergic diseases. *Clin Exp Allergy* 2001; 31:1862–6.
- 40 Katz KA, Pocock SJ, Strachan DP. Neonatal head circumference, neonatal weight, and risk of hayfever, asthma and eczema in a large cohort of adolescents from Sheffield, England. *Clin Exp Allergy* 2003; 33:737–45.
- 41 Sears MR. Epidemiology of childhood asthma. *Lancet* 1997; 350:1015–20.
- 42 Xu B, Pekkanen J, Jarvelin MR. Obstetric complications and asthma in childhood. *J Asthma* 2000; 37:589–94.
- 43 Kero J, Gissler M, Gronlund MM *et al*. Mode of delivery and asthma – is there a connection? *Pediatr Res* 2002; 52:6–11.
- 44 Hakansson S, Kallen K. Caesarean section increases the risk of hospital care in childhood for asthma and gastroenteritis. *Clin Exp Allergy* 2003; 33:757–64.
- 45 Debley JS, Smith JM, Redding GJ, Critchlow CW. Childhood asthma hospitalization risk after cesarean delivery in former term and premature infants. *Ann Allergy Asthma Immunol* 2005; 94:228–33.
- 46 Xu B, Pekkanen J, Hartikainen AL, Jarvelin MR. Caesarean section and risk of asthma and allergy in adulthood. *J Allergy Clin Immunol* 2001; 107:732–3.
- 47 Laubereau B, Filipiak-Pittroff B, von Berg A *et al*. Caesarean section and gastrointestinal symptoms, atopic dermatitis, and sensitisation during the first year of life. *Arch Dis Child* 2004; 89:993–7.
- 48 Negele K, Heinrich J, Borte M *et al*. Mode of delivery and development of atopic disease during the first 2 years of life. *Pediatr Allergy Immunol* 2004; 15:48–54.
- 49 Maitra A, Sherriff A, Strachan D, Henderson JALSPAC Study Team. Mode of delivery is not associated with asthma or atopy in childhood. *Clin Exp Allergy* 2004; 34:1349–55.
- 50 Gasparoni A, Maccario R, Chirico G *et al*. Neonatal B lymphocyte subpopulations and method of delivery. *Biol Neonate* 1992; 61:137–41.
- 51 Karmaus W, Botezan C. Does a higher number of siblings protect against the development of allergy and asthma? A review. *J Epidemiol Commun Health* 2002; 56:209–17.
- 52 Bernsen RM, de Jongste JC, van der Wouden JC. Birth order and sibship size as independent risk factors for asthma, allergy, and eczema. *Pediatr Allergy Immunol* 2003; 14:464–9.
- 53 Westergaard T, Rostgaard K, Wohlfahrt J, Andersen PK, Aaby P, Melbye M. Sibship characteristics and risk of allergic rhinitis and asthma. *Am J Epidemiol* 2005; 162:125–32.
- 54 Bernsen RM. Re: sibship characteristics and risk of allergic rhinitis and asthma. *Am J Epidemiol* 2006; 163:99–100.
- 55 Karmaus W, Arshad H, Mattes J. Does the sibling effect have its origin in utero? Investigating birth order, cord blood immunoglobulin E concentration, and allergic sensitization at age 4 years. *Am J Epidemiol* 2001; 154:909–15.

- 56 Devereux G, Barker RN, Seaton A. Antenatal determinants of neonatal immune responses to allergens. *Clin Exp Allergy* 2002; 32:43–50.
- 57 van Gool CJ, Thijs C, Dagnelie PC *et al*. Determinants of neonatal IgE level: parity, maternal age, birth season and perinatal essential fatty acid status in infants of atopic mothers. *Allergy* 2004; 59:961–8.
- 58 Lagadari M, Blois S, Margni R, Miranda S. Analysis of macrophage presence in murine placenta: influence of age and parity status. *Am J Reprod Immunol* 2004; 51:49–55.
- 59 Sunyer J, Anto JM, Harris J *et al*. Asthma multi-centre infants cohort study. Maternal atopy and parity. *Clin Exp Allergy* 2001; 31:1352–5.
- 60 Karmaus W, Arshad SH, Sadeghnejad A, Twiselton R. Does maternal immunoglobulin E decrease with increasing order of live offspring? Investigation into maternal immune tolerance. *Clin Exp Allergy* 2004; 34:853–9.
- 61 Sunyer J, Anto JM, Plana E *et al*. Maternal atopy and changes in parity. *Clin Exp Allergy* 2005; 35:1028–32.
- 62 Harris JM, White C, Moffat S, Mills P, Newman Taylor AJ, Cullinan P. New pregnancies and loss of allergy. *Clin Exp Allergy* 2004; 34:369–72.
- 63 Morin-Papunen L, Tiilikainen A, Hartikainen-Sorri AL. Maternal HLA immunization during pregnancy: presence of anti HLA antibodies in half of multigravida women. *Med Biol* 1984; 62:323–5.
- 64 Prescott SL, Taylor A, Roper J *et al*. Maternal reactivity to fetal alloantigens is related to newborn immune responses and subsequent allergic disease. *Clin Exp Allergy* 2005; 35:417–25.
- 65 MacKie RM, Dick HM. A study of HLA antigen distribution in families with atopic dermatitis. *Allergy* 1979; 34:19–23.
- 66 Burton GJ, Jauniaux E. Placental oxidative stress: from miscarriage to preeclampsia. *J Soc Gynecol Invest* 2004; 11:342–52.
- 67 Li DK, Wi S. Changing paternity and the risk of preeclampsia/eclampsia in the subsequent pregnancy. *Am J Epidemiol* 2000; 151:57–62.
- 68 Darmochwal-Kolarz D, Leszczynska-Gorzela B, Rolinski J, Oleszczuk J. Pre-eclampsia affects the immunophenotype of neonates. *Immunol Lett* 2001; 77:67–71.
- 69 Clausen CR, Munoz J, Bergman RK. Reaginic-type of antibody in mice stimulated by extracts of *Bordetella pertussis*. *J Immunol* 1969; 103:768–77.
- 70 Haus M, Weinberg EG. Specific IgE antibodies to *Bordetella pertussis* after immunisation in infancy. *Lancet* 1988; 1:1393.
- 71 Koppen S, de Groot R, Neijens HJ, Nagelkerke N, van Eden W, Rümke HC. No epidemiological evidence for infant vaccinations to cause allergic disease. *Vaccine* 2004; 22:3375–85.
- 72 Yoneyama H, Suzuki M, Fujii K, Odajima Y. The effect of DPT and BCG vaccinations on atopic disorders. *Arerugi* 2000; 49:585–92.
- 73 Martignon G, Oryszczyn MP, Annesi-Maesano I. Does childhood immunization against infectious diseases protect from the development of atopic disease? *Pediatr Allergy Immunol* 2005; 16:193–200.
- 74 Benke G, Abramson M, Raven J, Thien FC, Walters EH. Asthma and vaccination history in a young adult cohort. *Aust NZ J Public Health* 2004; 28:336–8.
- 75 Bremner SA, Carey IM, DeWilde S *et al*. Timing of routine immunisations and subsequent hay fever risk. *Arch Dis Child* 2005; 90:567–73.
- 76 Bernsen RM, de Jongste JC, van der Wouden JC. Lower risk of atopic disorders in whole cell pertussis-vaccinated children. *Eur Respir J* 2003; 22:962–4.
- 77 Bernsen RM, de Jongste JC, Koes BW, Aardoom HA, van der Wouden JC. Diphtheria tetanus pertussis poliomyelitis vaccination and reported atopic disorders in 8–12 year old children. *Vaccine* 2006; 24:2035–42.
- 78 Bernsen RM, van de Wouden JC. Re: no epidemiological evidence for infant vaccinations to cause allergic disease. Letter to the Editor. *Vaccine* 2005; 23:1427.
- 79 Laubereau B, Grote V, Holscher G *et al*. Vaccination against *Haemophilus influenzae* type B and atopy in east German schoolchildren. *Eur J Med Res* 2002; 7:387–92.
- 80 DeStefano F, Gu D, Kramarz P *et al*. Childhood vaccinations and risk of asthma. *Pediatr Infect Dis J* 2002; 21:498–504.
- 81 Mommers M, Weishoff-Houben M, Swaen GM *et al*. Infant immunization and the occurrence of atopic disease in Dutch and German children: a nested case-control study. *Pediatr Pulmonol* 2004; 38:329–34.
- 82 Bernsen RM, Koes BW, de Jongste JC, van der Wouden JC. The *Haemophilus influenzae* B vaccination and reported atopic disorders in 8–12 years old children. *Pediatr Pulmonol* 2006; 41:463–9.
- 83 Kamboj KK, King CL, Greenspan NS, Kirchner HL, Schreiber JR. Immunization with *Haemophilus influenzae* type b-CRM(197) conjugate vaccine elicits a mixed Th1 and Th2 CD(4+) T cell cytokine response that correlates with the isotype of antipoly-saccharide antibody. *J Infect Dis* 2001; 184:931–5.
- 84 Heaton T, Rowe J, Turner S *et al*. An immunoepidemiological approach to asthma: identification of in-vitro T-cell response patterns associated with different wheezing phenotypes in children. *Lancet* 2005; 365:142–9.
- 85 Ong KK, Petry CJ, Barratt BJ *et al*. Maternal-fetal interactions and birth order influence insulin variable number of tandem repeats allele class associations with head size at birth and childhood weight gain. *Diabetes* 2004; 53:1128–33.
- 86 Cote K, Blanchard R, Lalumiere ML. The influence of birth order on birth weight: does the sex of preceding siblings matter? *J Biosoc Sci* 2003; 35:455–62.
- 87 Karmaus W, Johnson CC. Invited commentary: sibship effects and a call for a comparative disease approach. *Am J Epidemiol* 2005; 162:133–8.
- 88 Blanchard R. Quantitative and theoretical analyses of the relation between older brothers and homosexuality in men. *J Theor Biol* 2004; 230:173–87.