

# The effect of prenatal exposure on total IgE at birth and sensitization at twelve months and four years of age: The PIAMA study

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There is increasing evidence that the development of the fetal immune system can be influenced by environmental exposure *in utero*. We investigated whether prenatal exposure is associated with a high neonatal total IgE level and sensitization at the age of 1 and 4 yr. Data from 1027 infants were collected in a Dutch birth cohort study (PIAMA study). Total IgE was measured in heel prick blood collected in the first week of life. Sensitization was defined as a specific IgE level in serum of  $\geq 0.35$  IU/ml against house dust mite, cat, dog, milk or egg. Logistic regression analysis was performed to study independent relationships between risk factors and a high neonatal total IgE ( $\geq 0.50$  IU/ml) or sensitization. A high neonatal total IgE was found in 12.2% of boys and 6.2% of girls. A dog at home during pregnancy was negatively associated with a high neonatal total IgE [odds ratio (95% CI) 0.5 (0.2–1.0)]. A cat at home [OR 0.6 (0.4–1.0) and maternal smoking (OR 0.4 (0.2–1.0)] were negatively associated with sensitization at 12 months, but not at 4 yr. The presence of older siblings, season of birth, birth weight, mode of delivery, gestational age and maternal age were not associated with a high neonatal total IgE or sensitization. The higher total IgE level and prevalence of sensitization at 4 yr in boys compared with girls was only present in children from allergic mothers. Our results suggest a short-lasting protective effect of prenatal exposure to pets on total IgE at birth and early sensitization.

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It has been shown that a high total immunoglobulin E (IgE) level in cord blood is a strong risk factor for the development of allergic diseases (1). This indicates that at least in some children the predisposition for allergic disease

has already been established at birth. Until recently, the intrauterine environment has been considered the place where genetic rather than environmental influences take place. However, there is increasing evidence that fetal immune responses to allergens may occur during gestation and that intrauterine conditions play an important role in the control of these responses (2).

Pregnancy is characterized by a down-regulation of the T helper lymphocytes type 1 (Th1) response and a polarization towards T helper lymphocytes type 2 (Th2) of the fetal immune system (3). Normally, immune responses deviate

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towards Th1 in the first years after birth to obtain a more balanced Th1 and Th2 response (4). Deviation from this immune-maturation process plays a role in the development of allergic diseases (5). One of the factors that may provoke an immune-deviation towards Th1 is the exposure to micro-organisms. This may constitute the mechanism of the generally accepted hypothesis that exposure to micro-organisms early in life protects against the development or progression of allergic diseases. The latter is supported by observations that children growing up on a farm with livestock (6) or in an environment with siblings (7) or pets (8–10) have a reduced risk of developing allergic diseases.

Allergen exposure is another factor that may influence the process of immune deviation towards a Th1-response. Fetal immune responses to inhalant allergens encountered by the mother during pregnancy have been demonstrated from 22 wk gestation (11) and a high exposure to allergens of the mother may enhance the Th2 biased immune response of the neonate.

Whether environmental exposures that have been shown to be associated with allergic disease are also operating *in utero* has hardly been examined. Furthermore, it is unclear whether these possible effects of intrauterine exposure on the total IgE production in the neonate have any significance with regard to the development of sensitization later in life.

We used data from the PIAMA study, a birth cohort study on Prevention and Incidence of Asthma and Mite Allergy, to investigate whether prenatal exposure is associated with a high total IgE level measured in heel prick blood in the first week of life and whether the intrauterine factors studied were consistently associated with sensitization at 12 months and 4 yr of age. We also studied whether a maternal history of allergy modifies the association of exposure with a high total IgE level or sensitization.

## Methods

### Subjects

The PIAMA study is a prospective birth cohort study on Prevention and Incidence of Asthma and Mite Allergy. The participating children were born between May 1996 and December 1997 in three regions of the Netherlands. They were selected using a validated screening questionnaire with questions on asthma and respiratory allergies administered to the mother at her first visit to the midwife (12). The inclusion procedure has been described in detail elsewhere

(13). The PIAMA cohort consists of 1327 children from allergic and 2819 children from non-allergic mothers (Fig. 1).

The cohort included 855 infants from mothers with asthma or respiratory allergy, participating in the intervention part of the PIAMA study, a double blind placebo-controlled study on the primary preventive effect of the use of mattress covers on the development of asthma or allergy. Besides, 472 infants from allergic and a random sample of 2819 infants from non-allergic women were included in the natural history part of the PIAMA study.

All 1327 allergic mothers and a random sample of 663 non-allergic mothers were selected for more extensive investigation, including the collection of dust at home-visits performed at 3 months of age. During these home-visits, the parents were asked written informed consent to the use of the blood spot on filter paper that was, as a common practice, left over from routine neonatal screening in the first week of life. In this way, informed consent was obtained from 1755 parents and total IgE was actually measured in heel prick blood from 1280 infants (Fig. 1).

Complete data on the studied risk factors and neonatal total IgE levels were obtained from 1027 infants. Missing data on IgE was either because of absence of home-visit (74), lack of informed consent ( $n = 108$ ) or blood spot ( $n = 475$ ), or a detection limit of measurement above 0.50 IU/ml ( $n = 150$ ). The additional missing data on risk factors were caused by non-response on one or more of the three questionnaires involved ( $n = 103$ ) (Fig. 1).

Specific IgE to house dust mite, cat, dog, grass, milk or egg was measured at 12 months of age in serum of 546 children with allergic mothers from the intervention part. At 4 yr of age, specific IgE to the same allergens was measured in 315 children from the intervention part and 175 and 227 children with allergic and non-allergic mothers respectively from the natural history part of the PIAMA study (Fig. 1). Informed consent to collect blood for specific IgE measurement was obtained from 1036 parents at 4 yr of age.

Complete data on the studied risk factors and sensitization at 12 months and 4 yr were obtained from 492 and 682 children respectively.

### Measurements

Data on the presence of pets in the home, maternal cigarette smoking and paternal history of allergy were collected by questionnaire in the last trimester of pregnancy. Data on sex, birth weight, gestational age, mode of delivery,

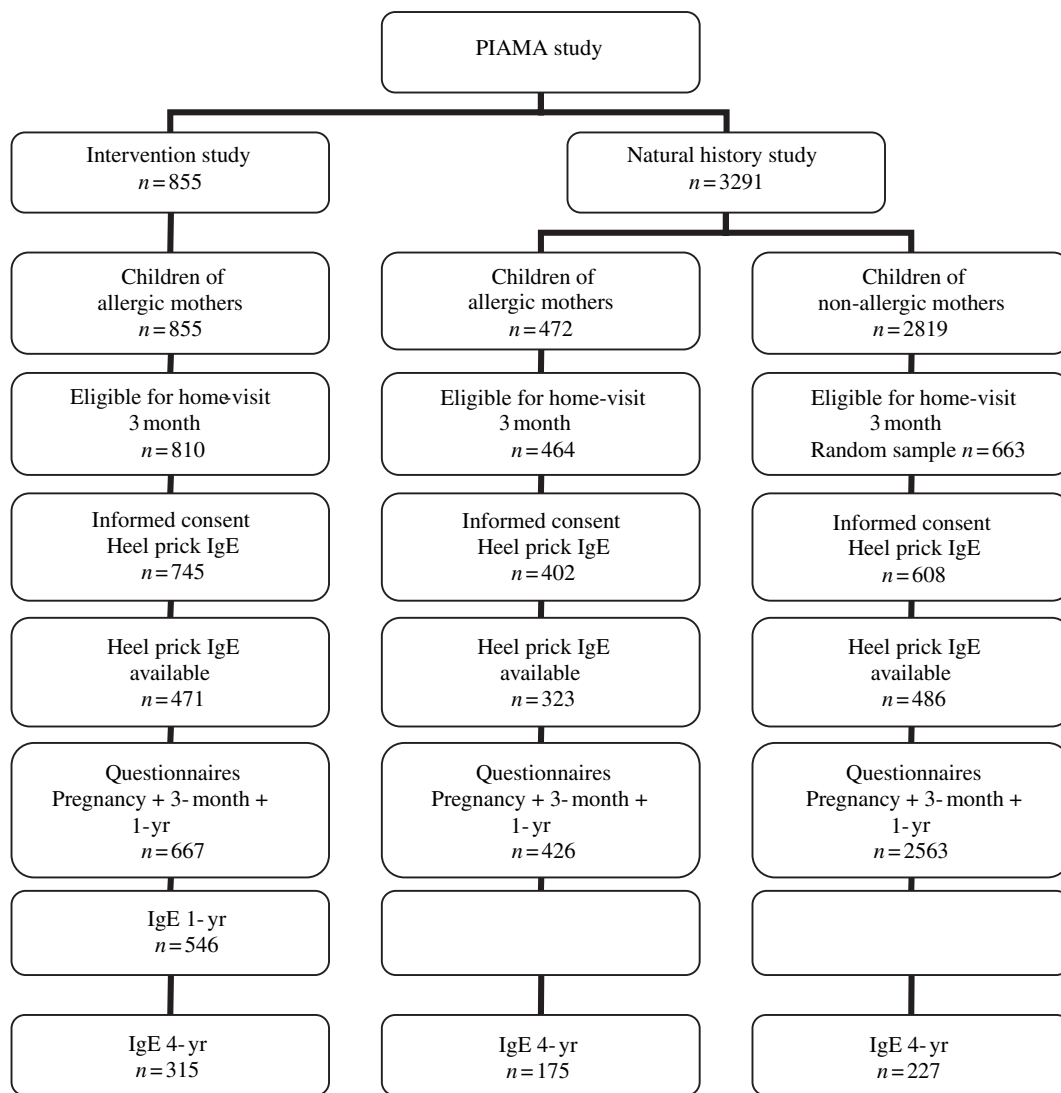


Fig. 1. Flow chart of the study population.

maternal age, and the presence of siblings were derived from the 3-month questionnaire. Data on educational level of the parents were obtained from the 12-month questionnaire.

Total IgE was measured in plasma obtained from a heel prick blood spot. Values of IgE below the detection limit were classified as negative. The limit of detection of IgE varied by the amount of plasma obtained from the spots, which, in turn, increased with the size of the blood spot. Eighty-eight percent of the measurements had a detection limit of 0.5 IU/ml or lower. When the detection limit was higher than the chosen cut off value of 0.5 IU/ml, a true positive value between 0.5 IU/ml and the detection limit will be falsely classified as negative. Therefore, all measurements with a detection limit above 0.5 IU/ml were excluded from the analysis.

Specific IgE was measured by Radio Allergo Sorbent Test and sensitization was defined as a specific IgE concentration higher than 0.35 IU/ml for at least one allergen (house dust mite, cat, dog, grass, milk or egg).

#### Analysis

Multiple logistic regression analysis was performed to estimate odds ratios (ORs) and 95% CIs of the association between a high neonatal total IgE level ( $\geq 0.50$  IU/ml) or sensitization (dependent variables) and individual risk factors adjusted for the other risk factors, region, and education level of the parents (independent variables). The following risk factors were studied: presence of cats or dogs in the home, presence of (older) siblings, season of birth, sex, being born by caesarian section, being born in hospital,

gestational age, birth weight, age of the mother and cigarette smoking of the mother. Gestational age was divided into four categories with 40 wk  $\pm$  7 days as the reference category and premature (< 37 wk), 37 or 38 wfk, and more than 41 wk as the other categories. Birth weight was divided into three categories ( $\leq$  3000, 3001–4000, and > 4000 grams) with 3001–4000 grams as reference category, which approximately corresponds to the interval of one standard deviation around the mean (2995–4055). Age of the mother was divided into quartiles:  $\leq$  28, 29–30, 31–33, and > 33 yr of age at birth of the child with the first quartile as reference.

To study whether a high total IgE level at birth or sensitization at 4 yr had different associations with risk factors in infants from allergic compared with infants from non-allergic mothers, multiple logistic regression analysis was performed stratified by allergy status of the mother. At 12 months of age no data of children from non-allergic mothers were available.

Observed differences were statistically tested by including an interaction term of allergy status of the mother and the risk factor in a combined model (not stratified by allergy status). The statistical package used for the analyses was SPSS version 11.0.1.

## Results

A high heel prick total IgE level ( $\geq$ 0.50 IU/ml) was found in 9.3% of the infants, 12.2% in boys and 6.2% in girls. Infants with a high total IgE more often had a mother or father with allergic disease than infants with lower or undetectable levels of total IgE (Table 1). There was a trend that infants with a high total IgE had less frequently pets (cats or dogs) at home (33% vs. 42%,  $p = 0.09$ ), which was less apparent for cats (26% vs. 30%,  $p = 0.41$ ) than for dogs (10% vs. 15%,  $p = 0.13$ ).

At 12 months of age, the prevalence of sensitization (29%) was measured only in the group of children from allergic mothers. At this age, sensitization was mainly found to milk or egg (28%) and rarely to the inhalant allergens house dust mite, cat or dog (3%).

Children with sensitization at 12 months had less frequently a mother who smoked during pregnancy (6% vs. 13%,  $p = 0.03$ ) or who had a cat at home during pregnancy (21% vs. 31%,  $p = 0.02$ ) than non-sensitized infants, whilst the presence of dogs at home during pregnancy was similar (13% vs. 12%,  $p = 0.91$ ). Furthermore, they were more frequently born in hospital (66% vs. 56%,  $p = 0.06$ ).

Table 1. Characteristics of infants with and without total IgE  $\geq$  0.50 IU/ml at birth ( $n = 1027$ ). Data are presented as mean (s.d.) or % ( $n$ )

|                                                 | IgE $\geq$ 0.50 IU/ml | IgE < 0.50 IU/ml | p-value* |
|-------------------------------------------------|-----------------------|------------------|----------|
| Number                                          | 95                    | 932              |          |
| Male gender, % ( $n$ )                          | 67 (64)               | 49 (459)         | <0.01    |
| Mean gestational age (wk)                       | 40 (2.0)              | 40 (1.5)         | 0.33     |
| Mean birth weight (g)                           | 3530 (552)            | 3513 (506)       | 0.77     |
| Mean age mother at birth (yr)                   | 30 (3.7)              | 30 (3.7)         | 1.00     |
| Mean age father at birth (yr)                   | 32 (4.4)              | 33 (4.3)         | 0.70     |
| Presence of sibling(s), % ( $n$ )               | 50 (47)               | 47 (438)         | 0.65     |
| Cigarette smoking mother pregnancy, % ( $n$ )   | 12 (11)               | 11 (104)         | 0.90     |
| Cat at home during pregnancy, % ( $n$ )         | 26 (25)               | 30 (283)         | 0.41     |
| Dog at home during pregnancy, % ( $n$ )         | 10 (9)                | 15 (86)          | 0.13     |
| Born by caesarian section, % ( $n$ )            | 7 (7)                 | 9 (87)           | 0.53     |
| Born in hospital, % ( $n$ )                     | 58 (55)               | 57 (528)         | 0.82     |
| Asthma or respiratory allergy mother, % ( $n$ ) | 75 (71)               | 60 (560)         | <0.01    |
| Asthma or respiratory allergy father, % ( $n$ ) | 39 (37)               | 29 (270)         | 0.04     |

\*Chi-squared test for proportions and  $t$ -test for continuous data.

At 4 yr of age, sensitization was present in 38% of the children: 40% in children from allergic mothers and 34% in children from non-allergic mothers. At this age, 28% was sensitized to milk or egg and 17% to inhalant allergens.

Smoking of the mother, a cat or dog at home and born in hospital were not differently distributed between sensitized and non-sensitized children. Children from allergic mothers with sensitization at 4 yr of age had more often a father with allergic disease than non-sensitized children from allergic mothers (42% vs. 29%,  $p < 0.01$ ), which was not found at 12 months of age (37% vs. 33%,  $p = 0.50$ ).

There were no significant sex differences in the prevalence of sensitization.

Association between prenatal risk factors and a high heel prick total IgE

The presence of cats or dogs at home during pregnancy was borderline significantly associated with a lower risk of a high total IgE level in heel prick blood of the infant in the first week of life [OR (95% CI) 0.6 (0.4–1.0),  $p = 0.06$ ]. Table 2 shows that this association was more outspoken for the presence of dogs than for cats [OR dogs 0.5 (0.2–1.0),  $p = 0.06$ ; OR cats 0.8 (0.5–1.3),  $p = 0.35$ ]. Exclusion of 208 subjects whose parents reported avoidance of pets at home because of allergic disease of one of the family members did not influence the associations [OR dogs 0.5 (0.2–1.1),  $p = 0.10$ ; OR cats 0.8 (0.5–1.5),  $p = 0.76$ ]. Exclusion of

Table 2. Association between a high neonatal total IgE ( $\geq 0.50$  IU/ml) and several risk factors (n = 1027)

| Risk factors                              | n   | Unadjusted odds ratio (95% CI) | Adjusted† odds ratio (95% CI) |
|-------------------------------------------|-----|--------------------------------|-------------------------------|
| Male gender                               | 523 | 2.1 (1.4–3.3)*                 | 2.4 (1.5–3.8)*                |
| Presence of siblings                      | 485 | 1.1 (0.7–1.7)                  | 1.0 (0.6–1.7)                 |
| Cat at home during pregnancy              | 305 | 0.8 (0.5–1.3)                  | 0.8 (0.5–1.3)                 |
| Dog at home during pregnancy              | 152 | 0.6 (0.3–1.2)                  | 0.5 (0.2–1.0)                 |
| Gestational age in weeks                  |     |                                |                               |
| <37                                       | 40  | 1.3 (0.5–3.3)                  | 1.6 (0.5–5.3)                 |
| 37–38                                     | 159 | 0.6 (0.3–1.2)                  | 0.6 (0.3–1.3)                 |
| 39–40                                     | 617 | Reference                      | Reference                     |
| >41                                       | 211 | 0.8 (0.4–1.3)                  | 0.8 (0.4–1.4)                 |
| Birth weight in gram                      |     |                                |                               |
| <3000                                     | 147 | 0.6 (0.3–1.2)                  | 0.5 (0.2–1.3)                 |
| 3000–3999                                 | 720 | Reference                      | Reference                     |
| >4000                                     | 160 | 1.0 (0.6–1.8)                  | 1.0 (0.5–1.8)                 |
| Age mother in years                       |     |                                |                               |
| ≤ 28                                      | 97  | Reference                      | Reference                     |
| 29–30                                     | 451 | 0.7 (0.3–1.4)                  | 0.7 (0.3–1.6)                 |
| 31–33                                     | 400 | 0.9 (0.4–1.8)                  | 1.0 (0.5–2.2)                 |
| >33                                       | 79  | 0.8 (0.3–2.1)                  | 0.9 (0.3–2.6)                 |
| Born in                                   |     |                                |                               |
| Spring                                    | 268 | Reference                      | Reference                     |
| Summer                                    | 193 | 1.2 (0.6–2.2)                  | 1.3 (0.7–2.5)                 |
| Autumn                                    | 302 | 1.0 (0.6–1.8)                  | 1.2 (0.6–2.2)                 |
| Winter                                    | 264 | 1.0 (0.6–1.8)                  | 1.0 (0.6–1.9)                 |
| Cigarette smoking mother during pregnancy | 115 | 1.0 (0.5–2.0)                  | 1.1 (0.5–2.3)                 |
| Born by caesarian section                 | 94  | 0.8 (0.3–1.7)                  | 0.8 (0.4–2.0)                 |
| Born in hospital                          | 583 | 1.1 (0.7–1.6)                  | 1.2 (0.7–1.9)                 |
| Asthma or respiratory allergy mother      | 631 | 2.0 (1.2–3.2)*                 | 1.9 (1.2–3.2)*                |
| Asthma or respiratory allergy father      | 307 | 1.6 (1.0–2.4)*                 | 1.6 (1.0–2.5)*                |

\*p &lt; 0.05.

†Adjusted for other risk factors, region and education level of parents.

434 infants whose parents reported a history of allergy to pets or whose siblings had asthma did not diminish the associations either [OR dogs 0.4 (0.1–1.3),  $p = 0.14$ ; OR cats 0.7 (0.4–1.5),  $p = 0.41$ ].

The presence of older siblings had no association with total IgE of the infant (Table 2). Neither was there an association between total IgE in heel prick blood and season of birth, cigarette smoking of the mother, birth weight, gestational age, maternal age and mode or place of delivery. Infants of allergic mothers had significantly more often a high heel prick total IgE [OR 1.9 (1.2–3.2),  $p = 0.01$ ]. The increased risk of infants from allergic fathers was borderline significant [OR 1.6 (1.0–2.5),  $p = 0.048$ ].

Additional adjustment for the presence and type of intervention with mattress covers did not change the associations.

Analysis stratified by allergy of the mother

The higher proportion of a high total IgE level at birth in boys compared with girls (Table 2) was only present in children from allergic mothers [OR 3.2 (1.8–5.7),  $p < 0.01$ ] (Table 3). The association between total IgE and the studied risk factors was not different according to the allergy status of the mother.

Association between prenatal risk factors and sensitization at 12 months and 4 yr

The presence of pets at home during pregnancy was associated with a lower risk of sensitization at 12 months of age [OR 0.6 (0.4–1.0),  $p = 0.06$ ], but not with sensitization at 4 yr of age. Furthermore, the negative association at 12 months was only present for cats [OR cats 0.6 (0.4–1.0),  $p = 0.04$ ] and not for dogs [OR dogs 1.3 (0.7–2.5),  $p = 0.40$ ] (Table 4). Excluding 171 subjects whose parents reported avoidance of pets at home because of allergic disease of one of the family members resulted in a non-significant association at 12 months of age [OR cats 0.7 (0.4–1.2),  $p = 0.19$ ].

The presence of siblings had no significant associations with sensitization at 12 months and 4 yr. Maternal smoking during pregnancy was negatively associated with sensitization at 12 months. In children from non-allergic mothers, a maternal age at birth older than 33 yr and being born in the autumn were significant risk factors for sensitization at 4 yr (Table 4).

Boys from allergic mothers were not having an increased risk of sensitization at 12 months of age, but at 4 yr they were [OR = 1.9 (1.2–2.8),  $p < 0.01$ ], whereas boys from non-allergic mothers had no increased risk at 4 yr. Asthma or allergy from the father was significantly associated with sensitization at 4 yr only [OR 1.5 (1.1–2.2),  $p = 0.02$ ], and specifically so if the mother was allergic as well [OR 1.7 (1.1–2.5),  $p = 0.02$ ].

## Discussion

We studied prenatal risk factors for the presence of high total IgE just after birth and sensitization at 12 months and 4 yr. We found that boys and children from an allergic or asthmatic mother or father had a significantly higher risk of high IgE levels in the first week of their lives. Boys also had a higher risk of allergic sensitization at age 4, and specifically when the mother was allergic. This was however, not the case at 12 months of age.

Table 3. Difference in the association between a high neonatal total IgE ( $\geq 0.50$  IU/ml) and several risk factors in children from allergic and non-allergic mothers

| Risk factors                                 | Non-allergic mother (n = 396),<br>odds ratio (95% CI)† | Allergic mother (n = 631),<br>odds ratio (95% CI)† | p-value interaction<br>term with allergy mother |
|----------------------------------------------|--------------------------------------------------------|----------------------------------------------------|-------------------------------------------------|
| Male gender                                  | 1.4 (0.6–3.3)                                          | 3.2 (1.8–5.7)*                                     | 0.11                                            |
| Presence of siblings                         | 0.5 (0.2–1.2)                                          | 1.4 (0.8–2.5)                                      | 0.13                                            |
| Cat at home                                  | 0.7 (0.2–1.8)                                          | 0.8 (0.4–1.5)                                      | 0.93                                            |
| Dog at home                                  | 0.2 (0.0–1.9)                                          | 0.6 (0.2–1.3)                                      | 0.13                                            |
| Gestational age in weeks                     |                                                        |                                                    |                                                 |
| <37                                          | 2.4 (0.2–29.1)                                         | 1.6 (0.4–6.9)                                      | 0.85                                            |
| 37–38                                        | 0.9 (0.2–3.7)                                          | 0.5 (0.2–1.3)                                      | 0.73                                            |
| 39–40                                        | Reference                                              | Reference                                          |                                                 |
| >41                                          | 0.8 (0.2–2.5)                                          | 0.7 (0.4–1.5)                                      | 0.76                                            |
| Birth weight in gram                         |                                                        |                                                    |                                                 |
| <3000                                        | 0.5 (0.1–2.5)                                          | 0.6 (0.2–1.7)                                      | 0.90                                            |
| 3000–3999                                    | Reference                                              | Reference                                          |                                                 |
| >4000                                        | 1.0 (0.3–3.9)                                          | 1.0 (0.5–2.1)                                      | 0.62                                            |
| Age mother in years                          |                                                        |                                                    |                                                 |
| ≤ 28                                         | Reference                                              | Reference                                          |                                                 |
| 29–30                                        | 0.5 (0.1–2.3)                                          | 0.8 (0.3–1.9)                                      | 0.61                                            |
| 31–33                                        | 1.7 (0.4–7.2)                                          | 0.9 (0.3–2.2)                                      | 0.58                                            |
| >33                                          | 1.0 (0.1–7.4)                                          | 0.8 (0.2–3.1)                                      | 0.99                                            |
| Born in                                      |                                                        |                                                    |                                                 |
| Spring                                       | Reference                                              | Reference                                          |                                                 |
| Summer                                       | 1.5 (0.4–5.4)                                          | 1.2 (0.5–2.7)                                      | 0.29                                            |
| Autumn                                       | 0.9 (0.3–2.9)                                          | 1.4 (0.7–2.9)                                      | 0.60                                            |
| Winter                                       | 0.7 (0.2–2.7)                                          | 1.2 (0.6–2.5)                                      | 0.53                                            |
| Cigarette smoking<br>mother during pregnancy | 1.7 (0.4–6.7)                                          | 1.0 (0.4–2.3)                                      | 0.73                                            |
| Born by caesarian section                    | 1.4 (0.3–7.4)                                          | 0.7 (0.3–2.1)                                      | 0.62                                            |
| Born in hospital                             | 0.6 (0.2–1.5)                                          | 1.5 (0.8–2.7)                                      | 0.23                                            |
| Asthma or respiratory allergy father         | 1.8 (0.7–4.7)                                          | 1.6 (0.9–2.7)                                      | 0.56                                            |

\*p < 0.05.

†Adjusted for other risk factors, region and education level of parents.

A borderline significant, negative association was found between the presence of pets at home during pregnancy and total IgE of the infant just after birth with a more pronounced effect from dogs than from cats. However, prenatal exposure to pets was not consistently associated with sensitization at 12 months and 4 yr of age. We observed that a cat at home during pregnancy was significantly associated with a lower prevalence of sensitization at 12 months, but could not exclude that selective avoidance of cats in families with a high risk of allergic disease was responsible for this association.

A protective effect of early-life exposure to pets on the development of allergic disease has been reported in several studies (8–10). Our findings suggest that this effect may already be operating *in utero*, but has no significance with regard to the development of sensitization in the first years of life. Several explanations must be considered for the negative association between the prenatal presence of pets and total IgE at birth. First, there could be a real protective effect. This may be explained by the fact that pets in the house are associated with higher levels of endotoxin (14).

There is increasing evidence that endotoxin exposure may protect against atopy by activating type-1 T-helper cell responses (15, 16). An alternative explanation may be that the lifestyle of households keeping pets is different in other aspects related to allergic disease. Also, parents of children with a genetic predisposition of allergic disease may selectively avoid keeping pets in the home. This explanation is unlikely for the effect on total IgE at birth, as in that case we would expect the negative association to diminish after exclusion of infants with a genetic predisposition of allergic disease, which did not occur in our population.

Neonatal IgE does not accurately predict development of allergic diseases later in life. However, there are strong arguments that a high total IgE measured at birth relates to atopy. Several studies have shown a high cord-blood IgE to be a risk factor for the development of allergic disease later in life (1, 17). In our data, infants with a high level of total IgE in heel prick blood had a higher prevalence of sensitization at the age of 12 months than infants with a low level of IgE (43% vs. 24%, p = 0.03). Thus,

Table 4. Association between sensitization at 12 months and 4 yr of age and several risk factors

| Risk factors                              | Odds ratio (95% CI)†                 |                            |                                     |                                 |
|-------------------------------------------|--------------------------------------|----------------------------|-------------------------------------|---------------------------------|
|                                           | 12 months allergic mothers (n = 492) | 4 yr total group (n = 682) | 4 yr non-allergic mothers (n = 222) | 4 yr allergic mothers (n = 460) |
| Male gender                               | 0.9 (0.6–1.3)                        | 1.3 (0.9–1.8)              | 0.6 (0.3–1.2)                       | 1.9 (1.2–2.8)*                  |
| Presence of siblings                      | 0.7 (0.5–1.2)                        | 0.8 (0.6–1.2)              | 0.8 (0.4–1.6)                       | 0.9 (0.6–1.3)                   |
| Cat at home during pregnancy              | 0.6 (0.4–1.0)*                       | 1.1 (0.8–1.6)              | 1.0 (0.5–1.9)                       | 1.1 (0.7–1.7)                   |
| Dog at home during pregnancy              | 1.3 (0.7–2.5)                        | 1.1 (0.7–1.7)              | 0.6 (0.2–1.6)                       | 1.5 (0.8–2.6)                   |
| Gestational age in weeks                  |                                      |                            |                                     |                                 |
| <37                                       | 0.8 (0.3–2.2)                        | 1.0 (0.4–2.4)              | 2.0 (0.4–10.9)                      | 0.7 (0.2–2.2)                   |
| 37–38                                     | 1.1 (0.6–2.0)                        | 0.7 (0.4–1.1)              | 1.0 (0.4–2.6)                       | 0.6 (0.4–1.2)                   |
| 39–40                                     | Reference                            | Reference                  | Reference                           | Reference                       |
| >41                                       | 1.6 (0.9–2.6)                        | 1.0 (0.7–1.6)              | 1.1 (0.5–2.5)                       | 1.0 (0.6–1.8)                   |
| Birth weight in gram                      |                                      |                            |                                     |                                 |
| <3000                                     | 1.7 (0.9–3.2)                        | 0.9 (0.5–1.5)              | 0.5 (0.2–1.6)                       | 1.2 (0.6–2.4)                   |
| 3000–3999                                 | Reference                            | Reference                  | Reference                           | Reference                       |
| >4000                                     | 1.4 (0.8–2.4)                        | 1.0 (0.6–1.6)              | 1.2 (0.5–2.9)                       | 1.0 (0.6–1.9)                   |
| Age mother in years                       |                                      |                            |                                     |                                 |
| ≤ 28                                      | Reference                            | Reference                  | Reference                           | Reference                       |
| 29–30                                     | 1.2 (0.5–2.6)                        | 0.8 (0.4–1.5)              | 1.1 (0.3–3.9)                       | 0.7 (0.3–1.5)                   |
| 31–33                                     | 1.2 (0.5–2.7)                        | 1.3 (0.7–2.5)              | 1.9 (0.6–6.9)                       | 1.2 (0.6–2.7)                   |
| >33                                       | 1.5 (0.6–4.2)                        | 1.9 (0.8–4.1)              | 6.7 (1.3–34.1)*                     | 1.2 (0.5–3.3)                   |
| Born in                                   |                                      |                            |                                     |                                 |
| Spring                                    | Reference                            | Reference                  | Reference                           | Reference                       |
| Summer                                    | 1.1 (0.6–2.0)                        | 1.1 (0.7–1.7)              | 0.6 (0.2–1.8)                       | 1.3 (0.7–2.2)                   |
| Autumn                                    | 1.2 (0.7–2.1)                        | 1.3 (0.8–2.0)              | 2.4 (1.0–5.4)*                      | 0.9 (0.5–1.5)                   |
| Winter                                    | 0.8 (0.4–1.6)                        | 0.8 (0.5–1.3)              | 1.5 (0.6–3.7)                       | 0.6 (0.3–1.1)                   |
| Cigarette smoking mother during pregnancy | 0.4 (0.2–1.0)*                       | 1.3 (0.8–2.2)              | 2.3 (0.8–6.5)                       | 1.1 (0.6–2.1)                   |
| Born by caesarian section                 | 1.1 (0.5–2.4)                        | 0.7 (0.4–1.2)              | 2.9 (0.8–10.7)                      | 1.2 (0.6–2.5)                   |
| Born in hospital                          | 1.2 (0.8–2.0)                        | 0.9 (0.6–1.2)              | 1.1 (0.5–2.8)                       | 0.8 (0.5–1.3)                   |
| Asthma or respiratory allergy mother      | Not applicable                       | 1.2 (0.9–1.7)              | Not applicable                      | Not applicable                  |
| Asthma or respiratory allergy father      | 1.2 (0.8–1.8)                        | 1.5 (1.1–2.2)*             | 1.3 (0.7–2.6)                       | 1.7 (1.1–2.5)*                  |

\*p &lt; 0.05.

†Adjusted for other risk factors, region and education level of parents.

some children may have an increased tendency to develop sensitization early in life, possibly expressed as elevated levels of total IgE at birth. However, neonatal IgE was not significantly associated with the prevalence of sensitization at the age of 4 yr (40% vs. 36%,  $p = 0.58$ ).

Risk factors for early sensitization may be different from risk factors for later sensitization. Besides, the risk of developing asthma may be higher for early sensitized children than for children with later sensitization. It has been reported that especially children from atopic parents with early persistent sensitization have a high risk to develop asthma (18).

Our results do not support the hypothesis that neonatal IgE levels decrease with increasing birth order (19). Devereux et al. (20) showed that the magnitude of proliferative responses to allergens in cord blood mononuclear cells decreased significantly with increasing birth order. We found a tendency of a negative association between the presence of siblings and neonatal IgE in infants from non-allergic mothers. Our population includes a relatively small proportion of children

from non-allergic mothers and a limited number of siblings. This may explain the absence of an association between total IgE and the presence of siblings in our study.

We found no association between the season of birth and neonatal total IgE. Results from previous studies on seasonal variation of cord blood IgE are inconsistent. Some studies reported no association (21, 22). Other studies reported lower levels of IgE in children born in the autumn (23, 24), while opposite effects, i.e. higher levels of IgE with birth during late autumn or winter, were also found (25, 26). Seasonal variation in neonatal IgE may be the result of variation in climatic conditions, which may explain these inconsistencies.

We found no significant association between neonatal total IgE and gestational age, birth weight, maternal age and cigarette smoking, consistent with previous reports (21, 23–25). It has recently been suggested that caesarean section delivery might be a risk factor for the development of atopic diseases because of its gut flora modulating properties (27). However, the

94 children of mothers who delivered their child by caesarean section did not have higher total IgE levels at birth or a higher prevalence of sensitization in the first 4 yr of life than children who were born by a vaginal delivery in our population.

Previous studies found maternal, but not paternal allergic disease to be associated with higher IgE levels in the neonate (19, 21, 22). We found both paternal and maternal allergy to increase the risk of a high total IgE level at birth.

Paternal, but not maternal allergy, was a risk factor for sensitization at 4 yr of age, consistent with the results from another birth cohort study (28). However, there are other studies that found the opposite (29).

Many studies have reported that boys have higher total IgE levels at birth than girls (18, 21, 25). Our observation is of importance, as we have measured total IgE in blood obtained in the first week of life. Previous studies measured IgE in cord blood and could not exclude contamination with maternal blood. Interestingly, we found that a sex-related difference in neonatal total IgE and sensitization is limited to infants from mothers with allergic disease. Recently, similar findings were reported from another birth cohort study with regard to persistent wheezing in the first 4 yr of life (30). An interpretation of our finding could be that there is stronger transmission of maternal allergy in boys. This could reflect genetic or non-genetic factors such as programming of the fetus or trans-placental passage of maternally derived cytokines. These effects may be stronger in boys than in girls for reasons unknown. Whether the observed effect may account for the higher prevalence of atopic disease in boys than in girls is as yet unknown. Our study has a long follow up and future analyses may reveal the importance of our finding.

Complete data were obtained from 1027 of the 1982 infants who were originally selected. The loss of data was in most cases caused by the absence of a heel prick blood spot that was large enough to measure IgE, although informed consent of the parents was obtained. This happened more often in children of allergic mothers than in children of non-allergic mothers, for which no plausible explanation could be found. It is unlikely that this may have biased our results, as neonatal screening and the subsequent storage of the extra spot occurred independent of our study. The remaining relatively high loss of subjects is caused by the fact that our study has a longitudinal design with several moments in time at which data were collected, which increases the

probability of data not being complete. At each moment the response rates were sufficiently high. Eighty-nine percent of the parents of the children that were initially included in the PIAMA study, gave informed consent to measure IgE in heel prick blood. The successive response rates of the questionnaires among subjects of whom IgE was available, were 99% at pregnancy, 96% at 3 months and 93% at 12 months of age.

In summary, we investigated whether previously reported risk factors of allergic disease during the last trimester of pregnancy are associated with higher neonatal IgE levels and found a negative, borderline significant, association between the presence of dogs in the home and a high total IgE level at birth. Moreover, we found that both maternal and paternal allergy increased the risk of an elevated total IgE level at birth. Sensitization at 12 months and 4 yr of age had different risk factors, i.e. cigarette smoking was protective at 12 months in children of allergic mothers, but not at age 4. In contrast, boys had a higher risk of sensitization at age 4, especially when the mother was allergic. Our results suggest that neonatal IgE depends on prenatal factors, especially the presence of dogs in the household. However, we found no indication that prenatal exposure has a prolonged effect on sensitization.

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